Aquaculture and pollution studies of an Australian fish silver perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae)

by

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Thesis submitted for the fulfilment of the requirements for the Degree of Doctor of Philosophy

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DECLARATION

I **Golam Kibria** declare that the thesis entitled

"Aquaculture and pollution studies of an Australian fish silver perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae)"

is my own work and has not been submitted previously, in whole or in part in respect of any academic award.

Signature of candidate

28.11.97

Date

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ABSTRACT

The silver perch, *Bidyanus bidyanus* is an Australian native fish of great aquaculture potential. Currently, silver perch represent a major freshwater aquaculture industry in Australia. There have been some research on the biology and nutrition of silver perch but until now there has been no research on the quality and quantity of wastes that may result from aquaculture of silver perch or other species in Australia. The present study was undertaken primarily to investigate the biological growth of silver perch juveniles fed on artificial and sewage grown zooplankton, and quantifies solid and nutrient load from rearing of silver perch at different temperatures. Silver perch juveniles were reared at three temperatures (20^{o}C, 25^{o}C and 30^{o}C) and fed on three commercial diets referred to as diet-1, diet-2 and diet-3. The protein and phosphorus content of three diets are : diet-1 (53% and 1.31%), diet-2 (45%, 1.16%), and diet-3 (36% and 1.28%) respectively.

Of the three diets fed to silver perch, diet-2 containing 45% protein, resulted comparatively higher growth rate though statistical analysis did not reveal any significant difference in weight gain of fish fed the three diets ($P>0.05$). The gain in weight decreased as diet-2>diet-1>diet-3. Silver perch reared at 25^{o}C resulted significantly better food conversion ratio (FCR) and specific growth rate (SGR) in the order of 25^{o}C>30^{o}C>20^{o}C ($P<0.05$). The analysis of sewage grown zooplankton revealed an attractive biochemical composition, containing 54.34%-64.8% protein, 7.29%-7.73% fat and 1.11%-1.14% phosphorus. Silver perch fed on *Daphnia carinata* resulted significantly higher gain in weight, SGR and FCR over *Moina australiensis* ($P<0.05$).

The diet which resulted in a comparatively higher growth and a better FCR, produced less suspended and dissolved solid waste to the environment ($P>0.05$). The solid waste production was significantly lower at 25^{o}C compared to fish reared at 30^{o}C and 20^{o}C ($P<0.05$). The average solid waste production at 25^{o}C was 284 kg.tonne^{-1} fish production. The main path of nitrogen loss in the aquaculture of silver perch was found to be via gill excretion (85.7%-90.2%) and via faeces (9.8%-14.3%). The hourly excretion rate of ammonia showed a sharp increase soon after a meal and a linear decrease during the remaining 24 hours. The daily ammonia excretion rate increased with an increase in temperature and was significantly higher at 30^{o}C than at 25^{o}C ($P<0.05$). A linear relationship was found between nitrogen intake and the loss of nitrogen in faeces. Faecal nitrogen loss was higher at 30^{o}C than at 25^{o}C ($P<0.05$). The nitrogen retention by silver perch was found significantly greater at 25^{o}C than at 30^{o}C ($P<0.05$), and average nitrogen retained at 25^{o}C and 30^{o}C were 43.1% and 29.4% respectively ($P<0.05$). The main path of phosphorus loss by silver perch was via faeces. At 25^{o}C, 66.7% of the phosphorus loss was in particulate form and the remaining 33.3% in dissolved form. Fish fed on diet-2 excreted comparatively less orthophosphate than either by diet-1 and diet-3 fed to fish ($P>0.05$). Similar to ammonia excretion, there was a sharp increase in orthophosphate excretion soon after a meal which also decreased linearly during the remaining 24 hours. The daily orthophosphate excretion increased with an increase in temperature and was

significantly higher at 30°C than at 25°C ($P<0.05$). The phosphorus retention by silver perch was significantly better at 25°C than at 30°C ($P<0.05$). The average phosphorus retained at 25°C was 49.1% and at 30°C it was 24.5% ($P<0.05$). The release of nutrients from fish food was depended upon pH and temperature of the environment. The release of orthophosphate and ammonium was higher at higher temperatures. The release of phosphate was accelerated in acidic media whereas ammonium in neutral to alkaline media. The fish food which contained the highest concentration of phosphorus generated faeces with most labile phosphorus. When silver perch juveniles were reared at 0 salinity, 4 salinity, 8 salinity and 12 salinity, the best growth rate and higher nutrient retention was achieved at 4 salinity ($P<0.05$).

The present study demonstrated that silver perch can grow significantly faster at a temperature close to it's optimum. It also show that sewage grown zooplankton was an inexpensive and alternative diet in the rearing of silver perch juveniles. The study reveals that culture of fish at their optimum temperature may enhance nutrient retention and a reduction in the discharge of nutrient to the environment. The study further indicated that rearing of silver perch in a slightly saline conditions may be another option for achieving better growth rates and provide a lower nutrient loading to the environment.

LIST OF PAPERS

Paper I. Aspects of phosphorus pollution from aquaculture. (1996). ***Naga, The ICLARM Quarterly***, 9(3), 20-24.

Paper II. Australian native species in Aquaculture. (1996). ***The Victorian Naturalist*** 113(5), 264-267.

Paper III. Pollution from aquaculture. (1997). ***Chemistry in Australia***. 64(1), 19-20.

Paper IV. Preliminary rearing trials of an Australian native fish, silver perch (*Bidyanus bidyanus*) (Mitchell) with reference to growth and production of solid waste in aquaculture. (1997). ***Asian Fisheries Science*** 9, 301-309.

Paper V. Australian native freshwater fish and crustaceans : environmental role and aquaculture potential. (1997). ***World Aquaculture***, 28 (4), 56-62.

Paper VI. Zooplankton : biochemistry and significance to aquaculture. (1997). ***Naga, The ICLARM Quarterly*** 20(2) : 8-15 (in press).

Paper VII. The nutrient content and the release of nutrients from fish food and faeces. (1997). ***Hydrobiologia*** 357 : 165-171.

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Paper X. Effect of temperature on nitrogen losses and nitrogen retention in silver perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae) fed on artificial diets. (1997). ***Journal of Aquaculture in the Tropics*** (revised ms submitted).

Paper XI. Biology and aquaculture of silver perch : A review. (1998). ***The Victorian Naturalist*** 115 : 230-236.

Paper XII. Nitrogen pollution from aquaculture : Can it be reduced?. (1998). ***Naga, The ICLARM Quarterly*** 21 (1) : (accepted).

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Paper XIV. Biological growth of silver perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae) reared at different temperatures. (1997). ***Journal of the World Aquaculture Society*** (submitted)

Abstracts Published

I. Phosphorus balance in a simulated aquaculture system : A case study with the native Australian fish silver perch, *Bidyanus bidyanus*. (1995). ***Fourth Asian Fisheries Forum. Organized by Asian Fisheries Society and The China Society of Fisheries***. Beijing, China 116-20 October 1995. 5p.

II. Patterns of nutrient discharge by silver perch *Bidyanus bidyanus* reared at different temperatures. World Aquaculture 97. (1997). In : *Book of abstracts* - ***The annual international conference and exposition of the World Aquaculture Society. Seattle, Washington, USA.*** February 19-23, 1997. 179p.

III. Nitrogen pollution from aquaculture : Nitrogen losses and nitrogen retention by silver perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae). (1997). ***Joint conference of Australian Society for Fish Biology (ASFB) and Fish and Aquatic Resources Management Association for Australasia (FARMAA)***. Darwin, Northern Territory, Australia. 15-19 July 1997. 65p.

IV. Sewage grown zooplankton as an alternative feed of Australian native fish silver perch, *Bidyanus bidyanus*. (1996). ***VI International Symposium on Nutrition and Feeding of Fish. College Station, Texas, USA***. August 11-15, 1996. Poster 14.

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GENERAL INTRODUCTION

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GENERAL INTRODUCTION

Aquaculture is an important food production industry and it is becoming increasingly popular for the increase in demand for fresh and healthy foods. Although aquaculture is an infant industry, interest in the culture of fish and other aquatic organisms has increased as a result of population increase, interest in research on native species, availability of research funding for aquaculture research (CRC for Aquaculture 1996/97), over-exploitation of some major commercial fisheries (Rowling 1990) and importation of large quantities of fish and fishery products (Jarzynski 1991).

Aquaculture in Australia is expanding steadily with a marked increase production in the last five years (Table 1.1.). There has been a 240% increase in aquaculture production since 1988-89 (O'Sullivan and Kiley 1996). The highly valued Atlantic salmon (*Salmo salar*), Rainbow trout (*Oncorhynchus mykiss*), barramundi (*Lates calcarifer*) and the giant tiger prawn (*Penaeus monodon*) have been the main species cultured recently (O'Sullivan and Kiley 1996). There is an increase in competition since these species (as indicated above) are costly to produce and they are also cultured in other countries in Europe and Asia (Rowland *et al.* 1995).

Table 1.1. Australian aquaculture production data for the financial years 1988/89 to 1994/95 (O.Sullivan & Kiley 1996)

Year	Tonnes (000's)	Value (A$) (million)
1988/89	11.9	135.9
1989/90	12.4	188.0
1990/91	14.3	237.5
1991/92	16.2	254.1
1992/93	16.8	255.5
1993/94	21.7	318.9
1994/95	23.5	418.7

Australia has few freshwater fish (180-190 species), of which 127 are endemic to Australia (Allen 1989). It is generally believed that the limited number of freshwater fish in Australia may be a reflection of a long geographical isolation of the continent. Nevertheless, the Australian native fish and native crayfish form the main freshwater aquaculture industry of the country (Kibria *et al.* 1996a). Among the native fish, four species have been identified as potential for aquaculture. These are the silver perch (*Bidyanus bidyanus*), Golden perch (*Macquaria ambigua*), Murray cod (*Maccullochella peeli*) and Freshwater catfish (*Tandanus tandanus*) (Hume and Barlow 1993).

The native silver perch is regarded as the species with highest potential for aquaculture in Australia (Allan and Rowland 1996) because it possesses a number of preferred characteristics including being easy to culture in earthen ponds (Rowland *et al.* 1995), utilizing of natural zooplankton as a food (Rowland 1984; 1986), rapid growth rates, acceptance of artificial feeds, and being amenable to crowding (Barlow 1986; Rowland and Barlow 1991; Allan and Rowland 1992; Rowland and Kearney 1992). It is one of the species of the Murray-Darling River system that is much sought after by commercial and recreational fishers (Cadwallader 1979). Considerable interest has been shown in culturing silver perch in countries like, China and Taiwan (Gooley and Rowland 1993) and I did receive great interest in silver perch following my talk to the 4th Asian Fisheries Forum held in Beijing, China (16-20 October 1995) (Kibria *et al.* 1996a). At present, silver perch farming is very popular as demonstrated by the increasing interest and investment in the culture of the species (Kibria *et al.* 1996a). The following pages contain a critical literature review on aspects of silver perch biology and aquaculture, biochemical composition of zooplankton and significance to aquaculture, solid waste production, and nitrogen and phosphorus pollution from aquaculture which are the main topics of my research study.

1.1 THE BIOLOGY AND CULTURE OF SILVER PERCH

History, natural habitats and status

Silver perch, *Bidyanus bidyanus* (Mitchell, 1838) (Teraponidae) is a native Australian freshwater fish (Figure 1.1.) endemic to most of the Murray-Darling river system (Merrick and Schmida 1984) except in the cool high upper reaches of streams (Lake 1967a; Merrick and Schmida 1984; Pollard *et al.* 1990). It is a potamodromous species, i.e., migrate wholly within freshwater habitat (Guo *et al.* 1995). The species was commonly consumed by the Aborigines and the scientific name was also derived from the Aboriginal name *bidyan* (Rowland 1995a) by the explorer Major Thomas Mitchell who named the fish after he caught it from the Namoi River in 1832 (Mitchell, 1838).

The species was once abundant and widespread throughout the Murray-Darling River system but its distribution and abundance have been greatly reduced and it is now uncommon in many areas (Rowland 1994). The decline in population of the species is thought to be due to increased competition for food from introduced cyprinids e.g. *Cyprinus carpio*, predation by the English perch, *Perca fluviatilis*, and the construction of dams which prevented upstream migration of the species (Cadwallader and Backhouse 1983). It is now categorized as a "Potentially Threatened" species (Jackson 1994) and its status may turn into "Endangered" unless measures are taken to increase its population (Rowland 1995a). Such measures might include aquaculture since *B. bidyanus* has a number of characteristics that makes the farming of the species a viable proposition for aquaculture.

1.1.2 Biological characteristics of the silver perch

The species can tolerate a wide range of temperature, from 2.0^{o}C to 32.0^{o}C (Cadwallader and Backhouse 1983), but the optimum growth temperature range is believed to be 23.0^{o}C - 28.0^{o}C (Rowland *et al.* 1995). Apart from extremes

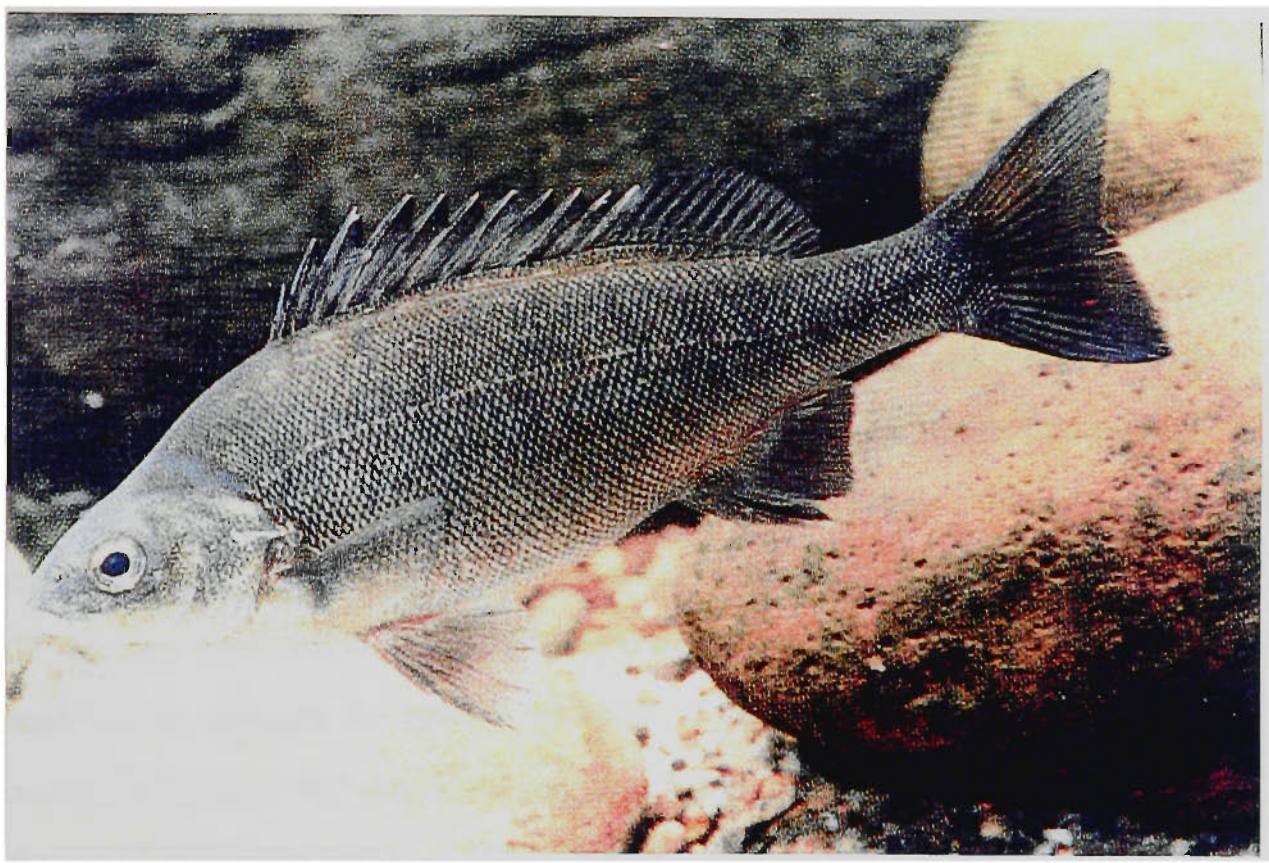

Figure 1.1. The Silver perch, *Bidyanus bidyanus* is a major freshwater aquaculture industry in Australia. Demands to culture the species is increasing both in Australia and overseas.

of temperature, higher ammonia concentration could significantly reduce the growth of silver perch during the hottest months (Rowland 1995b). Table 1.1.1. lists the water quality parameters that are recommended for culture of silver perch.

Table 1.1.1. Water quality variables recommended for intensive silver perch aquaculture (Rowland 1995b)

Variables	Recommended	Optimum for growth
Temperature (°C)	10-30	23-28
Dissolved oxygen (mg/l)	>4.5	
pH	7.0-9.5	
Total ammonia nitrogen (TAN) (mg/l)	<2.0	
NH_3 (mg/l) (unionised ammonia)	<0.1	

Male silver perch become mature in their second year (233 mm) while females mature in their third year (340 mm) (Cadwallader and Backhouse 1983; Merrick and Schmida 1984). However, adult fish may die after spawning (Lake 1967b; Cadwallader and Backhouse 1983). Spawning occurs in summer (November - January) when sexually mature fish migrate upstream to spawn in shallow warm waters (Cadwallader 1977; Merrick 1980; Cadwallader and Backhouse 1983; Reynolds 1983). Flooding is thought to be required for natural spawning of silver perch (Davis 1977). The fecundity is high and the number of eggs per female varies from 300,000 (Merrick 1980) to 500,000 eggs (Whitley 1960). Eggs are pelagic with a diameter of 2-8mm (Lake 1967c). Hatching occurs about 30 hours after eggs are laid at temperatures of 22.0-31.0^{o}C (Cadwallader and Backhouse 1983). The larvae are benthic (Lake 1967c) and juveniles form large schools (Lake 1967a; Merrick 1980) which often congregating below rapids, weirs (Merrick 1980) and fast flowing water with sand, or gravel bottom (Llewellyn 1983; Merrick and Schmida 1984; Starling 1992). Adults can live in extremely turbid waters (Cadwallader 1977) and the species is tolerant of high salt concentrations (Ingram *et al.* 1996). Silver perch is also a territorial and aggressive fish (Cadwallader and Backhouse 1983; Starling 1992). The average size at two years of age is around 180 mm (Cadwallader and Backhouse 1983) and common sizes usually caught are 350-410 mm (0.75kg - 2.5 kg) (Merrick and Schmida 1984). The

pre-spawning activities of *B. bidyanus* have been described by Merrick and Schmida (1984).

Food and feeding habitats

The young fish (larvae) starts feeding from the sixth day after hatching and feeds mainly on zooplankton including rotifers, copepod nauplii, small copepods and cladocerans (Cadwallader and Backhouse 1983; Rowland 1984; Thurstan and Rowland 1995). Adult fish are omnivorous and, at times, feed extensively on zooplankton, particularly the larger ostracods and cladocerans. Other food includes shrimps (*Macrobrachium* sp., atyids), yabbies, chironomid larvae, aquatic insects, earthworms, molluscs, filamentous algae, and aquatic plants (Lake 1967a; Cadwallader and Backhouse 1983; Merrick and Schmida 1984; Rowland 1994). The natural zooplankton identified from earthen ponds under *B. bidyanus* culture is comprised of *(Moina micrura, Daphnia carinata* (cladocerans), *Boeckella fluvialis* (copepod) and *Brachionus calyciflorus, Asplanchna sieboidi* (rotifers) (Culver and Geddes 1993).

Aquaculture of Silver Perch

There are a number of characteristics that make silver perch an ideal species for aquaculture, which are : a rapid and uniform growth in crowded conditions (Barlow 1986; Pollard 1986), (the current stocking densities in earthen ponds is 10,000 fingerling/ha with aeration and 5000 fingerlings/ha without aeration; Walker 1993), effective utilization of both plant proteins and meat meals (Allan and Rowland 1996), low production costs, its high fecundity and ready acceptance of low-protein diets (Barlow 1986; Rowland and Barlow 1991; Walker 1994a,1994b). Silver perch are also tolerant to high temperatures (Pollard 1986), and omnivorous feeding habits (Rowland and Kearney 1992). Silver perch farming is a major native fish culture industry in this country as demonstrated by its production figures (Table 1.1.2 and 1.1.3) and has the potential to achieve an annual production of up to 10 t/ha (Walker 1994a; Rowland 1995c). Major *B. bidyanus* farms are small (1-3 ponds) although

there are some medium sized farms (4-8 ponds) and a few large farms (6-17 hectares) (Walker and Caney 1996).

Table 1.1.2. Comparison of native silver perch and native finfish production during 1994/95 (O'Sullivan and Kiley 1996). [Key : NDA = No details available; NSW = New South Wales; NT=Northern Territory; TAS=Tasmania; SA=South Australia; QLD=Queensland; VIC=Victoria; WA=Western Australia].

	farm (tonnes)	hatchery (000's)	value ($,000)
FINFISH			
Statewise Silver Perch production			
Silver Perch (NSW)	17.3	1,807.3	635.8
Silver Perch (VIC)	1	NDA	10
Silver Perch (QLD)	34.4	400.0	331.9
Silver Perch (SA)	-	-	-
Silver Perch (WA)	-	-	-
Silver Perch (TAS)	-	-	-
Silver Perch (NT)	-	-	-
Silver Perch and other native finfish production during 1994/95			
Silver Perch (all states)	52.7	2,207.3	977.7
Golden perch (all states)	0.5	2,498.9	397.0
Murray cod (all states)	<0.1	196.7	101.3
Trout cod (all states)	no data	32.1	3.4
Australian bass (all states)	<0.1	274.3	95.6
Catfish (all states)	no data	44.5	42.6
Macquarie perch (all states)	no data	53.0	5.4
Mary River cod (all states)	no data	4.7	7.0

The aquaculture of silver perch is highest in New South Wales (NSW), and in Queensland (QLD); and these two states contributed 98% of *B. bidyanus* production in 1994/1995 (Table 1.1.2). The increase in freshwater fish production in Australia is mainly due to the interest and investment in growing *B. bidyanus* (Kibria *et al.* 1996a) (see also Table 1.1.3).

Table 1.1.3. Native fish production and contribution from silver perch to aquaculture from 1988 to 1995.

Year	Native fish farm production (t)	Silver Perch contribution (t)	Source
1988-89	10	-	Tradwell *et al.* (1992)
1991-92	43.9	26.6	O'Sullivan (1995)
1994-95	53.2	52.7	O'Sullivan & Kiley (1996)

To achieve the best result for raising silver perch, research has been undertaken by a number of different authors on the breeding, nutrition, growth performance in ponds, tanks, and saline waters. These findings are summarized below :

The artificial breeding of Silver Perch

Techniques on the artificial propagation of silver perch, along with other native species have been developed at the Inland Fisheries Research Station, Narrandera, Eastern Freshwater Fish Research Hatchery, Grafton in New South Wales, and at Snobs Creek Freshwater Fisheries Research Station and Hatchery in Victoria (Rowland 1983; Rowland 1984). The techniques involved inducing breeding of native fish using artificial hormone (human chorionic gonadotrophin, HCG) and a preparation of the pituitary gland from common carp. The optimum dose of HCG that induced a high hatching rate of eggs in silver perch was 200 IU/kg HCG (Rowland 1984). About three million silver perch fry are produced annually by induced breeding to stock farm dams, and other natural waters used for recreational fishing (Rowland *et al.* 1995).

Nutrition of silver perch

Research has been undertaken on the nutrition and production of silver perch by a number of authors (Rowland and Barlow 1991; Allan and Rowland 1992; Allan and Rowland 1994; O'Sullivan 1994; Allan 1995). Allan and Rowland (1991) obtained the fastest growth rate of *B. bidyanus* using the diet containing 36% protein using three experimental diets at 21%, 36% and 49% protein. Based on this trial, Allan and Rowland (1992) have developed the first reference diet for *B. bidyanus* which containing 36% protein, 5.5% fat, and 1.1% fatty acids. The total methionine and total lysine composition (the limiting amino acids in feed) of the reference diet was 7.4 and 22.6g/kg respectively. The food conversion ratio (FCR) was found to be affected by both the protein content of diets and rearing temperatures (Table 1.1.4). *B. bidyanus* fed on a diet containing 36% protein showed better food conversion ratio (FCR) than silver perch fed at other levels of protein (Allan and Rowland 1991).

Table 1.1.4. Effect of dietary protein levels and rearing temperature on food conversion ratio (FCR) of silver perch.

Protein content (%)	Feeding rate % BW[1]/day	Water temperature	FCR	Culture system	Source
20.7%	satiation	18.3°-22.8°C	3.0±0.2	Tank	Allan and Rowland (1991)
36%	satiation	18.3°-22.8°C	1.7±0.1	Tank	Allan and Rowland (1991)
49%	satiation	18.3°-22.8°C	2.4±0.3	Tank	Allan and Rowland (1991)
35%	5%	22.0°C-26.6°C	1.1-1.2	Pond	Rowland *et al.* (1994)
35%	4% & 3%	13.2°C-28.4°C	1.8-1.9	Pond	Rowland *et al.* (1995)
35%	3%	-	1.0-1.3	-	Allan and Rowland (1992)
35%	3%	12.5°C-30.3°C	2.3	Pond	Rowland (1995d)
50%	3%	22.0°C-31.3°C	0.7	Pond	Rowland (1994)*

[1]BW = body weight; * = Rowland commented that FCR was better apparently due to eating of natural pond food as well.

Research has also been undertaken on the use of Australian oilseeds (soybean, canola, cottonseed, and peanut) and grain legumes (lupins, chick pea, field pea and cow pea) as ingredient of silver perch diets and results are encouraging, since apparent digestibility coefficients (ADC's) of vegetable protein is similar to, or higher than that obtained with fish meal (Allan and Rowland 1994). The research demonstrated that silver perch is very efficient in digesting vegetable protein, and the best growth of the fish was obtained from peanut meal, followed by soybean meal, lupin and canola meal. However, research found that growth of silver perch decreased, and the FCR value deteriorated with an increase in plant protein content. Very poor FCRs was reported when silver perch were fed on diet with a high fibre content (O'Sullivan 1994). Studies were also conducted on the effects of varying protein and energy concentrations, with results showing that the growth of silver perch increased with increasing protein and energy level in diets (Allan *et al.* 1994). The fat deposition in *B. bidyanus* was directly related to the fat content of diets (Anderson and Arthington 1989; Hunter *et al.* 1994). Further experimental trials confirmed that silver perch require both the linolenic (18:3n-3) and linoleic (18:2n-6) series of fatty acids for optimal growth (Anderson and Arthington 1989). A synopsis of nutritional research on silver perch has been published by Allan and Rowland (1996).

Growth and production of Silver Perch

Growth of silver perch is affected by the rearing temperature. During the winter months (May-September), the growth rate is significantly slower than in the warmer months (October to March) (Table 1.1.5). Silver perch eat aggressively at a temperatures above 20°C and are less aggressive at lower temperatures. Although silver perch grow better in temperatures above 20°C, the prolonged exposure at 30°C and above appear to adversely affect the appetite, food conversion and growth of the species (Rowland 1995b).

Table 1.1.5. Growth of silver perch reared in ponds at different seasons.

Months	Seasons	Temperature	Growth (g/d)	Source
May-September	Winter	11.1-20°C	0.5	Rowland (1995c)
October-March	Summer	>20°C	2.3 (low density)	Rowland (1995c)
October-March	Summer	>20°C	2.1 (high density)	Rowland (1995c)

The silver perch fry grew faster at a stocking density of 25,000/ha than at 80,000/ha in earthen ponds (Rowland *et al.* 1994). When silver perch were reared at 43,000 fish/ha, the annual production figure was calculated to be 10.2 tonnes/ha (Rowland 1995d; Rowland *et al.* 1995). However, at higher densities, in particular during summer, the water quality may deteriorate resulting an increase of disease susceptibility (Rowland 1995d.).

Performance of Silver Perch in tanks/cages

McKinnon *et al.* (1996) obtained comparatively better growth and survival of silver perch in floating cages fixed in irrigation channels in integrated aquaculture-irrigation systems, while poor growth and survival resulted rearing the fish from cages fixed in tanks supplied with groundwater. The growth and FCR of silver perch was significantly lower in tanks than in earthen ponds (Rowland 1995c). Similarly, growth of *B. bidyanus* were found to be slower in aquaria compared to commercial ponds (Kibria *et al.* 1997a.).

Performance of Silver Perch in saline waters

Silver perch can tolerate salinity as high as 15 ppt (Guo *et al.* 1995) and larvae hatched at 6 salinity showed better survival rate than those hatched in freshwater (Guo *et al.* 1993). Ingram *et al.* (1996) reported a good survival and growth of silver perch when reared in cages at lower salinities of 8.0-15.3 ppt, whereas poor growth and survival resulted at higher salinities of 9.5-24.6 ppt. The growth and survival obtained at 8.0-15.3 salinities were better than the growth and survival of similar sized fish reared in freshwater cages (Ingram *et al.* 1996).

Information regarding other aspects of the aquaculture of *B. bidyanus* has been provided in a number of recent publications as given in Table 1.1.6.

Table 1.1.6 List of recent publications on silver perch aquaculture

	References
Culture in farm dams	Barlow (1995)
Site selection & design of ponds for silver perch culture	Ogburn *et al.* (1995)
Artificial breeding of silver perch	Thurstan & Rowland (1995)
Fingerlings and market size silver perch production	Rowland (1995c)
Water quality criteria for silver perch	Rowland (1995b)
Diseases of silver perch	Callinan & Rowland (1995)
Artificial diets for silver perch	Allan (1995); Allan & Rowland (1996)

1.2 ZOOPLANKTON : BIOCHEMICAL COMPOSITION AND SIGNIFICANCE TO AQUACULTURE

Zooplankton are the natural food of freshwater and marine fish and crustaceans. Practically all species of fish and prawn depends on zooplankton at one stage or another during their life span. Some species may feed exclusively on zooplankton during their entire life, for example, about 90% of the herring *(Clupea harengus)* food consists of zooplankton (Arrhenius and Hansson 1993). Success or failure in culturing of planktonivorous fish fry depends primarily on the zooplankton, their composition

and density (Geiger 1983; Fernando 1994). Zooplankton have been used to rear fry and larvae (De Pauw *et al.* 1981; Watanabe *et al* 1983) and as feed for species which normally do not accept artificial feed (Bryant and Matty 1980). Both live (Dabrowski 1984; Alam *et al.* 1993) and frozen zooplankton (Brett 1971; Sargent *et al.* 1979) have been used for commercial and experimental aquaculture. Zooplankton are a valuable source of protein, amino-acids, lipids, fatty-acids, minerals and enzymes and could be an inexpensive ingredient to replace expensive fishmeal. There have been few studies on the chemical composition of zooplankton although such information is vital to evaluate a species and it's suitability in aquaculture (Watanabe *et al.* 1983; Millamena *et al.* 1990).

Biochemical composition of zooplankton

Several researchers have demonstrated that the biochemical composition of zooplankton is attractive for aquaculture of finfish and crustaceans (Table 1.2.1, 1.2.2). The average protein content of different zooplankton species are, *Daphnia* sp.63.32±10.3, *Moina* sp. 67.49±6.25, (Crustaceans : Cladocerans) and *Brachionus* sp. 62.03±3.42 (Rotifers), *Cyclops* sp. 63.98±13.31 (Copepods). There is a wide variation in crude fat content, which can be influenced by the nutritional input in the culture media. The lipid content in *Daphnia* sp. is in the range of 4.5-23.6% whereas in *Moina* sp. it varied between 7.73-27.22% (Table 1.2.1.). The biochemical composition in natural zooplankton varies seasonally (Khan and Qayyum 1971; Donnelly *et al.* 1994) and could be affected by the level of nutrients in waters (Vijverberg and Frank 1976). The protein, lipid and phosphorus contents in most zooplankton appear to satisfy the requirements of fish (Table 1.2.1). The content of essential amino-acids (EAA) in zooplankton shows that it can match the EAA requirements of fish (Table 1.2.2). This agrees with the findings of Yurkowski and Tabachek (1979) who reported that the essential amino-acids content of the cladoceran, *Daphnia pulex*, copepods, *Diaptomus* sp. and *Cyclops* sp. is equal to or greater than the amino acid requirement of chinook salmon. Lysine and methionine are known to be the most limiting amino

Table 1.2.1. The relative amounts of moisture, protein, lipid, carbohydrate, ash, phosphorus and calcium expressed as percentages of the total organic matter in zooplankton used for aquaculture (dry matter basis). The nutrient requirement for omnivorous and carnivorous fish are also included for comparison [Legend : Mois=Moisture; CP=Crude protein; CF=Crude fat; CHO=Carbohydrate; P=Phosphorus; Ca=Calcium].

	Mois.	CP	CF	CHO	Ash	P	Ca	Source
Cladocerans								
Daphnia sp.	89.3	70.1	13.07	-	6.53	1.40	0.19	1
Daphnia sp.	-	45.0	04.5	-	-	-	-	8
Daphnia sp.	89.3	70.1	13.07	-	6.54	1.46	0.21	11
D. magna	-	68.0	13.1	17.9	-	-	-	5
D. magna	-	45-50	-	-	-	-	-	16
D. magna	-	56.3	10.7	-	-	-	-	9
D. pulex (natural pond)	91.2	65.6	23.6	-	-	-	-	6
D. pulex	94.0	50.0	16.66	-	20.0	-	-	1
D. pulex	94.0	49.7	16.3	4.9	19.3	-	-	2
D. pulex (horse manure)	89.8	66.8	20.9	-	-	-	-	6
D. pulicaria	-	78.1	14.6	7.30	-	-	-	5
D. longispina	-	75.6	12.2	12.2	-	-	-	5
D. obtusa	-	67.5	8.60	23.9	-	-	-	5
D. hyalina	-	69.4	24.3	6.3	-	2.0	-	7
D. carinata (sewage)	92.9	54.3	7.29	27.1	11.3	1.14	-	10
Moina sp. (bakers yeast)	87.2	68.55	22.13	-	-	1.41	0.08	1
Moina sp. (yeast+ manure)	89.0	77.85	11.81	-	-	1.1	0.09	1
Moina sp. (poultry manure)	87.9	59.12	27.22	-	-	1.32	0.16	1
M. micrura	-	65.1	8.7	-	-	-	-	4
M. asutraliensis (sewage)	93.7	64.8	7.73	20.65	6.82	1.11	-	10
M. micrura (chicken manure)	-	69.53	9.94	-	6.80	-	-	14
Copepods								
Cyclops sp.	-	69.3	14.8	-	-	-	-	9
C. vicinus	-	69.2	6.0	24.8	-	-	-	5
C. sphacricus	-	73.2	18.5	8.4	-	-	2.1	7
Diaptomus sp.	92.4	57.89	25.06	-	5.26	-	-	1
D. gracills	-	76.0	9.4	14.6	-	-	-	5
D. castor	-	85.0	9.5	5.5	-	-	-	5
Tigriopus japonicus (natural)	88.6	71.00	22.80	4.38	0.79	0.09	-	1
T. Japonicus (yeast)	87.2	69.48	20.48	4.67	0.94	0.16	-	1
T. Japonicus (yeast+chlorella)	86.6	67.16	23.81	3.73	0.97	0.15	-	1
Calanus plumchrus	89.0	58.20	21.80	0.50	9.40	-	-	15
Copepods (Sargasso sea)	86.15	51.25	-	-	-	0.75	-	19
Copepods (Continental)	87.00	36.87	-	-	20.50	0.60	-	19
Copepods (North Pacific)	78.45	56.87	-	-	04.00	-	-	19
Rotifers								
Brachionus plicatilis (yeast)	90.8	64.3	15.1	-	10.9	1.11	0.16	11
B. plicatilis (yeast+ chlorella)	87.9	63.7	23.1	-	6.78	1.38	0.11	11
B. plicatilis (chlorella)	86.4	58.1	27.4	-	6.62	1.54	0.15	11
B. plicatilis	-	56.92	12.8	16.68	13.60	1.42	-	17
Euphausiaceas								
Krillmeal	-	56.2	9.2	-	15.9	2.08	4.00	3
Krill meal	-	41.8	13.0	11.3	-	1.48	-	13
Euphausia pacifica	82.0	33.33	27.77	-	27.77	1.61	2.57	1
E. superba -	56.3	10.7	-	15.0	2.0	3.6	12	
Euphausids	80.50	38.44	-	-	20.50	1.05	-	19
Recommended dietary nutrient levels for fish								
Omnivorous fish								
Fry (0.05g)	-	42	8	-	-	1.0	2.5	18
Fingerling (0.5-10g)	-	39	7	-	-	0.9	2.5	18
Juvenile (10-50g)	-	37	7	-	-	0.8	2.0	18
Carnivorous fish								
Fry (0.05g)	-	52	16	-	-	1.0	2.5	18
Fingerling (0.5-10g)	-	49	14	-	-	0.8	2.5	18
Juvenile (10-50g)	-	47	14	-	-	0.8	2.0	18

1.Creswell (1993); 2.Yurkowski and Tabachek (1979); 3.Koops *et al.* (1979); 4.Tay *et al.* (1991);. 5. Blazka (1966); 6. Mims *et al.* (1991); 7.Vijverberg and Frank (1965); 8.Anon (1996); 9.Aqua Company, U.K (personal contact); 10. Present study (chapter 4); 11.Watanabe *et al.* (1983); 12.Lukowicz (1979); 13.Hilge (1979); 14.Alam *et al.* (1993); 15. Brett (1971).; 16. De Pauw *et al.* (1981); 17.Millamena *et al.* (1990); 18.Creswell (1993 quoted from Tacon 1990); 19.Parsons *et al.* (1984).

Table 1.2.2. Essential amino-acid composition of zooplankton (% total amino-acid as dry matter) compared with the essential amino-acid requirements of omnivorous and carnivorous fish. [Legend : Arg=Arginine; His=Histidine; Iso=Isoleucine; Leu=Leucine; Lys=Lysine; Met=Methionine; Phe=Phenylalanine; Thr=Threonine; Val=Valine]

	Arg	His	Iso	Leu	Lys	Met	Phe	Thr	Try	Val	Source
Cladocerans											
Daphnia sp	5.7	1.9	5.6	7.4	7.5	2.5	4.2	-	1.3	6.2	1
Daphnia sp	5.7	2.6	3.9	7.9	7.4	1.7	9.2	3.5	-	5.2	3
Daphnia sp. (*Chlorella*+yeast)	4.91	1.23	7.09	10.8	8.37	1.29	3.78	3.87	0.67	10.1	10
D. pulex	5.6	2.1	5.6	8.2	8.8	2.3	4.7	5.2	-	6.4	2
D. pulex (807-945 um)	2.67	1.15	1.72	2.8	3.34	0.94	2.31	2.67	-	2.44	4
D. pulex (854-1350um)	14.4	7.6	1.4	1.4	2.4	2.2	1.3	2.1	-	2.2	4
D. magna (waste water)	2.23	0.57	0.91	1.51	1.84	0.03	1.84	1.64	-	1.85	7
D. carinata (sewage grown)	3.37	1.65	2.60	5.22	3.35	1.46	2.52	2.86	0.71	3.27	9
Moina sp	5.1	1.6	2.5	6.0	5.8	1.0	3.6	-	1.2	3.2	1
Moina sp.	5.1	1.6	2.5	6.0	5.8	1.0	3.6	3.8	1.2	3.2	6
Moina sp.	7.0	2.2	3.4	8.3	8.0	1.4	4.9	5.2	1.6	4.4	8
Moina sp. (*Chlorella*+yeast)	11.7	1.94	5.9	11.1	6.9	1.11	3.31	6.1	0.95	8.42	10
Moina australiensis (sewage grown)	3.01	1.37	2.67	4.54	3.34	1.13	2.67	2.79	0.76	3.56	9
Copepods											
Cyclops strenus (540-730um)	4.69	1.45	2.69	4.29	5.52	1.14	2.97	2.88	-	3.31	4
C. strenus (810-1147um)	2.41	0.98	1.79	2.69	2.26	0.88	1.98	2.00	-	2.75	4
Tigriopus japonicus	5.2	1.6	2.5	5.0	5.7	1.1	3.5	-	1.1	3.2	1
T. japonicus	5.2	1.6	2.5	5.0	3.3	1.1	3.5	3.8	-	3.3	5
T. japonicus	6.9	2.1	3.3	6.6	7.5	1.5	4.6	5.0	1.5	4.3	8
Rotifers											
Brachionus sp. 200um	4.04	3.16	2.9	4.89	5.87	1.48	4.11	2.83	-	4.46	4
B. plicatilis	4.6	1.5	3.5	6.0	5.9	0.9	3.8	-	1.3	6.2	1
B. plicatilis	6.3	2.1	4.8	8.2	8.2	1.2	5.3	4.7	1.6	5.5	8
B. plicatilis (yeast)	4.2	1.4	2.9	5.5	5.7	0.8	3.5	3.5	3.0	3.6	6
B. plicatilis (Yeast+Chlorella)	4.5	1.4	2.8	5.3	5.8	0.8	3.4	3.1	3.0	3.5	6
Euphausiaceas											
Antartic Krill, *Euphausia superba*	6.22	2.30	5.10	7.77	8.58	3.03	6.47	4.70	1.50	5.90	5
Minimum essential amino-acids requirements in fish											
Omnivorous fish											
Fry (0.05g)	1.81	0.76	1.18	2.15	2.48	0.81	1.22	1.35	0.25	1.40	11
Fingerling (0.5-10g)	1.68	0.71	1.09	1.99	2.31	0.75	1.13	1.26	0.23	1.30	11
Juvenile (10-50g)	1.59	0.67	1.04	1.89	2.19	0.71	1.07	1.19	0.22	1.23	11
Carnivorous fish											
Fry (0.05g)	2.24	0.95	1.46	2.66	3.08	1.00	1.51	1.67	0.31	1.73	11
Fingerling (0.5-10g)	2.11	0.89	1.37	2.50	2.90	0.94	1.42	1.58	0.29	1.63	11
Juvenile (10-50g)	2.02	0.85	1.32	2.40	2.78	0.90	1.36	1.51	0.28	1.56	11

1. Hepher (1988); 2. Yurkowski and Tabachek (1979); 3. Block (1959); 4. Dabrowski. and Rusiecki (1983); 5. Suzuki (1981); 6. Watanabe *et al.* (1983); 7. Allen and Allen (1981); 8. Creswell (1993); 9. Present study (chapter 4); 10. Kokova *et al.* (1990); 11. Creswell (1993, Quoted from Tacon 1990).

-acids in feeds (Dabrowski and Rusiecki 1983; New 1987), although these two amino-acids are present in appreciable quantities in zooplankton studied (Table 1.2.2). There is a seasonal variation in the amino-acids content of zooplankton (Yurkowski and Tabachek 1979), moreover the trophic factor and taxonomic position could have an affect on the amino acids composition of zooplankton (Sadykhov *et al.* 1975). The fatty acids composition of zooplankton is influenced by the fatty acids composition of their diet (Watanabe *et al.* 1983; Proulx and Noüe 1985) and may change as the seasonal succession of phytoplankton species occurs (Jeffries 1970). The high ratio of unsaturated fatty acids to saturated fatty acids of zooplankton may denote that zooplankton is a high quality food for rearing of commercial fish larvae (Lokman 1994). Both docosahexaenoic acid (DHA) and eicosaptaenoic acid (EPA) are present in considerable amounts in zooplankton (Lokman 1994) and these two fatty acids are essential for the growth and development of fish (Kanazawa *et al.* 1979; Tucker 1992). However, Watanabe *et al.* (1983) reported that in some cases *Daphnia* sp. could be deficient of DHA whereas *Moina* sp. could be deficient in both DHA and EPA. An enrichment of zooplankters would improve the n-3 highly unsaturated fatty acid (HUFA) content of zooplankton (Watanabe *et al.* 1983; Tucker 1992; Fernnández-Reiriz, *et al.* 1993). Further information on lipids and the lipid classes of freshwater zooplankton can be found in Cavaletto *et al.* (1989), Watanabe *et al.* (1983), Mims *et al.* (1991).

Performance of fish fed on zooplankton

A faster growth rate was observed in Crayfish, *(Cherax albidus)* fed on live *Daphnia* sp. than fed on dry pellets (Jones *et al.* 1995a, 1995b); similarly, carp (*Cyprinus carpio*) and Atlantic salmon fed on zooplankton (Holm and Moller 1984; Kamler *et al.* 1992) performed better than those fed on formulated diets. LaBrasseur (1969) has established a high growth rate and good food conversion of Chum salmon (*Oncorhynchus nerka*) fed on live crustacean zooplankton. Watanabe *et al.* (1983)

reported an excellent protein efficiency ratio (PER) in rainbow trout fed with *Daphnia* sp. and *Moina* sp. Fish larvae in general, demonstrated a higher growth fed on live zooplankton than on artificial diets (Dave 1989).

Herring and trout offered frozen calanoid copepods, *Calanus finmarchicus* assimilated more than 90% of the dry matter in zooplankton (Sargent *et al.* 1979). Fish fed on frozen zooplankton were healthier (Sargent *et al.* 1979), with good survival (Dabrowski *et al.* 1984) and growth compared to artificial dry diets (Fermin and Boliver 1994). However, a comparatively lower growth and feed efficiency was reported for sockeye salmon with frozen zooplankton (Brett 1971), which according to author was unexpected. The protein efficiency ratio (PER) was also better with the krill meal (Koops *et al.* 1979). Frozen zooplankton represents an excellent food source for hatchlings of white fish, carp, and lake char (Bryant and Matty 1980).

Merits and demerits of using zooplankton

Zooplankton are regarded as an important source of carotene. Fish fed on copepods and krill (*Euphausia pacifica*) were found to be more pigmented than on commercial feed (Sargent *et al.* 1979; Spenelli 1979), a characteristic important for marketing of salmonids. The flavour and texture of fish have been improved when fed with zooplankton (Spenelli 1979). Live zooplankton contains enzymes (amylase, proteases, exonuclease, esterase) which could play an important role in larval digestion (Munilla-Moran *et al.* 1990). Both live and frozen zooplankton are attractive to fish. Zooplankton enhances the metamorphosis of larvae (Fluchter 1980), are nutritious, tastier, and easily digestible; furthermore, the chilled and frozen zooplankton floats and are therefore easily caught by fish (Tucker 1992). The carotenoid in zooplankton may have a positive effect on the growth of Atlantic salmon in the initial feeding phase (Torrissen 1984). The high contents of amino-acids (Dabrowski and Rusiecki 1983), enzymes (Horvath *et al.* 1979; Lauff and Hofer 1984) and water (Lemm 1983) in zooplankton are all positive qualities for start feeding (Holm and Moller 1984). Free

amino acids are present in the frozen fluid that surrounds the zooplankton and these constitute a powerful attractant and appetite stimulant for fish (Dabrowski and Rusiecki 1983; Mearns 1986; Tucker 1992).

Frozen zooplankton can release soluble material into the water through the freeze-damaged cells. As a result a large percentage of free amino acids could disappear from the food material. Free and protein bound amino acids could be lost in this way from zooplankton (Anon 1970; Grabner *et al.* 1981/1982) which may affect the growth of fish (Brett 1971). Rapid freezing might avoid both soluble and insoluble organic loss from fragmented pieces (Anon 1970). Pelleting the freeze dried zooplankton could be an alternative method that reduces the rate of leaching (Grabner *et al.* 1981/1982). The high fibre content in some zooplankton may depress the digestibility of other nutrients (Koops *et al.* 1979). The sulphur amino acids (methionine & cysteine) in most zooplankton are not sufficient to meet the expected requirements of fish (Yurkowski and Tabachek 1979; Dabrowski and Rusiecki 1983).

Zooplankton are rich in essential amino acids and fatty acids and should be sufficient as a source of nutrients which are required by fish for optimum growth. The high mortality of the young fish is thought to be a result of a diet deficient in essential nutrients, specially lipids (D'Abramo and Lovell 1991). In this regard zooplankton could be an inexpensive food for growing fish. Two fatty acids, EPA and DHA, which are essential for the growth and development of fish (Kanazawa *et al.* 1979) are present in zooplankton. Zooplankton are palatable and easy to handle. Zooplankton have been widely used for the rearing of larvae or fry and most studies have shown that the fry perform better when fed with live zooplankton than when fed with dry artificial diets (Dabrowski 1984; Dabrowski *et al.* 1984; Dave 1989). In larviculture, artificial diets may perform poorly due to poor digestibility (Dabrowski 1984; Lauff and Hofer 1984), deficiency of growth factors (Higgs *et al.* 1985), insufficient stimulation of feeding behaviour (Holm 1986) or pollution due to overfeeding (Dave 1989). The

zooplankton with a close to ideal biochemical composition (Watanabe *et al.* 1983) would therefore be a suitable first food for fish. Carotenoids in zooplankton may function as an antioxidant in eggs and larvae (Tacon 1981) in addition to pigmenting the flesh. There is already some indication that fish meal can be replaced by zooplankton based meal (Koops *et al.* 1979) and freshwater zooplankton (*Moina* sp.) could be a suitable substitute for expensive *Artemia* sp. (Alam *et al.* 1993). However, further research is needed on the development of zooplankton based dry diets (pellets) which could reduce the leaching of nutrients seen in frozen zooplankton. Experiments on total or partial replacement of fish meal by zooplankton meal could be another area for research. It is also essential to quantify the heavy metals, pesticides and levels other contaminants in fish fed on waste/sewage grown zooplankton before using them at large scale. In summary, zooplankton whether live or frozen constitutes a very useful supplements in the diets of farmed fish (Brett 1971).

1.3 SOLID WASTE PRODUCTION FROM AQUACULTURE

Solids waste is an important and basic measure of effluent quality. It is the major source of wastes in aquaculture (Cho 1993). Information on solid waste production are of paramount importance for waste management and the development of an ecologically sustainable aquaculture programme. The solid waste in aquaculture are waste feed (uneaten feed and fines), undigested feed (faeces), mucus, intestinal cells, bacteria and scales (Phillips and Beveridge 1986; Beveridge *et al.* 1991; Cripps 1993a,1993b; Costa-Pierce 1996; Cripps and Kelly 1996) (Figure 1.3.1). In general, the undigested starch from grain by-product, and ash from the animal bones are the main components of solid wastes from aquaculture (Cho 1993). About half of the phosphorus and 10-23% of the nitrogen of the fish diet can be lost as solid waste i.e., via uneaten food and faecal pellets (Hall *et al.* 1992; Cho 1993).

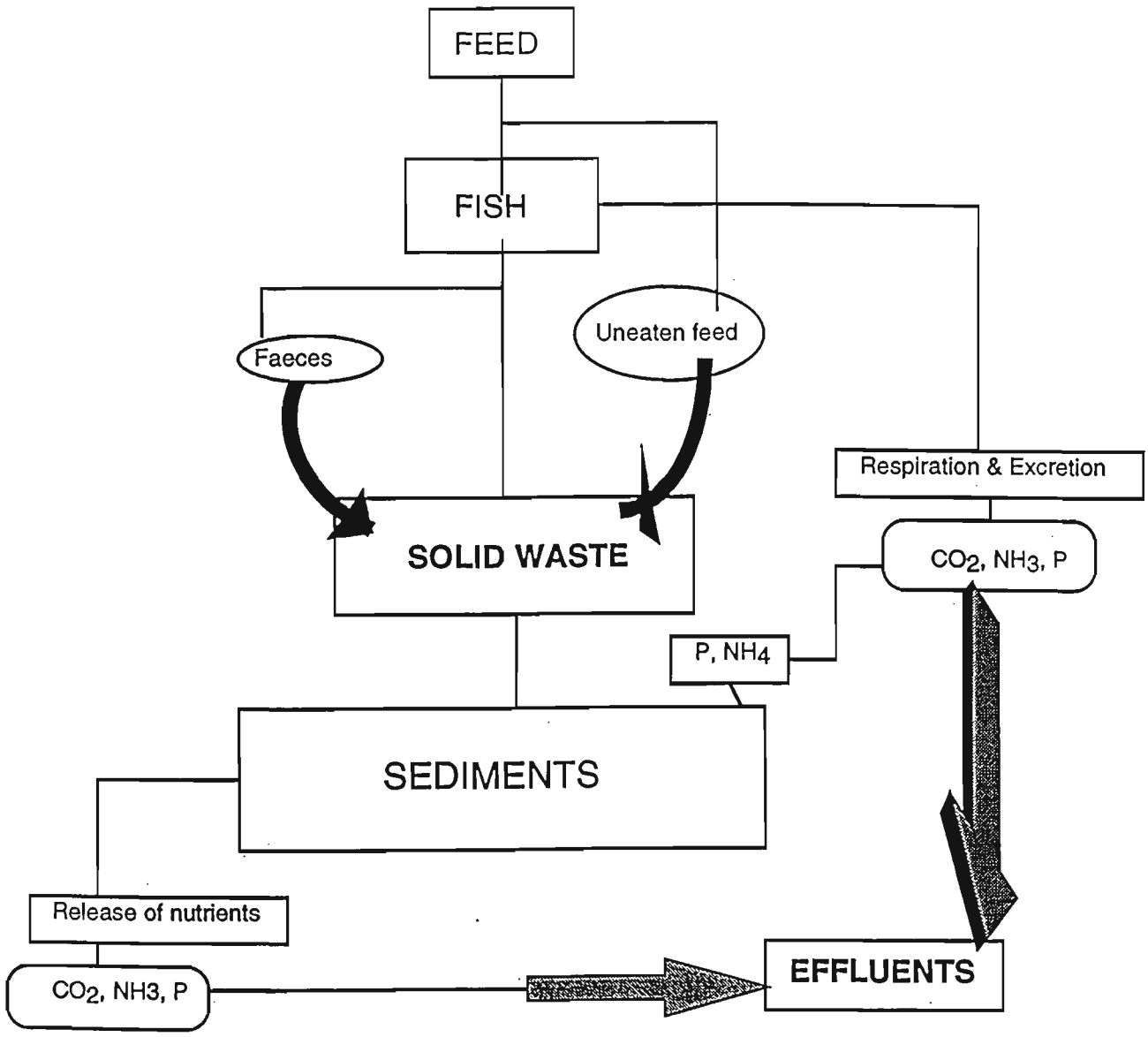

Figure 1.3.1. A general flow chart of origin of solid waste and nutrients from feeding of fish in ponds. A significant amount of solid waste produce will be accumulated into pond sediments. The nutrients generates from respiration and excretion of fish and from sediments containing solid waste of aquaculture. Nitrogen is lost mainly as ammonia via gill excretion and phosphorus via faecal pellets (Source : Boyd & Tucker 1995; Kibria *et al.* 1997b).

Factors related to solid waste production

The amount of solid waste depends on the system of culture, type of feed and management practised (Beveridge *et al.* 1991). For example, there could be a variation in solid output between tanks and ponds (Albaster 1982; Beveridge 1984) and between and within cage farms (Bergheim *et al.* 1982). Furthermore, methods used in collecting and analyzing of samples may cause further variation (Merican and Phillips 1985). The major proportion of solid output occurs during summer in the temperate zone and during harvesting in semi-intensive ponds (Boyd 1978; IOA 1990; Costa-Pierce 1996). The solid waste output can rise by 100-200 times during tank cleaning (Bergheim *et al.* 1982). The semi-intensive ponds produce environmentally insignificant amounts of solid waste both in temperate and tropical ponds (Boyd 1978; Edwards 1993; Schwartz & Boyd 1994) where the bulk of solid waste accumulates into the ponds sediments (Edwards 1993) (Figure 1.3.1). Factors such as, feeding rates (Merican and Phillips 1985; Liao 1970; Butz and Vens-Cappell 1982), digestibility of diet, food conversion ratio (FCR), pellet size, stocking densities, health of the fish, feeding schedules (Kilambi *et al.* 1976; Merican and Phillips 1985), and seasonal temperature (Kilambi *et al.* 1976) can influence the quantity of solid output. In addition to the above, water quality, species, size of fish, season of culture, efficiency of production and harvesting schedules may further influence the amount of solid wastes output (Boyd 1978; Bergheim *et al.* 1982; Merican and Phillips 1985; Beveridge 1987; IOA 1990; Beveridge *et al.* 1991; Schwartz & Boyd 1994). It was found that feed ingredients containing a high concentration of fibre, chitin and indigestible carbohydrate may increase the excretion of suspended solids (Lall 1991). Merican and Phillips (1985) found that feeding rate rather than fish size has been the most important determinant of solid waste production in rainbow trout.

Relationship between solids and particle size

The number of particles produced in solid wastes of aquaculture has been reported to be high, ranging from 1833-1,880,000 particles/l (Cripps 1993a). The nutrient

concentration (nitrogen and phosphorus) in suspended solids may be related to the particle size (Table 1.3.1.). The size of solid particles produced depends upon species cultured, and size of fish, the amount of mixing and shear force in holding facilities (Muir 1982).

Table. 1.3.1. Variation in the percentage of nutrients associated with the particulate fraction.

Particle size (μm)	Total nitrogen (TN)	Total phosphorus (TP)	Reference
> 0.45	26.2	30	Foy & Rosell (1991a)
> 0.45	20	25	Cripps (1995)
>0.60	7-32	47-84	Bergheim *et al.* 1993

Nutrient in solid waste and the release of nutrients

Merican and Phillips (1985) conducted extensive research on solid waste production and reported that solid waste from rainbow trout comprised 35.6% nitrogen and 65.9% phosphorus. The results of others suggest about 20-30% nitrogen (Bergheim *et al.* 1993; Cripps 1995) and 65% phosphorus (Yarris 1981) in solid waste from aquaculture. Ketola and Harland (1993) stated that phosphorus concentration in solid waste output is influenced by the type of diet and the level of phosphorus in diet. Factors such as, feeding rate, feed composition and water temperature may determine the proportion of nutrients lost through solid waste (Foy and Rosell 1991b; Kelly *et al.* 1994) (see also chapter 8 for nutrient content and the release of nutrients from solid waste). The nutrient leaching from solid waste to dissolved fractions depends on pH, temperature, ionic composition, nutrient concentration, flow of water and agitation of the water (Boersen & Westers 1986; Pontius 1990).

Solid waste production

Solid waste from cage culture could be substantial (Merican and Phillips 1985) and may account for 5-40% of the total food fed to fish (Phillips and Beveridge 1986). The feed loss from salmon and trout culture has been estimated to be 5-20%. It has been reported that even in balanced feed, 15-20% of eaten food is indigestible (Asgard *et al.*

1986). Feed wastes with trash fish and moist diets are significantly higher than that of commercial dry diets (Warren-Hansen 1982) and consequently result in a higher solid output (Warren-Hansen 1982). On the other hand extruded diets produced least solid waste output compared to fish fed on moist diets, the ratio of solid waste output between an extruded diet : solid waste was 1: 0.269% and with moist diet : solid waste was 1 : 1.08% (Stewart 1997).

Beveridge *et al.* (1991) calculated that faecal waste in salmonids, carp and shrimps are about 26-27% of ingested feed whereas with catfish it is 41%. The faecal output in rainbow trout was reported to be 259 g dry matter/kg food fed. (Butz and Vens-Cappell 1982). The majority of research on aquaculture wastes has been conducted in temperate Europe and North America where reported solid waste production (kg/t fish produced) markedly varied, from 243 kg/t (Cho *et al.* 1991) to a higher figure of 2153 kg/ t (NCC 1990) (Table 1.3.2.).

Table 1.3.2. Solid waste loadings (kg/tonne fish produced) from various aquaculture species.

Species	Solid waste	Reference
Salmonids	289-2153	NCC (1990)
Rainbow trout	290-655	Phillips & Beveridge (1986)
Rainbow trout	1350	Solbe (1982)
Rainbow trout in cages	389	Merican & Phillips (1985)
Rainbow trout in tanks	259	Butz & Vens-Cappell (1982)
Trout	550	Warren-Hansen (1982)
Brown trout	243	Cho *et al.* (1991)
Species not mentioned	510.7±207.3	IOA (1990)
Catfish	2302	Boyd & Tucker (1995)
Silver perch	284-549	Present study (chapter 5)

The suspended solids concentrations as effluents from fish farms are typically in the range of 3-14 mg/l. whereas data from shrimp farms appears to be much higher (Table 1.3.3).

Table 1.3.3. Reported suspended solids concentration in fish farm effluent.

Country	Species	Suspended solids (mg/l)	References
21 EIFAC farm	Trout & salmon	9.0	Albaster (1982)
Norway	Salmon	3.0	Bergheim *et al.* (1991a)
Norway	Salmon	1.6-14.1	Bergheim *et al.* (1993)
Sweden	-	6.9	Cripps (1995)
U.K.	-	5.0-50.0	Muir (1982)
31 UK farms	Rainbow trout	11.1	Solbe (1982)
Denmark	Rainbow trout	5-50	Warren-Hansen (1982)
USA	Channel catfish	>30.0	Boyd & Tucker (1995)
Thailand	Shrimp	30.0-190.0	Phillips *et al.* (1993)

Impacts of solid waste on the environment

Solid waste discharged from aquaculture may cause depletion of dissolved oxygen and disappearance of natural fish species from flowing rivers. Sludge worms, *Tubifex* may increase in numbers by actively feeding on organic sludge (solid matter) from the bottom. An increase in chironomid insects, snails numbers and high counts of protozoa and extensive algal growths could result in water bodies receiving suspended solids/organic matter (Connell 1993). By degradation, nutrients (nitrogen and phosphorus) from solid waste are released (see equation 1 & 2 below) which might cause further deterioration of water quality and eutrophication of the receiving water bodies.

1. Carbohydrate + protein + fat (organic waste) ---> $CH_4 + H_2O + NH^+_4 + NO^-_2$+ organic P (with low dissolved oxygen supply)

2. Carbohydrate + protein + fat (organic waste) ---> $CO_2 + H_2O + NO^-_3 + PO^{-3}_4$ (with high DO_2 supply) (Connell 1993).

Suspended solids increase the water turbidity in rivers and prevents light penetration necessary for primary production (Connell 1993). High or prolonged solid loads can cause physical, mechanical and chemical problems to the environment (Cripps and Kelly 1996). It can smother the benthos, reduce water opacity and may clog the respiratory apparatus of fish. The nutrients in solid waste may sediment out and under certain conditions release the dissolved fractions and thereby increase the

nutrient pool of the environment (Kelly 1992; Ackefors & Enell 1994). It is the dissolved fractions which have an immediate effects on the water quality of the environment (NCC 1990; Kelly 1993). Indeed, the water quality of freshwater bodies can be assessed based on the levels of suspended solids (Table 1.3.4).

Table 1.3.4. Criteria for assessing water quality of freshwater based on suspended solids (OCE 1988).

Excellent	<30 mg/l
Good	<35 mg/l
Moderate	<40 mg/l
Poor	<60 mg/l
Degraded	>60 mg/l

Changes in the water chemistry and an increase in ammoniacal nitrogen and phosphorus concentrations immediately downstream of the fish farms were reported (Alabaster 1982; Beveridge 1984; Munro *et al.* 1985; Phillips 1985; Phillips and Beveridge 1986). Solid waste material enriches the sedimentary nutrients immediately beneath the cages (Kelly 1993). Apart from the above, the cage aquaculture could elevate the carbon, nitrogen and phosphorus levels of water bodies and may encourage high nutrient tolerant algal growth (eg. Cynobacteria) (Eley *et al.* 1972; Kilambi *et al.* 1976; Hays 1980). This will ultimately affect the water quality and autotrophic food web. However, the impact of solid waste in ponds culture system could be minimal as static ponds has a tremendous capacity to assimilate organic waste and nutrients and consequently waste load from ponds are low (Edwards 1993; Boyd & Tucker 1995). For example, the nutrients released via solid wastes in ponds could be used up by phytoplankton and may return to the atmosphere as carbon dioxide, methane, ammonia, and nitrogen or adsorbed in the pond soil, or lost as pond effluents (Boyd and Tucker 1995) (Figure 1.3.1).

Reduction of solid waste

Selection of diets with digestible ingredients and avoiding excess nutrients could be a simple means of reducing solid waste from aquaculture (Cho 1993). In this respect high nutrient-dense diets (HND) which are highly digestible and have a well balanced protein : energy ratios in diets would be most desirable for a sustainable aquaculture (Cho 1993).

1.4 NITROGEN POLLUTION FROM AQUACULTURE

Proteins are the main source of nitrogen and essential amino acids, and the most expensive energy source (Pillay 1990). Dietary protein supplied in aquaculture is used to fulfil the three basic needs : growth, maintenance and repletion of depleted tissues (Cowey and Sargent 1972; Jauncey 1982). To maximize the nutrient utilization and minimize the solid and soluble waste load, it is essential to provide the optimum level of protein to the cultured fish (Cho 1993). Generally, nutrients absorbed in excess of requirements may be excreted as ammonia and urea which is derived from the catabolic breakdown of protein (Beveridge and Phillips 1993). When food wastage is high and the nitrogen retention and assimilation are poor, a majority of nitrogen added to the culture system may ultimately pollute the environment (Handy and Poxton 1993). The aim of aquaculture should therefore be to provide sufficient nitrogen through feed to promote a good growth of stock. Feeding in excess of requirements is uneconomical and may deteriorate the water quality and therefore may imbalance the ecology of the receiving waters (Poxton 1992).

There are three main ways that nitrogen pollution from aquaculture can occur, such as, overfeeding of fish /or feeding of fish at a time when they are not growing, feeding an unstable and highly soluble diet and feeding a diet of poor absorption and nitrogen retention or a combination of the above three factors (Handy and Poxton

1993). Ingested nitrogen may cause nitrogen pollution to the receiving waters from direct gill excretion and through faecal loss (Figure 1.4.1.). The nitrogen pollution from ingested food can be less, if adequate nitrogen is absorbed by the gut and adequate absorbed nitrogen is retained by the tissues (Handy and Poxton 1993).

Gill excretion

The main excretory product in teleost fish is total ammonia nitrogen (TAN), which is formed in the liver and excreted across the gills (Smith 1929; Randall and Wright 1987; Sayer and Davenport 1987; Ramnarine *et al.* 1987; Kelly *et al.* 1994). The gill excretion represents about 80%-90% of nitrogen losses from fish (Wood 1958; Sayer and Davenport 1987) and the faecal nitrogen loss accounts for 10-20% (Fivlstad *et al.* 1990). The other route of nitrogen loss is via uneaten feed or dust (Beveridge *et al.* 1991; Bergheim and Asgard 1996; Kibria *et al.* 1997b) (Figure 1.4.1.).

Hourly patterns of ammonia excretion

The post-prandial excretion rate has been reported to be initially high before returning slowly to the pre-feeding level (Brett and Zala 1975; Kaushik 1980; Kaushik and Oliva-Teles 1985; Ramnarine *et al.* 1987). When fish were offered two meals a day two peaks of excretion were observed in rainbow trout (*Oncorhynchus myskiss*) (Kaushik 1981) and in cod *Gadus morhus* (Ramnarine *et al.* 1987). Rychly (1980) found that post-prandial nitrogen excretion was higher in the first meal than the second meal. Kaushik and Cowey (1991) reported that the amplitude and time of appearance of peaks can vary depending upon fish size, amount of nitrogen intake and water temperature. In general, the peak of ammonia excretion was found to be four hours after feeding sockey salmon and American eel (Brett and Zala 1975; Gallagher and Matthews 1987; Jarboe 1995). In the catfish, the highest TAN production occurred at 3.5 hour and 2.5 hour after the first and second meals respectively (Jarobe 1995).

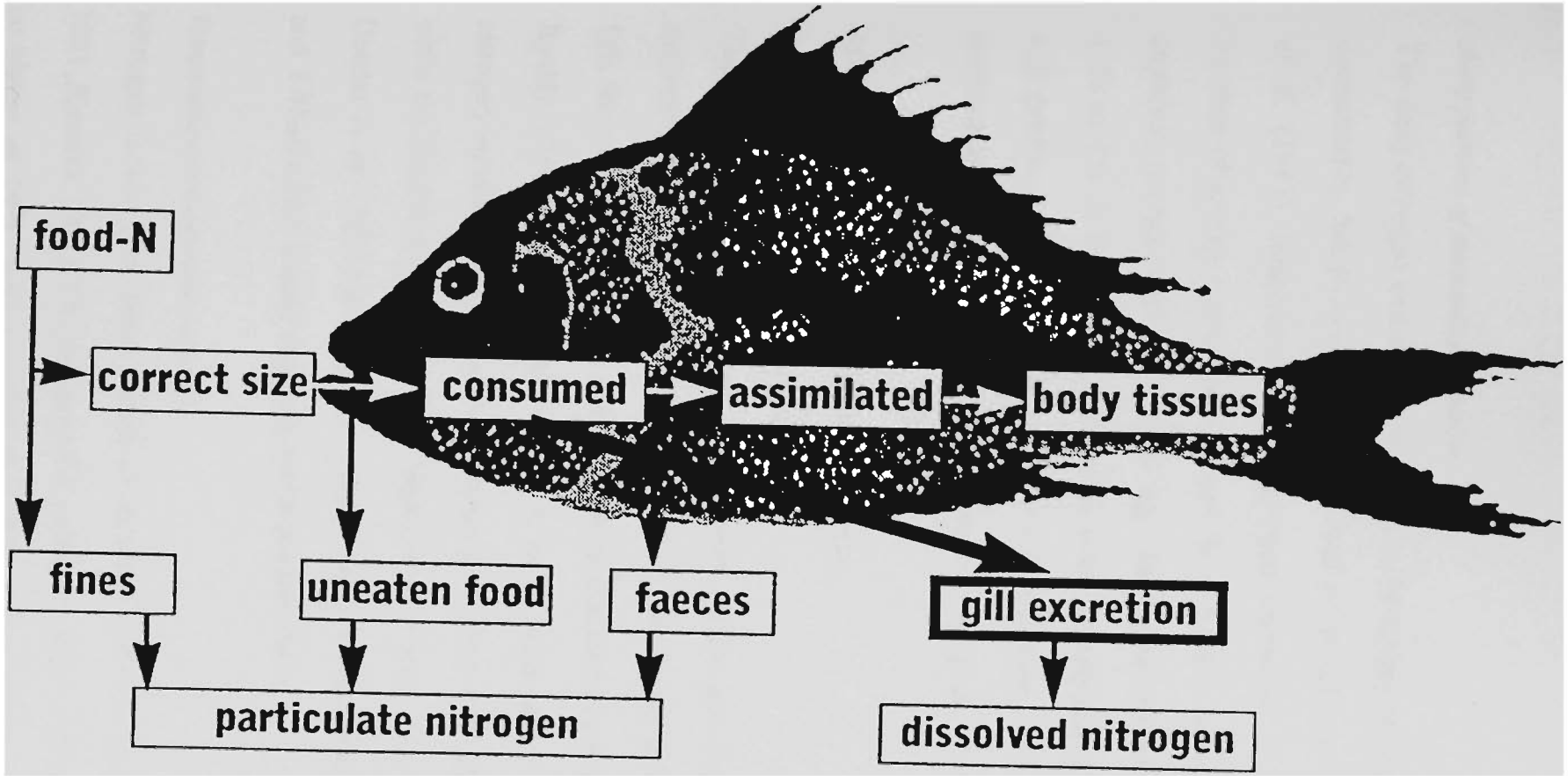

Figure 1.4.1. General flow chart of origin of nitrogen in aquaculture from feeding.

Daily patterns of ammonia production

The daily nitrogen excretion was found to be mainly related to the amount of nitrogen consumed by fish (Kaushik 1980) and feed composition (Johnsen *et al.* 1993). Johnsen *et al.* (1993) demonstrated that nitrogen excretion in fish were reduced with an increase of energy content and a decrease of protein content in the diet. This reduced ammonia excretion from 35 to 22 kg/t fish produced and faecal nitrogen loss from 11% to 8%. It has been reported that when protein supply is in excess or the amino acid profile does not correspond to the requirement of the fish, the excess nitrogen supplied through feed will simply be excreted (Bergheim and Asgard 1996).

Main factors related to ammonia excretion

Ingested nitrogen and ammonia production

The quality and quantity of the nitrogen intake are the most important factors determining the ammonia production. In fact, the higher the nitrogen ingestion by the fish, the more the ammonia and urea that is excreted (Savitz 1971; Savitz *et al.* 1977; Rychly 1980; Dosdat *et al.* 1995). A linear relationship was found between the nitrogen ingestion and ammonia excretion in rainbow trout (Nose 1971; Rychly 1980), while the relationship was linear or logarithmic in both trout and carp (Kaushik 1980). Dosdat *et al.* (1995) established a strong relationship between the ingested nitrogen and TAN for both hourly and daily ammonia excretion.

Temperature and ammonia excretion

Nitrogen losses have been shown to increase with increasing temperature (Jobling 1981; Kaushik 1981). The postprandial excretion rate in rainbow trout was reported to be higher at 18^{o}C than at 10^{o}C (Kaushik 1981). Similarly, the Japanese flounder released more ammonia at 25^{o}C than at lower temperatures in the order of 25^{o}C>20^{o}C>16^{o}C (Kikuchi *et al.* 1995). The rapid increase of temperature may stress the fish and therefore influence nitrogen intake, apparent digestibility of nitrogen compounds, and the excretion of nitrogen through branchial and urinary pathways. A

rise in temperature causes an increase in plasma ammonia concentration which in turn would increase the level of ammonia loss through renal/branchial paths (Pequin and Serfaty 1968). As a result both hourly and daily patterns of nitrogen excretion can be affected. (Kikuchi *et al.* 1995).

Diet and ammonia excretion

Dabrowski and Kaushik (1984) reported that the type of feed may affect the timing of excretion, for example, fish fed on live *Artemia* showed maximum excretion after two hours whereas for fish fed on a dry diet the peak of excretion was delayed for up to 4-6 hours. A lengthening of the excretion period was also observed with a diet containing higher protein levels (Ballestrazzi *et al.* 1994). The hourly excretion rate in the sea bass increased with increasing protein level and was not significantly different among the herring meal diets containing 44%, 49% and 54% protein ($P>0.05$) (Ballestrazzi *et al.* 1994). The nitrogen excretion in fish is related to the amount of nitrogen ingested by fish (Kaushik 1980) and higher at higher feeding rates (Dosdat *et al.* 1995) and higher protein level (Kaushik and Oliva-Teles 1985). This may demonstrate that the nitrogen excretion profile is a function of the ingested nitrogen.

Other factors affecting in ammonia excretion

Other factors that may influence the nitrogen excretion are : rearing conditions (Fromm and Gillette 1968; Olson and Fromm 1971; Sukumaran and Kutty 1977; Wilkie and Wood 1991), body weight (Jobling 1981; Kikuchi *et al.* 1995), species (Davenport *et al.* 1990; Gershanovitch and Pototskij 1992), intra-species families (Gallagher *et al.* 1984; Kaushik *et al.* 1984; Ming 1985), and physiological status (Wiggs *et al.* 1989). Additional factors include temperature (Speece 1973; Paulson 1980; Jobling 1981; Clark *et al.* 1985; Beveridge and Phillips 1993; Kelly *et al.* 1994), pH, ambient dissolved oxygen concentration, life stage, protein level and protein utilization efficiency, exercise or activity state, group-effect, species, time of day (Maetz 1972; Brett and Zala 1975; Rychly and Marina 1977; Leid and Bratton 1984;

Preez *et al.* 1986; Davenport *et al.* 1990; Paul *et al.* 1990; Cai and Summerfelt 1992), feeding rate, and stocking density (Rowland 1996). In summary, nitrogen excretion increases with increasing protein level (Ballestrazzi *et al.* 1994; Lanari *et al.* 1995a) and temperature (Jobling 1981) and decreases with high energy or extruded diets (Lanari *et al.* 1995b) or when protein:energy ratio in diet is optimum (Gallagher and Matthews 1987). A single factor or a combination of the above factors may make a result in a change in ammonia excretion.

Faecal loss of nitrogen

Iwata (1970) found a direct relationship between the nitrogen intake and faecal nitrogen release in crucian carp, *Carassius auratus*. Fish fed on a higher nitrogenous diet also had faeces of higher nitrogen content (Ogino *et al.* 1973a). Kaushik (1981) reported that the faecal nitrogen loss was higher at a higher temperature. Similarly, when flounder, (*Paralichthys olivaceus*) were reared at 16^{o}C, 20^{o}C and 25^{o}C, the faecal nitrogen loss was proportionately higher at higher temperatures (Kikuchi *et al.* 1995). The reported faecal nitrogen lost from fish is in the range of 5.5% to 15.7% of nitrogen intake (Kaushik 1980; Porter *et al.* 1987; Beveridge and Phillips 1993). In general, the faecal nitrogen loss could be one-third of nitrogen excreted by fish (Porter *et al.* 1987). The nitrogen assimilation efficiency is inversely related to the faecal loss of nitrogen, a lower nitrogen assimilation efficiency could elevate the faecal nitrogen output many fold (Table 1.4.1).

Table 1.4.1. The relationship between the assimilation efficiency and faecal nitrogen excretion.

Common name	N_2 assimilation efficiency (%)	Faecal excretion (mg N kg^{-1} h^{-1})	Reference
Atlantic salmon	68	17.49	Santos & Jobling (1991)
Atlantic salmon	95	02.73	Santos & Jobling (1991)
Atlantic salmon	99	00.54	Santos & Jobling (1991)
Turbot	68	04.62	Bromley (1987)
Turbot	95	00.72	Bromley (1987)
Turbot	99	00.14	Bromley (1987)

Nitrogen retention in fish

The amount of nitrogen retained in the carcass can be a useful means of evaluating a diet and the nitrogen retention efficiency of a fish (Brown *et al.* 1993). Three factors appears to play a significant role in nitrogen retention in fish. These are - diet, temperature and body weight of fish. The simple, most accurate and economical way to quantify the nitrogen retention in fish is to analyse the carcass using known biological and nutritional procedures (Cho 1993) (equation 1) and through a nitrogen balance equation (Rychly 1980; Hall *et al.* 1992) (equation 2). The nitrogen balance is determined from the proportion of nitrogen absorbed into the tissues (nitrogen retention) from the food and the metabolic output (nitrogen loss).

Equation 1 : N retention = [{final body nitrogen - initial body nitrogen} x 100 / total dietary nitrogen supplied] (Brown *et al.* 1993).

Equation 2 : N balance = N consumed - N retained - N excreted - Faecal N (Hall *et al.* 1992).

The average nitrogen retention efficiency in fish carcass is 35.4%. It is evident therefore that more than 60% of nitrogen fed to fish is lost through gill excretion or faeces (Table 1.4.2).

Table 1.4.2. Nitrogen balance (N retention and N losses) in fish fed on artificial diets.

N GAIN	[----------N LOSS----------]					
N retained	Dissolved N	Faecal N	Uneaten N	Diet	Species	Reference
27.8%	56.48%	15.74%	-	ns[1]	-	Ackefors & Enell (1994)
49.1%	37.3%	13.5%		HED[2]	-	Johnsen *et al.* (1993)
36.0%	-	-	-	Pellet	Rainbow trout	Lanari *et al.* (1995a)
48%	-	-	-	Extruded	Rainbow trout	Lanari *et al.* (1995a)
20.8%	48.74%	30.50%	-	Dry diet	Rainbow trout	Phillips & Beveridge (1986)
-	78%	22%	-	ns	Salmon	Enell (1995)
36.8%	28.1%	35.0%	-	Dry diet	Rainbow trout (15.7°C)	Oliva-Teles & Rodrigues (1993)
45.0%	32.1%	22.9%	-	Dry diet	Rainbow trout (21.5°C)	Oliva-Teles & Rodrigues (1993)
24.7%	60.3%	15%	-	ns	-	Hakanson *et al.* (1988)
36.0%	54.3%	-	-	Dry diet	Rainbow trout	Gomes *et al.* (1993)
23.44%	42.20%	14.40%	20.0%	ns	Tilapia	Beveridge & Phillips (1993)
28%	48%	23%	-	Dry diet	Rainbow trout	Hall *et al.* (1992)
43.1%	52.0%	5.6%	-	Dry diet	Silver perch (25°C)	(present study) (chapter 6)
29.4%	61%	9.86	-	Dry diet	Silver perch (30°C)	(present study) (chapter 6)

[1]ns = not stated; [2]HED = High energy diet.

Factors affecting nitrogen retention efficiency

Effect of diet on nitrogen retention

The diet and its composition is the most important factor determining the efficiency nitrogen retention in fish. Rainbow trout fed on an extruded diets retained 46-49% of dietary nitrogen compared to 35-36.5% retained when fed on normal diets of the same protein value (Lanari *et al.* 1995a). Diets containing high levels of raw starch or materials with low digestibility generally have a low nitrogen assimilation efficiency (Jobling 1986). Lanari *et al.* (1995a) demonstrated that the diet which resulted in better relative growth rate and food conversion ratio also resulted in better nitrogen retention in rainbow trout.

Effect of temperature on nitrogen retention

Rainbow trout utilized diet more efficiently at 21^{o}C than at 15.7^{o}C suggesting that at the higher temperature there was better digestibility and nitrogen retention (Kaushik and Oliva-Teles 1985). An improved diet digestibility at higher temperature was also reported by Choubert *et al.* (1982). However, Cho & Slinger (1979) and Luquet & Fauconneau (1979) did not find any affect of temperature on diet digestibility.

Effect of fish size and growth rate on nitrogen retention

Nitrogen retention can vary with the size of fish (Pandian 1967; Gerking 1971) or the growth rate (Brown *et al.* 1987). Adult fish with low growth rates may retain 15-20% of the absorbed nitrogen, whereas the rapidly growing fish can retain 40% of nitrogen supplied in diet (reviewed by Handy and Poxton 1993).

Impacts of nitrogen loading on the environment

Nitrogen is one of the primary limiting nutrients in freshwater (Kelly *et al.* 1994) and the marine environment (Ryther and Dunstan 1971; Makinen 1991; Kelly *et al.* 1994; Brodle 1995). Nitrogen from fish farm effluents may enter into an environment in

different nitrogenous forms (Table 1.4.3.) such as ammonia, total organic nitrogen, total nitrogen, nitrite, and nitrate (Erskine and Saynor 1995).

Table 1.4.3. Specific nitrogen loadings of feed-derived compounds

	TN	TAN	TON	Reference
Scotland (g/kg/d)	0.17-7.2	0.07-5.5	0-0.46	Hennessy *et al.* (1991)
Norway (g/kg/d)	0.17-0.81	0.05-0.09	-	Bergheim *et al.* (1991b)
Norway (g/kg/d)	0.29-0.49	0.15-0.18	-	Bergheim *et al.* (1991b)
Norway (mg/l)	0.5	-	-	Bergheim *et al.* (1991a)
Norway (mg/l)	0.43-0.70	-	-	Bergheim *et al.* (1993)
Sweden (mg/l)	0.70	-	-	Cripps (1995)
N. Ireland (mg/l)	0.531	-	-	Foy and Rosell (1991a)
U.K.(mg/l)	0.5-5.0	-	-	Muir (1982)
Denmark (mg/l)	3-20	-	-	Warren-Hansen (1982)

TN = total nitrogen; TAN = total ammonia nitrogen; TON =total organic nitrogen.

Studies conducted in temperate regions have revealed that an abundance of nitrogen in aquatic system caused a toxic effect upon resident biota (Carr and Goulder 1990a, 1990b; Foy and Rosell 1991a,1991b), stimulated primary production (Makinen 1991) and resulted in eutrophication of coastal waters (Ryther and Dunstan 1971). An abundance of nitrogen increased seagrasses, phytoplankton, benthic algae, epiphytic algal overgrowth, and encouraged the growth of toxic algal species, such as, *Chrysochromulina polyplepis* and *Phaeocystis pouchetti* (Cambridge *et al.* 1986) in marine waters which caused an offensive odour and resulted in the occurrence of slimy water (Lancelot *et al.* 1987). Wu *et al.* (1994) reported a decrease in dissolved oxygen and an increase in ammonia level of marine environment received fish farm effluents in Hong Kong. Nutrient discharged from cages or land-based farms changed the benthic environment (Merican & Phillips 1985) resulting in the losses of benthic invertebrates (Brown *et al.* 1987). Much of the nitrogen lost was utilized by either phytoplankton (Krom *et al.* 1989) or deposited in the culture system into sediment. The percentage of nitrogen deposited into sediments is reported to be around 30.4% (Phillips & Beveridge 1986), and 15.7% (Ackefors & Enell 1990). The nitrogen loading from aquaculture is in the range of 68-211 kg/t fish produced (Table 1.4.4.).

Table 1.4.4. Nitrogen loadings (total nitrogen) per tonne of fish production per year

Country	Species	Culture system	Diets	TN/t	Reference
Nordic country	Salmon	Cages		78	Enell (1995)
Norway	-	-	-	95-102	Hall *et al.* (1992)
Denmark	Rainbow trout	Ponds	Dry feed	75	Warren-Hansen (1982)
Poland	Rainbow trout	Cages	Moist diet	90	Penczak *et al.* (1982)
Sweden	Rainbow trout	Cages	Dry feed	81	Enell & Lof (1983)
Europe	Marine fish	-		190	Handy & Poxton (1993)
Europe	-	-	-	108.4±47.3	IOA (1990)
U.K	Rainbow trout	-	Dry diet	103.8	Phillips & Beveridge (1986)
UK	Salmonid	-	-	123	HRPB (1987)
Scotland	Rainbow trout	Cage			Merican & Phillips (1985)
Scotland	Rainbow trout	-	-	83-104	NCC (1990)
Scotland	Rainbow trout	Cages	Dry feed	99	Phillips (1985)
Japan	Yellowtail Cages	-		68	Watanabe (1991)
Japan	Yellowtail	Cages		109	Watanabe (1991)
Japan	Bream	Cages		211	Watanabe (1991)
Thailand	Marine shrimp	-	-	102.3	Briggs & Funge-Smith (1994)

An intensive system can generate 7-31 times more nitrogen load than a semi-intensive system (Edwards 1993). In the semi-intensive system, a significant part of unutilized nutrient is lost in the pond sediment (Pullin 1989; Bergheim and Asgard 1996) (Table 1.4.5.).

Table 1.4.5 Fate of nitrogen in semi-intensive and intensive aquaculture system (Edwards 1993)

	% N removed by fish	% N released to environment	% N accumulated in sediments
Semi-intensive	11-15	2-6	83
Intensive	21-53	47-49	0

Reduction of nitrogen loading from aquaculture

In order to reduce the nitrogen loading from aquaculture, a better food conversion ratio is essential. A reduction in nitrogen excretion in yellowtail and red sea bream was achieved as a result of better food conversion ratio (Watanabe 1991). To minimize the nutrient load to the environment, an FCR of 1.0-1.2 is highly desirable (Cowey and Cho 1991). It has been suggested that improved feed quality and feeding techniques are the primary factors for the reduction of nitrogen pollution from aquaculture (Eikerbrokk *et al.* 1991; Jensen 1991). High energy diets are known to increase the utilization of nutrients and as a consequence reduce the solid waste and nutrient load in the water (Johnsen and Wandsvik 1991). Nevertheless, extrusion is generally believed

to be a valid means of reducing nitrogen and phosphorus discharge from aquaculture (Table 1.4.6.) and this favourably improved growth rate, feed utilization and gross protein retention in rainbow trout (Lanari *et al.* 1995a). Furthermore, diets with optimum carbohydrate and protein levels may improve the nitrogen retention in fish (Alsted and Jokumsen 1989; Johnsen and Wandsvik 1991; Johnsen *et al.* 1993; Hillestad and Johnsen 1994; Bergheim and Asgard 1996).

Table 1.4.6. Effect of fat content on FCR and nitrogen loadings in rainbow trout

Diet	Av. fat content in diet (%)	Av. FCR obtained	Av. nitrogen load (kg/t)	Source
Normal pellet	20.45	1.25	47.8	Lanari *et al.* (1995a)
Extruded diet	28.0	0.89	26.6	Lanari *et al.* (1995a)

The type of diet fed to fish could determine the amount of nitrogen load expected, for example, Japanese yellowtail fed on a moist-pellet reduced nutrient load by 50% compared to a diet of raw fish. When fish were fed on dry pellets the nitrogen excretion was reduced by another 25% (Watanabe 1991). The dry pellet used in Sweden resulted in good food conversion and low nutrient losses in comparison to moist food types (Persson 1988).

Nutrient load can also be minimized by controlling the feeding regimes. At restricted feeding (*ad lib*), nutrient load could be lower since restricted feeding gives a higher nitrogen assimilation efficiency to fish (Usher *et al.* 1990). The lowering of nitrogen content in feed may be another alternative for reducing the nitrogen load into the effluent water (Handy and Poxton 1993; Lanari *et al.* 1995b.). Furthermore, alternate feeding of high and low protein diets may reduce nitrogen load with substantial savings of feed cost (De Silva *et al.* 1993). In Nordic countries nitrogen content in feed has been decreased from 7.8% (1974) to 6.8% (1994) by legislation which resulted in a reduction of 58% of nitrogen pollution from aquaculture with a consequential improvement of food coefficient (Enell 1995) (Figure 1.4.2.) .

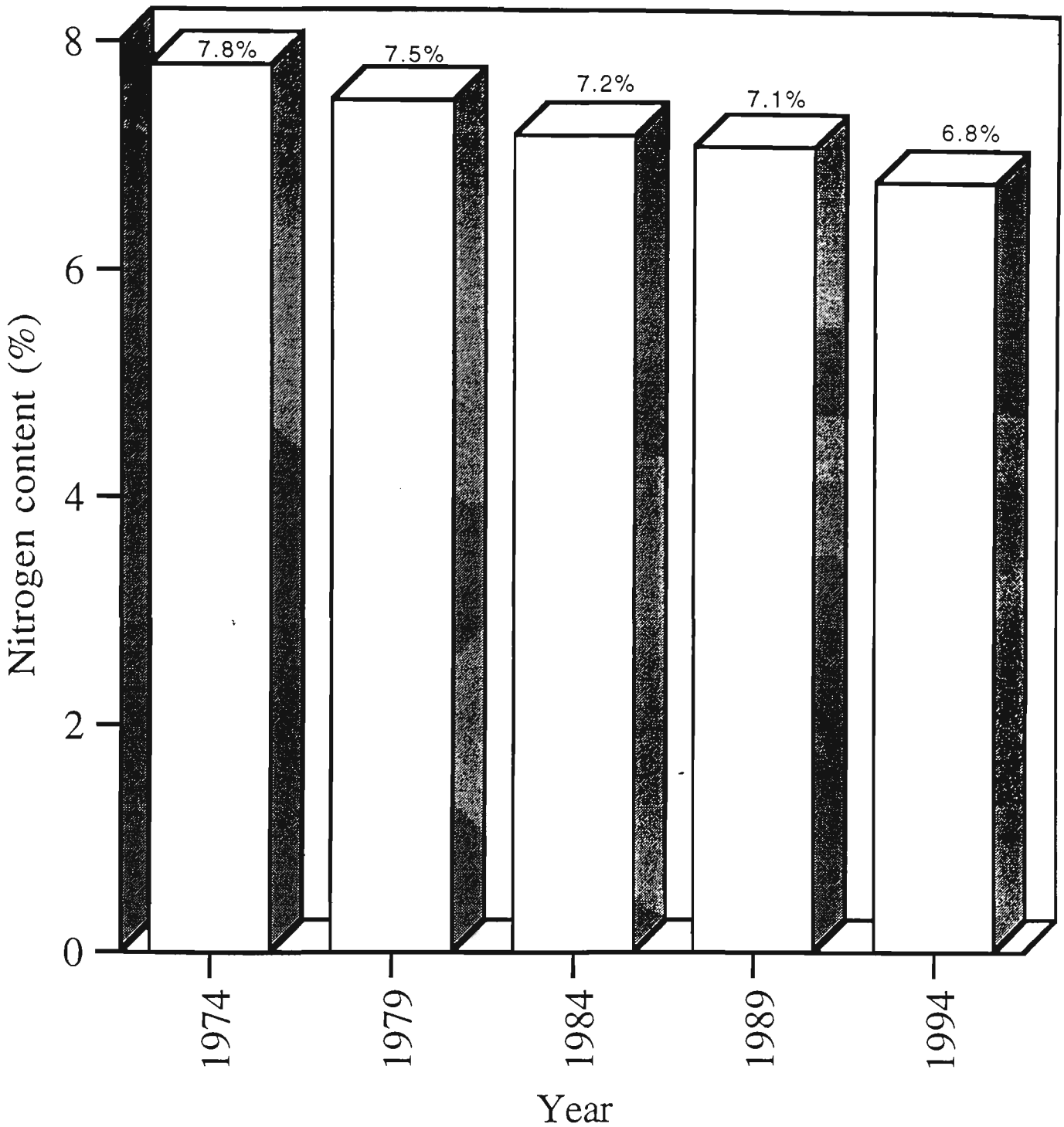

Figure 1.4.2. Reduction of Nitrogen content in fish feed by Nordic countries during 1974 to 1994. This has resulted in a reduction of nitrogen pollution by 58% with an improvement of feed coefficient (data from Enell 1995).

The main food wastage in aquaculture appears to be due to a mismatch of farming practice and not giving due consideration to the feeding behaviour and nutritional physiology of the species being cultured. Many fish species show feeding periodicity (Hall 1987; Sagar and Glova 1988), therefore feeding time should be matched to the time of maximal appetite. Some species feed exclusively during the day (haddock), some mostly at dusk (dab) (Hall 1987), and some feed continuously provided the stomach remains partly empty (Smith *et al.* 1989). Since optimum growth can be achieved below the maximum ration level (Brett and Groves 1979), the practice of avoiding the satiation ration can be applied to reduce the nitrogen pollution.

The uneaten food fraction is the one of the path of nitrogen wastage from aquaculture contributing 1-30% of food wastage (Beveridge *et al.* 1991). Food dispersion by automatic feeders may result in wastes up to 40.5% of food offered because the food is localised, resulting in a few dominant aggressive fish overfeeding close to the dispenser (Thorpe *et al.* 1990). Hand feeding minimizes wastage in salmonid culture and therefore may reduce the nutrient load (Beveridge *et al.* 1991). Polyculturing of fish rather than monoculture may save food wastage and reduce nitrogen pollution as leftover food from feeding a single fish could be consumed by bottom or column feeding fish. Avoiding feeding immediately after a stressful treatments, such as handling, or the administration of therapeutants could further reduce nutrient loads (Poxton 1992).

Some commercial feeds have a tendency to dissolve rapidly in water resulting in the instantaneous release of nutrients from solid waste. Pelleted feeds are relatively stable in water and therefore may reduce nutrient release to the environment (Hilton *et al.* 1981; Jayaram and Shetty 1981). Feed can be manufactured according to the feeding behaviour of fish, a floating pellet (extruded pellets) can be offered to a surface feeder and a sinking pellets (compressed pellets) to a column or bottom feeder (Hardy 1989). The disintegration of pellets can be delayed by coating or encapsulating the

food in agar, alginate's, or synthetic materials. (Hardy 1989). Use of less dust feed, higher feeding frequency, and feeding according to appetite would reduces food conversion by at least 0.4 units and consequently reduces nutrient loss (Persson 1988).

Quantification of nitrogen excretion in fish is of importance as it may indicate the amount of nitrogen that is released into the environment (Handy and Poxton 1993). Moreover, data on nitrogenous waste production helps to improve the dietary protein utilization of fish (Kaushik *et al.* 1984). Metabolic loss such as ammonia excretion is a valuable means of evaluating a diet and its ingredients (Brett and groves 1979). Research on the improvement of nitrogen utilization and simultaneous reduction of nitrogen loss is highly desirable for the development of sustainable aquaculture (Cho 1993). From this perspective, it is essential to identify the optimum protein requirements of a species that will assure a higher growth and a lower nitrogen waste to the environment. It is, however, clear that nitrogen load from aquaculture is minimal (4-13%) compared to agriculture and other point source (Table 1.4.7).

Table 1.4.7. Pollution (Total nitrogen) loading rates from various sources in different countries compared to aquaculture

Polluter	Country	Total N	TN/TP	Reference
Agriculture (t/d)	Japan	5.59	18.03	Watanabe (1991)
Domestic wastes (t/d)	Japan	3.56	10.47	Watanabe (1991)
Industrial effluents (t/d)	Japan	0.60	1.82	Watanabe (1991)
Aquaculture (t/d)	Japan	1.49	5.51	Watanabe (1991)
Aquaculture (as % of the total)		**13%**		**Watanabe (1991)**
Forestry (t/y)	Finland	505.5	6.6	FNBW (1981)
Industry (t/y)	Finland	3340	32.6	FNBW (1981)
Aquaculture (t/y)	Finland	240	4.0	FNBW (1981)
Aquaculture (as % of the total)		**4%**		**FNBW (1981)**

t/d = tonne per day; t/y = tonne per year.

1.5 PHOSPHORUS POLLUTION FROM AQUACULTURE

Phosphorus is an essential element for living organisms and exists in streams and other water ways as dissolved and particulate forms (Welch and Lindell 1980). Phosphorous is required for optimum growth, feed conversion efficiency, bone development and maintenance of acid-base regulation, lipid and carbohydrate metabolism of fish (Lovell 1978; Cowey and Sargent 1979; Lall 1989; Ketola and Richmond 1994). Growth and food conversion has been positively correlated with the dietary levels of phosphorus in fish (Shim and Ho 1989) and fish receiving a low phosphorus diet showed the poorest growth, gain in weight (Shim and Ho 1989) and depressed appetites (Lovell 1978). Dietary phosphorus is the main source of this element for fish (Nose and Arai 1978) since phosphorus concentration in natural waters is low (Boyd 1971; Lall 1991) and the rate of absorption from water is also low (Phillips *et al.* 1956). Fish must effectively absorb, mobilize and conserve phosphorus in order to meet the requirements for growth and metabolism, (Lall 1991).

The presence of high concentrations of phosphate in water is often an indication of pollution as it may accelerate plant growth (Beveridge 1987), and disrupt the aquatic ecosystem thereby benefiting certain species and altering species diversity in affected areas (Anon 1987, OCE 1988). Eutrophication of water bodies is often correlated with the phosphorus loading into the environment (Kaushik 1992) and aquaculture has been identified as one of the sources of phosphorus pollution (EPA 1995). Details of the impacts of eutrophication is given in Bernhardt (1981). Studies in Europe and Northern America have revealed a phosphorus surplus in most commercial feed above actual requirements (Tacon and De Silva 1983; Beveridge 1987) or is supplied in a form which is unavailable to the fish (Beveridge 1987). Discharge of phosphorus from fish farms and hatchery effluents have caused phosphorus pollution in Nordic countries, North America and Europe (Bernhardt 1981; Albaster 1982; Beveridge 1984; Enell 1987; Folke and Kautsky 1989; Ketola 1990; Bratten 1991; Foy and Rosell 1991b; Lall 1991).

Digestibility and bioavailability of phosphorus

The digestibility and bioavailability of phosphorus to fish differs among feed ingredients and inorganic phosphorus supplements (Lall 1991) and is influenced by chemical form, digestibility of diet, particle size, interaction with other nutrients, feed processing and water chemistry (Lall 1991). The more soluble the salt, the higher the availability of phosphorus. Salmonids utilize phosphorus from fishmeal more effectively than do carp or tilapias (Ogino *et al.* 1979; Watanabe *et al.* 1980). This is probably linked to the limited secretion of gastric juices by warmwater species (Yone and Toshima 1979; Ogino *et al.* 1979). The phosphorus requirement for fish is in the range of 0.3-0.8% (Table 1.5.1).

Table 1.5.1. Reported phosphorus requirements of various species.

Species	% Phosphorus in diet	Reference
Rainbow trout	0.7-0.8	Ogino & Takeda (1978)
Channel catfish	0.8-1.0	Ogino & Takeda (1978)
Channel catfish	0.45	Lovell (1978)
Carp	0.5-0.6	Watanabe *et al.* (1980)

Path of phosphorus loss

Feed is the main source of phosphorus loadings from aquaculture to the environment (Ketola 1990; Seymor and Bergheim 1991). There are a number of ways in which phosphorus can be lost during aquaculture operation, losses of phosphorus can occur through feed fines, uneaten food, faeces, dead fish and excretion (Forster and Goldstein 1969; Nakashima and Leggett 1980; Beveridge 1987) (see also Figure 1.5.1.). The main loading of phosphorus to the environment was reported to be via faecal pellets (Solberg and Bregnballe 1982; Enell 1987; Kristiansen and Hessen 1992; Pillay 1992; Kibria *et al.* 1997c accepted). It is reported that of the total phosphorus loss from aquaculture, 77% is in particulate form as faeces and 23% in dissolved form (Enell 1987; Ackefors and Enell 1990; Enell and Ackefors 1991; Cho 1993). The particulate form (faeces) settles on the bottom of the tank or accumulates in the sediment and is gradually released in a soluble form during anaerobic and other related

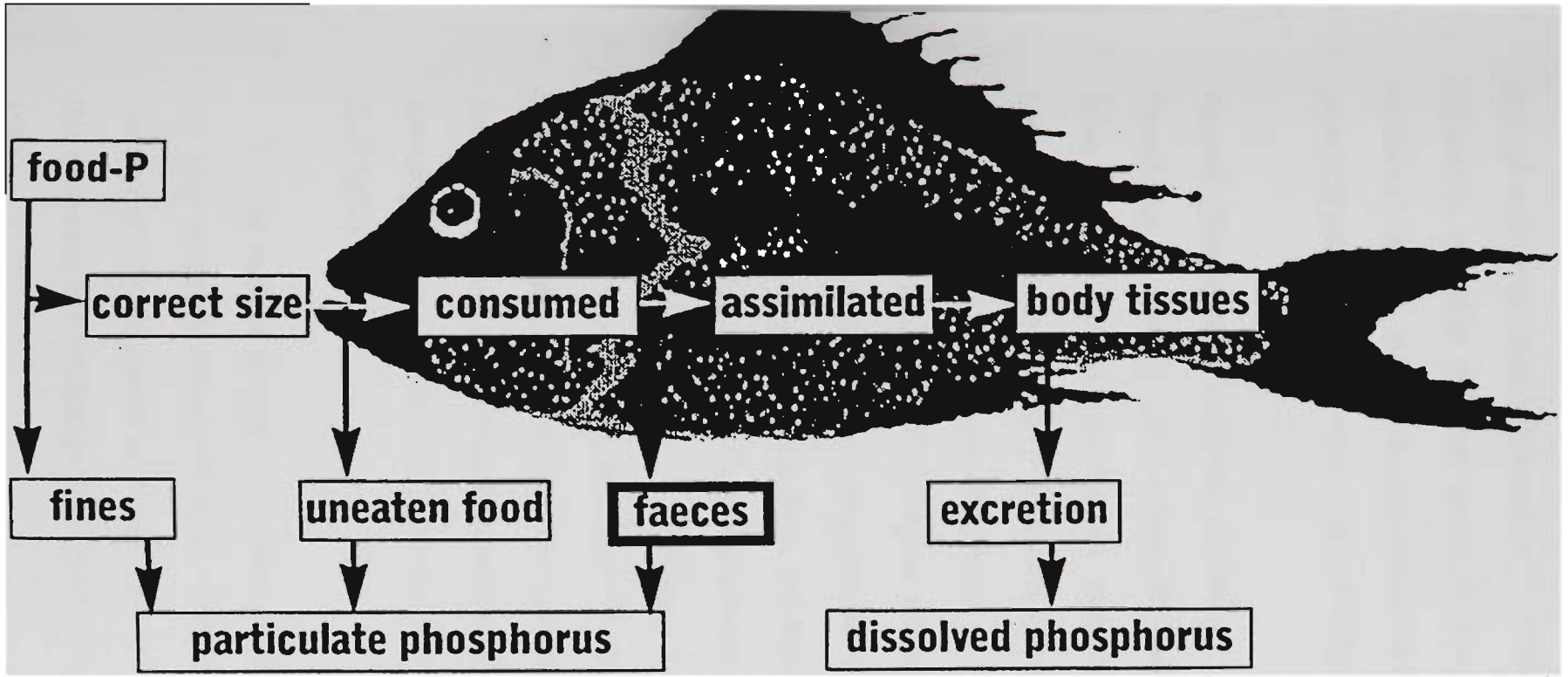

Figure 1.5.1. General flow chart of origin of phosphorus in aquaculture from feeding.

biological processes (Lall 1991) (see also Figure 1.3.1) The soluble form is lost through urine in the form of phosphate (Lall 1991; Pillay 1992). The soluble fraction is referred to as either dissolved inorganic phosphorus or orthophosphate or soluble reactive phosphate. It is the dissolved fraction that is the most assessable form for the growth of plants (Bostrom *et al.* 1988a; 1988b).

Factors affecting the phosphorus excretion

Phosphorus excretion in fish is affected by both temperature and diet. For instance, in noble crayfish (*Astacus astacus*), the phosphorus excretion rate doubled when the temperature increased from 15^{o}C to 20^{o}C (Kristiansen and Hessen 1992). An elevated excretion of phosphorus was also found at higher temperatures in bluegill sunfish (Savitz 1971). A comparatively higher phosphorus excretion was reported in noble crayfish fed on a diet rich in protein and fat (Kristiansen and Hessen 1992). Seabass when fed on corn gluten based diets, had significantly lower phosphorus excretion in comparison to herring meal diets (Ballestrazzi *et al.* 1994) and this may be related to the lower phosphorus content in corn gluten. Lall (1991) reported that both the quality and quantity of food fed may determine the amount of phosphate excreted by fish. The dissolved phosphorus excreted by fish is one of the end products of carbohydrate, lipid, nucleoprotein and phospho protein metabolism (Cho 1993). The form of phosphorus consumed by fish will affect the amount of soluble and particulate phosphorus excreted (Lall 1991).

The peak of phosphate excretion is related to feeding and activity periods and can vary cyclically (Hennessy *et al.* 1996; Kibria *et al.* 1997c accepted). A rapid increase in orthophosphate excretion was recorded soon after the feeding of trout, and this decreased and reached prefeeding levels within six hours (Solberg and Bregnballe 1982).

The food conversion ratio (FCR) can play a significant role in determining the level of phosphorus pollution expected since an increase in FCR value from 1.0 to 1.5 may increase pollution load to about 86% for total phosphorus (Storebakken and Austreng 1987a and 1987b). Therefore, improvement of FCR is vital to reducing phosphorus pollution from aquaculture. In addition, digestibility of feed, nutrient content of diet, the protein : energy ratio in the feed, and the feeding technique may influence the feed coefficient and phosphorus load from aquaculture (Enell 1995).

Phosphorus retention

Phosphorus retention can be affected by the source of phosphorus into the diet. For example, monobasic calcium phosphate and monobasic sodium phosphate were found to be highly digestible and resulted in the highest phosphorus retention in channel catfish while retention from corn and wheat middlings was lowest (Table 1.5.2.).

Table1.5.2. Effect of the source of phosphorus on net phosphorus retention by channel catfish.

Materials	Net P retention	Reference
Monobasic calcium phosphate	94%	Lovell (1978)
Monobasic sodium phosphate	90%	Lovell (1978)
Dibasic calcium phosphate	65%	Lovell (1978)
Fishmeals	40%	Lovell (1978)
Corn and wheat middlings	25-28%	Lovell (1978)
Soybean meals	5--54%	Lovell (1978)

The supplementation of fish feed with phytase reduced phosphorus excretion and improved phosphorus retention in carp (Schafer *et al.* 1995). Phosphorus retention was found to be significantly higher in trout and sunbass with a decreased level of dietary phosphorus content (Brown *et al.* 1993; Ketola and Richmond 1994). Nose and Arai (1979) reported that phosphorus from nonskeletal sources is highly available to fish. Few data however, are available on phosphorus retention in fish, but these limited data show that phosphorus retention in fish ranged from 14%-87% (Table 1.5.3).

Table 1.5.3. Phosphorus retention in different fish species

Species	P retention (%)	P in feed /source	Reference
Salmon	14-22%	-	Ketola (1975)
Rainbow trout	64-68%	-	Ogino & Takeda (1978)
Rainbow trout	33%	-	Ketola and Harland (1993)
Sunshine bass	69-87%	0.34-0.54%	Brown *et al.* (1993)
Sunshine bass	40-50%	0.64-1.04%	Brown *et al.* (1993)
Carp	30.4-47.1%	0.73-1.16	Schafer *et al.* (1995)
Coho salmon	14-22%	-	Ketola *et al.* (1991)
Channel catfish	39-40%	Menhaden & anchovy meal	Lovell (1978)
Silver perch	24.5%-49.1%	1.16%-1.31%	Present study (chapter 7) & Kibria *et al.* (1997c accepted)

Phosphorus retention efficiency varies according to species, feeding habits, diet, growth rates and amount of phosphorus in the diet (Ketola *et al.* 1991; Ketola and Richmond 1994) and the amount of phosphorus retained by fish can be calculated by the following equation :

Equation for phosphorus retention :

P retention = [(final body phosphorus - initial body phosphorus) x 100 / total dietary phosphorus supplied] (Brown *et al.* 1993)

The Phosphorus discharge/phosphorus pollution load to the environment can be estimated from data on retention in fish carcass (Ketola 1990). It is equal to the difference between what is added by the feed and what is utilized for fish production (Hakanson *et al.* 1988; Foy and Rosell 1991a). In order to determine the phosphorus retention in the carcass, growth, survival and feed conversion data must be determined initially. Next, phosphorus concentration must be analyzed in feed, fish carcass, uneaten food and faeces. Finally the amount of soluble and particulate phosphorus discharged in water should be determined using the following phosphorus balance equation :

Equation for phosphorus balance :

P balance = Amount of P fed - P retained in carcass - P in faeces - P in uneaten feed (Ketola and Harland 1994).

Table 1.5.4. gives an account of phosphorus retention and path of phosphorus loss from aquaculture of different species. It shows that the main path of phosphorus

loss in aquaculture is via faeces, therefore it is essential that the digestibility of phosphorus is improved to reduce phosphorus pollution from aquaculture.

Table 1.5.4. Phosphorus retention and phosphorus losses in fish (as feed fed basis).

P Retention	[---Path of P loss---] Faecal P	Non-faecal P	Total P excretion	Diet	Reference
40.8%	39.2%	22.1%	-	HND[1]	Cho (1993)
-	77%	23%	-	-	Enell (1995)
34.7%	-	-	65.3%	Normal diet	Schafer *et al.* (1995)
45%	-	-	55%	Diet+phytase	Schafer *et al.* (1995)
36.4%	54.5%	9.1%	-	HED[2]	Johnsen *et al.* (1993)
14.6%	58.80	26.60		Dry diet	Phillips & Beveridge (1986)
30%	70%	-			Hakanson *et al.* (1988)
49.1%	33.6%	17.3%	-	Dry diet	Present study (chapter 7)

[1]HND = High nutrient dense diet
[2]HED = high energy diet

Phosphorus pollution from aquaculture

Phillips and Beveridge (1986) estimated that 85% of phosphorus fed to fish was lost to the environment. The rate of phosphorus loss in different commercial aquaculture species is reported to be mostly in between 6-48 kg/tonne of fish production and more with carp (Table 1.5.5). Among the factors, type of feed is one of the most important factors in determining the level of phosphorus loss since a much higher phosphorus loss can be obtained if trash feed is given to fish compared to dry and moist feed (Warren-Hansen 1982).

Table 1.5.5. Comparison of phosphorus loss rates (TP kg/t fish produced/year) in aquaculture

Country	Species	Culture system	Diets	kg/t	Reference
Nordic	Salmon	Cages		9.5	Enell (1995)
Norway	Atlantic salmon	Ponds	-	9.0	Ibrekk (1989)
Denmark	Rainbow trout	Ponds	Dry feed	11	Warren-Hansen 1982
Poland	Rainbow trout	Cages	Moist diet	23	Penczak *et al.* (1982)
Sweden	Rainbow trout	Cages	Dry feed	13.5	Enell & Lof (1983)
Finland	Rainbow trout	Ponds	-	18.3	Sumari (1982)
U.K	Rainbow trout	Ponds	-	15.7	Solbe 1982
U.K	Rainbow trout	-	Dry feed	27	Phillips & Beveridge (1986)
Scotland	Rainbow trout	Cages	Dry feed	27	Phillips (1985)
N. Ireland	Rainbow trout	Tanks	Diet (herring meal)	25.6	Foy & Rosell (1991a)
Italy	Rainbow trout	-	Extruded diets	6.63	Lanari *et al.* (1995b)
Italy	Rainbow trout	-	Normal pellet	7.2	Lanari *et al.* (1995b)
USA	Rainbow trout	-	Dry feed	21	Ketola (1982)
USA	Rainbow trout	Jars	-	10-15	Ketola (1991)
Canada	Brown trout	Tanks	HND	6.0	Cho *et al.* (1991)
Thailand	Shrimp	Ponds	-	13-24.4	Phillips *et al.* (1993)
Thailand	Shrimp	Ponds	-	47.6	Briggs & Funge-Smith (1994)
Indonesia	Carp	-	-	90.2±90.4	Costa-Pierce & Roem (1990)

Phosphorus as an indicator of water quality

The phosphorus concentration in water can at any given time may be used to assess the quality of freshwater aquatic environment (Table 1.5.6.). On the basis of this, it appears that effluents from fish farms and hatcheries will be of degraded quality since the discharge from fish farms and hatcheries are reported to be 0.15 mg/l total phosphorus (Warren-Hansen 1982) and 0.1 mg/l dissolved phosphate (Albaster 1982). This leads to the possibility of eutrophication in waterbodies which receive direct fish farm effluents.

Table 1.5.6. Criteria for assessing water quality of freshwater based on phosphorus level (OCE 1988).

	Total phosphorus (mg/l)	Dissolved phosphorus (mg/l)
Excellent	<0.010	<0.008
Good	<0.025	<0.020
Moderate	<0.050	<0.040
Poor	<0.100	<0.080
Degraded	<0.100	<0.080

Identification of the "right Phosphorus"

As mentioned above, feed is the main source of phosphorus pollution in aquaculture. In plants, two thirds of phosphorus is bound in phytin that trout and salmon cannot digest (Ketola 1990). It has been demonstrated that phosphorus requirements are species-specific and surplus phosphorus provided through feed is either excreted, or passed out in the faeces (Beveridge 1987). Most animal and inorganic sources of phosphorus are readily available to fish (Ketola 1990). Ketola (1982) reported that the source of the phosphorus significantly influenced the retention as well as loss rate; for example, defluorinated rock phosphate (DRP) resulted in good growth of rainbow trout with a reduction of 46% in phosphorus discharge (Ketola 1985) in comparison to dicalcium phosphate (Ketola 1991). Additionally, the bioavailabilty of dietary phosphorus is influenced by the digestibility of diet, particle size, interaction with other nutrients, feed processing and water chemistry (Lall 1991). Salmonids utilize

phosphorus from fish meal more efficiently than do carp (Yone and Toshima 1979) whereas availability of phosphorus to tilapia is low (Watanabe *et al.* 1980).

Phosphorus release from food and faeces

The primary source of phosphorus losses in aquaculture is in particulate form such as faeces and uneaten food. In general, about 30-60% of phosphorus losses by fish will be permanently accumulated in sludge (Persson 1988). The release of phosphorus from food and faeces (solid waste) to the environment depends on physico-chemical characteristics of the environment such as pH, temperature, oxygen, turbulence and microbial activity (Persson 1988). Phosphorus release from fish food was observed to be accelerated in acidic rather than in neutral or alkaline media (see Chapter 8)

Overcoming phosphorus pollution from aquaculture

Fish farm effluents can increase phosphorus levels and consequently cause eutrophication of receiving waters. As a result, further use of such water bodies for recreational or domestic or industrial purpose will be seriously affected. As phosphorus requirements are species specific it is vital that phosphorus be provided in feed at a level that maximizes growth of fish but minimizes pollution of the environment. In this context research on the development of "low pollution diets" are of utmost importance. The nutrient from aquaculture can be minimized by retaining waste water (effluents) in holding ponds and by reusing waste water on lands for growing crops in an integrated aquaculture-agriculture system. The other alternative would be to develop high energy diets which reduce phosphorus discharge to the environment compared to normal feed (Bohl *et al.* 1992). However, although aquaculture has been identified as one of the point sources of phosphorus pollution, phosphorus loadings to the environment from fish farms is minimal compared to other sources such as agriculture (Table 1.5.7).

Table 1.5.7. Typical nutrient concentration for various point source discharges (EPA 1995).

Source	Typical phosphorus (mg/l)
Dairy shed effluent	340
Feedlot effluent	150
Fish farms	0.07

To reduce phosphorus pollution, the following are important as suggested in Kibria *et al.* (1996b).

1. Estimation of phosphorus balance of species under aquaculture (Ketola 1991);

2. Feed composition and type (e.g., extruded feeds are more digestible and generate less dust and solid waste and also result in better FCR (Warren - Hansen 1982; Matty 1990);

3. Feeding techniques (e.g. avoiding overfeeding and adjusting feed amount and frequency to the culture temperature) (Seymour and Bergheim 1991);

4. Formulation of diets to meet nutrient requirements and proper choice of dietary ingredients (Kaushik 1992);

and

5. Reduction of phosphorus levels in feeds without affecting growth, feed efficiency, health and reproduction (Lall 1991; Kendra 1991), in this regard fish meal may be partially replaced by other low phosphorus protein sources to reduce phosphorus content of diet (such as with soybean meal) (Bergheim and Asgard 1996). In Nordic countries phosphorus levels in feed has been progressively reduced which resulted in a reduction of phosphorus pollution by 85% (Figure 1.5.2.). Additionally, improvements in diet digestibility may further reduce phosphorus pollution from aquaculture.

It is evident from the above review of literature that the past research on silver perch has been concentrated on its biology (Rowland 1984; Rowland and Barlow 1991; Rowland and Ingram 1991; Rowland and Kearney 1992; Rowland 1994; Rowland and Allan 1994; Rowland *et al.* 1995) and nutrition (Allan and Rowland 1992; Allan and Rowland 1994; Allan *et al.* 1993; Allan *et al.* 1994). To date there has been no attempt to study the types of waste entering the environment from aquaculture in Australia. The Environment Protection Authority (EPA) has expressed concern about projected environmental impacts from expansion of aquaculture in Australia since discharge from fish farms may change the chemistry of receiving waters (EPA

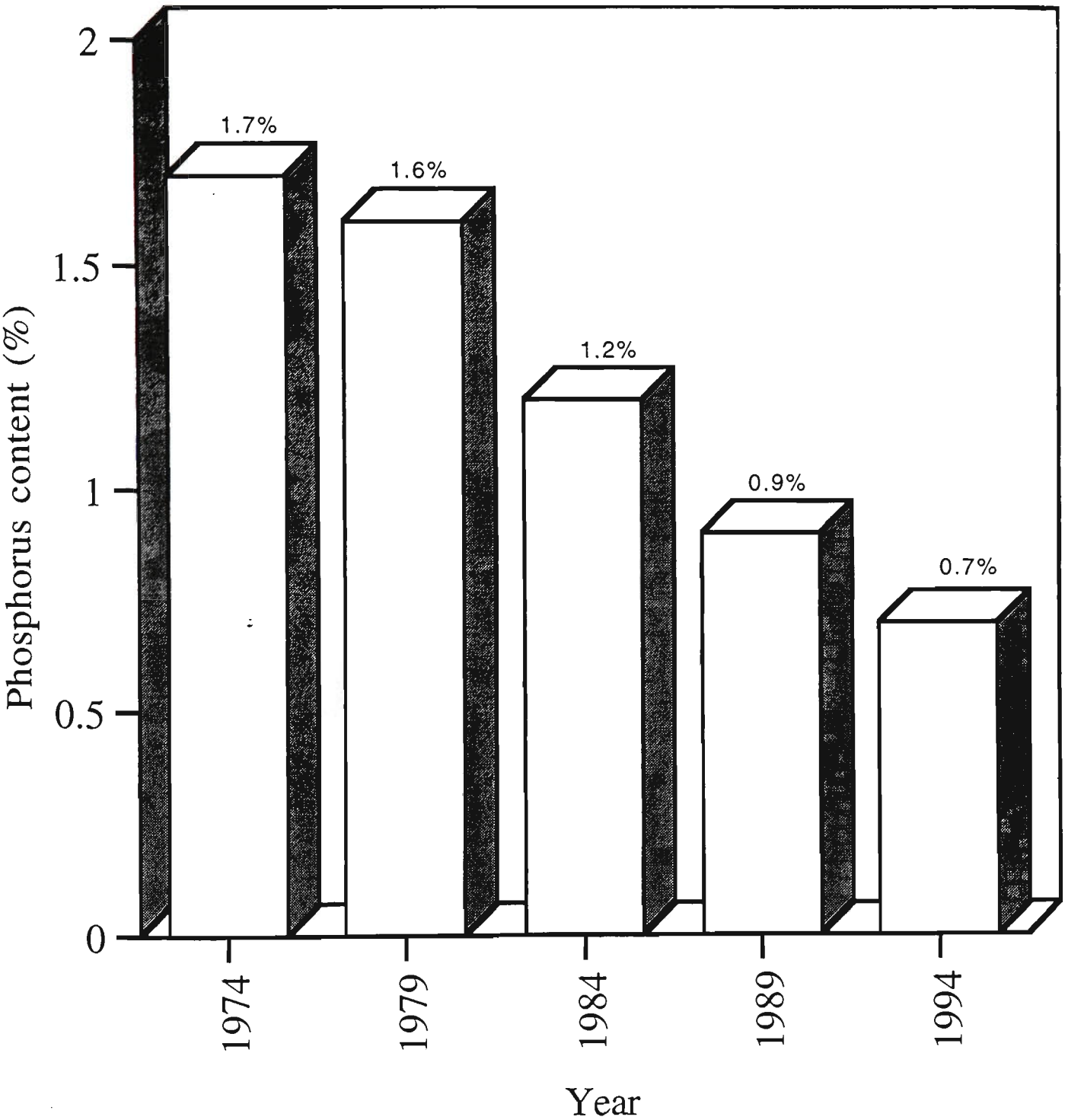

Figure 1.5.2. Reduction of phosphorus content in fish feed by Nordic countries during 1974 to 1994. This has resulted in a reduction of phosphorus pollution by 85% with an improvement of feed coefficient (data from Enell 1995).

1995). This has been further aggravated with the knowledge that aquaculture effluents are known to be one of the point sources of water pollution (State of Victoria 1995). As a result EPA, the Department of Conservation and Natural Resource Management and the Fisheries and Aquaculture sectors are interested in obtaining further information on the physical, chemical and biological data on fish farm effluents. Research conducted in Europe and North America has demonstrated a strong relationship between eutrophication in lakes and rivers and the discharge of aquaculture effluents (Albaster 1982; Penczak *et al.* 1982; Foy and Rosell 1991a, 1991b; Handy and Poxton 1993; Lanari *et al.* 1995a.,1995b). There is a need to reduce the feeding and metabolic waste from aquaculture so that aquaculture can be an environmentally sustainable development programme (Kibria *et al.* 1997b). Since feed costs account for about 60% of aquaculture production (Stickney 1986; Manzi 1989) it is also economically desirable that feed loss be minimized. There have been no previous studies on the quality and quantity of waste load from aquaculture of silver perch although such data are vital for environmental and nutritional management strategies and the planning and development of diets of low pollution potential. Similarly, there are few studies on the effects of temperature on the growth of silver perch and till now there has been no research on the use of inexpensive food such as natural zooplankton for alternative feed. The current study is the first systematic research conducted on the biological growth and pollution potential from aquaculture of silver perch. The main objectives of this study were :

1. To study the biological growth of silver perch, *Bidyanus bidyanus* at different rearing temperatures fed on artificial diets;

2. To evaluate the biochemical composition of sewage grown zooplankton for use as an alternative feed in the aquaculture of silver perch;

3. To analyse the quality and quantity of solid waste produced by silver perch at different rearing temperatures;

4. To investigate nitrogen retention by silver perch and nitrogen losses to the environment when culturing *B. bidyanus* at different rearing temperatures;

5. To study phosphorus retention by silver perch and phosphorus losses to the environment when culturing *B. bidyanus* at different rearing temperatures;

6. To analyse the nutritional composition of solid waste and the release of nutrients from solid waste;

7. To evaluate the effect of salinity on growth and pollution load of silver perch.

Chapter 2

GENERAL MATERIALS AND METHODS

2

GENERAL MATERIALS AND METHODS

The Australian native fish silver perch *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae) was selected as the test species in this study. Silver perch juvenile were reared at room temperatures and at 20°C, 25°C and 30°C and fed on three artificial commercial diets to carry out experiments on the quality and quantity of waste load in reference to solid waste, and on aspects of nitrogen and phosphorus pollution from aquaculture of silver perch. Fractionation of nutrient content in solid waste and the release of nutrients from fish food and faeces were studied under different experimental situations. An evaluation was made on the biochemical composition of sewage grown zooplankton and the performances of silver perch fed on zooplankton. Experiments were also conducted on growth and nutrient load by silver perch at 0 salinity, 4 salinity, 8 salinity and 12 salinity.

Experiments on the biological growth of silver perch and on the production of solid waste, phosphorus and nitrogen pollution, nutrient content in solid waste and the release of nutrients from solid waste were conducted at the wet laboratory at the Victoria University of Technology. Experiments on the utilization of sewage zooplankton were carried out at the Zootech multipurpose hatchery located at the Werribee, Melbourne. Details of the design of each experiment and relevant procedures have been provided in the relevant chapters. This chapter gives a summary of the general materials and methods adopted on biochemical analysis of fish food, fish, faeces and zooplankton, and analysis of water quality.

The silver perch *B. bidyanus* were purchased from a local native fish farm where fish were grown in earthen ponds. Fish were first acclimatized in the laboratory in large holding tanks (46 x 30 x 30 cm) for four weeks before use in experiments. They were reared in small glass aquaria (30 x 16 x 17 cm) and the water in aquaria was maintained at the required temperature by heating water with thermostatically controlled heater in a large tank (70 x 60 x 30 cm) in which small aquaria were placed. Fish were subjected to a short salt bath ($10g \cdot l^{-1}$) for 60 minutes prior to start of feeding trials (Rowland and Ingram 1991). The three commercial diets used in experiments referred to as diet-1, diet-2 and diet-3 containing 53%, 45% and 36% protein and 1.36%, 1.16% and 1.28% phosphorus respectively. The main objectives of this study was to quantify the waste load from aquaculture of silver perch according to the protocols outlined by Persson (1988) of Swedish Environment Protection Authority who stated that use of static aquaria are the best and most appropriate to study pollution load from aquaculture were followed. In aquaria it has been shown that the loss of uneaten feed and faecal materials can be minimized. Moreover, wastes are clearly visible, easier to collect, and any number of replicates may be used.

Domestic tap water was used after dechlorination with recommended conditioner (Sera Aquatan). For all experiments, the water quality was maintained at pH 7.5-8.0 using sodium bicarbonate or sodium bi-phosphate to adjust the pH, the dissolved oxygen level was above 6 $mg \cdot l^{-1}$, and the hardness between 80-100 $mg \cdot l^{-1}$. The pH was measured with a bench-top pH meter (Orion model SA520), dissolved oxygen by using a dissolved oxygen meter (YSI model 58), hardness by a test kit (Aquasonic, NSW) and the salinity by a conductivity meter (YSI model 33). The water temperature and fish health was monitored daily. A biological filter was used in each aquarium/tank to enhance the culture environment.

2.1 Biochemical analysis of feed, fish, zooplankton and faeces

The proximate composition (moisture, crude protein, crude fat, carbohydrate, mineral matter or ash,) of diets, fish and zooplankton were determined following AOAC (1990a; 1990b), as modified by New (1987) and O'Brien (1994).

2.1.1. Moisture determination

The moisture content was determined by drying samples in a flat, aluminium dish in an oven at 100^{o}C until a constant weight was recorded (New 1987).

$$\text{Moisture content (\%)} = \frac{\text{Weight of fresh sample - Weight dry sample}}{\text{Weight of fresh sample}} \times 100$$

2.1.2. Crude protein determination

Crude protein (N x 6.25) in feed, fish and faeces was determined by the semi-micro Kjeldahl digestion method. About 0.2g of samples were placed into a 50 ml digestion tubes with 1 Kjeldahl catalyst tablet (Ajax Cat No. 1509), and 5 ml concentrated sulphuric acid (H_2SO_4). The samples were then heated on the digestion block for 30 minutes at 150^{o}C, then for 30 minutes at 250^{o}C and then at 30 minutes or more at 360^{o}C until the solution was clear. After digestion, the solution was allowed to cool and after cooling, deionized water (Milli-Q-H_2O) was added to bring the total volume to 50 ml. The solution was then vortexed for one minute for constant mixing on a vortex mixture. 0.1 ml aliquot of digested sample was transferred into a spectro tubes where 8.9 ml of salicylate reagent {(sodium nitroprusside [$(Na_2(Fe(CN)_5NO)2H_2O)$] + sodium salicylate [$(C_6H_4(OH)COONa)$]} and 1.0 ml of cyanurate reagent {(sodium dichloroisocyanurate ($C_2N_2O_2Cl_2Na.2H_2O$) + sodium hydroxide (NaOH)} was added and left for 30 minutes for colour development. The colour developed was read at 697 nm on a spectrophotometer and the percentage of nitrogen was calculated as follows :

Nitrogen content of sample (%) = $\frac{\text{Absorbance - Blank}}{\text{Slope}} \times \frac{A}{B} \times 10^4$ (O'Brien 1994)

where
A = Final volume
B = Weight of sample taken

2.1.3. Crude fat analysis

Crude fat of feed, fish and zooplankton was determined by the ether extraction method (New 1987). 2g of dried samples was placed into an extraction thimble and thimble containing the samples was inserted into a soxhlet apparatus. A dry solvent flask was placed beneath the soxhlet apparatus, 200 ml of petroleum ether (b.p. 40-60^{o}C) was added and the flask containing the ether was heated for 16 hours. After 16 hours the flask was placed in a water bath to remove the ether and dried at 105^{o}C for 30 minutes. The flask was cooled in a desiccator and weighed in an analytical balance. The crude fat was calculated using the following formula :

Crude fat (% of DM) = $\frac{\text{Weight of fat}}{\text{Weight of sample}} \times 100$ (New 1987)

2.1.4. Crude ash determination

Ash was determined after placing 2g of the dry sample in a dry, tared porcelain dish in a muffle furnace and burning at 600^{o}C until the residue was free of organic matter (New 1987). It was cooled in a desiccator and weighed using an analytical balance. The ash (%) was calculated as :

Ash (%) = $\frac{\text{Weight of ash}}{\text{Weight of sample}} \times 100$ (New 1987)

2.1.5. Carbohydrates

The carbohydrates or nitrogen free extract (NFE) was calculated by the difference between the dry matter of the sample and the sum of the determined crude protein, ether extract and ash :

NFE : 100 - crude protein (%) - crude fat (%) - ash (%) (Lanari *et al.* 1995a).

2.1.6. Phosphorus determination

Samples for analysis of phosphorus were digested following the protein digestion methods described above (2.1.2). Next 0.2 ml aliquots of digested samples were transferred into spectro tubes, and mixed with 1.0 ml of modified Murphy and Riley (M & R) reagents {(ammonium molybdate $[(Mo)(NH_4)_6Mo_7O_{24} \times 4H_2O)$ + conc. sulphuric acid $[(H_2SO_4)]$, + Milli-Q-$H_2O)]$}. After mixing, 8.8 ml of Milli-Q-H_2O was added using an automatic dispenser. The samples were then left overnight for full colour development. The colour developed was read at 827 nm on a spectrophotometer and the percentage of phosphorus was calculated as follows :

$$\text{Phosphorus (\%)} = \frac{\text{Absorbance - Blank}}{\text{Slope}} \times \frac{A}{B} \times 10^4 \quad \text{(O'Brien 1994)}$$

where
A = Final volume
B = Weight of sample taken

2.1.7. Proximate composition and amino acids content of diets

The three commercial diets referred to as diet-1, diet-2 and diet-3 were used to carryout experiments on the biological growth of silver perch, and on the production of solid waste, and pollution load. The biochemical composition of the three diets determined are given in Table 2.1. The specifications of the three diets obtained from the feed companies are described in Table 2.2.

Table 2.1. Proximate composition of diets used for studies on biological growth and nutrient load.

	Diet-1	Diet-2	Diet-3
Dry matter, DM (%)	91.80	90.12	88.99
Crude protein (% DM)	53.00	45.00	36.00
Ether extract (% DM)	06.92	08.50	04.96
Ash (% DM)	13.06	09.84	13.00
N-free extract (% DM)[1]	27.02	36.66	46.04
Nitrogen (%)	08.48	07.20	05.74
Phosphorus (% DM)	01.31	01.16	01.28
Gross energy (MJ/Kg DM)[2]	19.89	20.28	18.40
Digestible energy (MJ/kg DM)[3]	17.24	17.34	15.39
DP:DE (g/MJ DM)	30.74	25.95	23.39

[1] The Nitrogen free extract = 100 - Crude protein - Ether extract - Ash (Lanari *et al.* 1995a).

[2] Gross energy contents of the diets were estimated by using values of 0.0236 MJ/g protein, 0.0395 MJ/g lipid and 0.0172 MJ/g carbohydrate (NFE) (Silver *et al.* 1993).

[3] The digestible energy contents of the diets were estimated by using values of 0.02213 MJ/g protein (assuming 90% is digestible) 0.0356 MJ/g lipid (assuming 90% lipid is digestible), and 0.0129 MJ/g carbohydrate (assuming 75% is digestible) (Silver *et al.* 1993).

Table 2.2. Specification of three diets used in different experiments (information as available from the feed companies).

Diet - 1	
Feed type :	A feed for fast growing warm water silver perch fry until juveniles reach 5g in weight. A concentrated diet high in vitamins and energy.
Ingredients :	Fish meal, blood meal, soybean meal and fish oil
Feeding rate :	3% of body weight.
Diet - 2	
Feed type :	An extruded dry diet of semi-floating crumble for feeding juveniles until silver perch are about 6 cm long
Ingredients :	Grain cereals, and cereal by-products, fish, meat and meal by-products, whole grain legumes, methionine, vitamins, minerals and natural organic acids.
Feeding rate :	twice per day; 3% of body weight.
Diet - 3	
Feed type :	For silver perch (0.5-15g), 2mm crumbles are recommended for 0.5-15g silver perch.
Ingredients :	Grains, grain by products, soybean meal, fish meal, blood meal, synthetic amino acids, fish oil, vitamins, trace minerals and an antioxidant.
Feeding rate :	3% of body weight.

The amino acid of the three diets were analyzed at the State Chemistry Laboratory, Melbourne and are given in Table 2.3.

Table 2.3. Essential and non-essential amino acids composition of the three diets used in feeding trials ($g.100^{-g}$ dry weight).

	Diet-1	Diet-2	Diet-3
Essential-amino acids			
Arginine (Arg)	2.91	2.42	2.08
Histidine (His)	1.79	1.29	1.06
Isoleucine (Iso)	1.97	1.66	1.54
Leucine (Leu)	4.68	3.25	2.98
Lysine (Lys)	4.20	2.65	2.23
Methionine (Met)	1.18	0.80	0.84
Phenylalanine (Phe)	2.59	1.86	1.60
Threonine (Thr)	2.32	1.68	1.43
Tryptophan (Try)	0.59	0.43	0.40
Valine (Val)	3.18	2.36	1.95
Non-essential amino acids			
Alanine (Ala)	2.37	2.27	2.07
Asparatic acid (Asp)	3.75	3.54	3.19
Cysteine (Cys)	0.57	0.44	0.43
Glutamic acid (Glu)	6.87	6.36	5.54
Glycine (Gly)	3.03	2.03	1.7
Proline (Pro)	3.13	2.03	1.78
Serine (Ser)	2.35	1.88	1.66
Tyrosine (Tyr)	1.37	1.17	1.05

2.2. Water quality analysis

Nitrogen and phosphorus in water

Water samples for ammonium, nitrite, nitrate, total nitrogen, and orthophosphate were determined using a Tecator flow injection analyzer (FIA). The methods was as described in the Tecator flow injection instruction manual, Tecator (1990). All water samples were first filtered through a 0.45µm membrane filter prior to analysis.

2.2.1. Ammoniacal nitrogen (NH_4^+-N)

In the flow injection analysis, samples containing ammonium ions (NH_4-N) were first mixed with sodium hydroxide (0.5 M NaOH) which resulted in an alkaline medium where gaseous ammonia is formed ($NH_4^+ + OH \leftrightarrows NH_3(g) + H_2O$). The ammonia gas is diffused through a gas permeable membrane into an indicator (N red) stream,

composed of a mixture of acid-base indicators. A colour change which represented a shift in pH was measured spectophotometrically at 590 nm.

2.2.2. Nitrite (NO_2^--N)

The nitrite/nitrate was determined by the ammonium chloride (NH_4Cl), sulphanilamide ($C_6H_8N20_2S$) and N-(1-naphthyl)-Ethylene Diamine Dihydrochloride ($C_{12}H_{14}N_2$ x 2 HCl) method as described in the Tecator instruction manual (Tecator 1990). The sample containing nitrite (NO_2-N) was injected into a stream of ammonium chloride, which was then reacted with an acidic sulphanilamide and N-(1-naphthyl)-Ethylene Diamine Dihydrochloride (NED) solution to form a reddish-purple azo dye. The absorbance of this reddish colour was determined at 540 nm.

2.2.3. Nitrate (NO_3^--N)

The water samples containing nitrate (NO_3-N) were injected into a stream of ammonium chloride through a cadmium reductor which reduced nitrate to nitrite. The nitrite reacted with an acidic sulphanilamide and N-(1-naphthyl)-Ethylene Diamine Dihydrochloride (NED) forming a purple azo dye. This purple azo dye was measured at 540 nm.

2.2.4. Total nitrogen (NO_3^--N)

5 ml of potassium persulphate ($K_2S_2O_8$) solution (prepared following instruction) was added to each water samples used for total nitrogen determination. The samples were then heated for 30 minutes in an autoclave at a pressure of 200 kPa. After cooling the samples, 0.15 ml of 1 M sulphuric acid, and one drop of phenolphthalein ($C_{20}H_{12}O_4$) solution were added to each sample. The samples were then neutralized with 0.12 M sodium hydroxide (NaOH) until a pale pink colour developed indicating that nitrogen had been converted to nitrate. The autoclaved samples were then analyzed for nitrate following the procedures described above.

2.2.5. Orthophosphate (PO_4^{3-}-P)

The orthophosphate and total phosphorus in water was determined by the molybdate [$(Mo)(NH_4)_6Mo_7O_{24}$ x $4H_2O$], hydrazine ($N_2H_6SO_4$) and stannous chloride ($SnCl_2$) method as described in the instruction manual (Tecator 1990). The sample containing orthophosphate was reacted first with ammonium molybdate to form the heteropoly molybdophosphoric acid, which was subsequently reduced to phosphomolybdenum blue by stannous chloride in a sulphuric acid medium. The intensive blue colour developed was then measured spectrophotometrically at 690 nm.

2.2.6. Total phosphorus (PO_4-P) in waters

0.15 ml of 4 M sulphuric acid and 3 ml of potassium persulphate was added to samples (15.0 ml) for total phosphorus determination in order to convert all phosphorus species to orthophosphate. The samples were then heated for 30 minutes in an autoclave at 200 kPa. The digested samples containing orthophosphate were then measured spectrophotometrically as above.

2.3. Cleaning of glassware used for phosphorus and nitrogen determination

For routine water quality analysis, glassware, centrifugation tubes and plastic containers were soaked in decon, rinsed three times with tap water and finally two times with distilled water. For phosphorus and nitrogen determination, glassware had a light wash with $NaHCO_3$ solution, followed by rinsing in tap and distilled water, soaked overnight in an acid bath containing 6N HCL and washed again with tap and distilled water (Adams 1990). When necessary glassware was soaked in chromic acid solution ($Na_2Cr_2O_7$) ($70g \cdot l^{-1}$) followed by soaking into $NaHCO_3$ solution. Pipettes used for drawing reagents were rinsed with deionised water and later with the solution to be transferred. All spectrophotometer cells were rinsed with deionized water prior to use.

2.4. Preservation of samples

Water samples collected were analyzed as soon as possible. If there was any delay in analysis, the samples were preserved and stored in appropriate containers following the methods of American Public Health Association (APHA 1989). Where there was a need for transportation of samples, samples were placed in crushed or cubed ice blocks before shipment. Samples of faeces, uneaten food, and fish were kept in a deep freezer at -25^{o}C until analyzed.

2.5. Statistical analysis

The sample mean, standard deviation, and standard error were calculated according to the method of Zar (1984). Percentage data were transformed to arc sin values prior to analysis to achieve homogeneity of variances. One way analysis of variance (ANOVA) was conducted to compare the means of specific growth rate (SGR), food conversion ratio (FCR), protein efficiency ratio (PER) and nutrient load using an IBM compatible MS Excel 5.0 programme. Curves were fitted (linear or exponential or polynomial as appropriate) to indicate the trend or correlations in data on the release of nutrients of ammonium and orthophosphate, nutrient intake vs faecal nutrient lost. The coefficient of determination (r^2) was used to determine the strength of the correlation between variables and curves were fitted to data as appropriate using Cricket graph-III (version 1.0, Computers Associates International).

2.6. Formulae used

Apparent net protein utilization (ANPU) = $\frac{\text{final body nitrogen - initial body nitrogen}}{\text{amount of nitrogen consumed}}$ (Jauncey 1982)

Feed conversion ratio (FCR) = $\frac{\text{weight of feed fed}}{\text{wet weight gain}}$ (Laird & Needham 1988)

Gain in weight or absolute growth = $W_2 - W_1$ (Chiu 1989)

where W_2 is the final weight of fish
W_1 is the initial weight of fish

Nutrient retention = $\frac{(\text{final body nutrient - initial body nutrient}) \times 100}{\text{total nutrient fed}}$ (Brown *et al.* 1993)

Percentage weight gain = $\frac{\text{Final average weight}}{\text{Initial average weight}} \times 100$ (Ballestrazzi *et al.* 1994)

Protein efficiency ratio (PER) = $\frac{\text{g wet weight gain}}{\text{g crude protein fed}}$ (EIFAC 1980; De Silva & Anderson 1995);

Specific growth rate (SGR) = $\frac{\ln \text{final weight} - \ln \text{initial weight}}{\text{total days of experiment}}$ (Laird and Needham 1988)

where ln = natural logarithm

Definitions of abbreviations, terms and units used in this thesis are given in Appendix II (pages 262-265).

Chapter 3

EFFECT OF TEMPERATURE ON THE BIOLOGICAL GROWTH OF SILVER PERCH FED ON ARTIFICIAL DIETS

3

EFFECT OF TEMPERATURE ON THE BIOLOGICAL GROWTH OF SILVER PERCH FED ON ARTIFICIAL DIETS

INTRODUCTION

The silver perch (*B. bidyanus*) is an Australian native fish with high potential for aquaculture (Rowland *et al.* 1995). It is one of the four principal species of the Murray-Darling River system and is much sought after by commercial and recreational fishers (Cadwallader 1979). Although aquaculture is an infant industry in Australia, interest in the culture of silver perch is growing both in Australia and in nearby Asia (Gooley and Rowland 1993). Silver perch currently represents a major native aquaculture industry in the country (Kibria *et al.* 1996a)

Previous research on *B. bidyanus* has concentrated on its production (Rowland and Barlow 1991; Allan and Rowland 1992; Rowland and Allan 1994; and Rowland *et al.* 1995) and nutrition (Allan and Rowland 1992; Allan *et al.* 1994 and Allan and Rowland 1994) (see also review on silver perch biology and aquaculture in chapter 1.1). There is however only limited information on the effects of temperature on silver perch growth although temperature is known to be a most important factor controlling growth, metabolic rate, food conversion and the survival of fish and crustaceans (Farmanfarmaian and Moore 1978; Barlow 1986; Cho and Kaushik 1990). In addition, the rearing temperature may also determine the amount of solid and soluble waste load from aquaculture (see chapter 5, 6, 7 and 8). There is a paucity of data and information on the effects of temperature on the growth of Australian species including the silver perch (Barlow 1986; Maguire and Allan 1992). The silver perch is known to tolerate a wide range of temperature from 2-32^{o}C (Cadwallader and Backhouse 1983); however the optimum temperature for growth of Australian native fish and crayfish is not fully

known (Barlow 1986). Silver perch can grow at temperature as low at 12^{o}C (Barlow and Bock 1981) and a reasonable growth was reported when the temperature exceeds 14^{o}C (Barlow 1986). The current experiments were conducted to evaluate the effects of a range of temperature on gain in weight, specific growth rate (SGR), and food conversion ratio (FCR) of silver perch juveniles fed on three artificial diets.

MATERIALS AND METHODS

The experiment was conducted by rearing silver perch juveniles in glass aquaria (30 x 16 x 17 cm). Fish were acclimatized and the water quality parameters was maintained in the same way as described in earlier chapter (chapter 2). Fish were fed on the three commercial diets referred to as diet-1, diet-2 and diet-3 with protein content of 53%, 45% and 36% respectively. Further information and specifications of the three diets are given in table 2.1, 2.2, and 2.3 (chapter 2.). Fish were fed on silver crumbles/starter (appropriate for juveniles) at the rate of 3% of their body weight as recommended by the feed manufacturer. Proximate analysis of the diets was determined following AOAC protocols (1990a,1990b). Protein was analyzed by the semi-Kjeldahl method (Nx6.25), fat by ether extraction, moisture by drying in an oven overnight at 100^{o}C for 24 hours, ashing by burning samples in a muffle furnace at 600^{o}C overnight, and carbohydrate calculated by the difference in weight (details are given in chapter 2.1.).

Two experiments were conducted to evaluate the effect of temperature on the growth of silver perch juveniles. The first experiment was conducted at the room temperature (18-20^{o}C) and called here as a 'preliminary experiment' and the second experiment was conducted by setting water temperature at 20^{o}C, 25^{o}C and 30^{o}C. The experimental details are described below :

Trial-1 (Preliminary experiment) : Effects of feed on growth at room temperature

Fish were reared in glass aquaria at room temperature (18-20^{o}C) for four weeks. Individual fish were kept in separate aquaria to study individual growth performance with respect to gain in weight, food conversion ratio (FCR) and specific growth rate (SGR). Fish were fed on three diets (diet-1, diet-2 and diet-3) at the calculated rate of 3% of their body weight. The feed was divided into two portions and fed twice a day (0900 and 1600 hours respectively), six days a week for four weeks. Unfed fish (controls) were kept in separate aquaria to compare growth performance of fed fish. Sampling was conducted once a week to measure gain in weight and to adjust the amount of feed to be fed per day. The experimental details are provided in Table 3.1.

Trial 2 : Effects of temperature (20^{o}C, 25^{o}C and 30^{o}C) on growth, FCR and SGR

The silver perch juveniles were reared at 20^{o}C, 25^{o}C and 30^{o}C for four weeks to study the effect of temperature on growth, food conversion and specific growth rate. The experimental details are given in Table 3.2. The water temperature for this trial was maintained by heating water with a thermostatically controlled heater in a large tank (70 x 60 x 30 cm) in which the six replicate aquaria were placed. Fish were fed on the three diets (diet-1, diet-2 and diet-3) at the rate of 3% of their body weight. The feed was divided into two portions and fed twice a day (0900 and 1600), and six days a week for four weeks. Sampling was conducted bi-weekly when the feed amount was adjusted with respect to mean weight of fish per aquarium.

Results were analyzed by one way analysis of variance (ANOVA) or a 2 sample student t test (Zar 1984) using an IBM compatible MS Excel programme 5.0. The weight gain, percentage weight gain, SGR, FCR and PER were calculated using the appropriate formulae given in the chapter 2.6.

Table 3.1. Experimental details of feeding trial-1 conducted at room temperature (18-20^{o}C) with silver perch juveniles fed on three diets.

Trial no.	Temperature (^{o}C)	Tank size (cm)	Diet no.	No tanks per diet	No fish per tank	No fish per treatment	Average initial size (mg)
1	18-20	30 x16 x17	1	10	1	10	1,540
1	18-20	30 x16 x17	2	10	1	10	1,140
1	18-20	30 x16 x17	3	10	1	10	1,440

Table 3.2. Experimental details of feeding trial-2 conducted at three different temperatures (20^{o}C, 25^{o}C and 30^{o}C) with silver perch juveniles fed on three diets.

Trial no.	Temperature (^{o}C)	Tank size (cm)	Diet no.	No tanks per diet	No fish per tank	No fish per treatment	Average initial size (mg)
2	20	30 x16 x17	1	6	4	24	625
2	20	30 x16 x17	2	6	4	24	630
2	20	30 x16 x17	3	6	4	24	627
2	25	30 x16 x17	1	6	4	24	615
2	25	30 x16 x17	2	6	4	24	622
2	25	30 x16 x17	3	6	4	24	631
2	30	30 x16 x17	1	6	4	24	630
2	30	30 x16 x17	2	6	4	24	628
2	30	30 x16 x17	3	6	4	24	634

RESULTS

Trial 1 : Preliminary experiment

Silver perch juveniles fed on diet-2 showed a trend of slightly better growth rate (Figure 3.1). However, statistical analysis did not reveal any significant difference ($P>0.05$) in weight gain between the fish fed on the three different diets. Moreover, a statistically significant feed conversion ratio (FCR) and specific growth rate (SGR) were achieved in fish fed on diet-2. than fish fed on the other diets (Table 3.3). Fish fed diets 1 and 3 had significantly lower SGR and higher FCR values than those fed diet 2. The survival in this preliminary experiment was 100%.

Table 3.3. Weight gain, specific growth rate and food conversion ratio of individual silver perch fed three diets and reared at room temperatures for 28 days in feeding trial-1.

	Diet-1	Diet-2	Diet-3
Initial average weight (mg)	1540	1,140	1,440
Final average weight (mg)	1,980	1,600	1,830
Average gain in weight (mg)	440 ± 19^{a}	460 ± 22^{a}	390 ± 50^{a}
Percentage weight gain	29 ± 1.40^{a}	40 ± 2.2^{b}	27 ± 1.70^{a}
Specific growth rate (SGR)	1.05 ± 0.07^{a}	1.41 ± 0.03^{b}	0.99 ± 0.04^{a}
Food conversion ratio (FCR)	2.97 ± 0.06^{a}	2.24 ± 0.10^{b}	3.20 ± 0.04^{a}
Protein efficiency ratio (PER)	0.69 ± 0.02^{1a}	0.99 ± 0.07^{b}	0.89 ± 0.06^{a}
Survival (%)	100	100	100

Values are mean±SE; n=10.
Values in the same row with common superscripts are not significantly different ($P>0.05$).

Trial 2 : Effect of temperature

Silver perch showed the highest growth at 25°C and the least growth at 20°C in the order of 25°C>30°C>20°C (Figure 3.2). The gain in weight of silver perch was significantly different at different temperatures being significantly higher at 25°C (Table 3.4) ($P<0.05$). The specific growth rate was also found to be significantly greater at 25°C. Fish grown at 25°C had better FCR and PER than at either 30°C or 20°C (Table 3.4). The survival at 20°C and 25°C was 100% but there were some mortalities at 30°C.

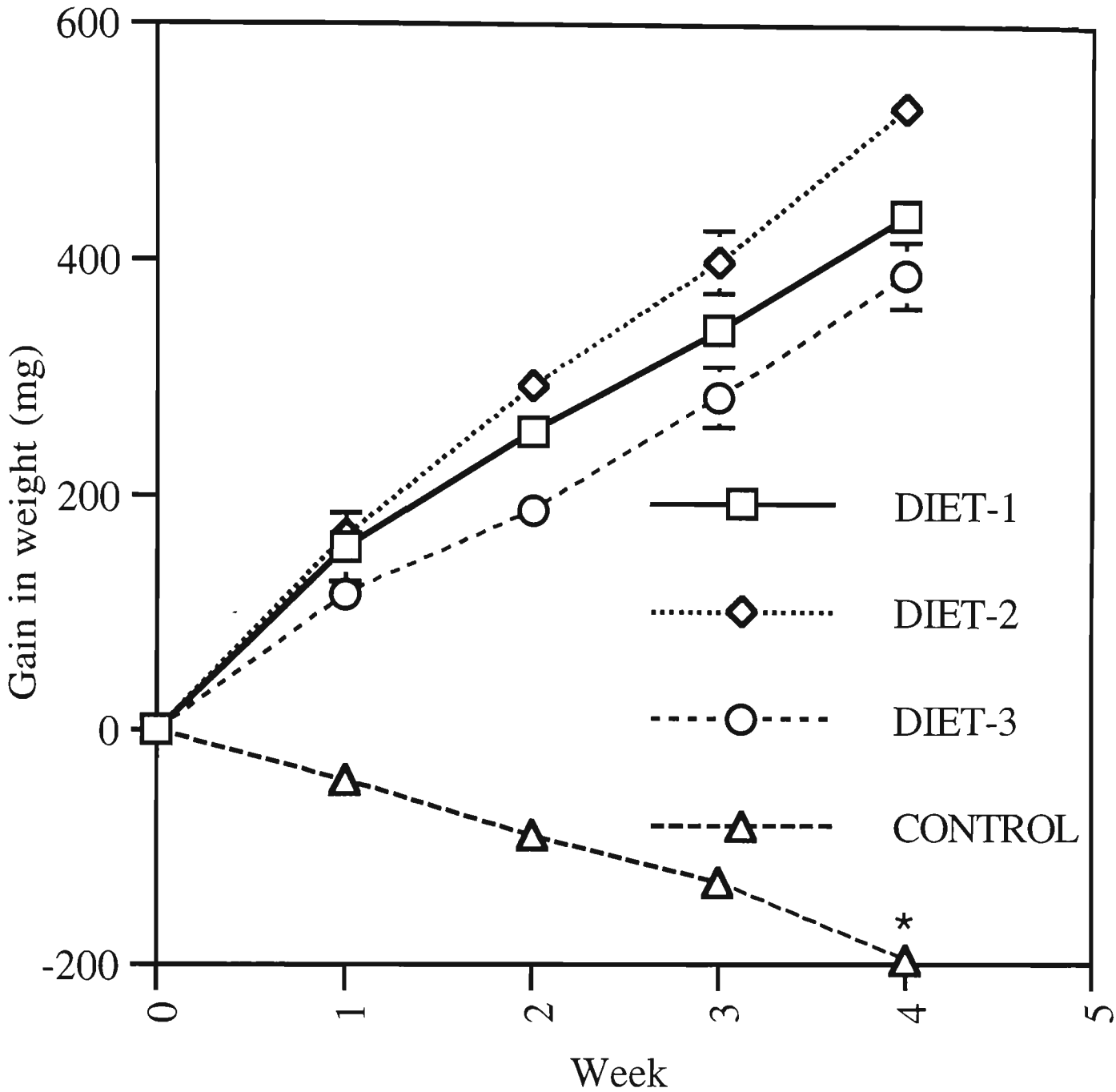

Figure 3.1. Gain in weight of silver perch juveniles fed three diets and reared at room temperatures (18-20 degree celcius) for 28 days. There was no significant difference in weight gain of fish fed the three diets (P>0.05). The control group was unfed (mean±SE; n=10).

* = denotes significantly different

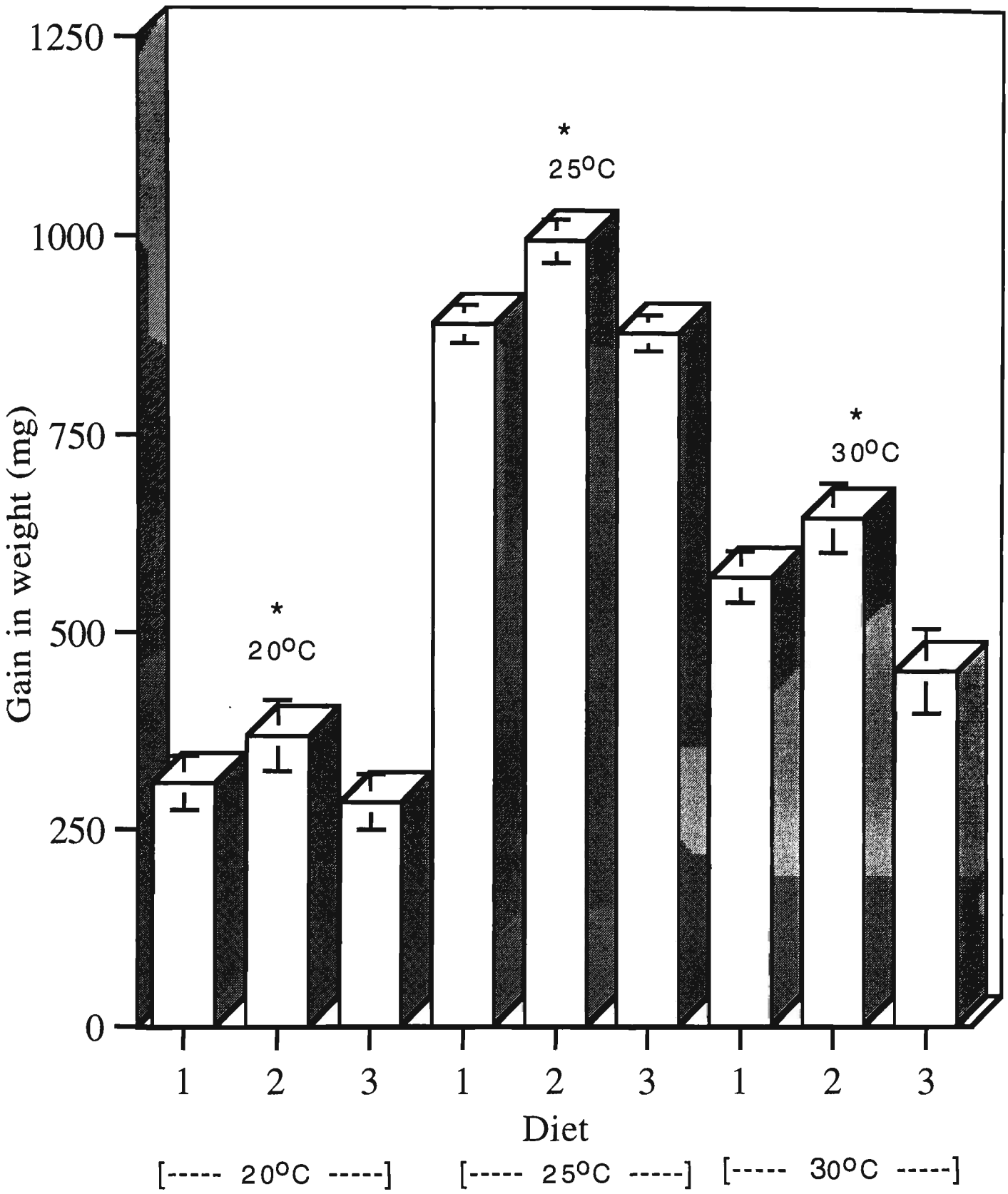

Figure 3.2. Gain in weight of silver perch juveniles fed three diets (here indicated as 1, 2, 3) and reared at 20, 25 and 30 degree celcius for 28 days. The gain in weight was significantly different in fish held at the three temperatures ($P<0.05$)(mean$\pm$SE; n=6).

* = denotes significantly different

Table 3.4. Performance of silver perch fed on three diets and reared at 20°C, 25°C and 30°C for 28 days in trial-2.

	[----------20°C----------]			[----------25°C----------]			[----------30°C----------]			
	Diet-1	Diet-2	Diet-3	Diet-1	Diet-2	Diet-3	Diet-1	Diet-2	Diet-3	Pooled SE
Initial average weight (mg)	625	630	627	615	622	631	630	628	634	
Final average weight (mg)	934	999	912	1501	1618	1519	1203	1276	1087	
Percentage weight gain[1]	49.44[a]	58.57[a]	45.45[a]	144.9[b]	160.2[b]	139.6[b]	90.95[c]	103.2[c]	71.45[c]	4.3
Specific growth rate (SGR)[2]	1.67[a]	1.92[a]	1.56[a]	3.72[b]	3.98[b]	3.66[b]	2.70[c]	2.95[d]	2.25[c]	0.016
Food conversion ratio (FCR)[3]	2.17[a]	1.91[a]	2.32[a]	1.05[b]	1.00[b]	1.10[b]	1.40[c]	1.29[c]	1.64[c]	0.11
Protein efficiency ratio (PER)[4]	0.87[a]	1.16[a]	1.19[a]	1.79[b]	2.22[b]	2.58[b]	1.35[c]	1.73[c]	1.68[c]	0.09
Survival (%)	100	100	100	100	100	100	91.70	96.00	91.70	

[1]Percentage weight gain = $\frac{\text{final average weight}}{\text{initial average weight}} \times 100$ (Ballestrazzi *et al.* 1994)

[2]Specific growth rate (SGR) = $\frac{\ln \text{final weight} - \ln \text{initial weight}}{\text{total days of experiment}}$ (Laird and Needham 1988)

where ln = natural logarithm

[3]Feed conversion ratio (FCR) = $\frac{\text{weight of feed fed}}{\text{wet weight gain}}$ (Laird & Needham 1988)

[4]Protein efficiency ratio (PER) = $\frac{\text{g wet weight gain}}{\text{g crude protein fed}}$ (EIFAC 1980; De Silva & Anderson 1995);

[5]Values are mean±SE; n=6;

[6]Values in the same row with common superscripts are not significantly different from each other (P>0.05).

DISCUSSION

In both the preliminary experiment (trial-1) and the temperature trial (trial-2), fish fed diet-2 containing 45% protein showed a slightly higher gain in weight compared to fish fed diet-1(53%) or diet-3(36%) although statistical analysis did not reveal any significant differences in gain in weight of fish fed the three diets ($P>0.05$). The result obtained showed a trend that at all temperature diet-2 consistently achieved a slightly higher gain in weight to silver perch compared to other two diets ($P>0.05$) (Figure 3.2). Although optimum protein requirements of silver perch has not yet been demonstrated, the results of this study have indicated a trend in that small silver perch juveniles may perform slightly better on a 45% protein diet although as indicated no significant differences were found.

The maximum growth of silver perch was found to be at 25^{o}C which suggests that this temperature is close to the optimum growing temperature of silver perch ($P<0.05$) (Figure 3.2). Barlow (1986) has reported that the optimum growing temperature is between 20^{o}C and 30^{o}C (Barlow 1986) while Rowland *et al.* (1995) indicated a figure of 23^{o}C and 28^{o}C. It is well known that all species demonstrate a peak growth at their optimum temperature and that the growth rate falls at temperatures other than the optimum (Brett 1979; Hepher 1988). In poikilothermic animals, growth increases with an increase in temperature but decreases above the optimum temperature (Brett 1979). At low temperatures, the rate of digestion is greatly reduced and gross conversion efficiency could be less as a result of reduced digestive efficiency and fish consuming less feed (Brett 1979; Cho and Kaushik 1990). At higher than optimum temperature appetite and activity of fish is less which could suppress the growth and food conversion efficiency (Brett 1979; Cossins and Bowler 1987). In fact, the optimum growth of aquatic animals occurs over a reasonable and fixed narrow range of temperature (Hepher 1988; Jones 1991) and the growth rate is gradually suppressed at lower and higher of optimum temperature (Cossins and

Bowler 1987; Jones 1991). For example, bluntnose minnows, *Pimephales notatus* grew faster at 25°C (close to optimum), more slowly at 30°C and most slowly at 15°C (Gill and Weatherley 1984). In crayfish, *Cherax quadricarinatus*, growth was found to be best at 28°C and significantly less at 32°C. The author did suggest that poor growth of crayfish at 24°C and 32°C may represent low and high extremes for good growth in this species (Jones 1991). The lower growth at 30°C and least growth at 20°C found in this study may represent two extremes from the optimum growing temperature (25°C) of silver perch (Table 3.4; Figure 3.2).

The growth of silver perch is primarily determined by the temperature (Rowland *et al.* 1994; Rowland 1995b; Rowland 1995c) which is also reflected in the present study. Allan and Rowland (1991) reported that silver perch juveniles grow slowly at 18.3-22.8°C. Similarly, silver perch juveniles grow at a lower rates during the coldest winter months in earthen ponds (Rowland *et al.* 1995) and at a higher rate during the warmer months (Rowland 1995a; Rowland 1995b). When silver perch are exposed to temperatures of 30°C and in excess of 30°C for 3-4 weeks there is an affect on appetite, food conversion and growth (Rowland and Bryant 1995). This may account for the lower growth and FCR obtained with silver perch at 30°C compared to 25°C in this study (trial-2; Table 3.4). In addition to temperature, ammonia level could be higher at a higher temperature (see chapter 6) and may have affected the growth of silver perch since higher ammonia level in the culture system could stress or may cause mortalities (Boyd 1990; Hart and O'Sullivan 1993).

The medium of culture may affect growth of silver perch. It appears that the growth of silver perch in aquaria is slower than in commercial ponds which may indicate that the artificial tanks may not be a preferable culture medium to achieve the maximum growth of silver perch. Growth of silver perch in artificial medium (tanks) was reported to be significantly less than in fish cultured in earthen ponds (Rowland and Bryant 1995; McKinnon *et al.* 1996). Similarly, crayfish have shown a poor

growth in concrete and plastic-lined ponds whereas earthen ponds were very effective in crayfish rearing (Tradwell *et al.* 1992). This may well account for the relatively less weight gain demonstrated by juvenile silver perch in the current study.

It is concluded that there was a trend for the 45% protein diet to give a higher weight gain compared with the other two diets tested although the result was not significant. It was also shown that silver perch grew significantly better at 25^{o}C than at either 20^{o} or 30^{o}C ($P<0.05$) indicating that the optimum temperature for silver perch is close to 25^{o}C.

Chapter 4

UTILIZATION OF SEWAGE GROWN ZOOPLANKTON : BIOCHEMISTRY OF ZOOPLANKTON AND THE PERFORMANCE OF SILVER PERCH FED ON SEWAGE ZOOPLANKTON

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INTRODUCTION

Zooplankton are the natural food of many marine and freshwater fish and prawn. It has been extensively used in the rearing of larvae and fry (De Pauw *et al.* 1981; Tay *et al.* 1991). Both live (Dabrowski 1984; Alam *et al.* 1993) and frozen zooplankton (Brett 1971; Sargent *et al.* 1979) have been used in the rearing of finfish and crustaceans. Previous research has demonstrated that fry and juveniles perform better when fed on zooplankton than on artificial dry diets (Dabrowski 1984; Dave 1989). Frozen and freeze dried zooplankton represent an excellent food source for hatchlings of white fish, carp, Lake char and species which normally require live plankton for growth (Bryant and Matty 1980).

Zooplankton are regarded as a valuable source of protein, amino acid, lipid, fatty acid and enzymes in aquaculture (Ogino 1963; Pillay 1990; Millamena *et al.* 1990; Munilla-Moran *et al.* 1990) (see also review on biochemical composition of zooplankton in chapter 1.2). Yurkowski and Tabachek (1979) reported that the essential amino acids in cladocerans, *Daphnia pulex* and copepods, *Diaptomus sp.* is equal to or greater than those normally required by fish. Studies on the nutrient composition of natural zooplankton are important as these would aid in determining the suitability of the organisms as feed ingredients in aquaculture (Yurkowski and Tabachek 1979). The nutritional quality of living organisms can be evaluated by determining their proximate and amino-acids composition, and by the protein efficiency ratio (PER) and net protein utilization (Watanabe *et al.* 1978). Zooplankton grows

abundantly in the nutrient rich Werribee Sewage Treatment Lagoons (WSTL), Melbourne but the resource is underutilized. There appear to be no previous reported studies on the utilization of this resource for aquaculture in Australia. Currently fish meal is the main protein source for fish feeds (Lovell 1989), however it is becoming expensive because of high demand and a reduction in supply (Barlow 1989). There is a need therefore, to evaluate ingredients which are cheaper to use and could be an alternative protein source in the culture of fish. Silver perch is an Australian native fish of high aquaculture potential and represents the main native freshwater aquaculture industry in Australia (Kibria *et al.* 1996a). Zooplankton are the major natural food of young and adult silver perch (Lake 1967a; Cadwallader and Backhouse 1983; Rowland 1984) (see also chapter 1.2 for natural food of silver perch). There has been to date no published research report on the performance of silver perch fed on zooplankton in a controlled environment. Experiments were undertaken in this study to examine the proximate composition of sewage grown zooplankton and the performance *B. bidyanus* fed on two species of cladoceran zooplankton namely *Daphnia carinat*a and *Moina australiensis*.

MATERIALS AND METHODS

Source and production of zooplankton

Zooplankton grows abundantly in the last stage of sewage treatment process at the Werribee Sewage Treatment Lagoons (WST). The WSTL is located 35 km southwest of Melbourne, Australia. The zooplankton utilize algae from the ponds and remove nitrogen and phosphorus (pollutants) from the waste water. The sequence of zooplankton production at the WSTL is given in Figure 4.1. During 1995-96 (summer), the Zootech (a private company researching on zooplankton) used a special harvester "Baleen" and harvested 40-84 kg/hour of zooplankton from the lagoons. The harvested zooplankton were kept frozen at -20^{o}C in blocks until used.

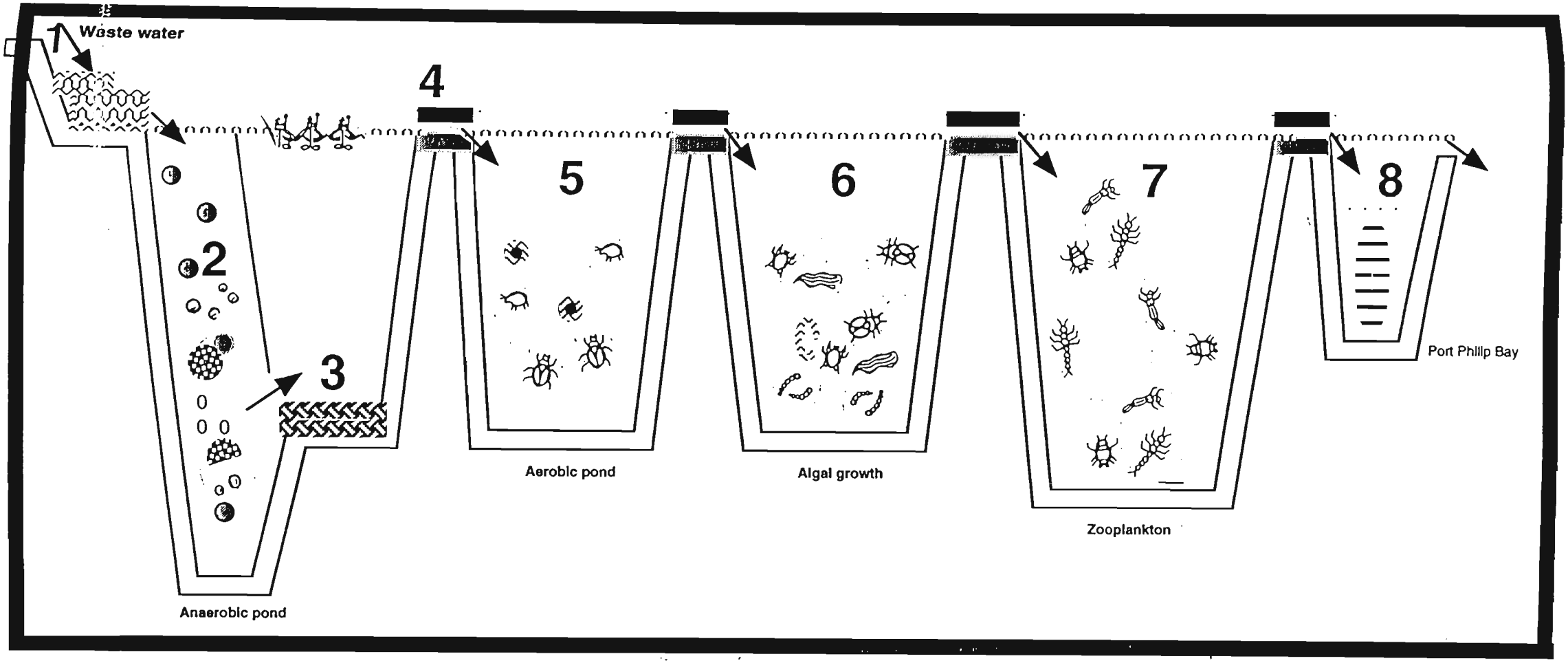

Figure 4.1. Sequence of zooplankton production at Werribee Sewage Treatment Lagoons.

1. Waste water (raw sewage) enters the aerobic ponds;
2. Bacteria digest the organic matter;
3. Sludge containing the heavy metals and other chemicals settle out;
4. Waste water moves into the aerobic ponds;
5. Algae grow in nutrient rich water;
6. Bacteria and algae remove nitrogen from water;
7. Zooplankton grow and feed on algae and bacteria;
8. Treated waste water along with zooplankton flows into Port Phillip Bay;

The dominant zooplankton at the WSTL are *Daphnia carinata* King, and *Moina australiensis* Saris (Lai 1994). This natural and invaluable resource is underutilized, and has been to date drained into the Port Philip Bay by the Melbourne Water Authority.

Fish husbandry and experimental procedures

The rearing experiments was conducted at the Zootech multipurpose hatchery located at the Werribee Treatment Complex. Silver perch (*B. bidyanus*) juveniles were bought from a local native fish hatchery. Fish were acclimatized in the hatchery environment for three weeks before the start of the experiment. Rectangular (dark coloured) P.V.C tanks (41 x 84 x 27 cm) were selected randomly for each of the seven feeding treatments (Table 4.1.). Each tank was stocked with 15 fish (av. weight 698 mg) with three replicates per treatment, or 45 fish per treatment.

Table 4.1. The different feeding regimes used in feeding silver perch with zooplankton.

Feeding regimes	Code
1 Control (commercial diet-2) (100%)	CON
2 *Daphnia carinata* (100%)	DAP
3 *Moina australiensis* (100%)	MOI
4 *D. carinata* (50%) and *M. australiensis* (50%)	DAP+MOI
5 *D. carinata* (33.33%) + *M. australiensis* (33.33%) + Commercial diet (33.33%)	DAP+MOI+CON
6 *D. carinata* (50%) + Control (50%)	DAP+CON
7 *M. australiensis* (50%) + Control (50%)	MOI+CON

The fish were reared at 25^{O}C, since in a previous study, the best growth of silver perch juveniles was obtained at this temperature (see chapter 3.). The water temperature of the tank was maintained at 25^{O}C using a thermostatically controlled heater. The water quality was maintained to a level optimum for fish growth (chapter 2.). The light regime was 12 h light and 12 h dark. Fish were fed on either frozen single zooplankton (*D. carinata* / or *M. australiensis*) or a combination of frozen *D. carinata* and *M. australiensis*. The 45% protein diet (commercial diet-2) was selected as control diet (see Table 2.1 in chapter 2 for proximate composition of the diet). Fish were fed @ 3%

of their body weight (dry matter). The calculated feed was divided into two portions and fed twice a day, six days a week for four weeks. The experimental design is given in Table 4.2.

Biochemical analysis

Fish and zooplankton samples were stored at -20^{o}C until analyzed. Triplicate samples of zooplankton (*D. carinata* and *M. australiensis*), and feed were subjected to chemical analysis (moisture, crude protein, fat, ash and phosphorus determination) (see chapter 2, section 2.1 for details of analytical methodology). At the completion of the experiment pooled fish samples were dried and ground using mortar and pestle for crude protein, fat, ash and phosphorus determination. Amino-acids of zooplankton was analyzed at the State Chemistry Laboratory, Melbourne.

Heavy metals analysis

The fish carcass was analyzed for the heavy metals (zinc, cadmium and lead) in order to ascertain the level of these contaminants which bioaccumulated as a result of feeding of sewage grown zooplankton. Samples were dried in an oven and crushed gently in a pestle and mortar. Dried samples were digested in analytical grade nitric acid (HNO_3) at 100^{o}C in a block heater (Corp and Morgan 1991). Metals were analyzed on a varian spectra AA-400 Flame Absorption Spectrophotometer with background correction using procedural blanks.

Handling of data and statistical procedures

Specific growth rate (SGR), food conversion ratio (FCR), protein efficiency ratio (PER) and apparent net protein utilization (ANPU) were calculated using appropriate formulae as described in chapter 2.6. All statistical analysis were performed following Zar (1984) as described in chapter 2.5.

Table 4.2. Experimental details of feeding trials conducted with zooplankton grown at sewage lagoons and fed to silver perch. Fish were reared at 25^{0}C for 28 days.

Feeding regime	Temperature (°C)	Tank size (cm)	No tanks	No fish per tank	No fish per treatment	Average initial size (mg)
1. Control (Silver perch diet) (100%)	25	41x84x27	3	15	45	696
2. *Daphnia* sp. (100%)	25	41x84x27	3	15	45	705
3. *Moina* sp. (100%)	25	41x84x27	3	15	45	691
4. *Daphnia* (50%) + *Moina* (50%)	25	41x84x27	3	15	45	703
5. *Daphnia* (33.3%) + *Moina* (33.3%) + control (33.3%)	25	41x84x27	3	15	45	698
6. *Daphnia* (50%) + Control (50%)	25	41x84x27	3	15	45	710
7. *Moina* (50%) + Control (50%)	25	41x84x27	3	15	45	687

RESULTS

Proximate composition of zooplankton

The proximate analysis of *Daphnia carinata* and *Moina australiensis* revealed that the sewage grown zooplankton had a very good biochemical composition (Table 4.3.). The protein content in *D. carinata* and *M. australiensis* were 54.34% and 64.80% respectively, which is higher than that of the control commercial silver perch diet. The lipid content were in the range of 7.29-7.73% (Table 4.3.). The percentage of phosphorus in the two species of zooplankton ranged from 1.11-1.14%.

Table 4.3. Proximate analysis of control diet and zooplankton (*Daphnia carinata* and *Moina australiensis)* harvested from the Werribee Sewage Treatment Lagoons.

	Dry diet (control)	*D. carinata*	*M. australiensis*
Crude protein (%)	45.00 ± 1.50	54.34 ± 2.83	64.80 ± 1.48
Crude fat (%)	08.50 ± 0.49	07.29 ± 0.03	07.73 ± 0.06
Crude ash (%)	09.84 ± 0.02	11.26 ± 0.01	06.82 ± 0.07
Carbohydrate (%)	36.66 ± 1.04	27.10 ±1.56	20.65 ± 0.97
Moisture (%)	09.88 ± 0.10	92.87 ± 0.09	93.72 ± 0.11
Nitrogen (%)	07.20 ±0.50	08.69 ± 0.31	10.37 ± 0.17
Phosphorus (%)	01.16 ± 0.08	01.14 ± 0.02	01.11 ± 0.01
Digestible energy (Kcal. kg^{-1})	5273	5250	5700
DE/P (g. $kcal^{-1}$)	117	97	88

[1]Values are mean±SE; n=3;
[2]Gross energy was calculated using kilocalorie values of 5.5/g protein, 9.1/g fat and 4.1/g carbohydrate (New 1987).

Amino-acids content in zooplankton

The amino-acid analysis reveals that both *D. carinata* and *M. australiensis* contained an appreciable amount of both essential and non-essential amino acids (Table 4.4.). Both species of zooplankton appear to be slightly superior in amino acid composition to commercial silver perch diet (control) although there was no statistical differences. The most limiting amino acids in feeds are lysine and methionine (New 1987); these two amino acids were present in higher concentration compared with levels in the control diet although the difference was not significant. The lysine content in *D. carinata* and *M. australiensis* were 3.35% and 3.34%, while control diet contained 2.65%.

Table 4.4. Essential and non-essential amino acid composition of commercial diet-2 (control) and zooplankton harvested from the Sewage Treatment Lagoons and used in feeding trials ($g.100^{-g}$).

	Commercial diet	*D. carinata*	*M. australiensis*
Essential-amino acids			
Arginine (Arg)	2.42	3.37	3.01
Histidine (His)	1.29	1.65	1.37
Isoleucine (Iso)	1.66	2.60	2.67
Leucine (Leu)	3.25	5.22	4.94
Lysine (Lys)	2.65	3.35	3.34
Methionine (Met)	0.80	1.46	1.13
Phenylalanine (Phe)	1.86	2.52	2.67
Threonine (Thr)	1.68	2.86	2.79
Tryptophan (Try)	0.43	0.71	0.76
Valine (Val)	2.36	3.27	3.56
Non-essential amino acids			
Alanine (Ala)	2.27	3.23	3.77
Asparatic acid (Asp)	2.42	4.70	5.12
Cystine (Cys)	0.44	0.71	0.60
Glutamine acid (Glue)	6.36	6.39	6.57
Glycine (Gly)	2.03	2.48	2.59
Proline (Pro)	2.03	2.20	2.36
Serine (Ser)	1.88	2.82	2.62
Tyrosine (Tyr)	1.17	2.14	2.38

Performance of silver perch fed on zooplankton

Silver perch fed on the control diet (treatment-1) and on *D. carinata* (treatment-2) resulted in a higher specific growth rate ($P<0.05$) compared with the SGR of fish from among the seven treatments (Table 4.5.). The gain in weight resulting from feeding the control diet (treatment-1) and *D. carinata* (treatment-2) was not significantly different ($P>0.05$). Fish fed on either *M. australiensis* or a combination of *M. australiensis* showed comparatively less SGR (treatment-3, 4, 5, 7) compared with the control or *D. carinata* diets. The food conversion ratio (FCR) and protein efficiency ratio (PER) were significantly higher in fish on control and *D. carinata* diets compared to fish fed on other treatments (Table 4.5.). Similarly, ANPU was also significantly better with control and *D. carinata* diets compared with other treatments ($P<0.05$).

Table 4.5. Mean performance of silver perch juveniles fed on either single zooplankton or a mixed zooplankton or a combination of zooplankton plus commercial diet. The fish were reared at 25°C for 28 days and fed @ 3% of body weight, twice a day, six days a week.

	CON[1]	DAP[2]	MOI[3]	DAP+MOI	DAP+MOI+CON	DAP+CON	MOI+CON
	100%	100%	100%	50% + 50%	33.3%+33.3%+ 33.3%	50%+50%	50%+50%
	1	2	3	4	5	6	7
Initial av. weight (mg)	696	705	691	703	698	710	687
Final av.weight (mg)	1668	1628	1410	1371	1438	1548	1380
Av. gain in weight (mg)	973[a]	923[a]	719[c]	668[c]	740[c]	838[b]	693[c]
Percentage weight gain (%)	140	131	104	95	106	118	101
SGR[4]	3.64±0.04[a]	3.45±0.16[a]	2.97±0.0[c]	2.78±0.05[c]	3.02±0.11[c]	3.25±0.04[b]	2.91±0.02[c]
SGR as % of control	100.0	94.7	84.9	76.4	82.3	89.3	79.9
FCR[5]	1.14±0.07[a]	1.26±0.12[a]	1.66±0.04[c]	2.15±0.47[c]	1.56±0.09[b]	1.34±0.12[b]	1.94±0.13[c]
PER[6]	2.10±0.14[a]	1.89±0.11[a]	1.32±0.09[c]	1.17±0.11[d]	1.47±0.06[c]	1.7±0.02[b]	1.34±0.05[c]
ANPU[7]	57.04±2.1[a]	50.00±4.0[a]	35.50±3.10[c]	27.6±2.13[d]	38.4±0.48[c]	43.86±1.77[b]	31.5±0.91[c]

[1]CON = control (commercial diet-2).
[2]DAP = *Daphnia carinata*
[3]MOI = *Moina australiensis*
[4]SGR = Specific growth rate = ln final weight - ln initial weight/ total days of experiment (Laird and Needham 1988).
[5]FCR = Food conversion ratio = weight of feed fed/ wet weight gain (Laird & Needham 1988).
[6]PER = Protein efficiency ratio = g wet weight gain/g crude protein fed (EIFAC 1980; De Silva & Anderson 1995).
[7]ANPU = Apparent net protein utilization = body nitrogen at the end of test - body nitrogen at the beginning of test/amount of nitrogen consumed (Jauncey 1982).
[8]Values are mean±SE; n=3.
[9]Values in the same with row with common superscripts are not significantly different from each other (P>0.05).

Effect of different feeding regimes on the chemical composition of fish

The carcass composition of fish at the end of experiments is shown in Table 4.6. In general, the moisture level was reduced whilst protein, lipid and ash content had increased compared to the initial values in all treatments. Fish fed on the control diet had higher protein and lipids content than that of other zooplankton based treatments, whereas fish fed on zooplankton had higher ash and phosphorus content ($P<0.05$) compared with the control diet (Table 4.6). Fish received *M. Australianises* based treatment had a lower protein and lipid content compared to *D. carinata* based treatments.

Carcass analysis for heavy metals contaminants

The carcass analysis revealed a lower level of zinc, cadmium and lead in carcass fed on sewage zooplankton than the concentration recommended in fish as food by the National Health and Medical Research Council (Table 4.7.). Fish fed on mixed zooplankton (treatment-4) had a higher level of zinc than that of other treatments ($P<0.05$). Cadmium levels was negligible and there was no significant difference among treatments ($P>0.05$).

DISCUSSION

The proximate analysis of zooplankton harvested from the Werribee sewage treatment lagoons (WSTL) revealed a very good biochemical composition for a fish food source. The protein levels in *D. carinata* and *M. australiensis* were 54.34% and 64.80% respectively, which are close to that of other cladocerans (see Table 1.2.1, in chapter 1.2 on biochemical composition of zooplankton). Previous studies have reported protein values in the range of 49.70% to 70% for *Daphnia* sp. (Blazka 1966; Yurokowski and Tabachek 1979; Watanabe *et al.* 1983) and between 59.00% and 77.85% for *Moina* sp. (Tay *et al.* 1991; Creswell 1993). Indeed, the relative amount of protein varies greatly for freshwater zooplankton (Vijverberg and Frank 1976). Both

Table 4.6. Effect of zooplankton based feeding regimes on the chemical composition of silver perch (moisture, protein, lipid, ash and phosphorus). The fish were reared at 25°C for 28 days and fed @ 3% body weight, twice a day, six days a week.

Feeding regimes	Moisture (%)	Crude protein (%)	Crude lipid (%)	Ash (%)	Phosphorus (%)
1. Control (commercial diet-2) (100%)	75.97±0.61[a]	65.21±0.18[a]	10.72± 0.09[a]	18.50±0.09[a]	2.56±0.16[a]
2. Daphnia (100%)	76.33±0.19[b]	65.01±0.19[a]	09.92±0.03[b]	20.05±0.03[b]	3.04±.0.12[b]
3. Moina (100%)	78.12±0.24[c]	62.43±0.28[c]	09.16±0.12[b]	21.40±0.06[b]	2.95±0.05[b]
4. Daphnia+Moina (50%+50%)	79.02±0.34[c]	62.03±0.03[c]	09.06±0.14[c]	24.34± 0.03[c]	3.45±0.27[c]
5. Daphnia+Moina+Control (33.33%+33.33%+33.33%)	78.02±0.09[c]	64.72±0.03[b]	09.63±0.08[b]	22.56±0.05[b]	3.05±0.01[b]
6. Daphnia+Control (50%+50%)	77.54±0.25[b]	64.82±0.04[b]	10.23±0.11[b]	18.62±0.04[b]	2.69±0.04[b]
7. Moina+Control (50%+50%)	78.50±0.29[c]	62.40±0.02[c]	09.57±0.11[b]	18.86±0.01[b]	2.80±0.11[b]

[1]Body composition at the beginning of the experiment was moisture, 79.52, protein 61.95, lipid 9.07%, ash 17.63%, phosphorus 2.51%.
[2]Values are mean±SE; n=3.
[3]Values in the same column with common superscripts are not significantly different from each other (P>0.05).

Table 4.7. Metal concentrations (μg g^{-1} dry weight) in the carcass of silver perch reared for 28 days and fed on zooplankton and zooplankton plus commercial diet. The zooplankton was harvested from the Werribee sewage treatment lagoons.

Feeding regimes	Zinc (Zn)	Cadmium (Cd)	Lead (Pb)
1. Control (commercial diet-2) (100%)	207.6±15.43[a]	3.77±0.06[a]	Not detected (ND)
2. Daphnia (100%)	226.37±8.05[a]	3.91±0.03[a]	ND
3. Moina (100%)	214.84±15.96[a]	3.91±0.02[a]	ND
4. Daphnia+Moina[1,2] (50%+50%)	287.27±8.10[b]	3.96±0.06[a]	ND
5. Daphnia+Moina+Control (33.33%+33.33%+33.33%)	278.54±8.09[a]	3.84±0.79[a]	ND
6. Daphnia+Control (50%+50%)	233.69±0.00[a]	3.85±0.02[a]	ND
7. Moina+Control (50%+50%)	238.69±7.64[a]	3.82±0.76[a]	ND
Maximum permitted conc. in fish as food (mg kg^{-1})[1]	750.00	1.00	12.5

[1]National Health and Medical Research Council, Australia, Standard A 12, Metals and contaminants in food. Commonwealth of Australia, Gazette no. P 27. 1987.
[2]Values are mean±SE; n=3.
[3]Values in the same column with common superscripts are not significantly different from each other(P>0.05).

D. carinata and *M. australiensis* are the dominant species at the WSTL and could be a valuable source of protein for use in aquaculture. The higher protein levels observed (Table 4.3) in zooplankton may be linked to the nutrient rich sewage waters of WSTL. Vijiverberg and Frank (1976) reported a positive correlation between the nitrate and ammonium concentration in the water and the nitrogen content of zooplankton.

Watanabe *et al.* (1983) analyzed various species of zooplankton, and reported 13% lipid levels in *Daphnia* sp., and in the range of 12-27% for *Moina* sp.. The lipid content of both species of zooplankton of WSTL appears to be slightly lower (7.29-7.73%) compared to other species of zooplankton (see Table 1.2.1. in chapter 1.2). The biochemical composition of freshwater zooplankton is known to be extremely variable (Vijverberg and Frank 1976) and depends on the food eaten by zooplankton (Proulx and Noue 1985). The fatty acids composition of *D. carinata* and *M. australiensis*, was not analyzed in this study. However, Nichols *et al.* (1993) reported appreciable levels of both EPA (eicosapentaenoic acid) and DHA (docosahexaenoic acid) of zooplankton harvested from WSTL. Tucker (1992) noted that *Daphnia* sp. could be deficient in DHA and *Moina* sp. deficient in both DHA and EPA. In general, the fatty acids composition of zooplankton are influenced by the fatty acids composition of their diet (Watanabe *et al.* 1983; Proulx and Noue 1985) and related to the seasonal succession of phytoplankton species in water bodies (Jeffries 1970). It appears therefore, that the nutritional value of zooplankton may vary with the season.

Phosphorus is an important element for growth and food conversion of fish and dietary phosphorus is the main source for fish (Lall 1991). Proximate analysis showed that both species of zooplankton harvested from the WSTL contained a desirable level of phosphorus (1.11-1.14%) (Table 4.3.). Creswell (1993) and Watanabe *et al.* (1983) reported 1.40-1.46% phosphorus in *Daphnia* sp. and 1.1-1.41% in *Moina* sp. Khan and Qayyum (1971) found that zooplankton grown in sewage or waste waters may

contain higher phosphorus and that this fact may be related to the efficient nitrogen and phosphorus removal capacity by zooplankton in wastewaters (Kawasaki *et al.* 1992). The WSTL is also nutritionally enriched, therefore *D. carinata* and *M. australiensis* harvested from that site may have caused the higher nitrogen and phosphorus content.

Both *D. carinata* and *M. australiensis* revealed a well balanced composition of amino-acids which match the essential amino acids requirements of fish reported by Tacon (1990) and Creswell (1993). However, concentrations of some of the essential amino acids in *D. carinata* and *M. australiensis* harvested from WSTL are lower than that of values reported by Yurkowski and Tabachek (1979), Hepher (1988) on *Daphnia* sp. and by Hepher (1988), Creswell (1993) on *Moina* sp. The lower values could be a result of seasonal variations (Holm and Walther 1988) or the foods eaten by the zooplankton (Watanabe *et al.* 1983) or the growing medium (Allen and Allen 1981) (see Table 1.2.2 in chapter 1.2 for more information on amino-acids content of other zooplankton). Other researchers have found that the essential amino-acids content of zooplankton can meet the essential-amino acids requirement of fish, for example, essential-amino acids content in *Daphnia pulex* was found to be equal or greater than the requirement of chinook salmon (Yurkowski and Tabachek 1979). Both *D. carinata* and *M. australiensis* have a relatively similar composition of essential and non-essential amino acids (average 51.5% essential amino acids). Holm and Walther (1988) reported that 50.6% of amino acids in daphnids were essential amino acids. The cladocerans, *D. carinata* and *M. australiensis* were found to contain a relatively similar proportion of polar to non-polar amino acids, viz, 0.89 in *D. carinata* and 0.80 in *M. australiensis*. A higher proportion of polar to non-polar amino acids (1.57) was calculated for other species of daphnids by Holm and Walthier (1988) which could represent the better nutritional status of those species.

The performance results obtained (Table 4.5.) indicate that silver perch may have a preference for *D. carinata* over *M. australiensis* although further experiments would be necessary to confirm this hypothesis. The specific growth rate (SGR), food conversion ratio (FCR), protein efficiency ratio (PER) and Apparent net protein utilization (APNU) were significantly higher in fish fed on *D. carinata* (treatment-2) than in other zooplankton based treatments ($P<0.05$) (Table 4.5.). Fish that received *M. australiensis* based feeding treatments had significantly lower SGR, FCR, PER and ANPU compared to results obtained from feeding of fish of *D. carinata*. The SGR, FCR. PER of fish fed the control diet and *D. carinata* were similar ($P>0.05$). Jones (1995a, 1995b) observed a faster growth in crayfish (*Cherax albidus*) fed on *Daphnia* sp. than in those fed dry pellets. Similarly, carp and Atlantic salmon fed with zooplankton performed better than those fed formulated diets (Holm and Moller 1984; Kamler *et al.* 1992). Herring and trout offered frozen copepods assimilated more than 90% of the dry matter in zooplankton (Sargent *et al.* 1979). Fish fed on frozen zooplankton were found to be healthier (Sargent *et al.* 1979), resulting in good growth and survival in aquaculture (Dabrowski *et al.* 1984; Fermin and Boliver 1994). Confer and Lake (1987) established that yellow perch (*Perca flavescens*) have a higher affinity for *Diaptomus sicilis* than other species of zooplankton. Similarly, Atlantic salmon (*Salmo salar*) showed a preference for daphnids (Holm and Walther 1988). This current study indicates that silver perch has a higher affinity for *D. carinata* over *M. australiensis* and performed equally to commercial diet. Furthermore, the species and their origin (Watanabe *et al.* 1978), and seasonal changes in the biochemical composition in zooplankton could influence their nutritional value to fishes (Sadykhov *et al.* 1975; Jeffries 1979). Therefore further research on seasonal biochemical changes of zooplankton of WSTL would provide important information on the appropriate time of harvesting of zooplankton for use in aquaculture.

The maximum PER values obtained with *D. carinata* (treatment-2) was 1.89. Trout fed on krill zooplankton (*Euphausia* sp.) had a PER of 1.92 (Koops *et al.* 1979)

whereas with channel catfish it was 1.03 (Hilge 1979). Watanabe *et al.* (1983) fed rainbow trout with *Daphnia* sp. and *Moina* sp. and obtained a higher PER of 3.9 with *Daphnia* sp. and 2.6 with *Moina* sp.. These zooplankton were raised by artificially feeding nutritionally enriched foods which might have resulted in the higher PER. It is noteworthy that silver perch fed sewage grown *D. carinata* showed encouraging performance results as demonstrated by the significantly higher SGR, FCR, PER, APNU values over *M. australiensis* but these results are not significantly different from those obtained with fish reared on the control commercial silver perch diet (Table 4.5.). It has been reported that growth rate and feed efficiency were higher when trout were fed on Krillmeal than fishmeal (Koops *et al.* 1979). Similarly, carp had a significantly higher SGR when fed with zooplankton than carp fed formulated diets (Kamler *et al.* 1992). All performance indicators such as SGR, FCR, PER and ANPU were better when carp fed on 50% Krillmeal suggesting that part of fish meal can be replaced with zooplankton (Lukowicz 1979). Future research should be conducted on part or full replacement of expensive fishmeal with zooplankton meal for rearing of silver perch juveniles.

Rainbow trout fed on zooplankton, *Euphausia pacifica* showed a higher ash and lower fat content than those fed commercial diets (Spenelli 1979). Similarly, silver perch fed on zooplankton had significantly higher ash, and phosphorus and lower fat content in the present study (Table 4.6.). Koops *et al.* (1979) also found a higher phosphorus content in fish fed on krillmeal (a zooplankton from the Antarctica) than fishmeal. Carp were found to have accumulated more protein and lipid when fed on krillmeal than on artificial diet (Pffener and Meske 1978). Koops *et al* (1979) found that trout fed on krillmeal had a higher retention of phosphorus than when fed on fishmeal based diet. Therefore, the results of present study are comparable with previous studies conducted by Spenelli (1979), and Koops *et al.* (1979).

Accurate data on the potential contaminants of zooplankton are of paramount important when fish were reared on sewage grown zooplankton as there could be some concern regarding the potential bioaccumulation of heavy metals in fish. However, silver perch fed on either *D. carinata* sp. or *M. australiensis* did not show higher zinc, cadmium and lead levels in the carcass in most treatments compared to control diets (Table 4.7.). Lukowicz (1979) reported a lower level of cadmium, lead, arsenic and mercury in carp fed on krillmeal. The results on the concentration of heavy metals demonstrate that zinc, cadmium and lead levels in the silver perch carcass are within the safe range as set out by the National Health and Medical Council of Australia (1987) for human consumption. This preliminary data may indicate that zooplankton from the Werribee Sewage Treatment Lagoons may be safe in the culture of fish and other aquarium species though further possibly seasonal investigation would be essential.

It is concluded that zooplankton from the Werribee Sewage Treatment Lagoons (WSTL) are rich in protein, essential amino acids, lipid, and phosphorus content. Moreover, silver perch performed significantly better with *Daphnia carinata* compared with *Moina australiensis* ($P<0.05$). The specific growth rate (SGR), food conversion ratio (FCR), and protein efficiency ratio (PER) were not significantly different compared to fish fed on control commercial silver perch diet and *D. carinata* (Table 4.5.). This may indicate that zooplankton could be an alternative diet for silver perch.

Chapter 5

EFFECT OF DIET AND TEMPERATURE ON SOLID WASTE PRODUCTION IN THE AQUACULTURE OF SILVER PERCH

5

EFFECT OF DIET AND TEMPERATURE ON SOLID WASTE PRODUCTION IN THE AQUACULTURE OF SILVER PERCH

INTRODUCTION

Solid waste is defined here as waste feed (uneaten food and dust), and faeces containing mucus, discarded intestinal cells, and bacteria that may generate from feeding and metabolic activities of fish (Beveridge *et al.* 1991; Beveridge and Phillips 1993; Costa-Pierce 1996). The concentration of suspended solids derived from fish farms is an important and basic measure of effluent quality (Cripps and Kelly 1996). Knowledge of solid waste production is essential to control possible aquatic pollution (Merican and Phillips 1985) and to design proper guidelines for an ecological sustainable aquaculture programme (Cho *et al.* 1991).

Production of solid waste from aquaculture varies between different systems of rearing and fish species (Albaster 1982; Beveridge 1984), for example, waste loss could be substantial from cage aquaculture amounting about 5-40% of food fed (Merican and Phillips 1985; Phillips and Beveridge 1986). Faecal output from the culture of salmonids, carp, and shrimps is reported to be 26-27% of ingested feed whereas for catfish it is 41% (Beveridge *et al.* 1991). Solid waste contains nutrients (nitrogen and phosphorus) (Bergheim *et al.* 1993; Cripps 1995) which may be released when environmental conditions, such as, pH, or temperature (Boersen and Westers 1986; Pontius 1990; see also chapter 8) are appropriate and thereby cause eutrophication or water pollution of the receiving waters (Kelly 1993; Ackefors and Enell 1994). The amount of solid waste load in receiving waters is influenced by the type of diet fed to the farmed species (Warren-Hansen 1982) and could be higher with moist diets than with extruded diets (Stewart 1997). Among other factors, feeding

rates (Liao 1970; Butz and Vens-Cappell 1982; Merican and Phillips 1985), the digestibility of diets, food conversion ratio, pellet size (Kilambi *et al.* 1976; Merican and Phillips 1985), process technology (e.g. extruded diets), feeding practice (Blyth and Burser 1993), the level of fat in the diet (Eikebrokk *et al.* 1991; Bergheim and Asgard 1996) and rearing temperatures (Kilambi *et al.* 1976) may determine in the quantity of solid output.

Until now there have been no attempt to study the amount or types of waste entering the environment from the rearing of silver perch or other species in Australia (Kibria, *et al.* 1997a). This is important since a review of literature clearly demonstrates that interest and investment in silver perch farming is increasing and silver perch is a major native aquaculture industry in the country (Kibria *et al.* 1996a). Moreover, research in Europe and North America demonstrates a strong correlation between water pollution and the loading of solid waste from fish farms (see chapter 1.3, solid waste production). This chapter describes the first in a series of planned experiments to study waste production from rearing of the silver perch. The objectives of present rearing trials were to compare the amount of suspended and dissolved solid waste load from feeding and to quantify solid waste production by *B. bidyanus* at different rearing temperatures.

MATERIALS AND METHODS

The experiment was conducted in the wet laboratory at the Victoria University of Technology. Fish for experiments were purchased from a local native fish farm and were acclimatized in the laboratory in large holding tanks for four weeks before use in experiments (see chapter 2). Two experiments were conducted to study the production of solid waste. The first preliminary experiment was conducted to evaluate the effects of feeding on the production of suspended and dissolved solids. In this study, fish were

reared at room temperature (18-20^{o}C) and fed two diets (diet-1 and diet-2) and the resulting solid waste load was quantified without separating uneaten food from faeces. The second experiment was conducted to observe the effect of three temperatures (20^{o}C, 25^{o}C and 30^{o}C) on the quantity of uneaten food and the production of faecal waste. In the second experiment, silver perch were fed three diets (diet-1, diet-2 diet-3) (details on the proximate composition of the three diets are given in Table 2.1, in chapter 2.). In both experiments, fish were fed on silver perch crumbles/starter @ 3% of their body weight. The calculated feed was divided into two portions and fed twice a day, six days a week for four weeks. Fish were acclimatized and water quality in aquaria was maintained similarly as mentioned in chapter 2.

Preliminary experiment 1 : Effects of feeding on the production of suspended and dissolved solids

In this experiment, individual fish (av. weight 2,050 mg) were kept in separate aquaria (30 x 16 x17 cm) and fed on two commercial diets, diet-1 and diet-2. Ten aquaria were allocated per diet to study the load of suspended and dissolved solids produced over a four week period. Each aquarium was siphoned once every morning using a 5 mm hose to collect both uneaten food and faeces. Collected solids were filtered first through a fast filter (Whatman 512) and later through a standard 0.45µm glass fibre filter (APHA 1989). The residue retained on both filters was dried in an oven at 100^{o}C for 1 hour, cooled in a desiccator and weighed using an analytical balance. The increase in weight was defined to be the total suspended solids and is expressed as suspended solids/fish. The filtrate from the total suspended solids was used for total dissolved solids determination following APHA guidelines (APHA 1989). An aliquot of the filtered sample was dried to constant weight in an oven at 180^{o}C, cooled in a desiccator and weighed (APHA 1989). The increase in dish weight was considered as representative of the total solids (APHA 1989). Unfed fish (control) were kept in separate aquaria to compare suspended and dissolved solid production compared to fed fish.

The total dissolved solids was calculated using the following formula :

Total dissolved solids (mg/l) $\frac{(A - B - C) \times 1000}{\text{sample volume, ml}}$ (APHA 1989; Adams 1990)

where :
A = weight of dry residue + beaker, (mg);
B = weight of beaker, mg;
C = blank (final weight of blank beaker - initial weight of blank beaker)

Experiment 2 : Effects of temperature on the production of uneaten food and faeces

Silver perch juveniles (mean weight 688 mg) were reared in aquaria (30 x 16 x17 cm) for four weeks to study the effect of temperature on solid waste production at 20^{o}C, 25^{o}C and 30^{o}C and fed on three commercial diets referred to as diet-1, diet-2 and diet-3 (see Table 2.1 for proximate composition). Fish were stocked at the rate of four per aquarium, and six aquarium were allocated for testing the effect of each diet at each temperature, i.e., 24 fish per diet per temperature. Fish were fed at the rate of 3% of their body weight. The calculated feed was divided into two portions and fed twice a day (0900 and 1400), six days a week for four weeks.

Aquaria were siphoned with a 5mm hose each morning and each evening to collect accumulated faecal pellets, and before and after each feed in order to collect and separate fractions of faeces and uneaten food. Collected faeces and food were centrifuged (Econospin-Sorvall Instruments) at 2000 r.p.m. for 10 minutes to separate solids from the liquid (Law 1984; Hajen *et al.* 1993). The supernatant was discarded and solids were dried in an oven overnight at 100^{o}C. Dried samples were pooled to determine the amount of solid waste (uneaten food and faeces) per diet and per temperature since sample quantity was small.

RESULTS

Preliminary experiment at room temperatures

In the preliminary experiment with two diets, diet-2 resulted in slightly higher growth and feed conversion than diet-1 although the results were not significantly different ($P>0.05$). Marginally less suspended and dissolved solid waste was produced by fish fed diet-2 (Figure 5.1. and Figure 5.2). Suspended solid waste production from diet-2 and diet-1 was 350 mg · $fish^{-1}$ and 400 mg · $fish^{-1}$, respectively but the results were not significantly different ($P>0.05$). The dissolved solids produced were 1,647 mg · l^{-1} with diet-2, and 1,711 mg · l^{-1} with diet-1. Unfed fish (control) produced the least amount of suspended and dissolved solids and this was significantly different from that produced by the fed fish ($P<0.05$) (Figure 5.1, 5.2).

Solid waste (uneaten food and faeces) production at 20°C, 25°C and 30°C

The solid waste produced at the three rearing temperatures (20°C, 25°C and 30°C) is shown in Table 5.1. The results demonstrate that total solid waste production (uneaten food plus faeces) in silver perch was significantly higher ($P<0.05$) when fish were reared at 20°C compared with the waste from fish reared at 25°C. At 20°C, the dominant waste was uneaten food whereas at 25°C all food offered was consumed by the fish. At 30°C a small fraction of food (5%) remained uneaten. The faecal production was in the order of 30°C>25°C>20°C and similarly a higher faecal nitrogen and faecal phosphorus loss was also observed at 30°C (see chapter 6 and 7). The estimated solid waste produced at 20°C, 25°C and 30°C were 549 kg · $tonne^{-1}$, 284 kg · $tonne^{-1}$ and 393 kg · $tonne^{-1}$ fish production respectively ($P<0.05$). Diet-2 fed to fish resulted less solid waste compared to diet-1 and diet-3 at all temperatures though the difference of fish fed the three diets was not significant ($P>0.05$) (Table 5.1).

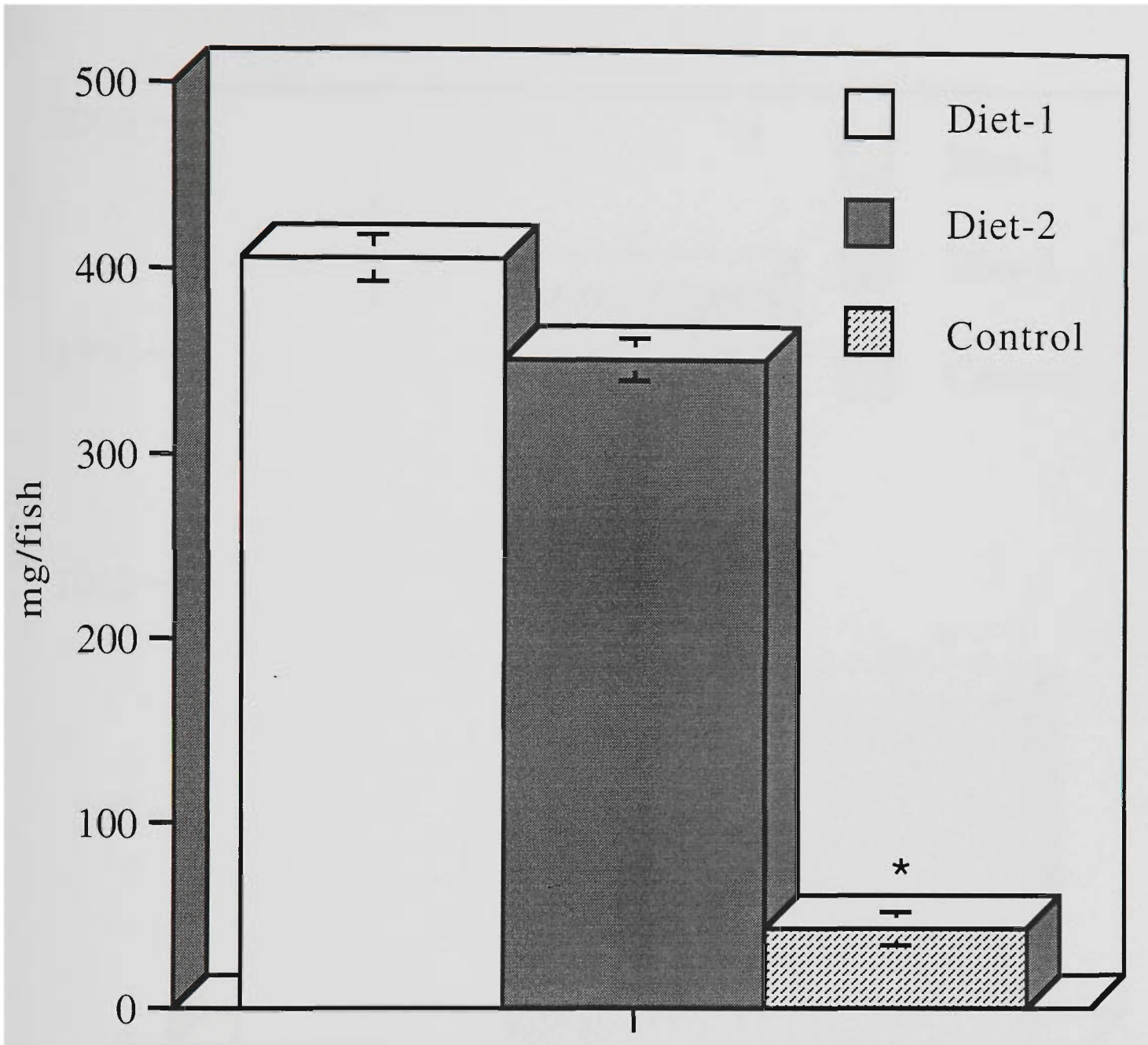

Figure 5.1. Suspended solids produced by silver perch juveniles (2.00-2.13g) fed two diets in preliminary experiment and reared for 28 days at 18-20 degree celcius. The control group was unfed. Suspended solids production was significantly lower with control group ($P<0.05$) (mean$\pm$SE; n=10).

* denotes significantly different

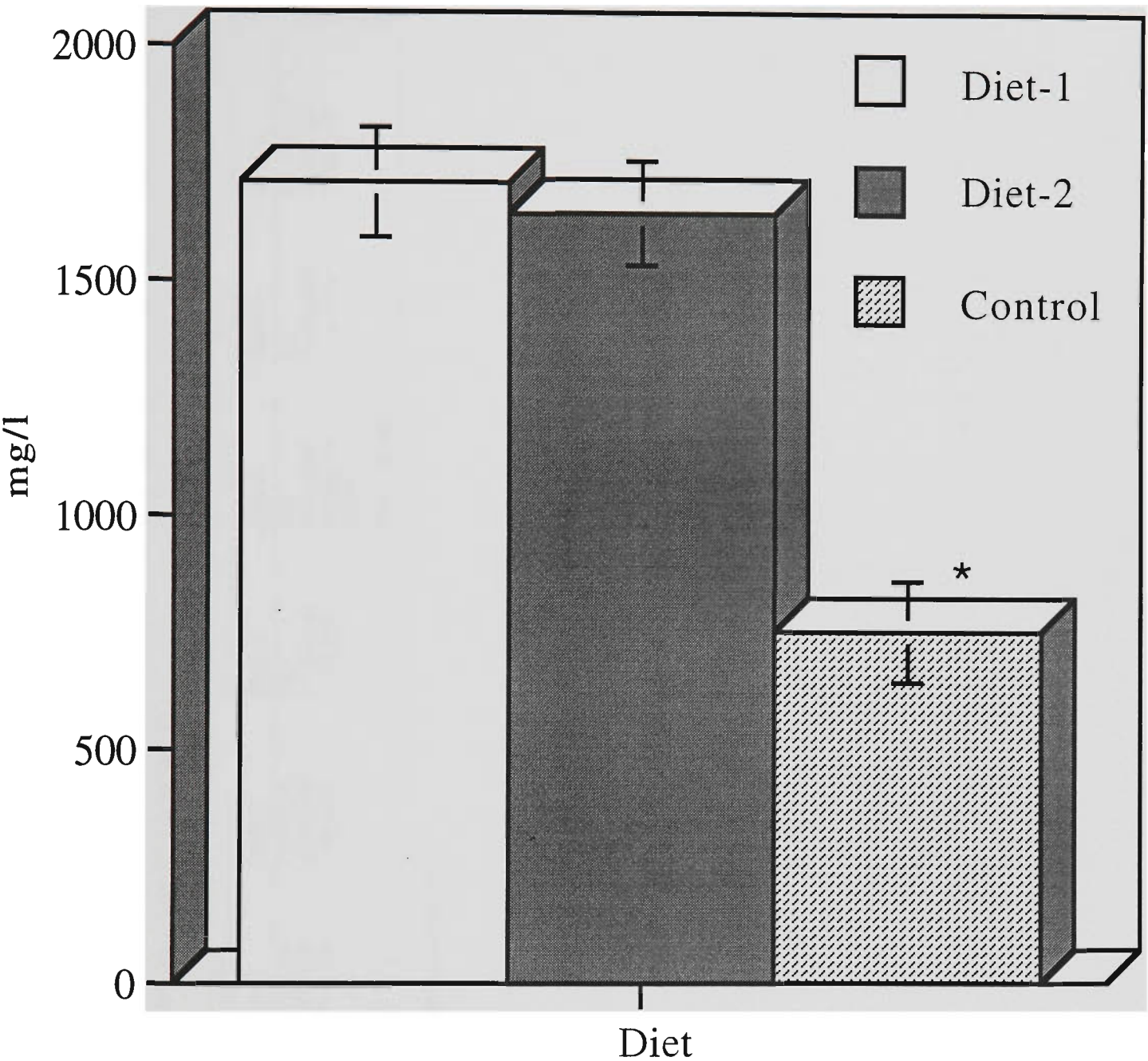

Figure 5.2. Dissolved solids produced by silver perch juveniles (2.00-2.13g) fed two diets in preliminary experiment and reared for 28 days at 18-20 degree celcius. The control group was unfed. Dissolved solids production was significantly lower with control group (mean±SE; n=10).

* denotes significantly different

Table 5.1. Showing fraction of solid waste (uneaten food and faeces) produced from rearing of silver perch at 20°C, 25°C and 30°C for 28 days. Fish were fed on three diets @ 3% of body weigh twice a day and six days a week. Estimation was made from pooled dried samples (as fed basis).

	[----------20°C----------]			[----------25°C----------]			[----------30°C----------]		
	Diet-1	Diet-2	Diet-3	Diet-1	Diet-2	Diet-3	Diet-1	Diet-2	Diet-3
A. Uneaten food (% feed fed)	27.65±1.3[a]	21.51±0.95[a]	26.50±1.4[a]	0.00	0.00	0.00	0.4.50±0.67[b]	0.3.60±0.35[b]	0.6.80±0.84[c]
B. Faeces (% feed fed)	07.80±0.87[a]	07.48±0.54[a]	12.28±0.89[b]	17.60±0.96[c]	13.60±0.67[c]	22.20±0.76[d]	19.40±0.82[e]	17.80±0.65[e]	22.50±0.74[e]
C. Total solid waste (A+B) (% of feed fed)	35.45±2.2[a]	29.00±1.49[a]	38.80±2.3[a]	17.60±0.96[b]	13.60±0.67[b]	22.20±0.76[b]	23.90±1.5[c]	21.40±1.0[c]	29.30±1.6[c]
D. Mean Total solid waste (% of feed fed)		34.41[a]			17.7[b]			24.87[c]	
E. Mean solid waste (kg/tonne fish production)		549±46.00[a]			284± 38.50[b]			393±35.80[c]	

[1]Values are mean±SE.

[2]Values in the same row with common superscripts are not significantly different from each other (P>0.05).

DISCUSSION

The present trials measured the production of solid wastes from silver perch juveniles reared in glass aquaria. In the preliminary experiment, diet-2 produced less suspended and dissolved solid waste (Figure 5.1, 5.2) in comparison to fish fed on diet-1 although this difference was not significant ($P>0.05$). Lall (1991) reported that ingredients containing a high concentration of fibre, chitin and indigestible carbohydrate may increase the excretion of suspended solids. It has been reported by Asgard *et al.* (1986) that even in balanced feed about 15-20% of eaten food is indigestible. The preliminary experiment was carried out at lower temperatures (18-20^{o}C) and at low temperature fish must be less active resulting in poor food intake and consequently increased more solid waste production. Rearing of silver perch at a higher growth temperature could be a means of reducing solid waste discharge into the environment. The next experiments was therefore conducted at higher than ambient temperatures, since optimum temperature for silver perch is reported to be in between 20^{o}C and 30^{o}C (Barlow 1986) and results in chapter 3. supports Barlow's results as higher growth was achieved at a temperature above of 20^{o}C.

In the second experiment, silver perch showed significantly higher solid waste load at 20^{o}C compared with the waste load at 25^{o}C and at 30^{o}C (<0.05). At 20^{o}C, a substantial amount of food (25% of food) offered was uneaten resulting in a higher solid waste load to the environment. This may be related to lower feed intake at 20^{o}C compared with the feed intake at 25^{o}C and 30^{o}C. At 25^{o}C, all food offered was eaten by the fish while at 30^{o}C only a small amount of food was uneaten. Thus supporting the view that the optimum growing temperature of silver perch is around 25^{o}C. The feed loss from salmon and trout culture is reported to be 1-30% of food fed, which depends upon the system of culture, type of feed fed and other management practice

(Beveridge *et al.* 1991). It is further reported that feed losses as uneaten food is much higher in cages than in tanks or ponds (Beveridge 1984; Merican and Phillips 1985).

Faecal production was found to be higher at 30°C compared to 25°C, this could be due to better digestibility at 25°C. The rate of digestion in fish is known to vary with temperature (National Academy of Sciences 1977; Webb 1978) and in particular, nutrient in feed are more digestible up to a certain temperature (Stickney and Lovell 1977). Cho and Slinger (1978) showed that the digestibility coefficient did not improve with increasing temperature greater than 15°C for rainbow trout. Interestingly, digestibility of total nutrients by common carp increased with increasing temperature, but beyond the optimum temperature, digestibility of protein remain unchanged or decreased (Shcherbina and Kazlauskene 1971). The typical faecal production in carnivorous fish is 250 to 259 g · kg^{-1} food fed (dry weight) and higher in omnivorous/herbivorous species (Butz and Vens-Cappell 1982; Beveridge and Phillips 1993). The faecal production in the current study was 136-222 g · kg^{-1} food fed and 178-225 g · kg^{-1} food fed at 25°C and 30°C respectively (Table 5.1.). The restricted feeding regime adopted in this experiment may have resulted in a lower faecal production of silver perch. The faecal production in fish was reported to increase at higher feeding rates (Butz and Vens-Cappell 1982). It is also reported that the quantity of uneaten food, and faecal production could vary with species, type of food fed (trash, moist and dry) and may be influenced by body size and season of culture (Beveridge and Phillips 1993). Therefore, small silver perch juveniles and dry feed fed to fish may be other reason of lower faecal production obtained in the current study.

The main solid output found in this study consists of uneaten food and faecal pellets, which is similar to results of previous researchers (Kendra 1991; Kaushik 1992; Pillay 1992). The maximum solid waste production obtained with silver perch at 20°C was 549 kg · $tonne^{-1}$ and the minimum 284 kg · $tonne^{-1}$ at 25°C ($P<0.05$) in the current

study. Though solid waste production from rearing of fish ranges widely from 289 to 2153 kg · tonne^{-1} (NCC 1990), the more reliable estimate appears to be 510±210 kg solids of each tonne of fish produced (IOA 1990) (see also Table 1.3.2 in chapter 1.3). The typical example of solid waste from aquaculture of rainbow trout was reported to be 290-655 kg · tonne^{-1} (Phillips *et al.* 1990). The solid waste production in the present study with silver perch is within the range of the reported value. Cho *et al.* (1991) reported solid waste production from brown trout (*Salmo trutta*) culture to be 204 kg · tonne^{-1} and suggested that use of high nutrient diet (HND) or "low pollution diet" can reduce solid load further to 183 kg · tonne^{-1} fish production. They suggested that modifications of diet formulations and re-examining the feeding strategies could reduce solid waste load from aquaculture.

In the present study, lower solid waste load obtained at 25^{O}C may indicate that fish all being reared close to their optimum temperature may produce less solid output to the environment. In addition, the restricted feeding strategy adopted here may have further reduced solid output since a study by Ballestrazzi, *et al.* (1994) demonstrated a reduction of waste output with restricted feeding in sea bass. Diet-2 fed to fish resulted in the lowest solid output at each temperature regime ($P>0.05$) which is most likely related to the extruded nature of the diet. This view is supported by previous observations that extruded diet/ingredients improves the utilization of foodstuffs and lead to reduction in suspended solids in the water (Johnsen and Wandsvik 1991). To reduce pollution load from fish feed, improvement in feed conversion ratio (FCR) is of utmost important (Alsted 1991). Previous studies have indicated that high energy diet reduce the solid waste load by improving the feed conversion and by increasing digestibility of organic matter (Johnsen and Wandsvik 1991). The FCR was found to be significantly better in fish reared at 25^{O}C ($P<0.05$) compared to that in fish reared at 30^{O}C and 20^{O}C (see chapter 3). The better FCR obtained at 25^{O}C may possibly have caused less solid output at 25^{O}C.

It is concluded that in the preliminary trials, diet-2 gave a slightly higher gain in weight and FCR to fish compared to diet-1 which may have caused less suspended and dissolved solid waste to the environment. In addition the present results show that silver perch reared at 25^{o}C resulted significantly less solid waste output than either 30^{o}C and 20^{o}C ($P<0.05$), which indicates that the culture of fish close to their optimum temperature could be a simple means of reducing solid waste to the environment.

Chapter 6

EFFECT OF TEMPERATURE ON NITROGEN LOSSES AND NITROGEN RETENTION IN SILVER PERCH

6

EFFECT OF TEMPERATURE ON NITROGEN LOSSES AND NITROGEN RETENTION IN SILVER PERCH

INTRODUCTION

In intensive aquaculture, sufficient nitrogen is provided through feed to promote a good growth of fish. However, feeding in excess of requirements is uneconomical and may deteriorate the water quality (Poxton 1992). In order to maximize the nitrogen retention and minimize the nitrogen load to the environment, it is essential to provide the optimum level of protein (N x 6.25) to the cultured fish (Cho 1993). Generally, nitrogen absorbed in excess of requirements may be excreted as ammonia and the rate of excretion depends upon consumption rate (Rychly 1980; Kaushik and Cowey 1991), water temperature (Jobling 1981; Beveridge and Phillips 1993), body size of fish (Jobling 1981; Beveridge and Phillips 1993), protein content of the diet (Rychly 1980) and the species cultured (Beveridge and Phillips 1993).

There are few studies on nitrogen pollution from feeding of fish (Handy and Poxton 1993). Gill excretion and faecal output are the main routes of nitrogen loss from fish (Smith 1929; Fivlstad *et al.* 1990). The additional route of nitrogen loss is via uneaten food or food fine (Bergheim and Asgard 1996). The urinary nitrogen loss in fish (via kidneys) is minimal accounting for 2-14% (Smith 1929; Evans 1993). Nitrogen is a limiting nutrient in the aquatic system, and an abundance of nitrogen may cause algal blooms or eutrophication in waterbodies receiving fish farm effluents, resulting in the fluctuation of oxygen and pH levels (Phillips and Beveridge 1986; Laird and Needham 1988). As a consequence, there are regulations on the level of nitrogen that can be released as fish farm effluents to coastal waters in some countries (Handy and Poxton 1993). Moreover, to overcome the deleterious impacts of nitrogen pollution, the nitrogen content in commercial feed have been reduced in Nordic countries (Enell 1995).

The nitrogen pollution from ingested food can be minimized, if adequate nitrogen is absorbed by the gut and a substantial quantity of absorbed nitrogen is retained by the tissues. The amount of nitrogen retained can be quantified through nitrogen assimilation efficiency (N absorbed / N ingested) (Handy and Poxton 1993). Indeed, nitrogen retention is a good indicator of the growth of fish (Gerking 1955) and is a useful means of evaluating a diet. The nitrogen retention and nitrogen loss ratio also allows a quantification of the amount of nitrogenous wastes entering into an environment. There is a need to reduce nitrogen losses from aquaculture since studies made in other countries have found a strong relationship between water pollution and discharge of nitrogen from fish farms (see chapter 1.4, on nitrogen pollution).

Although aquaculture is an infant industry in Australia it is expanding because of an increase in interest and investment for culturing silver perch (Kibria *et al.* 1996a). To date, there has not been any research on nitrogen losses and nitrogen retention from the aquaculture of silver perch though such information are vital to assess dietary protein utilization by fish (Kaushik *et al.* 1984) and to quantify nitrogen load into an environment. The present study was an attempt to study nitrogen loss and nitrogen retention by silver perch, *Bidyanus bidyanus* at 25^{o}C and 30^{o}C.

MATERIALS AND METHODS

Silver perch juveniles (av.size 656 gm) were bought from a local native fish farm and acclimatized similarly as mentioned in chapter 2. They were reared in glass aquaria (30 x 16 x 17 cm) at two temperatures, 25^{o}C and 30^{o}C. These temperatures were selected since fish reared at 25^{o}C and 30^{o}C showed better growth and less solid waste output in earlier experiments (see Chapter 3 and Chapter 5). Fish were fed on three commercial silver perch diets (silver crumbles/starter) at the rate of 3% of their body weight. The feed was divided into two portions and fed twice a day (at 0900 and 1600 hours), six days a week for four weeks in order to study the nitrogen losses and

nitrogen retention by silver perch. The three diets referred to as diet-1, diet-2 and diet-3, contained 53%, 45% and 36% protein respectively (see Table 2.1. for proximate composition of the three diets). Fish reared at 25°C had significantly better food conversion ratios (FCR) than fish reared at 30°C ($P<0.05$). Of the three diets fed to fish, fish fed diet-2 showed a trend of slightly better FCR than other diets ($P>0.05$).

Fish were stocked at the rate of four fish per aquarium and each diet had six replicates., i.e., 24 fish per diet per temperature. The water quality was maintained similarly as described in chapter 2. Each aquarium was siphoned a number of times per 24 hours to collect faeces by using a 5 mm hose. It includes siphoning of aquarium soon before the feeding and 5-10 minutes after the feeding in order to remove any uneaten food. Collected faeces were first centrifuged at 2000 r.p.m for 10 minutes to separate solids from the liquid (Econospin-Sorvall Instruments), the supernatant was discarded and the precipitate was dried in an oven at 100°C (Law 1984; Hajen *et al.* 1993). Water in aquaria was sampled every morning using a plastic syringe to record the daily variations in ammonia concentrations (total ammonia nitrogen) with respect to temperature and diets fed to fish. An hourly pattern of ammonia excretion was also recorded by analyzing aquaria water at two hourly intervals for 24 hours on three consecutive days (at the end of above trial). The nitrogen balance is determined from the proportion of nitrogen absorbed into the tissues from the food (nitrogen gain) and metabolic output (nitrogen loss) :

N balance = N consumed - N retained - N excreted - Faecal N (Hall *et al.* 1992).

The nitrogen intake of fish was calculated from the nitrogen content of the feed and amount fed to fish.

Samples were weighed using an analytical balance (Mettler AE 200). Proximate analysis of fish, and faeces (moisture, nitrogen, fat, carbohydrate, ash and phosphorus)

was done following AOAC (1990a, 1990b) (see chapter 2 for methodology). The nitrogen retained in the fish carcass was determined by subtracting initial nitrogen content in the carcass from the final carcass nitrogen content (Brown *et al.* 1993). Ammonia in water was determined with a Tecator flow injection analyzer (Aquatec 5400 analyzer) following the Aquatec instruction manual (Tecator 1990) (see chapter 2.2, for details on methodology).

Mean, standard deviation and standard error of nitrogen losses and nitrogen retention per diet were calculated following Zar (1984). One way analysis of variance (ANOVA) were used to compare nitrogen retention and nitrogen losses at 25^{o}C and 30^{o}C since there was no significant differences in nitrogen retention of three diets fed to fish. All calculations were performed on an IBM compatible MS Excel programme (version 5.0).

RESULTS

Hourly patterns of nitrogen excretion

The hourly patterns of ammonia excretion is presented in Figure 6.1. The figure shows that there was a sharp increase of ammonia level soon after a meal and a linear decrease during the remaining 24 hours. The rate of ammonia excreted was affected by the protein level of the diet. Fish fed diet-1 containing 53% protein showed a trend of slightly more ammonia excretion than fish fed either 45% and 36%, although there was no significant difference in the hourly ammonia excretion rate of the three diets fed to fish ($P>0.05$).

Daily patterns of ammonia excretion

In a comparison of the three diets, there was trend for diet-1 to be discharge more ammonia compared to fish fed diet-2 and diet-3, though differences in daily ammonia excretion between the three diets were not significantly different ($P>0.05$) (Figure 6.2, 6.3). The daily ammonia excretion rate increased with increase in temperature and was significantly higher at 30^{o}C ($P<0.05$) (Figure 6.4.).

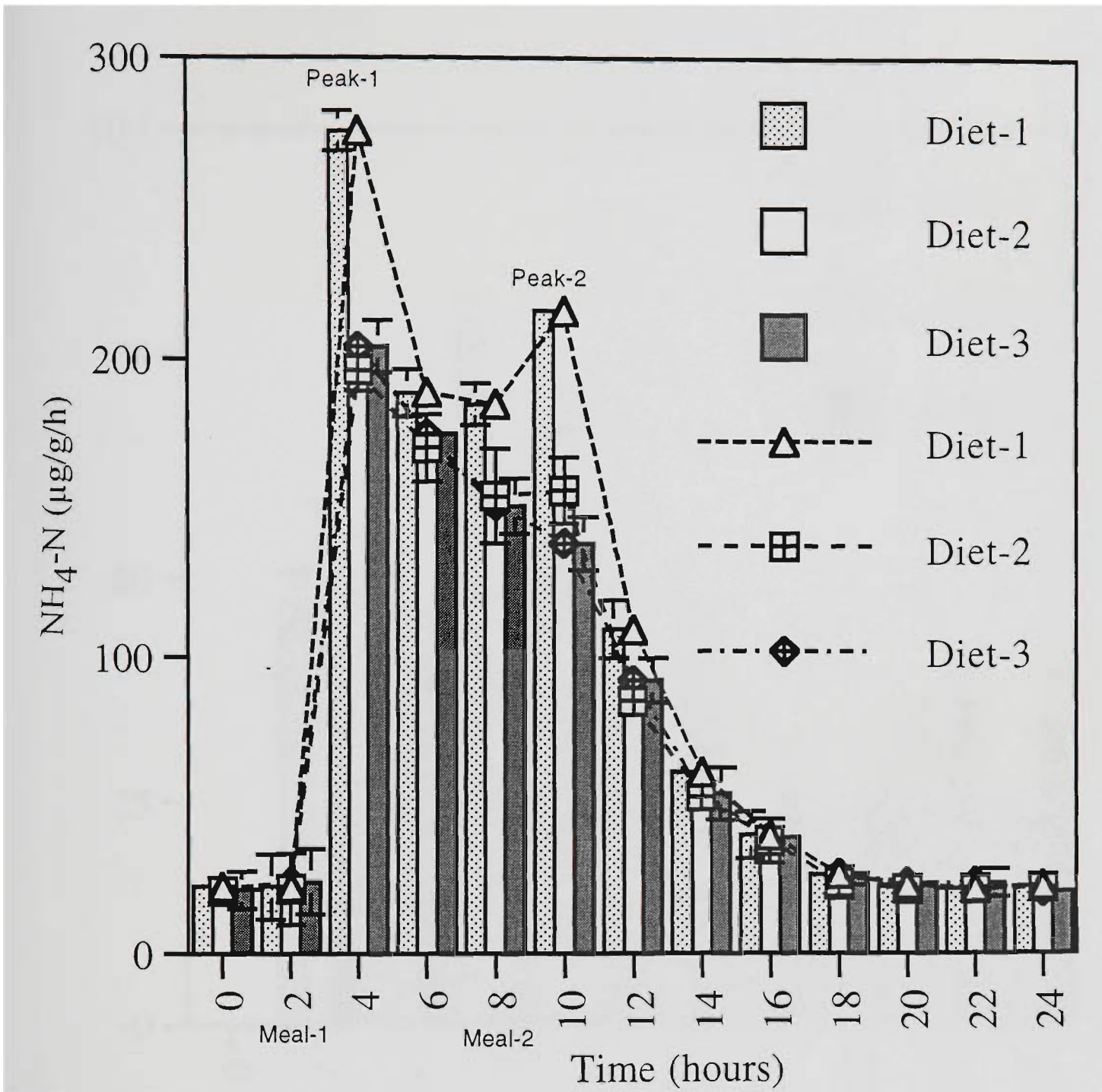

Figure 6.1. Postprandial patterns of ammonia excretion by silver perch juveniles fed three diets and reared at 25 degree celcius. There was no significant difference in hourly ammonia excretion in fish fed on the three diets ($P>0.05$). The protein content of the three diets are : Diet-1(53%) diet-2(45%), and diet-3(36%) (mean±SE; n=3).

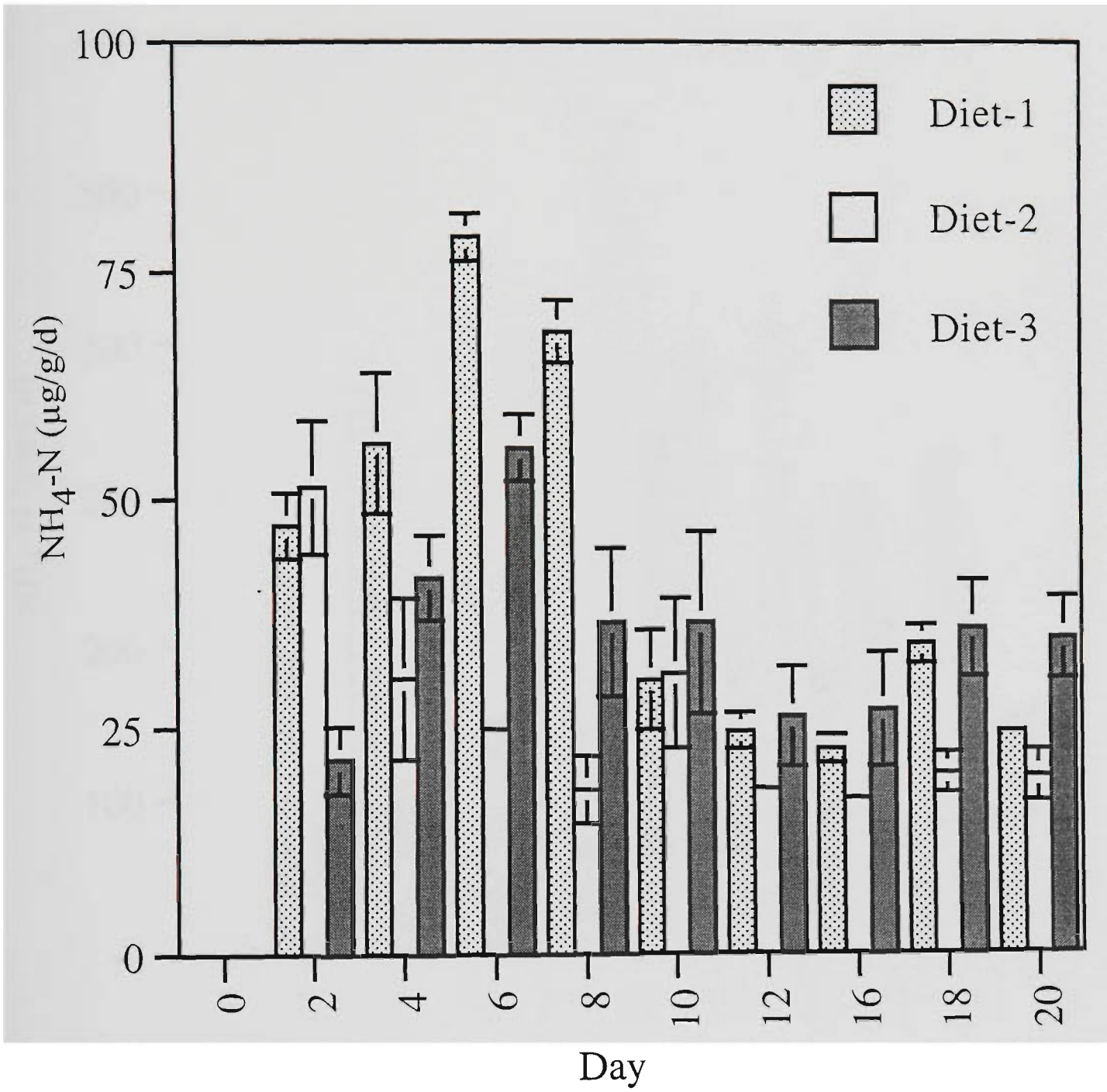

Figure 6.2. Day to day variation in ammonia excretion by silver perch juveniles fed three diets and reared at 25 degree celcius. There was no significant difference in ammonia excretion in fish fed on the three diets at 25 degree celcius ($P>0.05$) (mean±SE; n=6).

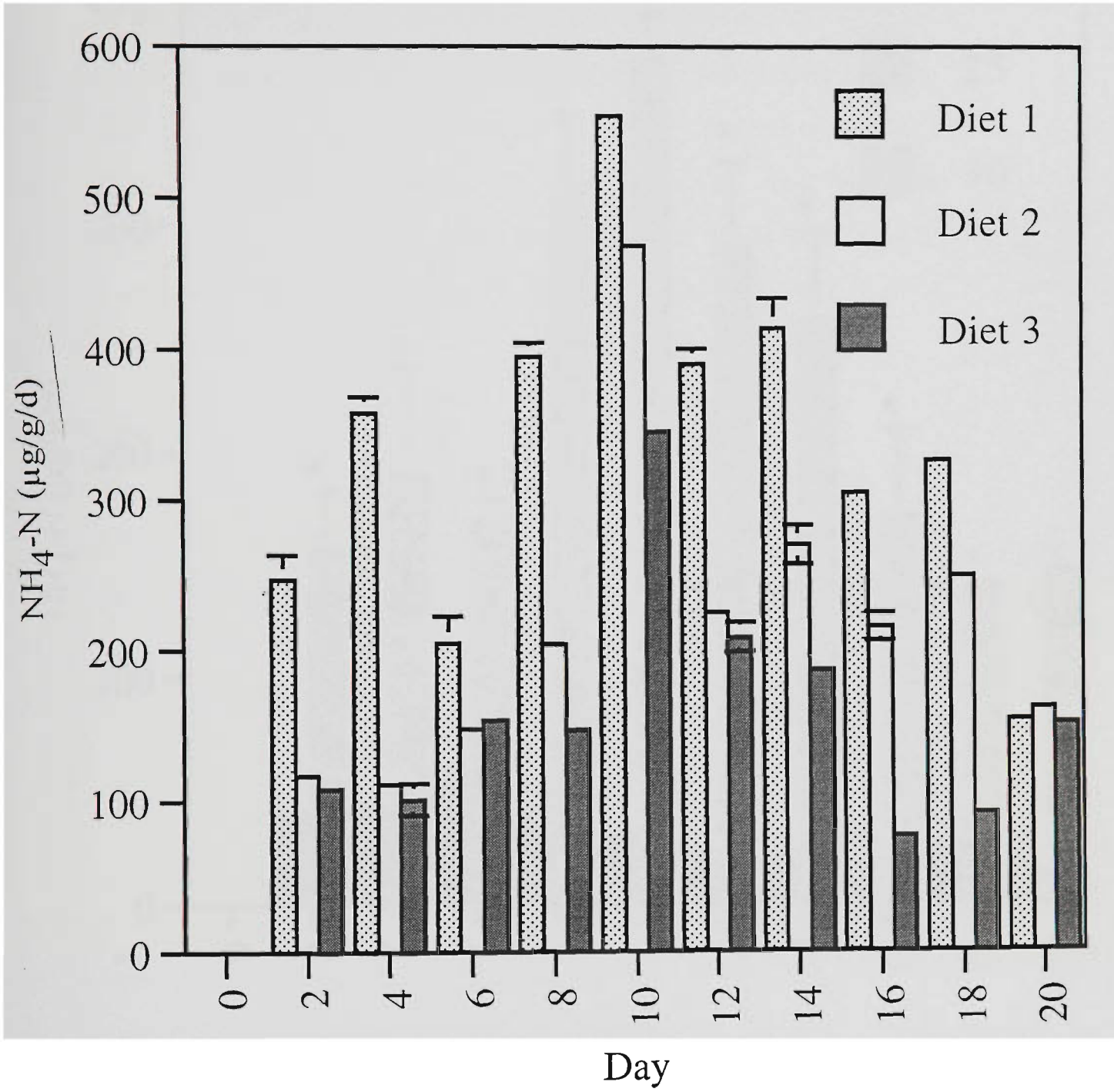

Figure 6.3. Day to day variation in ammonia excretion by silver perch juveniles fed three diets and reared at 30 degree celcius. There was no significant difference in ammonia excretion in fish fed on the three diets at 30 degree celcius ($P>0.05$) (mean±SE; n=6).

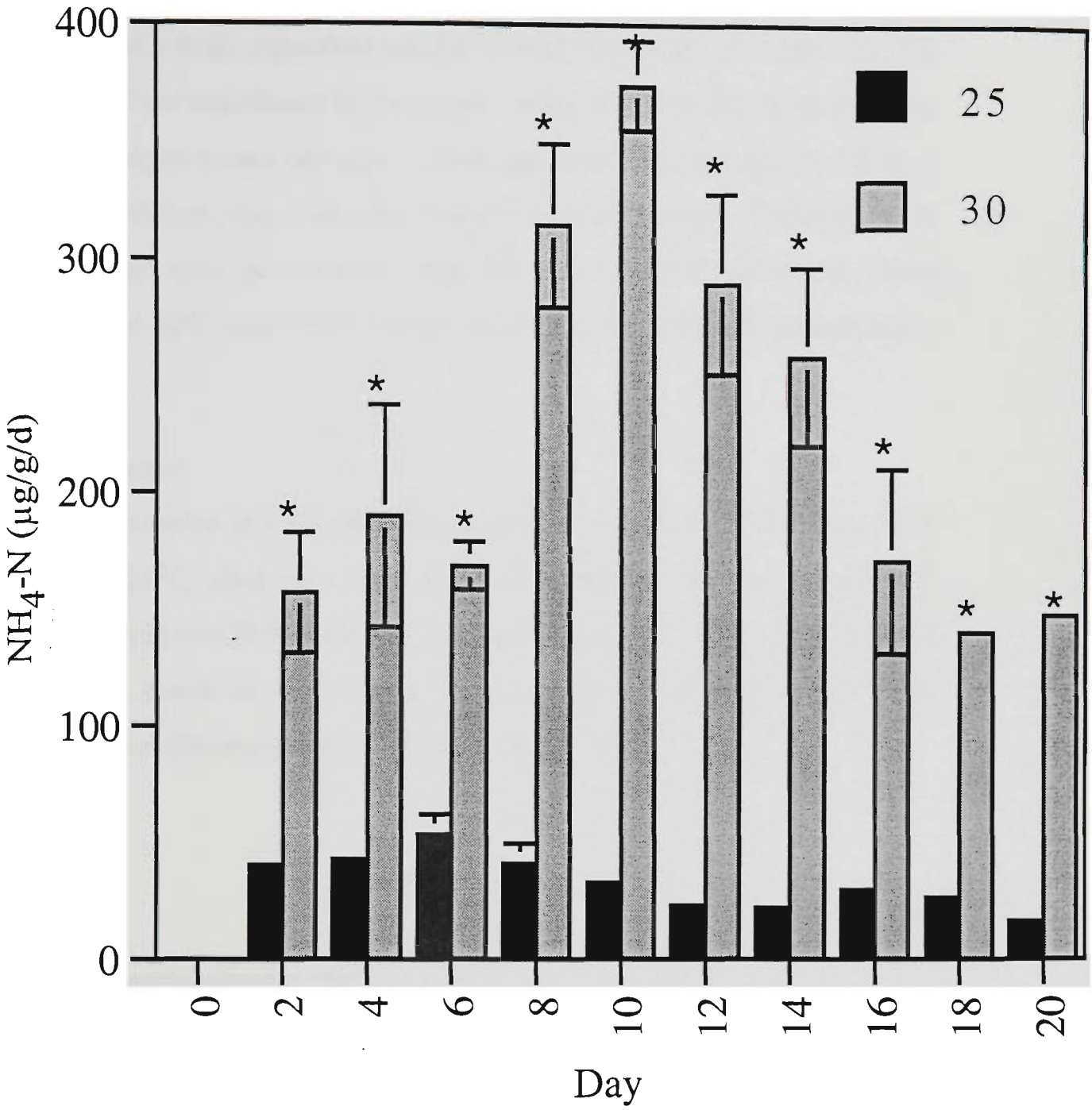

Figure 6.4. Day to day variation of ammonia excretion by silver perch juveniles reared at 25 and 30 degree celcius. Ammonia excretion was significantly higher at 30 degree celcius ($P<0.05$) (mean±SE; n=18).

* denotes significantly different

Faecal nitrogen loss

There was a positive correlation between nitrogen intake and faecal nitrogen losses in silver perch and a linear regressions could be plotted (Figure. 6.5, 6.6 and 6.7.). The faecal nitrogen loss was affected by the protein content of the diet and the temperature. Fish fed on a higher protein diet (diet-1) discharged more faecal nitrogen than diets of lower protein content (diet-2 and diet-3) at the same temperature. Similarly, fish at 30^{o}C discharged more faecal nitrogen than fish reared at 25^{o}C, the average faecal nitrogen loss at 30^{o}C was 9.86% whereas at 25^{o}C it was 5.84% of nitrogen intake (Table 6.1.).

Nitrogen retention

The nitrogen retention in silver perch was significantly higher at 25^{o}C than at 30^{o}C ($P<0.05$). At 25^{o}C, silver perch retained 43.1% of ingested nitrogen and at 30^{o}C nitrogen retention was 29.4% of ingested nitrogen (Table 6.1.). Of the three diets fed to silver perch, diet-2, showed a trend of higher nitrogen retention at both 25^{o}C and 30^{o}C though the differences were not significant ($P>0.05$).

DISCUSSION

Maximum ammonia nitrogen excretion in silver perch was found soon after the first meal. A similar trend was found in trout by Rychly (1980). There appear to be two peaks of ammonia excretion soon after feeding of each meal to silver perch in this experiment (Figure 6.1.). Kaushik (1981) also reported two peaks of nitrogen excretion in rainbow trout *(Oncorhynchus myskiss)* fed on two meals. Similarly, cod (*Gadus morhus*) showed two peaks of ammonia excretion fed on two meals (Ramnarine *et al.* 1987). The hourly patterns of ammonia excretion observed in silver perch was similar to the observations made on other species (Brett and Zala 1975; Dabrowski and Kaushik 1984; Ballestrazzi *et al.* 1994), who reported that post-prandial excretion rate increases rapidly after a meal before returning slowly to

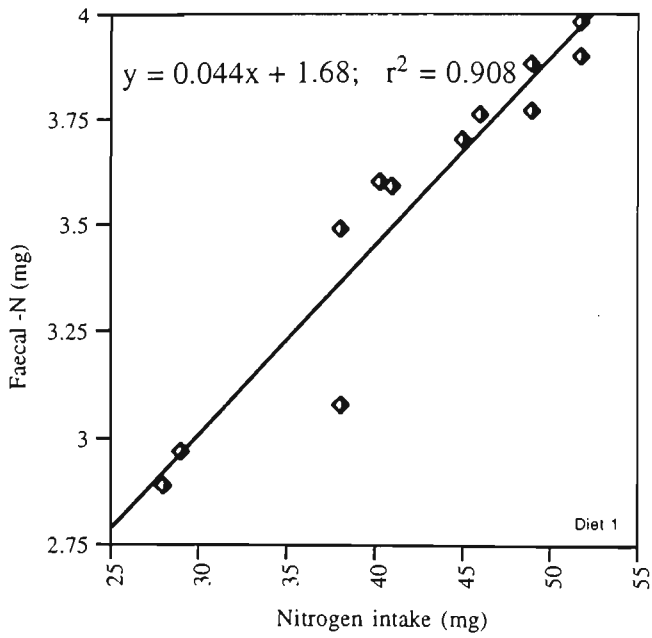

Figure 6.5. The relationship between nitrogen intake and faecal nitrogen loss from feeding of diet-1 to silver perch at 25 degrees. Diet-1 contained 53% protein.

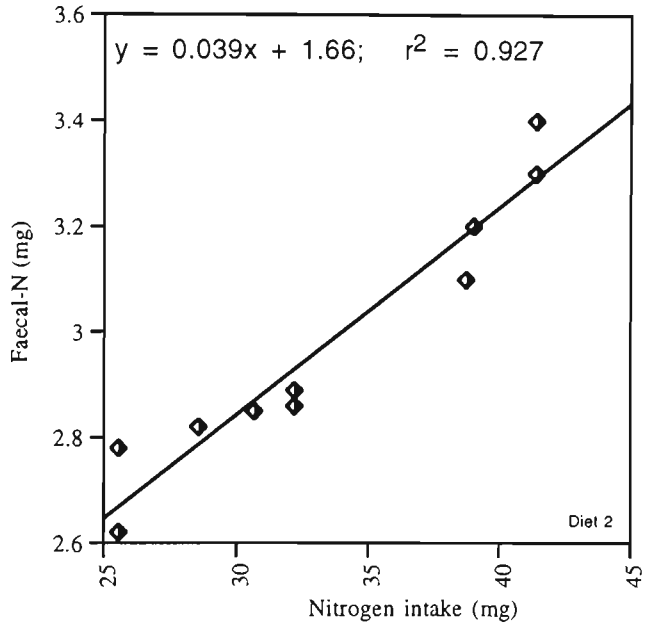

Figure 6.6. The relationship betweeen nitrogen intake and faecal nitrogen loss from feeding of diet-2 to silver perch at 25 degrees. Diet-2 contained 45% protein.

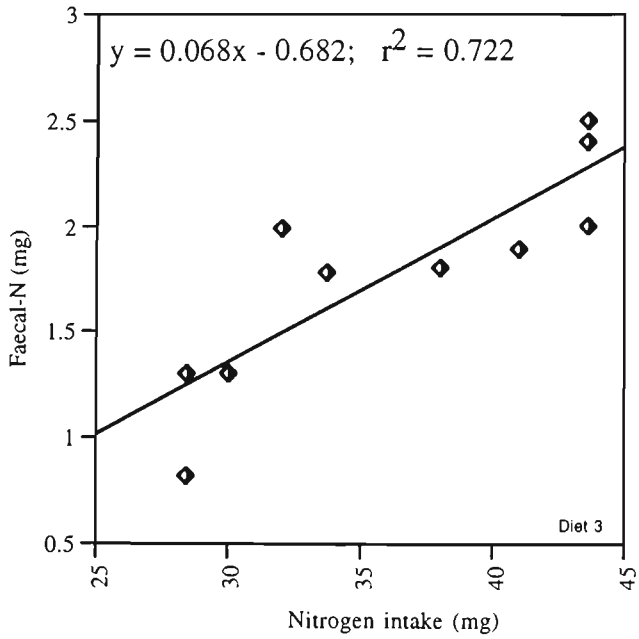

Figure 6.7. The relationship between nitrogen intake and faecal nitrogen loss from feeding of diet-3 to silver perch at 25 degrees. Diet-3 contained 36% protein.

Table 6.1. Nitrogen losses and nitrogen retention (g / kg body weight) and percentage of nitrogen retained in silver perch juveniles reared at 25^{0}C and 30^{0}C for 28 days. Fish were fed on three diets @ 3% of body weight, twice a day, six days a week. Diet-1, diet-2 and diet-3 contains 53%, 45% and 36% protein respectively.

	[----------25^{0}C----------]			[----------30^{0}C----------]		
	Diet-1	Diet-2	Diet-3	Diet-1	Diet-2	Diet-3
N Ingested (g /kg BW per day)	3.79±0.2	3.23±0.1	2.60±0.3	3.89±0.4	3.25±0.3	2.60±0.4
N retained (carcass) (g /kg BW per day)	1.42±0.08^a	1.60±0.35^a	1.1±0.26^a	1.03±0.31^b	1.02±0.28^b	0.79±0.22^b
Faecal nitrogen (g /kg BW per day)	0.24±0.01^a	0.16±0.03^a	0.15±0.01^a	0.40±0.01^b	0.34±0.01^b	0.23±0.02^b
Non-faecal nitrogen (g /kg BW per day)	2.14	1.47	1.35	2.46	1.89	1.60
N Retained / N Ingested (%)	37.50	49.50	42.40	26.48	31.38	30.38
Mean nitrogen retained (%)		43.13±3.48^a			29.41±1.35^b	

[1]Nitrogen retention = $\frac{\text{(carcass nitrogen content at the end of experiment - carcass nitrogen content at the start of experiment)}}{\text{total nitrogen intake}}$ x 100 (Hardy 1989; Brown *et al.* 1993)

[2]Values are mean±SE; n=6.

[3]Values in the same row with common superscripts are not significantly different from each other (P>0.05).

the pre-feeding level. Moreover, the amplitude and time of appearance of peak rates are known to depends upon the size of fish, amount of nitrogen intake and water temperature (Kaushik and Cowey 1991). Ballestrazzi *et al.* (1994) found that hourly ammonia excretion in the sea bass increased with increasing protein level, and that it was affected by the timing of the meal though there was no significant differences in nitrogen excretion of diets containing 41%, 48.6% and 53.6% protein. Similarly silver perch fed diet-1 (53% protein) showed maximum ammonia excretion compared to fish fed diet-2 (45% protein) and diet-3 (53% protein) although the excretion rate was not significantly different ($P>0.05$) (Figure 6.1.).

The daily ammonia excretion with silver perch was found to be significantly higher at 30^{o}C than at 25^{o}C ($P<0.05$) (Figure 6.4.). Kaushik (1981) and Jobling (1981) also reported an increase of ammonia excretion of rainbow trout (*Salmo gairdneri*) and young plaice, *Pleuronectes platessa* reared at higher temperature. Similarly, Japanese flounder released more ammonia at 25^{o}C than at lower temperature in the order of 25^{o}C>20^{o}C>16^{o}C (Kikuchi *et al.* 1995). Therefore the results obtained in this study with silver perch also agree with the findings of other researchers that at a higher temperature there was a higher ammonia excretion (Jobling 1981; Kaushik 1981; Kikuchi *et al.* 1995). A rise in temperature increases plasma ammonia excretion which increases the level of ammonia through renal and branchial paths (Pequin and Serfaty 1968); thus both hourly and daily patterns of nitrogen excretion can be affected (Kikuchi *et al.* 1995). Indeed, nitrogen excretion in fish can be used to evaluate the diet (Cho 1993) and the dietary protein utilization (Kaushik *et al.* 1984). Most of the nitrogen in fish is excreted in the form of ammonia and the amount of excretion varies depending upon body weight (Jobling 1981; Kikuchi *et al.* 1990), species (Davenport *et al.* 1990), temperature (Beveridge and Phillips 1993; Jobling 1981; Kelly *et al.* 1994; Rowland 1996), nitrogen intake (Rychly 1980; Kaushik and Cowey 1991), protein content of the diet (Iwata 1970; Ogino *et al.* 1973a; Rychly 1980), quality of diet (Dosdat *et al.* 1995; Lanari *et al.* 1995a),

protein/energy ratio (Gallagher and Matthews 1987), feeding rates (Dosdat *et al.* 1995), rearing conditions (Fromm and Gillette 1968; Olson and Fromm 1971; Sukumaran and Kutty 1977), pH and dissolved oxygen level (Brett and Zala 1975; Maetz 1972). The present study also confirms that temperature, protein content of the diet and nitrogen intake could affect the quantity of nitrogen excretion in silver perch (see Figure 6.2, 6.3, 6.4, 6.5, 6.6, 6.7).

In the present study, the main path of nitrogen losses were via gill excretion (85.7%-90.2%) and faeces (9.8%-14.3%). This is comparable to previous research where the reported nitrogen loss via gill excretion was 80%-90% (Wood 1958; Sayer and Davenport 1987) whereas nitrogen loss through faeces was between 10%-20% (Fivlsad *et al.* 1990). Nitrogen lost from aquaculture could stimulate algal growth (Ryther and Dunstan 1971; Carr and Goulder 1990a, 1990b), bring a change in the chemistry of the recipient waters (Albaster 1982; Munro *et al.* 1985) and may change the benthic environment (Merican and Phillips 1985). The nitrogen excretion in fish can be reduced by increasing energy content and decreasing the content of protein in the diet (more information on reduction on nitrogen discharge is given in chapter 2.4). For example, a reduction of ammonia excretion from 35 to 22 kg/t fish and faecal nitrogen loss from 11% to 8% was achieved from the feeding a high energy diet to salmon (Johnsen *et al.* 1993). It is believed that when the protein supply is in excess of requirements or when the amino acid profile does not corresponds to the requirement of the fish, the excess nitrogen supplied through feed will simply be excreted (Bergheim and Asgard 1996). The present study also demonstrated that the discharge of nitrogen via gill excretion and faecal loss by silver perch fed on commercial diets could be substantial (Table 6.1), therefore research on the optimum protein requirements of silver perch is essential in order to reduce the nitrogen output into the environment.

In the present study faecal nitrogen loss was found to be related to the amount of nitrogen in feed, for example, the diet containing higher nitrogen also produced more faecal nitrogen in silver perch (Figure 6.5, 6.6, 6.7.). Ogino *et al.* (1973a) reported a higher faecal nitrogen losses in carp fed on a diet containing higher nitrogen diet. A linear relationship between the nitrogen intake and faecal nitrogen release was also found in crucian carps by Iwata (1970) and Nose (1967). Ogino *et al.* (1973a), Kim (1974) and Nose (1967) reported a linear relationship between the faecal nitrogen loss and the protein content of the diet fed to carp and rainbow trout respectively. Silver perch raised at 30^{o}C released significantly more faecal nitrogen than raised at 25^{o}C and the faecal nitrogen losses at 25^{o}C and 30^{o}C were 5.68$\pm$0.4% and 9.86$\pm$0.5% of nitrogen intake respectively ($P<0.05$). The rainbow trout, flounder and carp released more faecal nitrogen at higher temperature than at lower temperature (Ogino *et al.* 1973a; Kaushik 1981; Kikuchi *et al.* 1995) and in general the metabolic faecal nitrogen excretion in fish increases with an increase in temperature (Kim 1974; Hepher 1988). The faecal nitrogen loss in fish (rainbow trout, tropical species) is reported to be in the range of 13-15.7% of nitrogen intake (Kaushik 1980; Beveridge and Phillips 1993) whereas in sea bream the loss of nitrogen through faeces was from 5.5 to 13.6% (Porter *et al.* 1987). Values of faecal nitrogen loss obtained in this study falls within these reported results. In short, the faecal nitrogen loss could be one-third of nitrogen excreted by fish (Porter *et al.* 1987).

Silver perch reared at 25^{o}C showed significantly higher ($P<0.05$) nitrogen retention than reared at 30^{o}C (Table 6.1). This may be related to the better FCR and better growth obtained at 25^{o}C (see chapter 3 for silver perch growth at 25^{o}C). It has been suggested that fish grown above their optimum temperature may have a lower food conversion efficiency due to lower appetite and growth of fish (Brett 1979; Cossins and Bowler 1987) and these may have resulted to the lower nitrogen retention at 30^{o}C with silver perch. Furthermore, the better nitrogen retention at 25^{o}C may have been due to significantly lower nitrogen excretion observed at this temperature (Figure

6.4.). It is known that temperature, diet, and body weight of fish could play a significant role in nitrogen retention in fish (Cho 1993). Kaushik and Oliva-Teles (1985) observed that rainbow trout utilized diets more efficiently at their optimum temperature signifying that at optimum temperature there appears to be better digestibility and better nutrient retention. The diet and its composition is another important factor determining the nitrogen retention efficiencies. For example, extruded diets resulted in much higher nitrogen retention in rainbow trout than normal diets (Lanari *et al.* 1995a, 1995b). The nitrogen retention with extruded diets was 46-49% while with normal diet varied between 35-36.5% (Lanari *et al.* 1995a). In the current experiment, however extruded diet, diet-2 did not result significantly higher nitrogen retention in silver perch compared to other two diets fed to fish ($P>0.05$). Therefore, further experiment may be conducted to see whether diet-2 result significantly better nitrogen retention when reared for longer period.

A study conducted by Lanari *et al.* (1995a) demonstrated that the diet which resulted in better growth and FCR also resulted in maximum nitrogen retention in rainbow trout. The nitrogen retention in rainbow trout was reported to be higher with a diet higher in protein content (Rychly 1980). In the current experiment, diet 1-containing 53% protein did not result in higher nitrogen retention in silver perch, which may indicate that protein requirement of silver perch is lower than 53% and this may be related to lower weight gain, SGR, and FCR obtained with diet-1 in earlier experiments (see chapter 3; and Kibria *et al.* 1997a).

It is concluded that the above experiments on nitrogen losses and nitrogen retention shows that culture of fish at their optimum temperature may enhance nitrogen retention and a reduction in the discharge of nitrogen to the environment. In addition, silver perch reared at $25^{o}C$ resulted significantly higher nitrogen retention (43.1%) than reared at $30^{o}C$ (29.4%) ($P<0.05$).

Chapter 7

EFFECT OF TEMPERATURE ON PHOSPHORUS LOSSES AND PHOSPHORUS RETENTION IN SILVER PERCH

7

EFFECT OF TEMPERATURE ON PHOSPHORUS LOSSES AND PHOSPHORUS RETENTION IN SILVER PERCH

INTRODUCTION

Fish require phosphorus for optimum growth, feed conversion efficiency, bone development, maintenance of acid-base regulation, and lipid and carbohydrate metabolism (Lovell 1978; Ogino and Takeda 1978; Lall 1991). Diets deficient in phosphorus can suppress the appetite and may lead to the death of fish (Lall 1979). Since concentration of phosphorus is low in both freshwater and seawater (Boyd 1971; Weatherly and Gill 1987), and phosphorus is absorbed only at a very low rate from the water (Phillips *et al.* 1956), the main source of phosphorus for fish is dietary.

Dietary phosphorus supplied and unavailable to fish will be evacuated from the gut in the faeces, whilst phosphorus surplus to requirements will be excreted via the kidney and gills (Forster and Goldstein 1969; Nakashima and Leggett 1980). The phosphorus released from uneaten food and faeces should be minimized because it stimulates algal growth or eutrophication in receiving waters (Walker and Hillmann 1982; Lall 1991). Studies conducted in Europe and North America demonstrate a strong correlation between water pollution and discharge of phosphorus from fish farm effluents since it can deteriorate the water quality of the recipient water bodies (see chapter 1.5 for impacts of phosphorus pollution). In order that environmental impacts are minimized information on the absorption, metabolism and excretion of phosphorus by cultured species of fish is essential (Lall 1991). There is very limited information available on phosphorus losses and retention (Kibria *et al.* 1996b; Kibria *et al.* 1997c. accepted) and studies are required on phosphorus retention at various stages of development in fish (Lall 1991).

The silver perch, *Bidyanus bidyanus* is an Australian native fish of the highest aquaculture potential (Allan & Rowland 1996) and the industry is growing rapidly with the yield growing from 26.6 tonnes in 1991-92 to 52.7 tonnes in 1994-95 (O'Sullivan 1995; O'Sullivan and Kiley 1996) as a result of interest and investments in culturing this species (Kibria, *et al.* 1996a). Although there is some concern about the environmental impacts of aquaculture, until now there is no research either on the quality or quantity of phosphorus that may be discharged from the aquaculture of silver perch. The objectives of this study were to evaluate the effects of temperatures on phosphorus losses and phosphorus retention in silver perch *Bidyanus bidyanus*.

MATERIALS AND METHODS

The experiment was conducted in the wet laboratory at the Victoria University of Technology. Juveniles of silver perch of similar size (av. wt. 928 mg) were bought from a local native fish farm where fish were grown in earthen ponds. They were acclimatized in the laboratory in large holding tanks similarly as mentioned in chapter 2. They were reared in small glass aquaria (30 x 16 x 17 cm) at two temperatures 25^{o}C and 30^{o}C since optimal temperature for silver perch is believed to be in the range of 23^{o}C to 28^{o}C (Rowland *et al.* 1995), and also confirmed in the present study as shown in chapter 3. Fish were fed on three commercial silver perch diets (silver crumbles/starter) at the rate of 3% of their body weight (as recommended). The feed was divided into two portions and fed twice a day (at 0900 and 1600 hours), six days a week for four weeks to study the phosphorus losses and phosphorus retention in this species. The three diets fed to fish referred to as diet-1, diet-2 and diet-3. The protein and phosphorus content of the three diets are : diet-1 (53% & 1.3%), diet-2 (45% & 1.16%) and diet-3 (36% & 1.28%) respectively (Table 2.1. for proximate composition of three diets). Four fish were stocked in each glass aquarium and each diet was replicated in six aquaria, i. e., 24 fish per diet per temperature. Individual aquaria were aerated with air stones to enhance dissolve oxygen content. The water quality was

maintained similarly as described in chapter 2. Each aquarium was siphoned a number of times per 24 hours by using a 5 mm hose, to collect faeces. Collected faeces were centrifuged as per the method described in chapter 6. Water samples were collected each morning using a plastic syringe to record the daily variations in orthophosphate concentrations with respect to temperature and diet fed to fish. Data were also collected on the hourly pattern of phosphate excretion for the three consecutive days of fish fed three diets at 25^{o}C. The food conversion ratio (FCR) was determined and the FCR was found significantly better at 25^{o}C than fish reared at 30^{o}C. The FCR at 25^{o}C and 30^{o}C were 1.09 and 1.66 respectively. The phosphorus (P) balance was determined using the following phosphorus balance equation :

P balance = Amount of P fed - P retained in carcass - P in faeces - P in uneaten feed (Ketola and Harland 1994).

Samples were weighed using an analytical balance (Mettler AE 200). Proximate analysis of food, fish and faeces was done following the methods as described in chapter 2.1. The phosphorus retained in the fish carcass was determined by subtracting initial carcass phosphorus content from the final carcass phosphorus content. Orthophosphate in water was determined with a Tecator flow injection analyser (Aquatec 5400 analyzer) following the Aquatec instruction manual (Tecator 1990) as described in chapter 2.

Mean, standard deviation and standard error were calculated following the method described by Zar (1984). All percentage data were transformed to arc sin values prior to analysis using an IBM computer. One way analysis of variance (ANOVA) was used to compare the phosphorus loss and phosphorus gain at 25^{o}C and 30^{o}C since there was no significant difference of phosphorus retention of three diets fed to fish. All calculations were performed on an IBM compatible MS Excel programme (version 5.0).

RESULTS

Hourly patterns of phosphate excretion

The hourly patterns of orthophosphate excretion is presented in Figure 7.1. The figure shows that there was a rapid increase in phosphate level soon after the first meal reaching a maximum at 6 hours and this decreased linearly during the remaining 24 hours. There was no second peak following the second meal. However, the excretion rate was not significantly different between silver perch fed on the three different diets containing 53%, 45% and 36% protein ($P>0.05$).

Daily patterns of phosphate excretion

Fish fed diet-2 containing 45% protein excreted comparatively less orthophosphate than either fish fed diet-3 (53% protein) and diet-3 (36% protein) (Figure 7.2, 7.3). However, there were no significant differences in phosphate discharged by fish fed the three diets ($P>0.05$). Silver perch reared at $30^{o}C$ showed greater orthophosphate excretion than those reared at $25^{o}C$ and the excretion rate was significantly higher at $30^{o}C$ than at $25^{o}C$ ($P<0.05$) (Figure 7.4.).

Phosphorus retention

The phosphorus retention in silver perch was significantly higher at $25^{o}C$ than at $30^{o}C$ ($P<0.05$) which may be related to the optimum growing temperature of silver perch at $25^{o}C$. The average phosphorus retained at $25^{o}C$ was 49.1% and at $30^{o}C$ it was 24.5% (Table 7.1). At $25^{o}C$, 66.7% of the phosphorus loss was in particulate form and the remaining 33.3% was in dissolved form.

Body composition of fish

The body composition of fish varied with the rearing temperature (Table 7.2.) and fish reared at $25^{o}C$ had significantly higher body protein, ash and phosphorus content than fish reared at $30^{o}C$ ($P<0.05$).

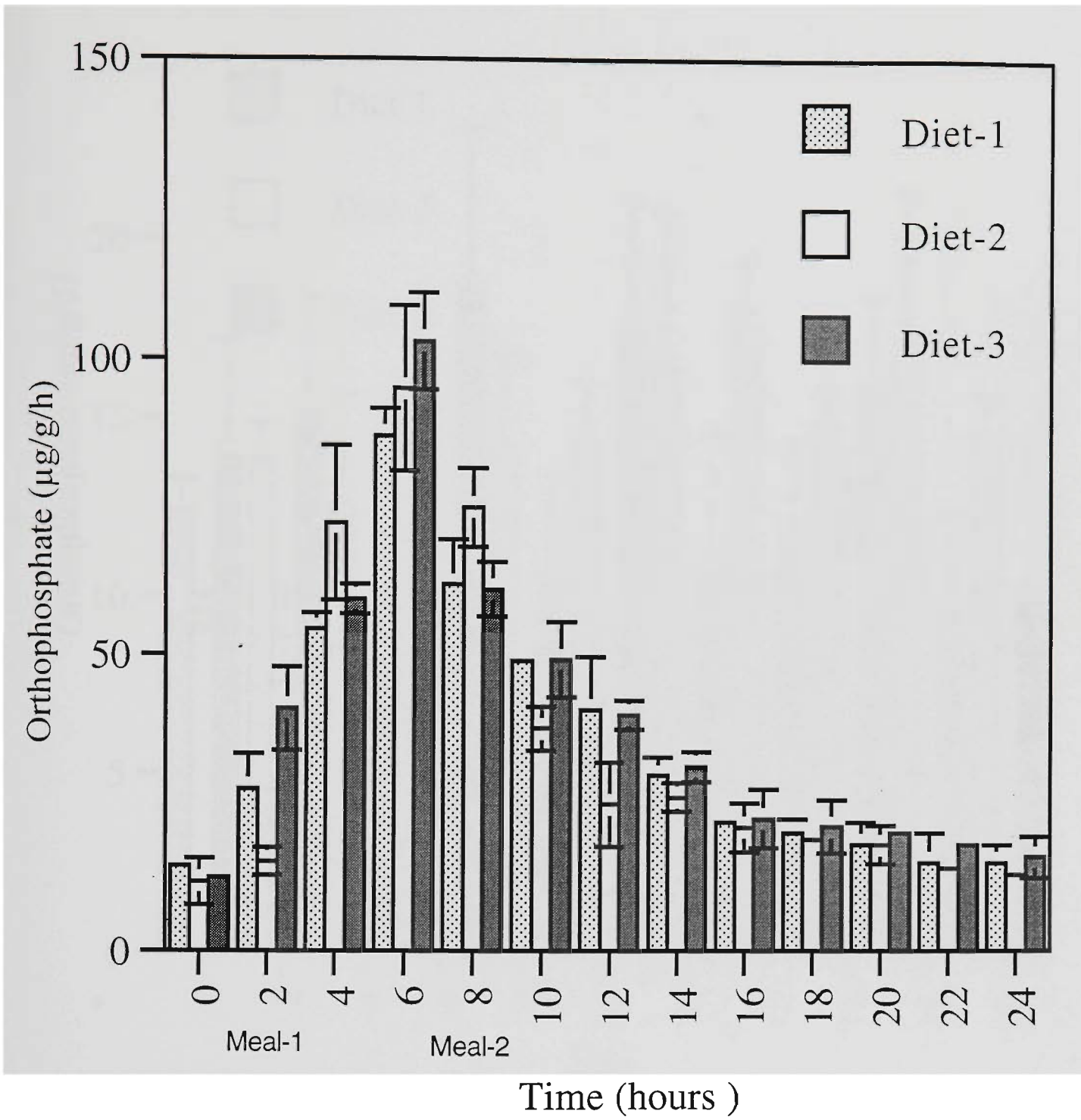

Figure 7.1. Postprandial patterns of orthophosphate excretion by silver perch juveniles fed three diets and reared at 25 degree celcius. There was no significant difference in hourly phosphate excretion in fish fed on the three diets (P>0.05) (mean±SE; n=3).

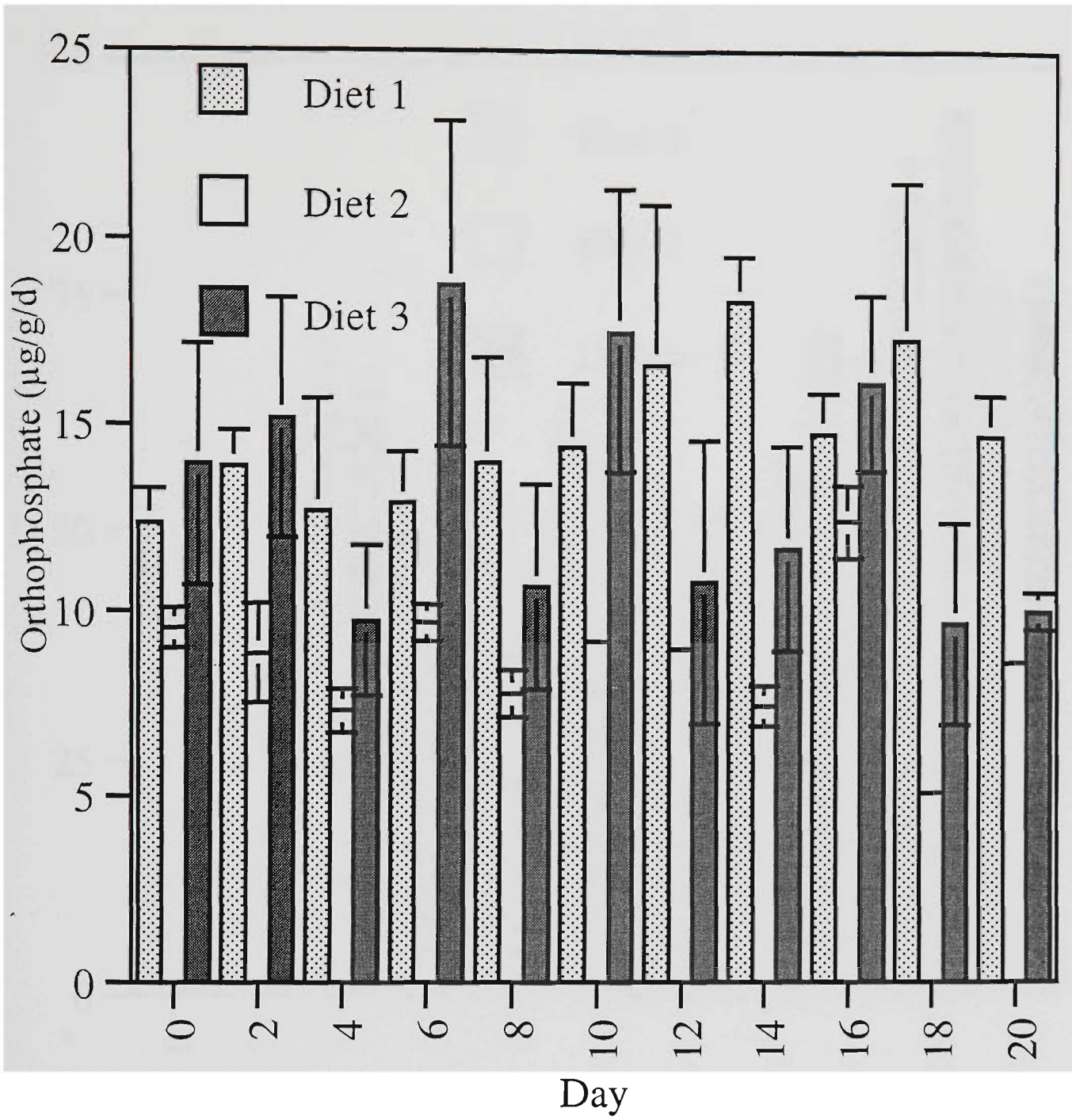

Figure 7.2. Day to day variation in orthophosphate excretion by silver perch juveniles fed three diets and reared at 25 degree celcius. There was no significant difference in phosphate excretion in fish fed on the three diets at 25 degree celcius ($P>0.05$) (mean$\pm$SE; n=6).

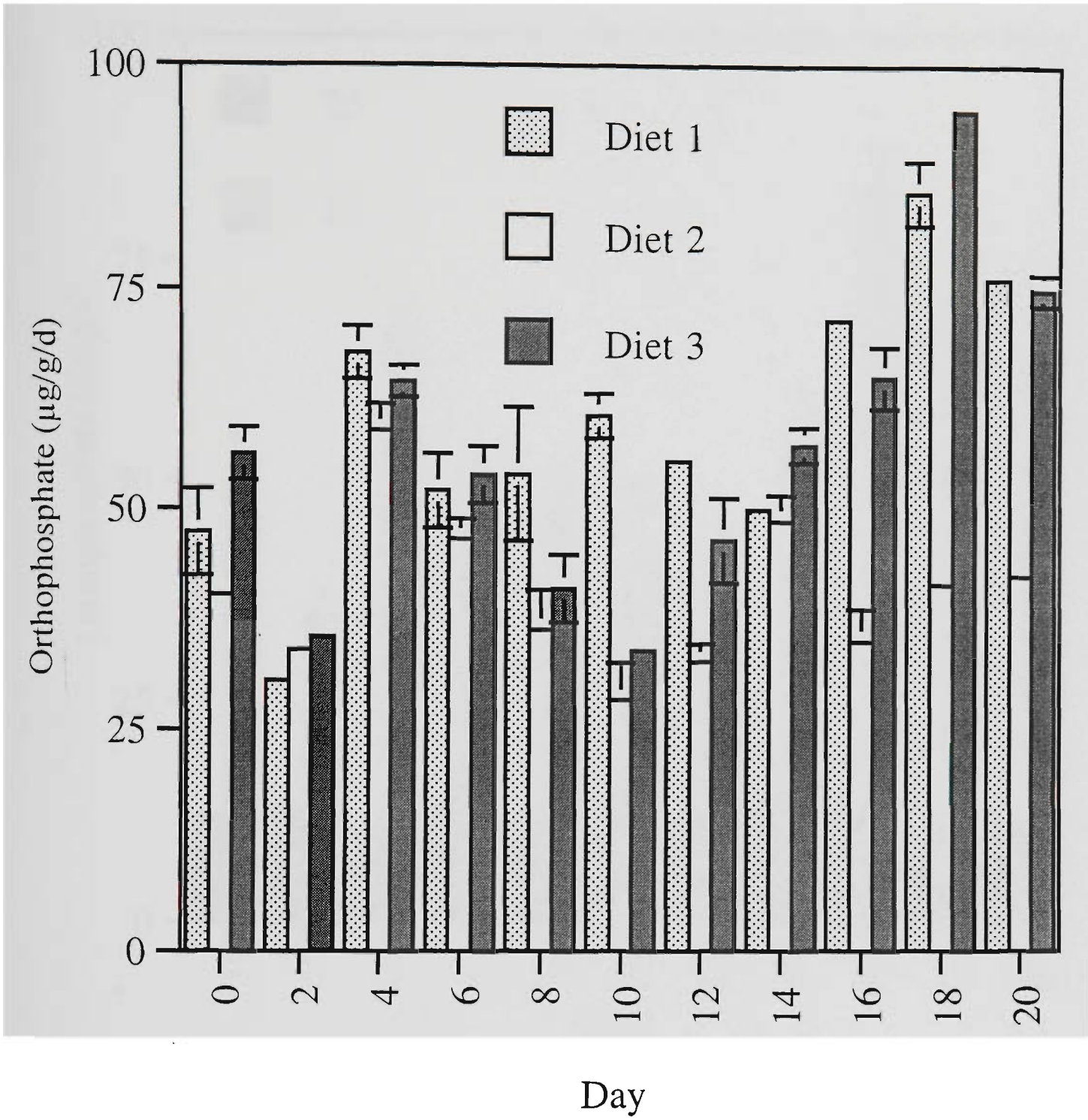

Figure 7.3. Day to day variation in orthophosphate excretion by silver perch juveniles fed three diets and reared at 30 degree celcius. There was no significant difference in orthophosphate excretion in fish fed on the three diets at 30 degree celcius ($P>0.05$).(mean±SE; n=6).

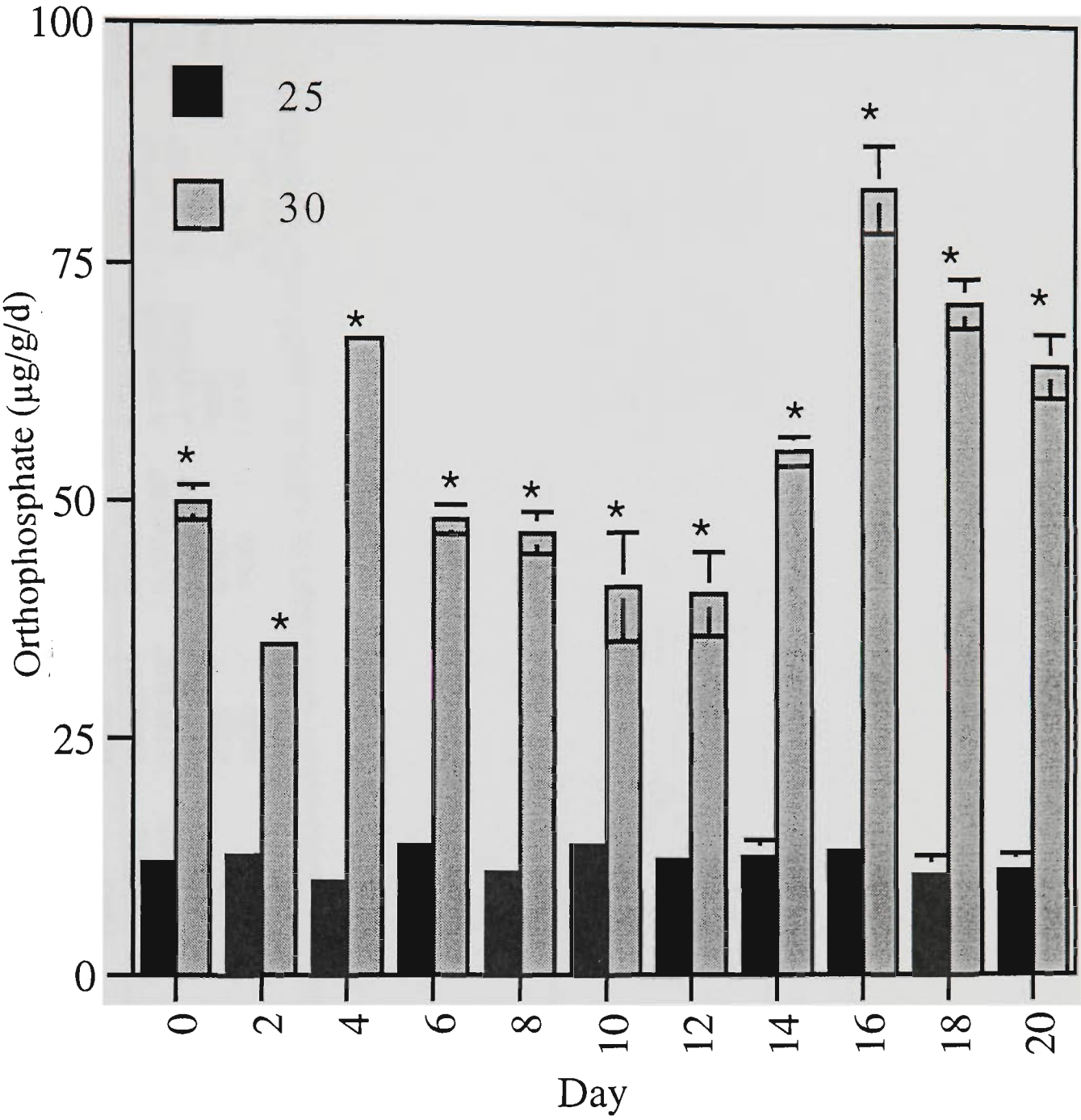

Figure 7.4. Day to day variation of orthophosphate excretion by silver perch juveniles reared at 25 and 30 degree celcius. Phosphate excretion was significantly higher at 30 degree celcius ($P<0.05$) (mean±SE; n=18).

* denotes significantly different

Table 7.1. Phosphorus losses and phosphorus retention (g / kg of body weight per day) and percentage of phosphorus retained in silver perch juveniles reared at 25°C and 30°C for 28 days. Fish were fed on three diets @ 3% body weight, twice a day, six days a week. Diet-1, diet-2 and diet-3 contains 53%, 45% and 36% protein respectively.

	[----------25°C----------]			[----------30°C----------]		
	Diet-1	Diet-2	Diet-3	Diet-1	Diet-2	Diet-3
P Ingested (g /kg BW per day)	0.524±0.02	0.466±0.03	0.514±0.01	0.524±0.04	0.464±0.05	0.464±0.03
P Retained (carcass) (g /kg BW per day)	0.285±0.02[a]	0.223±0.02[a]	0..231±0.04[a]	0.143±0.02[b]	0.115±0.06[b]	0.10±0.09[b]
Faecal phosphorus (g /kg BW day)	0.190±0.01[a]	0.143±0.01[b]	0.174±0.02[a]	0.300±0.01[c]	0.201±0.01[d]	0.21±0.02[d]
Non-faecal phosphorus (g /kg BW day)	0.049	0.100	0.109	0.081	0.148	0.154
P Retained / P Ingested (%)	54.30	48.00	45.00	27.29	24.78	21.55
Mean phosphorus retained (%)		49.10±2.74[a]			24.54±1.66[b]	

[1]Phosphorus retention = (carcass phosphorus content at the end of experiment - carcass phosphorus content at start of experiment) x 100 / total phosphorus intake (Hardy 1989; Brown *et al.* 1993).
[2]Values are mean±SE; n=6.
[3]Values in the same row with common superscripts are not significantly different from each other(P>0.05).

Table 7.2. Chemical composition and phosphorus content of the whole body at the end of feeding experiment.

		[----------25°C----------]			[----------30°C----------]		
	Initial	Diet-1	Diet-2	Diet-3	Diet-1	Diet-2	Diet-3
Moisture (%)	80.21	78.71	78.36	78.23	79.10	79.15	79.10
Crude protein (% dry weight)	58.83	63.59±0.58[a]	65.59±01.5[a]	63.87±0.51[a]	59.31±01.26[b]	60.46±01.40[b]	59.43±0.1[b]
Ash (% dry weight)	16.37	18.24±0.19[a]	17.43±0.22[b]	18.94±0.04[a]	17.16±0.21[c]	16.64±0.26[d]	16.01±0.2[d]
Phosphorus (% dry weight)	2.49	2.83±0.02[a]	2.76±0.06[a]	2.82±0.09[a]	2.50±0.27[b]	2.56±0.13[b]	2.51+0.13[b]

[1]Initial sample size was 34 fish.
[2]Values are mean±SE; n=6.
[3]Values in the same row with common superscripts are not significantly different from each other(P>0.05).

DISCUSSION

In silver perch orthophosphate excretion peaked soon after the first meal, a similar trend was found in sea bass by Ballestrazzi *et al.* (1994). Orthophosphate excretion in fish fed on three diets containing 53%, 45% and 36% protein was not significantly different ($P>0.05$). Ballestrazzi *et al.* (1994) also did not find any significant differences in orthophosphate excretion in sea bass fed on three diets containing 54%, 49% and 44% protein. Similarly, there was a rapid increase in orthophosphate concentration soon after the feeding of trout, which decreased and reached nonfeeding levels within six hours (Solberg and Bregnballe 1982). Peaks of orthophosphate excretion are thought to be related to feeding and activity of fish (Hennessy, *et al.* 1996) and is also dependent on the quantity and quality of food supplied (Lall 1991). Since all the commercial diets used in this experiment were of high quality, this may have caused nonsignificant orthophophate excretion in fish reared in this experiment.

Daily phosphate excretion showed that silver perch fed on diet-2 containing 45% protein, discharged comparatively less orthophosphate than fish fed on other two diets, although there was no significant differences in phosphate discharges of the three diets fed to silver perch (Figure 7.2, 7.3). It shows that there was no affect of protein level in diets in the discharge of phosphate in silver perch. Ballestrazzi *et al.* (1994) also did not find any affect of protein level in the release of phosphate in seabass. Fishmeal is the only source of protein and phosphorus for the three diets which could be the reason that orthophosphate excretion did not differ in the three diets tested. The phosphorus excretion rate was reported to be affected by both protein (Ballestrazzi *et al.* 1994) and phosphorus source (Ketola and Harland 1993).

Ballestrazzi *et al.* 1994 found that, when fish were fed on gluten based diets, orthophosphate excretion was significantly reduced apparently as a result of their less phosphorus in gluten compared to herring meal diets. Ketola and Harland (1993) reported that defluorinated rock phosphate as source of dietary phosphorus decreased about 40-51% phosphorus discharge in rainbow trout.

Silver perch reared at 25^{o}C showed less orthophosphate excretion than those reared at 30^{o}C and the excretion rate was significantly higher at 30^{o}C in fish fed on all three diets ($P<0.05$) (Figure 7.4.). The increased phosphate excretion at 30^{o}C may be linked to the higher metabolic rate at higher temperatures. Kristiansen and Hessen (1992) reported an increase in orthophosphate excretion of noble crayfish, *Astacus astacus* reared at higher temperature and Savitz (1971) observed an elevated phosphorusexcretion in bluegill sunfish, *Lepomis macrocephalus* at higher temperature. Therefore present results with silver perch agrees with the findings of Kristiansen and Hessen (1992) and Savitz (1971).

The phosphorus retention in silver perch was significantly more at 25^{o}C than at 30^{o}C ($P<0.05$) which may be related to the optimum growing temperature of silver perch and higher growth achieved at 25^{o}C (see chapter 3.). The phosphorus retention at 25^{o}C was 49.1% and at 30^{o}C it was 24.5%. (Table 7.1.). The reported phosphorus retention varies markedly between species, from 14-22% in coho salmon (Ketola, *et al.* 1991), 61-81% in rainbow trout (Ogino and Takeda 1978), 39-40% in channel catfish (Lovell 1978), 36.40% in Atlantic salmon (Johnsen, *et al.* 1993), 33% in rainbow trout (Ketola and Harland 1991) and 69-87% in sunshine bass (Brown, *et al.* 1993). In the present study with silver perch, phosphorus retention was found to be correlated with the rearing temperature and maximum phosphorus retention was obtained was 49% at 25^{o}C (close to optimum temperature).

The variation in phosphorus retention in fish appears to be due to diet, growth rates, source and level of phosphorus fed to fish (Ketola *et al.* 1991; Lall 1991). Lanari *et al.* (1995a) reported a significantly higher phosphorus retention by rainbow trout on a diet low in phosphorus. Brown *et al.* (1993) showed that sunshine bass fed with diets containing 0.34-0.54% available phosphorus had a phosphorus retention of 69-87% compared to 40-50% when fed with diets containing higher phosphorus of 0.64-1.04%. Similarly, rainbow trout fed on a lower phosphorus diet (0.89% phosphorus) had a phosphorus retention of 27%, while with a higher phosphorus diet (1.2% phosphorus), the phosphorus retention was 17% (Eskelnen 1984). Fish reared in cages gave a lower phosphorus retention apparently due to lower FCR values (Holby and Hall 1991). In the current study, the marginal differences in the phosphorus content of the three diets may have attributed in the non-significant retention of phosphorus in fish fed the three diets. Perhaps if the diets contained very different phosphorus content, the results of phosphorus retention could have shown significant differences.

The significantly higher phosphorus retention obtained with silver perch at 25^{o}C may have been a result of the better FCR achieved at 25^{o}C. The FCR is known to play a significant role in determining the level of phosphorus load expected (Storebakken and Austreng 1987a, 1987b; Enell 1995), since an increase in FCR value from 1.0 to 1.5 may increase phosphorus load to about 86% for total phosphorus (Storebakken and Austreng 1987a, 1987b) (see also chapter 1.5 for other factors contributing to phosphorus loading from aquaculture). In addition to FCR, digestibility of feed, nutrient content in diet, the feeding technique may influence the feed coefficient and phosphorus load from aquaculture (Enell 1995). Matty (1990) and Gavine *et al.* (1995) emphasized that a good FCR is essential in reducing the phosphorus pollution in salmonids and trout. Similarly, nutrient loss rates were found to be dependent upon nutrient content of the diet and the species cultured (Foy and

Rosell 1991a). Furthermore, the greater retention of phosphorus at 25°C may have been a result of lower phosphate excretion and lower faecal phosphorus losses obtained at 25°C (Figure 7.4; Table 7.1.).

Of the total phosphorus loss from aquaculture, a major loss has been reported to be in particulate form accounting to about 77.0-85.7%, a minor loss which accounts for 14.3-23.0% was in dissolved form (Enell 1987; Ackefors and Enell 1990; Enell and Ackefors 1991; Johnsen *et al.* 1993). The main path of phosphorus loss in silver perch in this study was via faeces accounting for 66.7% of the total phosphorus lost and the remaining 33.5% lost was in dissolved form.

The significantly higher body protein, ash and phosphorus content obtained at 25°C (Table 7.2.) may be related to the better phosphorus retention observed at 25°C and could be an effect of temperature adopted. Hasan, *et al.* (1993) found that the fish group that showed better FCR also had higher carcass protein and ash content. The body composition of fish has been reported to be affected by the rearing temperature (Brett, *et al.* 1969; Windell, *et al.* 1978; Singh, *et al.* 1979), ration size (Huisman, *et al.* 1979), experimental diets and protein level (Ballestrazzi *et al.* 1994; Haiqing and Xiqin 1994), and fish size (Brett *et al.* 1969). In the present study silver perch reared at 25°C also had a better FCR which may have caused higher carcass protein and carcass ash content at this temperature. Elliott (1976) reported that in brown trout, *(Salmo trutta)* changes in body constituents were affected by temperature, since they were very low at lower temperature (5.6°C) and maximum at optimum temperature (12.8°C). For example, the protein and ash content (wet weight basis) was highest when trout were reared at 12.8°C and lower at a temperature higher than optimum (Elliott 1976).

In conclusion, the results obtained in this study show that growing of silver perch at optimum temperature can enhance phosphorus retention and cause a reduction in the discharge of phosphorus to the environment. Silver perch reared at $25^{o}C$ resulted significantly higher phosphorus retention rate (49.1%) compared with $30^{o}C$ (24.5%) ($P<0.05$).

Chapter 8

THE NUTRIENT CONTENT AND THE RELEASE OF NUTRIENTS FROM FISH FOOD AND FAECES

8

THE NUTRIENT CONTENT AND THE RELEASE OF NUTRIENTS FROM FISH FOOD AND FAECES

INTRODUCTION

Fish food and faeces are known to be the main wastes in intensive aquaculture. The production of feed wastes as dust and uneaten food have been estimated by Beveridge (1996) to be close to 10%. Previous researchers also reported a figure of 10-30% of waste uneaten food alone from intensive aquaculture (Hoelzi and Vens-Cappell 1980; Penczak *et al.* 1982). The faecal output on a dry weight basis is reported to be 260 g per kg of food fed to fish (Butz and Vens-Cappell 1982). Uneaten food along with faeces (solid wastes) may increase sedimentation and enrich the nutrient pool of the receiving waters (Beveridge 1987). Under appropriate conditions phosphorus and nitrogen could be released from the sediments underlying receiving waters (see chapter 1.3 for factors related in the release of nutrients from solid waste) and may stimulate algal growth (Pettersson 1988).

Both phosphorus and nitrogen are essential ingredients incorporated into formulated feed to achieve a good growth of fish. These two nutrients are also required for algal growth in water bodies (State of Victoria 1995). In freshwater, phosphorus is the limiting nutrient and orthophosphate is the form that is readily available to plants (Chamberlain and Shapiro 1969; Welch and Lindell 1980; Bostrom *et al.* 1988a,1988b; Boyd 1990). Both phosphorus and nitrogen are normally limiting nutrients in seawater (Boyd 1990). Fish farm effluents containing phosphorus and nitrogen have been reported to have caused eutrophication of receiving waters (Laird and Needham 1988; Foy and Rosell 1991b). However, few researchers have studied

the fractional composition of nutrients and factors important in the release of nutrients from fish food and faeces (Kibria *et al.* 1977d. accepted). Such information is essential in planning strategies for nutrient management and pollution control. The present study investigates the fractional composition of nutrients and the effect of temperature and pH on the release of nutrients from fish food and faeces in the rearing of the Australian native fish silver perch *B. bidyanus*.

MATERIALS AND METHODS

Silver perch *Bidyanus bidyanus* juveniles (av. wt. 0.590g) were fed on two commercial diets referred to as diet-1 and diet-2 for four weeks at 25^{o}C. Fish were reared in small aquarium (70x60x30cm). The water quality of aquaria was maintained similarly as described in chapter 2. The total number of fish per treatment was twenty four i.e., the number of replicate tanks per diet was six. Fish were fed at the rate of 3% body weight. The feed was divided into two portions and fed to fish twice a day and six days a week for four weeks. Faeces was siphoned off a number of times a day by using a 5mm hose. Collected faeces was dried overnight in an oven at 100^{o}C. Pooled faeces of each 'diet group' were used for nutrient fractionation and to conduct experiments on nutrient release. Experiments were also conducted on the effect of temperatures (20^{o}C & 25^{o}C) and pH (4,7,10) on the release of nutrients from fish food since uneaten food is the major source of aquaculture wastes (also confirmed in the present study as demonstrated in chapter 5.). Diet-1 contained 8.48% nitrogen (53% protein) and 1.31% phosphorus while diet-2 contained 7.2% nitrogen (45% protein) and 1.16% phosphorus (see Table 2.1 for proximate composition of the two diets). Protein (N x 6.25) and total phosphorus in feed, fish and faeces were determined following AOAC (1990a, 1990b) (see Chapter 2 for details on methodology). Total nitrogen, ammonium (NH_4^+), nitrite (NO_2^-), nitrate (NO_3^-), total P and orthophosphate (PO_3^-) of water samples were analyzed with a Tecator flow injection analyzer (Aquatec 5400 analyzer)

following the Aquatec instruction manual (Tecator 1990) (details are given in chapter 2.2). pH was measured using a pH meter (Orion model SA 520). All food and faeces were dried at 100°C overnight for moisture analysis.

Fractionation of nutrients

Phosphorus fractionation

The inorganic phosphorus in fish food and faeces were fractioned following the extraction methods of Pettersson (1988) in order to separate the orthophosphate fractions that are directly available to plants (labile fractions) and the fractions bound in the sediments (iron, aluminium and calcium bound phosphorus) (Table 8.1.). 25 mg of food and faeces was incubated with 10 ml of solution in centrifugation tubes (number of replicates were four). After centrifugation, orthophosphate was determined in the supernatant.

Table 8.1. Extraction of orthophosphate from fish food and faeces.

Extraction no	Extraction methods	Extractable phosphorus
Extraction-1*	Distilled water four times for 90 minutes	Water soluble phosphorus
Extraction-2*	NH_4CL two times, 120 minutes each	Water soluble phosphorus
Extraction-3	NaOH (0.1 mol/l) for 16 hours	Fe and Al bound phosphorus
Extraction-4	HCl (0.5 mol/l) for 24 hours	Ca bound phosphorus

* phosphorus extracted is directly available to plants.

Nitrogen fractionation

Dried faeces collected from the two diet groups was fractioned into nitrogen composition (total nitrogen, nitrite, nitrate and ammonium) following the methods described in Kristiansen and Hessen (1992) (number of replicates were three). The faeces were crushed into a fine mass of particles using a mortar and then mixed with 2 decilitres of water and sonicated (Cole-Palmer 8853). Before analyzing water was

added to yield a concentration of 1 g dry faeces l^{-1} and nitrogen content analyzed using a Tecator flow injection analyzer (Tecator 1990).

Release of nutrients from faeces and foods

Release of nutrients from faeces

The release of phosphate and ammonium from faeces was determined by incubating 75 mg faeces in 50 ml distilled water in an Erlenmeyer flasks at 25°C for a week following the method of Pettersson (1988) (number of replicates were four). Water was sampled everyday using a plastic syringe.

Effect of temperature and pH on the release of nutrients from fish food

300 mg of fish food (diet-2) was incubated with 200 ml distilled water for seven days in glass bottles. The food was incubated at each of two temperatures [(20°C and 25°C) at three pH levels (4.0, 7.0 and 10.0)] in order to observe the effect of temperature and pH on the release of nutrients (number of replicates were four). The fractions measured were dissolved phosphorus (orthophosphate) and ammonium, the nutrient fraction which impact most on water quality and aquatic living organisms according to Bostrom *et al.* (1988a, 1988b), and Welch and Lindell (1988).

Statistical analysis

Mean, standard deviation and standard error of phosphorus and nitrogen fractions of fish food and faeces were calculated following Zar (1984). One way analysis of variance (ANOVA) was used to compare the release nutrients (orthophosphate and ammonium) from faeces from the two diet groups. ANOVA's were calculated using an IBM compatible MS Excel programme. Best curves were fitted as appropriate to indicate the trend or correlations in data on the release of ammonium and orthophosphate (Cricket Graph-III, version 1.0).

RESULTS

Fractional composition of phosphorus in feed and faeces

Water soluble phosphorus (extraction-1) varied between 1.87-2.57 mg P g^{-1} dw in diets. The diet with the highest total phosphorus content also had the highest concentration of water-soluble phosphorus ($P>0.05$; $n=4$). This fraction ranged from 28.8 to 29.0% of the total phosphorus content. Both fish food and faeces contained major phosphorus fraction in a labile form represented by the fraction extracted by water and ammonium chloride (Table 8.2).

Table 8.2. Fractional composition of phosphorus (orthophosphate) in fish food and faeces (mg P g^{-1} dry weight). Faeces-1 and faeces-2 are from the diet-1 and diet-2 respectively fed to silver perch. (Details on extraction procedures are given in Table 8.1).

	Diet -1	Diet -2	Faeces-1	Faeces-2
H_2O-P	2.57 ± 0.09	1.87 ± 0.05	1.03 ± 0.04	0.94 ± 0.04
NH_4Cl- P	1.68 ± 0.19	1.44 ± 0.05	1.46 ± 0.21	0.94 ± 0.12
NaOH-P	2.33 ± 0.31	1.68± 0.14	1.10 ± 0.17	0.68 ± 0.25
HCl-P	2.25 ± 0.14	1.50 ± 0.33	1.00 ± 0.18	0.71± 0.01

[1]Values are mean±SE.

[2]n=4.

Nutrient content in faeces

Faecal analysis revealed a higher total phosphorus content and lower nitrogen content in faeces of silver perch in diets. Faeces of diet group-1 and diet group-2 contained 3.08±0.20% and 2.00±0.18% phosphorus and 3.29±0.29% and 3.21±0.51% nitrogen by weight. However, there was no significant differences in faecal phosphorus and faecal nitrogen content of the two diet groups tested ($P>0.05$). This may confirm that the main path of phosphorus loss in fish is through faeces.

Fractional composition of nitrogen in faeces

Various nitrogen components in faeces of silver perch are summarized in Table 8.3. It reveals that ammonium (5.65-6.0%), nitrite (2.35-2.38%) and nitrate (5.23-6.31%) are relatively low in the faeces compared to total N (86.44-85.62%) (dry weight basis).

Table 8.3. Nitrogen content of dry faeces of silver perch reared for four weeks at 25^{O}C and fed two diets (mg N g^{-1} DW).

Diet-group	Total-N	NO_3-N	NO_2-N	NH_4-N
1	25.90 ± 0.10	1.92 ± 0.21	0.71 ± 0.10	1.71 ± 0.04
2	25.76 ± 0.13	1.56 ± 0.28	0.72 ± 0.07	1.78 ± 0.03

[1]Values are mean±SE.
[2] n=3.

Release of nutrients from faeces

There was a rapid and momentary release of phosphorus from faeces for the first few days and thereafter a growth of bacteria appears to slowed down the release of phosphate (Figure 8.1.). Ammonium release from faeces was slow and increased slowly with time (Figure 8.1.).

Effect of temperatures and pH on the release of nutrients

Release of nutrient from the fish feed was directly related to temperature of incubation with the rate of release significantly higher at a higher temperature ($P<0.05$). The release of phosphorus and ammonium were accelerated at 25^{O}C in comparison to release at 20^{O}C (Figure 8.2, 8.3, 8.4, 8.5, 8.6, 8.7). The release of orthophosphate was higher in acidic medium (pH 4.0) (Figure 8.2, 8.3, 8.4) whereas ammonium release was accelerated in neutral to alkaline media (pH 7.0 and 10.0) (Figure 8.6, 8.7, 8.8.).

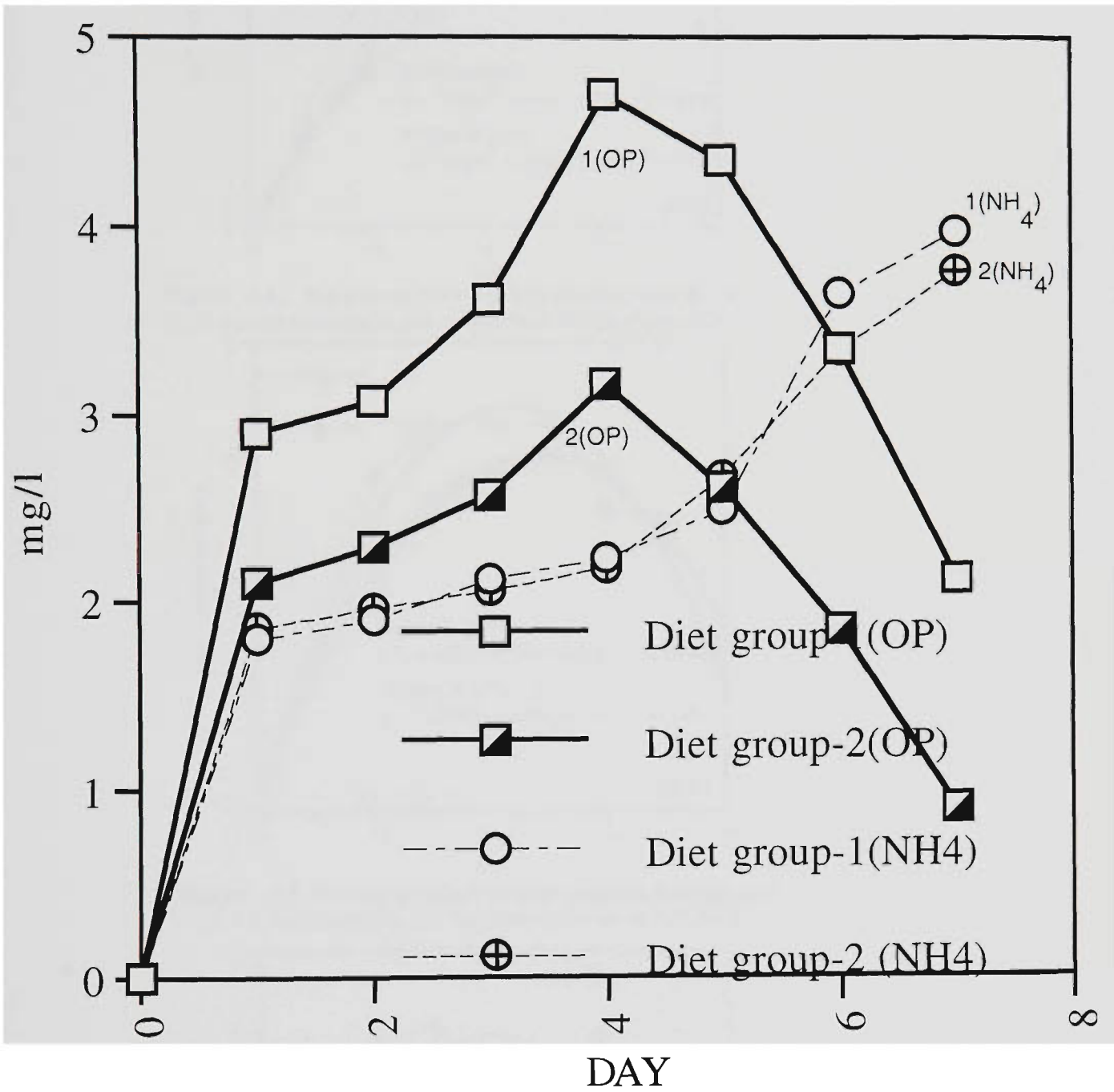

Figure 8.1. The average release of orthophosphate and ammonium from faeces of silver perch. The faeces were incubated at 25 degree celcius for seven days. There was no significant difference in the release of either orthophosphate or ammonium between the two diet groups ($P>0.05$) (n=4). [Legend : OP = orthophosphate; NH_4 = ammonium].

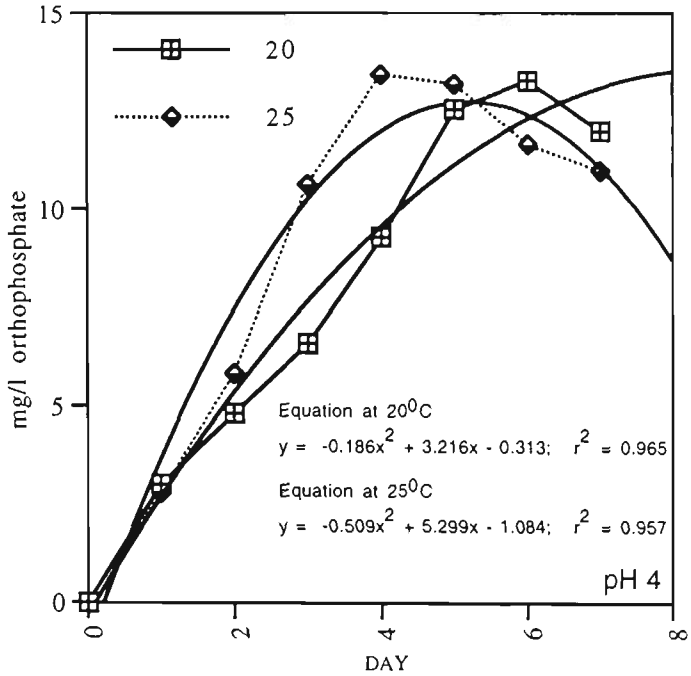

Figure 8.2. The average release of orthophoshate from fish food at pH 4.0 and incubated at two temperatures for seven days (n=4).

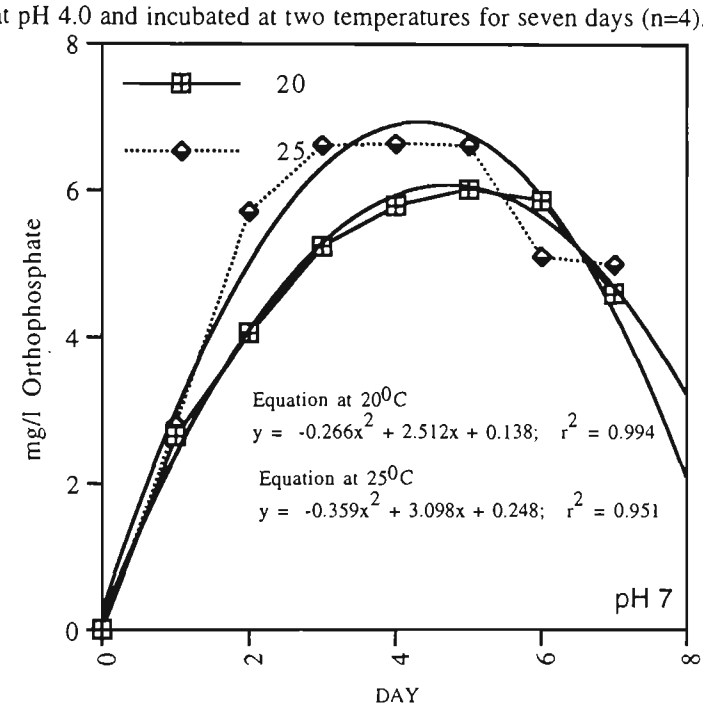

Figure 8.3. The average release of orthophosphate from fish food at pH 7.0 and incubated at two temperatures for seven days (n=4).

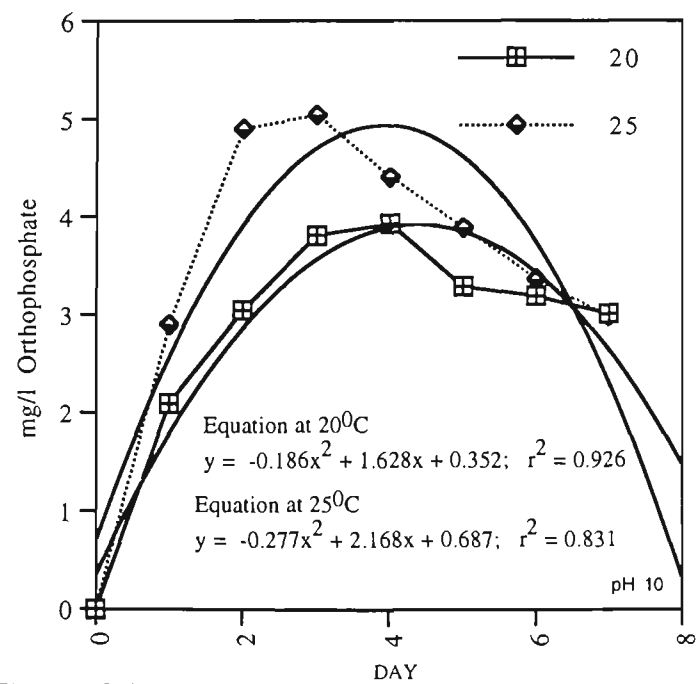

Figure 8.4. The average release of orthophosphate from fish food at pH 10.0 and incubated at two temperatures for sevendays (n=4).

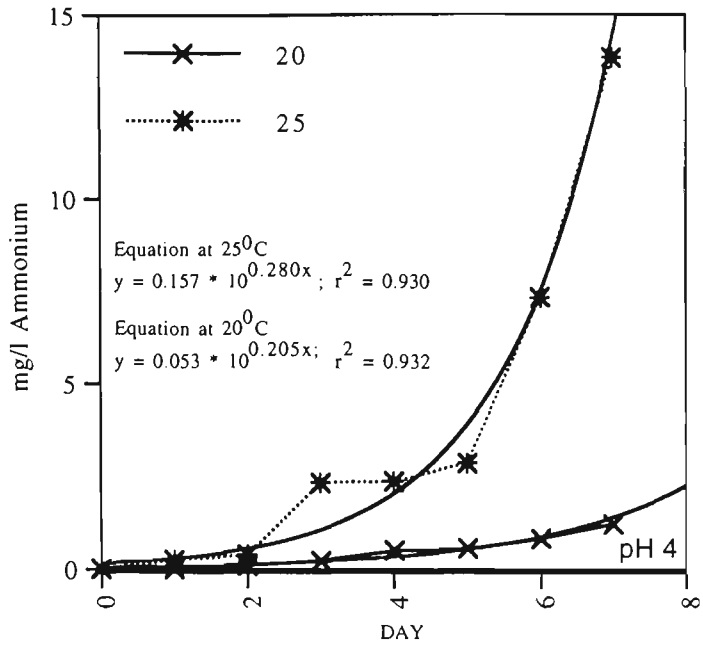

Figure 8.5. The average release of ammonium from fish food at pH 4.0 and incubated at two temperatures for seven days (n=4).

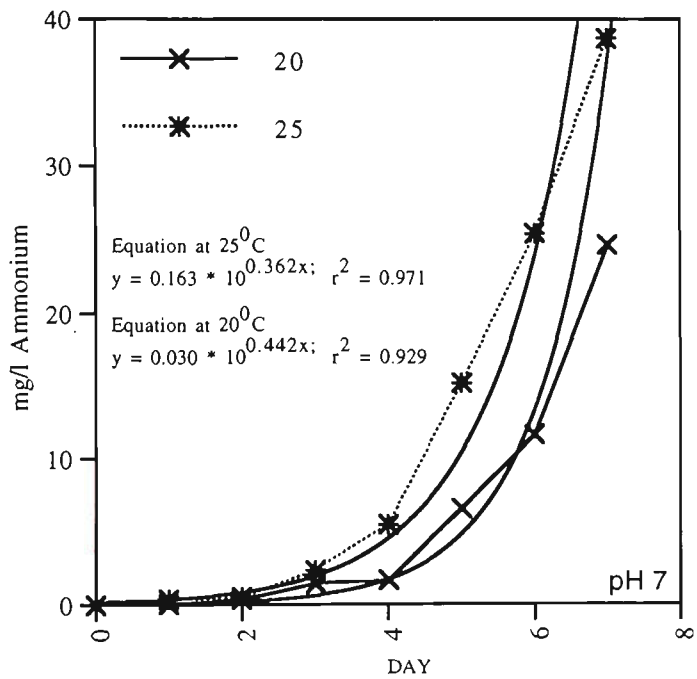

Figure 8.6. The average release of ammonium from fish food at pH 7.0 and incubated at two temperatures for seven days (n=4).

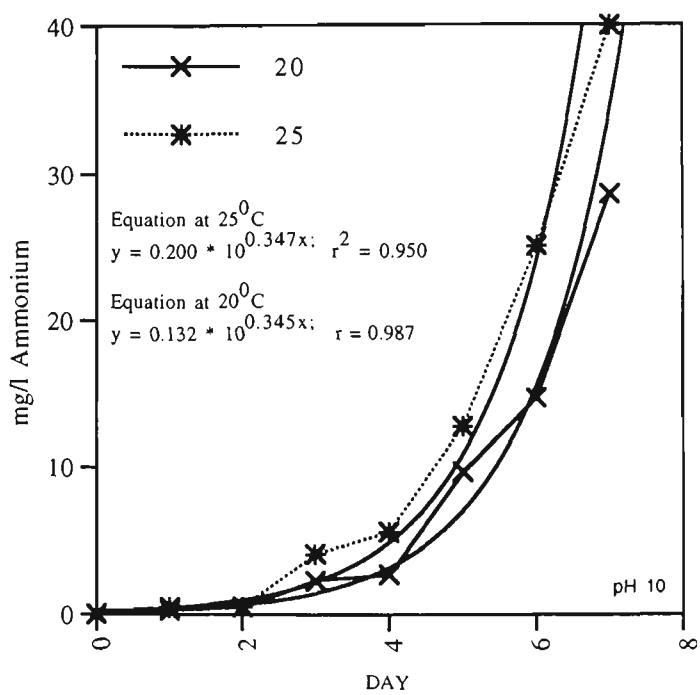

Figure 8.7. The average release of ammonium from fish food at pH 10.00 and incubated at two temperatures for seven days (n=4).

DISCUSSION

The fractional composition of phosphorus revealed a higher water soluble fraction in both fish food and faeces and this was higher with a diet containing a higher phosphorus content (Table 8.2). Pettersson (1988) found a higher concentration of water soluble phosphorus in diets containing the higher total phosphorus. In the current study fish food and faeces contained the major phosphorus fraction in a labile form represented by the fraction extracted by water and ammonium chloride (Table 8.2.). The maximum labile fraction in rainbow trout food was reported to be 26-38% and in faeces 15-54% by Pettersson (1988). The present experimental results also show that uneaten food and faeces may contain a majority of phosphorus in a labile form (49.00%-51.00% in fish food; 54.24%-57.14% in faeces) and the labile forms are believed to be readily available to plants for their growth (Butz and Vens-Cappell 1982).

The analysis of faeces revealed that silver perch faeces contains a higher phosphorus content than nitrogen. A high phosphorus loss through faeces was also found by Pettersson (1988) in rainbow trout, and Johnsen *et al.* (1993) in Atlantic salmon. Hakanson *et al.* (1988) calculates that of the total phosphorus and nitrogen fed to fish, 70% phosphorus and 15% nitrogen loss was through faeces. Kristiansen and Hessen (1992) reported 4.0% phosphorus and 2.3% nitrogen in faeces of Atlantic salmon (*Salmo salar*) and a loss of 3.5% phosphorus and 4.1% nitrogen in noble crayfish (*Astacus astacus*) faeces, while the faecal analysis of rainbow trout (*Salmo gairdneri*) yielded 1.59% phosphorus and 3.93% nitrogen (Penczak *et al.* 1982). The average phosphorus and nitrogen lost in silver perch through faeces were 2.54% and 3.25% respectively. The phosphorus and nitrogen ratio in silver perch diets were 1:6.5 and 1:6.2 in diet-1 and diet-2 while the P : N ratios in the faeces were 1 : 1.1 and 1:

1.61 for diet group-1 and diet group-2 respectively (dry weight basis). This agrees with the results of Pettersson (1988) who found that the phosphorus : nitrogen ratio in the foods was 1 : 5 while in faeces it was 1: 1-2.

In the fractional analysis of silver perch faeces, contribution of ammonium accounts for 5.6-6.0% of the nitrogen content. The ammonium contribution was also minimal in crayfish and salmon faeces being 4.0-4.4% of total nitrogen (Kristiansen and Hessen 1992). Foy and Rosell (1991a) fractioned nitrogen loadings from a Northern Ireland fish farm where they estimated that nitrite and nitrate contributed about 2.4% of the total nitrogen loadings to the environment. In the current study, the nitrite contribution in silver perch faeces was 2.35%-2.38%.

The experiments on the release of nutrients from faeces demonstrated that leaching of nutrients from faeces or food could be instantaneous and therefore nutrient enrichment of the environment occurs almost immediately (Figure 8.1). The present results is in agreement of Pettersson (1988) who also observed a rapid release of phosphate from fish food during the first few days and thereafter a growth of bacteria decreased the phosphate concentration. Both Makinen *et al.* (1988) and Phillips *et al.* (1993) stated that an efficient and quick removal of solid wastes is essential if nutrient loadings to the environment are to be controlled.

Both pH and temperature were found to influence in the release of nutrients (orthophosphate and ammonium) from silver perch food (Figure 8.2, 8.3, 8.4, 8.5, 8.6, 8.7). Persson (1988) also found that the release of nutrients from fish food and faeces depends upon pH, temperature, oxygen, water turbulence and the microbial activity of the environment (see chapters 1.4 and 1.5 on nitrogen and phosphorus pollution for more information). The patterns of phosphate and nitrogen release observed in the current study were similar to the results of Pettersson (1988). Pettersson (1988) also

reported a much higher phosphorus release in acidic medium (pH 5.0) and found that the leakage of ammonium from fish food was temperature dependent. The release of ammonium and orthophosphate from silver perch food was similar to the release from salmon excreta being much higher at higher temperature (Kristiansen and Hessen 1992).

In conclusion, the results obtained in this study show that the main path of phosphorus loss to the environment in the culture of silver perch is via faeces. The maximum release of phosphorus and nitrogen from solid wastes depends upon temperatures and pH of the environment, being higher at 25^{o}C than at 20^{o}C. The release of phosphorus increased with decrease in pH whereas ammonium release increased with increase in pH.

Chapter 9

EFFECT OF SALINITY ON BIOLOGICAL GROWTH AND NUTRIENT RETENTION IN SILVER PERCH

9

EFFECT OF SALINITY ON BIOLOGICAL GROWTH AND NUTRIENT RETENTION IN SILVER PERCH

INTRODUCTION

The silver perch is native to the Murray-Darling River system (Merrick and Schmida 1984) and migrates entirely within freshwater (Guo *et al.* 1995). The fish and crayfish of the Murray-Darling River system are known to tolerate considerably high salinities (Guo *et al.* 1993). There is a wide variation in the ability of fish to acclimate to increasing or decreasing salinity (Stickney 1991). Silver perch is least tolerant to high salinities (Guo *et al.* 1995). It is reported that freshwater species inhabit brackishwater generally without an adverse effect on the species (Morrissy 1978; Mills and Geddes 1980). There is an advantage in the aquaculture of a freshwater species which can tolerate certain range of salinity since the species has a wider environmental tolerance (Jones 1990).

The silver perch is a freshwater species that can tolerate salinity up to 15 ppt (Guo *et al.* 1995). A better survival and growth rate were reported when both larvae and juvenile were reared in slightly saline waters (Guo *et al.* 1995; Ingram *et al.* 1996). Furthermore, there was no significant effect on the development of silver perch eggs in a salinity of up to 9 ppt (Guo *et al.* 1993). Silver perch is an Australian native fish of highest aquaculture potential (Allan and Rowland 1996) and interest to culture the species is growing in both Australia and nearby Asia (Gooley and Rowland 1993; Kibria *et al.* 1996a). Until now there has been no attention paid to the effect of salinity on the growth and nutrient retention by silver perch. The objectives of this preliminary study were to examine the effect of different salinities on the growth, food conversion and nutrient retention in silver perch, *Bidyanus bidyanus* reared at 25^{o}C.

MATERIALS AND METHODS

The experiment was conducted in the wet laboratory at the University. Juveniles of silver perch (av. wt. 445 mg) were bought from a local native fish farm where fish were grown in earthen ponds. They were acclimatized in the laboratory in four separate freshwater holding tanks for four weeks and thereafter the salinity level of holding tanks was increased slowly as appropriate. Fish were kept to the desire test salinity levels for two weeks prior to the start of feeding trials. They were reared in small glass aquaria (30 x 16 x 17 cm) at 25^{O}C, since the best growth and nutrient retention in silver perch were obtained at this temperature (see chapter 3, 6 and 7). Fish were fed on a commercial silver perch diet (diet-2) containing 45% protein and 1.16% phosphorus (see Table 2.1 for proximate composition) at the rate of 3% of their body weight (as recommended). The feed was divided into two portions and fed twice a day (0900 and 1600), six days a week for four weeks to study on the growth, food conversion and nutrient (nitrogen and phosphorus) retention at four salinities (0 salinity, 4 salinity, 8 salinity and 12 salinity). Artificial sea salt (Red Sea Salt, Red Sea Fish pHarm, Israel) were used to prepare the required salinities following instructions on the packaging.

Four fish were stocked in individual glass aquaria and six replicated aquaria were prepared for each salinity regime ie., 24 fish per salinity. Individual aquaria were fitted with air stones to enhance dissolved oxygen content. The water quality was maintained similarly as described in chapter 2. The relative gain in weight, specific growth rate (SGR) and food conversion ratio (FCR) were calculated following appropriate formulae given in chapter 2.6. Samples were weighed using an analytical balance (Mettler AE 200). Proximate analysis was done following AOAC (1990a, 1990b), as given in chapter 2.

Mean, standard deviation and standard error were calculated following Zar (1984). All percentage data were transformed to arc sin values prior to analysis using an IBM computer. One way analysis of variance (ANOVA) were used to compare nutrient retention at different salinities using an IBM compatible MS Excel programme (version 5.0).

RESULTS

The figure 9.1. shows the growth of silver perch at different salinities. Faster growth was observed at 4 salinity compared to other salinities ($P<0.05$). The specific growth rate (SGR), and food conversion ratio (FCR) were also significantly better at 4 salinity (Table 9.1.). No mortality was observed of fish reared at different salinities.

Both nitrogen and phosphorus retention were significantly higher at 4 salinity, compared to other salinities ($P<0.05$). At 4 salinity 61% nitrogen and 65% phosphorus fed to fish were retained. Overall nutrient retention was better at salinities above 0 (Table. 9.1, 9.2).

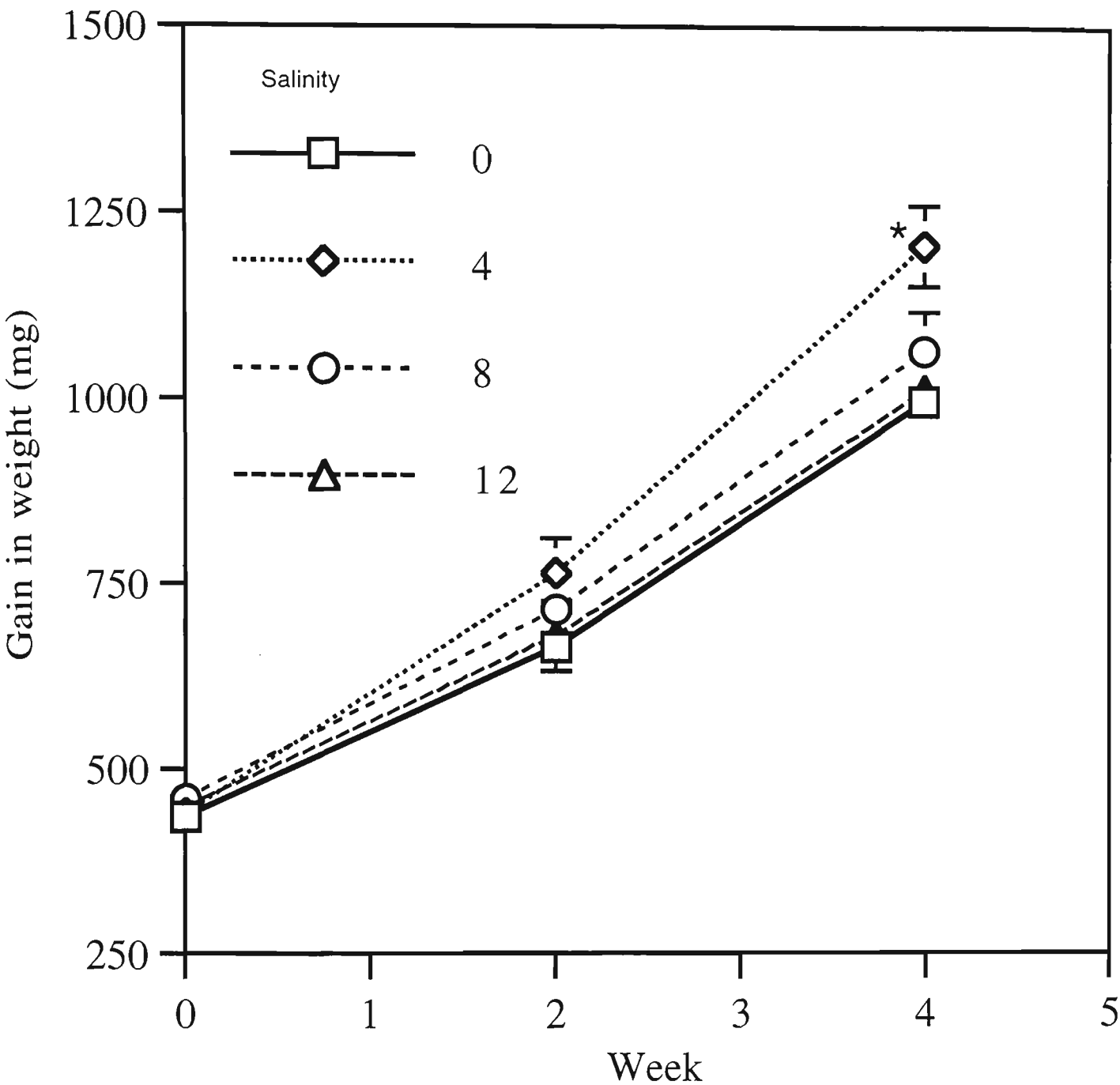

Figure. 9.1. Gain in weight of silver perch juveniles at different salinities. Fish were fed on a 45% protein diet and reared at 25 degree celcius for 28 days. The gain in weight was significanlty higher at 4 salinity ($P<0.05$) (mean$\pm$SE; n=6).

* = denotes significantly different

Table 9.1. Performance of silver perch at four different salinities. Fish were fed on a 45% protein diet @ 3% of body weight, twice a day and six days a week and reared at 25°C for 28 days.

Salinity---------->	0	4	8	12
Initial average weight (mg)	435	439	459	448
Final average weight (mg)	997	1206	1065	1012
Gain in weight (mg)	562±26[a]	767±16[b]	606±22[c]	564±24[a]
Relative weight gain	129[a]	175[b]	132[a]	126[a]
Specific growth rate (SGR)	3.45±0.08[a]	4.21±0.04[b]	3.51±0.07[a]	3.39±0.07[a]
Food conversion ratio (FCR)	1.18±0.06[a]	1.09±0.12[b]	1.17±0.06[a]	1.22±0.08[a]
Survival (%)	100	100	100	100

[1]Values are mean±SE; n=6.

[2]Values in the same row with common superscripts are not significantly different from each other (P>0.05).

Table 9.2. Nitrogen ingested, retained and released (g/kg body weight per day) by silver perch reared at different salinities.

Salinity---------->	0	4	8	12
N ingested (g/kg bw/d)	2.16±0.3	2.16±0.14	2.16±0.24	2.16±0.28
N retained (g/kg bw/d)	0.93±0.01[a]	1.32±0.02[b]	1.22±0.06[c]	0.97±0.03[a]
N released (g/kg bw/d)	1.23	0.84	0.94	1.19
N retained/N ingested (%)	43.15±1.28[a]	61.34±1.63[b]	56.34±4.63[c]	45.12±3.58[a]

[1]Values are mean±SE; n=6.

[2]Values in the same row with common superscripts are not significantly different from each other (P>0.05).

Table 9.3. Phosphorus ingested, retained and released (g/kg body weight per day) by silver perch reared at different salinities.

Salinity---------->	0	4	8	12
P ingested (g/kg bw)	0.347±0.05	0.347±0.06	0..347±0.03	0.347±0.01
P retained (g/kg bw)	0.157±0.01[a]	0.225±0.04[b]	0.168±0.02[a]	0.163±0.04[a]
P released (g/kg bw)	0.190	0.122	0.179	0.184
P retained/P ingested (%)	45.42±0.41[a]	65.00±3.6[b]	48.36±3.53[a]	47.00±2.36[a]

[1]Values are mean±SE; n=6.

[2]Values in the same row with common superscripts are not significantly different from each other (P>0.05).

DISCUSSION

The results show that weight gain, specific growth rate and food conversion ratio were higher in saline water than in freshwater. The best performance was achieved at 4 salinity. (Table 9.1.). Smith and Thorpe (1976) reported a faster growth of trout in salt water than in freshwater. A better survival and growth of silver perch larvae and juveniles were also found in slightly saline water (6 salinity) by Guo *et al.* (1993). Ingram *et al* (1996) reported a good survival and growth of silver perch when reared at lower salinities of 8.0-15.3, whereas a poor growth and survival resulted at higher salinities of 9.5-24.6. In general, most freshwater species under aquaculture showed a better performance in slightly saline water ranging from 2 to 5 salinities than in freshwater (Stickney 1991), for example, channel catfish at 5 salinity (Burnside *et al.* 1975), silver carp at 3-4 salinities (Oertzen 1985) and common carp at 3 salinity (Lam and Sharma 1985). This preliminary study also showed a better performance of silver perch at a salinity of 4 ($P<0.05$), which is close to the results obtained by other researchers (Burnside *et al.* 1975; Lam and Sharma 1985; Oertzen 1985).

The FCR was significantly better at 4 salinity than at other salinities tested. The rainbow trout showed the best food conversion efficiency in brackish water than in freshwater (Murai and Andrews 1973). Coho salmon also resulted a better food conversion at 5-10 salinities than either in freshwater or in higher salinities of more than 10 (Otto 1970). Therefore a better FCR obtained with silver perch at lower salinity is close to the results of Otto (1970) and Murai and Andrews (1973).

There was no mortality of silver perch reared at different salinities. The better growth of silver perch in slightly saline waters may be related to inhibition of diseases in saline environment (Guo *et al.* 1995). Saltwater (3 salinity) has been used to control diseases of silver perch eggs and a bath of 10 salinity was recommended to kill parasites in larvae and fry (Rowland and Ingram 1991). Therefore, better performance

of silver perch observed in slightly saline water in this study could be due to more healthier environment and a possible lacking of parasites and diseases incidence. Moreover, the better growth at lower salinities may be related to the ability of silver perch to maintain a stable internal environment at lower salinities (Eckert *et al.* 1988).

The nutrient retention with silver perch was higher in saline waters than in freshwater (Table 9.2, 9.3.). This is encouraging since a higher nutrient retention is desirable for a profitable farming and for a sustainable aquaculture programme. The nitrogen retention efficiency was also reported to be higher when trout were grown in saltwater than in freshwater (Smith and Thorpe 1976).

It is concluded that silver perch can tolerate saline conditions with no adverse effect in salinities as high as 12 salinity and growth and nutrient retention were enhanced at 4 salinity. Freshwater species which can be cultured in slightly saline waters provide a significant advantage in that they can be used to expand aquaculture in brackish waters (Smith *et al.* 1986). This is particularly important since Australia has vast areas of brackish waters and freshwater systems are limited in this country. Further research using saline water will be required to confirm the suitability of brackish water sites for the aquaculture of silver perch.

Chapter 10

GENERAL CONCLUSION AND RECOMMENDATIONS

10

GENERAL CONCLUSION AND RECOMMENDATIONS

The present study has investigated the biological growth [(weight gain, specific growth rate (SGR), food conversion ratio (FCR), protein efficiency ratio (PER)] and pollution potential from aquaculture of Australian fish silver perch, *Bidyanus bidyanus*. This study also investigated the utilization of sewage grown zooplankton with reference to biochemical composition of zooplankton and performance of silver perch fed on two dominant species of sewage zooplankton. A preliminary investigation was also conducted on the biological growth and nutrient retention by silver perch at different salinities. Silver perch juveniles were fed on three commercial diets referred to as diet-1, diet-2 and diet-3 and fed at the rate of 3% of their body weight. The rearing temperatures were 20^{o}C, 25^{o}C and 30^{o}C in different experimental situations (chapter 3-9). The protein and phosphorus content of the three diets are : diet-1 (53% protein, 1.31% phosphorus), diet-2 (45% protein, 1.16% phosphorus) and diet-3 (36% protein, 1.28 phosphorus). Silver perch was chosen in this study since silver perch is a major native aquaculture industry in the country and there was no previous research on the quality and quantity of waste discharge from aquaculture of silver perch.

Biological growth of silver perch fed on commercial diets

Of the three diets fed to silver perch, fish fed on diet-2 showed a slightly higher weight gain at each temperature of 20^{o}C, 25^{o}C and 30^{o}C; although weight gain of fish fed the three diets was not significantly different (P>0.05) (chapter 3). A trend of higher SGR, FCR and PER were also obtained with diet-2 at all temperatures (P>0.05). Results on biological growth may indicate that silver perch juveniles do not require the

higher protein diet used in this study to achieve the best growth and a diet containing 45% protein may be sufficient to achieve a good growth rate.

When silver perch juveniles were reared at different temperatures, a significantly higher growth rate and a significantly better SGR, FCR, PER were obtained at 25°C. The maximum growth achieved at 25°C may suggest that the optimum growing temperature of silver perch is close to this temperature. This result supports the view that the optimal temperature for silver perch lies between 20°C-30°C (Barlow 1986) or more precisely inbetween 23°C-28°C (Rowland *et al.* 1995). Fish reared at 20°C and 30°C had a lower weight gain than those reared at 25°C. Further research at 1°C or 2°C interval may pinpoint the exact optimum temperature of silver perch.

Utilization of sewage grown zooplankton

At the Werribee Sewage Treatment Lagoons (WSTL), two species of zooplankton are abundantly available, these are, *Daphnia carinata* and *Moina australiensis*. These two zooplankton were subjected to analysis. The analysis reveals that both zooplankton had a biochemical composition which was useful for aquaculture of silver perch. The protein content in *D. carinata* and *M. australiensis* were 64.80% and 54.34% respectively. The other biochemical composition also revealed that the zooplankton contained an appreciable concentration of lipid (7.29-7.73%), and phosphorus (1.11-1.16) (Chapter 4). The amino acids contents of both zooplankton matched the amino-acids requirement of fish. Lysine and methionine, the most limiting amino acids in normal fish feed are also present in appreciable levels in both zooplankton.

Fish fed on frozen *D. carinata* grew at the same rate as fish fed on commercial silver perch diet, and the growth, SGR, and FCR were not significantly different between fish fed on *D. carinata* and silver perch diet ($P>0.05$) (Chapter 4). Fish fed on

M. australiensis alone or in combination with other diets performed poorly. This may indicate that silver perch juveniles may have a preference for *D. carinata* over *M. australiensis*. Therefore, the current study provided some indication that zooplankton could be an alternative diet in the rearing of silver perch. Results also indicate that there was no increase in the heavy metals within the carcass of fish fed sewage zooplankton in the short term.

In nature, the main food of silver perch are zooplankton (chapter 2.1), therefore more research should be undertaken on the utilization of this vast and unutilized volume of zooplankton grown at WSTL which is being drained into Port Phillip Bay by the Melbourne Water Authority. During 1995-96, an average of 40-84 kg/h zooplankton was harvested from WSTL (source : Zootech Private Research Group, Werribee). Utilization of this resource through aquaculture would recycle waste to produce a valuable aquatic food source. Future research programmes may include the feasibility of using zooplankton meal as an ingredient of fish feed, and total or partial replacement of expensive protein meal by zooplankton meal. Success of such research may enable the production of less expensive silver perch diets and thus reduce production costs of silver perch. It may also be useful to investigate the seasonal variation in protein, lipid and other mineral contents of zooplankton, to investigate ways to minimize the loss of nutrients from frozen zooplankton, and to study the removal of the indigestible chitinous fraction from zooplankton. All this information is necessary in the development of a zooplankton based dry diet and the use of sewage grown zooplankton in the aquaculture or aquarium industry.

Solid waste production from silver perch aquaculture

Two experiments were conducted on the production of solid waste load (uneaten food and faeces) by rearing silver perch juveniles at different temperatures (chapter 5). The first experiment conducted on the production of suspended and dissolved solids

revealed that the diet which resulted in slightly higher weight gain and a better FCR to fish, produced less suspended and dissolved solids ($P>0.05$). In the second experiment, a higher solid output was obtained at $20^{o}C$ and the lowest solid output at $25^{o}C$ from the rearing of fish at $20^{o}C$, $25^{o}C$ and $30^{o}C$. At $20^{o}C$, the dominant waste was uneaten food accounting for 25% of food offered. The faecal production was found to be higher at $30^{o}C$ in the order of $30^{o}C>25^{o}C>20^{o}C$. Fish fed on diet-2 consistently produced less solid waste output among fish fed on three diets at $20^{o}C$, $25^{o}C$ and $30^{o}C$. Fish reared at $25^{o}C$ produced significantly lower solid waste output. The estimated solid waste at $20^{o}C$, $25^{o}C$ and $30^{o}C$ were 549kg · $tonne^{-1}$, 284 kg · $tonne^{-1}$ and 393 kg · $tonne^{-1}$ fish produced respectively ($P<0.05$).

The study shows that the production of solid waste in silver perch culture could be affected by diets and rearing temperature. This finding agrees with other researchers who found that diet, feeding rate and rearing temperature can influence in the quantity of solid waste output (Kilambi *et al.* 1977; Butz-and Vens-Cappell 1982; Merican and Phillips 1985). Fish reared at $25^{o}C$ produced lowest solid waste load, and this could be related to the higher growth and better FCR obtained at $25^{o}C$. The rearing experiment conducted showed that if silver perch were reared at a lower temperature, solid output could be higher since a substantial amount of food offered would remain uneaten. Results also show that rearing fish at a temperature above optimum may increase the solid output through increase in faecal loss. This has implications for the environmental pollution of freshwater habitats from the aquaculture of silver perch.

Nitrogen loss and nitrogen retention in silver perch

In order to investigate nitrogen losses and nitrogen retention, silver perch juveniles were fed on three commercial diets and reared at two temperatures ($25^{o}C$ and $30^{o}C$) (Chapter 6). The main path ways of nitrogen loss was found to be gill excretion (85.7%-90.2%) and faeces (9.8%-14.3%). The hourly ammonia excretion rate showed

a sharp increase soon after a meal and a linear decrease during the remaining 24 hours and this rate was not significantly different in fish fed on the three different diets at 25°C ($P>0.05$). The daily ammonia excretion in the culture of silver perch was significantly higher at 30°C than at 25°C ($P<0.05$), similarly the faecal nitrogen loss was also higher at 30°C than at 25°C ($P<0.05$) which may confirm the results obtained by others showing that at higher temperature the nitrogen losses can also be higher (Jobling 1981; Kaushik 1981).

Nitrogen retention by silver perch was found to be significantly greater at 25°C than at 30°C ($P<0.05$), and average nitrogen retained at 25°C and 30°C were 43.1% and 29.4% respectively ($P<0.05$). The higher nitrogen retention at 25°C may be related to the better food conversion ratio (FCR) and lower nitrogen loss obtained at 25°C with silver perch. The above experiments on nitrogen losses and nitrogen retention with silver perch shows that culture of fish at their optimum temperature may enhance nitrogen retention and result in a reduction in the discharge of nitrogen to the environment. Since nitrogen is an expensive ingredient in fish feed a higher nitrogen retention is also desirable for profitable farming. Additionally the amount of nitrogen retained by fish could also be used to evaluate a diet (Cho 1993). The present study demonstrates that growing of fish close to their optimum temperature could be a simple way of achieving higher nitrogen retention and a lower nitrogen output to the environment. Further studies are required to determine the optimum protein requirements of silver perch that result in maximum nitrogen retention and minimum nitrogen pollution of the environment. The study should use different sizes of silver perch and different feeding strategies (for example alternate feeding of low and high protein diets as suggested by De Silva *et al.* 1993).

Phosphorus losses and retention in silver perch

Experiments conducted on phosphorus losses and phosphorus retention reveal that the main path of phosphorus loss in the culture of silver perch is faecal output (64.67%-66.61%) (Chapter 7). There was a sharp increase in phosphate excretion soon after a meal similar to the ammonia excretion rate (Figure 8.1) and a linear decrease during the remaining 24 hours. The daily phosphate excretion increased with increase in temperature. The phosphate excretion was found to be significantly higher at 30^{o}C than at 25^{o}C ($P<0.05$). This could be due to the lower FCR and lower phosphorus retention obtained at 30^{o}C. Of the three diets fed to silver perch, fish fed diet-2 containing 45% protein excreted comparatively less orthophosphate than either diet-1 and diet-2 at both 25^{o}C and 30^{o}C. This fact could be related to the slightly lower phosphorus content in diet-2 or due to the slightly better FCR obtained with diet-2, though the excretion rate did not differ significantly in fish fed on the three diets ($P>0.05$). Silver perch reared at 25^{o}C retained significantly more phosphorus than those reared at 30^{o}C ($P<0.05$), which may be due to the higher growth and better FCR achieved with silver perch at 25^{o}C. At 25^{o}C, phosphorus retention was 49% and at 30^{o}C it was 24.5%.

Results obtained in the current study agrees with the findings of others who reported that phosphorus discharge and retention in finfish are influenced by the FCR (Storebakken and Austreng 1987a, 1987b; Seymour and Bergheim 1991; Bohl *et al.* 1992; Cho 1993; Johnsen *et al.* 1993), type of diet (Ketola and Harland 1993; Matty 1990), and temperature (Kristiansen and Hessen 1992; Ballestrazzi *et al.* 1994). In order to minimize the phosphorus losses and maximize the phosphorus retention, further research is needed on the optimum phosphorus requirements of silver perch.

Nutrient fractionation and the release of nutrients from solid waste

Fish food (silver perch diet) and faeces were fractioned to reveal nutrient composition of solid waste from silver perch (Chapter 8). The fractionation of phosphorus showed that the diet containing the highest total phosphorus also had the highest concentration of water soluble phosphorus. Both fish food and faeces contained a major phosphorus fraction in a labile form, the fraction that is readily available to plants. Faecal analysis reveals that ammonium, nitrite and nitrate are relatively low and total nitrogen component was the main component in the faeces comprising 86.44% to 85.62% (dry weight basis). The fractionation results obtained here were similar to the composition results reported by Pettersson (1988) on phosphorus fractionation and Kristiansen and Hessen (1992) on nitrogen fractionation on fish food and faeces.

Experiments carried out on the release of nutrients showed a rapid release of phosphate and a slow release of ammonium from silver perch faeces, a similar pattern was also reported by Pettersson (1988) in rainbow trout faeces. The release of nutrients from fish food and faeces was found to be affected by temperature and pH of the environment. The release rate was accelerated at higher temperatures. Phosphate release was higher in acidic medium whereas ammonium release was higher in neutral to alkaline media. The release of nutrients from rainbow trout food and faeces were also reported to be affected by pH (Persson 1988; Pettersson 1988), and temperature (Persson 1988; Kristiansen and Hessen 1992). The present experiments demonstrate that fish food and faeces may contain a major phosphorus and nitrogen fraction in a labile form and that the leaching of nutrients from solid waste could be instantaneous. Therefore an efficient and quick removal of solid waste is essential if nutrient loading to the environment is to be controlled. Further experiments are needed on the minimization of total and labile nutrient content in silver perch diets in order to reduce the phosphorus and nitrogen load to the environment without negatively affecting the growth of the silver perch.

Effect of salinity on nutrient retention

A preliminary study was conducted on the rearing of silver perch in slightly saline water which showed a better nutrient retention at 4 salinity (chapter 9). Freshwater species which can be cultured in slightly saline waters provide a significant advantage in expanding aquaculture in brackish water. This is particularly important since Australia has a vast brackish water zone and freshwater is very limited. However further research in saline water will be required to confirm the suitability of silver perch for culture in brackish water sites.

In summary, the study demonstrates that the growth of silver perch was significantly better at a temperature close to its optimum and the zooplankton could be an alternative diet for silver perch culture. The study also shows that solid waste and nutrient load could be significantly less if silver perch are cultured at 25^{o}C. The nutrient composition study reveals that silver perch food and faeces contain a major fraction in a labile form and that the release of nutrients from fish food is affected by the temperature and pH of the environment. In order to have maximum growth, a minimum solid and nutrient load and therefore a sustainable silver perch aquaculture programme, silver perch should be cultured at a temperature close to its optimum.

Based on the above study the following future investigations are recommended

I. Experiments conducted at 1 to 2^{o}C interval between 20-30^{o}C to determine the exact optimum temperature of silver perch;

II. A detailed investigation on heavy metals and other contaminant levels in the zooplankton harvested from the Werribee sewage treatment lagoons to determine their suitability for use in aquaculture;

III. Further experiments on the optimum nitrogen and phosphorus requirements of silver perch;

IV. Research by alternate feeding of high and low nutrient diets in order to investigate reduction of the nutrient load from silver perch culture;

V. A through study of the factors affecting the release of nutrients from sediments containing silver perch wastes;

VI. Comparative studies on the biological growth and nutrient retention by silver perch in freshwater and saline waters;

Chapter 11

REFERENCES

REFERENCES

Ackefors, H. & Enell, M. (1990). Discharge of nutrients from Swedish fish farming to adjacent sea areas. *Ambio* **19**, 28-35.

Ackefors, H. & Enell, M. (1994). The release of nutrients and organic matter from aquaculture systems in Nordic countries. *Journal of Applied Ichthyology* **10(4)**, 225-241.

Adams, V. D. (1990). *Water and wastewater examination manual.* Lewis Publishers Inc., Michigan, U.S.A. 247p.

Alam, M. J., Ang, K. J. & Cheah, S. H. (1993). Use of *Moina micrura* (de Man) as an *Artemia* substitute in the production of *Macrobrachium rosenbergii* (de Man) post-larvae. *Aquaculture* **109**, 37-349.

Albaster, J. S. (1982). *Report of the EIFAC Workshop on fish-farm effluents.* Silkebog, Denmark, 26-28 May, 1981. EIFAC Technical paper 41. FAO, Rome. 166p.

Allan, G. L. (1995). Development of artificial diets for silver perch. In : *Silver Perch Culture, Proceedings of Silver Perch Workshops, Grafton and Narrandera, April 1994.* (ed. by S. J. Rowland & C. Bryant), pp. 77-87. *Austasia Aquaculture*, Hobart for NSW Fisheries, Sydney.

Allan, G. L. & Rowland, S. J. (1991). Growth of silver perch *Bidyanus bidyanus* on diets with different levels and sources of protein. *Proceeding of the Nutritional Society of Australia*, **16**: 211.

Allan, G. L. & Rowland, S. J. (1992). Development of an experimental diet for silver perch (*Bidyanus bidyanus). Austasia Aquaculture* **6**, 39-40.

Allan, G. L. & Rowland, S. J. (1994). The use of Australian oil seeds and grain legumes in aquaculture. In : *Proceedings of the Third Asian Fisheries Forum, Singapore, 26-30 October 1992.* (ed. by L. M. Chou., A. D. Munro., T. J. Lam., T. W. Cheong., J. K. Ding., K. K. Hooi., H. W. Khoo., V. P. E. Phang., K. F. Shim, & C. H. Tan), pp. 667-670. *Asian Fisheries Society*, Manila, Philippines, .

Allan, G. L. and Rowland, S. J. (1996). The silver perch industry : research progress and priorities toward developing cost-effective diets. *Austasia Aquaculture* **10(2)**, 37-40.

Allan, G. L., Rowland, S. J. & Parkinson, S. A. (1993). Replacement of fish meal with soybean meal, peanut meal, canola meal or lupins in diets for silver perch *Bidyanus bidyanus* (Mitchell). *Proceedings of the VI International Symposium on Fish Nutrition and Feeding , 4-7 October 1993*, Hobart, Australia (Abstract).

Allan, G. L., Warner-Smith, R. J. & Frances, J. (1994). The effects of varying protein and energy concentrations on growth, protein efficiency ratio and food conversion ratio for silver perch (*Bidyanus bidyanus* Mitchell). In : *Proceeding of the Nutritional Society of Australia*, **18**, 53.

Allen, G. (1989). The freshwater fishes of Australia. T. F. H. (Australia) Pty. Ltd. NSW, Australia

Allen, W. A. & Allen, G. H. (1981). Amino acids in the food web of a wastewater aquaculture system for rearing juvenile salmon. *Progressive Fish Culturist* **43(4)**,178-182.

Alsted, N. S. (1991). Study on the possibilities of reducing the discharge of organic matter from fish farming by changing the diet composition. In : *Proceedings of the International Symposium on Feeding Fish in Our Water*. Nutritional Strategies in Management of Aquaculture Waste. June 5-8th 1990, Guelph, Ontario, Canada.

Alsted, N. & Jokumsen, A. (1989). The influence of dietary protein on the growth of rainbow trout - The current state of fish nutrition in aquaculture. In : *Proceedings of the Third International Symposium on Feeding and Nutrition in Fish. Toba, Japan, 28 August-1 September* (ed. by M. Takeda & T. Watanabe), pp. 77-90.

Anderson, A. J. and Arthington, A. H. 1989. Effect of diet on the fatty acid composition of silver perch (*Leiopotherapon bidyanus*) lipids. *Comparative Biochemistry and Physiology* **93B**, 715-720.

Anon. (1970). News briefs : Culture of fish food organisms. *American Fish Farmer* **1**, 4.

Anon. (1987). Model permit package for fish culture industry. US Environment Protection Agency, New Jersey, 45p.

Anon. (1996). Daphnia - live fish food. *Aquarium Life*, Melbourne, Australia, edition no. **51**.

AOAC. (1990a). *Official methods of analysis of the Association of Official Analytical Chemists.* (ed. by K. Helrich). Fifteenth Edition. Volume I. Association of Official Analytical Chemists, Arlington, Virginia, U.S.A. 684p.

AOAC. (1990b). *Official methods of analysis of the Association of Official Analytical Chemists.* (ed. by K. Helrich). Fifteenth Edition. Volume II. Association of Official Analytical Chemists, Arlington, Virginia, U.S.A. 1298p.

APHA. (1989). *Standard methods for the examination of water and waste water.* (ed. by L . S. Clesceri., A. E. Greenberg, & R. R. Trussel), (17th edition). American Public Health Association (APHA) - American Water Works Association(AWWA) - Water Pollution Control Board (WPCB), Washington D. C. 1527p.

Arrhenius, F. & Hansoon, S. (1993). Food consumption of larval, young and adult herring and sprat in the Baltic Sea. *Marine Ecology Progressive Series* **96**, 125-137.

Asgard, T., Storebakken, T. & Austreng, E. (1986). Reduction of self-pollution by correct feeding. In : *Miljohandbook for fiskeoppdrett* (ed. J. I. Vikan), pp. 35-46. Poster at the 3rd International Conference on Aquafarming. 'AQUACOLTURA'86', Verona, Italy, 9-10 October 1986.Austreng, E. and Asgard, T. 1991. Foring av fisk i harmoni med miljoet. Aqua Books, Oslo, Norway.

Ballestrazzi, R., Lanari, D., D'Agaro, E. & Mion, A. (1994). The effect of dietary protein level and source on growth, body composition, total ammonia and reactive phosphate excretion of growing sea bass (*Dicentrachus labrax*). *Aquaculture* **127**, 197-206.

Barlow, C. G. (1986). Fish in farm dams and implications for extensive aquaculture, pages 33-43. In : *Proceedings of the First Freshwater Aquaculture Workshop, Narrandera, NSW* (ed. by L.F. Reynolds). Agriculture and Fisheries, Sydney, Australia.

Barlow, S. (1989). Fish meal-World outlook to the year 2000. *Fish Farmer* **Sept/Oct**, pp.40-43.

Barlow, C. G. (1995). Culture of silver perch in farm dams : limitations and problems. In : *Silver Perch Culture : Proceedings of Silver Perch Workshops, Grafton and Narrendera, April 1994.* (ed. by S. J. Rowland & C. Bryant), pp. 13-17. Austasia Aquaculture, Hobart for NSW Fisheries, Sydney.

Barlow, C. G. & Bock, K. (1981). *Fish for farm dams.* NSW Fisheries, Sydney.

Bergheim, A. & Asgard, T. (1996). Waste production from aquaculture. In : *Aquaculture and Water Resource Management* (ed. by D. J. Baird., M.C. M. Beveridge., L. A. Kelly and J. M. Muir). pp. 50-80. Blackell Science Ltd. Oxford, U.K.

Bergheim, A., Silvertsen, A. & Selmer-Olsen, A. (1982). Estimated pollution loadings from Norwegian Fish farms. I. Investigations 1978-79. *Aquaculture* **28**, 347-361.

Bergheim, A., Tyvold, T. and Seymour, E. A. (1991a). Effluent loadings and sludge removal from landbased salmon farming tanks. *In : Proceedings of Aquaculture Europe 9. Aquaculture and the Environment* (compiled by N. de Pauw & J. Joyce). EAS Special Publication, 14, EAS, Bredene, Belgium.

Bergheim, A., Aabel, J. & Seymour, E. (1991b). Past and present approaches to aquaculture waste management in Norwegian net pen culture operations. In : *Nutritional Strategies and Aquaculture Waste* (ed. by C. B.Cowey & C. Y Cho), pp 117-1136. University of Guelph, Ontario.

Bergheim, A., Sanni, S., Indrevik, G. & Holland, P. (1993). Sludge removal from salmonid tank effluent using rotating microsieves. *Aquacultural Engineering* **12**, 97-109.

Bernhardt, H. (1981). Z. Wasser Abwasser Forsch. 14 nr. I.S, 14-26.

Beveridge, M. C. M. (1984). Cage and pen farming : carrying capacity models and environmental impact. *FAO Fisheries Technical Paper* **255**. FIRI/T255, 131p.

Beveridge, M.C. M. (1996). *Cage Aquaculture*. Fishing News Books, Farnham, England.

Beveridge, M. C. M. & Phillips, M. J. (1993). Environmental impact of tropical inland aquaculture. In : *Environment and Aquaculture in Developing Countries*. (ed. by R.S. V. Pullin, H. Rosenthal & J. L. Maclean). pp. 213-236. ICLARM Conference Proceedings. **31**.

Beveridge, M. C. M., Phillips, M. J. & Clark, R. M. (1991). A quantitative and qualitative assessment of wastes from aquatic animal production. In : *Aquaculture and water quality. Advances in World Aquaculture.* Vol. 3 (ed. by D. E. Brune & J. R. Tomasso), pp. 506-533. The World Aquaculture Society, Baton Rouge.

Blazka, P. (1966). The ratio of crude protein, glycogen and fat in the individual steps of the production chain. Hydrobiological studies. I. Prague (ed. by J. Hrbacck), pp. 395-409. Academia Publishing House of the Czechoslovak Academy of Sciences, Prague.

Block, R. J. (1959). The approximate amino acid composition of wild and hatchery trout (*Salvelinus fantinalis*) and some of their principal foods (*Gammarus* and *Hexagenia bilineata*), *Boyce Thomson Institute of Plant Research Contribution* **20**, 103-105.

Blyth, P. J. & Burser, P. (1993). Detection of the feeding behaviour of seacaged Atlantic salmon, *Salmo salar*, using "new"adaptive' feeder technology. In : *Proceedings of the 6th International Symposium on Fish Nutrition and Feeding*, Hobart, Australia, 4-7 October 1993. (abstract)

Boersen, G. & Westers, H. (1986). Waste solids control in hatchery raceways. *Progressive Fish Culturist* **48**, 151-154.

Bohl, M., Moav, B., Hilge, V. & Rosenthal, H. (1992). The influences of feeds on trout pond effluents. In : *Progress in Aquaculture Research.* (ed. by B. Moav, V. Hilge and H. Rosenthal), pp.109-118. Special Publication European Aquaculture Society. 17.

Bostrom, B., Persson, G. & Broberg, B. (1988a). Bioavailability of different phosphorus forms in freshwater systems. *Hydrobiologia* 170, 133-155.

Bostrom, B., Anderson, J. M., Fleischer, F & Janson, M. (1988b). Exchange of phosphorus across sediment - water interface. *Hydrobiologia* 170, 229-244.

Boyd, C. E. (1971). Phosphorus dynamics in ponds. Proceedings of the Annual Conference of Southeast Association of Game and Fish Commission. **25**, 418-426.

Boyd, C. E. (1978). Effluents from catfish ponds during fish harvest. *Journal of Environmental Quality*. 7, 59-62.

Boyd, C. E. (1985). Chemical budgets for channel catfish ponds. *Transactions of the American Fisheries Society.* **11**, 191-298.

Boyd, C. E. (1990). Water quality in ponds for aquaculture. Alabama Agricultural Experiment Station, Auburn University, Birmingham, Alabama. 482p.

Boyd, C. E. & Lichtkoppler, F. (1979). Water quality management in pond fish culture. International Centre for Aquaculture, Agricultural Experiment Station, Research and Development Series No. 22, AID/DSAN-G 0039.30p.

Boyd, C. E. & Tucker, C. S. (1995). Sustainability of channel catfish farming. *World Aquaculture.* **26(3)**, 45-53.

Bratten, B. (1991). Status of pollution from aquaculture in six Nordic countries, release of pollutants, effects and waste water treatment. In : *Aquaculture and Environment* (ed. by N De Pauw and J. Joyce), pp. 40-41. European Aquaculture Society Special Publication. 14.

Brett, J. R. (1971). Growth responses of young sockeye salmon (*Oncorhynchus nerka*) to different diets and planes of nutrition. *Journal of the Fisheries Research Board of Canada* **28**, 1635-1643.

Brett, J. R. (1979). Environmental factors and growth. In : *Fish Physiology, volume VIII.* (ed. by S. W. Hoar, D. J. Randall & J. R. Brett). pp. 599-675. Academic Press, USA.

Brett, J. R. & Zala, C. A. (1975). Daily pattern of nitrogen excretion and oxygen consumption of sockey salmon (*Oncorhynchus nerka*) under controlled conditions. *Journal of the Fisheries Research Board of Canada*. **32**, 2479-2486.

Brett, J. R. & Groves, T. D. D. (1979). Physiological energetics. In : *Fish Physiology, volume VIII*. (ed. by W. S. Hoar, D. J. Randall & J. R. Bretts). pp. 279-352. Academic press Inc, London.

Brett, J. R., Shelbourn, J. E. & Shoop, C. T. (1969). Growth rate and body composition of fingerlings sockeye salmon *Oncorhynchus nerka* in relation to temperature and ration size. *Journal of the Fisheries Research Board of Canada* 26, 2363-2394.

Briggs, M. R. P. & Funge-Smith, S. J. (1994). A nutrient budget of some intensive marine shrimp ponds in Thailand. *Aquaculture and Fisheries Management*. **25**, 789-811.

Brodle, J. (1995). The problems of nutrients and eutrophication in the Australian marine environment. In : *State of marine environment report for Australia* (ed. by L. P. Zann & D. C. Sutton). pp. 1-29, Technical annex 2. Great Barrier Reef Marine Park Authority. Australia.

Bromley, J. R. (1987). The effects of food type, meal size and body weight on digestion and gastric evacuation in turbot, *Scophthalamus maximus* L. *Journal of Fish Biology* **30**, 501-512.

Brown, J. R., Gowen, R. J. & McLusky, D. S. (1987). The effect of salmon farming on the benthos of a Scottish sea loach. *Journal of the Experimental Marine Biology and Ecology*. **109**, 39-51.

Brown, M. L., Jaramillo, F. Jr., & Gatlin, D. M. (1993). Dietary phosphorus requirement of juvenile sunshine bass, *Morone chryops* male x *M. saxtilis* females. *Aquaculture* **113**, 355-363.

Bryant, B. L. & Matty, A. J. (1980). Optimisation of Artemia feeding rate for carp larvae (*Cyprinus carpio* L.). *Aquaculture* **21(3)**, 203-212.

Burnside, M. C., Avault, J. W. & Perry, W. G. (1975). Comparison of a wild and a domestic strain of channel catfish grown in brackish water. *The Progressive Fish-Culturist*. **37**, 52-54.

Butz, I and Vens-Cappell, B. (1982). Organic load from the metabolic products of rainbow trout fed with dry food. In : *Report of the EIFAC Workshop on fish farm effluents*. (ed. by J.S.Albaster). Silkeborg, Denmark, 26-28 May 1981. EIFAC Technical Paper. **41**, 57-70.

Cadwallader, P. L. (1977). J.O. Langrtry's 1949-50 Murray River investigations. Fisheries and Wildlife Paper, Victoria. No. 13. 70pp.

Cadwallader, P. L (1979). Distribution of native and introduced fish in Seven Creeks River system. Victoria, Australia. *Australian Journal of Ecology*. **4**, 361-385.

Cadwallader, P. L. & Backhouse, G. N. (1983). *A guide to the freshwater fish of Victoria*. Government Printer. Melbourne. 249p.

Cai, Y. & Summerfelt, R. C. (1992). Effects of temperatures and size on oxygen consumption and ammonia excretion by walleye. *Aquaculture* **104**, 127-138.

Callinan, R. B., & Rowland, S. J. (1995). Diseases of silver perch. In : *Silver Perch ulture : Proceedings of Silver Perch Workshops, Grafton and Narrendera, April 1994*. (ed. by S. J. Rowland & C. Bryant), pp. 67-75. Austasia Aquaculture, Hobart for NSW Fisheries, Sydney.

Cambridge, M. L., Chiffings, A. W., Brittan, C., Moore, L. & McComb, A. J. (1986). The loss of seagrass in Cockburn Sound, Western Australia. Possible causes of seagrass decline. *Aquatic Botany*. **24**, 269-285.

Carr, O. J. & Goulder, R. (1990a). Fish-farm effluents in rivers-1: Effects on bacterial populations and alkaline phosphate activity. *Water Research* **24(5)**, 631-638.

Carr, O. J. & Goulder, R. (1990b). Fish farm effluents in rivers-II. Effects on inorganic nutrients, algae, and the macrophyte *Ranunculus pencillatus*. *Water Research*. **24(5),** 639-647.

Cavaletto, J. F., Vanderploeg, H. A. & Gardner, W. S. (1989). Waxesters in two species of freshwater zooplankton. *Limnology and Oceanography* **34(4)**, 785-789.

Chamberlain, W. & Shapiro, J. (1969). On the biological significance of phosphate analysis. Comparison of standard and new methods with a bioassy. *Limnology and Oceanography* **14**, 921-927.

Chiu, Y. N. (1989). Considerations for feeding experiments to quantify dietary requirements of essential nutrients in fish. In : *Fish Nutrition Research in Asia. Proceedings of the Third Asian Fish Nutrition Network Meeting*, (ed. by S.S.De Silva). pp. 46-57. Asian Fisheries Society Special Publication 4, Asian Fisheries Society, Manila, Philippines.

Cho, C. Y. (1993). Digestibility of feedstuffs as a major factor in aquaculture waste management. In : *Fish nutrition in practice*. (ed. by S. J. Kaushik & P. Luquet). IV International symposium on fish nutrition and feeding. Biarritz, France, June 24-27, 1991. Ed. INRA, Les Colloques. no. 61.

Cho, C. Y. & Kaushik, S. J. (1990). Nutritional energetics in fish : Energy and protein utilization in rainbow trout (*Salmo gairdneri*). In : Aspects of food production, consumption and energy values. (ed. by G. H. Bourne). pp. 132-172. *World Reviews of Nutrition and Dietetics* 61.

Cho, C. Y. & Slinger, S. J. (1978). Apparent protein digestibility measurement in feedstuffs for rainbow trout. *Proceedings of the World Symposium on Finfish Nutrition and Fishfeed Technology*. **2**, 239-247.

Cho, C. Y., Hynes, K. R. & Yashida, H. K. (1991). Quantification of fish culture wastes by biological (nutritional) and chemical (limnological) methods : the development of high nutrient dense (HND) diets. In : *Nutritional Strategies and Aquaculture Waste* (ed. by C. B. Cowey & C. Y. Cho). pp. 37-50. Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste. University of Guelph, Ontario, Canada.

Choubert, G., Fauconneau, B & Luquet, P. (1982). Influence d'une elevation de la temperature de l'eau sur la digestibilite de la matiere seche, de l'azote et de ll'energie de l'aliment distribue a la truite arc en-ciel (*Salmo gairdneri* Rich). *Reproduction Nutrition and Development* **22**, 941 -949.

Clark, E. R., Harman, J. P. & Forster, J. R. M. (1985). Production of metabolic waste products by intensively farmed rainbow trout, *Salmo gairdneri* Richardson. *Journal of Fish Biology* **27**, 381-393.

Confer, J. L. & Lake, G. J. (1987). Influence of prey type on growth of young yellow perch (*Perca flavescens*). *Canadian Journal of Fisheries and Aquatic Sciences* **44**, 2028-2033.

Connell, D. W. (1993). Water pollution-causes and effects in Australia and New Zealand. University of Queensland Press, St Lucia, Queensland.

Corp, N. & Morgan, A. J. (1991). Accumulation of heavy metals from polluted soils by the earthworm, *Lumbricus rubellus* : Can laboratory exposure of 'control' worms reduce biomonitoring problems? *Environmental Pollution* **74**, 39-52.

Cossins, A. R. & Bowler, K. (1987). Temperature biology of animals. Chapman and Hall, London, U. K. 339p.

Costa-Pierce, B. A. (1996). Environmental impacts of nutrients from aquaculture : Towards the evolution of sustainable aquaculture. In : *Aquaculture and Water Resource Management*. (ed. by D. J. Baird., M. C. M. Beveridge., L. A. Kelly & J. M. Muir), pp. 81-113. Blackell Science Ltd. Oxford, U. K.

Costa-Pierce, B. A. & Roem, C. M. (1990). Waste production and efficiency of feed use in floating net cages in a eutrophic tropical reservoir. In : *Reservoir Fisheries and Aquaculture Development for Resettlement in Indonesia*. (ed. by B. A. Costa-Pierce & O. Soemarwoto), pp. 257-271. ICLARM, Manila, Philippines.

Cowey, C.B & Cho, C.Y. (1991). (editors) Nutritional strategies and aquaculture waste. *Proceeding of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste*. pp 37-50. University of Guelph,Ontario,Canada,

Cowey, C. B. & Sargent, J. R. (1972). Fish nutrition. *Advances in Marine Biology* **10**, 383-492.

Cowey, C. B. and Sargent, J. R. (1979). Nutrition. In : *Fish Physiology*, Vol. 8. (ed. by W. S. Hoar, D. J. Randall and J. R. Brett), pp 1-69. Academic Press, New York.

CRC for Aquaculture. (1996/97). Research on new finfish species for aquaculture. *Austasia Aquaculture* **10(5)**, 70-74.

Creswell, R. L. (1993). Aquaculture desk reference. AVI Book, New York. 206p.

Cripps, S. J. (1993a). The application of suspended particle characterization techniques to aquaculture systems. In : *Techniques for Modern Aquaculture* (ed. by J-K Wang), pp. 26-34. Proceedings of an Aquacultural Engineering Conference, 21-23 June 1993, Spokane, Washington, USA.

Cripps, S. J. (1993b). Minimising outputs : treatment. *Journal of Applied Ichthyology* **10(4)**, 284-294.

Cripps, S. J. (1995). Serial particle size fractionation and characterisation of an aquacultural effluent. *Aquaculture* **133**, 323-339.

Cripps, S. J. & Kelly, L. A. (1996). Reductions in wastes from aquaculture. In : *Aquaculture and Water Resource Management* (ed. by D. J. Baird., M. C. M. Beveridge, L. A. Kelly & J. M. Muir), pp. 166-201. Blackell Science Ltd. Oxford, U. K.

Culver, D. A. & Geddes, M. C. (1993). Limnology of rearing ponds for Australian fish larvae : relationships among water quality, phytoplankton, zooplankton, and the growth of larval fish. *Australian Journal of Marine and Freshwater Research* **44**, 537-551.

D'Abramo, L. R. & Lovell, R. T. (1991). Aquaculture research needs for the year 2000 : fish and crustacean nutrition. *World Aquaculture* **22**, 57-82.

Dabrowski, K. (1984). The feeding of fish larvae : present " state of the art" and perspective's. *Reproduction Nutrition and Development* **24**, 807-823.

Dabrowski, K. & Kaushik, S. J. (1984). Rearing of coregonid (*Coregonus schinzi palea* Cuv.) larvae using dry and live food. II. Oxygen consumption and nitrogen excretion. *Aquaculture* **41**, 333-344.

Dabrowski, K. & Rusiecki, M. (1983). Content of total and free amino acids in zooplanktonic food of fish larvae. *Aquaculture* **30**, 31-42.

Dabrowski, K., Charlon, N., Bergot, P. & Kaushik, S. (1984). Rearing of coregonid (*Coregonus schinzi palea* CUVETVAL.) larvae using dry and live food. I. Preliminary data. *Aquaculture* **41**, 11-20.

Dave, G. (1989). Experiences with waste water-cultured *Daphnia* in the start-feeding of rainbow trout (*Salmo gairdneri*). *Aquaculture* **79**, 337-343.

Davenport, J., Kjorsvik, E. & Haug, T. (1990). Appetite, gut transit, oxygen uptake and nitrogen excretion in captive Atlantic halibut, *Hippoglossus hippoglossus* L., and lemon sole, *Microstomus kitt* (Walbaum). *Aquaculture* **90**, 267-277.

Davis, T. L. O. (1977). Reproductive biology of the freshwater catfish, *Tandanus tandanus* Mitchell, in the Gwydir River, Australia. II, Gonadal Cycle and fecundity. *Australian Journal of Marine and Freshwater Research* **28**, 159-169.

De Pauw, N., P. Laureys & J. Morales. (1981). Mass cultivation of *Daphnia magna* (Straus) on ricebran. *Aquaculture* **25**, 141-152.

De Silva, S. S. & Anderson, T. A. (1995). Fish nutrition in Aquaculture. Chapman & Hall, London. 316p.

De Silva, S.S., Mitchell, B. D. & Nandeesha, M. C. (1993). Reducing feed costs in semi-intensive finfish culture : An update on mixed feeding schedules and an idea for enhancing endogenous food supply in ponds. *Naga, The ICLARM Quarterly* **16(1)**, 24-26.

Devendra, C. (1989). Nomenclature, terminology and definitions appropriate to animal nutrition. p. 1-10. In : *Fish Nutrition Research in Asia* (ed. by S. S. De Silva). Proceedings of the Third Asian Fish Nutrition Network Meeting, Asian Fisheries Society. Special Publication. 4, 166p. Asian Fisheries Society, Manila, Philippines.

Donnelly, J., Torres, J. J. & Hopkins, T. L. (1994). Chemical composition of Antartic zooplankton during fall and winter. *Polar Biology* **14**, 171-183.

Dosdat, A., Metailler, R., Tetu, N., Servais, F., Chartois, R., Huelvan, C. & Desbruyeres, E. (1995). Nitrogenous excretion in juvenile turbot, *Scophthalmus maximus* (L.), under controlled conditions. *Aquaculture Research* **26**, 619-650.

Eckert, R., Randall, D., and Augustine, G. (1988). 'Animal physiology' : Mechanisms and Adaptations.' Freeman : NewYork. 683p.

Edwards, P. (1993). Environmental issues in integrated agriculture-aquaculture and wasteawater-fed fish culture systems. In : *Environment and Aquaculture in developing countries* (ed. by R. S. V. Pullin, H. Rosenthal & J. L. Maclean), pp. 139-170. ICLARM Conference Proceedings, 31.

Eikerbrokk, B., Flogstad, H., Hergheim, A. & Asgard, T. (1991). Prospects and perspectives for the development of green production technologies in Northern Seas Agriculture. In : *Seminar on Agriculture and Aquaculture* (ed. by J. M. Debois & B. Bratten), pp. 85-100. 1st ENS Conference, 26-30 August 1991, Stavanger, Norway.

EIFAC. (1980). Report of the EIFAC, IUNC, and ICES working group on standardization of methodology in fish nutrition research. (ed. by J.D. Castell and K. Tiews). *European Inland Fisheries Advisory Commission*. EIFAC/T36. EIFAC/FAO. 24p.

Eley, R.L., Carrol, J. H. & Woody, D. De. (1972). Effects of caged catfish culture on water quality and community metabolism. *Proceedings of the Oklahama Academy of Science* **51,** 10-15.

Elliott, J. M. (1976). Body composition of brown trout (*Salmo trutta* L.) in relation to temperature and ration size. *Journal of Animal Ecology* **45**, 273-289.

Enell, M. (1987). Environmental impact of cage fish farming with special references to phosphorus and nitrogen. *ICES/1987/FF*, 44.

Enell, M. (1995). Environmental impact of nutrients from NORDIC countries. *Water Science* **31(10)**, 61-71.

Enell, M. & Ackefors, H. (1991). Nutrient discharges from aquaculture operations in Nordic countries into adjacent sea areas. *ICES C. M./199/F*:56, ref. MEQC.

Enell, M. & Lof, J. (1983). Environmental impact of aquaculture - sedimentation and nutrient loadings from fish cage culture farming. *Vatten* **39**, 364-374.

EPA (Environment Protection Authority). (1995). Protecting water quality in Port Phillip Bay. State Environment Protection Policy (Waters of Victoria). Publication No. 434. Environment Protection Authority, Melbourne, Australia.

Erskine, W. D. & Saynor, M. (1995). The influence of waterway management on water quality with particular reference to suspended solids, phosphorus and nitrogen. Benilla Printing. State of Victoria. 82p.

Eskelnen, P. (1984). The phosphorus balance of rainbow trout (Vesihallituksen Monisterarja), National Water Administration, Finland, 241, 33-42. *Translated by Canadian (Translation) Fish and Aquatic Science*.

Evans, D. H. (1993). Chapter 13 : Ammonia and urea metabolis and excretion. p. 379-425. The physiology of fishes. Marine Science Series, CRC Press Inc.,

Farmanfarmaian, A. & Moore, R. (1978). Diseasonal thermal aquaculture. 1. Effect of temperature and dissolved oxygen on survival and growth of *Macrobrachium rosenbergii*. *Proceedings of the Annual Meeting of the World Mariculture Society* **9**, 55-66.

Fermin, A. C. & Boliver, M. E. C. (1994). Feeding live or frozen *Moina macrocopa* (Strauss) to Asian sea bass, *Lates calcarifer* (Bloch) larvae. *Israeli Journal of Aquaculture - Bamidgeh* **46(3)**, 132-139.

Fernando, C. H. (1994). Zooplankton, fish and fisheries in tropical freshwaters. *Hydrobiologia* **272**, 105-123.

Fernnandez-Reiriz, M. J., Labarta, U. & Ferreiro, M. J. (1993). Effects of commercial enrichment of diets on the nutritional value of the rotifer (*Brachionus plicatilis*). *Aquaculture* **112**, 195-206.

Fivlstad, S., Thomassen, J. M., Smith, M. J., Kjartansson, H. & Sando, A-B. (1990). Metabolic production rates from Atlantic salmon (*Salmo salar* L.) and Arctic char (*Salvelinus alpinus* L.) reared in single pass land-based brackish water and sea water systems. *Aquacultural Engineering* **9**, 1-21.

Fluchter, J. (1980). Review of the present knowledge of rearing whitefish (Coregonidae) larvae. *Aquaculture* **19**, 191-208.

FNBW (Finland National Board of Waters). (1981). Industrial water statistics in 1977-1978. National Board of water, Helsinki, Finland (in Finnish with English Summary).

Forftner, U. & Wittmann, G. T. W. (1979). Metal pollution in the aquatic environment. Springer- Verlag, Berlin.

Forster, R. P. & Goldstein, L. (1969). Formation of excretory products, In : *Fish Physiology* (ed. by W. S. Hoar and D. J. Randall), pp.313-350. vol. I. Academic Press, New York, USA.

Folke, C. & Kautsky, N. (1989). The role of ecosystems for a sustainable development of aquaculture. *Ambio* **18**, 234-243.

Foy, R. H. & Rosell, R. (1991a). Loadings of nitrogen and phosphorus from a Northern Ireland fish farm. *Aquaculture* **96**, 17-30.

Foy, R. H. & Rosell, R. (1991b). Fractionation of phosphorus and nitrogen loadings from a Northern Ireland fish farm. *Aquaculture* **96**, 31-42.

Fromm, P. O. & Gillette, J. R. (1968). Effect of ambient ammonia on blood ammonia and nitrogen excretion of rainbow trout (*Salmo gairdneri*). *Comparative Biochemistry and Physiology* **26**, 887-896.

Gallagher, M. E. & Matthews, A. M. (1987). Oxygen consumption and ammonia excretion of the American eel *Anguilla rostrata* fed diets with varying protein energy ratios and protein levels. *Journal of the World Aquaculture Society* **18(2)**, 107-112.

Gallagher, M. I., Kane, E. & Courtney, J. (1984). Differences in oxygen consumption and ammonia production among american elvers (*Anguilla rostrata*). *Aquaculture* **40**, 183-187.

Geiger, J. G. (1983). Zooplankton production and manipulation in striped bass rearing ponds. *Aquaculture* **35**, 331-351.

Gavine, F. M., Phillips, M. J. & Murray, A. (1995). Influence of improved feed quality and food conversion ratios on phosphorus loadings from cage culture of rainbow trout, *Oncorhynchus mykiss* (Walbaum), in freshwater. *Aquaculture Research* **26**, 483-495.

Gerking, S. D. (1955). Influence of rate of feeding on body composition and protein metabolites of bluegill sunfish. *Physiology and Zoology* **28**, 267-283.

Gerking, S. D. (1971). Influence of rate of feeding and body weight on protein metabolism of bluegill sunfish. *Physiology and Zoology* **44**, 9-19.

Gershanovitch, A. D. & Pototskij, I. V. (1992). The peculiarities of nitrogen excretion in sturgeons (*Acipenser ruthenus*) (Pisces, Acipenseridae) - I. The influence of ration size. *Comparative Biochemistry and Physiology* **103A**, 609-612.

Gill, H. S. & Weatherley, A. H. (1984). Protein, lipid and caloric contents of bluntnose minnow, *Pimephales notatus* Rafinesque,during growth at different temperatures. *Journal of Fish Biology* **25**, 491-500.

Gomes, E. F., Corraze, G. & Kaushik, S. (1993). Effects of dietary incorporation of a co-extruded plant protein (rapeseed and peas) on growth, nutrient utilization and muscle fatty acid composition of rainbow trout (*Oncorhynchus mykiss*). *Aquaculture* **113**, 339-353.

Gooley, G. & Rowland, S. (1993). Murray-Darling Finfish. Current Developments and Commercial Potential. *Austasia Aquaculture* 7, 35-38.

Grabner, M., Wiser, W. & Lackner, R. (1981/82). The suitability of frozen and freeze dried zooplankton as food for fish larvae : A biochemical test programme. *Aquaculture* **26**, 85-94.

Guo, R., Mather, P. & Capra, M. F. (1993). Effect of salinity on the development of silver perch (*Bidyanus bidyanus)* eggs and larvae. *Comparative Biochemistry and Physiology* **A103**, 531-535.

Guo, R., Mather, P. B., & Capra, M. F. (1995). Salinity tolerance and osmoregulation in the silver perch, *Bidyanus bidyanus* (Mitchell) (Teraponidae), an endemic Australian freshwater teleost. *Marine and Freshwater Research* **46**, 947-952.

Haiqing, S. & Xiqin, H. (1994). Effects of dietary animal and plant protein ratios and energy levels on growth and body composition of bream (*Megalopbrama skolkovii* Dybowski). *Aquaculture* **127**, 189-196.

Hajen, W.E., Beames, R. M., Higgs, D. M. & Dosanjh, B. S. (1993). Digestibility of various feed stuffs by post-juveniles chinook salmon (*Oncorhynchus tshawytscha*) in sea water. I. Validation of technique. *Aquaculture* **112**, 321-332.

Hakanson, L., Ervik, A., Makinen, T. & Moller, B. (1988). Basic concepts concerning assessments of environmental effects of marine fish farms. Nordic Council of Ministers, Nord 1988, 90.

Hall, S. J. (1987). Maximum daily ration and the pattern of food consumption in haddock, *Melanogrammus aeglefinnus* (L.) and dab, *Limanda limanda* (L.). *Journal of Fish Biology* **31**, 479-491.

Hall, P. O. J., Holby, O., Kollberg, S. & Samuelsson, M-O. (1992). Chemical fluxes and mass balances in marine fish cage farm. IV. Nitrogen. *Marine Ecology Progress Series* 89, 81-91.

Handy, R. D. & Poxton, M. G. (1993). Nitrogen pollution in mariculture : toxicity and excretion of nitrogenous compounds by marine fish. *Reviews in Fish Biology and Fisheries* **3**, 205-241.

Hardy, R. W. (1989). Diet preparation. In : *Fish Nutrition* (ed. by J. E. Halver). pp. 475-548. 2nd edition. San Diego, Academic Press.

Hart, P. & O'Sullivan, D. (1993). Recirculation systems : Design, Construction and Management. *National Key Centre for Teaching and Research in Aquaculture*. University of Tasmania at Launceston, Launceston, Tasmania. Workshop Series. 130p.

Hasan, M. A., Islam, M. N., & Alim, M. A. (1993). Evaluation of silkworm pupae meal as dietary protein source for catfish (*Heteropneustes fossilis* Bloch). In : *Fish Nutrition Practice*, (ed : S. J. Kaushik and P.Luquet), pp. 785-791, INRA, Paris.

Hays, T. (1980). Impact of net pen culture on water quality and fish populations in Bull Shoals reservoir. Completion report to the Arkansas Game and Fish Commission, Fayetteville, Arkansass. 10p.

Hennessy, M., Wilson, L. & Struthers, W. (1991). Management strategies for salmon farm effluents. A report prepared for the Scottish salmon growers association, Perth, Scotland.

Hennessy, M.M., Wilson, L., Struthers, W. & Kelly, L. A. (1996). Waste loadings from two freshwater Atlantic salmon juvenile farms in Scotland. *Water, Air and Soil Pollution.* **86**, 235-249.

Hepher, B. (1988). Nutrition of pond fishes. Cambridge University Press, Cambridge.

Higgs, D. A., Markert, J. R., Plonikoff, M. D., McBride, J. R. & Dosanjh, B. S. (1985). Development of nutritional and environmental strategies for maximising the growth and survival of juvenile pink salmon (*Oncorhynchus grobuscha*). *Aquaculture* **47**, 113-130.

Hilge, V. (1979). Preliminary results with krill meal and fish meal in diets for channel catfish (*Ictalurus punctatus* Raff.). In : *Proceedings of the World Symposium on Finfish Nutrition and Fishfeed Technology.* (ed. by J.E. Halver and K. Tiews). p. 167-171. Hamburg 20-23 June 1978, volume II, Merlin.

Hillestad, M. & Johnsen, F. (1994). High energy/low protein diets for Atlantic salmon : effects on growth, nutrient retention and slaughter quality. *Aquaculture* **124**, 109-116.

Hilton, J. W., Cho, C. Y. & Slinger, S. J. (1981). Effect of extrusion processing and steam pelleting diets on pellet durability, pellet water absorption, and the physiological response of rainbow trout (*Salmo gairdneri* R.). *Aquaculture* **25**, 185-194.

Hoelzi, A. & Vens-Cappell, B. (1980). Profitability of food-fish production in net cages. *Fisch. Teichwirt* **32**, 2-5.

Holby, O. & Hall, P. O. J. (1991). Chemical fluxes and mass balances in marine fish cage farm. II. Phosphorus. *Marine Ecology Progress Series* **70**, 263-272.

Holm, J. C. (1986). Yolk sac absorption and early food selection in Atlantic salmon feeding on live prey. *Aquaculture* **54**, 173-188.

Holm, J. C. & Moller, D. (1984). Growth and prey selection by Atlantic salmon yearlings reared on live freshwater zooplankton. *Aquaculture* **43**, 401-412.

Holm, J.C. & Walther, B. T. (1988). Free aminoacids in live freshwater zooplankton and dry feed : Possible importance for first feeding in Atlantic salmon fry (*Salmo salar*). *Aquaculture* **71**, 223-234.

Horvath, L., Tamas, G. & Szabo, E. (1979). Nutrition-biological questions of bred cyprinids in the first month of their life. In : *Finfish Nutrition and Fish feed Technology* (ed. by *J. E. Halvers and K. Tiews)*, pp 467-478. Proceedings of the World Symposium on Finfish Nutrition and Fish feed Technology, Hamburg 20-23 June 1978, Vol. I.

Howes, J. (1995). The Pocket Environment Dictionary - An introduction to common environment terms. The Five Miles Press Pty Ltd., Victoria, Australia. 128p.

HPRB (Highland River Purification Board). (1987). Policy for controlling pollution from fish farms. Dingwall : HRPB, Tech. Rep. No. TRA/87. 10p.

Hume, D. & Barlow, C. (1993). Bibliography : Four Australian Fish that have potential for aquaculture : Freshwater catfish (*Tandanus tandanus*), Golden perch (*Macquaria ambigua*), Murray cod (*Maccullochella peeli*), and Silver perch (*Bidyanus bidyanus*). Department of Primary Industries, Queensland Government, Brisbane, Australia.

Hunter, B. J., Roberts, D. C. K. & Allan, G. L. (1994). The effects of varying dietary protein/energy ratios in the fat content and fatty acid composition of silver perch (*Bidyanus bidyanus*). *Proceedings of the Nutritional Society of Australia* **18**, 54.

Huisman, E. A., Breteler, J. G. P, Vismans, M. M. & Kanis, E. (1979). Retention of energy, protein, fat and ash in growing carp (*Cyprinus carpio* L.) under different feeding and temperature regimes. In : *Finfish Nutrition and Fishfeed Technology* (ed. by J. E. Halvers & K. Tiews). pp. 175-188. Proceedings of the World Symposium on Finfish Nutrition and Fish Feed Technology, Hamburg 20-23 June 1978, Volume 1.

Ibrekk, H. O. (1989). Model for determining pollution loading to Norwegian coastal waters (i.e., Lenzka-zones) NIV Aonotat 0-88145, Oslo.

Ingram, B., Gooley, G. & McKinnon, L. (1996). Potential for inland mariculture in Victorian saline groundwater evaporation basins. *Austasia Aquaculture* **10(2)**, 61-63.

IOA (Institute of Aquaculture) (1990). Institute of freshwater Biology and Institute of Terrestrial Ecology. Fish farming and the Scottish Freshwater Environment. Nature Conservancy Council, Edinburgh.

Iwata, K. (1970). Relationship between food and growth in young crucian carps, *Carassius auratus cuvieri*, as determined by the nitrogen balance. *Japan Journal of Limnology* **31**, 129-151.

Jackson, P. (1994). Australian threatened fishes - 1994 supplement. *Australian Society for Fish Biology Newsletter* **24(2)**, 18-26.

Jarboe, H. H. (1995). Diel dissolved oxygen consumption and total ammonia nitrogen production by fingerlings channel catfish following feeding at different times. *The Progressive Fish Culturist* **57**, 156-160.

Jarzynski, S. (1991). Background fisheries statistics. Australian Fisheries Service, Canberra, Australia.

Jauncey, K. (1982). Carp (*Cyprinus carpio*) Nutrition - A review. In : *Recent Advances in Aquaculture*. (ed. by J. F. Muir and R. J. Roberts). pp. 217-263. Croom Helm, London.

Jayaram, M. G. & Shetty, H. P. C. (1981). Formulation, processing and water stability of two new pelleted fish feeds. *Aquaculture* **23**, 355-359.

Jeffries, H. P. (1970). Seasonal composition of temperate plankton communities fatty acids. *Ecology* **15**, 419-426.

Jeffries, H. P. (1979). Biochemical correlates of seasonal changes in marine communities. *American Naturalist* **113**, 643-658.

Jensen, J. B. (1991). Environmental regulations of fresh water fish farms. In : *Nutritional Strategies and Aquaculture Waste* (ed. by C. B. Cowey & C. Y. Cho), pp. 51-63. Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste. University of Guelph, Guelph, Ontario, Canada, 1990.

Jobling, M. (1981). Some effects of temperature, feeding and body weight on nitrogenous excretion in young plaice, *Pleuronectes platessa* L. *Journal of Fish Biology* **18**, 87-96.

Jobling, M. (1986). Gastrointestinal overload- a problem with formulated feeds. *Aquaculture* **51**, 257-263.

Johnsen, F. & Wandsvik, A. (1991). The impact of high energy diets on pollution control in the fish farming industry. In : *Nutritional Strategies and Aquaculture Waste* (eds. C. B. Cowey and C. Y. Cho), pp. 51-63. Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste. University of Guelph, Guelph, Ontario, Canada, 1990.

Johnsen, F., Hillestad, M. & Austreng, E. (1993). High energy diets for Atlantic salmon. Effect on pollution. In : *Fish Nutrition in Practice* (eds. S. J. Kaushik & P. Luquet), pp. 391-401. INRA, Paris.

Jones, C. (1990). Crayfish biology - getting down to basics. *Australian Fisheries* 49(11) : 3-7.

Jones, C. M. (1991). The biology and aquaculture potential of the tropical freshwater crayfish *Cherax quadricarinatus*. Queensland Department of Primary Industries, Queensland Government, Brisbane.

Jones, C. M. (1995a). Production of juvenile redclaw crayfish, *Cherax quadricarinatus* (Von Martens) (Decapoda,Parastacidae) - 1. Development of hatchery and nursery procedures. *Aquaculture* **138(1-4)**, 221-238.

Jones, C. M (1995b). Production of juvenile redclaw crayfish, *Cherax quadricarinatus* (Von Martens) (decapoda, Parastacidae) - 2. Juvenile nutrition and habitat, *Aquaculture* **138(1-4)**, 239-245.

Kailola, P. J., Williams, M. J., Stewart, P. C., Reichelt, R. E., McNee, A & Grieve, C. (1993). *Australian Fisheries Resources.* Bureau of Resource Science, Department of Primary Industries and Energy, and the Fisheries Research and Development Corporation. Canberra, Australia. 422p.

Kamler, E., Szlaminska, M., Raciborski, K., Barska,B., Jakubas, M., Kuozynski, M & Przybyl. A. (1992). Bioenergetic evaluation of four formulated diets for carp (*Cyprinus carpio*) larvae as compared with zooplankton fed and starved larvae. *Journal of Animal Physiology* **67**, 1-15.

Kanazawa, A., Teshima, S. & Ono, K. (1979). Relationship between essential fatty acid requirements of aquatic animals and the capacity for bioconversion of linolenic acid to highly unsaturated fatty acids. *Comparative Biochemistry and Physiology* **63B**, 295-298.

Kaushik, S. J. (1980). Influence of nutritional status on the daily patterns of nitrogen excretion in the carp (*Cyprinus carpio* L.) and the rainbow trout. *Reproduction Nutrition and Development* **20**, 1751-1765.

Kaushik, S. J. (1981). Influence of a rise in temperature on the nitrogen excretion of rainbow trout (*Salmo gairdneri* R.). *Proceedings of the World Symposium on Aquaculture in Heated Effluents and Recirculation Systems.* Stavanger 28-30 May 1980, vol. I, Berlin 1981. p. 77-85.

Kaushik, S. J. (1992). Recent trends in the development of high-energy diets for salmonids. 2e Conference Internazionale " Produzione di alimenti per animali", Piacenza, Italie, 25-26 fev. 1992.

Kaushik, S. J. & Cowey, C. B. (1991). Dietary factors affecting nitrogen excretion by fish. In : *Nutritional strategies and aquaculture wastes.* (ed. by C. B. Cowey & C. Y. Cho). pp. 3-19. Proceedings of the first International symposium on nutritional strategies in management of aquaculture waste (NSMAW). Fish Nutrition Research Laboratory, Department of Nutritional Sciences, University of Guelph, Canada and Fisheries Branch, Ontario Ministry of Natural Resources, and International Union of Nutritional Sciences (IUNS).

Kaushik, S. J. & Oliva-Teles, A. de. (1985). Effect of digestible energy on nitrogen and energy balance in rainbow trout. *Aquaculture* **50**, 89-101.

Kaushik, S. J., Fauconneau, B., & Blanc, J. M. (1984). A study of nitrogen excretion and oxygen consumption in five half-sib families of rainbow trout. *Reproduction Nutrition and Development* **24**, 431-438.

Kawasaki, L. Y., Tarifeno-Silva, Yu, D. P. & Gordan, M. S. (1992). Aquacultural approaches to recycling of dissolved nutrients in secondarily treated domestic wastewaters - 1 : Nutrient uptake and release by artificial food chain. *Water Research* **16**, 37-49.

Kelly, L. A. (1992). Dissolved reactive phosphorus release from sediments beneath a freshwater cage aquaculture development in West Scotland. *Hydrobiologia* **235/236**, 569-572.

Kelly, L. A. (1993). Release rates and biological availability of phosphorus released from sediments receiving aquaculture wastes. *Hydrobiologia* **253**, 367-372.

Kelly, L. A., Bergheim, A. & Hennessy, M. M. (1994). Predicting output of ammonium from fish farms. *Water Research* **28(6)**, 1403-1405.

Kendra, W. (1991). Quality of salmonid effluents during a summer low-flow season. *Tansaction of the American Fisheries Society* 120, 43-51.

Ketola, H. G. (1975). Requirements of Atlantic salmon for dietary phosphorus. *Transaction of the American Fisheries Society* **104**, 548-551.

Ketola, H. G. (1982). Effects of phosphorus in trout diets on water pollution. *Salmonid* 6(2), 12-15.

Ketola, H. G. (1985). Effect of phosphorus in trout and salmon feeds on water pollution. In : *Nutrition and feeding in fish.* (ed. by A. M. Mackie & J. G. Bell). pp.465-473. Aberdeen, U.K.

Ketola, H. G. (1990). Dietary control of phosphorus in hatchery effluents. Proceedings of the 41st Annual Northwest Fish Culture Conference, Idaho Department of Fish and Game, Boise, Idaho.

Ketola, H. G. (1991). Managing fish hatchery discharges of phosphorus through nutrition. In : *Proceedings from the Aquaculture Symposium.* Engineering aspects of Intensive Aquaculture. pp.187-197. Northeast Regional Agricultural Engineering Service Publication 49. Cornell University, Ithaca, New York.

Ketola, H. G. & Harland, B. F. (1993). Influence of phosphorus in rainbow trout diets on phosphorus discharges in effluent water. *Transactions of the American Fisheries Society* 122, 1120-1126.

Ketola, H. G. and Richmond, M. E. (1994). Requirement of rainbow trout for dietary phosphorus and its relationship to the amount discharged in hatchery effluents. *Transactions of the American Fisheries Society* **123,** 587-594.

Ketola. H. G., Westers H., Houghton, W. and Pecor, C. (1991). Effect of diet on growth and survival of coho salmon and on phosphorus discharges from a fish hatchery. *American Fisheries Society Symposium* **10**, 402-409.

Khan, J. A. and A. D. Qayyum. (1971). Water, nitrogen and phosphorus in freshwater plankton. *Hydrobiologia* **37**, 531-536.

Kibria, G., Nugegoda, D., Fairclough, R. & Lam, P. (1996a). Australian native species in Aquaculture. *Victorian Naturalist* **113(5)**, 264-267.

Kibria, G., Nugegoda, D., Lam, P. & Fairclough, R. (1996b). Aspects of phosphorus pollution from aquaculture. *Naga, The ICLARM Quarterly*, **9(3)**, 20-24.

Kibria, G, Nugegoda, D., Lam, P. & Fairclough, R. (1997a). Preliminary rearing trials of an Australian native fish silver perch (*Bidyanus bidyanus*) (Mitchell) with reference to growth and the production of solid wastes in aquaculture. *Asian Fisheries Science* **9**, 301-307.

Kibria, G., Nugegoda, D., Fairclough, R. & Lam, P. (1997b). Pollution from aquaculture. *Chemistry in Australia* **64(1)**, 19-20.

Kibria, G., Nugegoda, D., Fairclough, R. & Lam, P. (1997c. accepted). Effect of temperature on phosphorus losses and phosphorus retention in silver perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae). *Aquaculture Research.*

Kibria, G., Nugegoda, D., Fairclough, R. & Lam, P. (1997d accepted). The nutrient content and the release of nutrients from fish food and faeces. *Hydrobiologia.*7, 1-7 (in press).

Kikuchi, K., Sato, T., Iwata, N., Sakaguchi, I. and Deguchi, Y. (1995). Effects of temperatures on nitrogenous excretion of Japanese flounder. *Fisheries Science* **61(4)**, 604-607.

Kilambi, R., Hoffman, C., Brown, A., Adams, J. & Wickizer, W. (1976). Effects of cage culture fish production upon the biotic and abiotic environment of crystal lake, Arkansas. Department of Zoology, University of Arkansas, Fayetteville, Arkansas.

Kim, Y. K. (1974). Determination of true digestibility of dietary proteins in carp with chromic oxide containing diet. *Bulletin of the Japan Society of Science and Fisheries* **40**, 651-653.

Kokova, V. Ye., Barashkov, V. A. & Trubachev, I. N. (1990). Free amino acids of certain aquatic invertebrates. *Hydrobiologia* **26(1)**, 77-79.

Koops, H., Tiewes, K., Gropp, J. & Beck, H. (1979). Krill in trout diets. In : *Proceedings of the World Symposium on Finfish Nutrition and Fish feed Technology*. (ed. by J. E. Halvers & K. Tiewes). pp. 281-292.Hamburg 20-23 June 1978.

Kristiansen, G. & Hessen, D. O. (1992). Nitrogen and phosphorus excretion from the noble crayfish, *Astacus astacus* L., in relation to food type and temperature. *Aquaculture* 102, 45-264.

Krom, M. D., Erez, J., Porter, C. B. & Ellner, S. (1989). Phytoplankton nutrient uptake dynamics in earthen marine fishponds under winter and summer conditions. *Aquaculture* **76**, 237-253.

LaBrasseur, R. J. (1969). Growth of juvenile chum salmon (*Oncorhynchus keta*) under different feeding regimes. *Journal of the Fisheries Research Board of Canada* **26**, 1631-1645.

Lai, P. C. (1994). An ecological study of waste stabilization ponds in Werribee, Australia, with special reference to nutrient and plankton dynamics. M.Sc thesis. Department of Environmental Management, Victoria University of Technology, Australia (unpublished).

Laird, L. M. & Needham, T. (1988). Salmon and trout farming. Ellis Horwood Limited. England. 271p.

Lake, J. S. (1967a). Principles fishes of the Murray-Darling River system. In : *Australian Inland Waters and their Faunas* (ed. by A.H.Weatherly). pp. 192-213. Australian National University Press, Canberra, pp. 192-213.

Lake, J. S. (1967b). Rearing experiments with five species of Australian freshwater fishes. I. Inducement to spawning. *Australian Journal of Marine and Freshwater Research* **18**, 137-153.

Lake, J. S. (1967c). Rearing experiments with five species of Australian freshwater fishes. II. Morphogenesis and ontogeny. *Australian Journal of Marine and Freshwater Research* **18**, 155-173.

Lall, S. P. (1979). Minerals in finfish nutrition. In : *Finfish Nutrition and Fishfeed Technology* (ed. by J. E. Halver & K. Tiewes), pp. 85-93. Proceedings of the World Symposium on Finfish Nutrition and Fish feed Technology, Hamburg 20-23 June 1978, Vol. I

Lall, S. P. (1989). The minerals. In : *Fish Nutrition*. (ed. by J. E. Halver). pp. 86-97. 2nd edition. Academic Press Inc., San Diego, California.

Lall, S. P. (1991). Digestibility, metabolism and excretion of dietary phosphorus in fish. In : *Nutritional Strategies and Aquaculture Waste* (ed. by C. B. Cowey & C. Y. Cho), pp.21-36. Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste. University of Guelph, Guelph, Ontario, Canada, 1990.

Lam, T. J. & Sharma, R. (1985). Effects of salinity and thyroxine on larval survival, growth and development in the carp, *Cyprinus carpio*. *Aquaculture* **44**, 201-212.

Lanari, D., D'Agaro, E. & Ballestrazzi, R. (1995a). Effect of dietary DP/DE ratio on apparent digestibility, growth and nitrogen and phosphorus retention in rainbow trout, *Oncorhynchus mykiss* (Walbaum). *Aquaculture Nutrition* **1**, 105-110.

Lanari, D., D'Agaro, E. & Ballestrazzi, R. (1995b). Dietary N and P levels, effluent water characteristics and performance in rainbow trout. *Water Science and Technology* **31(10)**, 157-165.

Lancelot, C., Billen, G., Sourina, A., Weisse, T., Colijn, F., Veldhuis, M. J. M., Davies, A. & Wassman, P. (1987). "*Phaeocystis* blooms and nutrient enrichment in the continental coastal zones of the North Sea'. *Ambio* **16**, 38-46.

Lauff, M. & Hofer, R. (1984). Proteolytic enzymes in fish development and the importance of dietary enzymes. *Aquaculture* **37**, 335-346.

Law, A. T. (1984). Nutritional study of jelawat, *Leptobarbus hoevenii* (Bleeker), fed on pelleted feed. *Aquaculture* 41, 227-233.

Leid, E. & B. Bratton. (1984). The effect of feeding and starving and different ratios of protein energy to total energy in the feed on the excretion of ammonia in Atlantic cod (*Godus morhua*). *Comparative Biochemistry and Physiology* **78B**, 49-52.

Lemm, C. A. (1983). Growth and survival of Atlantic salmon fed semi-moist or dry starter diets. *Progressive Fish Culturist* **9**, 291-296.

Liao, P. B. (1970). Pollution potential of salmonid fish hatcheries. *Water and Sewage Works*. August, 291-297.

Llewellyn, L. C. (1983). The distribution of fish in New South Wales. *Australian Society of Limnology*. Special Publication No. 7, 23p.

Lokman, H. S. (1994). Lipid and fatty acid composition of indigenous zooplankton from Terenggau waters of the south China. *Journal of Aquaculture Tropical* **9**, 292-296.

Lovell, R. T. (1978). Dietary phosphorus requirements of channel catfish (*Ictalurus punctatus*) *Transaction of the American Fisheries Society* **107**, 617-621.

Lovell, T. (1989). Nutrition and feeding of fish. Van Rnostrand Reinhold, New York.

Lukowicz, M. V. (1979). Experiences with krill (*Euphausis superba* Dana) in the diet for young carp (*Cyprimus carpio*). In : *Proceedings of the World Symposium on Finfish Nutrition and Fish feed Technology* (ed by J. E. Halvers & K. Tiewes). pp. 293-302. Hamburg 20-23 June 1978.

Luquet, P. & Fauconneau, B. (1979). Influence de la temperature sur quelques aspects de l'alimentation des poissons. *Bull. Cent. Etud. Sci. Biarritz* **12**, 475-492.

Maetz, J. (1972). Branchial sodium exchange and ammonia excretion in the goldfish *Carassius auratus*. Effects of ammonia-loading and temperature changes. *Journal of the Experimental Biology* **56**, 601-620.

Makinen, T. (1991). Nutrient load from marine aquaculture. In : *Marine Aquaculture and the Environment* (ed. by T. Makinen), pp. 1-8. Nordic Council of Ministers, Copenhagen.

Makinen, T, Lindgren, S, & Eskelinen, P. (1988). Sieving as an effluent treatment method for aquaculture. *Aquaculture Engineering* 7**(6)**, 367-377.

Manzi, J. J. (1989). Aquaculture research priorities for the 1990's. *World Aquaculture* **20(2)**, 29-32.

Matty, A. J. (1990). Feeds in the fight against pollution. *Fish Farming International.* January, 16-17.

Maguire, G. B. & Allan, G. L. (1992). Effects of temperature on growth, food consumption and food conversion of *Penaeus monodon*, *Penaeus plebejus* and *Metapenaeus macleay*. In : *Proceedings Nutritional Society Workshop*, Salamander Bay, 15-17 April 1991. (ed. by G. L. Allan and W. Dall). pp. 97-99. NSW Fisheries, Brackish Water Culture Research Station, Salamander Bay, Australia.

McKinnon, L., Gooley, G. & Gasior, R. (1996). Integrated silver perch aquaculture trials in the Golburn-Murray irrigation district of Victoria. *Austasia Aquaculture* **10(1)**, 45-47.

Mearns, K. J. (1986). Sensitivity of brown trout (*Salmo trutta* L.) fry to amino acids at the start of exogenous feeding. *Aquaculture* **55**, 191-200.

Merican, Z. & Phillips, M. (1985). Solid waste production from rainbow trout, *Salmo gairdneri* Richardson, cage culture. *Aquaculture and Fisheries Management* **1**, 55-69.

Merrick, J. R. (1980). Family Teraponidae. Freshwater Grunters or Perches. In : *Freshwater Fishes of South-Eastern Australia.* (ed. by R. M. McDowell). (Reed : Sydney), pp. 150-152.

Merrick, J. R. & Schimida, G. E. (1984). Australian freshwater fishes - biology and management. Griffin Press Ltd. South Australia.

Millamena, O. M., Penaflorida, V. D., & Subosa, P. F. (1990). The macronutrient composition of natural food organisms mass cultured as larval feed for fish and prawns. *Israeli Journal of Aquaculture - Bamidgeh* **42(3)**, 77-83.

Mills, B. J. & Geddes, C. (1980). Salinity tolerance and osmo-regulation of the Australian crayfish *Cherax destructor* Clark (Decapoda:Parastacidae). *Australian Journal of Marine and Freshwater Research* **31**, 667-676.

Mims, S. D., Webster, C. D., Tidwell, J. H. & Yancey, D. H. (1991). Fatty acid composition of *Daphnia pulex* cultured by two different methods. *Journal of the World Aquaculture Society* **22 (2)**, 153-156.

Ming, F. W. (1985). Ammonia excretion rate as an index for comparing efficiency of dietary protein utilization among rainbow trout (*Salmo gairdneri*) of different strains. *Aquaculture* **46**, 27-35.

Mitchell, T. L. (1838). Three expeditions into the interior of Australia, with description of the recently explored region of Australia Felix and of the present colony of New South Wales. T & W Boone, London.

Morrissy, N. M. (1978). The past and present distribution of marron *Cherax tenuimanus* (Smith), in Western Australia. *Fisheries Research Bulletin of Western Australia* **22**, 1-32.

Muir, J. F. (1982). Economic aspects of waste treatment in fish culture. In : *Report of the EIFAC Workshop on Fish-farm effluents* (ed. by J. S. Albaster), pp 123-135. *EIFAC Technical Paper* **41**.

Munilla-Moran, R., Stark, J. R. & Barbour, A. (1990). The role of exogenous enzymes in digestion in cultured larvae (*Scophthalmus maximus* L.). *Aquaculture* **88**, 337-350.

Munro, K. A., Samis, S. C. & Nassichuk, M. D. (1985). The effects of hatchery effluents on water chemistry, periphyton and benthic invertebrates of selected British Columbia Stream. *Canadian Fisheries and Aquatic Sciences.* MS Report 1830. 147p.

Murai, T. & Andrews, J. W. (1973). Growth and food conversion of rainbow trout reared in brackish and freshwater. *Fishery Bulletin* **70**, 1203-1295.

Nakashima, B. S. & Leggett, W. C. (1980). Natural sources and requirements of channel catfish (*Ictalurus punctatus*). *Transactions of the American Fisheries Society* 107, 617-621.

National Academy of Sciences. (1977). Nutrient requirements of warm water fishes. Nutrient requirement of domestic animals. National Academy of Sciences. Washington, D. C. 78p.

NCC. (1990). Fish farming and the Scottish Freshwater Environment. Nature Conservancy Council, Edinburgh.

New, M. (1987). Feed and feeding of fish and shrimp - A manual on the preparation and presentation of compound feeds for shrimp and fish in aquaculture. Aquaculture Development and Coordination Programme, United Nations Development Programme and Food and Agriculture Organization of the United Nations. FAO and UNEP. Rome, Italy. *ADCP/REP/87/26* : 275p.

Nichols, D. S., Nichols, P. D. & Volkman, J. K. (1993). Oil, proximate and fatty acid composition of freshwater copepods and daphnids obtained from Werribee treatment complex using the frish polyculture harvester. CSIRO Division of Oceanography, Report 93--Zootech-1.

Nose, T. (1967). On the metabolic faecal nitrogen in young rainbow trout. *Bulletin of the Freshwater Research Laboratory* **17**, 97-105.

Nose, T. (1971). Determination of nutritive value of food protein in fish. III. Nutritive value of casein, white fish meal and soybean meal in rainbow trout fingerlings. *Bulletin Freshwater Fisheries Research Laboratory* **21**, 85-98.

Nose, T. & Arai, S. (1979). Recent advances in studies on mineral nutrition of fish in Japan. In : *Advances in Aquaculture*. (ed. by T. V. R. Pillay & W. A. Dill). pp. 584-590. Fishing News Books Ltd. Farnham, Surrey, England.

O'Brien, K. 1994. Semi-microdigestion for nitrogen, phosphorus and potassium. In : *Proceedings of the Analytical Techniques Workshop*. Brisbane, 13 April 1994. (ed. by G. Allan and J. Frances). pp.18-20.

OCE. (1988). State of Environment Report : Victoria's Inland Waters. Office of the Commissioner of the Environment, Melbourne. 655p.

Oertzen, Jorg-A, V. (1985). Resistance and capacity adaptation of juvenile silver carp, *Hypothalmichrhys molitrix* (Val.), to temperature and salinity. *Aquaculture* **44**, 321-332.

Ogburn, D. M., Rowland, S. J., Mifusd, C. & Creighton, G. (1995). Site selection, design and operation of pond-base aquaculture systems. In : *Silver Perch Culture : Proceedings of Silver Perch Workshops, Grafton and Narrendera, April 1994*. (ed. by S. J. Rowland & C. Bryant), pp. 19-28. Austasia Aquaculture, Hobart for NSW Fisheries, Sydney.

Ogino, C. (1963). Studies on the chemical composition of some natural foods of aquatic animals. *Bulletin of the Japan Society of Science and Fisheries* **29**,459-462.

Ogino, C., Kakino, J. & Chen, M-S. (1973a). Protein nutrition in fish-II. Determination of metabolic faecal nitrogen and endogenous nitrogen excretion of carp. *Bulletin of the Japan Society of Science and Fisheries.* **39**, 519-523.

Ogino, C., Kakino, J. & Chen, M. S. (1973b). Protein nutrition in fish. V. Relation between biological value of dietary proteins and their utilization in carp. *Bulletin of the Japan Society of Science and Fisheries* **39**, 955-959.

Ogino, C. & Takeda, H. (1978). Requirements of rainbow trout for dietary calcium and phosphorus. *Bulletin of the Japan Society of Science and Fisheries* **44**, 1019-1022.

Ogini, C., Takeuchi, L., Takeda, H. & Watanabe, T. (1979). Availability of dietary phosphorus in carp and rainbow trout. *Bulletin of the Japan Society of Science and Fisheries.* **45**, 1527-1532.

Oliva-Teles, A. & A. M. Rodrigues, A. M. (1993). The effect of high temperatures and diet protein level on the metabolic utilization of diets by rainbow trout. In : *Fish Nutrition in Practice*. (ed. by S. J. Kaushik & P. Luquet). pp. 301-305. IV International Symposium on Fish Nutrition and Feeding, Biarritz, France, June 24-27, 1991. INRA 61

Olson, K. R. & Fromm, P. O. (1971). Excretion of urea by two teleosts exposed to different concentrations of ambient ammonia. *Comparative Biochemistry and Physiology* **40A**, 997-1007.

O'Sullivan, D. (1994). Research underway on replacement of fishmeal in aquaculture diets. *Austasia Aquaculture* 8**(2),** 44-45.

O'Sullivan, D. (1995). Status of Australian native finfish aquaculture production in 1991-92. In : *Silver Perch Culture : Proceedings of Silver Perch Workshops, Grafton and Narrandera, April 1994.* (ed. by S.J.Rowland & C. Bryant), pp. 5-8. Austasia Aquaculture, Hobart for NSW Fisheries, Sydney.

O'Sullivan, D. & Kiley, T. (1996). Status of Australian aquaculture in 1994/95. *Austasia Aquaculture Trade Directory* 1996 : 2-12.

Otto, R. G. (1970). Effects of salinity on the survival and growth of pre-smolt coho salmon (*Oncorhynchus kisutch*). *Journal of Fisheries Research Board of Canada* **28**, 343-349.

Pandian, T. J. (1967). Intake, digestion, absorption and conversion of food in the fishes *Megalops cyprinoides* and *Ophiocephalus striatus*. *Marine Biology* **1**, 16-32.

Parsons, T. R., Maita, Y. & Lalli, C. M. (1984). A manual of chemical and biological methods for seawater analysis. Pergamon Press Ltd, Oxford. 173p.

Paul, A. I., Paul, J. M. & Smith, R. L. (1990). Rates of oxygen consumption of yellowfin sole (*Limanda aspera* (Pallas) relative to body size, food intake, and temperature. *Journal du Conseil International pour l' Exploration de la Mer* **47**, 205-207.

Paulson, L. J. (1980). Models of ammonia excretion for brook trout (*Salvelinus fontinalis*) and rainbow trout (*Salmo gairdneri*). *Canadian Journal of Fish and Aquatic Science* **37**, 1421-1425.

Penczak, T., Galicka, W., Molinski, E., Kusto, E. & Zalewski, M. (1982). The enrichment of a mesotrophic lake by carbon, phosphorus and nitrogen from the cage aquaculture of rainbow trout *Salmo gairdneri*. *Journal of Applied Ecology* **19**, 371-393.

Pequin, I. & Serfaty, A. (1968). La regulation hepatique at interstinale de l' ammoniemie chex la carpe. *Arch. Sci. Physiol* **22**, 449-459.

Persson, G. (1988). Relationships between feed, productivity and pollution in the farming of large rainbow trout (*Salmo gairdneri)*. Swedish Environmental Protection Agency, Stockholm,PM 3534, 48pp.

Pettersson, K. (1988). The mobility of phosphorus in fish-foods and fecals. *Verh. Internat. Verein. Limnol* **23**, 200-206.

Pffener, E. & Meske, C. (1978). Untersuchungen aber Casein and Krillmehl als einzige proteinquelle im Alleinfutter fur Karpfen Z Tierphysiol Tierenahrg, *Futtermittelkde* **40**, 76-91.

Phillips, M. J. (1985). The environmental impact of cage culture on Scotish Freshwater Lochs. Unpublished report to the Highlands and Islands Development Borad. Institute of Aquaculture, University of Stirling. 106p.

Phillips, M. J. & Beveridge, M. (1986). Cages and the effect on water condition. *Fish Farmer* **(May/June),** 17-19.

Phillips, A. M., Podoliak, Jr. H. A., Brockway, D. R., & Blazer, G. C. (1956). Cortland hatchery report 25. New York State of Conservation, Department of Fisheries Research Bulletin., Cortland hatchery report 25, 20p.

Phillips, M. J., Beveridge, M. C. M., & Macintosh, D. J. (1990). The impact of aquaculture on the coastal environment. Proceedings of ISCZCZ 1988, China Ocean Press, Beijing 1990 : p. 537-545.

Phillips, M. J., Kwei, L., & Beveridge, M. C. M. (1993). Shrimp culture and the environment : lessons from the World's most rapidly expanding warmwater aquaculture sector. In : *Environment and Aquaculture in Developing Countries* (ed. by R. S. V. Pullin, H. Rosenthal & J. L. Maclean), pp.171-197. ICLARM Conference proceedings, 31.

Pillay, T. V. R. (1990). Aquaculture - Principles and Practices. Fishing News Books, London

Pillay, T. V. R. (1992). Aquaculture and the Environment. Fishing News Books, Oxford, UK. 184p.

Pollard, J. (1986). Fishing Australia. The Book Company International Pty Ltd, Sydney.

Pollard, D. A., Ingram, B. A., Harris, J. H. & Reynolds, L. F. (1990). Threatened fishes in Australia - an overview. *Journal of Fish Biology* **37(supp. A)**, 67-78.

Pontius, F. W. (ed.) (1990). Water quality and treatment : A handbook of community water supplies (4th edn). McGraw-Hill, New York.

Porter, C. B., Krom, M. D., Robbins, M. G., Brickell, L. & Davidson, A. (1987). Ammonia excretion and total N budget for gilthead seabream (*Sparus aurata*) and its effect on water quality conditions. *Aquaculture* **66**, 287-297.

Proulx, D. and Noüe, J. de la. (1985). Growth of *Daphnia magna* on urban wastewaters tertiarily treated with *Scenedesmus* sp. *Aquaculture Engineering* **4**, 93-111.

Poxton, M. G. (1992). Water quality fluctuations and monitoring in intensive fish culture. In : *Aquaculture and the Environment 1992.* (ed. by De Pauw, N. and J. Joyce). pp. 121-143. European Aquaculture Society, Ghent, Belgium : Special Publication No 16.

Preez, H. H., Strydom, W. & Winder, P. E. D. (1986). Oxygen consumption of two marine telesots *Lithognathus lithognathus* (Cuvier 1830) (Teleosti : Sparidae). *Comparative Biochemistry and Physiology* **85B**, 313-331.

Pullin, R. S. V. 1989. Third-world aquaculture and the environment. *NAGA, The ICLARM Quarterly* **12**, 10-13.

Purchase, S. (1997). Australian writers' dictionary. Oxford University Press, Melbourne.

Ramnarine, I. W., Pirie, J. M., Johnstone, A. D. F. & Smith, G. W. 1987. The influence of ration size and feeding frequency on ammonia excretion by juvenile Atlantic cod, *Gadus morhua* L. *Journal of Fish Biology* **31**, 545-559.

Randall, D. J. & Wright, P. A. (1987). Ammonia distribution and excretion in fish. *Fish Physiology and Biochemistry* **3**, 107-120.

Reynolds, L. F. (1983). Migration patterns of five species in the Murray-Darling River system. *Australian Journal of Marine and Freshwater Research* **34**, 857-871.

Rosenthal. H., Hilge, V., Ackefors, H., Bucke, D., Stewart, J. E. & Castell, J. D. (1990). A proposed glossary on biological and technical terms relevant to aquaculture. International Council for the Exploration of the Sea. Council Meeting C. M. Mariculture Committee. First draft. 140p.

Rowland, S. J. (1983). Design and operation of an extensive aquaculture system for breeding warmwater fishes. In : *Proceedings of the first Australian freshwater aquaculture workshop*, Narrandera, NSW, February 1993. pp.121-131.

Rowland, S. J. (1984). The hormone-induced spawning of silver perch, *Bidyanus bidyanus* (Mitchell) (Teraponidae). *Aquaculture* **42**, 83-86.

Rowland, S. J. (1986). The hormone induced spawning and larval rearing of Australian native fish with particular emphasis on the golden perch, *Macquaria ambigua*, In : *Proceedings of the First freshwater Aquaculture Workshop.* (ed. by L. F. Reynolds). pp. 22-32. Department of Agriculture, NSW, Sydney, Australia.

Rowland, S. J. (1994). Preliminary evaluation of the Australian freshwater fish silver perch, *Bidyanus bidyanus* for pond culture. *Journal of Applied Aquaculture* **4(1)**, 40-48

Rowland, S. J. (1995a). The silver perch, *Bidyanus bidyanus*, and its potential for aquaculture. In : *Silver Perch Culture : Proceedings of Silver Perch Workshops, Grafton and Narrandera, April 1994.* (ed. by S.J.Rowland & C. Bryant), pp.9-11. *Austasia Aquaculture*, Hobart for NSW Fisheries, Sydney.

Rowland, S.J. (1995b). Water quality in the intensive pond culture of silver perch. In : *Silver Perch Culture : Proceedings of Silver Perch Workshops, Grafton and Narrandera, April 1994.* (ed. by S.J.Rowland & C. Bryant), pp. 41-49. *Austasia Aquaculture*, Hobart for NSW Fisheries, Sydney.

Rowland, S. J. (1995c). Production of fingerling and market-size silver perch in earthen ponds. In : *Silver Perch Culture : Proceedings of Silver Perch Workshops, Grafton and Narrandera, April 1994.* (ed. by S.J.Rowland & C. Bryant), pp. 41-49. *Austasia Aquaculture*, Hobart for NSW Fisheries, Sydney.

Rowland, S. J. (1995d). High density pond culture of silver perch, *Bidyanus bidyanus*. *Asian Fisheries Science* **8**, 73-79.

Rowland, S. J. (1996). Water quality in the intensive culture of silver perch. *Austasia Aquaculture* **10(1)**, 35-42.

Rowland, S. J. & Allan, G. L. (1994). Production of fingerling, *Bidyanus bidyanus* (Teraponidae) at two densities in earthen ponds in Australia. *The Progressive Fish Culturist* **56**, 296-298.

Rowland, S. J. & Barlow, C. G. (1991). Fish biology the right prerequisites : a case study with freshwater silver perch (*Bidyanus bidyanus*). *Austasia Aquaculture* **5**, 27-30.

Rowland, S. J. & Bryant, C. (1995). (Editors). Silver perch Culture. Proceedings of Silver Perch Workshops, Grafton and Narrandera, April 1994. *Austasia Aquaculture*, Hobart for NSW Fisheries, Sydney.

Rowland, S. J. & Ingram, B. A. (1991). Diseases of Australian Native Freshwater Fishes with particular emphasis on the ectoparasitic and fungal diseases of Murray cod (*Maccullochella peeli*), Golden perch (*Macquaria ambigua*) and silver perch (*Bidyanus bidyanus*). *Fisheries Bulletin* 4, NSW Fisheries, Sydney, 33p.

Rowland, S. J. & Kearney, R. E. (1992). The prospects for the commercial aquaculture of silver perch (*Bidyanus bidyanus*). A research project arising from a review of freshwater aquaculture. NSW Fisheries, Sydney. 16p.

Rowland, S. J., Allan, G. L., Clark, K., Hollis, M. & Pontifex, T. (1994). Production of fingerling *Bidyanus bidyanus* (Teraponidae) at two densities in earthen ponds in Australia. *The Progressive Fish Culturist* **56**, 296-298.

Rowland, S. J., Allan, G. L., Hollis, M. & Pontifex, T. (1995). Production of the Australian freshwater silver perch, *Bidyanus bidyanus* (Mitchell), at two densities in earthen ponds. *Aquaculture* **130**, 317-328.

Rowling, K. R. (1990). Changes in the stock composition and abundance of spawning gemfish *Rexea solandri* (Cuvier), Gempylidae, in south-eastern Australian waters. *Australian Journal of Marine and Freshwater Research* **41**, 145-163.

Rychly, J. (1980). Nitrogen balance in trout II. Nitrogen excretion and retention after feeding diets with varying protein and carbohydrate levels. *Aquaculture* **20**, 3 43-350.

Rychly, J. & Marina, B. A. (1977). The ammonia excretion of trout during a 24 hour period. *Aquaculture* **11**, 173-175.

Ryther, J. H. & Dunstan, W. M. (1971). Nitrogen, phosphorus and eutrophication in the marine environment. *Science* **171**, 1008-1013.

Sadykhov, D. A., I. B. Bogatova & Filatov, V. L. (1975). Amino acid composition of certain freshwater zooplankton representative. *Gidrobiol Zh* **11**, 53-57. (In Russain).

Sagar, P. M. & Glova, G. L. (1988). Diel feeding periodicity, daily ration and prey selection of a riverine population of juvenile chinook salmon, *Oncorhynchus tshawyscha* (Walbaum). *Journal of Fish Biology* **33**, 643-653.

Santos, J. D. & Jobling, M. (1991). Factors affecting gastric evacuation in cod, *Gadus morhua* L. for single meals of natural prey. *Journal of Fish Biology* **38**, 697-713.

Sargent, J. R., Mcintosh, R., Bauermeister, A. & Blaxter, J. H. S. (1979). Assimilation of the was esters of marine zooplankton by herring (*Clupea harengus*) and rainbow trout (*Salmo gairdneri*). *Marine Biology* **51**, 203-207.

Sayer, M. D. & Davenport, J. (1987). The relative importance of the gills to ammonia and urea excretion of five seawater and one freshwater teleost species. *Journal of Fish Biology* **31**, 561-570.

Savitz, I. (1971). Nitrogen excretion and protein consumption of the bluegill sunfish (*Lepomis macrochirus*). *Journal of the Fisheries Research Board of Canada* **28**, 449-457.

Savitz, J., Albanese, E., Evinger, M. J. & Kolasinski, P. (1977). Effect of ration level on nitrogen excretion, nitrogen retention and efficiency of nitrogen utilization for growth in largemouth bass (*Micropterus samonides*). *Journal of Fish Biology* **11**, 185-192.

Schafer, A., Koppe, W. M., Meeyre-Burgdorff, K.-H. and Gunther, K. D. (1995). Effects of a microbial phytase on the utilization of native phosphorus by carp in a diet based on soybean meal. *Water Science Technology* **31**, 149-155.

Schwartz, M. & Boyd, C. E. (1994). Effluent quality during harvest of channel catfish from watershed ponds. *Progressive Fish Culturist* **56**, 25-32.

Seymour, E. A. & Bergheim, A. (1991). Towards a reduction of pollution from intensive aquaculture with reference to the farming of salmonids in Norway. *Aquaculture Engineering* **10**, 73-88.

Shcherbina, M. A. & Kazlauskene, O. P. (1971). Water temperature and the digestibility of nutrient substances by carp. *Hydrobiologia Journal (USSR)*, **7(3)**, 40-44.

Shim, K. F. & Ho, C. S. (1989). Calcium and phosphorus requirements of guppy, *Poecilia reticulata*. *Nippon Suisan Gakkaishi* **55(1),** 1947-1953.

Silver, G. R., Higgs, D. A., Dosajh, B. S., Mckeown, B. A., Deacon, G. & French, D. (1993). Effect of dietary protein to lipid ratio on growth and chemical composition of chinook salmon (*Oncorhynchus tshawytscha*) in sea water. In : *Fish Nutrition in Practice* (ed. by ed. by S. J. Kaushik & P. Luquet). pp. 459-468. IV International Symposium on Fish Nutrition and Feeding, Biarritz, France, June 24-27, 1991. INRA 61.

Singh, B. N., Sinha, V. R. P. & Chakraborty, D. P. (1979). Effects of protein quality and temperature on the growth of fingerlings of rohu, *Labeo rohita* (Hamilton). In : *Finfish Nutrition and Fishfeed Technology* (ed. by J. E. Halver & K. Tiewes), pp. 303-310. Proceedings of the World Symposium on Finfish Nutrition and Fishfeed Technology, Hamburg, June 1978, vol. II.

Smith, H. W. (1929). The excretion of ammonia and urea by gills of fish. *Journal of Biology and Chemistry* **81**, 727-742.

Smith, M. A. K. & Thorpe, A. (1976). Nitrogen metabolism and trophic input in relation to growth in freshwater and saltwater *Salmo gairdneri*. *Biological Bulletin* **150**, 139-151.

Smith, M. A. K., Jenkins, W. E. & Haaggerty, R. W. (1986). Growth and survival of juvenile striped bass (*Morone saxatilis*) x white bass (*M. chrysops*) hybrids reared at different salinities. Proceedings Annula Conference of Southeast Association of Fish and Wildlife Agencies **40**, 143-151.

Smith, R.L., Paul, J. M. & Paul, A. I. (1989). Gastric evacuation in walleye pollock, *Therogrn chaclogramma*. *Canadian Journal of Fisheries and Aquatic Science* **46**, 489-493.

Solbe, J. F.de L G. (1982). Fish farm effluents : a United Kingdom survey. In : *Report of the EIFAC workshop on fish farm effluents,* Silkeborg,Denmark, 26-28 May 981. *EIFAC Technical Paper* **41**, 29-55.

Solberg, S. O. & Bregnballe, F. (1982). Pollution from trout feed with minced trash f fish. In : *Report of the EIFAC workshop on fish-farm effluents, Silkeborg, Denmark, 26-28 May 1981.* (ed. by J. S. Albaster), pp. 65-71. *FAO/EIFAC Technical Paper* **41**.

Speece, R. E. (1973). Trout metabolism characteristics and the rational design of nitrification facilities for water reuse in hatcheries. *Transaction of the American Fisheries Society* **2**, 323-324.

Spenelli, J. (1979). Preparation of salmonid diets containing zooplankton and their effect on organoleptic properties of pen-reared salmonids. In : *Proceedings of the World Symposium on Finfish Nutrition and Fish Feed Technology, Hamburg 20-23 June 1978* (ed. by J. E. Halver & K. Tiews editors). pp. 383-392. Volume II, Berlin 1979.

Starling, S. (1992). The Australian Fishing Book. Sandipiper Press (NSW) Ltd, 512p.

State of Victoria. (1995). Nutrient management strategy for Victorian Inland Waters. State of Victoria., Australia. 73p.

Stewart, J. E. (1997). Environmental impacts of aquaculture. *World Aquaculture*. **28(1)**, 47-52.

Stickney, R. R. (1986). Culture of non-salmonid freshwater fishes. CRC Press, Boca Raton, Florida.

Stickney, R. R. (1991). Effects of salinity on aquaculture production. In : *Aquaculture and Water Quality.* (ed. by D. E. Brune and J. R. Tomasso). p.105-132. Advances in World Aquaculture, volume 3. The World Aquaculture Society, LA., USA.

Stickney, R. R. & Lovell, R. T. (1977). Nutrition and feeding of channel catfish. A report from the Nutrition subcommittee of regional Research Project S-83. Southern Cooperative Series. Bulletin 218. 67p.

Storebakken, T. & Austreng, E. (1987a). Ration level of salmonids. I. Growth, survival, body composition and feed conversion in Atlantic salmon fry and fingerlings. *Aquaculture* **60**, 189-206.

Storebakken, T. & Austreng, E. (1987b). Ration level of salmonids. II. Growth, feed intake, protein digestibility, body composition and feed conversion in Rainbow trout weighing 0.5-1.0 kg. *Aquaculture* **60**, 207-221.

Sukumaran, N. & Kutty, M. N. (1977). Oxygen consumption and ammonia excretion in the catfish *Mystus armatus* with special reference to swimming speed and ambient oxygen. *Proceedings of the Indian Academy of Sciences* **86B**, 195-206.

Sumari, O. (1982). A report on fish farm effluents in Finland. Report of the EIFAC Workshop on Fish Farm Effluents. *EIFAC Technical Paper* **41**, 21-27.

Suzuki, T. (1981). Fish and krill protein : Processing Technology. Applied Science Publishers Ltd, London.

Tacon, A. (1981). Speculative review of possible carotenoid function in fish. *Progressive Fish Culturist* **43**, 205-208.

Tacon, A. G. J. (1990). Standard methods for the nutrition and feeding of farmed fish and shrimp, vol. I. The essential nutrients, Argent Laboratories Press, Redmond, Washington, USA.

Tacon, A. G. J. & De Silva, S. S. (1993). Mineral composition of some commercial fish feeds available in Europe. *Aquaculture* **31**, 11-20.

Tay, S. H., Rajbanshi, V. K., Ho, W. H., Chew, J. & Yap, E. A. (1991). Culture of cladoceran *Moina micrura* kurz using agroindustrial wastes. In : *Asian Fish Nutrition Workshop, Vijayawada India, September 1990.* (ed. by S. S. de Silva). pp. 135-141. Fish Nutrition Research in Asia. Proceedings of the Fourth Asian Fish Nutrition Workshop. no.5.

Tecator. (1990). Instruction manual. The Aquatec System. Hoganas : Sweden. 49p.

Thorpe, J. E., Talbot,C., M. S. Miles, M. S., Rawlings, C. & Keay, D. S. (1990). Food consumption in 24 hours by Atlantic salmon *Salmo salar L.* in a sea cage. *Aquaculture* **90**, 41-47.

Thurstan, S. & Rowland, S. J. (1995). Techniques for the hatchery production of silver perch. In : *Silver Perch Culture : Proceedings of Silver Perch Aquaculture Workshops. Grafton and Narrandera, April, 1994.* (ed. by S. J. Rowland & C. Bryant). pp. 29-39. Austasia Aquaculture, Hobart for NSW Fisheries, Sydney.

Torrissen, O. J. (1984). Pigmentation of salmonids - effect of carotenoids in eggs and start feeding diet on survival and growth rate. *Aquaculture* **43**, 185-193.

Tradwell, R., McKelvie, L., & Maguire, G. B. (1992). Potential of Australian aquaculture. Australian Bureau of Agricultural and Resource Economics. (ABARE), ABARE Research Report No. 92.2. Commonwealth of Australia, Canberra. 81p.

Tucker, J. W. (1992). Feeding intensively-cultured marine fish larvae. In : *Proceedings of the Aquaculture Nutrition Workshop, Salamander Bay, 15-17 April 1991.* (ed. by G. L. Allan & W. Dall). pp. 129-146. NSW Fisheries, Brackish Water Fish Culture Research

Usher, M. L., Talbot, C. & Eddy, F. B. (1990). Effects of transfer to seawater on digestion and gut function in Atlantic salmon smolts (*Salmo salar* L.). *Aquaculture* **90**, 85-96.

Vijverberg, J. and Frank, T. H. (1976). The chemical composition and energy contents of copepods and cladocerans in relation to their size. *Freshwater Biology* **6**, 333-345.

Walker, T. (1993). Silver perch : Giving small farmers a slice of the action. *Austasia Aquaculture* **7(5),** 3-6.

Walker, T. (1994a). Moratorium on silver perch all for a good cause says Government. *Austasia Aquaculture* **8(5)**, 3-6.

Walker, T. (1994b). Catfish model for silver perch put under microscope. *Austasia Aquaculture* **8(5),** 24-36.

Walker, K. F., & Hillmann, T. J. (1982). Phosphorus and nitrogen loads associated with the River-Murray near Albury-Wodonga and their effects on phytoplankton populations. *Australian Journal of the Marine and Freshwater Research* **33**, 223-243.

Walker, T & Caney, D. (1996). Silver perch extension officer reflects on progress. *Austasia Aquaculture* **10(2),** 34-36.

Warren-Hansen, I. (1982). Evaluation of matter discharged from trout farming in Denmark. In : *Report of the EIFAC workshop on fish farm effluents*, Silkeborg, Denmark, 26-28 May 1981. *EIFAC Technical Paper* **41**, 57-63

Watanabe, T. (1991). Past and present approaches to aquaculture waste management in Japan. In : *Nutritional Strategies and Aquaculture Waste*. (ed. by C. B. Cowey & C. Y. Cho), pp. 137-154. Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste. University of Guelph, Guelph, Ontario, Canada, 1990.

Watanabe, T., Arakawa, C., Kitajima, C. & Fujita, S. (1978). Nutritional quality of living feed from the view point of essential fatty acids for fish. *Bulletin of the Japan Society of Science and Fisheries* **44**, 1223-1227.

Watanabe, T., Murakami, A., Takeuchi, L., Nose, T. & Ogino, C. (1980). Requirement of chum salmon held in freshwater for dietary phosphorus. *Bulletin of the Japan Society Science and Fisheries* **46**, 361-367.

Watanabe, T., Kitajima, C. & Fujita, S. (1983). Nutritional values of live organisms used in Japan for mass propagation of fishes : A review. *Aquaculture* **34**, 115-143.

Weatherly, A. H. & Gill, H. S. (1987). The biology of fish growth. Academic Press. London. 443p.

Webb, P. W. (1978). Partitioning of energy into metabolism and growths. In : *Ecology of freshwater fish production* (ed. by S. D. Gerking). pp.184-214. Blackell Scientific Publications, London.

Welch, E. B. and Lindell, T. (1980). Ecological effects of wastewater.- Applied limnology and pollutant effects. chapter 4. Nutrient cycles. p. 54-81.

Whitley, G. P. (1960). "Native Freshwater Fishes of Australia". (Jacaranda Press : Brisbane).

Wiggs, A. J., Henderson, E. B., Saunders,R. I. & Kutty, M. N. (1989). Activity, respiration, and excretion of ammonia by Atlantic salmon (*Salmo salar*) smolt and postmolt. *Canadian Journal of Fisheries and Aquatic Science* **46**, 790-795.

Wilkie, M. P. & Wood, C. M. (1991). Nitrogenous waste excretion, acid-base regulation, and ionicregulation in rainbow trout (*Oncorhynchus mykiss*) exposed to extremely alkaline water. *Physiological Zoology* 64, 790-795.

Windell, J. T., Foltz, J. W. & Sarokon, J. A. (1978). Effect of size, temperature, and amount fed on nutrient digestibility of a pelleted diet by rainbow trout, *Salmo gairdneri*. *Transactions of the American Fisheries Society* **107(4)**, 613-616.

Wood, J. D. (1958). Nitrogen excretion in some marine teleosts. *Canadian Journal of Biochemistry and Physiology* **36**, 1237-1242.

Wu, R. S. S., Lam, K. S., MacKay, D. W., Lau, T. C. & Yam, V. (1994). Impact of marine fish farming on water quality and bottom sediment : A case study in the sub-tropical environment. *Marine Environmental Research* **38**, 115-145.

Yarris, L. (1981). Trout manure as fertilizer. *Agriculture Research* (USDA). **30**, 9.

Yone, Y. & Toshima, T. (1979). The utilization of phosphorus in fish meal by carp and black sea bream. *Bulletin of the Japan Society of Science and Fisheries* **45**, 753-756.

Yurkowski, M. & Tabachek, J. L. (1979). Proximate and amino-acid composition of some natural fish foods. In : *Proceedings of the World Symposium on Finfish Nutrition and Fishfeed Technology,* (ed. by J. E. Halver & K. Tiews). pp. 435-448. Hamburg 20-23 June, 1978, vol. I. Bergin.

Zar, J. H. (1984). Biostatistical analysis. second edition. Prentice-Hall International,Inc, Emglewood,Cliffs, New-Jersey. 620P.

APPENDIX -I

PUBLISHED /ACCEPTED PAPERS

Editorial

The Network of Tropical Aquaculture Scientists (NTAS) and the *Aquabyte* section of *Naga* have come under a new management. The outgoing NTAS Coordinator and Editor of *Aquabyte* Dr. R.S.V. Pullin and Secretary Ms. Maan P. Bimbao have done a great job, who, in this issue, trace the history of the network since its inception.

With ever increasing demand for fish, all eyes are focused on increasing fish production through aquaculture. Much aquaculture research has been and is being done in developing and developed countries and the results of these studies are not easily available to researchers in developing countries who lack access to good libraries. *Naga*, with its wide circulation, could communicate research results to a large section of the research community. NTAS encourages young scientists to publish their research results in *Aquabyte* along with those of the Specialists.

M.V. Gupta

Aspects of Phosphorus Pollution from Aquaculture

Golam Kibria, Dayanthi Nugegoda, Paul Lam and Robert Fairclough

Introduction

Phosphorus is an essential element for living organisms and exists in waterbodies as dissolved and particulate forms. Phosphorus is required for optimum growth, feed efficiency, bone development and maintenance of acid-base regulation in fish (Lall 1989; Ketola and Richmond 1994). The presence of high concentration of phosphates in water may indicate presence of pollution as it may accelerate plant growth (Beveridge 1987) and disrupt the aquatic ecosystem thereby benefiting certain species and altering species diversity in affected areas (Anon. 1987; OCE 1988). Eutrophication of waterbodies is often correlated with the phosphorus loading into the environment (Kaushik 1992) and aquaculture has been identified as one of the sources of phosphorus pollution (EPA 1995). Details of the impacts of eutrophication is given in Bernhardt (1981). Phosphorus must be provided in fish feed because of its low concentration in water (Lall 1991). Studies made in Europe and Northern America have revealed a phosphorus surplus in most commercial feeds which is above actual requirements (Tacon and de Silva 1983; Beveridge 1987); or is supplied in a form which is unavailable to the fish (Beveridge 1987). Surplus phosphorus is excreted, while unavailable phosphorus is passed out in the feces (Beveridge 1987). Discharge of phosphorus from fish farms and hatchery effluents have caused phosphorus pollution in Nordic countries, North America and Europe (Bernhardt 1981; Alabaster 1982; Beveridge 1984; Enell 1987; Folke and Kautsky 1989; Ketola 1990; Bratten 1991; Foy and Rosell 1991a; Lall 1991). This article examines the path of phosphorus pollution, quantification/prediction of phosphorus load from aquaculture and remedial measures.

Path of Phosphorus Loss

Feed is the main source of phosphorus loadings from aquaculture to the environment (Ketola 1990; Seymour and Bergheim 1991). There are a number of ways in which phosphorus can be lost during aquaculture operation such as feed fines, uneaten food, feces, dead fish and excretion (Fig. 1). However, the main loading of phosphorus to the environment was reported to be via fecal pellets (Pillay 1992; Kibria et al. 1995). Fish excrete phosphorus in soluble and particulate forms (Lall 1979). The particulate form (feces) settles to the bottom of the tank or accumulates in the sediment and soluble form is lost through urine in the form of phosphate (Lall 1991; Pillay 1992). The soluble fraction is referred to as either dissolved inorganic phosphorus or orthophosphate or soluble reactive phosphate. It is the dissolved fraction that is most available for plant growth (Bostrom et al. 1988).

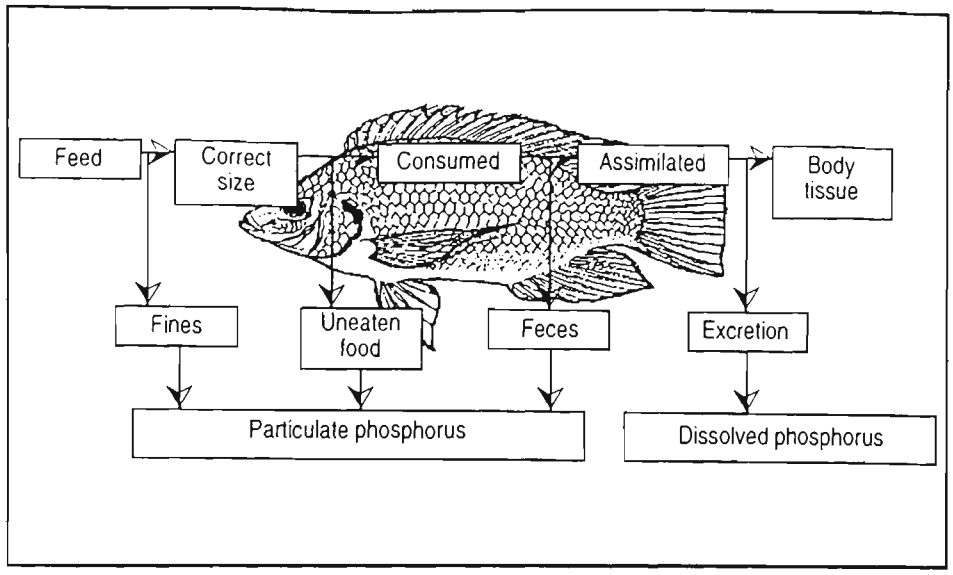

Fig. 1. Flowchart of origin of phosphorus in aquaculture from feeding.

Table 1. Comparision of phosphorus loss rates (kg fish produced) in aquaculture.

Country	Phosphorus loss (kg/t)	Species	Culture method	Reference
Denmark	11.0	Rainbow trout	Ponds	Warren-Hansen (1982)
Finland	18.3	Rainbow trout	Ponds	Sumari (1982)
UK	15.7	Rainbow trout	Ponds	Solbe (1982)
N. Ireland	25.6	Rainbow trout	Tanks	Foy and Rosell (1991b)
Norway	9.0	Atlantic salmon	Ponds	Ibrekk (1989)
USA	10-15	Rainbow trout	Jars	Ketola (1991)
Poland	23	Rainbow trout	Cages	Penczak et al. (1982)
Norway	13.5	Rainbow trout	Cages	Enell and Lof (1983)
UK	27	Rainbow trout	Cages	Phillips (1985)
Spain	39.2	Rainbow trout	Ponds	Tarazona et al. (1993)
Canada	6.0	Brown trout	Tanks	Cho et al. (1991)

Data on Phosphorus Pollution from Aquaculture

Phillips et al. (1990) estimated that 85% of phosphorus fed to fish was lost to the environment. In a study made with native Australian silver perch (*Bidyanus bidyanus*), Kibria et al. (1995) demonstrated that phosphorus loss from aquaculture can vary with temperature, being higher at a lower temperature (95% lost at 20°C) versus (80% lost at 25°C). The phosphorus loss rates of different commercial aquaculture species is reported to be mostly between 9 and 40 kg of fish production (Table 1). The feed type could also determine the level of phosphorus loss since a much higher phosphorus loss could result if trash feed is supplied to fish compared to dry and moist feed (Warren-Hansen 1982).

Quantification of Phosphorus Loss from Aquaculture

The phosphorus loss/pollution load to the environment can be estimated from data on retention in fish carcass using feed/gain data (Ketola 1990). It is equal to the difference between what is added by the feed and what is utilized for fish production (Foy and Rosell 1991b). In order to determine the phosphorus retention in carcass growth, survival and feed conversion data are needed to be determined initially. Next, phosphorus concentration is to be analyzed in feed, fish carcass, uneaten food and solid wastes (feces). Then the amount of soluble and suspended phosphorus (P) discharged in water could be determined using the following phosphorus balance equation:

P discharged in water = P supplied as feed - (P deposited in body tissues + P in uneaten food + P in solid wastes)

The food conversion ratio (FCR) can play a significant role in

determining the level of phosphorus pollution expected since an increase in FCR value from 1.0 to 1.5 may increase pollution load to about 86% for total phosphorus (Storebakken and Austreng 1987a, 1987b). Therefore, improvement of FCR is vital for reducing phosphorus pollution from aquaculture. A strong correlation was found between the phosphorus loss rates and the FCR with silver perch: the better the FCR, the lower the rates of phosphorus loss (Fig. 2).

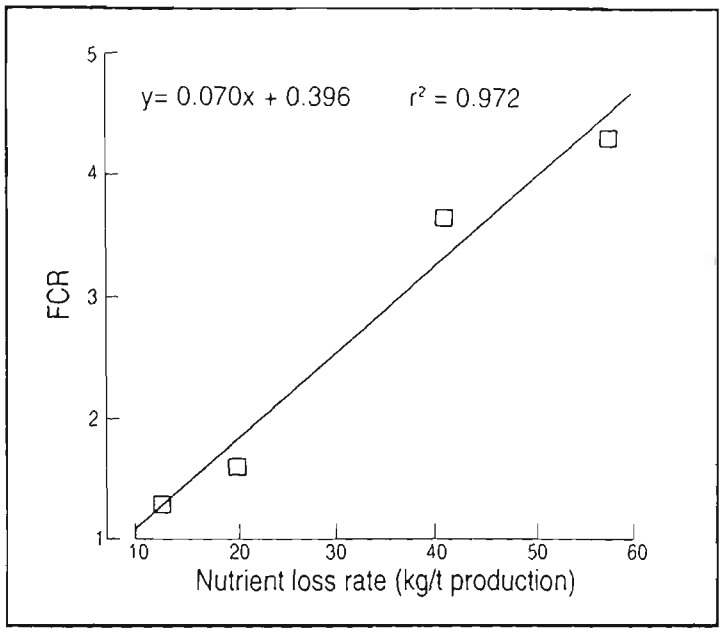

Fig. 2. Relationship between the food conversion ratio (FCR) and average phosphorus loss rate per tonne of silver perch (Bidyanus bidyanus) production. The lower FCR value obtained at 25°C when good growth is achieved and FCR at 20°C when comparatively poorer growth resulted (Kibria et al. 1995).

Simple Calculation of Phosphorus Retention

If a commercial diet contains 1.7% phosphorus and retention of phosphorus is estimated to be 21%, then phosphorus wasted per ton of feed supplied can be calculated as follows:

The amount of P present per ton of feed	=	1 000 kg x 0.017 kg= 17.0 kg
If P retention is 21%, the amount of P retained	=	17 kg x 0.21= 3.57 kg
The amount of P discharged (as solid and dissolved) to environment	=	17-3.57 = 13.43 kg/t of feed fed
Values at different FCR		
At FCR 1.0:1, the amount of P present per ton of feed	=	1 000 kg x 0.017 kg =17.0 kg P
At FCR 1.5:1, the amount of P present per ton of feed	=	1 500 kg x 0.017 kg = 25.5 kgP
At FCR 2.0:1, the amount of P present per ton of feed	=	2 000 kg x 0.017 kg = 34.0 kg P
Plosses to environment		
at FCR 1.0:1 → (17.0-3.57) →	=	13.43 kg P/t feed fed
at FCR 1.5:1 → (25.5-5.35) →	=	20.14 kg P/t feed fed
at FCR 2.0:1 → (34.0-7.14) →	=	26.86 kg P/t feed fed

Phosphorus as an Indicator of Water Quality

The phosphorus concentration at any given time may be used to assess the quality of freshwater aquatic environment (Table 2). Based on this, it appears that effluents from fish farms and hatcheries will be of degraded quality since the discharge from fish farms and hatcheries are reported to contain 0.15 mg/l total phosphorus (Warren-Hansen 1982) and 0.1 mg/l dissolved phosphate (Alabaster 1982), which leads to the possibility of eutrophication in waterbodies which receive direct fish farm effluents.

Identification of the "Right Phosphorus"

As mentioned above, feed is the main source of phosphorus pollution in aquaculture. In plants, two-thirds of phosphorus is bound in phytin that trout and salmon cannot digest (Ketola 1990). It has been demonstrated that phosphorus requirements are species-specific and surplus phosphorus provided through feed is either excreted. Most of the animal and inorganic sources of phosphorus are readily available to fish (Ketola 1990). Ketola (1982) reported that the source of the phosphorus significantly influenced the retention as well as loss rate, e.g., defluorinated rock phosphate (DRP) resulted in good growth of rainbow trout with a reduction of 46% in phosphorus discharge (Ketola 1985) in comparison to dicalcium phosphate (Ketola 1991). Additionally, the bioavailability of dietary phosphorus is influenced by the digestibility of diet, particle size, interaction with other nutrients, feed processing and water chemistry (Lall 1991). Salmonids utilize phosphorus from fish meal more efficiently than do

carp (Yone and Toshima 1979) whereas availability to tilapia is low.

Phosphorus Release from Food and Feces

The majority of phosphorus loss is in particulate form such as feces and uneaten food. However, release to the environment of phosphorus from food and feces depends on physico-chemical characteristics of the environment such as pH, temperature, oxygen, turbulence and microbial activity (Persson 1988). Phosphorus release from fish food was observed to be accelerated in acidic rather than in neutral or alkaline medium (Fig. 3).

The bioavailability of phosphorus from fish food and feces depends on proportion of labile phosphorus there. The fish food having the highest total phosphorus content has been found to have the highest concentration of labile phosphorus (Kibria et al. 1995).

Overcoming Phosphorus Pollution from Aquaculture

To reduce phosphorus pollution, the following are important:

1. estimation of phosphorus balance of species under aquaculture (Ketola 1991);
2. feed composition and type (e.g., extruded feeds are more digestible and generate less dust and solid waste and also result in better FCR (Warren-Hansen 1982; Matty 1990);
3. feeding techniques (e.g., avoiding overfeeding and adjusting feed amount and frequency to the temperature (Seymour and Bergheim 1991);
4. formulation of diets to meet nutrient requirements and proper choice of dietary ingredients (Kaushik 1992); and
5. reduction of phosphorus levels in feeds without affecting growth, feed efficiency, health and reproduction (Kendra 1991; Lall 1991).

Table 2. Criteria for assessing water quality of freshwater based on phosphorus level (OCE 1988).

	Total phosphorus (mg/l)	Dissolved phosphorus (mg/l)
Excellent	<0.010	<0.008
Good	<0.025	<0.020
Moderate	<0.050	<0.040
Poor	<0.100	<0.080
Degraded	<0.100	<0.080

Fish farm effluents can increase phosphorus levels and consequently cause eutrophication in receiving waters. As a result, further use of such waterbodies for recreational or domestic or industrial purpose will be seriously affected. In Asia, where aquaculture is a booming industry and demand to use water for agriculture and domestic purposes is high, it is essential that more research on phosphorus pollution is conducted. As phosphorus requirements are species-specific, it is vital that phosphorus be provided in feed at a level that maximizes growth of fish but minimizes pollution of the environment. In this context, research on the development of "low pollution diets" are of utmost importance. The nutrient from aquaculture can be minimized by retaining wastewater (effluents) in holding ponds and by reusing wastewater on lands for growing crops in integrated aquaculture-agriculture system. The other alternative would be to develop high energy diets which reduce phosphorus discharge to the environment compared to normal feed (Bohl et al. 1992). Though aquaculture has been identified as one of the point sources of phosphorus pollution, phosphorus loadings to the environment from fish farms is minimal compared to other sources such as agriculture (Table 3).

Table 3. Typical nutrient concentration for various point source discharges (EPA 1995).

Source	Total phosphorus (mg/l)
Fish farms	0.07
Dairy shed effluent	34.0
Feedlot effluent	1.50

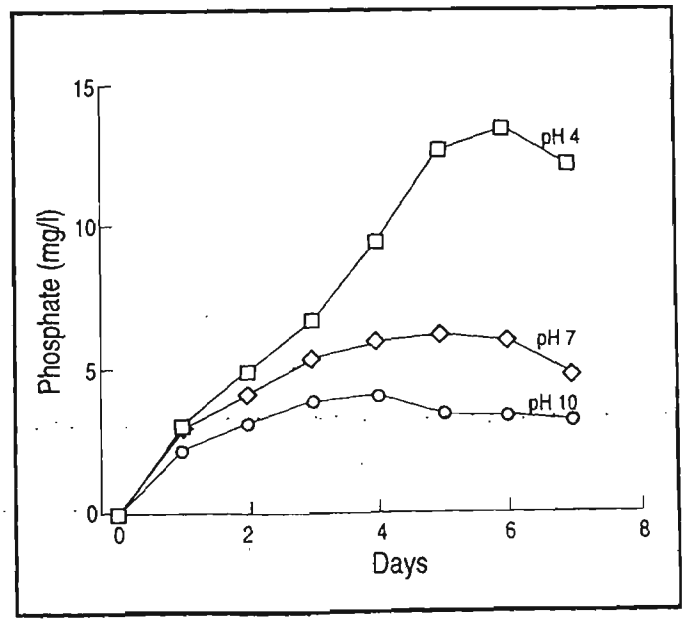

Fig. 3. The release of phosphate from fish food at different pH levels. The food was incubated at 20°C for seven days.

References

Alabaster, J.S. 1982. Report of the EIFAC Workshop on Fish-farm Effluents. FAO/EIFAC Tech. Pap. 41, 166 p.

Anon. 1987. Model permit package fish culture industry. US Environment Protection Agency, New Jersey. 45 p.

Bernhardt, H. 1981. Z. Wasser Abwasser Forsch. 14 Nr. 1.S.:14-26.

Beveridge, M.C.M. 1984. Cage and pen farming: carrying capacity models and environmental impact. FAO Fish. Tech. Pap. 255. FIRI/T255, 131 p.

Beveridge, M.C.M. 1987. Cage aquaculture. Fishing News Books Ltd., England. 352 p.

Bohl, M., B. Moav, V. Hilge and H. Rosenthal. 1992. The influence of feeds on trout pond effluents, p. 109-118. *In* B. Moav, V. Hilge and H. Rosenthal (eds.) Progress in aquaculture research. Spec. Publ. Europ. Aquacult. Soc. 17.

Bostrom, B., G. Persson and B. Broberg. 1988. Bioavailability of different phosphorus forms in freshwater systems. Hydrobiologia 170:133-155.

Bratten, B. 1991. Status of pollution from aquaculture in six Nordic countries, release of pollutants, effects and waste water treatment, p. 40-41. *In* N. De Pauw and J. Joyce (eds.) Aquaculture and the Environment. Eur. Aquacult. Soc. Sp. Pub. 14.

Cho, C.Y., J.D. Hynes, K.R. Wood and H.K. Yoshida. 1991. Quantification of fish culture wastes by biological (nutritional) and chemical (limnological) methods: the development of high nutrient dense (HND) diets, p. 37-50. *In* C.B. Cowey and C.Y. Cho (eds.) Nutritional strategies and aquaculture waste. Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste. University of Guelph, Ontario, Canada.

Enell, M. 1987. Environmental impact of cage fish farming. Rep. Inst. Freshwat. Res. 63:106.

Enell, M. and J. Lof. 1983. Environmental impact of aquaculture sedimentation and nutrition loadings from fish cage culture farming. Vatten/Water 39(4):364-375.

EPA. 1995. Protecting water quality in Port Phillip Bay. State Environment Protection Policy (Waters of Victoria). Publication No. 434. Environment Protection Authority, Melbourne, Australia.

Folke, C. and N. Kautsky. 1989. The role of ecosystems for a sustainable development of aquaculture. Ambio 18:234-243.

Foy, R.H. and R. Rosell. 1991a. Fractionation of phosphorus and nitrogen loadings from a Northern Ireland fish farm. Aqua-culture 96:31-42.

Foy, R.H. and R. Rosell. 1991b. Loadings of nitrogen and phosphorus from a Northern Ireland fish farm. Aquaculture 96:17-23.

Ibrekk, H.O. 1989. Model for determining pollution loading to Norwegian coastal waters (i.e., Lenka-zones) NIVAonotat O-88145, Oslo.

Kaushik, S.J. 1992. Recent trends in the development of high-energy diets for salmonids. 2e Conference Internazionale "Produzione di alimenti per animali", Piacenza, Italie, 25-26 fev. 1992.

Kendra, W. 1991. Quality of salmonid effluents during a summer low-flow season. Trans. Am. Fish. Soc. 120:43-51.

Ketola, H.G. 1982. Effect of phosphorus in trout diets on water pollution. Salmonid 6(2):12-15.

Ketola, H.G. 1985. Effect of phosphorus in trout and salmon feeds on water pollution, p. 465-473. *In* C.B. Conwey, A.M. Mackie and J.G. Bell (eds.) Nutrition and feeding in fish. Aberdeen, U.K.

Ketola, H.G. 1990. Dietary control of phosphorus in hatchery effluents. Proceedings of the 41st Annual Northwest Fish Culture Conference. Idaho Department of Fish and Game, Boise, Idaho.

Ketola, H.G. 1991. Managing fish hatchery duscharges of phosphorus through nutrition, p. 187-197. *In* Proceedings from the Aquaculture Symposium, Engineering Aspects of Intensive Aquaculture. Northeast Regional Agricultural Engineering Service Publication 49. Cornell University, Ithaca, New York.

Ketola, H.G. and M.E. Richmond. 1994. Requirement of rainbow trout for dietary phosphorus and its relationship to the amount discharged in hatchery effluents. Trans. Am. Fish. Soc. 123:587-594.

Kibria, G., D. Nugegoda, P. Lam and R. Fairclough. 1995. Phosphorus balance in a simulated aquaculture system: a case study with the native Australian fish silver perch *Bidyanus bidyanus* (Mitchell) Teraponidae. Paper presented at the Fourth Asian Fisheries Forum, 16-20 October 1995, Beijing, China.

Lall, S.P. 1979. The minerals, p. 86-97. *In* J.E. Halver and K. Tiews (eds.) Fish nutrition and feed technology. Vol. 1. Heenemann, Berlin.

Lall, S.P. 1989. The minerals, p. 219-257. *In* J.E. Halver (ed.) Fish nutrition. 2nd edition. Academic Press Inc., San Diego, California.

Lall, S.P. 1991. Digestibility, metabolism and excretion of dietary phosphorus in fish. p. 163-185. *In* C.B. Cowey and C.Y. Cho (eds.) Nutritional strategies and aquaculture waste. Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste. University of Guelph, Ontario, Canada.

Liao, P.B. and R. Mayo. 1972. Salmonid hatchery water re-use systems. Aquaculture 1:317-355.

Matty, A.J. 1990. Feeds in the fight against pollution. Fish Farming International, January:16-17.

Penczak, T., W. Galicka, M. Molinski, E. Kusto and M. Zalewski. 1982. The enrichment of a mesotrophic lake by carbon, phosphorus and nitrogen from the cage aquaculture of rainbow trout *Salmo gairdneri*. J. App. Ecol. 19: 371-393.

Persson, G. 1988. Realtionships between feed, productivity and pollution in the farming of large rainbow trout (*Salmo gairdneri*). PM 3534, Swedish Environmental Protection Agency, Stockholm, Sweden. 48 p.

Phillips, M.J. 1985. The environmental impact of cage culture on Scottish freshwater Lochs. Institute of Aquaculture, Stirling. 106 p.

Phillips, M.J., M.C.M. Beveridge and D.J. Macintosh. 1990. The impact of aquaculture on the coastal environment, p. 537-545. Proceedings of ISCZC 1988, Chino Ocean Press, Beijing.

Pillay, T.V.R. 1992. Aquaculture and the environment. Fishing News Books, England. 184 p.

Seymour, E.A. and A. Bergheim. 1991. Towards a reduction of pollution from intensive aquaculture with reference to the farming of salmonids in Norway. Aquacult. Eng. 10:73-88.

Solbe, J.F. de L.G. 1982. Fish farm effluents: a United Kingdom survey, p. 29-55. *In* Report of the EIFAC Workshop on fish farm effluents. EIFAC Tech. Pap. 41. Silkeborg, Denmark.

OCE. 1988. State of the Environment Report: Victoria's inland waters. Office of the Commissioner of the Environment, Melbourne. 655 p.

Storebakken, T. and E. Austreng. 1987a. Ration level of salmonids. I. Growth, survival, body composition and feed conversion in Atlantic salmon fry and fingerlings. Aquaculture 60:189-206.

Storebakken, T. and E. Austreng. 1987b. Ration level for salmonids. II. Growth, feed intake, protein digestibility, body composition and feed conversion in Rainbow trout weighing 0.5-1.0 kg. Aquaculture 60:207-221.

Sumari, O. 1982. A report on fish farm effluents in Finland. Report of the EIFAC Workshop on Fish Farm Effluents. FAO/EIFAC Tech. Pap. 41:21-27.

Tacon, A.G.J. and S.S. De Silva. 1983. Mineral composition of some commercial fish feeds available in Europe. Aquaculture 31:11-20.

Tarazona, J.V., J.A. Ortiz, M. Carballo and M.J. Munoz. 1993. Pollution generated by fish farms: A system dynamics model. Freseniuns Environ. Bull. 2:84-89.

Warren-Hansen, I. 1982. Evaluation of matter discharged from trout farming in Denmark. Report of the EIFAC Workshop on Fish Farm Effluents. FAO/EIFAC Tech. Pap. 41:57-63.

Yone, Y. and T. Toshima. 1979. The utilisation of phosphorus in fish meal by carp and black sea bream. Bull. Jap. Soc. Sci. Fish. 45:753-756.

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Australian Native Species in Aquaculture

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Abstract
Australian native freshwater fish and crayfish possess good food, recreational and commercial values. Although aquaculture is an infant industry, expanding rapidly due to recent local and overseas (Asian)demands for native species. This article gives a glimpse of the status of native freshwater aquaculture in Australia. (*The Victorian Naturalist* 113, 1996, 264-267)

Introduction

Australian native fish and crayfish form the main freshwater aquaculture industry of the country. Australia has few freshwater fish (180-190 species) (Merrick and Schmida 1984), most of which are native to Australia. Among them only four native fish possess potential for aquaculture, Silver Perch *Bidyanus bidyanus*) Golden Perch *Macquaria ambigua*, Murray Cod *Maccullochella peeli* and Freshwater Catfish *Tandanus tandanus* (Hume and Barlow 1993)

Silver Perch

Silver Perch farming is booming in the country and there is an interest in cultivating the species in countries like China, and Taiwan. The farms are spread over the warmer parts of New South Wales, Queensland and Victoria. New South Wales has the highest number of Silver Perch farms followed by Victoria and Queensland. Factors contributing to the expansion of the Silver Perch industry include good growth rates, their acceptance of low-protein diets (Barlow 1986; Rowland and Barlow 1991), the ease of culture in earthen ponds (Rowland *et al.* 1994) and their omnivorous feeding habits (Rowland and Barlow 1991). The demand for Silver Perch farming is so great that at this stage three commercial feed companies (Kinta, Janos, Barstock) are manufacturing Silver Perch feeds. Further stimulus for Silver Perch arose with the huge recent shipment of fry and fingerlings to China. It is believed that the Chinese are interested in rearing Silver Perch in their traditional ponds since it is an ideal species for Chinese pond polyculture systems. At the recent international conference held in Beijing (Fourth Asian Fisheries Forum, 16-20 October 1995), a number of enquires were made regarding Silver Perch (Fig. 1).

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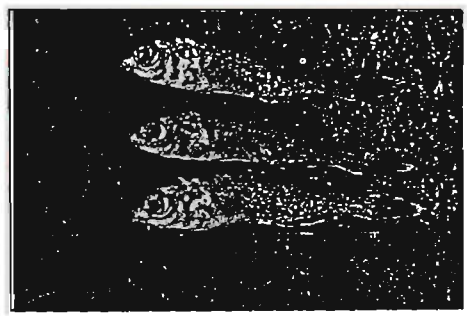

Fig. 1 Silver Perch fingerlings are in great demand both in Australia and in overseas (e.g. China) to stock in ponds and dams.

Above all, freshwater fish production is increasing in Australia mainly due to an increasing interest and investment in growing Silver Perch (Table 1).

Table 1. Native Aquaculture Production (value $000).
Source : O'Sullivan(1994)

Year	Native Fish	Native Crayfish
1989-90	2,888	1,599
1990-91	2,913	2,339
1991-92	4,355	2,235

Golden Perch

Golden Perch farming has not been so popular although it is more attractive to consumers than Silver Perch. Trials are being conducted by the government research institutes to develop Golden Perch diets (Arumugam and Geddes 1987). Once commercial Golden Perch feeds become available in the market, then Golden Perch farming would become a popular aquacul-

ture industry since consumer demand for Golden Perch is high and can therefore fetch higher market prices. Some private companies are conducting research to develop Golden Perch feed from sewage-grown zooplankton (Zootech News 1994).

Freshwater Crayfish

Australia has the most diverse collection of freshwater crayfish in the southern hemisphere, three of which used in aquaculture. These are, Yabby *Cherax destructor*, Marron *Cherax tenuimanus* and Redclaw *Cherax quadricarinatus*. Yabby farms are located in South Australia, Victoria and New South Wales (Kailola *et al.* 1993). Marron have been commercially cultured in Western Australia for the last 20 years (Kailola *et al.* 1993). Redclaw requires more tropical conditions and is cultured mainly in Queensland. The natural distribution of freshwater species can be seen in Fig. 2. There is a big domestic market for crayfish but both live and frozen crayfish are also being exported to nearby Asian countries. A summary of biological information on native Aquaculture species is given in Table 2.

Conclusion

The demands for freshwater and marine foods in Australia are increasing as a result of population increase, Asian migration and health consciousness. It is predicted that Australian native aquaculture industry would become a lucrative primary industry in food production (Gooley and Rowland 1993).

However, it should be noted that effluents from aquaculture industry may cause water pollution as nutrients discharged may cause eutrophications to water bodies (Foy and Rosell 1991; Ketola *et al.* 1991). An increase in water turbidity and oxygen demand in natural systems may come from the solid wastes of aquaculture. Therefore it is essential to monitor the level of nutrient discharged from aquaculture industry to the natural system in order to prevent any environmental disasters.

Since natural populations of Australian native freshwater species are either threatened or in decline due to physical, chemical and biological reasons (Cadwallader 1978; Scott 1989), the demands of Australian aquaculture for large numbers of fingerlings, to supply the overseas market and to stock dams and ponds in Australia, could also offer the opportunity to restock natural Australian freshwater systems and reduce the necessity of fishing them. Thus, this programme would also offer the opportunity to help reverse the decline in natural populations.

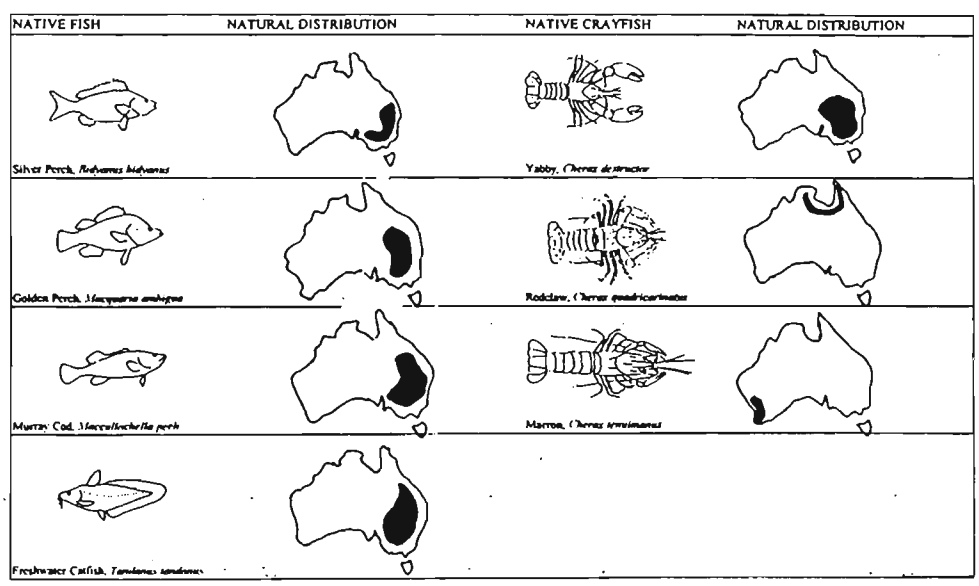

Fig. 2. Natural distribution of native Australian species under aquaculture

Table 2. Summary of biological information on native species.
Legend: C=Celsius; CL=Carapace length; F.W= Freshwater; WT=Water temperature; T= temperature
A= Breeding season; B= Age and size at maturity;CC= Breeding stimulus; D= Temperature tolerance;
E= Optimum growth temperature; F= Fecundity; G= Feeding habits
Figures in parenthesis denotes source
Sources: 1.Lake (1967a); 2. Merrick (1980); 3.Lake(1967b); 4. Rowland(1992a);5.Whiteley(1960);6.Backhouse et al.(1991a); 7.Lake(1967c);8. Cadwallader(1977); 9.Lake(1967d);10. Backhouse et al,(1991b);11.Kailola et al.(1993); 12.Llewellyn and Macdonald(1980); 13.Cadwallader(1978); 14.Rowland(1988); 15. Mosig(1982); 16. Rowland(1992b); 17. Llewellyn and Pollard(1980); 18.Davis(1977); 19.Macleans(1975); 20.Johnson(1988); 21.Merrick and Lambert(1990); 22.O'Sullivan((1992); 23.Mitchell and collins(1989); 24.Jones(1990).

SPECIES	A	B	CC	D	E
Silver Perch	Oct-Dec(1)	M-2 yrs;F-3 yrs(2)	T >23.3 C + increase in water level(3	2-37 C(4)	23-28 C(4)
Golden Perch	Oct.-Mar(8)	2-3 years(3),	T 23.5 C 4- & flooding(9,1)	38 C(4)	23-28 C(4)
Murray Cod	Oct.-Dec(12)	4 years(13,1)	T > 21 C , water level rise not essential (3,14)	2-33 C(15)	20 C(4)
F.W Catfish	Oct.-Dec(1)	few at 2 years most by 5 years(17)	T 24 C (18)	1-38 C(15)	19-25(1)
Yabby	Oct.-Mar(11)	0.2-0.3 year (30-50mmCL)(11)	T>15 C (20) increase in daylength	1-38 C(21) 28 C (23)	20-23 C (22)
Redclaw	All year(11)	1.0 year(11)	WT above 20 C & increment in day length(11)	5-42 C(24)	24-30 C(23)
Marron	Sep.-Oct(11)	1-3 years, (25-30mmCL)(11)	rise in water temp.(11)	5-32 C(21)	24-30 C(22), 24 C(11)

F	G
Silver Perch 50,000 (1.8-2.0 kg)(5)	omnivore-consists of zooplankton(ostracods),shrimps,small aquatic insects,molluscs,earth worms & plant material(6,7);larvae feed on both phyto & zooplankton(7)
Golden Perch 500,000(2.2-2.4kg)(3)	carnivore-mainly crustaceans(yabbies) ,aquatic insects,molluscs and fish(10). Young golden feed on zooplankton on recently inundated floodplains(11)
Murray Cod 30,000-50,000 for 1-2kg fish(16)	carnivore-adults feed on crustaceans,molluscs,fish, occasionally amphibians and reptiles(11,14), larval feed consists of copepods,cladocerans(16),
F.W Catfish 18,000(1.25kg)-26,000(2.27kg(3)	adults are omnivorous, young eat zooplankton & worms(19),adults carnivores & bottom feeders feed on molluscs & crustaceans(1)
Yabby 1000(large female)(11)	juvenile - filter feeder, adult feeds on detritus,plant material and small invertebrates(23)
Redclaw 300-1000(24)	larvae diet includes zooplankton & detritus and adult diet mainly detritus(24)
Marron 40-2400(21)	opportunistic scavenger-detritus,plant and animal material & aquatic insects(11)

References

Arumugam, P.T. and Geddes, M.C. (1987). Feeding and growth of Golden Perch larvae and fry *Macquaria ambigua* Richardson. *Transactions of the Royal Society of South Australia* **111**, 59-65.

Backhouse, G., Cadwallader, P. and Raadik,T. (1991a). 'Freshwater fish of Victoria - Silver Perch'. *Department of Conservation and Environment, Victoria. Infosheet-69*

Backhouse, G., Cadwallader, P. and Raadik, T. (1991b). Freshwater fish of Victoria - Golden Perch. *Department of Conservation and Environment, Victoria. Infosheet-51*

Barlow, C.G. (1986). Fish in farm dams and implications for extensive aquaculture. pages. 33-34. *In* 'Proceedings of the First Freshwater Aquaculture Workshop'. Ed. L.F.Reynolds. (Narrendera, NSW Agriculture and Fisheries: Sydney, Australia).

Cadwallader, P.L. (1978). Some causes of the decline in range and abundance of native fish in the Murray-Darling River system. *Proceeding Royal Society of Victoria* **90**, 211-224.

Cadwallader, P.L. (1977). I.O.Langrey's 1949-50, Murray River Investigations. *Fish.Wild.Pap. Victoria*. No. 13.

Davis, T.L.O. (1977). Reproductive biology of the Freshwater Catfish, *Tandanus tandanus* Mitchell, in the Gwyd River, Australia, and effects associated with impoundment of this river by the Copeton Dam. *Australian Journal of Marine and Freshwater Research* **28**, 455-485.

Foy, R.H. and Rosell, R. (1991). Fractionation of phosphorus and nitrogen loadings from a Northern Ireland fish farm. *Aquaculture* **96**, 31-42.

Gooley, G. and Rowland, S. (1993). Murray-Darling Finfish: Current developments and commercial

potential. *Austasia Aquaculture* 7(3), 35-36.
Hume, D. and Barlow, C. (1993). 'Bibliography of four Australian native fish that have potential for aquaculture: Freshwater Catfish, Golden Perch, Murray Cod and Silver Perch'. (Department of Primary Industry: Queensland Government).
Johnston, H.T. (1988). Crayfish farming. *In* 'Crayfish Farming'. Ed G. Ryder. (NSW, Department of Agriculture and Fisheries: Sydney).
Jones, C.M. (1990). 'The biology and aquaculture potential of the tropical freshwater crayfish, *Cherax quadricarinatus*'. (Queensland Government).
Kailola, P.J., Williams, M.J. and Stewart ,P.C., Reichelt, R.E., McNee, A. and Grieve, C. (1993). 'Australian Fisheries Resources'. (Commonwealth of Australia, Bureau of Resource Sciences and the Fisheries Research development Corporation).
Ketola, H.G., Westers, H., Houghton W., and Pecor, C. 1991. Effect of diet on growth and survival of Coho Salmon and on phosphorus discharges from a fish hatchery. *American Fisheries Society Symposium* **10**, 402-409.
Lake, J.S. (1967a). Freshwater fishes of the Murray-Darling River system. *In* 'Australian inland waters and their fauna'. Ed A.H.Weatherley. (Australian National University Press:Canberra).
Lake ,J.S. (1967b). Rearing experiments with five species of Australian freshwater fishes. I. Inducement to spawning. *Australian Journal of Marine and Freshwater Research* **18**, 137-153.
Lake, J.S. (1967c). Rearing experiments with five species of Australian freshwater fishes. II. Morphogenesis and Ontogeny. *Australian Journal of Marine and Freshwater Research* **18**, 155-173.
Lake, J.S. (1967d). Freshwater fishes of the Murray-Darling River system. *Chief Secretary's Department, NSW State Fisheries Research Bulletin* 7.
Llewellyn, L.C. and Macdonald,M.C. (1980). Family Percichthyidae. Australian Freshwater Basses and Cods. *In* 'Freshwater Fishes of South-Eastern Australia'. Ed R.M. McDowall. (Reed:Sydney).
Llewellyn, L.C. and Pollard,D.A. (1980). Family Plotosidae. Elletailed Catfishes. *In* 'Freshwater Fishes of South-Eastern Australia'. Ed R.M. McDowall. (Reed:Sydney).
Macleans, J. (1975). The potential for aquaculture in Australia. *Australia Fisheries Paper No. 21.* (Australian Fisheries Service: Canberra).
Merrick, J.R. (1980). Family Teraponidae. Freshwater Grunters of Perches. *In* 'Freshwater Fishes of South-Eastern Australia'. Ed R.M. McDowall. (Reed:Sydney).
Merrick, J.R. and Lambert, C.N. 1990. 'The yabby, marron and redclaw - production and marketing'. (John R. Merrick Publications, NSW: Australia).
Merrick, J.R. and Schmida, G.E. (1984). 'Australian freshwater fishes - biology and management'. (Griffin Press Ltd.: South Australia).
Mitchell ,B.D. and Collins, R. 1989. Developments of field-scale intensive culture techniques for the commercial production of the yabby (*Cherax destructor*). Centre for Aquatic Sciences, Warrnmbool Institute of Advanced Education, Unpublished report.
Mosig, J. 1982. Native fish for farm dams. *Trees and Victoria's resources* **24**, 3-4.
O'Sullivan, D. (1994). Production and value continue to climb. *Austasia Aquaculture* 8 (4), 2.
O'Sullivan, D. (1992). Freshwater crayfish farms - site selection. *Austasia Aquaculture*, 6 (5), 16-18.
Rowland, S. (1988). 'Murray Cod'. (NSW Agriculture and Fisheries. Agfacts F3.2.4).
Rowland, S. (1992a). 'Water quality in freshwater aquaculture'. (Fish Facts: NSW Fisheries).
Rowland, S. (1992b). Diet and feeding of Murray Cod (*Maccullochella peeli*) larvae. *Proceedings of the Linnean Society of NSW* **113**(3), 193-201.
Rowland, S., Allan, G., Hollis ,M. and Pontifex, T. (1994). Production of the Australian freshwater Silver Perch, *Bidyanus bidyanus* (Mitchell), at two densities in earthen ponds. *Aquaculture* 30, 317-328.
Rowland ,S. and Barlow ,C.G. (1991). Fish biology-the right prerequisite, a case study with freshwater Silver Perch(*Bidyanus bidyanus*). *Austasia Aquaculture* 5 (5), 27-30.
Rowland ,S. and Kearnery, R.E. (1992). The prospects for the commercial Aquaculture of Silver Perch, *Bidyanus bidyanus*. A Research Project arising from a review of
freshwater aquaculture, NSW Fisheries, Sydney.
Scott ,D. (1989). 'State of the Environment report 1988 - Victoria's Inland Waters'. (Office of the Commissoner for the Environment).
Whitely ,G.P. (1960). 'Native Freshwater Fishes of Australia'. (Jacaranda Press: Brisbane).
Zootech News. 1994. 'Zootech Newsletter 1 (May)'. (Melbourne: Australia).

The Fauna of Tasmania: Mammals

by R.H. Green

Publisher: *Potoroo Publishing, Launceston, [1994].*
Paperback, 15 x 22 cm, viii + 56 pp. and 64 coloured plates. RRP $14.95

This attractively presented little book is printed on good quality paper and its cover features a colour photograph of a Tasmanian endemic, the Long-tailed Mouse, *Pseudomys higginsi*. The Introduction confirms what the title suggests, that this is the first of a series; the second is on birds, and subsequent volumes on reptiles, frogs and freshwater fishes are intended. The contents are tallied most succinctly on the rear cover; '2 monotremes, 20 marsupials, 38 eutherians, including 13 marine mammals [of about 32 known from local waters], and 10 introduced mammals. Giving information on evolution, relationship[s], identification, distribution, habitat, abundance, food, behaviour and breeding. Illustrated with 64 photographs [mainly by the author].'

Introductory sections entitled 'A special

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Pollution from aquaculture

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AQUACULTURE IS an important aquatic food production industry which is becoming popular in Australia as a result of increasing demand for fresh and healthy foods. The bulk of current aquaculture production comes from salmonids, prawn, native fish and freshwater crayfish. To achieve rapid growth and a profitable production, formulated artificial food is used in the rearing of fish. During aquaculture operations wastes are generated in the form of solids and soluble products that may cause water pollution to the receiving waters. These wastes are uneaten food, faeces, scales, mucus (solid wastes) and dissolved nutrients (soluble waste) (Figs 1 & 2) may result from the feeding and metabolic processes of the culture organisms. The two most important nutrients incorporated in fish food are the nitrogen and phosphorus essential for biological growth of fish but an abundance of these nutrients could cause algal blooms or eutrophication in natural waters. Phosphorus is the key nutrient for the growth of algae in freshwater, and nitrogen as ammonia is toxic to fish and is also an important nutrient required by algae.[1] It is therefore imperative that the digestive and metabolic wastes are reduced to a minimum so that aquaculture can be an environmentally sustainable development program. Since feed cost accounts for about 60% of aquaculture production, it is also economically desirable that feed loss be minimised. This article discusses the factors associated with pollution from aquaculture and possible impacts of aquaculture wastes on our environment.

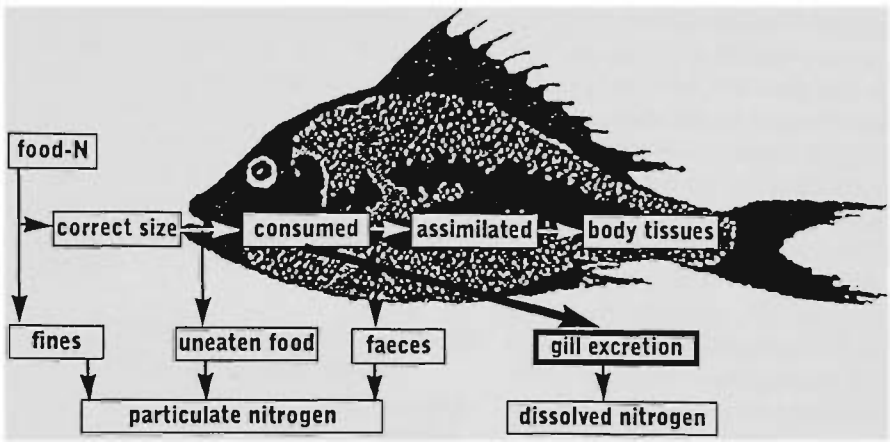

Figure 2. General flow chart of origin of nitrogen in aquaculture from feeding.

Results and discussion

Research conducted in our laboratory found that temperature and food conversion (kg food fed/kg fish produced) could affect the amount of loading from aquaculture. The native fish silver perch *Bidyanus bidyanyus* when reared in different temperatures produced less solid and nutrient load at 25°C compared to other temperatures in the order of 25°C <30°C < 20°C. The better growth rate and good food conversion ratio (FCR) at 25°C may be related to the comparatively low pollution load achieved at this temperature. Furthermore, a good nitrogen and phosphorus retention in the carcass may have attributed to the lower nitrogen and phosphorus pollution at 25°C. Nonetheless, the average nitrogen and phosphorus pollution was found to be more than 50% of food fed to fish at 25°C. The main path of phosphorus and nitrogen loss was found to be via faecal pellets and gill excretion respectively (Figs 1 & 2). The daily ammonia and orthophosphate discharges increased at the higher temperature and were affected by the type of diet. The nutrient release from solid waste was observed to be dependent upon pH of the environment, for example the release of phosphorus was accelerated in acidic media whereas ammonium in neutral-to-alkaline media (Figs 3 & 4). The fish food having the highest concentration of phosphorus generated faeces with most labile phosphorus as characterised by the distilled water and ammonium chloride extractable fractions (Table 1).

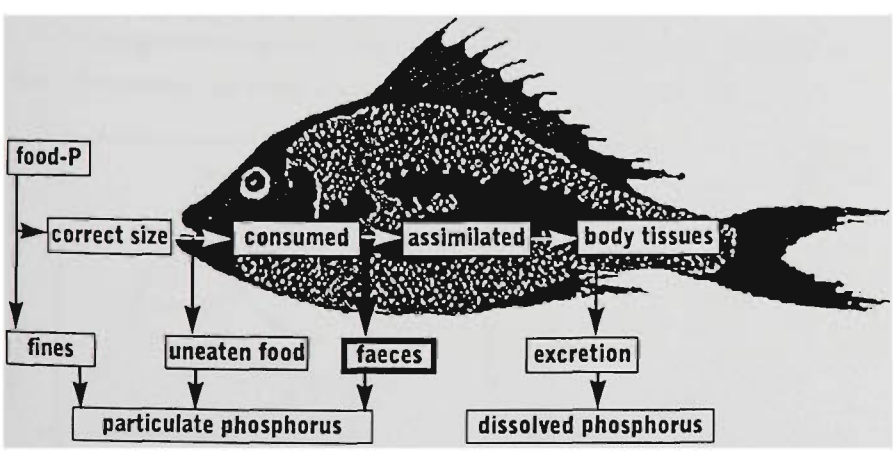

Figure 1. General flow chart of origin of phosphorus in aquaculture from feeding.

Impact of solid wastes

The solids produced per tonne of fish production is reported to be in the range of 300–1000 kg.[2] We have estimated a figure of 308 kg/tonne from silver perch production based on our results at 25°C, and at 20°C it was almost doubled, apparently due to poor growth. The accumulation of solid wastes could lead to the formation of hydrogen sulfide in the nearby environment which is highly toxic to fish, in addition the organic waste may cause a high demand for oxygen[1] and increase in bacterial populations.[3] Suspended solids could affect light penetration and increase in siltation in river/lake beds, lower dissolved oxygen levels and produce hydrogen sulfide and methane.[4] From our experiments and others, it appears that the quantity of solids and dissolved organics produced from aquaculture is directly related to the quantity of food absorbed. Poor quality of ingredients added to feed may result in decreased availability of nutrients and increase in faecal losses.[5]

Impact of nutrient

The phosphorus loss to the environment has been estimated to be 66–70% of feed fed[6] whereas nitrogen loss through gill excretion and faecal output could be 78%.[7] At the best growth rate the phosphorus lost in our experiments ranged from 63–80% and nitrogen lost varied between 50% to 63%. An increase in nutrient load could stimulate algal blooms or eutrophication in water bodies receiving fish farm effluents, resulting in the fluctuation of oxygen and pH levels, and may put a great strain on water-treatment facilities.[8]

Relationships between FCR and nutrient loss

The nutrient loss rates were reported to be dependent on FCR and the nutrient content of diet.[9] At 25°C, the better FCR obtained in our experiments may have caused less pollution load. Feed is the main source of pollution in aquaculture and the FCR determines the level of pollution, for example an increase of FCR from 1.0 to 1.5 is reported to have increased the phosphorus and nitrogen pollution to about 86% and 70% respectively.[10]

Fractionational composition of nutrient and algal growth

Our experiment demonstrated that both fish food and faeces may contain a significant proportion of labile phosphorus (Table 1). Since the algal growth is related to the proportion of labile phosphorus in the input,[9] it is therefore of paramount importance to reduce the total phosphorus as well as the labile phosphorus content in fish foods[11] in order to minimise the phosphorus pollution from aquaculture.

Conclusion

Our research shows that growing of fish at optimum temperature may reduce the solid and nutrient load to the environment. A good food conversion ratio (FCR) could reduce the pollution load which is also vital for economical production. In order to reduce the environmental impacts of aquaculture, development of low-polluting diets is of utmost important. The simple alternative would be to feed fish according to demand or raising of fish during optimum growing temperature or seasons. However, further research on the optimal nitrogen and phosphorus requirements of fish that will maximise the fish growth but minimise the pollution load to environment is essential. Experiments are also needed on the labile fraction of nitrogen and phosphorus in commercial aquaculture diets and factors related to the release of nutrient from the sediments containing the aquaculture wastes. All of this information is vital for environmental management strategy planning and the development of diets of low pollution potential.

Acknowledgments

We are grateful to the Department of Environmental Management, Victoria University of Technology for providing all the facilities required to conduct this extensive research.

Extraction methods	Diet 1	Diet 2	Faeces 1	Faeces 2
Distilled water	2.57	1.87	1.03	0.94
Ammonium chloride	1.68	1.44	1.46	1.0
Sodium hydroxide	2.33	1.68	1.1	0.68
Hydrochloric acid	2.25	1.5	1.0	0.71

Note: Phosphorus content in diet 1 and diet 2 are 1.3% and 1.16% respectively;
Faeces 1 and faeces 2 are from the diet 1 and diet 2 fed to fish;
The fraction extracted by distilled water and ammonium chloride is readily available to plants

Table 1. Fractionational composition of phosphorus in fish food and faeces(mg P/g dry weight)

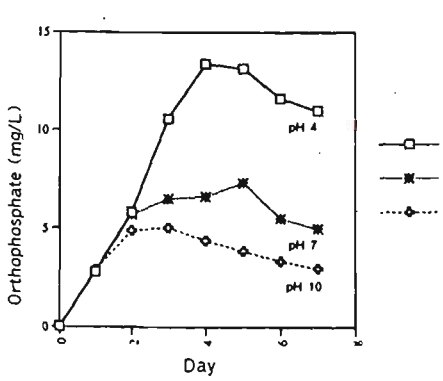

Figure 3. The release of phosphate from fish food at different pH. The food was incubated at 25°C for seven days

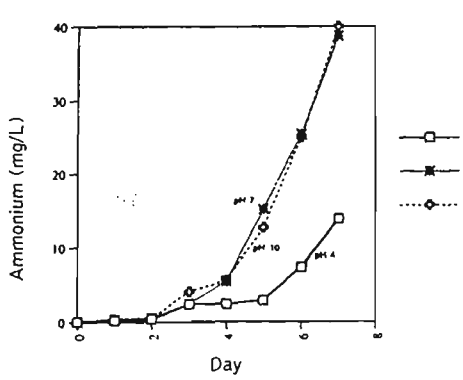

Figure 4. The release of ammonium from fish food at different pH. The food was incubated at 25°C for seven days.

References

1. M. Phillips and M. Beveridge, *Fish Farmer* 1986, 5(6), 17–19.
2. M.C.M. Beveridge., M.J.Phillips and R.M.Clarke, "A quantitative and qualitative assessment of wastes from aquatic animal production", in *Advances in world aquaculture*, (eds D.E.Brune and J.R.Tomasso), (World Aquaculture Society: Baton Rouge, 1991, 3, 506–533).
3. C.Folke and N. Kautsky, *Ambio.*, 1989, 18, 234–243.
4. G. Mather, G. *Aquaculture – An Australian environmental review*. Total Environment Centre Inc. Sydney. 1993, p. 88.
5. S.J. Kaushik, *Conference Internazionale ' Produzione di almenti per animalia*, Piacenza, Italic, 25-26 Fev.,1992, 1–2.
6. H.G. Ketola, *Salmonia*, 1982, 6(2), 12–15.
7. R.J. Gowen and N.B. Bradbury, "The ecological impact of salmonid farming in coastal waters: a review", in *Oceanography and marine biology: an annual review* (ed M. Barnes), 1987.
8. L.M. Laird and T. Needham, *Salmon and trout farming*, (Ellis Horwood Limited: England, 1988, p. 271).
9. R.H. Foy and R. Rosell, *Aquaculture*, 1991, 96, 17–23.
10. T. Storebakken and E. Austreng, *Aquaculture*, 1987, 60, 207–221.
11. K. Pettersson, *Verh. Internat. Verein. Limnol.*, 1988, 23, 220.

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Asian Fisheries Society, Manila, Philippines

Preliminary Rearing Trials of an Australian Native Fish, Silver Perch *(Bidyanus bidyanus)* (Mitchell) with Reference to Growth and Production of Solid Waste in Aquaculture[1]

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Abstract

The native silver perch *Bidyanus bidyanus* has good potential as an aquaculture species in Australia. Juveniles of this species (1.14-2.13 g) were reared in glass aquaria at room temperatures (18-20°C) with aeration. Fish were kept individually in separate aquaria for the study of growth and solid waste production. In the first experiment, three commercial diets referred to as diet 1, diet 2 and diet 3 were offered to fish for four weeks in order to study the gain in weight and food conversion ratio. The gain in weight decreased in the order of fish fed diet 2 > diet 1 > diet 3 ($P>0.05$). In the second experiment, diets 2 and 1 were fed to fish for four weeks in order to study the relationship between growth of fish and production of solid wastes (suspended and dissolved) in the culture system. Diet 2 resulted in slightly better gain in weight ($P>0.05$) and less solids production in comparison to diet 1.

Introduction

The silver perch (*Bidyanus bidyanus*) is an Australian native fish of high aquaculture potential (Rowland et al. 1995). It is one of four principal species of the Murray-Darling River system and is much sought after by commercial and recreational fishers (Cadwallader 1979). Although aquaculture is an infant industry in Australia, interest in the culturing of silver perch is growing both in Australia and in nearby Asia (Gooley and Rowland 1993).

Previous research on *B. bidyanus* has concentrated on its biology (Rowland and Barlow 1991; Allan and Rowland 1992; Rowland and Allan 1994; Rowland et al. 1995) and nutrition (Allan and Rowland 1992; Allan et al. 1994; Allan and Rowland 1995). Until now there have been no attempts to study the

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amount or types of waste entering the environment from the rearing of silver perch or other species in Australia. This information is required since research in Europe and North America demonstrates a strong relationship between water pollution and the discharge of fish farm effluents (Albaster 1982; Ketola 1982; Penczak et al. 1982; Solbe 1982; Bregheim et al. 1984; Phillips 1985; Carr and Goulder 1990; Beveridge et al. 1991; Cowey and Cho 1991; Foy and Rosell 1991a, 1991b; Persson 1991; Pillay 1992; Ketola and Richmond 1994). This is the first in a series of planned experiments to study waste production from rearing of silver perch. These preliminary trials were designed to study growth and waste production at 18-20°C (room temperature) since previous research has demonstrated that silver perch can grow at temperatures as low at 12°C (Barlow and Bock 1981; Rowland 1995). Based on these results, further studies will be conducted on other forms of pollution from aquaculture of silver perch. The objectives of these preliminary rearing trials were:

- To evaluate growth performances (gain in weight and feed conversion) of silver perch fed on three locally available commercial diets;
- To compare the amount of solid waste (uneaten food and feces) load from the feeding of silver perch with reference to growth achieved.

Materials and Methods

The experiments were conducted in the wet laboratory at the Victoria University of Technology. The experimental fish (1,140-2,130 mg) were purchased from a local native fish farm where fish were grown in earthen ponds. They were acclimatized in the laboratory in large holding tanks (46 x 30 x 30 cm) for four weeks before use in experiments. All experiments were conducted at room temperatures of 18-20°C. Domestic tap water was used after dechlorination with an appropriate conditioner (Sera Aquatan). pH was monitored using a pH meter (Orion SA 520), and water temperature and fish health were monitored routinely. The pH of water was maintained at 7.5-8.5 using sodium bicarbonate or sodium bi-phosphate to adjust the pH. Fish were fed silver crumbles/starter (appropriate for fingerlings) at the rate of 3% body weight as recommended. They were fed twice per day (0900 and 1600), 6 d per week. Sampling was conducted once a week to measure gain in weight and to adjust feed amount. The experimental details are given in Table 1. Proximate analysis was done following AOAC (1990) and Chiu (1989). Protein was analyzed by Kjeldahl method (N x 6.25), fat by ether extraction (New 1987), moisture by drying in an oven overnight at 105°C, ashing by burning samples in a muffle furnace at 600°C overnight, and carbohydrate by difference in weight. Gross energy of diets was calculated using kilocaloric values of $5.5 \cdot g^{-1}$ protein, $9.1 \cdot g^{-1}$ lipid and $4.1 \cdot g^{-1}$ carbohydrate (New 1987).

Experiment 1: Effects of Feed on Growth and Feed Conversion

The experiment ran for four weeks and tested three commercial diets, referred to as diet 1, diet 2 and diet 3 with protein content of 53, 45 and 36%, respectively. Table 2 shows the proximate composition of the diets. Fish (1.14-1.54 g) were kept individually in separate aquaria (30 x 16 x 17 cm) to study individual growth performance with respect to gain in weight, food conversion ratio (FCR) and specific growth rate (SGR). Unfed fish (control) were kept in separate aquaria to compare growth performance of fed fish. A biological filter was used in each tank to enhance the culture environment.

Table 1. Experimental details of feeding trials 1 and 2 conducted at room temperatures (18-20°C) with silver perch juveniles. Trial 1 tested three diets, while trial 2 tested two diets. Both experiments ran for 28 d.

Trial no.	Temperature (°C)	Tank size (cm)	Diet no.	No. tanks/diet	No. fish/tank	No. fish/treatment	Average initial size (mg)
1	18-20	30 x 16 x 17	1	10	1	10	1,540
1	18-20	30 x 16 x 17	2	10	1	10	1,140
1	18-20	30 x 16 x 17	3	10	1	10	1,440
2	18-20	30 x 16 x 17	1	10	1	10	2,000
2	18-20	30 x 16 x 17	2	10	1	10	2,130

Table 2. Proximate composition and other information of the three diets fed to silver perch juveniles in trials 1 and 2. Trial 1 used all three diets while trial 2 used diets 1 and 2 only.

	Diet 1	Diet 2	Diet 3
Protein	53.00	45.00	35.89
Fat (%)	06.92	08.50	4.96
Carbohydrate (%)	27.02	36.66	46.15
Ash (%)	13.06	09.84	13.00
Nitrogen (%)	08.48	07.20	05.74
Phosphorus (%)	01.31	01.16	01.28
Fiber (%)	01.42	03.00	02.00
Dry matter (%)	91.80	90.12	88.99
Moisture (%)	08.20	09.88	11.01
Digestible energy (Kcal·kg^{-1})	5,027	5,273	4,851
DE/P (Kcal·kg^{-1})	95	117	135

Experiment 2 : Effects of Feeding on Suspended and Dissolved Solids Production

Here, as above, individual fish were kept in separate aquaria but fed diets 1 and 2 which sustained the best gain in weight in experiment 1. This experiment was designed to study the amount of suspended and dissolved solids produced over a 4-week growth period. No biological filter was used in the aquaria, however aeration was provided and dissolved oxygen level was more than 6 $mg \cdot l^{-1}$. One-third of the water was exchanged daily. Each aquarium was siphoned once every morning to collect uneaten food and feces (solid waste) using a 5-mm hose. Collected solids were filtered first through a fast filter (Whatman 512) and later through a standard 0.45-μm glass fiber filter (APHA 1989). The residue retained on both filters was dried in an oven at 105°C for 1 h, cooled in a desiccator and weighed using an analytical balance. The increase in weight was considered as total suspended solids and was determined as suspended solids/fish. The filtrate from the total suspended solids was used for total dissolved solids determination following APHA (1989).

The following formulae were used:

Weight gain (g) :Final weight of fish - Initial weight of fish (Chiu 1989)

Feed conversion ratio (FCR) : $\frac{\text{Weight of feed fed}}{\text{Weight gained by the fish}}$ (Laird and Needham 1988: Chiu 1989)

Specific growth rate (SGR) : $\frac{\ln w_t - \ln w_o \times 100}{d}$ (Laird and Needham 1988; Chiu 1989)

where, ln = natural logarithm; wt = average final weight; w_o = average initial weight; d = total days of experiment;

Protein efficiency ratio (PER) :Wet fish weight gain/Dietary protein intake (EIFAC 1980)

Total dissolved solids (mg/l) : $\frac{(A - B) \times 1{,}000}{\text{sample volume, ml}}$ (APHA 1989)
where A = weight of dry residue + dish,(mg);
B = weight of dish, mg

Results were analyzed by one-way analysis of variance (ANOVA) or a 2-sample student t-test using an IBM-compatible MS Excel program. The level of significance was set at 0.05 for all statistical tests performed.

Results

Of the three diets, diet 2 resulted in the best growth rate (Fig. 1.). However, statistical analysis did not reveal any significant difference ($P>0.05$) in weight gain among the fish fed the three different diets. Better FCR and SGR were achieved in diet 2 ($P<0.05$) than in the other diets (Table 3.). Diet 3 had the poorest FCR and SGR, and was not used in trial 2. The survival rate in experiment 1 was 100%.

Table 3. Weight gain, specific growth rate and food conversion of individual silver perch fed three diets and reared at room temperatures for 28 d in trial 1.

	Diet 1	Diet 2	Diet 3
Average initial weight (mg)	1,540	1,140	1,440
Average final weight (mg)	1,980	1,600	1,830
Gain in weight (mg)	440±19[1]	460±22[1]	390±50[1]
Percentage gain in weight	29±1.40[1]	40±2.2[2]	27±1.70[1]
Specific growth rate (SGR)	1.05±0.07[1]	1.41±0.03[2]	0.99±0.04[1]
Food conversion ratio (FCR)	2.97±0.06[1]	2.24±0.10[2]	3.20±0.04[1]
Protein efficiency ratio (PER)	0.69±0.02[1]	0.99±0.07[2]	0.89±0.06[2]
Survival	100	100	100

[1] Values are mean±SE; n=10.
[2] Values in the same row with common superscripts are not significantly different (P>0.05).

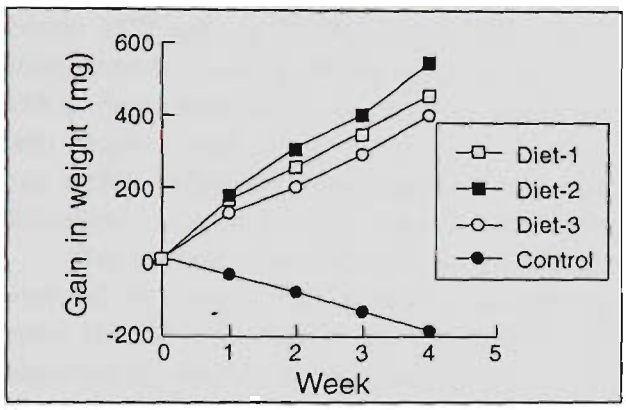

Fig. 1. Gain in weight/fish (mean±SE; n=10) of silver perch juveniles reared for four weeks and fed three diets @ 6 d•week^{-1} at room temperatures (18-20°C). There were no significant differences in weight gain of fish fed three diets (P>0.05). The control group was unfed.

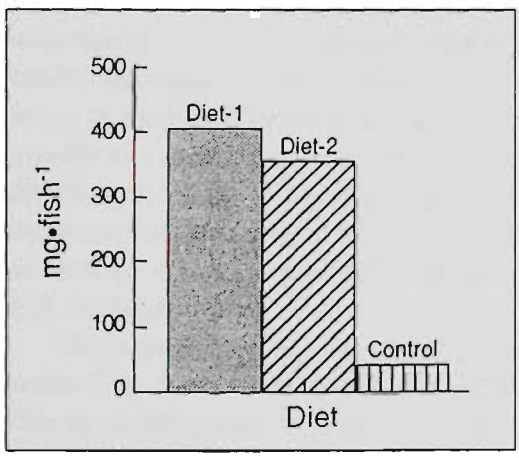

Fig. 2. Mean suspended solids produced by silver perch (2.00-2.13 g) fed two diets in experiment 2 and reared for four weeks at 18-20°C (n=10). The control group was unfed.

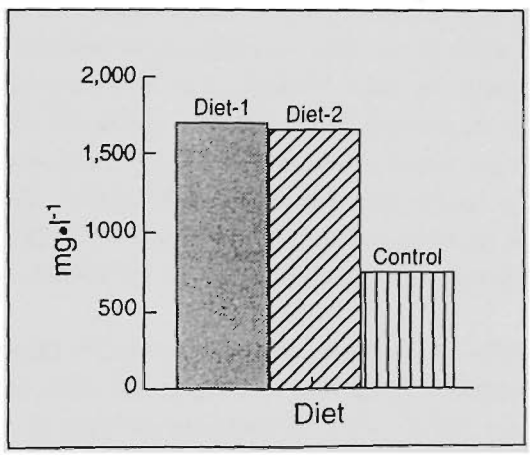

Fig. 3. Mean dissolved solids (±SE) produced by silver perch (2.00-2.13 g) fed two diets in experiment 2 and reared for four weeks at 18-20°C (n=10). The control group was unfed.

In the second experiment with two diets, diet 2 again resulted in slightly better SGR and FCR than diet 1, though the results were not significantly different (P>0.05). Slightly less suspended and dissolved solid waste was produced by fish fed diet 2 (Figs. 2 and 3). Solid waste production from diets 2 and 1 was 350 mg·fish^{-1} and 400 mg·fish^{-1}, respectively, and was not significantly different (P>0.05). The dissolved solids produced was 1,647 mg·l^{-1} with diet 2,

and 1,711 mg·l^{-1} with diet 1. Unfed fish (control) produced the least amount of suspended and dissolved solids and this was significantly different from that produced by fed fish ($P<0.05$) (Figs. 2 and 3).

Discussion

In both experiments, fish fed diet 2 (45% protein) showed better gain in weight compared to diet 1 (53%) or diet 3 (35%) although statistical analysis did not show consistent differences across the two experiments. This may indicate that omnivorous silver perch do not require the higher levels of protein tested in this experiment. Allan and Rowland (1991) found that silver perch gained similar weight with 35.7% and 49% protein fed diets ($P>0.05$) but gained less weight with 20.7% protein ($P<0.05$). In our experiment, FCR was better with a 45% protein diet. However, Allan and Rowland (1991) reported better FCR with a 35.7% protein diet. The optimum protein requirements of silver perch juveniles is reported to be closer to 32-35% (Allan and Rowland 1991). From studies made on other freshwater species, it appears that the protein requirement of omnivorous silver perch could be closer to those of channel catfish (Allan and Rowland 1992) or common carp (Cadwallader 1979) and therefore lower than that used in diets 1 and 2.

The growth rate achieved in our experiments was low compared to commercial situations. This could be due to the effect of lower rearing temperatures (18-20°C) or culture methods. Slow growth of silver perch juveniles was reported by Allan and Rowland (1991) at 18.3-22.8°C. It appears that growth of silver perch in aquaria is much slower, which may indicate that artificial tanks may not be an ideal culture system for achieving maximum growth of silver perch. Rowland (1995) obtained a significantly slower growth rate of silver perch in tanks compared to earthen ponds. Similarly, crayfish have shown poor growth in concrete and plastic-lined ponds whereas earthen ponds were very effective in crayfish rearing (Tredwell et al. 1992). Moreover, the objectives of these preliminary trials was to measure the production of solid wastes in a laboratory culture situation, and was not meant to achieve maximum growth of *B. bidyanus*.

In experiment 2, diet 2 produced less suspended and dissolved solid waste (Fig. 2) in comparison to diet 1. This may be related to the slightly better gain in weight and FCR achieved in diet 2 with experiment 2. Lall (1991) reported that ingredients containing a high concentration of fiber, chitin and undigestible carbohydrate may increase the excretion of suspended solids. Total solid production from cages was estimated to be 290-655 kg dry weight·tonne^{-1} of rainbow trout (Phillips et al. 1990). The typical annual figure for solid discharge as effluents from Danish trout farms is reported to be 550 kg·tonne^{-1} (Warren-Hansen 1982). Though caution must be exercised when extrapolating our laboratory results to field aquaculture situations, the estimated solid waste production (dry-weight basis) in diets 1 and 2 were 600 and 551 kg·tonne^{-1} fish produced, respectively ($P>0.05$). The estimated solid waste production was based on rearing of silver perch at 18-20°C and feeding at the rate

of 3% body weight. It is reported that even in balanced feed, 15-20% of eaten food is undigestible (Asgard et al. 1986). The present experiment was carried out at lower temperatures (18-20°C.), and at this temperature fish must be less active resulting in poor food intake, and consequently produced more solid waste. Rearing silver perch at a higher growth temperature could be a means of reducing solid waste discharge into the environment.

Minimization of solid discharge from fish farms is important because solids increase the turbidity in receiving waters. Waters high in suspended solids are unsatisfactory for bathing, and waters with high dissolved solids are of inferior palatability and may induce an unfavorable physiological reaction in the transient consumer (APHA 1989). It is therefore essential that further research be conducted on solids production at the higher temperature optimum for growth of silver perch which is probably in the range of 23-28°C (Rowland et al. 1995). Investigations are also necessary on the chemistry of solid wastes produced, since research in other countries (Albaster 1982; Persson 1988; Beveridge et al. 1991; Foy and Rosell 1991a, 1991b) demonstrates a direct relationship between phosphorus and nitrogen levels in solid wastes from aquaculture and algal blooms or eutrophication in lakes or rivers. Our further experiments will be a more detailed study on solid waste, separating uneaten food from feces, and on aspects of phosphorus and nitrogen pollution from rearing of silver perch.

Acknowledgment

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References

Albaster, J.S. 1982. Report of the EIFAC workshop on fish-farm effluents, Silkeborg, Denmark, 26-28 May 1981. FAO/EIFAC Technical Paper 41. 166 pp.

Allan, G.L. and S.J. Rowland. 1995. The use of Australian oilseeds and grain legumes in aquaculture diets. The Third Asian Fisheries Forum: 667-669.

Allan, G., S. Rowland, K. O'Brien, A. Anderson and N. Preston. 1994. Fishmeal replacement in aquaculture feeds for silver perch. In: Replacement of fishmeal in aquaculture diets. Fisheries Research and Development Corporation. Sub-Program 93/120. Year one progress report - November 1994.

Allan, G.L. and S.J. Rowland. 1992. Development of an experimental diet for silver perch (*Bidyanus bidyanus*). Austasia Aquaculture 6: 39-40.

Allan, G.L. and S.J. Rowland. 1991. Growth of silver perch *Bidyanus bidyanus* on diets with different levels and sources of protein. Proceedings of the Nutritional Society of Australia 16: 211.

AOAC. 1990. Official methods of analysis. Fifteenth edition, vol. 1. The Association of Official Analytical Chemists (ed. K. Helrich). Virginia, Arlington. 682 pp.

APHA. 1989. Standard methods for the examination of water and waste water. Seventh edition. American Public Health Association, American Water Works Association and Water Pollution Control Board, Washington D.C. 1,527 pp.

Asgard, T., T. Storebakken and E. Austreng. 1986. Reduction of self-pollution by correct feeding. Poster at the 3rd International Conference on Aquafarming. AQUACOLTURA '86, Verona, Italy, 9-10 October 1986.

Barlow, C.G. and K. Bock. 1981. Fish for farm dams. NSW Fisheries, Sydney.

Bergheim, A., H. Hustveit and A. Selmer-Olsen. 1984. Estimated pollution loadings from Norwegian fish farms. II. Investigations 1980-1981. Aquaculture 36: 157-168.

Beveridge, M.C.M., M.J. Phillips and R.M. Clarke. 1991. A quantitative and qualitative assessment of wastes from aquatic animal production. In: Aquaculture and water quality, Advances in world aquaculture, volume 3 (eds. D.E. Brune and J.R. Tomasso), pp. 506-533. The World Aquaculture Society.

Cadwallader, P.L. 1979. Distribution of native and introduced fish in Seven Creeks River system, Victoria, Australia. Australian Journal of Ecology 4: 361-385.

Carr, O.J. and R. Goulder. 1990. Fish-farm effluents in rivers-1: Effects on bacterial populations and alkaline phosphate activity. Water Research 24(5): 631-638.

Chiu, Y.N. 1989. Considerations for feeding experiments to quantify dietary requirements of essential nutrients in fish. Fish nutrition research in Asia. Proceedings of the Third Asian Fish Nutrition Network Meeting: 46-57.

Cowey, C.B and C.Y. Cho, Editors. 1991. Nutritional strategies and aquaculture waste. Proceedings First International Symposium on Nutritional Strategies in Management of Aquaculture Waste: 37-50.

EIFAC. 1980. Report of the EIFAC, IUNC, and ICES working group on standardization of methodology in fish nutrition research. (eds. J.D. Castell and K. Tiews). European Inland Fisheries Advisory Commission. EIFA/T36. EIFAC/FAO. 24 pp.

Foy, R.H. and R. Rosell. 1991a. Loadings of nitrogen and phosphorus from a Northern Ireland fish farm. Aquaculture 96: 17-23.

Foy, R.H. and R. Rosell. 1991b. Fractionation of phosphorus and nitrogen loadings from a Northern Ireland fish farm. Aquaculture 96: 31-42.

Gooley, G. and S. Rowland. 1993. Murray-Darling finfish current developments and commercial potential. Austasia Aquaculture 7:35-38.

Ketola, H.G. 1982. Effect of phosphorus in trout diets on water pollution. Salmonid 6(2): 12-15.

Ketola, H.G. and M.E. Richmond. 1994. Requirement of rainbow trout for dietary phosphorus and its relationship to the amount discharged in hatchery effluents. Transactions of the American Fisheries Society 123: 587-594.

Lall, S.P. 1991. Digestibility, metabolism and excretion of dietary phosphorus in fish. Nutritional strategies and aquaculture waste. Proceedings First International Symposium on Nutritional Strategies in Management of Aquaculture Waste: 163-185.

Laird, L.M. and T. Needham. 1988. Salmon and trout farming. Ellis Horwood Limited, London. 271 pp.

New, M.B. 1987. Feed and feeding of fish and shrimp - A manual. FAO/UNEP. ADCP/REP/87/26, Rome. 274 pp.

Penczak, T., W. Galicka, M. Molinski, E. Kusto and M. Zalewski. 1982. The enrichment of a mesotrophic lake by carbon, phosphorus and nitrogen from the cage aquaculture of rainbow trout *Salmo gairdneri*. Journal of Applied Ecology 19: 371-393.

Persson, G. 1991. Eutrophication resulting from salmonid fish culture in fresh and salt waters, Scadinavian experiences. Nutritional strategies and aquaculture waste. Proceedings First International Symposium on Nutritional Strategies in Management of Aquaculture Waste: 163-185.

Persson, G. 1988. Relationships between feed, productivity and pollution in the farming of large rainbow trout (*Salmo gairdneri*). Swedish Environmental Protection Agency, Stockholm, PM 3534. 48 pp.

Phillips, M.J. 1985. The environmental impact of cage culture on Scottish freshwater lochs. Institute of Aquaculture, Stirling. 106 pp.

Phillips, M.J., M.C.M. Beveridge and D.J. Macintosh. 1990. The impact of aquaculture on the coastal environment. Proceedings of ISCZCZ 1988: 537-545.

Pillay, T.V.R. 1992. Aquaculture and the environment. Fishing News Books, Oxford. 184 pp.

Rowland, S.J. 1995. Production of fingerling and market-size silver perch in earthen ponds. Silver perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrendrea, April 1994. Austasia Aquaculture, Hobart for NSW Fisheries, Sydney: 41-49.

Rowland, S.J. and G.L. Allan. 1994. Production of fingerling, *Bidyanus bidyanus* (Teraponidae) at two densities in earthen ponds in Australia. Progressive Fish Culturist. 56:296-298.

Rowland, S.J. and C.G. Barlow. 1991. Fish biology - the right prerequisite. A case study with freshwater silver perch (*Bidyanus bidyanus*). Austasia Aquaculture 5: 27-30.

Rowland, S.J., G.L. Allan, K. Clark, M. Hollis and T. Pontifex. 1994. Production of fingerling *Bidyanus bidyanus* (Teraponidae) at two densities in earthen ponds in Australia. Progressive Fish Culturist. 56: 296-298.

Rowland, S., G. Allan, M. Hollis and T. Pontifex. 1995. Production of the Australian freshwater silver perch, *Bidyanus bidyanus* (Mitchell), at two densities in earthen ponds. Aquaculture 130 : 317-328.

Solbe, J.F. de L.G. 1982. Fish farm effluents: a United Kingdom survey. Report of the EIFAC Workshop on fish farm effluents, 26-28 May, Silkeborg, Denmark. EIFAC Technical Paper 41: 29-55.

Tredwell, R., L. McKelvie and G.B. Maguire. 1992. Potential of Australian aquaculture. Australian Bureau of Agricultural and Resource Economics. ABARE Research Report No. 92.2. Commonwealth of Australia, Canberra. 81 pp.

Warren-Hansen, I. 1982. Evaluation of matter discharged from trout farming in Denmark. In: Report of the EIFAC Workshop on fish farm effluents, 26-28 May. Silkeborg, Denmark. EIFAC Technical Paper 41: 57-63.

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The nutrient content and the release of nutrients from fish food and faeces

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Abstract

The fish food and faeces were fractioned into the different components of phosphorus and nitrogen. There was a rapid release of phosphorus from the fish food and faeces and a decrease thereafter whereas ammonium release was slow at first with the rate increasing with time. Both temperature and pH affected the release of nutrients from fish food and faeces. The release of phosphorus and nitrogen was higher at higher temperatures. The maximum release of phosphorus was at pH 4.0 whereas nitrogen release was maximum at neutral (7.0) to alkaline (10.0) media.

Introduction

Fish food and faeces are known to be the main wastes in intensive aquaculture. The production of feed wastes as dust and uneaten food have been estimated to be 20% (Beveridge, 1987) whereas other studies reported a figure of 10–30% of waste uneaten food alone from intensive aquaculture (Hoelzi & Vens-Cappell, 1980; Penczak et al. (1982). The faecal input on a dry weight basis is reported to be 260 g per kg of food (Butz & Vens-Cappell, 1982). This uneaten food along with faeces (solid wastes) may increase the sedimentation and enrich the nutrient pool of the receiving waters (Beveridge, 1987). Under appropriate conditions phosphorus and nitrogen could be released from the sediments and may stimulate algal growth (Pettersson, 1988).

Both nitrogen and phosphorus are the essential ingredients incorporated in formulated feed to achieve a good growth of fish. These two nutrients are also required for algal growth in water bodies (State of Victoria, 1995). In freshwater, phosphorus is the limiting nutrient and orthophosphate is the form that is readily available to plants (Chamberlain & Shapiro, 1969; Welch & Lindell, 1980; Bostrom et al., 1988; Boyd, 1990). Both nitrogen and phosphorus normally are limiting nutrients in seawater (Boyd, 1990). Fish farm effluents containing phosphorus and nitrogen have been reported to have caused eutrophication of receiving waters (Laird & Needham, 1988; Foy & Rosell, 1991b). However, few authors have reported on the fractional composition of nutrients and factors related in the release of nutrients from fish food and faeces. Such information is essential in planning strategies for nutrient management and pollution control. The silver perch, *Bidyanus bidyanus* is an Australian native fish of highest aquaculture potential (Allan & Rowland, 1996) and the industry is booming in the country due to interest and investments of culturing the species (Kibria et al., 1996). Although there is some concern on the environmental impacts of aquaculture, until now there is no research either on the quantity or quality of nutrients that may discharge from aquaculture of silver perch. The present study investigates the fractional composition of nutrients and the release of nutrients from fish food and faeces in the rearing of the Australian native fish silver perch *B. bidyanus*.

Materials and methods

Silver perch *Bidyanus bidyanus* fingerlings (0.50–0.68 g) were fed on two commercial diets referred to

as diet-1 and diet-2 for four weeks at 25 °C. These two diets were selected since they resulted better nutrient retention in silver perch in our previous experiments. Fish were reared in small aquarium (70 × 60 × 30 cm) with four fish per aquarium. The total number of fish per treatment was twenty four i.e. the number of replicate tanks per diet was six. Fish were fed at the rate of 3% body weight, twice a day and six days a week. Faeces was siphoned off a number of times a day by using a 5 mm hose. Collected faeces was dried overnight in an oven at 105 °C. Pooled faeces of each 'diet group' were used for nutrient fractionation and to conduct experiments on nutrient release. Experiments were also conducted on the effect of temperatures and pH on the release of nutrients from fish food since uneaten food is the major source of aquaculture wastes. Diet-1 contained 53% protein (8.48% nitrogen) and 1.31% phosphorus while diet-2 contained 45% protein (7.2% nitrogen) and 1.16% phosphorus. Protein (N ×6.25) and total phosphorus in feed, fish and faeces were determined following AOAC (1990). Total nitrogen, ammonium (NH_4^+), nitrite (NO_2^-), nitrate (NO_3^-), total P and orthophosphate (PO_3^-) of water samples were analysed with a Tecator flow injection analyzer (Aquatec 5400 analyzer) following the Aquatec instruction manual (Tecator, 1990). pH was measured using a pH meter (Orion model SA 520). All food and faeces were dried at 105 °C overnight for moisture analysis.

Fractionation of nutrients

Phosphorus fractionation

The inorganic phosphorus in fish food and faeces were fractioned following the extraction methods of Pettersson (1988) in order to separate the orthophosphate fractions that are directly available to plants (labile fractions) and the fractions bound in the sediments (iron, aluminium and calcium bounded phosphorus) (Table 1).

Nitrogen fractionation

Dried faeces collected from the two diet groups was fractioned into nitrogen composition (total nitrogen, nitrite, nitrate and ammonium) following the methods described in Kristiansen and Hessen (1992).

Release of nutrients from faeces and foods

Release of nutrients from faeces

The release of phosphate and ammonium from faeces were determined by incubating 75 mg faeces in 50 ml distilled water in Erlenmeyer flasks at 25 °C for a week following Pettersson (1988).

Effect of temperatures and pH on the release of nutrients from fish food

300 mg of fish food (diet-2) was incubated with 200 ml distilled water for seven days in glass bottles. The food was incubated [at each of two temperatures (20 °C and 25 °C)] at three pH levels (4.0, 7.0 and 10.0) in order to observe the effect of pH and temperature on the release of nutrients. The fractions measured were dissolved phosphorus (orthophosphate) and ammonium, the most important nutrient fraction which impact on water quality and aquatic living organisms (Bostrom et al., 1988; Welch & Lindell, 1988).

Statistical analysis

Fish food and faeces for nutrient fraction and nutrient release experiments had a replicate of three or more. Mean standard deviation and standard error of the mean (s.e.) were calculated from all the measured variables of phosphorus and nitrogen fractions following Zar (1984). One way analysis of variance (ANOVA) was used to compare the release nutrients from faeces from two diet groups. ANOVA's were calculated using a IBM compatible MS Excel programme, setting the significance level at 0.05. Best curves was fitted to find the trend or correlations of variables/data series on the release of orthophosphate and ammonium from fish food at different pH. The coefficient of determination (r^2) were determined to find the relationship between the x and y data points in the fitted data series. All curves and correlation calculation was done using Cricket graph-III (version 1.0, Computers Associates International, USA).

Table 1. Extraction of orthophosphate from fish food and faeces

Extraction no	Extraction methods	Extractable phosphorus
* Extraction-1	Distilled water four times for 90 minutes	Water soluble phosphorus
* Extraction-2	NH_4CL two times, 120 minutes each	Water soluble phosphorus
Extraction-3	NaOH (0.1 mol/l) for 16 hours	Fe and Al bound phosphorus
Extraction-4	HCl (0.5 mol/l) for 24 hours	Ca bound phosphorus

* phosphorus extracted are directly available to plants.

Table 2. Fractional composition of phosphorus (orthophosphate) in fish food and faeces (mg P g^{-1} dry weight) ($n = 4$; mean ± s.e.). Faeces-1 and faeces-2 are from the diet-1 and diet-2 respectively fed to silver perch. Details on extraction procedures are given in Table 1

	Diet-1	Diet-2	Faeces-1	Faeces-2
H_2O-P	2.57 ± 0.09	1.87 ± 0.05	1.03 ± 0.04	0.94 ± 0.04
NH_4Cl-P	1.68 ± 0.19	1.44 ± 0.05	1.46 ± 0.21	0.94 ± 0.12
NaOH-P	2.33 ± 0.31	1.68 ± 0.14	1.10 ± 0.17	0.68 ± 0.25
HCl-P	2.25 ± 0.14	1.50 ± 0.33	1.00 ± 0.18	0.71 ± 0.01

Results and discussion

Fractional composition of phosphorus in feed and faeces

Water soluble phosphorus varied between 1.87–2.57 mg P g^{-1} dw in diets. The diet with the highest total phosphorus content also had the highest concentration of water-soluble phosphorus ($P > 0.05$, $n = 4$). This fraction ranged from 28.8 to 29% of the total phosphorus content. Pettersson (1988) also found the highest concentration of water soluble phosphorus in diets containing the highest total phosphorus. Both fish food and faeces contained major phosphorus fraction in a labile form represented by the fraction extracted by water and ammonium chloride (Table 2). The maximum labile fraction in rainbow trout food is reported to be 26–38% and in faeces 15–54% (Pettersson, 1988). Our experimental results show that uneaten food and faeces may contain a majority of phosphorus in a labile form and the labile forms are believed to be readily available to plants for their growth (Butz & Vens-Cappell, 1982).

Nutrient content in faeces

Faecal analysis revealed a higher total phosphorus and lower nitrogen content in faeces of silver perch than those in diets. Faeces of diet group 1 and diet group 2 contained 3.08 ± 0.20% and 1.99 ± 0.18% phosphorus and 3.29±0.29% and 3.21±0.51% nitrogen by weight. However, they were no significantly differences in faecal phosphorus and nitrogen content between the two diet groups ($P > 0.05$). This may confirm that the main path of phosphorus loss in fish is through faeces. A high phosphorus lost through faeces was also found by Pettersson (1988) in rainbow trout, and Johnsen et al. (1993) in Atlantic salmon. Hakanson et al. (1990) calculates that of the total phosphorus and nitrogen fed to fish, 70% phosphorus and 15% nitrogen was lost through faeces. Kristiansen and Hessen (1992) reported a value of 4.0% phosphorus and 2.3% loss of nitrogen through faeces in Atlantic salmon (*Salmo salar*) and a loss of 3.5% phosphorus and 4.1% nitrogen in noble crayfish (*Astacus astacus*) faeces, while the faecal analysis of rainbow trout (*Salmo gairdneri*) showed 1.59 ± 0.49% phosphorus and 3.93 ± 1.04% nitrogen (Penczak et al., 1982). The phosphorus and nitrogen ratio in silver perch diets were 1 : 6.5 and 1 : 6.2 in diet 1 and diet 2 while the P : N ratios in the faeces was 1 : 1.1 and 1 : 1.61 for diet group 1 and diet group 2 respectively (weight basis). This agrees with the results of Pettersson (1988) who found that the phosphorus : nitrogen ratio in the foods was 1 : 5 while in the faeces it was 1 : 1–2. Furthermore, above faecal P : N ratio show that nitrogen lost through faeces in silver perch is minimal. Fivelsad et al. (1990) stated that faecal nitrogen loss could be 10–20% of food fed to fish.

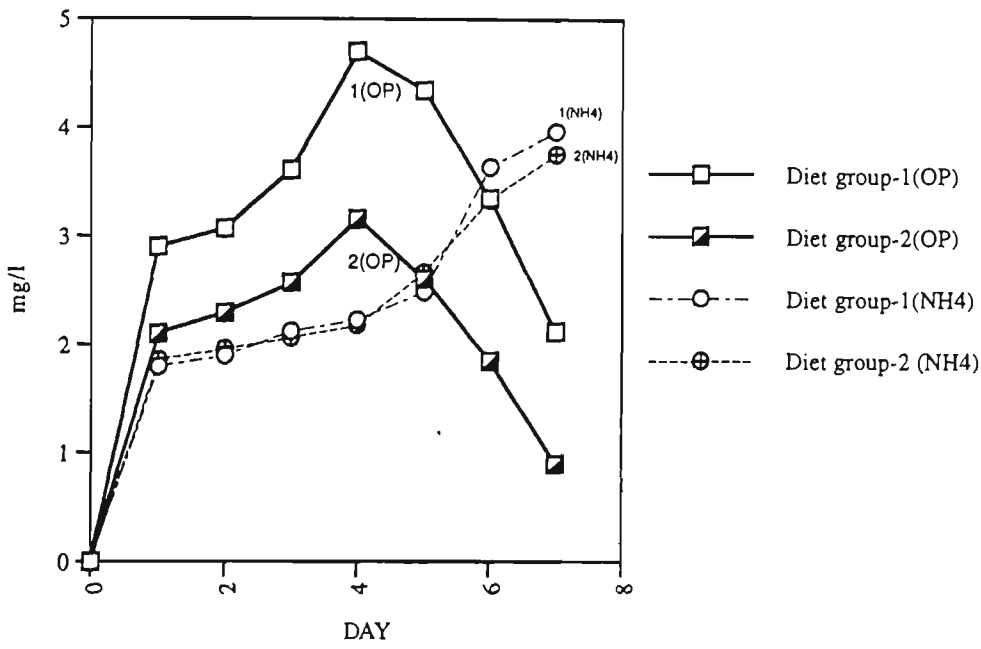

Figure 1. The average release of orthophosphate (OP) and ammonium (NH_4) from faeces of silver perch. The faeces were incubated at 25 °C for seven days ($n = 4$). There was no significant differences in the release of either orthophosphate ($F = 2.91$; F critical = 4.6) or ammonium ($F = 0.005$; F critical = 4.6) between the two diet groups ($p > 0.05$).

Table 3. Nitrogen content of dry faeces of silver perch reared for four weeks at 25 °C and fed two diets (mg N g^{-1} DW) ($n = 3$; mean ± s.e.)

Diet-group	Total-N	NO_3-N	NO_2-N	NH_4-N
1	25.90 ± 0.10	1.92 ± 0.21	0.71 ± 0.10	1.71 ± 0.04
2	25.76 ± 0.13	1.56 ± 0.28	0.72 ± 0.07	1.78 ± 0.03

Fractional composition of nitrogen in faeces

The contributions of faeces for the various nitrogen components are summarised in Table 3. It showed that ammonium (5.65–6.0%), nitrite (2.35–2.38%) and nitrate (5.23–6.31%) are relatively low in the faeces compared to total N (86.44–85.62%) (dry weight basis). The ammonium contribution was also minimal in crayfish and salmon faeces being 4.0–4.4% of total nitrogen (Kristiansen and Hessen, 1992). Foy and Rosell (1991a) fractioned nitrogen loadings from a Northern Ireland fish farm where they estimated that nitrite and nitrate contributed to about 2.4% of the total nitrogen loadings to the environment.

Release of nutrients from faeces

There was a rapid and momentary release of phosphorus from faeces for the first few days and at the later part the bacterial growth may have slowed down the release of phosphate (Figure 1). Our results is in agreement of Pettersson (1988) who also observed a rapid release of phosphate from fish food during the first few days and thereafter a growth of bacteria decreased the phosphate concentration. Ammonium release from faeces was slow and increased slowly with time (Figure 1). These experiments demonstrate that leaching of nutrients from faeces or food could be instantaneous and therefore nutrient enrichment of the environment occurs almost immediately. Both Makinen et al. (1988) and Phillips et al. (1993) stated that an efficient and quick removal of solid wastes is essential if phosphorus loadings to the environment are to be controlled.

Effect of temperatures and pH on the release of nutrients

Release of nutrient from the fish feed was found to be related to temperatures, and the rate of release was higher at a higher temperature ($P < 0.05$). The

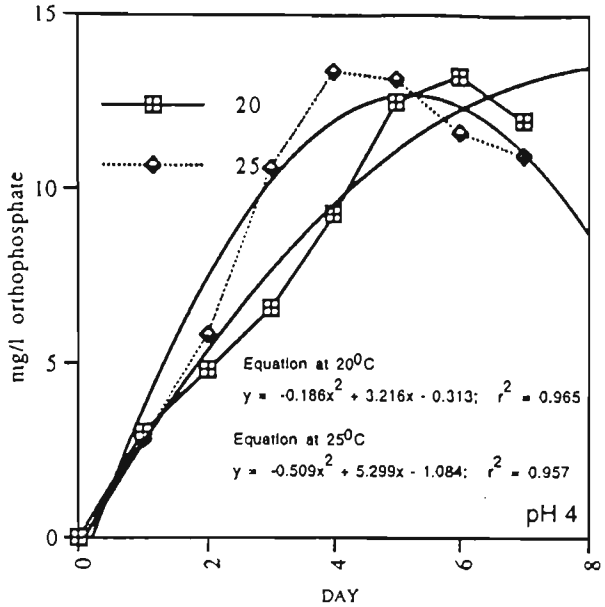

Figure 2. The average release of orthophosphate from fish food at pH 4.0 and incubated at two temperatures for seven days ($n = 4$).

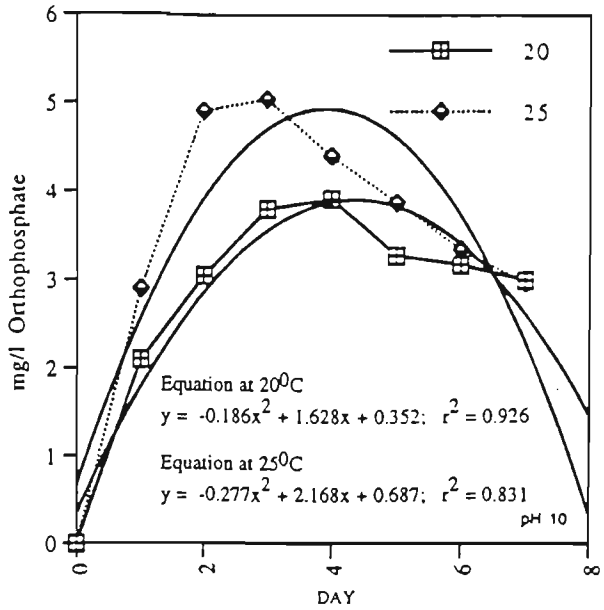

Figure 4. The average release of orthophosphate from fish food at pH 10.0 and incubated at two temperatures for seven days ($n = 4$).

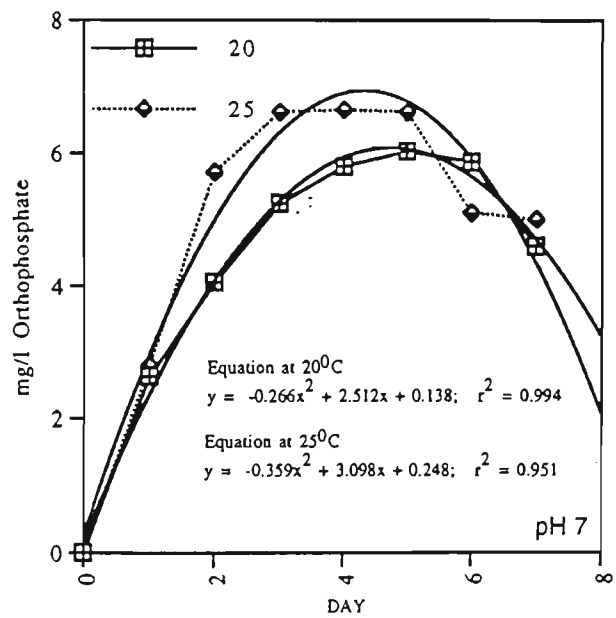

Figure 3. The release of orthophosphate from fish food at pH 7.0 and incubated at two temperatures for seven days ($n = 4$).

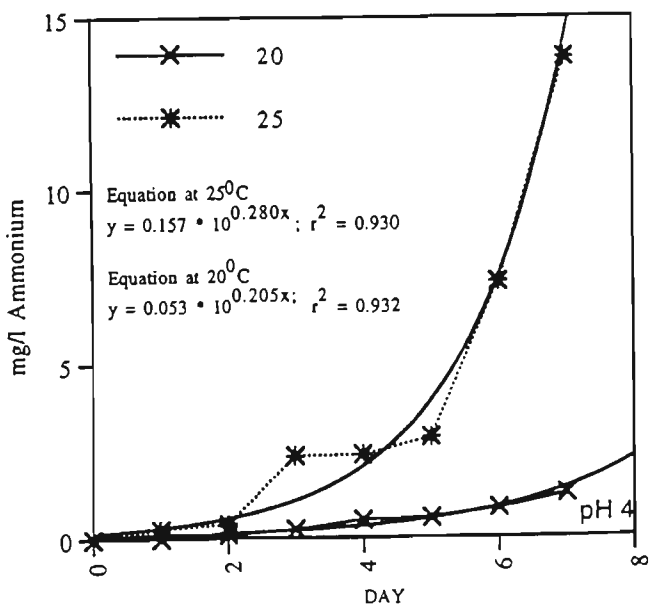

Figure 5. The average release of ammonium from fish food at pH 4.0 and incubated at two temperatures ($n = 4$).

release of phosphorus and ammonium were accelerated at 25 °C in comparison to 20 °C (Figure 2, 3, 4, 5, 6, 7). The release of orthophosphate was higher in acidic medium (pH 4.0) (Figure 2, 3, 4) whereas ammonium release was accelerated in neutral to alkaline media (pH 7.0 and 10.0) (Figure 5, 6, 7)). Persson (1988) stated that release of nutrients from fish food and faeces depends upon pH, temperature, oxygen, turbulence and microbial activity of the environment. The patterns of phosphorus and nitrogen released observed were similar to those recorded by Pettersson (1988). Pettersson (1988) also reported a much higher phosphorus release in acidic medium (pH 5.0) and found that the leakage of ammonium from fish food was temperature dependent. The release of ammonium and orthophosphate

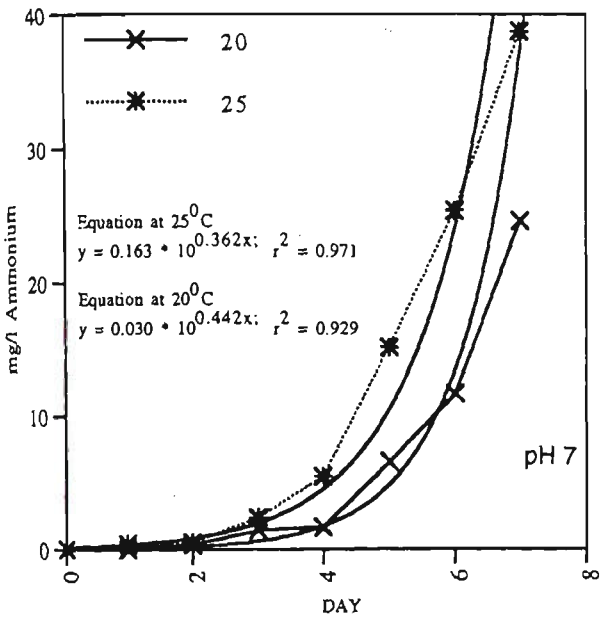

Figure 6. The average release of ammonium from fish food at pH 7.0 and incubated at two temperatures ($n = 4$).

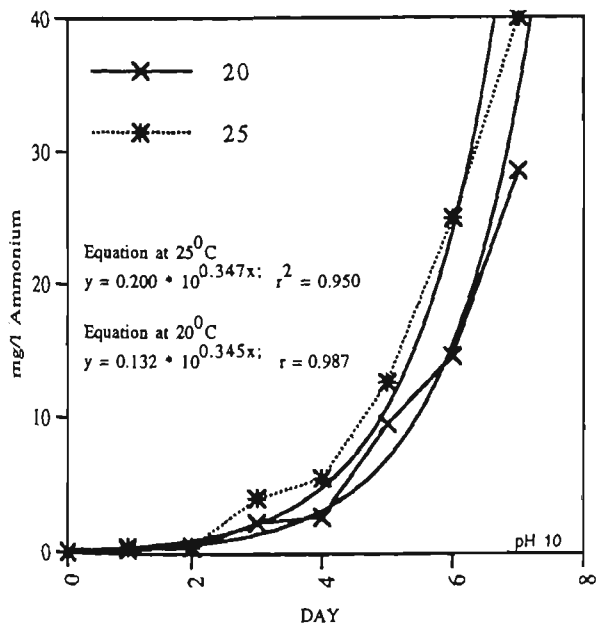

Figure 7. The average release of ammonium from fish food at pH 10.00 and incubated at two temperatures ($n = 4$).

from salmon excreta were also higher at higher temperatures (Kristiansen & Hessen, 1992).

Conclusion

The results show that the main path of phosphorus loss to the environment in the culture of silver perch is via faeces. The maximum release of phosphorus from solid wastes depends upon temperatures and pH of the environment, being higher at 25 °C than at 20 °C. The release of phosphorus increases with decrease in pH whereas ammonium release increases with increase in pH. Both fish food and faeces may contain a major phosphorus content in a labile form, therefore further experiments are needed to investigate the minimisation of total and labile phosphorus content in commercial diets in order to reduce the phosphorus load to environment without affecting the growth of the cultured species. Nitrogen loss through faeces are minimal however, uneaten food is probably the major input of nitrogen (as solid waste) to environment (Penczak et al., 1982; Beveridge, 1987), and measures are needed to minimise the feed wastes in aquaculture.

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References

Allan, G. L. & S. J. Rowland, 1996. The silver perch industry: research progress and priorities toward developing cost-effective diets. Austasia Aquaculture 10: 37–40.

AOAC, 1990. Official methods of analysis of the Association of Official Analytical Chemists (AOAC) (Editor K. Helrich). 15th edn., vol. 1. Virginia, USA.

Beveridge, M., 1987. Cage Aquaculture. Fishing News Ltd. Farnham, England: 351 pp.

Bostrom, B., G. Persson & B. Broberg, 1988. Bioavailability of different phosphorus forms in freshwater systems. Hydrobiologia 170: 133–155.

Boyd, C. E., 1990. Water quality in ponds for aquaculture. Alabama Agricultural Experiment Station. Auburn University and Birmingham Publishing Co. Alabama, USA, 462 pp.

Butz, I. & B. Vens-Cappell, 1982. Organic load from the metabolic products of rainbow trout fed with dry food. In Albaster, J. S. (ed.), Report of the EIFAC Workshop on Fish Farm Effluents. Silkeborg, Denmark, 26–28 May 1981. EIFAC Tech. Pap. 41: 57–70.

Chamberlain, W. & J. Shapiro, 1969. On the biological significance of phosphate analysis: Comparison of standard and new methods with a bioassy. Limnol. Oceanogr. 14: 921–927.

Fivlstad, S., J. M. Thomassen, M. J. Smith, H. Kjartansson & A-B. Anderson, 1990. Metabolic production rates from Atlantic salmon (*Salmo salar* L.) and Arctic char (*Salvelinus alpinus* L.) reared in single pass land based brackish water and sea water systems. Aquacult. Engin. 9: 1–21.

Foy, R. H. & R. Rosell, 1991a. Fractionation of phosphorus and nitrogen loadings from a Northern Ireland Fish Farm. Aquaculture 96: 31–42.

Foy, R. H. & R. Rosell, 1991b. Loadings of nitrogen and phosphorus, suspended solids and BOD from a Northern Ireland fish farm. Aquaculture 96: 17–30.

Hakanson, L., A. Wrvik, T. Makinene & B. Molleg, 1988. Basic concepts concerning assessments of environmental effects of marine fish farms. Nordic Council of Ministers, Copenhagen, 103 pp.

Hoelzi, A. & B. Vens-Cappell, 1980. Profitability of food-fish production in net cages. Fisch. Teichwirt. 32: 2–5.

Johnsen, F., M. Hillestad & E. Austreng, 1993. High energy diets for Atlantic salmon. Effect on pollution. In Kaushik, S. J. & P. Luquet (eds), Fish Nutrition in Practice. INRA, Paris: 391–401.

Kibria, G., D. Nugegoda, R. Fairclough & P. Lam, 1996. Australian native species in aquaculture. Victorian Nat. 113: 264–267.

Kristiansen, G. & D. O. Hessen, 1992. Nitrogen and phosphorus excretion from the noble crayfish, Astacus astacus L., in relation to food type and temperature. Aquaculture 102: 245–264.

Laird, L. M. & T. Needham, 1988. Salmon and Trout Farming. Ellis Horwood Limited, England, 271 pp.

Makinen, T., S. Lindgren & P. Esklinen, 1988. Sieving as an effluent treatment for aquaculture. Aquacult. Engin. 7: 367–377.

Penczak, T., W. Galicka, M. Molinski, E. Kusto & M. Zalewski, 1982. The enrichment of a mesotrophic lake by carbon, phosphorus and nitrogen from the cage aquaculture of rainbow trout, *Salmo gairdneri*. J. Appl. Ecol. 19: 371–393.

Persson, G., 1988. Relationships between feed, productivity and pollution in the farming of large rainbow trout (*Salmo gairdneri*). Swedish Environmental Protection Agency, Stockholm, PM 3534, 48 pp.

Pettersson, K., 1988. The mobility of phosphorus in fish-foods and fecals. Verh. int. ver. Limnol. 23: 200–206.

Phillips, M., R. Clarke & A. Mowat, 1993. Phosphorus leaching from Atlantic salmon diets. Aquacult. Engin. 12: 47–54.

State of Victoria, 1995. Nutrient management strategy for Victorian Inland Waters. State of Victoria, Australia, 73 pp.

Tecator, 1990. Instruction manual. The Aquatec System. Hoganas: Sweden, 49 pp.

Welch, E. B. & T. Lindell, 1980. Ecological effects of wastewater. – Applied limnology and pollutant effects. Chapter 4. Nutrient cycles: 54–81.

Zar, J. H., 1984. Biostatistical analysis. Prentice-Hill, Englewood Cliffs, New Jersey, 620 pp.

Australian native freshwater fish and crustaceans:

Environmental role and aquaculture potential

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Paul Lam
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Australia loses 80% of its precipitation through evaporation, and this makes it the driest continent after Antarctica. It has 180-190 species of freshwater fishes,[65] of which 127 are endemic to Australia.[6] The diversity of fishes appears to be low when compared with other southern continents. For example, in Africa there are more than 1900 species.[51] It is generally believed that the limited number of freshwater fish species in Australia reflects the long geographical isolation of the continent.

Because of the rainfall distribution pattern in Australia, freshwater aquaculture is restricted to the coastal belts, particularly in New South Wales, Queensland, Victoria and Western Australia.

The demand for freshwater and marine aquaculture products has been increasing by 3% annually since 1980.[39] This increasing demand of aquatic foods has led to an increase in activities aiming to development of new farming areas with new species, and utilization of large offshore fisheries. The need to balance the potential environmental impact of aquaculture activities and aquatic food production has resulted in a renewed interest in freshwater aquaculture. The native freshwater fish and crustaceans, apart from their inherent conservation values, represent a huge potential for aquatic food production. This article reviews the importance of Australian native freshwater fish and crustaceans in the context of conservation and aquaculture with particular emphasis on the silver perch.

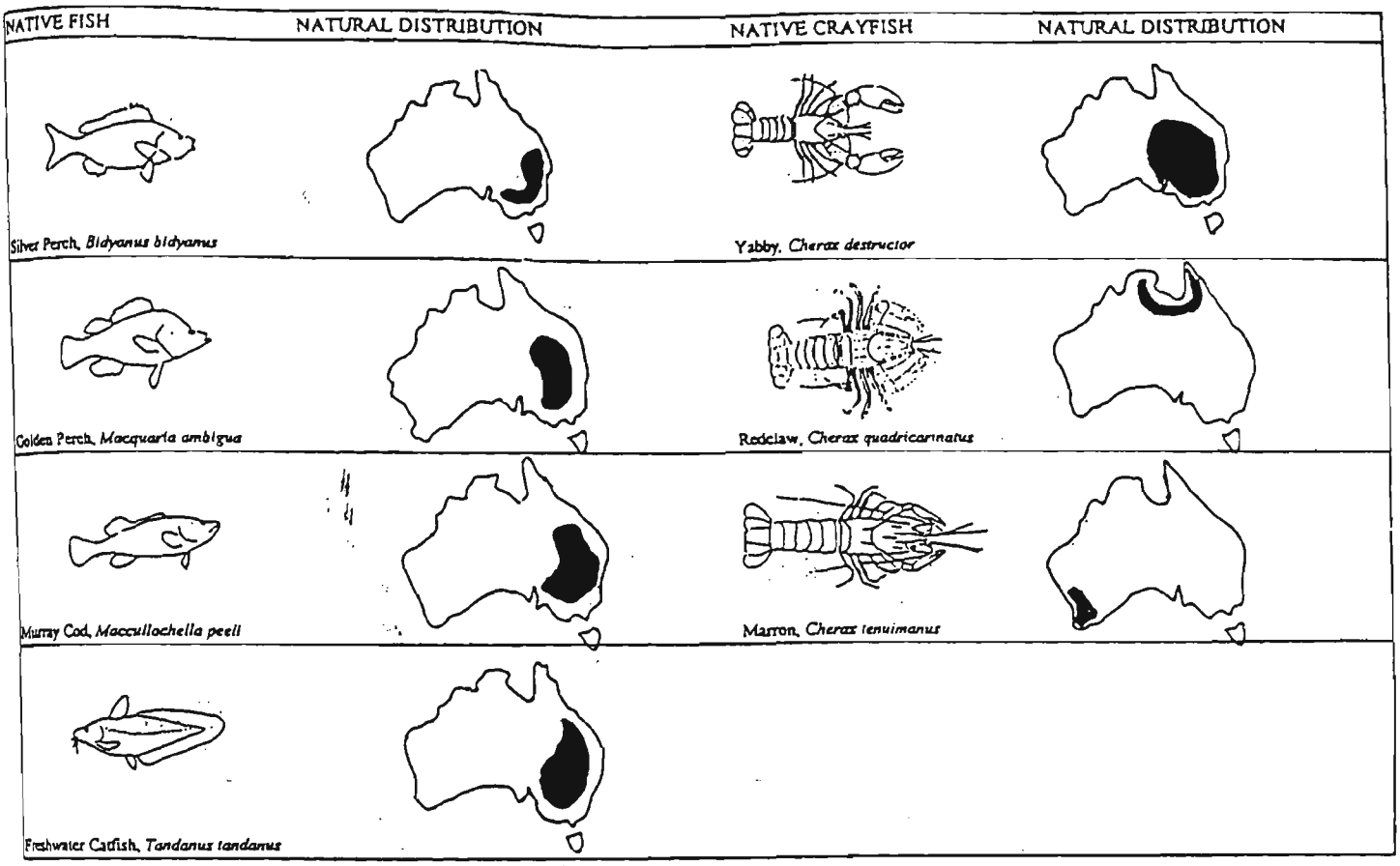
Native fauna and flora have value as biological indicators of inland water quality because many of them are sensitive to short- and long-term changes in the aquatic environment. In general, a robust and diverse native fish population indicates a healthy aquatic environment.Conversely, an environment that has no native fish or only a few exotic species may be seriously degraded. Native fauna, faced with a deteriorating aquatic environment, must either emigrate or die, leaving the environment to hardy exotic species, (e.g. introduced carps) that can survive in water quality that would kill most native species.

Rapid decline in the populations of native fish species has been reported in the MurrayDarling river system.[45,48,51] The main threats are dams and barriers, runoff from agriculture and sewage or industrial effluents, overfishing, loss of genetic variability and inbreeding and introduction of exotic species,[27,36,48,51] as well as overexploitation of the resources. These physical, chemical and biological factors have had enormous impacts on the local fish fauna and a large number of native fish considered to be endangered, vulnerable or threatened (Table 1).

Aquaculture potential

Aquaculture is a recent addition to the primary industry (agriculture, livestock) in Australia and is expanding rapidly.[70] Aquaculture production has increased steadily, especially the culture of native fish, and a further rise in the production of native fish and crayfish is expected because of the increased local and foreign demand. Private hatcheries in New South Wales (NSW), Victoria (VIC), Queensland (QLD), and South Australia (SA) produce silver perch, golden perch, Murray cod and crayfish for sale to the public for stocking private dams, and to state and territorial organizations for stocking public waterways. Native fish farming is becoming more attractive. For example, there were only 55 farms in Australia in 1988/89, but by 1994 the number of farms had increased by two to three times (Table 3). This rise is attributed to the increased interests and investment in growing silver perch.

Recent reviews of the status of aquaculture in Australia[28,35,57,59,60,86] have indicated that four native fish and three crustaceans (crayfish) have good potential for aquaculture in inland waters. The biology of these species is well understood and the culture techniques are well established. In addition, there is local as well as overseas demand for these species, and their profitability in-



Natural distribution of Australian native fish and crayfish under aquaculture.[35,67,85]

dex is attractive. The four fish in this category are found in MurrayDarling river systems: the silver perch (*Bidyanus bidyanus*), golden perch (*Macquaria ambigua*), Murray cod (*Maccullochella peeli*) and freshwater catfish (*Tandanus tandanus*). The three crustaceans are crayfish: the yabby (*Cherax destructor*), redclaw (*Cherax quadricarinatus*) and marron (*Cherax tenuimanus*). There are over 100 species of Australian freshwater crayfish[30] but these three have the most promise because their biology and culture technology are well known.

Among the native fish, the silver perch (*Bidyanus bidyanus*) has the greatest potential for aquaculture in Australia;[3,24,73,79] followed by the golden perch, Murray cod and catfish.[90]

Based on production, marketability and profitability, silver perch and redclaw appear to have the best prospects for aquaculture development. Generally, the aquaculture potential of a species depends mainly on its market demand. However, the characteristics required for profitable aquaculture should be compared with growth and production performance of the fish species in order to evaluate its aquaculture potential. Noteworthy characteristics are controlled breeding, readily acceptance of artificial food, tolerance to fluctuation of water quality and others. An example of a scheme for evaluating the aquaculture potential of Australian native species is given in Table 2 . Potential to be bred artificially and stocked successfully at any time of the year at a reasonable cost are prime determining factors in species screening. The availability of growout technology in the local environment and the demands of domestic and export markets are additional important criteria in species ranking.[86]

Biology and culture

Silver perch

The silver perch *(Bidyanus bidyanus)* is endemic to most of the MurrayDarling river system except the higher altitudes.[43] The natural distribution of silver perch along with other native species is given in Figure 1. The species was commonly consumed by Aboriginies and the scientific name was also derived from the Aboriginal name bidyan.[74] Silver perch is a potamodromous species, migrating wholly in freshwater.[27] The species can tolerate a wide range of temperature, from 20°C to 32°C[18] but the optimum temperature for growth is probably in the range of 23–28°C.[76] The important water quality parameters for silver perch culture have been summarized.[75] The larvae are benthic,[42] and juveniles form large schools,[43,50] often congregating below rapids[50] and fast flowing water with sand or gravel bottom.[51,85] It can also live in extremely turbid waters,[15] it is salt tolerant,[31] and both territorial and aggressive.[18,85] The average silver perch can reach 180 mm in two years time,[18] and specimens of 350–410 mm (0.75kg–2.5 kg).[52] are commonly caught.[51]

Table 1. Current status of native fish species in Victoria.

Status	Species
Extinct No longer found in the wild	1. Assiz's chanda perch *(Ambassis agassizi)* 2. Southern purple-spotted gudgeon *(Mogurunda adspersa)*
Endangered Immediate danger of extinction	1. Trout cod *(Maccullochella macquariensis)* 2. Brown galaxias *(Galaxias fuscus)* 3. Freshwater herring *(Potamalosa richmondia)* 4. Ewen's pigmy perch *(Nannoperca variagata)*
Vulnerable Species can move into endangered category soon	1. Freshwater catfish *(Tandanus tandanus)*** 2. Macquaria perch *(Macquaria australiasica)* 3. Tasmanian mudfish *(Galaxias cleaveri)* 4. Silver perch *(Bidyanus bidyanus)*** 5. Murray cod *(Maccullochella peeli)*** 6. Australian grayling *(Protoctes maraena)*
Potentially Threatened Species currently not endangered. Vulnerable, but could be at risk	1. Australian bass *(Macquaria novemaculeata)* 2. Broad-finned galaxias *(Galaxias brevipinnis)* 3. Dwarf galaxias *(Galaxiella pusilla)* 4. Spotted galaxias *(Galaxias truttaceus)* 5. Pouched lamprey *(Geotria australis)* 6. Golden perch *(Macquaria ambigua)*** 7. Yarra pigmy perch *(Edelia obscura)*
Indeterminate Species endangered/vulnerable or potentially threatened	1. Cox's gudgeon *(Gobiomorphus coxii)* 2. Striped gudgeon *(Gobiomorphus australis)* 3. Freshwater hardyhead *(Craterocephalus stercusmuscarum)* 4. Lake Eyre hardyhead *(Craterocephalus eyresii)* 5. River blackfish *(Gadopsis marmoratus)* 6. Flat headed galaxias *(Galaxias rostratus)* 7. Mountain galaxias *(Galaxias olidus)*

**Potential aquaculture species
Sources: Cadwallader,[16] OCE(1988), Koehn & Morison(1990)

Flooding is essential for natural spawning of silver perch.[19] The eggs are 2-8 mm in diameter and pelagic.[42] Spawning occurs in summer (November January) when sexually mature fish migrate upstream to spawn in warm, shallow waters.[15,50,66] Males become mature in their second year (233 mm) and the females mature in their third years (340 mm).[18,51] Fish often die after spawning.[18] The pre-spawning activities of silver perch have been discussed in Merrick and Schmida.[51] The fecundity is high and the number of eggs per female varies from 300 000[50] to 500 000.[93] Hatching occurs after about 30 hours at temperatures of 22–31°C.[18]

The young fish (larvae) start feeding on the sixth day, mainly on zooplankton (copepods and cladocerans).[18,71] Adult fish are omnivorous and at times may feed extensively on zooplankton, particularly the larger ostracods and cladocerans. The additional food includes shrimps (*Macrobrachium* spp., atyids), yabbies, chironomid larvae, aquatic insects, earthworms; molluscs, filamentous algae, and aquatic plants.[18,43,51,73] Some consider the silver perch to be the only species in Australia with tremendous potential for aquaculture,[4] and it is one of the four fish in the MurrayDarling River system[18] that are much sought after by commercial and recreational fishermen. Several characteristics make silver perch an ideal species for

Table 2. Rating of native fish and crayfish based on aquaculture characteristics.* Score (1-10) for each characteristic.

Characters	Silver Perch	Golden Perch	MurrayCod	FW Catfish	Yabby	Redclaw	Marron
Can be cultured legally	8	8	8	10	9	9	9
Biology, feeding, breeding behavior & distribution known	9	7	5	9	9	9	9
Perceived market value	7	9	9	5	6	8	8
Possibility of captive breeding	8	8	8	9	9	9	9
Withstand high population densities in artificial situations	8	7	5	8	9	7	7
Utilize wide range of readily available cheap sources of nutrients	9	5	7	9	9	9	9
Taste very well	7	8	9	9			
Grow to marketable size in less than a year	9	8	8	9	6	6	6
Attractive appearance	6	7	8	5	7	7	7
Good processing and storage characteristics	8	8	8	8			
Flourish in moderate or slow water flow rates	8	8	8	8			
Flourish in moderate levels of dissolved oxygen	7	7	7	9	6	5	3
Withstand moderate levels of dissolved ammonia	7	7	7	8	6	6	5
Withstand variation in pH and salinity	7	8	7	7	8	8	6
Excellent charateristics of nutrition & health; presence of omega 3 & omega 6 fatty acids	8	8	8	9			
Carcass free of bones when prepared for human consumption	8	8	8	9	9	9	9
Carcass without poisonous & unpalatable parts	10	10	10	9			
Attains early maturity	8	8	9	9	7	9	7
Closely related to species under intensive culture outside Australia	8	7	7	9	7	8	7
Less aggression and cannibalism	7	9			4	7	4

*Adapted from O'Sullivan,[57] Merrick and Lambert,[52] and Smith[84]

aquaculture: rapid and uniform growth crowded conditions;[12,64] ability to ize both plant proteins and meat meal ectively,[4] low production cost; high undity; ready acceptance of low-pro- in diet,[12,78,88,89] good growth in then ponds,[81,82] tolerance to high mperatures,[64] omnivorous feeding bit,[80] schooling characteristics of ju- niles and adults,[80] and small head [57] and high market demand to cul- e in Australia and nearby Asia.[24] The in reasons contributing to the decline ilver perch populations in its natural ge are: a) competition for food from troduced cyprinids, especially *rinus carpio*, predation by the Eng- perch, *Perca fluviatilis*, and the sical barrier to upstream migration ated by the construction of dams, ch also altered the flow and thermal mes of the rivers.[18] About three million fry are produced annually to stock farm dams and natural waters used for recreational fishing.[82] The current stocking densities in earthen ponds is 10 000 fingerling/ha managed with aeration and 5000 fingerlings/ha without aeration.[87] Silver perch farming is the major native fish culture industry in the country and has the potential of achieving an annual production of 10 000 tonnes/ha.[88] Silver perch aquaculture is concentrated in NSW where about 17 tonnes was produced in 1994/1995. Most farms are small (1–3 ponds) although there are some medium sized farms (4–8 ponds) and a few large farms (6–17 hectares).[91]

Golden perch

Golden perch *(Macquaria ambigua)* is highly prized by anglers. It occurs naturally throughout most of the Murray-Darling river system, except streams at elevations above 600m,[35] and is long-lived—up to 20 years.[13] The species can withstand salinities upto 33 ppt.[11] It tends to remain under cover most of the time (rocks, logs, stumps), moving out quickly to take passing prey. They are particularly active at night.[64]

Golden perch are carnivorous and consume insects, small fish and yabbies. The fry and fingerlings feed on zooplankton.[37] Eggs are pelagic and spawning is triggered by a rise in water level and temperature above 23°C.[41] Golden perch are reared by private and government hatcheries in Victoria, New South Wales and Queensland,[64] and the fry and fingerlings produced from these hatcheries are stocked in public waters and farm dams.

Table 3. Native aquaculture industry in Australia.

Species	Number of Farms		State	Total Number of Farms		Farm Production		Value ($000)	
	Registered	Productive		1988/89	1991/92	1988/89	1991/92	1988/89	1991/92
Freshwater Native Fish				55	231	10	43.9	173	4 345
Silver perch	42	5	VIC						
	73	5	NSW						
	<10		SA						
	39		QLD						
Total silver perch							26.6		
Golden perch	33	1	NSW						
	0		SA				6.1		
	21		QLD						
Total golden perch									
Murray Cod	8		QLD				11.0		
FW catfish	5		QLD						
Freshwater Crayfish				283	>400	>47.0	171.8	>764	2 235
Yabby	67	14	NSW						
	100, 150		VIC						
	300, 480		SA						
Total yabby						33.41			
Redclaw	37	4	NSW						
	90, 113		QLD						
	80,50		QLD						
Total redclaw							39.90		
Marron	19	1	NSW						
	150		SA						
	10		QLD						
Total marron									

Information obtained from various sources

Murray cod

The Murray cod *(Maccullochella peeli)* is one of the largest freshwater fish in the world. The largest cod ever caught weighed 113.5 kg (1.8m) and was a 1.8 m in length. Specimens of 20–40 kg are regularly captured by fishermen. They are found naturally in most of the MurrayDarling River system (Figure 1) and prefer habitats containing rocks, timber, stumps, clay banks or overhanging vegetation. Sexual maturity is reached at the age of four.[16,41] The spawning season extends from October to December[45] and spawning occurs at 20°C provided there is a slight run off of water into the pond. The eggs are demersal and adhesive and are laid in hollow logs or other substrates.[41,42,72] The species spawns naturally in earthen ponds[69].

The Murray cod is a carnivore and feeds on invertebrate and vertebrate organisms including yabbies, shrimp, crayfish, native and introduced fish (carps, redfin), and occasionally amphibians and reptiles.[35] The larvae feed on zooplankton (copepods and cladocerans), blood worms and aquatic insects[72]. The decline of Murray cod from the natural habitats appears to be due to construction of dams and weirs, overfishing, predation by introduced fish redfin (*Preca fluviatilis*), and siltation.

Freshwater catfish

Tandanus tandanus is distributed throughout most of the Murray Darling River system and in the streams of the east coast from Sydney to Rockhampton.[44] Adult catfish are carnivorous bottom feeders, and their main food is invertebrates, including molluscs and crustaceans.[20,40] Spawning season extends from October to December.[43] A temperature of 25°C or above is the primary stimulus for spawning and, unlike other native fish, flooding is not required for spawning.[41] Each female can produce from 2000 to 20 600 eggs.[19] Male and female catfish mature at the age of five.[19] The species makes a gravel nest of about 0.7 m in diameter and lays demersal eggs whcih the male then guards.[41,92] Catfish in farm dams grow at a faster rate than in rivers due to the more abundant food supply.[19]

Yabby

Yabbies *(Cherax destructor)* are distributed throughout central and southern inland Australia and are farmed commercially in Victoria, Western Australia, South Australia, and New South

Wales.[56] There are five distinct varieties of yabby,[10] and all inhabit turbid, slowflowing shallow waters.[35] Juveniles are often associated with macrophytes. During drought, the yabby survives by burrowing into damp soil and remaining there until the next rain.[22,32] Maximum growth can be achieved around 28°C.[53,54] Crowded conditions and low temperatures can seriously affect the growth rate. Cannibalism is particularly common during molting. Local markets prefer animals 30-45g (60 mm carapace length) whereas overseas markets (Hong Kong, Japan, Korea) prefer larger individuals (more than 40g).[35] Yabby are currently produced in shallow ponds, and dams in South Australia are comonly used for yabby culture, from where production figure of 50 kg/dam has been reported.[9] A backyard yabby hatchery can be set up with simple equipment such as heater, aerators, some flurotubes with programmable timer, a good water supply, and some shelter.[47]

Redclaw

Redclaw (*Cherax quadricarinatus*) is the largest freshwater crayfish in the world.[55] They are restricted to rivers of northern Australia[27] in habitats with abundant vegetation, and can be found in shallow, clear, fast-flowing creeks and slow-moving, deep and turbid waters. Hutchinson[29] reported twelve different strains of redclaw from a variety of habitats in Queensland.

Redclaw spawn throughout the year with low seasons in May and June. Higher water temperatures (>20°C) and long daylength tend to trigger spawning. Zooplankton and detritus are the main food of larval redclaw[35] while adults are detritivores.[33] Both survival and growth are influenced by temperature, and the best growth can be achieved at 28°C[33] when the dissolved oxygen level is above 5 mg/L.[35] The species can tolerate salinity up to 12 ppt.[33] For backyard redclaw culture, a series of small swimming pools, a source of clean water, aeration and a supply of crayfish are all that is required.[30]

The species is attractive in color and form, tolerant of environmental extremes, and fast-growing on inexpensive materials. They are non-aggressive compared to other species and are easy to breed in captivity, and for these reasons are considered the best of the three Australian crayfish species (yabby, marron and redclaw) for culture.[33,62] The redclaw is being farmed mainly in the sub-tropical southeast Queensland[38] and Northern Territory[46] in earthen ponds ranging in sizes from 500 m to 1 ha and 1-2 m deep.[35] Commercial aquaculture of this species commenced commenced in 1985,[35] and annual production in 1992 was 50 tonnes from 80 farms.[7] The animal is sold mainly live to restaurants, and most of the production is consumed locally (80%) and the rest is exported.[35]

Marron

Marron *(Cherax.tenuimanus)* inhabit fresh to brackish (salinity less than 6-8 ppt) river systems of south-west of Western Australia. The ideal temperature for survival and growth of marron in backyard pools or dams appears to be in the range of 13–24°C.[63] Marron grow slowly, and can not tolerate high temperature.[36] This species has been introduced to the United States, Zimbabwe, South Africa, China and Japan for experimental culture.[55] Marron have been cultured commercially in earthen ponds in Western Australia for 20 years.[35] Other methods being investigated include intensive rearing of juveniles in individual compartments housed within a larger tank ("battery culture").[63]

Culturing species endemic to Australia can reduce potential competition and pressure from overseas producers[24]. In addition, culturing native animals can help reverse the decline in populations of native species, significantly increase fisheries production, and help to restore degraded fisheries. Sale of cultured fish can help meet the local demand for live fish and reduce reliance on imports of aquatic foods. Apart from their inherent conservation and commercial values, native fish species also serve as an indicator of environmental quality of inland waters. Stock enhancement of native fish species will satisfy the recreational fishermen. There is also a growing demand for native species such as silver perch and redclaw in local and overseas markets, which demonstrates the potential of the freshwater aquaculture industry. Silver perch have feeding habits similar to the common carp[17] and could be an ideal culture species for Asian ponds. China and Taiwan have already shown an interest for the species.[24] Experimental culture of Australian crayfish is being carried out in several countries including Zimbabwe, U.S.A., and Japan. Research on the environmental impacts of native fish and crayfish aquaculture is essential as discharge of phosphorus and nitrogen may cause eutrophication or water pollution in receiving waters. Current aquaculture is based on monoculture, but polyculture of native fish and crayfish is an important area for future research. Brummett and Alon[14] have cultured Australian redclaw with nile tilapia (*Oreochromis niloticus*), and the results are promising. It is important to realize that Australian native freshwater fish and crustacean species have enormous significance in the context of conservation and aquatic food production.

Notes and References

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3. **Allan GL**, Rowland SJ, 1992. Austasia Aquaculture, 6:39–40.
4. **Allan GL**, Rowland SJ, 1996. Austasia Aquaculture 10(2):37–40.
5. **Allan GL**, WarnerSmith RJ, Frances J, 1994. Proceeding Nutrition Society of Australia 18:53.
6. **Allen G**, 1989. TFH (Australia) Pty Ltd, NSW.
7. **Anon**, 1992. Austasia Aquaculture 5:22.
8. **Anon**, 1994. Yabby marketing cooperative for state, South Australia. Austasia
9. **Austasia Aquaculture**, 1990. Austasia Aquaculture 5(1):23–24.
10. **Austasia Aquaculture Magazine**, 1990. Austasia Aquaculture Magazine 4(5):12.
11. **Backhouse G**, Cadwalladar P, Raddik T, 1991. Freshwater fishes of Victoria-Golden perch. Department of conservation and Environment Victoria, infosheet 51D:\INFO\FOVGOLDP\DOC\91.
12. **Barlow CG**, 1986. Pages 33-43 *in* Proceedings of the First Freshwater Aquaculture Workshop, Narrendera (L F Reynolds ed). NSW Agriculture and Fisheries, Sydney, Australia.
13. **Battaglene S**, Prokop F, 1987. Department of Agriculture, New South Wales, Agfact A3.2.2. 7p.
14. **Brummett RE**, Alon NC, 1994. Aquaculture 122:47–54.
15. **Cadwallader PL**, 1977. JO. Langtry's 1949–50 Murray River investigations. Fisheries and Wildlife Paper, Victoria. No. 13. 70pp.
16. **Cadwallader PL**, 1978. Proceeding Royal Society of Victoria 90:211–224.
17. **Cadwallader PL**, 1979. Australian Journal of Ecology 4:361–385.
18. **Cadwallader PL**, Backhouse, GN 1983. Government Printer, Melbourne. 249p.
19. **Davis TLO**, 1977. Australian Journal of Marine and Freshwater Research 28:159–169.

20. Davis TLO, 1977. Australian Journal of Marine and Freshwater Research 28:455–465.

21. Fallu R, 1995. Department of Agriculture, Victoria. 48p.

22. Frost JV, 1975. Pages 87-95 *in* Freshwater crayfish, Volume 2, papers from the Second International Symposium on Freshwater Crayfish, 1974 (JW Avault Jr., ed). Louisiana State University, Baton Rouge, Louisiana, USA.

23. Gehrke PC, Revell MB, Philbey AW, 1993. Journal of Fish Biology, 43:265279.

24. Gooley G, Rowland S, 1993. Austasia Aquaculture, 7(3):35–37.

25. Gooley G, Anderson TA, 1992. Pages 45-47 *in*: Proceedings of the Aquaculture Nutrition Workshop, Salamander Bay, 15–17 April 1991, (G L Allan, W Dall eds). NSW Fisheries, Brackish Water Fish Culture Research Station, Salamander Bay, Australia.

26. Grant E, 1972. Department of Primary Industries, Brisbane Australia.

27. Guo R, Mather PB, Capra MF, 1995. Marine and Freshwater Research 46:947–952.

28. Hume D, Barl ow C, 1993. . Department of Primary Industries, Queensland Government 1993. Brisbane.

29. Hutchinson R, 1987. Austasia Aquaculture Magazine 1(12):12–13.

30. Hutchinson R, 1989. Austasia Aquaculture Magazine 3(8):12.

31. Ingram B, Gooley G, McKinnon L, 1996. Austasia Aquaculture, 10(2):61–63.

32. Johnson HT, Mills B, Morrissy NM, 1988. *In:* Crayfish Farming, (G Ryder ed). Sydney, NSW Department of Agriculture and Fisheries.

33. Jones CM, 1990. The biology and aquaculture potential of the tropical freshwater crayfish, *Cherax quadricarinatus*. Brisbane, Queensland Department of Primary Industries. 130pp.

34. Jones CM, 1990. Australian Fisheries 49(11):3–7.

35. Kailola PJ, Williams MJ, Stewart PC, Reichelt RE, McNee A, Grieve C, 1993. Australian Fisheries Resources. Commonwealth of Australia. Bureau of Resource Sciences and the Fisheries Research and Development Corporation. 422p.

36. Kartamulia L, Rouse DB, 1992. Journal of the World Aquaculture Society 23(2):169–172.

37. Kiley T, 1996. Austasia Aquaculture 10(3):55.

38. King CR. 1994, Aquaculture 122:75–80.

39. Kumar JM, 1993. Austasia Aquaculture, 7(1):32–33.

40. Lake JS, 1959. Research Bulletin of Fisheries. NSW. 5.

41. Lake J, 1967. Australian Journal of Marine and Freshwater Research 18:137–153.

42. Lake JS, 1967. Australian Journal of Marine and Freshwater Research 18:155–173

43. Lake JS, 1967, *In* Australian Inland Waters and their Faunas (AHWeatherley, ed). Australian National University Press, Canberra.

44. Lake JS, 1971. Freshwater fishes and rivers of Australia, Nelson, Sydney.61pp.

45. Llwellyn L, McDonald MC, 1980. Pages 142-149 *in* Freshwater Fishes of SouthEastern Australia (RM McDowall ed) Reed, Sydney.

46. LoyaJavellana GN, Fielder DR, Thorne MJ, 1993. Aquaculture 118:299–308.

47. Lunnay J, 1988. Austasia Aquaculture Magazine 3(4):9 & 15.

48. Malcolm B, 1986.Pages 121-124 *in* Aquaculture in Australia—Fish Farming for Profit.Rural Press, Queensland Pvt Ltd..

49. Mather G, 1993. AquacultureAn Australian Environmental Review. Total Environment Centre Inc. Sydney.88p.

50. Merrick JR, 1980. Family Teraponidae. Pages 150-152 *in* Freshwater Fishes of SouthEastern Australia (RM McDowell ed). Reed, Sydney.

51. Merrick JR, Schmida GE, 1984. Australian freshwater fishes–biology and management. Griffin Press Ltd, South Australia.

52. Merrick JR, Lambert CN, 1991. The yabby, marron and redclaw: production and marketing p.180, Macarthur Press Pty. Limited., NSW.

53. Mills BJ, McCloud PL. 1983, Aquaculture 34:51–72.

54. Morrissy NM, 1990. Australian Journal of Marine and Freshwater Research 41:735–746.

55. Morrissy NM, Evans LE, Huner JV, 1990. World Aquaculture 21(2):113–122.

56. O'Connor PN. Rayns ND, 1993. Aquaculture prospects for freshwater crayfish. NSW Fisheries Fishnote. Fishnote DF/10.

57. O'Sullivan D, 1990. Pages 43-104 *in:* Aquaculture in Asia. (Joseph Mohan, editor), p. 43-104. Asian Fisheries Society.

58. O'Sullivan D, 1991. Status of Australian aquaculture in 1989–90. Austasia Aquaculture, June, 213.

59. O'Sullivan D, 1994. Austasia Aquaculture 8(2):44–45.

60. O'Sullivan D, 1994. Aquaculture source book no. 10. Turtle Press, Tasmania.

61. O'Sullivan D, 1995. Pages 5-8 *in* Silver perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrandera, April, 1994, (SJ Rowland and C Bryant eds). Austasia Aquaculture, Hobart for NSW Fisheries.

62. Pinto GF, Rouse DB, 1996. Journal of the World Aquaculture Society 27(2):187–193.

63. Platt J, 1989. Austasia Aquaculture Magazine 3(6):10–11.

64. Pollard J, 1986. Fishing Australia. The Book Company International Pty Ltd, Sydney.

65. Pollard DA, Ingram BA, Harris JH, Reynolds LF, 1990. Journal .Fish Biology. 37(supp. A):67–78.

66. Reynolds LF, 1983. Australian Journal of Marine and Freshwater Research 34:857871.

67. Riek EF, 1969. Australian Journal of Zoology 17:855–918.

68. Ross L, TaylorMoore N, Gillespie J, 1994. Chapter 13. Pages 157-170 *in* Queensland Year Book 1994. Australian Bureau of Statistics.

69. Rowland SJ, 1983. Journal of Fish Biology 23:525–534.

70. Rowland SJ, 1984. Aquaculture 42:83–86.

71. Rowland SJ, 1988. Aquaculture 70:371–389.

72. Rowland SJ, 1992. Proceedings of the Linnean Society of New South Wales 113(3):193–201.

73. Rowland SJ, 1994. Journal of Applied Aquaculture, 4(1):39–48.

74. Rowland SJ, 1995. Pages 9-11 *In* Silver perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrandera, April, 1994. (S J Rowland and C Bryant eds). Austasia Aquaculture, Hobart NSW, Australia.

75. Rowland SJ, 1995. Pages 51-65 *In* Silver perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrandera, April, 1994 (SJ Rowland, C Bryant, eds). Austasia Aquaculture, Hobart NSW, Australia.

76. Rowland SJ, 1995.Pages 41-49 *In* Silver perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrandera, April, 1994 (SJ Rowland, C Bryant, eds). Austasia Aquaculture, Hobart NSW, Australia

77. Rowland SJ, 1996. Environmental Biology of Fishes 46:350.

78. Rowland SJ, Barlow CG, 1991. Austasia Aquaculture, 5:27–30.

79. Rowland SJ, Bryant C (editors), 1995. Pages 41–49 *in* Austasia Aquaculture, Hobart for NSW Fisheries.

80. Rowland SJ, Kearney RE, 1992. The prospects for the commercial aquaculture of silverperch (*Bidyanus bidyanus*). A research project arising from a review of freshwater aquaculture. NSW Fisheries, 16p.

81. Rowland SJ, Allan GL, Clark K, Hollis M. Pontifex T, 1994. The Progressive Fish Culturist, 56:296–298.

82. Rowland SJ, Allan GL, Hollis M, Pontifex T, 1995. Aquaculture 130:317–328.

83. OCE. 1988. State of the Environment Report, 1988. Victoria's Inland Waters'. Office of the Commissioner for the Environment), Melbourne. 655p.

84. Smith T, 1993. Austasia Aquaculture 7(6):38–40.

85. Starling S, 1992. The Australian Fishing Book. Sandipiper Press(NSW) Ltd, 512p.

86. Treadwell R, McKelvie L, Maguire GB, 1992. Potential of Australian Aquaculture. Australian Bureau of Agricultural and Resources Economics(ABARE) Research Report 92.2. Commonwealth of Australia.Canberra .81p.

87. WalkerT, 1993. Austasia Aquaculture, 7(5):3–6.

88. Walker T, 1994. Austasia Aquaculture 8(5):3–6.

89. Walker T, 1994. Austasia Aquaculture 8(5):24–36.

90. Walker T, 1995. Austasia Aquaculture 9(1):3–4.

91. Walker T and Caney D, 1996. Austasia Aquaculture 10(2):34–36.

92. Whitely GP, 1941. Australian Museum Magazine 7(9):306–313.

93. Whitely GP, 1960. Native freshwater fishes of Australia. Jacandra Press: Brisbane. 127p.

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Date: Fri, 11 Jul 1997 16:26:00 -0700 (PDT)
From: Natalie Macawaris <N.MACAWARIS@CGNET.COM>
To: Golam Kibria <s9337499@cougar.vut.EDU.AU>
Subject: MS for Naga

Dear Mr Kibria,

Greetings from ICLARM. This has reference to your article "Zooplankton: its biochemistry..." to appear in the Naga. The magazine has been layouted however the editors have noted a few more questions:

1] In the Tables, the legend for P and Ca are lacking. Do these correspond with Phosphorus (or Potassium) and Calcium respectively?

2] Please check accuracy of spelling in the following species:

Daphnia obtisa (or obtusa?)
Cyclops vicinis (or vicinus?)
Date: Fri, 11 Jul 1997 16:26:00 -0700 (PDT)sktop printer...]
From: Natalie Macawaris <N.MACAWARIS@CGNET.COM>
To: Golam Kibria <s9337499@cougar.vut.EDU.AU>
Subject: MS for Naga

Date: Fri, 07 Nov 1997 11:42:00 -0800 (PST)
From: Natalie Macawaris <N.MACAWARIS@CGNET.COM>
To: Golam Kibria <s9337499@cougar.vut.edu.au>
Subject: Urgent: query on Article for Aquabyte/Naga

Dear Mr Kibria,

My apologies for overlooking your query (email dated 17 September 1997) on the Naga issue number in which your article on zooplankton will be published. The issue of Naga where your ms is featured is still in press (indeed we have experienced much difficulties in the release of this issue owing to the crash of the computer files and we had to reconstruct everything!). However, we can now give you the bibliographic entry as follows: <authors> 1997. Zooplankton: its biochemistry and significance in aquaculture. Naga, The ICLARM Quarterly 20 (2): 8-15.

Aquaculture Research, 1998, 29, 000–000

Effect of temperature on phosphorus losses and phosphorus retention in silver perch, *Bidyanus bidyanus* (Mitchell 1838), (Teraponidae) fed on artificial diets

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Abstract

Silver perch, *Bidyanus bidyanus* (Mitchell 1838), were reared in glass aquaria and fed on three diets containing 53%, 45% and 36% protein, and 1.31%, 1.16% and 1.28% phosphorus, respectively, in order to investigate the phosphorus losses and phosphorus retention at 25 and 30°C. The main path of phosphorus loss was found to be via faeces and was significantly higher at 30°C ($P < 0.05$). There was a sharp increase in orthophosphate excretion soon after meals, which decreased linearly during the remaining 24 h. The daily orthophosphate output was observed to increase at the higher temperature and was significantly higher at 30°C compared to 25°C ($P < 0.05$). Phosphorus retention by silver perch was significantly better at 25°C than at 30°C ($P < 0.05$).

Introduction

Fish require phosphorus for optimum growth, feed efficiency, bone development, maintenance of acid-base regulation, and lipid and carbohydrate metabolism (Lovell 1978; Ogino & Takeda 1978; Lall 1991). Diets deficient in phosphorus can suppress the appetite and may lead to the death of fish (Lall 1979). Because of the low phosphorus concentration in both fresh water and sea water (Boyd 1971; Weatherly & Gill 1987), and the low rate of absorption from the water (Phillips, Podoliak, Brockway & Balzer 1956), dietary phosphorus is the main source for fish.

Dietary phosphorus supplied and unavailable to fish will be evacuated from the gut in the faeces, while phosphorus surplus to requirements will be excreted via the kidney and gills (Forster & Goldstein 1969; Nakashima & Leggett 1980). Phosphorus is primarily discharged as undigested matter though faeces (Kristiansen & Hessen 1992). The phosphorus released from uneaten food and faeces should be minimized because it stimulates algal growth or eutrophications in receiving waters (Walker & Hillmann 1982; Lall 1991). Studies conducted in Europe and North America demonstrate a strong relationship between water pollution and the discharge of phosphorus from fish farm effluents since effluents can deteriorate the water quality of

1998, 29,

the recipient water bodies (Albaster 1982; Penczak, Galicka, Molinski, Kusto & Zalewski 1982; Foy & Rosell 1991a, b; Lanari, D'Agaro & Ballestrazzi 1995). In order to minimize the environmental impact, information on absorption, metabolism and excretion of phosphorus is essential (Lall 1991). There is very limited information available on phosphorus losses and retention, and studies are needed on phosphorus retention at various stages of development (Lall 1991).

The silver perch, *Bidyanus bidyanus* (Mitchell 1838), is an Australian native fish of the highest aquaculture potential (Allan & Rowland 1996), and the industry is growing rapidly (26.6 t in 1991–1992 and 52.7 t in 1994–1995) (O'Sullivan 1995; O'Sullivan & Kiley 1996) in the country because of interest and investments in culturing the species (Kibria, Nugegoda, Fairclough & Lam 1996). Although there is some concern on the environmental impacts of aquaculture, until now there has been no research into either the quantity or quality of phosphorus that may be discharged from aquaculture of silver perch. The objectives of the present study were to evaluate the effects of temperatures and feeding on phosphorus losses and phosphorus retention by silver perch.

Materials and methods

The experiment was conducted in the wet laboratory at the Victoria University, Melbourne, Victoria, Australia. Juveniles of silver perch of similar size (average weight 928 mg) were bought from a local native fish farm where fish were grown in earthen ponds. The animals were acclimatized in the laboratory in large holding tanks for 4 weeks prior to the start of feeding trials. The fish were reared in small glass aquaria (30 × 16 × 17 cm) at two temperatures, 25 and 30°C, since the optimal temperature for silver perch is believed to be in the range of 23–27°C (Rowland, Allan Hollis & Pontifex 1995). Fish were fed on three commercial silver perch diets (silver crumbles/starter) at the rate of 3% of body weight (as recommended), twice a day (0900 and 1600 h), 6 days a week for 4 weeks, to study phosphorus losses and phosphorus retention in silver perch. The three diets, referred to as diet 1, diet 2 and diet 3, contained 53%, 45% and 36% protein and 1.31%, 1.16% and 1.28% phosphorus, respectively (Table 1). Four fish were stocked in each glass aquarium and each diet was replicated in six aquaria, i.e. 24 fish per diet per temperature. Individually, aquaria were fitted with air stones to enhance dissolved oxygen content. The water quality was maintained at pH 7.5–8.0, dissolved oxygen level > 6 mg L^{-1} and hardness 80–100 mg L^{-1}. The pH was measured with a bench-top pH meter (Orion model SA520 Orion), dissolved oxygen by using a dissolved oxygen meter (YSI model 58, YSI) and hardness with a test kit (Aquasonic, NSW, Australia). The required water temperature was obtained by heating water with a thermostatically controlled heater in a large tank (70 × 60 × 30 cm) in which small aquaria were placed. The large tank was covered with a glass lid to reduce evaporation and to maintain the required specific temperature. Each aquarium was siphoned a number of times every 24 h to collect faeces. Collected faeces were first centrifuged at 2000 r.p.m. for 10 min (Econospin-Sorvall Instruments), the supernatant was discarded and the precipitate was dried in an oven (Law 1984; Hajen, Beames, Higgs, & Dosanjh 1993). Water samples were collected each morning using a plastic syringe to record the daily variations in ortho-phosphate concentrations with respect to temperature and diet fed to fish. Data were also collected on the hourly pattern of phosphate

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Table 1 Proximate composition, gross energy, phosphorus content and digestible energy ratio of experimental diets

	Diet 1	Diet 2	Diet 3
Dry matter (DM; %)	91.80	90.12	88.99
Crude protein (% DM)	53.00	45.00	36.00
Ether extract (% DM)	6.92	8.50	4.96
Ash (% DM)	13.06	9.84	13.00
N-free extract (% DM)[1]	27.02	36.66	46.04
N (% DM)	08.48	07.20	05.40
P (% DM)	1.31	1.16	1.28
Gross energy (MJ kg^{-1} DM)[2]	19.89	20.28	18.40
Digestible energy (MJ kg^{-1} DM)[3]	17.24	17.34	15.39
DP:DE (g MJ^{-1} DM)	30.74	25.95	23.39

[1]The Nitrogen free extract = 100 – crude protein – ether extract – ash.

[2]Gross energy contents of the diets were estimated by using values of 0.0236 MJ g $protein^{-1}$, 0.0395 MJ g $lipid^{-1}$ and 0.0172 MJ g $carbohydrate^{-1}$ (NFE).

[3]The digestible energy contents of the diets were estimated by using values of 0.02213 MJ g $protein^{-1}$ (assuming 90% protein is digestible), 0.0356 MJ g $lipid^{-1}$ (assuming 90% lipid is digestible) and 0.0129 MJ g $carbohydrate^{-1}$ (assuming 75% is digestible).

excretion for the three consecutive days of fish fed three diets at 25°C. Food conversion ratio (FCR) was calculated by the amount of food fed/fish weight gained. The FCR was found to be significantly better in fish reared at 25°C than in fish reared at 30°C ($P < 0.05$). The FCR calculated at 25 and 30°C was 1.09 and 1.66, respectively.

Samples were weighed using an analytical balance (Mettler AE 200). Proximate analysis was done following AOAC (1990): protein was analysed by the Kjeldahl method (N × 6.25), fat by ether extract in a soxhlet apparatus (AOAC 1990), carbohydrate by difference, moisture by drying in an oven at 105°C for 24 h, and ash by burning samples in a muffle furnace at 600°C overnight. Phosphorus content in dried food, fish and faeces was determined following AOAC (1990). The phosphorus retained in the fish carcass was determined by subtracting initial carcass phosphorus content from the final carcass phosphorus content. Orthophosphate in water was determined with a Tecator flow injection analyser (Aquatec 5400 Analyser) following the Aquatec instruction manual (Tecator 1990).

Mean, standard deviation and standard error of phosphorus loss and phosphorus gain per diet were calculated following Zar (1984). All percentage data were transformed to arc sin values prior to analysis (Systat Programme 5.0) using an IBM computer. One-way analyses of variance (ANOVA) were used to compare the means of phosphorus loss and gain using an IBM-compatible Microsoft Excel programme and setting the significance level at $P < 0.05$.

Results and discussion

Hourly patterns of phosphate production

The hourly patterns of orthophosphate excretion are presented in Fig. 1. Figure 1 shows that there was a rapid increase in phosphate level soon after the meal, which decreased linearly during the remaining 24 h. There was no second peak following the second meal. However, the excretion rate was not significantly different between the three diets fed to silver perch containing 53%, 45% and 36% protein ($P > 0.05$). Ballestrazzi, Lanari, D'Agaro & Moin (1994) also did not find any significant differences in orthophosphate excretion in fish fed on three diets containing 41%, 48.6% and 53.6% protein. The peak of excretion in silver perch was observed soon after the first meal and a similar trend was found in sea bass by Ballestrazzi *et al.* (1994).

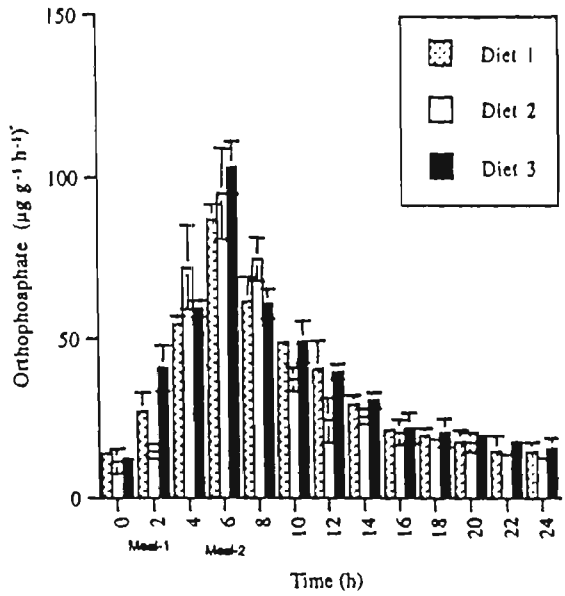

Figure 1 Postprandial patterns of orthophosphate excretion by silver perch juveniles fed three diets and reared at 25°C. There were no significant differences in phosphate excretion of the three diets fed to fish ($P > 0.05$). The protein and phosphorus content of the three diets are, respectively: (1) 53% and 1.33%; (2) 45% and 1.16%; and (3) 36% and 1.28% (mean ± SEM; $n = 3$).

Similarly, there was a rapid increase in orthophosphate concentration soon after the feeding of trout, which decreased and reached nonfeeding levels within 6 h (Solberg & Bregnballe 1982). Peaks of orthophosphate excretion are thought to be related to feeding and activity of fish (Hennessy, Wilson, Struthers & Kelly 1996), and are also dependent on the quantity and quality of food supplied (Lall 1991). These facts also reflected in the present study.

Day-to-day variation of phosphate excretion

Out of the three diets fed to fish, diet 2, containing 45% protein, excreted comparatively less orthophosphate than either the 53% and 36% protein diets at both temperatures. This could be a result of the slightly lower phosphorus content in diet 2, although there were no significant differences in phosphate discharges between the three diets fed to fish ($P > 0.05$) (Fig. 2). Nevertheless, fishmeal is the only source of protein for the three diets which might have caused non-significant orthophosphate excretion of the three diets tested. Ballestrazzi *et al.* (1994) found that a diet with lower phosphorus content also resulted in lower phosphorus excretion

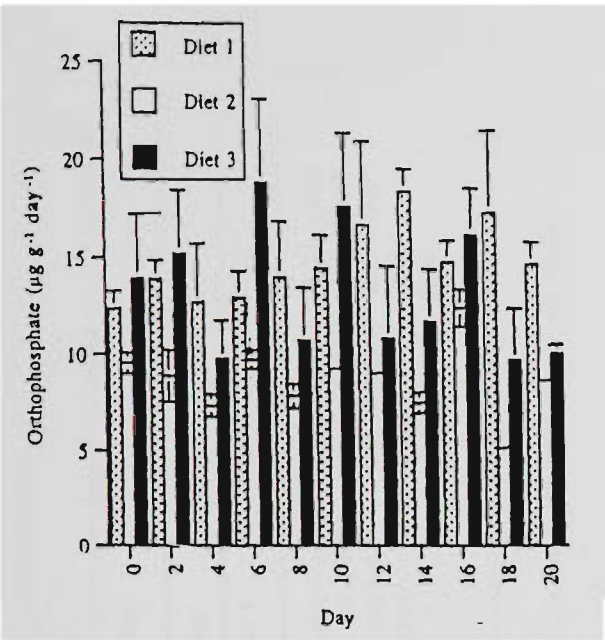

Figure 2 Day-to-day variation in orthophosphate excretion by silver perch juveniles fed three diets and reared at 25°C. There were no significant differences in the phosphate excretion of the three diets fed to fish at 25°C ($P > 0.05$) (mean ± SEM; $n = 6$).

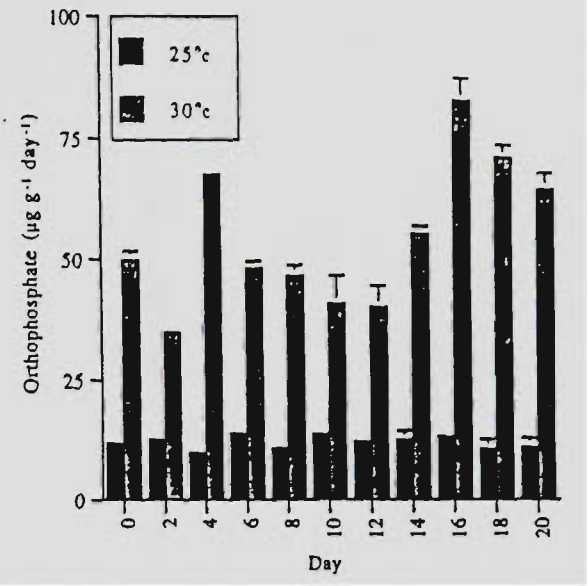

Figure 3 Day-to-day variation in orthophosphate excretion by silver perch juveniles reared at 25 and 30°C. Phosphate excretion was significantly higher at 30°C ($P < 0.05$) (mean ± SEM; $n = 18$).

and they also did not find any significant differences in orthophosphate excretion in seabass fed on herring meal based diets. Moreover, the phosphorus excretion rate was reported to be affected by the type of diet (Ketola, Westers, Houghton & Pecor 1991; Kristiansen & Hessen 1992) and the source of protein (Ballaestrazzi *et al.* 1994), phosphorus (Ketola & Harland 1993). For example, corn gluten diets significantly reduced orthophosphate excretion in seabass in comparison to herring meal diets and this could be a result of less phosphorus in gluten than fishmeal (Ballaestrazzi *et al.* 1994). Similarly, deflourinated rock phosphate decreased phosphorus discharges in rainbow trout by about 40–51% when compared to other phosphorus sources (Ketola & Harland 1993). Because diet 2 was an extruded diet, this may have caused better digestibility and a lower phosphorus excretion in silver perch since extruded diet is known to be more digestible, and to generate less solid and soluble waste discharge (Warren-Hansen 1982; Matty 1990).

Silver perch reared at 25°C showed less orthophosphate excretion than fish reared at 30°C and the excretion rate was significantly higher at 30°C ($P < 0.05$) (Fig. 3). Kristiansen & Hessen (1992) reported an increase in orthophosphate excretion of noble crayfish, *Astacus astacus*, reared at higher temperature, and Savitz (1971) observed an elevated phosphorus excretion in bluegill sunfish, *Lepomis macrocephalus*, reared at higher temperature. The FCR is reported to play a significant role in determining the level of phosphorus load expected, since an increase in FCR value from 1.0 to 1.5 may increase phosphorus load to about 86% for total phosphorus (Stroebakken & Austreng 1987a, b). The FCR for silver perch was significantly better at 25°C, which may be related to the significantly lower orthophosphate excretion observed at 25°C ($P < 0.05$). In addition to the digestibility of feed and the nutrient content in diet, the feeding technique may influence the feed coefficient and phosphorus load from aquaculture (Enell 1995).

authority?

Phosphorus retention

Phosphorus retention in silver perch was significantly greater at 25°C than at 30°C ($P < 0.05$), which may be related to the optimum growing temperature of silver perch at 25°C. The average phosphorus retained at 25°C was 49% and at 30°C it was 24.5% (Table 2.). The significantly higher phosphorus retention obtained with silver perch at 250°C may have been a result of the better FCR achieved at 25°C. Moreover, the higher phosphorus retention at 25°C may have been caused by lower phosphate excretion and lower faecal

Table 2 Phosphorus retention and phosphorus losses (g kg body weight^{-1} day–1), and percentage of phosphorus retained in silver perch fingerlings reared at 25 and 30°C for 28 days. Values with different superscripts in the same row are significantly different from each other ($P < 0.05$) (mean + SEM)

	25°C			30°C		
	Diet 1	Diet 2	Diet 3	Diet 1	Diet 2	Diet 3
P ingested (g kg BW^{-1} day–1)[1]	0.524 + 0.02	0.466 + 0.03	0.514 + 0.01	0.524 + 0.04	0.464 + 0.05	0.464 + 0.03
P retained (carcass) (g kg BW^{-1} day–1)[2]	0.285 + 0.02^{a}	0.223 + 0.02^{a}	0..231 + 0.04^{a}	0.143 + 0.02^{b}	0.115 + 0.06^{c}	0.10 + 0.09^{c}
Faecal phosphorus (g kg BW^{-1} day–1)	0.190 + 0.01^{a}	0.143 + 0.01^{b}	0.174 + 0.02^{a}	0.300 + 0.01^{c}	0.201 + 0.01^{d}	0.21 + 0.02^{d}
Non-faecal phosphorus (g kg BW^{-1} day–1)	0.049	0.100	0.109	0.081	0.148	0.154
P retained/P ingested (%)	54.30	48.00	45.00	27.29	24.78	21.55
Mean P retained	49.10 + 2.74^{a}	24.54 + 1.66^{b}				

[1]Theoretical phosphorus intake.

[2]Phosphorus retention = (carcass phosphorus content at the end of experiment – carcass phosphorus content at start of experiment) × 100/total phosphorus intake.

Table 3 Chemical composition and phosphorus content of the whole body at the end of feeding experiment. Values in the same row with different superscripts are significantly different (mean + SEM)

		25°C			30°C		
	Initial	Diet 1	Diet 2	Diet 3	Diet 1	Diet 2	Diet 3
Moisture (%)	80.21	78.71	78.36	78.23	79.10	79.15	79.10
Crude protein (% dry weight)	58.83	63.59 + 0.58^{a}	65.59 + 01.5^{a}	63.87 + 0.51^{a}	59.31 + 01.26^{b}	60.46 + 01.40^{b}	59.43 + 0.1^{b}
Ash (% dry weight)	16.37	18.24 + 0.19^{a}	17.43 + 0.22^{b}	18.94 + 0.04^{a}	17.16 + 0.21^{c}	16.64 + 0.26^{d}	16.01 + 0.2^{d}
Phosphorus (% dry weight)[1]	2.49	2.83 + 0.02^{a}	2.76 + 0.06^{a}	2.82 + 0.09^{a}	2.50 + 0.27^{b}	2.56 + 0.13^{b}	2.51 + 0.13^{b}

[1]The initial sample size was 34 fish.

phosphorus losses obtained at 25°C (Fig. 2, Table 2). Furthermore, the restricted feeding at 3% food ration may have an affect on comparatively lower phosphorus retention at 30°C. The reported phosphorus retention markedly varies between species: 14–22% in coho salmon, *Oncorhynchus kisutch* (Walbaum), (Ketola *et al.* 1991); 61–81% in rainbow trout, *Oncorhynchus mykiss* (Walbaum), (Ogino & Takeda 1978); 39–40% in channel catfish (Lovell 1978); 36.40% in Atlantic salmon, *Salmo salar* L., (Johnsen, Hillestad & Austreng 1993); 33% in rainbow trout (Ketola & Harland 1991); and 69–87% in sunshine bass (Brown, Jaramillo & Gatlin 1993). The other reasons for variation in phosphorus retention appear to be the result of diets, growth rates, source and levels of phosphorus fed to fish (Ketola *et al.* 1991; Lall 1991).

At 25°C, about 66.7% of the phosphorus lost was in particulate form and the remaining 33.5% was in dissolved form. Out of the total phosphorus loss from aquaculture, a major loss was reported to be in particulate form, accounting for about 77–85.7%, and a minor loss, accounting for 14.3–23%, was in dissolved form (Enell 1987; Ackefors & Enell 1990; Enell & Ackefors 1991; Johnsen *et al.* 1993). The present study also found that the main path of phosphorus loss in silver perch was via faeces.

There was a direct relationship between the feed conversion efficiency and phosphorus load to environment: the better the FCR the less the phosphorus load (Enell 1995). Matty (1990) mentioned that a good FCR is essential in reducing the phosphorus pollution in salmonids and trout. Similarly, nutrient loss rates were found to be dependent on FCR, nutrient content of the diet and the fish produced (Foy & Rosell 1991a). Therefore, a good FCR obtained at 25°C may be related to the lower phosphorus excretion and lower faecal phosphorus loss obtained in this study.

Body composition of fish

The body composition of fish varied by the rearing temperatures (able 3), and those fish reared at 25°C had significantly higher body protein, ash and phosphorus content than the fish reared at 30°C ($P < 0.05$). This could be related to the better phosphorus retention observed at 25°C and may be an effect of optimum growing temperature. The body composition of fish has been reported to be affected by the rearing temperatures (Brett, Shelbourn & Shoop 1969; Singh, Sinha & Chakraborty 1979; Windell, Foltz & Sarokon 1978), ration size (Hulsman, Breteler, Vismans & Kanis 1979), experimental diets and protein level (Ballestrazzi *et al.* 1994; Haiqing & Xiqin 1994), and fish size (Brett *et al.* 1969). Hasan, Islam & Alim (1993) found that the group of fish that showed better FCR also had higher carcass protein and ash content. In the present study, fish reared at 25°C also had a better FCR, which may have caused higher carcass protein and carcass ash content in silver perch at this temperature.

Summary and conclusion

The results show that growing of fish at an optimum temperature can enhance phosphorus retention and a reduction in the discharge of phosphorus to the environment. In order to maximize the phosphorus retention and minimize the phosphorus losses, further research is needed on the optimum phosphorus requirements of silver perch.

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References

Ackefors H. & Enell M. (1990) Discharge of nutrients from Swedish fish farming to adjacent sea areas. *Ambio* **19**, 28–35.

Albaster J.S. (1982) Report of the EIFAC Workshop on Fish-Farm Effluents, Silkeborg, Denmark, 26–28 May 1981. *FAO/EIFAC Technical Paper* **41**, 166p.

Allan G.L. & Rowland S.J. (1996) The silver perch industry: research progress and priorities toward developing cost-effective diets. *Australia Aquaculture* **10**, 37–40.

Association of Official Analytical Chemists (AOAC) (1990) *Official Methods of Analysis*, 15th edn, Vol. 1 (ed. by K. Helrich). AOAC, Arlington, VA.

Ballestrazzi R., Lanari D., D'Agaro E. & Mion A. (1994) The effect of dietary protein level and source on growth, body composition, total ammonia and reactive phosphate excretion of growing sea bass (*Dicentrachus labrax*). *Aquaculture* **127**, 197–206.

Boyd C.E. (1971) Phosphorus dynamics in ponds. *Proceedings of the Annual Conference of Southeast Association of Game and Fish Commission* **25**, 418–426.

Brett J.R., Shelbourn J.E. & Shoop C.T. (1969) Growth rate

excretion for the three consecutive days of fish fed three diets at 25°C. Food conversion ratio (FCR) was calculated by the amount of food fed/fish weight gained. The FCR was found to be significantly better in fish reared at 25°C than in fish reared at 30°C ($P < 0.05$). The FCR calculated at 25 and 30°C was 1.09 and 1.66, respectively.

Samples were weighed using an analytical balance (Mettler AE 200). Proximate analysis was done following AOAC (1990): protein was analysed by the Kjeldahl method (N × 6.25), fat by ether extract in a soxhlet apparatus (AOAC 1990), carbohydrate by difference, moisture by drying in an oven at 105°C for 24 h, and ash by burning samples in a muffle furnace at 600°C overnight. Phosphorus content in dried food, fish and faeces was determined following AOAC (1990). The phosphorus retained in the fish carcass was determined by subtracting initial carcass phosphorus content from the final carcass phosphorus content. Orthophosphate in water was determined with a Tecator flow injection analyser (Aquatec 5400 Analyser) following the Aquatec instruction manual (Tecator 1990).

Mean, standard deviation and standard error of phosphorus loss and phosphorus gain per diet were calculated following Zar (1984). All percentage data were transformed to arc sin values prior to analysis (Systat Programme 5.0) using an IBM computer. One-way analyses of variance (ANOVA) were used to compare the means of phosphorus loss and gain using an IBM-compatible Microsoft Excel programme and setting the significance level at $P < 0.05$.

Results and discussion

Hourly patterns of phosphate production

The hourly patterns of orthophosphate excretion are presented in Fig. 1. Figure 1 shows that there was a rapid increase in phosphate level soon after the meal, which decreased linearly during the remaining 24 h. There was no second peak following the second meal. However, the excretion rate was not significantly different between the three diets fed to silver perch containing 53%, 45% and 36% protein ($P > 0.05$). Ballestrazzi, Lanari, D'Agaro & Moin (1994) also did not find any significant differences in orthophosphate excretion in fish fed on three diets containing 41%, 48.6% and 53.6% protein. The peak of excretion in silver perch was observed soon after the first meal and a similar trend was found in sea bass by Ballestrazzi *et al.* (1994).

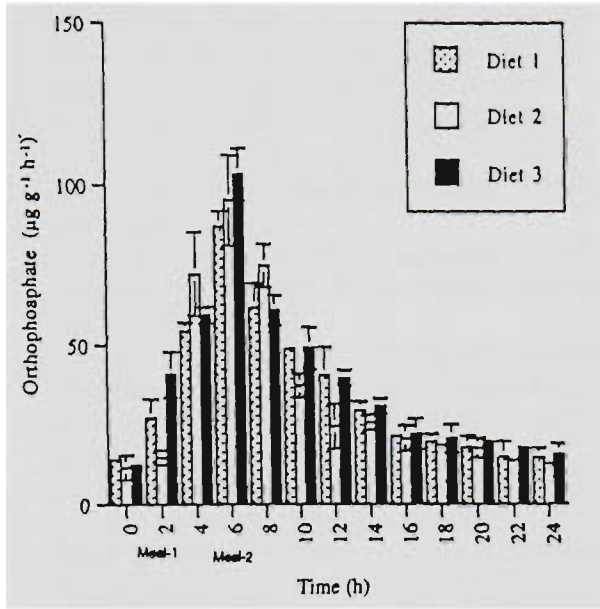

Figure 1 Postprandial patterns of orthophosphate excretion by silver perch juveniles fed three diets and reared at 25°C. There were no significant differences in phosphate excretion of the three diets fed to fish ($P > 0.05$). The protein and phosphorus content of the three diets are, respectively: (1) 53% and 1.33%; (2) 45% and 1.16%; and (3) 36% and 1.28% (mean ± SEM; $n = 3$).

Similarly, there was a rapid increase in orthophosphate concentration soon after the feeding of trout, which decreased and reached nonfeeding levels within 6 h (Solberg & Bregnballe 1982). Peaks of orthophosphate excretion are thought to be related to feeding and activity of fish (Hennessy, Wilson, Struthers & Kelly 1996), and are also dependent on the quantity and quality of food supplied (Lall 1991). These facts also reflected in the present study.

Day-to-day variation of phosphate excretion

Out of the three diets fed to fish, diet 2, containing 45% protein, excreted comparatively less orthophosphate than either the 53% and 36% protein diets at both temperatures. This could be a result of the slightly lower phosphorus content in diet 2, although there were no significant differences in phosphate discharges between the three diets fed to fish ($P > 0.05$) (Fig. 2). Nevertheless, fishmeal is the only source of protein for the three diets which might have caused non-significant orthophosphate excretion of the three diets tested. Ballestrazzi *et al.* (1994) found that a diet with lower phosphorus content also resulted in lower phosphorus excretion

and body composition of fingerlings sockeye salmon *Oncorhynchus nerka* in relation to temperature and ration size. *Journal of the Fisheries Research Board of Canada* **26**, 2363–2394.

Brown M.L., Jaramillo F., Jr & Gatlin D.M. (1993) Dietary phosphorus requirement of juvenile sunshine bass, *Morone chryops* male × *M. saxtilis* females. *Aquaculture* **113**, 355–363.

Enell M. (1987) Environmental impact of cage fish farming with special references to phosphorus and nitrogen. *ICES/1987/FF* **44**.

Enell M. (1995) Environmental impact of nutrients from NORDIC fish farming. *Water Science* **31**, 61–71.

Enell M. & Ackefors H. (1991) Nutrient discharges from aquaculture operations in Nordic countries into adjacent sea areas. *ICES C.M./199/F:* **56**, ref. MEQC.

Forster R.P. & Goldstein L. (1969) Formation of excretory products. In: *Fish Physiology*, Vol. I (ed. by W. S. Hoar & D. J. Randall), pp. 313–350. Academic Press, New York, NY.

Foy R.H. & Rosell R. (1991a) Loadings of nitrogen and phosphorus from a Northern Ireland Fish farm. *Aquaculture* **96**, 17–30.

Foy R.H. & Rosell R. (1991b) Fractionation of nitrogen and phosphorus loadings from a Northern Ireland fish farm. *Aquaculture* **96**, 31–42.

Haiqing S. & Xiqin H. (1994) Effects of dietary animal and plant protein ratios and energy levels on growth and body composition of bream (*Megalopbrama skolkovii* Dybowski). *Aquaculture* **127**, 189–196.

Hajen W.E., Beames R.M., Higgs R.M. & Dosanjh B.S. (1993) Digestibility of various feedstuffs by post-juveniles chinook salmon (*Oncorhynchus tshawytscha*) in seawater. I. Validation of techniques. *Aquaculture* **112**, 321–332.

Hennessy M.M., Wilson L., Struthers W. & Kelly L.A. (1996) Waste loadings from two freshwater Atlantic salmon juvenile farms in Scotland. *Water Air, and Soil Pollution* **86**, 235–249.

Hasan M.A., Islam M.N. & Alim M.A. (1993) Evaluation of silkworm pupae meal as dietary protein source for catfish (*Heteropneustes fossilis* Bloch). In: *Fish Nutrition Practice* (ed. by S. J. Kaushik & P. Luquet), pp. 785–791. INRA, Paris.

Huisman E.A., Breteler J.G.P., Vismans M.M. & Kanis E. (1979) Retention of energy, protein, fat and ash in growing carp (*Cyprinus carpio* L.) under different feeding and temperature regimes. In: *Finfish Nutrition and Fishfeed Technology*, Proceedings of the World Symposium on Finfish Nutrition and Fish feed Technology, Hamburg, 20–23 June 1978, Vol. I (ed. by J. E. Halvers & K. Tiews), pp. 175–188.

Johnsen F., Hillestad M. & Austreng E. (1993) High energy diets for Atlantic salmon. Effect on pollution. In: *Fish Nutrition in Practice* (ed. by S. J. Kaushik & P. Luquet), pp. 391–401. INRA, Paris.

Ketola H.G., Westers H., Houghton W. & Pecor C. (1991) Effect of diet on growth and survival of coho salmon and on phosphorus discharges from a fish hatchery. *American Fisheries Society Symposium* **10**, 402–409.

Ketola H.G. & Harland B.F. (1993) Influence of phosphorus in rainbow trout diets on phosphorus discharges in effluent water. *Transactions of the American Fisheries Society* **122**, 1120–1126.

Kibria G., Nugegoda D., Fairclough R. & Lam P. (1996) Australian native species in Aquaculture. *Victorian Naturalist* **113**, 264–267.

Kristiansen G. & Hessen D.O. (1992) Nitrogen and phosphorus excretion from the noble crayfish, *Astacus astacus* L., in relation to food type and temperature. *Aquaculture* **102**, 245–264.

Lall S.P. (1979) Minerals in finfish nutrition. In: *Finfish Nutrition and Fishfeed Technology*, Proceedings of the World Symposium on Finfish Nutrition and Fish feed Technology, Hamburg, 20–23 June 1978, Vol. I (ed. by J. E. Halver & K. Tiewes), pp. 85–93.

Lall S.P. (1991) Digestibility, metabolism and excretion of dietary phosphorus in fish. In: *Nutritional Strategies and Aquaculture Waste*, Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste, University of Guelph, Ontario, Canada (ed. by C. B. Cowey by C. Y. Chong), pp. 163–185.

Lanari D., D'Agaro E. & Ballestrazzi R. (1995) Effect of dietary DP/DE ratio on apparent digestibility, growth and nitrogen and phosphorus retention in rainbow trout, *Oncorhynchus mykiss* (Walbaum). *Aquaculture Nutrition* **1**, 105–110.

Law A.T. (1984) Nutritional study of jelawat, *Leptobarbus hoevenii* (Bleeker), fed on pelleted feed. *Aquaculture* **41**, 227–233.

Lovell R.T. (1978) Dietary phosphorus requirements of channel catfish (*Ictalurus punctatus*). *Transactions of the American Fisheries Society* **107**, 617–621.

Matty A.J. (1990) Feeds in the fight against pollution. *Fish Farming International* January, 16–17.

Nakashima B.S. & Leggett W.C. (1980) Natural sources and requirements of channel catfish (*Ictalurus punctatus*). *Transactions of the American Fisheries Society* **107**, 617–621.

Ogino C. & Takeda H. (1978) Requirements of rainbow trout for dietary calcium and phosphorus. *Bulletin of the Japan Society Science and Fisheries* **44**, 1019–1022.

O'Sullivan D. (1995) Status of Australian native finfish aquaculture production in 1991–92. In: *Silver Perch Culture*, Proceedings of Silver Perch Aquaculture Workshops, Grafton and Narrendera, April 1997 (ed. by S. J. Rowland & C. Bryant), pp. 5–8. Austasia Aquaculture for NSW Fisheries.

O'Sullivan D. & Kiley T. (1996) Status of Australian aquaculture in 1994/95. *Austasia Aquaculture Trade Directory* 2–12.

Penczak T., Galicka W., Molinski E., Kusto E. & Zalewski

M. (1982) The enrichment of a mesotrophic lake by carbon, phosphorus and nitrogen from the cage aquaculture of rainbow trout *Salmo gairdneri*. *Journal of the Applied Ecology* **19**, 371–393.

Phillips A.M., Podoliak H.A., Jr. Brockway D.R. & Balzer G.C. (1956) *New York State of Conservation, Department of Fisheries Research Bulletin, Cortland hatchery Report* **25**, 20 pp.

Rowland S.J., Allan G., Hollis M. & Pontifex T. (1995) Production of the Australian freshwater silver perch, *Bidyanus bidyanus* (Mitchell), at two densities in earthen ponds. *Aquaculture* **130**, 317–328.

Savitz I. (1971) Nitrogen excretion and protein consumption of the bluegill sunfish (*Lepomis macrochirus*). *Journal of the Fisheries Research Board of Canada* **28**, 449–457.

Singh B.N., Sinha V.R.P. & Chakraborty D.P. (1979) Effects of protein quality and temperature on the growth of fingerlings of rohu, *Labeo rohita* (Hamilton). In: *Finfish Nutrition and Fishfeed Technology*, Proceedings of the World Symposium on Finfish Nutrition and Fishfeed Technology, Hamburg, June 1978, Vol. II (ed. by J. E. Halver & K. Tiewes), pp. 303–310.

Solberg S.O. & Bregnballe F. (1982) Pollution from trout feed with minced trash fish. In: *Report of the EIFAC Workshop on Fish-Farm Effluents*, Silkeborg, Denmark, 26–28 May 1981 (ed. by J. S. Albaster), pp. 65–71. *FAO/EIFAC Technical Paper* **41**, 166 pp.

Storebakken T. & Austreng E. (1987a) Ration level of salmonids. I. growth, survival, body composition and feed conversion in Atlantic salmon fry and fingerlings. *Aquaculture* **60**, 189–206.

Storebakken T. & Austreng E. (1987b) Ration level of salmonids. II. growth, feed conversion, protein digestibility, body composition and feed conversion in Rainbow trout weighing 0.5–1.0 kg. *Aquaculture* **60**, 207–221.

Tecator (1990) *Instruction Manual, The Aquatec System*. Hoganas, Sweden.

Walker K.F. & Hillmann T.J. (1982) Phosphorus and nitrogen loads associated with the River Murray near Albury-Wodonga and their effects on phytoplankton populations. *Australian Journal of Marine and Freshwater Research* **33**, 223–243.

Warren-Hansen I. (1982) Evaluation of matter discharged from trout farming in Denmark. In: *Report of the EIFAC Workshop on Fish-Farm Effluents*, Silkeborg, Denmark, 26–28 May 1981 (ed. by J. S. Albaster), pp. 57–63. *FAO/EIFAC Technical Paper* **41**, 166 pp.

Weatherly A.H. & Gill H.S. (1987) *The Biology of Growth*. Academic Press, London.

Windell J.T., Foltz J.W. & Sarokon J.A. (1978) Effect of size, temperature, and amount fed on nutrient digestibility of a pelleted diet by rainbow trout. *Salmo gairdneri*. *Transactions of the American Fisheries Society* **107**, 613–616.

Zar J.H. (1984) *Biostatistical Analysis*. Prentice-Hill, Englewood Cliffs, NJ.

INTERNATIONAL CENTER FOR LIVING AQUATIC RESOURCES MANAGEMENT, INC.
MCPO BOX 2631, 0718 MAKATI CITY, PHILIPPINES

5 September 1997

Our ref NTAS-120-L97

Mr Golam Kibria
9 Keith Street
Coburg 3058
Australia

Dear Mr Kibria,

Your article *Nitrogen pollution from aquaculture: Can it be reduced?* has been forwarded to us for possible publication in the Aquabyte section of the Naga. The article will be sent to the reviewers and we shall notify you of its progress.

Thank you very much for your continued interest in the activities of the NTAS.

With best regards.

Yours sincerely,

[redacted]

Natalie D Macawaris
NTAS Secretary
Assistant Editor, Aquabyte - Naga

Date: Fri, 07 Nov 1997 11:42:00 -0800 (PST)
From: Natalie Macawaris <N.MACAWARIS@CGNET.COM>
To: Golam Kibria <s9337499@cougar.vut.edu.au>
Subject: Urgent: query on Article for Aquabyte/Naga

Your article 'Nitrogen Pollution from Aquaculture: Can it be Reduced' will be published in the January 1998 issue of Naga. In this regard, below are some queries re the article:

(a) We need the source (journal/book) title for

Iwata, K. 1970. Relationship between food and growth of young crucian carp, Carassinus auratus cuvieri, as determined by the nitrogen balance.

(b) We need the publishers name and place for

Watanabe, T. 1991. Past and present approaches to aquaculture waste management in Japan. In Nutritional strategies and aquaculture wastes, C.B. Cowley and C.Y. Cho (eds.)

On behalf of Dr Gupta, the NTAS Coordinator, we would like to convey our appreciation for your continued interest in the activities of the network.

More power to you and best regards.

Sincerely,

Natalie

JOURNAL OF APPLIED ICHTHYOLOGY

Editor in Chief: Prof. Dr. Dr. h.c. Dr. h.c. Harald Rosenthal
Düsternbrooker Weg 20, Insitute for Marine Science, University of Kiel, 24105 Kiel, Germany

Kiel, 26. November 1997

Dr. Godam Kibria
84 Munro Street
Coburg, VIC 3058
Australia

Re: revised manuscript JAI-97-50 : Effects of salinity on the growth and nutrient retention in silver perch *Bidyanus bidyanus*

Dear Dr. Kibria,

I am in receipt of the revised version of the above mentioned manuscript. After careful reading and comparison with the previous version and the referee comments I consider it now acceptable for publication in the Journal of Applied Ichthyology. Congratulations for providing a valuable contribution to aquatic sciences.

I have initiated today the processing of your paper. However, be aware that it will take quite some time before it will appear because we have at the moment a number of papers in the mill. I assume it will appear in the summer issue of 1998. You will receive the galley proof pages early in the next year.

Sincerely yours

[redacted]

Harald Rosenthal

Biology and Aquaculture of Silver Perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae): A Review

Golam Kibria[1], Dayanthi Nugegoda[1], Robert Fairclough[2] and Paul Lam[3]

Abstract

The Silver Perch *Bidyanus bidyanus* is the most important fish contributing to major endemic freshwater aquaculture production in Australia. The demand to culture the species is growing in nearby Asia. This is a review of its natural distribution, food and feeding habits, important biological characteristics, and on different aspects of Silver Perch aquaculture including breeding, nutrition, growth performance in freshwater and slightly saline waters. The review also includes data on Silver Perch production and the quality and quantity of wastes generated from aquaculture of Silver Perch. (*The Victorian Naturalist* 115, 1998,).

Introduction

Silver Perch *Bidyanus bidyanus* is an Australian endemic species with a high aquaculture potential (Allan and Rowland 1996). It is one of the species of the Murray-Darling River system that is much sought after by commercial and recreational fishers (Cadwallader 1979). The species was once abundant and widespread throughout the Murray-Darling River system but its distribution and abundance have been greatly reduced, and the fish is now uncommon in many areas (Rowland 1995a). Demand to cultivate the species is increasing both in Australia and in nearby Asia (Gooley and Rowland 1993) and, currently, *B. bidyanus* represent the main endemic freshwater aquaculture industry in Australia. There are some reports on *B. bidyanus* biology and aquaculture, however, this information is scattered in different journals, books and magazines. The present paper reviews and collates all the important information published on Silver Perch biology and aquaculture.

History, Natural Habitats and Status

Bidyanus bidyanus (Mitchell 1838) (Teraponidae) is endemic to most of the Murray-Darling river system (Merrick and Schmida 1984) except in the cool, high upper reaches of streams (Lake 1967a; Pollard *et al.* 1990; Merrick and Schmida 1984). It is a potamodromous species, i.e. migrates within the freshwater habitat (Guo *et al.* 1995). The species was commonly consumed by the Aborigines, and the scientific name was also derived from the Aboriginal name *bidyan* (Rowland 1995a) by the explorer Major Thomas Mitchell who named the fish after he caught it in the Namoi River in 1832 (Mitchell 1838).

The species was once abundant but its population has been greatly reduced due to competition for food from introduced cyprinids e.g. Common Carp *Cyprinus carpio*, predation by the English Perch *Perca fluviatilis*, and the construction of dams that prevented the upstream migration of *B. bidyanus* (Cadwallader and Backhouse 1983). It is now a 'Potentially Threatened' species (Jackson 1994) and may become 'Endangered' unless measures are taken to increase its population (Rowland 1994). Such measures might include aquaculture since *B. bidyanus* has a number of characteristics that make it a viable proposition for aquaculture.

Biological Characteristics of Silver Perch

This species can tolerate a wide range of temperature, from 2.0°–32°C (Cadwallader and Backhouse 1983), but the optimum growth temperature range is believed to be 23°–28°C (Rowland 1995b). Apart from the temperature, growth may also be affected by the ammonia levels (NH_3). Since fish can tolerate only a low level of ammonia (Table 1), higher ammonia levels in the culture system may stress and cause mortalities to fish (Boyd 1990; Hart and O'Sullivan 1993). Prolonged exposure of *B. bidyanus* at a concentration of over 0.1 mg/l total ammonia caused a significant reduction in growth (Rowland 1995c; Rowland *et al.* 1995). The important water quality parameters for *B. bidyanus* are given in Table 1.

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Table 1. Water quality variables recommended for intensive Silver Perch aquaculture (Rowland 1995c).

Variables	Recommended	Optimum for growth
Temperature (°C)	10-30	23-28
Dissolved oxygen (mg/l)	>4.5	
pH	7.0-9.5	
Total ammonia nitrogen (TAN) (mg/l)	<2.0	
NH_3 (mg/l) (unionised ammonia)	<0.1	

Males become mature in their second year (233 mm) while females mature in their third year (340 mm) (Cadwallader and Backhouse 1983; Merrick and Schmida 1984). However, adult fish may die after spawning (Lake 1967b; Cadwallader and Backhouse 1983). Spawning occurs in summer (November–January) when sexually mature fish migrate upstream to spawn in shallow, warm waters (Cadwallader 1977; Merrick 1980; Cadwallader and Backhouse 1983; Reynolds 1983). Flooding is thought to be required for natural spawning of *B. bidyanus* (Davis 1977). The fecundity is high and the number of eggs per female varies from 300,000 (Merrick 1980) to 500,000 eggs (Whitley 1960). Eggs are pelagic with diameter of 2–8mm (Lake 1967c). Hatching occurs after about 30 hours at temperatures of 22°–31°C (Cadwallader and Backhouse 1983). The larvae are benthic (Lake 1967b) and juveniles form large schools (Lake 1967a; Merrick 1980) often congregating below rapids, weirs (Merrick 1980) and fast flowing water with sand and gravel bottom (Llewellyn 1983; Merrick and Schmida 1984; Starling 1992). Adults can live in extremely turbid waters (Cadwallader 1977) and the species is tolerant of high salt concentrations (Ingram *et al.* 1996). Silver Perch is also a territorial and aggressive fish (Cadwallader and Backhouse 1983; Starling 1992). Within two years of hatching the average size could be 180 mm (Cadwallader and Backhouse 1983) and common sizes usually caught are 350–410 mm (0.75–2.5 kg) (Merrick and Schmida 1984). The pre-spawning activities of *B. bidyanus* have been described by Merrick and Schmida (1984).

Food and Feeding Habitats

The young fish (larvae) starts feeding from the sixth day after hatching, mainly on zooplankton including rotifers, copepod nauplii, small copepods and cladocerans (Cadwallader and Backhouse 1983; Rowland 1984; Thurstan and Rowland 1995). However, adult fish are omnivorous and, at times, feed extensively on zooplankton, particularly the larger ostracods and cladocerans. Other food includes shrimps (*Macrobrachium* spp., atyids), yabbies, chironomid larvae, aquatic insects, earthworms, molluscs, filamentous algae, and aquatic plants (Lake 1967a; Cadwallader and Backhouse 1983; Merrick and Schmida 1984; Rowland 1994). The natural zooplankton identified from earthen ponds under *B. bidyanus* culture comprised *Moina micrura*, *Daphnia carinata* (cladocerans), *Boeckella fluvialis* (copepod) and *Brachionus calyciflorus*, *Asplanchna sieboidi* (rotifers) (Culver and Geddes 1993).

Aquaculture of Silver Perch

Bidyanus bidyanus has been identified as the only wholly freshwater species in Australia with a tremendous potential for aquaculture (Allan and Rowland 1996). There are a number of characteristics that make *B. bidyanus* an ideal species for aquaculture: a rapid and uniform growth in crowded conditions (Barlow 1986; Pollard 1986) (the current stocking densities in earthen ponds is 10,000 fingerling/ha with aeration and 5000 fingerlings/ha without aeration; Walker 1993); its effective use of both plant proteins and meat meal (Allan and Rowland 1996); its low production cost, high fecundity and ready acceptance of low-protein diet (Barlow 1986; Rowland and Barlow 1991; Walker 1994a, 1994b); tolerance of high temperatures (Pollard 1986); omnivorous feeding habit (Rowland and Kearney 1992); and high demands for its culture in Australia and Asia (Gooley and Rowland 1993). *B. bidyanus* farming is a major endemic fish culture industry in this country as demonstrated by its production figures (Tables 2 and 3) and has the potential to achieve an annual production of 10 t/ha (Rowland

Table 2. Comparison of native Silver Perch and native finfish production during 1994/95 (O'Sullivan and Kiley 1996). Key: NDA = No details available; NSW = New South Wales; NT=Northern Territory; TAS=Tasmania; SA=South Australia; QLD=Queensland; VIC=Victoria; WA=Western Australia

	farm (tonnes)	hatchery (000's)	value ($,000)
FINFISH			
Statewise Silver Perch production			
Silver Perch (NSW)	17.3	1,807.3	635.8
Silver Perch (Vic)	1	NDA	10
Silver Perch (Qld)	34.4	400.0	331.9
Silver Perch (SA)	-	-	-
Silver Perch (WA)	-	-	-
Silver Perch (Tas)	-	-	-
Silver Perch (NT)	-	-	-
Silver Perch and other native finfish production during 1994/95			
Silver Perch (all states)	52.7	2,207.3	977.7
Golden perch (all states)	0.5	2,498.9	397.0
Murray cod (all states)	<0.1	196.7	101.3
Trout cod (all states)	0	32.1	3.4
Australian bass (all states)	<0.1	274.3	95.6
Catfish (all states)	0	44.5	42.6
Macquarie perch (all states)	0	53.0	5.4
Mary River cod (all states)	0	4.7	7.0

Table 3. Native fish production and contribution from Silver Perch during 1988 to 1995.

Year	Native fish farm production (t)	Silver Perch contribution (t)	Source
1988-89	10	-	Treadwell *et al.* (1992)
1991-92	43.9	26.6	O'Sullivan (1995)
1994-95	53.2	52.7	O'Sullivan & Kiley (1996)

1995b; Walker 1994a). Major *B. bidyanus* farms are small (1–3 ponds) although there are some medium sized farms (4–8 ponds) and a few large farms (6–17 hectares) (Walker and Caney 1996).

The aquaculture of *B. bidyanus* is dominant in Queensland (Qld) and in New South Wales (NSW). These two states contributed 98% of *B. bidyanus* production in 1994/1995 (Table 2). In fact, the increase in freshwater fish production in Australia is mainly due to the interest and investment in growing *B. bidyanus* (Kibria *et al.* 1996) (see also Table 3).

To achieve the best result for raising *B. bidyanus*, research has been undertaken by a number of different authors on the breeding, nutrition, growth performance in ponds, tanks, and saline waters. The potential effects of pollution from rearing of Silver Perch has also been examined. Findings of the above research are summarised below:

Research in the Artificial Breeding of Silver Perch

Techniques on the artificial propagation of *B. bidyanus*, along with other endemic species have been developed at the Inland Fisheries Research Station, Narrandera, Eastern Freshwater Fish Research Hatchery, Grafton, both in New South Wales, and at Snobs Creek Freshwater Fisheries Research Station and Hatchery in Victoria (Rowland *et al.* 1983; Rowland 1984). The techniques involved the induction of breeding of endemic fish by using an artificial hormone (Human Chorionic Gonadotrophin, HCG) and a preparation of the pituitary gland from Common Carp (Carp Pituitary Gland, CPG). The optimum dose of HCG that induced a high hatching rate of eggs in *B. bidyanus* was 200 IU/kg HCG (Rowland 1984). About three million *B. bidyanus* fry are produced annually by induced breeding for stocking farm dams and other natural waters used for recreational fishing (Rowland *et al.* 1995).

Research on Silver Perch Nutrition

Research has been undertaken on the nutrition and production of *B. bidyanus* by a number of authors, including Allan and Rowland (1991), Rowland and Barlow (1991), Allan and Rowland (1992), Allan and Rowland (1994). Out of three experimental diets tested (21%, 36% and 49% protein), Allan and Rowland (1991) obtained the fastest growth rate of *B. bidyanus* using a diet containing 36% protein. Based on this trial, Allan and Rowland (1992) developed the first reference diet for *B. bidyanus* which contained 35.6% protein, 5.5% fat, and 1.1% fatty acids. The total methionine and lysine composition (the limiting amino acids in feed) of the reference diet was 7.4 and 22.6 g/kg respectively. *B. bidyanus* fed a diet containing 35% protein showed better Food Conversion Ratio (FCR) than when fed a diet containing other levels of protein (Allan and Rowland 1991) whereas Kibria *et al.* (1997a.) found comparatively higher growth and FCR of *B. bidyanus* using a diet containing 45% protein (Table 4).

Research has also been undertaken on the use of Australian oilseeds (Soybean, Canola, Cottonseed, and Peanut) and grain legumes (Lupins, Chick Pea, Field Pea and Cow Pea) as ingredients of *B. bidyanus* diets and results are encouraging, since apparent digestibility coefficients (ADC's) of vegetable protein is similar to, or higher than, that obtained with fish meal (Allan and Rowland 1994). The research found that *B. bidyanus* is good at digesting vegetable protein, and the best growth of the fish was obtained from peanut meal. The next best was soybean meal followed by lupin and canola meal. However, research also found that growth of *B. bidyanus* decreased, and the FCR value deteriorated with the increase of plant protein content. Very poor FCRs resulted when *B. bidyanus* were fed with a high fibre diet (O'Sullivan 1994). Studies were also carried out on the effects of varying protein and energy concentrations, the result of which show that growth of *B. bidyanus* increased with increasing protein and energy level in diets (Allan *et al.* 1994). The fat deposition in *B. bidyanus* was directly related to the fat content of diets (Anderson and Arthington 1989; Hunter and Roberts 1994). Further experimental trials confirmed that *B. bidyanus* require in their diet fatty acids of both the linolenic (18:3n–3) and linoleic (18:2n–6) series (Anderson and Arthington 1989). A synopsis of nutritional research on *B. bidyanus* is given in Allan and Rowland (1996).

Growth and Production of Silver Perch

Growth of *B. bidyanus* is also affected by the rearing temperature. During winter months (May–September), the growth rate is significantly slower than in the warmer months (October–March) (Table 5). *B. bidyanus* eat aggressively at a temperature above 20°C and are less aggressive in winters. Although *B. bidyanus* grow better in temperatures above 20°C, prolonged exposure at 30°C and above adversely affects the appetite, food conversion and growth of the species (Rowland 1995c).

The *B. bidyanus* fry grew faster at a stocking density of 25,000/ha than at 80,000/ha in earthen ponds (Rowland *et al.* 1994). When *B. bidyanus* were reared at a higher density (43,000 fish/ha), the annual production figure was calculated to be 10.2 tonnes/ha (Rowland 1995d; Rowland *et al.* 1995). However, at higher density culture,

Table 4. Effect of dietary protein levels, and rearing temperature on food conversion ratio (FCR) of Silver Perch.

Protein content (%)	Feeding rate % BW[1]/day	Water temperature	FCR system	Culture	Source
20.7%	satiation	18.30-22.80C	3.0+0.2	Tank	Allan and Rowland (1991)
35.7%	satiation	18.30-22.80C	1.7+0.1	Tank	Allan and Rowland (1991)
49%	satiation	18.30-22.80C	2.4+0.3	Tank	Allan and Rowland (1991)
36%	3%	18.00-20.00C	3.20	Aquaria	Kibria *et al.* (1997a)
45%	3%	18.00-20.00C	2.24	Aquaria	Kibria *et al.* (1997a)
53%	3%	18.00-20.00C	2.97	Aquaria	Kibria *et al.* (1997a)
35%	5%	22.00C-26.60C	1.1-1.2	Pond	Rowland *et al.* (1994)
35%	4% & 3%	13.20C-28.40C	1.8-1.9	Pond	Rowland *et al.* (1995)
35%	3%	-	1.0-1.3	-	Allan and Rowland (1992)
35%	3%	12.50C-30.30C	2.3	Pond	Rowland (1995d)
50%	3%	22.00C-31.30C	0.7	Pond	*Rowland (1994)

[1]BW = body weight; * = Rowland commented that FCR was better apparently due to eating of natural pond food as well.

particularly during summer, the water quality may deteriorate resulting in an increase of disease susceptibility (Rowland 1995d).

Performance of Silver Perch in Tanks/ Cages

McKinnon *et al.* (1996) obtained comparatively better growth and survival of *B. bidyanus* in floating cages fixed in irrigation channels in integrated aquaculture-irrigation systems, while poor growth and survival resulted from cages fixed in tanks supplied with groundwater. The growth and FCR of *B. bidyanus* was significantly lower in tanks than in earthen ponds (Rowland 1995b). Similarly, growth of *B. bidyanus* was found to be slower in aquaria compared to commercial ponds (Kibria *et al.* 1997a).

Performance of Silver Perch in Saline Waters

Bidyanus bidyanus can tolerate salinity up to 15 parts per thousand (salinity) (Guo *et al.* 1995) and larvae hatched at six salinity showed a better survival rate than those hatched in freshwater (Guo *et al.* 1993). Ingram *et al.*(1996) reported a good survival and growth of *B. bidyanus* when reared in cages at a salinity of 8.0–15.3 salinity, whereas poor growth and survival resulted at higher salinities of 9.5–24.6 salinity. The growth and survival obtained at 8.0–15.3 salinity were better than those of similar sized fish reared in freshwater cages (Ingram *et al.* 1996). Recently, we reared *B. bidyanus* fingerlings at different salinity levels (0 salinity, 4 salinity, 8 salinity and 12 salinity) in order to evaluate growth and nutrient retention efficiency. *B. bidyanus* grew faster and had a better FCR and nutrient retention at 4 salinity than at other salinities (Kibria *et al. unpubl. data*).

Pollution Potential from Aquaculture of Silver Perch

Kibria *et al.* (1997b) reared *B. bidyanus* fingerlings at 20°C, 25°C and 30°C to investigate the solid waste production and nutrient loading to the environment. *Bidyanus bidyanus* grown at 25°C produced significantly less solid waste and nutrient load ($P<0.05$) compared with other temperatures in the order of 25°C< 30°C<20°C. In another experiment we found that the diet which resulted in *B. bidyanus* having comparatively higher weight gain, specific growth rate (SGR) and food conversion ratio (FCR) produced less suspended and dissolved solid waste to the environment (Kibria *et al.* 1997a). *Bidyanus bidyanus* grown at 25°C produced less nitrogen and phosphorus load to the environment and this could be due to the better growth, good FCR and lower faecal and metabolic loss obtained at that temperature (Kibria *et al.* 1997b; Kibria *et al.* 1997c).

Conclusion

The endemic fish *B. bidyanus* has great potential as a freshwater aquaculture species in Australia. The biology and nutrition of the species are well established. Recent study shows that growing *B. bidyanus* at their optimum temperature may enhance nutrient retention and a reduction in the discharge of nutrient to the environment. For a sustainable aquaculture programme, development of low polluting *B. bidyanus* diet and experiments on polyculturing of *B. bidyanus* would be important areas for future research. Moreover, further experiments on rearing of *B. bidyanus* in slightly saline waters may provide pertinent data of its suitability of culturing in brackish water zones.

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References

Allan, G.L. and Rowland, S.J. (1991). Growth of Silver Perch *Bidyanus bidyanus* on diets with different levels and sources of protein. *Proceedings of the Nutritional Society of Australia* **16**, 211.

Allan, G.L and Rowland, S.J. (1992). Development of an experimental diet for Silver Perch (*Bidyanus bidyanus*). *Austasia Aquaculture* **6**, 39-40.

Allan, G.L. and Rowland, S.J. (1994). The use of Australian oilseeds and grain legumes in aquaculture diets. *In* 'Proceedings of the Third Asian Fisheries Forum', Singapore, 26-30 October 1992, pages 667-670. Eds L.M. Chou, A.D. Munro, T.S. Lam, T.W. Chen, L.K.K. Cheong, J.K. Ding, K.K. Hooi, H.W. Khoo, V.P.E. Phang, K.F. Shim and C.H. Tan. (Asian Fisheries Society: Manila).

Allan, G.L. and Rowland, S.J. 1996. The Silver Perch industry: research progress and priorities toward developing cost-effective diets. *Austasia Aquaculture* **10**(2), 37-40.

Allan, G.L., Warner-Smith, R.J. and Frances, J. (1994). The effects of varying protein and energy concentrations on growth, protein efficiency ratio and food conversion ration for Silver Perch (*Bidyanus bidyanus* Mitchell). *Proceedings of the Nutritional Society of Australia* **18**, 53.

Anderson, A.J. and Arthington, A.H. (1989). Effect of diet on the fatty acid composition of Silver Perch (*Leiopotherapon bidyanus*) lipids. *Comparative Biochemistry and Physiology* **93B**, 715-720.

Barlow, C.G. (1986). Fish in farm dams and implications for extensive aquaculture. *In* 'Proceedings of the First Freshwater Aquaculture Workshop', Narrendera, pages 33-43. Ed. L.F. Reynolds. (NSW Agriculture and Fisheries: Sydney).

Boyd, C. (1990). Water quality in ponds for aquaculture. (Alabama Agricultural Experimental Station, Auburn University: Alabama).

Cadwallader, P.L. (1977). J.O. Langtry's 1949-50 Murray River investigations. Fisheries and Wildlife Paper, Victoria. No. 13.

Cadwallader, P.L. (1979). Distribution of native and introduced fish in seven creeks River system, Victoria, Australia. *Australian Journal of Ecology* **4**, 361-385.

Cadwallader, P.L. and Backhouse, G.N. (1983). A guide to the freshwater fish of Victoria, 249p. (Government Printer: Melbourne).

Culver, D.A. and Geddes, M.C. (1993). Limnology of rearing ponds for Australian fish larvae: relationships among water quality, phytoplankton, zooplankton, and the growth of larval fish. *Australian Journal of Marine and Freshwater Research* **44**, 537-551.

Davis, T.L.O. (1977). Reproductive biology of the freshwater Catfish, *Tandanus tandanus* Mitchell, in the Gwydir River, Australia. II, Gonadal Cycle and fecundity. *Australian Journal of Marine and Freshwater Research* **28**, 159-169.

Gooley, G. and Rowland, S. (1993). Murray-Darling Finfish-Current Developments and Commercial Potential. *Austasia Aquaculture* **7**(3), 35-37.

Guo, R., Mather, P. and Capra, M.F. (1993). Effect of salinity on the development of Silver Perch (*Bidyanus bidyanus*) eggs and larvae. *Comparative Biochemistry and Physiology* **A103**, 531-535.

Guo, R., Mather, P.B., and Capra, M.F. (1995). Salinity tolerance and osmoregulation in the Silver Perch, *Bidyanus bidyanus* Mitchell (Teraponidae), an endemic Australian freshwater teleost. *Australian Journal of Marine and Freshwater Research* **46**, 947-952.

Hart, P. and O'Sullivan, D. (1993). Recirculation systems: design, construction and management. National Key Centre for Teaching and Research in Aquaculture. (University of Tasmania at Launceston, Australia).

Hunter, B.J.and Roberts, D.C.K. (1994). The effects of varying dietary protein/energy ratios in the fat content and fatty acid composition of Silver Perch (*Bidyanus bidyanus*). *Proceedings of the Nutritional Society of Australia*, **18**, 54.

Ingram, B., Gooley, G. and McKinnon, L. (1996). Potential for inland mariculture in Victorian saline groundwater evaporation basins. *Austasia Aquaculture* **10**(2), 61-63.

Jackson, P. (1994). Australian threatened fishes - 1994 supplement. *Australian Society for Fish Biology Newsletter* **24**(2), 18-26.

Kibria, G., Nugegoda, D., Fairclough, R. and Lam, P. (1996). Australian native species in aquaculture *The Victorian Naturalist* **113**(5), 264-267.

Kibria, G., Nugegoda, D., Lam, P. and Fairclough, R. (1997a.) Preliminary rearing trials of an Australian native fish, Silver Perch (*Bidyanus bidyanus*) (Mitchell) with reference to growth and production of solid waste in aquaculture. *Asian Fisheries Science* **9**, 301-309.

Kibria, G., Nugegoda, D., Fairclough, R. and Lam, P. (1997b.) Pollution from aquaculture. *Chemistry in Australia* **64**(1), 19-20.

Kibria, G., Nugegoda, D., and Fairclough, R. (1997c). Nitrogen pollution from aquaculture: Nitrogen losses and nitrogen retention by Silver Perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae). Paper presented at the joint conference of Australian Society for Fish Biology (ASFB) and the Fish and Aquatic Resource Management Association of Australasia (FARMAA) held in Darwin, NT, Australia (15-19 July 1997). (abstract). 65-65.

Lake, J. S. (1971). Freshwater fishes and rivers of Australia. (Nelson: Sydney).

Lake, J. S. (1967a). Principal fishes of the Murray-Darling River system. *In* 'Australian Inland Waters and their Faunas', pages 192-213 Ed A.H. Weatherley. (Australian National University Press: Canberra).

Lake, J.S. (1967b) Rearing experiments with five species of Australian freshwater fishes. I. Inducement to spawning. *Australian Journal of Marine and Freshwater Research* **18**, 137-153.

Lake, J.S. (1967c). Rearing experiments with five species of Australian freshwater fishes. II. Morphogenesis and ontogeny. *Australian Journal of Marine and Freshwater Research* **18**, 155-173.

Llewllyn, L. C. (1983). The distribution of fish in New South Wales. Australian Society of Limnology. Special Publication No. 7, 23p.

McKinnon, L., Gooley, G. and Gasior, R. (1996) Integrated Silver Perch aquaculture trials in the Goulburn-Murray irrigation District of Victoria. *Austasia Aquaculture* **10**(1), 45-47.

Merrick, J.R. (1980). Family Teraponidae Freshwater Gunters or Perches. *In* 'Freshwater Fishes of South Eastern Australia', pp. 150-152. Ed. R.M. McDowell. (Reed: Sydney).

Merrick, J.R. and Schmida, G.E. (1984). 'Australian freshwater fishes - biology and management'. (Griffin Press Ltd: South Australia)

Mitchell, T.L. (1838). Three expeditions into the interior of Australia, with description of the recently explored region of Australia Felix and of the present colony of New South Wales. (T. & W. Boone, London).

O'Sullivan, D. (1994). Research underway on replacement of fishmeal in aquaculture diets *Austasia Aquaculture* **8**(2), 44-45.

O'Sullivan, D. (1995). Status of Australian native finfish aquaculture production in 1991-92. *In* 'Silver Perch culture: Proceedings of Silver Perch workshops, Grafton and Narrandera, April, 1994', pp 5-8. Eds S. J. Rowland and C. Bryant. (Austasia Aquaculture, Hobart for NSW Fisheries: Sydney).

O'Sullivan, D. and Kiley, T. (1996). Status of Australian aquaculture in 1994/95. Austasia Aquaculture Trade Directory 1996, 2-12

Pollard, J. (1986). 'Fishing Australia'. (The Book Company International Pty Ltd: Sydney)

Table 5. Growth of Silver Perch reared in ponds at different seasons.

Months	Seasons	Temperature	Growth (g/d)	Source
May-September	Winter	11.1-20°C	0.5	Rowland (1995b)
October-March	Summer	>20°C	2.3 (low density)	Rowland (1995b)
October-March	Summer	>20°C	2.1 (high density)	Rowland (1995b)

Pollard, D.A., Ingram, B.A., Harris, J.H. and Reynolds, L.F. (1990). Threatened fishes in Australia - an overview. *Journal of Fish Biology* 37 (supp. A), 67-78.
Reynolds, L.F. (1983). Migration patterns of five fish species in the Murray-Darling River system. *Australian Journal of Marine and Freshwater Research* 34, 857-871.
Rowland, S.J. (1984). The hormone-induced spawning of Silver Perch, *Bidyanus bidyanus* (Mitchell) (Teraponidae). *Aquaculture* 42, 83-86.
Rowland, S.J. (1992). Diet and feeding of Murray cod (*Maccullochella peeli*) larvae. *Proceedings of the Linnean Society of New South Wales* 113(3), 193-201.
Rowland, S. J. (1994). Preliminary evaluation of the Australian freshwater fish Silver Perch, Bidyanus bidyanus, for pond culture. *Journal of Applied Aquaculture* 4(1), 39-48.
Rowland, S. J. (1995a). The Silver Perch, *Bidyanus bidyanus*, and its potential for aquaculture. *In* 'Silver Perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrandera, April, 1994', pages 9-11. Eds S.J. Rowland and C. Bryant. (Austasia Aquaculture, Hobart for NSW Fisheries: Sydney).
Rowland, S.J. (1995b). Production of fingerling and market size Silver Perch in earthen ponds. *In* 'Silver Perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrandera, April, 1994', pages 41-49. Eds S.J. Rowland and C. Bryant. (Austasia Aquaculture, Hobart for NSW Fisheries: Sydney).
Rowland, S.J. (1995c) . Water quality in the intensive pond culture of Silver Perch. *In* 'Silver Perch culture: Proceedings of Silver Perch Workshops, Grafton and Narrandera, April, 1994', pages 51-65. Eds S.J. Rowland and C. Bryant. (Austasia Aquaculture, Hobart for NSW Fisheries: Sydney).
Rowland, S.J. (1995d). High density pond culture of Silver Perch, *Bidyanus bidyanus*. *Asian Fisheries Science* 8, 73-79.
Rowland, S.J., Diron, J. and Selose, P. (1983). Production and stocking of Golden and Silver Perch in NSW. *Australian Fisheries* September, 24-28.
Rowland, S.J. and Barlow, C.G. (1991). Fish biology the right prerequisites: a case study with freshwater Silver Perch (Bidyanus bidyanus). *Austasia Aquaculture* 5(5), 27-30.
Rowland, S.J. and Kearney, R.E. (1992). The prospects for the commercial aquaculture of Silver Perch (*Bidyanus bidyanus*). A research project arising from a review of freshwater aquaculture. NSW Fisheries, 16p.
Rowland, S.J., Allan, G.L., Clark, K., Hollis, M. and Pontifex, T. (1994). Production of fingerling, *Bidyanus bidyanus* (Teraponidae) at two densities in earthen ponds. *The Progressive Fish Culturist* 56, 296-298.
Rowland, S.J., Allan, G.L., Hollis, M. and Pontifex, T. (1995). Production of the Australian freshwater Silver Perch, *Bidyanus bidyanus* (Mitchell), at two densities in earthen ponds. *Aquaculture* 130, 317-328.
Starling, S. (1992). 'The Australian Fishing Book', 512p. (Sand Piper Press Ltd: NSW).
Treadwell, R., McKelvie, L. and Maguire, G.B. (1992). Potential of Australian Aquaculture. Australian Bureau of Agricultural and Resources Economics (ABARE) Research Report 92.2. Commonwealth of Australia. Canberra .81p.
Thurstan, S. and Rowland, S.J. (1995). Techniques for the hatchery production of Silver Perch. *In* 'Silver Perch Culture: Proceedings of Silver Perch Aquaculture Workshops, Grafton and Narrandera, April, 1994', pages 29-39. Eds S.J. Rowland and C. Bryant. (Austasia Aquaculture, Hobart for NSW Fisheries: Sydney).
Walker, T. (1993). Silver Perch: Giving small farmers a slice of the action. *Austasia Aquaculture* 7(5), 3-6.
Walker, T. (1994a). Moratorium on Silver Perch all for a good cause says Government. *Austasia Aquaculture* 8(5), 3-6.
Walker, T. (1994b). Catfish model for Silver Perch put under microscope. *Austasia Aquaculture* 8(5), 24-36.
Walker, T. and Caney, D. (1996). Silver Perch extension officer reflects on progress. *Austasia Aquaculture* 10(2), 34-36.
Whitely, G. P. (1960). 'Native Freshwater Fishes of Australia'. (Jacaranda Press: Brisbane).

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March 5, 1998

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ABSTRACTS

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Phosphorus Balance in A Simulated Aquaculture System: A Case Study With the Native Australian Fish Silver Perch *(Bidyanus bidyanus)*

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Phosphorus is the key nutrient in freshwater eutrophication. studies were made on the loadings of phosphorus to the environment form rearing of native freshwater silver perch, *Bidyanus bidyanus*. Trials were conducted at two temperatures (20±1℃and 25±1℃) and fish are being fed two commercial diets referred to as diet(1) and diet(2). Total phosphorus was determined in the fish caracass, feed, faeces and in uneaten feed in order to determine phosphorus gain by fish (as bioaccumulation) and phosphorus loss to the environment (as pollutants).

Keywords: Aquaculture, Phosphorus, Feed, Pollution

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PATTERNS OF NUTRIENT DISCHARGE BY SILVER PERCH *Bidyanus bidyanus* REARED AT DIFFERENT TEMPERATURES.

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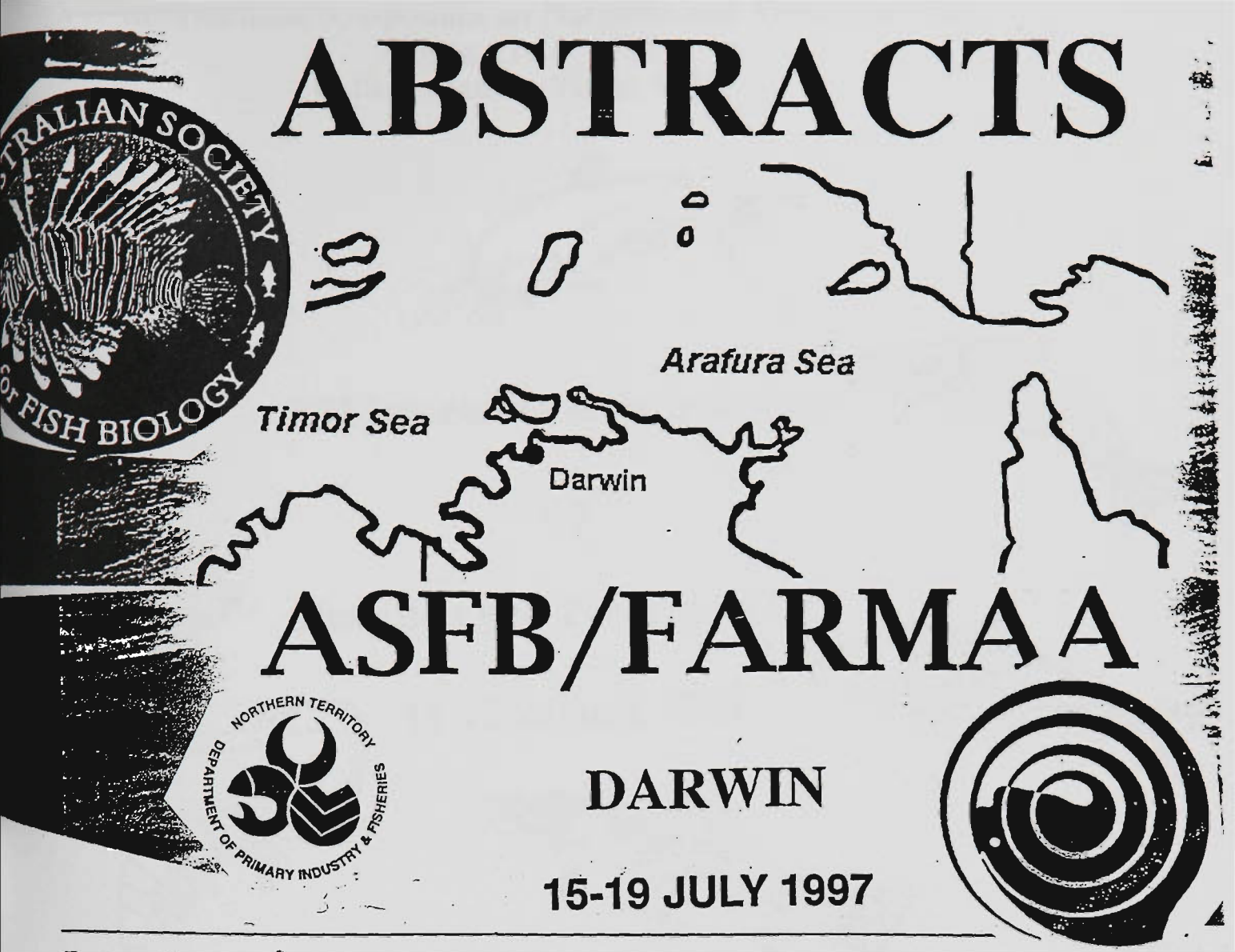
The native fish silver perch *(Bidyanus bidyanus)* were fed on three artificial diets and the hourly and daily patterns of ammonia and orthophosphate discharge at 25°C and 30°C were observed. There was a sharp increase in ammonia and phosphorus production soon after meals (Figure 1 and 2), which decreased linearly during the remaining 24 hours of the day. There was no significant difference between nutrient discharge in the fish fed the three different diets($P>0.05$). The daily ammonia and phosphorus excretion rate was observed increase at the higher temperature (Figure 3 and 4) and was significantly higher at 30°C ($P<0.05$). The daily patterns of ammonia and orthophosphate discharge was also affected by the diets and amount of feed consumed by the fish. The feed that resulted comparatively better nutrient retention in fish discharged less ammonia and orthophosphate to environment.



Poster: 11

Nitrogen pollution from aquaculture : nitrogen losses and nitrogen retention by silver perch, *Bidyanus bidyanus* (Mitchell 1838) (Teraponidae)

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Nitrogen is the most important and an expensive ingredient in fish feed. Nitrogen is required for normal growth of fish, but an abundance of nitrogen may cause nitrogen pollution in both freshwater and marine environments. There are no previous studies on pollution load from silver perch culture although silver perch is a major freshwater aquaculture industry in the country. Silver perch juveniles were reared in glass aquaria and fed on three diets containing 36%, 45% and 53% protein to quantify the nitrogen losses and nitrogen retention at 250C and 300C. The main path of nitrogen loss was found to be via gill excretion (85.7%-90.2%) and via faeces (9.8%-14.3%). The hourly excretion rate of ammonia showed a sharp increase soon after a meals and a linear decrease during the remaining 24 hours. This rate was not significantly different in fish fed on the three different diets at 250C ($P>0.05$). The daily excretion of ammonia was significantly higher at 300C than at 250C ($P<0.05$). A linear relationship was found between the nitrogen intake and the loss of nitrogen in faeces. Faecal nitrogen loss was higher at 300C than at 250C ($P<0.05$). The nitrogen retention by silver perch was found to be significantly greater at 250C than at 300C ($P<0.05$), and the average nitrogen retained at 250C and 300C was 43.2% and 29.5% respectively ($P<0.05$). The higher nitrogen retention at 250C may be related to the better food conversion ratio (FCR) and lower nitrogen loss obtained at 250C with silver perch. The study shows that culture of fish at their optimum temperature may enhance nitrogen retention and a reduction in the discharge of nitrogen to the environment.

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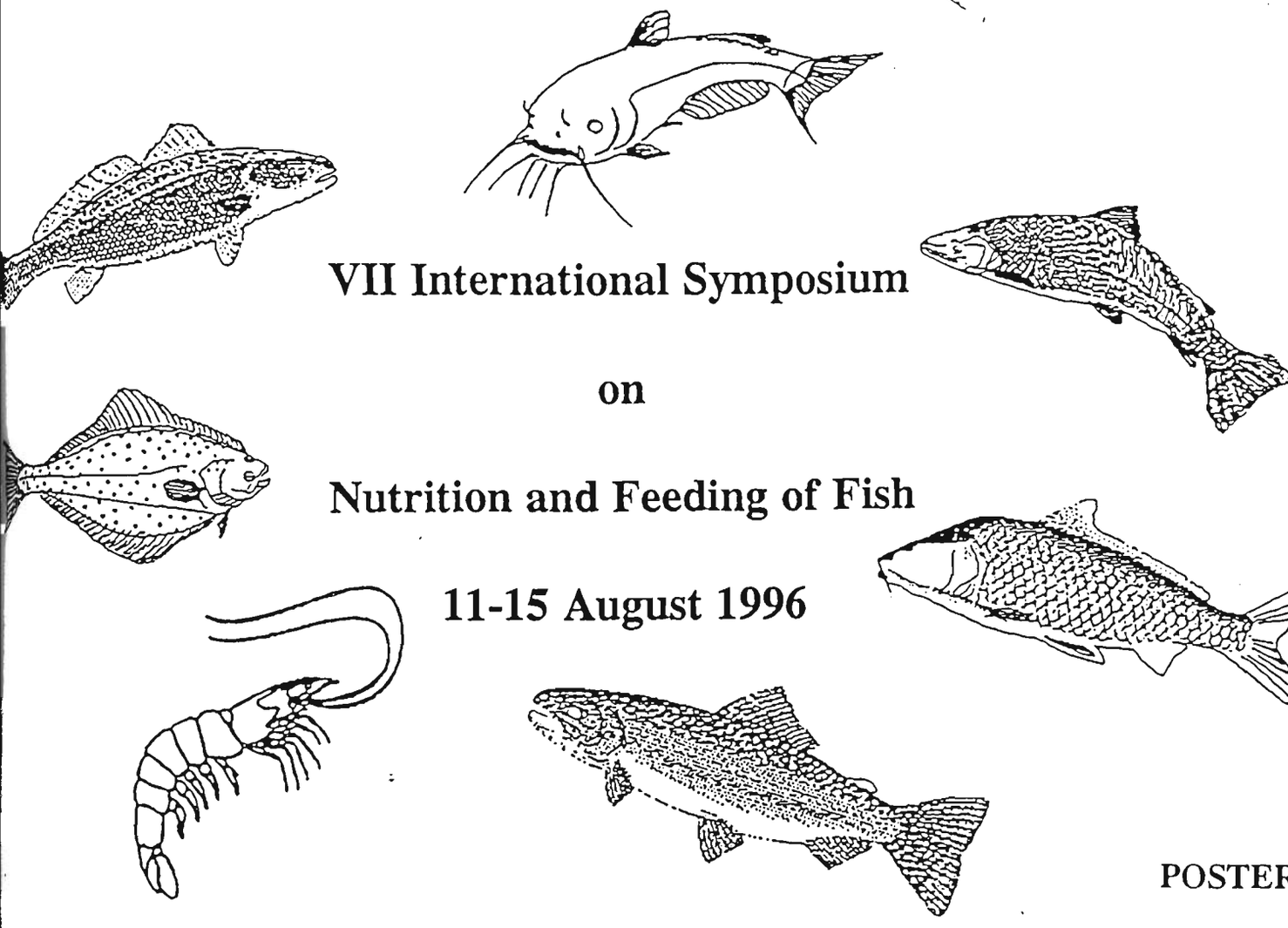

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SEWAGE GROWN ZOOPLANKTON AS AN ALTERNATIVE FEED OF AUSTRALIAN NATIVE FISH SILVER PERCH, BIDYANUS BIDYANUS

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Zooplankton are the essential food of juvenile fish and crustaceans. It contain high percentage of protein and fat. Zooplankton grows abundantly in the nutrient rich Werribee sewage lagoons., Melbourne and the resource is unutilised. The silver perch fingerlings were fed on live, frozen and dry zooplankton in order to evaluate performance of zooplankton based feed in comparision to commercial silver perch diets.

Appendix II. Definitions of abbreviations, terms and units

Acclimatization = the adaptation to a new environment (Rosenthall *et al.* 1990)

Aeration = the mixing of air and water by mechanical means (Boyd & Lichtkoppler 1979; Rosenthall *et al.* 1990);

Ammonia = a form of nitrogen/nutrient found in water and can be toxic to fish, it is the major waste product of protein or nitrogenous metabolism of aquacultured animals (Boyd & Lichtkoppler 1979; Rowland 1996);

Ammonium = positively charged ion resulting from the reaction of ammonia in water, NH_4 (Boyd & Lichtkoppler 1979);

Aquaculture = the farming of aquatic organisms (fish, molluscs, crustaceans, other invertebrates and aquatic plants) using extensive, semi-intensive or extensive methods in order to increase the production or yield per unit volume to a level above that obtained naturally in a particular aquatic environment (Rosenthal *et al* 1990);

Apparent net protein utilization (APNU)
= final body nitrogen - initial body nitrogen / amount of nitrogen consumed (Jauncey 1982);

Ash = essential minerals, non-essential minerals, toxic elements (Hardy 1989; De Silva & Anderson 1995);

Carbohydrates = starches, sugars, cellulose and gums (New 1987);

Crude protein = (N x 6.25), essential amino acids, non-essential amino acids, amines, nucleic acids (De Silva & Anderson 1995);

Crude fat (ether extract)
= the material extractable by ether extraction containing triglycerides, phospholipids, sterols, waxes (New 1987; Devendra 1989; De Silva & Anderson 1995);

Crumbles = small particles produced by cracking pellets and screening (Hardy 1989);

Diet = A controlled mixture or combination of feed ingredients or foods (Hardy 1989);

Crude fibre = indigestible carbohydrates fractions, cellulose, lignin, chitin (New 1987);

Digestibility = the proportion of a feed which is not excreted in the faeces and is assumed to be absorbed by the animal (Devendra 1989);

Dissolved solids = the portion of solids (material) that passes through a standard glass fibre filter (0.45um) and remains after evaporation (APHA 1989; Adams 1990), continuously distributed, ammonia, urea, orthophoshate, phosphorus (Muir 1982; Hennessy *et al.* 1996);

Dissolved oxygen = the elemental oxygen gas contained in a body of water to support the fish life (Boyd & Lichtkoppler 1979);

Dry matter (DM) = refers to moisture-free residue of a sample, which is determined by keeping a sample in an oven at 105^{o}C until it reaches a constant weight (Devendra 1989);

Ecology = the inter-relationships between living organisms and the environment (Clark 1990);

Ecologically sustainable development (ESD)
= development of an natural resource in such a way that the ecology (ecosystem) can continue to function in a natural way (Howes 1995);

Effluent = waste water discharged into water body;

Essential amino acids
= amino acids which are essential to the animal and which the animal body cannot synthesize fast to meet their requirements (New 1987; Devendra 1989);

Eutrophic = lake or water bodies over-supplied with nutrients that promotes excessive growth of algae and other plants (Clark 1990);

Fines = dust and small particles that result from pellet disintegration (Hardy 1989);

Faeces/faecal = excreted from the intestine (Butz and Vens-Cappell 1982);

Feed = a mixture or combination of ingredients (Hardy 1989);

Feed coefficient = the feed consumed per unit weight of increase (Rosenthall *et al.* 1990);

Fish food = any material containing nutrients that can be consumed, absorbed, and used by the body (Hardy 1989);

Fish meal = protein- rich animal feed product based on fish (Kailola *et al.* 1993);

g = gram(s) (Adams 1990);

Hardness = the concentration of calcium and magnesium in a water sample expressed as mg/l of equivalent of $CaCo_3$ (Boyd 1990);

Heavy metals = metals with an atomic mass greater than that of calcium; also known as trace metals (Forftner and Wittmann 1979);

kg = kilogram(s) (Adams 1990);

kPa = kilopascal (Purchase 1997)

l = litre(s) (Adams 1990);

Intake = amount of feed consumed and available for digestion and expressed in DM (Devendra 1989);

Leaching = dissolved and washed out, and the rate of leaching depends upon physical characteristics of waste (Hennessy *et al.* 1996);

Meal = physical form of a feed that has been reduced to a particle size (Devendra 1989);

Moisture = water (New 1987; Hardy 1989);

Moisture content (%)

= $\frac{\text{weight of fresh sample - weight of dry sample}}{\text{weight of fresh sample}} \times 100$ (New 1987);

M = molar concentration or mole or gram molecular weight per litre (Adams 1990; Parsons et al. 1984); moles of solute/litres of solution;

Non-faecal = excreted from the gills, kidney and skin and dissolved in water (Butz and Vens-Cappell 1982);

µg/l = micrograms per litre; 1 x 10^{-6}g or µM/l (Adams 1990; Parsons *et al.* 1984);

mg/l = milligrams per litre; 1 x 10^{-3}g (Adams 1990);

µl = microlitres(s) (Adams 1990);

µm = micrometer (0.001 millimetre or 10^{-3}mm) (Parsons *et al.* 1984);

µmhos / cm = specific conductance / or conductance - micromhos / centimetre (Adams 1990);

ml = millilitres(s) (Adams 1990);

nm = nanometre(s) 10^{-9}m (Adams 1990);

N = normality; gram equivalent/ litres of solution (Adams 1990);

Nitrogen = essential nutrient for both plants and animals;

Nitrite = is an intermediate oxidation state of nitrogen;

Non-essential amino acids

= amino acids which are not needed in the diet but which are essential to the animal. it's main role in diet palatability (New 1987; Devendra 1989);

Nutrient = substance necessary for the growth and reproduction of organisms (New 1987; Boyd & Lichtkoppler 1979);

Orthophosphate or reactive phosphorus

= phosphate that pass through 0.45 µm-pore diameter membrane filter and determines without hydrolysis or oxidative digestion, orthophosphate is readily available to plants (Boyd 1990);

Pellets = a physical form of feed or combination of feeds which are compacted by mechanical means (Devendra 1989);

pH = - log[H^+], it is a measure of the hydrogen ion concentration and indicates whether water is acidic or basis (Adams 1990; Rowland 1996);

Point source = from a specific locations (sewage treatment, dairy sheds, fish farms) (Erskine *et al.* 1995);

Pollution = the entry into a system of any material that will harm that environment or make it dangerous for any organism living in it (Howes1995)

ppt = 1 part per thousand or 1 gram per litre (Kailola *et al.* 1993);

Ration = a share or allotment of feed over 24 hour (Hardy 1989);

Salinity = total concentration of all ions dissolved in a water sample expressed in milligrams per litre or parts per million (Boyd 1990);

Solid waste = discontinuous and physically separable waste, which may leach out further, for example, dust, uneaten food, undigested food, mucus, intestinal cells, bacteria, faeces, scales (Muir 1982; Phillips and Beveridge 1986; Hennessy *et al.* 1996);

Soluble waste = non-faecal loss (Cho *et al* 1991);

Suspended solids = the portion of solids (residues) retained by a glass fibre filter after filtration it provides data on the concentration or load of a particulate matter suspended in the water column(APHA 1989; Adams 1990);

Sustainable = a natural resources/renewable resources that can be replaced naturally (Howes 1995);

Trash fish = fish of no commercial value that have been caught but are discarded (Kailola *et al.* 1993);

Water quality = all the physical, chemical and biological properties of a water body (Boyd & Lichtkoppler 1979);

Zooplankton = microscopic aquatic animals suspended in water (Boyd & Lichtkoppler 1979);