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AN INVESTIGATION INTO THE BEHAVIOUR AND PERFORMANCE OF A  
RECIRCULATING SYSTEM FOR INTENSIVE FISH CULTURE

A thesis towards the  
Degree of Doctor of Philosophy  
submitted by  
G J MANTLE, B.Sc.  
to the Open University

Systems Discipline  
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## ABSTRACT

The role of fish culture in the provision of fish for food is examined with special reference to Western Europe. The use of recirculating systems for intensive fish culture is considered and the need for establishing their feasibility for commercial culture recognised. The lack of suitable guidelines for the design and operation of such systems is seen as a major limitation. A research programme was initiated, resulting in the development of a simulation model.

Firstly a number of recirculating systems were designed, constructed and commissioned and their behaviour and performance monitored. Three areas for further investigation were noted and experiments to consider growth and food conversion, the production and accumulation of wastes and filter performance were conducted. Based on the understanding gained a simulation model was constructed. The model was verified using graph theory, by inspection of standard runs for maximum, minimum, mean and variation and by sensitivity analysis. The model was validated by the comparison of model output with the actual behaviour of the system using a range of stocking densities and ration levels. To improve agreement between predicted and measured diurnal variations in water quality, modifications to the simulation of ammonia production and short-circuiting in the filter were considered.

The use of the model is considered; to indicate areas of inadequate knowledge, to improve understanding of the behaviour of the laboratory systems and to develop guidelines for determining carrying capacity and optimum design of filters for recirculating systems. Use of the model for predictive purposes posed a number of difficulties, and it is concluded that the main value of the model is as an educational tool.

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## 1. THE ROLE OF FISH CULTURE

### 1.1 Fish in the diet

World fish harvests are presently around seventy million tonnes a year, of which twenty-one million tonnes are "industrial" fish used for fish meal and oil, contributing only indirectly to human nutrition (FAO, 1979). This gives an annual average per capita consumption of about thirteen kilogrammes, i.e. five to six per cent of protein consumed in the world and eighteen per cent of the animal protein (Pimentel *et al*, 1975). These global figures, however, conceal important variations in fish consumption. Although fish plays a generally less important role in the diet of developed countries than in that of developing countries, consumption per capita and total is greatest in the former (Table 1.1).

Table 1.1 Food fish consumption 1972/74 (three year average)

	Fish consumption 1972/74		
	million tonnes	%	Kg per capita
Developed countries	19.2	38	25.7
Developing countries	14.0	28	7.6
Centrally planned economies	16.7	34	13.6
World	49.9	100	13.1

After Krone (1979)

Both global and regional increases in the demand for fishery products have been estimated by the Food and Agricultural Organisation of the United Nations (FAO), (Table 1.2). This Table and Table 1.1 cannot be compared directly since Table 1.2 refers to total fishery products whilst Table 1.1 refers only to fish used directly for human consumption.

Table 1.2 Fishery Products: Actual and projected production and demand  
(thousand tonnes)

	Production			Total Demand	
	Actual 1972/74	Projected 1976	Projected 1985	Consumption 1972/74	Projected 1985
Developing Countries	30768	33867	39200	25526	41150
Latin America	6917	8417	8300	3511	5550
Africa	3278	3079	4090	2654	4210
Near East	737	780	1140	718	1370
Far East	10032	11756	13360	9460	15130
Asian CPE	9525	9590	11830	9082	14730
Other Developing Countries	279	245	480	101	160
Developed Countries	37133	39736	38420	42237	56550
North America	3812	4140	5360	5460	6760
Western Europe	11376	12131	11980	13551	16500
E. Europe and USSR	9753	11510	9810	11242	16800
Oceania	195	184	390	331	410
Other	11997	11771	10880	11653	16080
<u>World</u>	67901	73603	77620	67763	97700

Source: FAO (1978)

The production and consumption figures for European countries in Table 1.2 show that during 1972/74, 85 per cent of the demand in fishery products was met by production, but by 1985 this is expected to fall to 65 per cent. This is reflected in FAO projections of international trade in fishery products (FAO, 1978), which show that although during 1972/74 Europe was a net exporter of fishery products, by 1985 it will become a net importer. From Table 1.2 the reduced self sufficiency in fishery products can be seen as the result of both a decrease in production and an increase in demand.

During the period 1970-1976 the percentage of industrial fish caught by developed countries increased from 34 to 38 per cent (FAO, 1978). No breakdown of imports and exports into food and non-food fish for each group of countries is currently available. It is therefore uncertain how much of the predicted increases in demand for fishery products are from increased use of fish meal. Krone (1979) using FAO agricultural commodity projections, predicts that per capita consumption of food fish in developed countries will increase to 28.4 Kg in 1985 and 30.9 Kg in the year 2000. Since Krone's assumptions in making these calculations are not given, these projections should be viewed with caution and are not necessarily representative of individual countries. For example, in the United Kingdom estimates of the contribution of fish to the daily diet for the past ten years show a continuing decrease from a per capita consumption of 8.4 Kg in 1970 to 5.8 Kg (HMSO, 1980).

A number of possibilities exist for increased catches and the FAO estimate a total potential catch from conventional marine resources of 105 million tonnes, with 60 per cent of the increase expected to come from increased fishing effort, and 40 per cent from improved management (reduction of catch discarded at sea) (FAO, 1979). For example, the NE Atlantic is the world's second largest fishery, contributing over 21 per cent of the total fish harvest (FAO, 1979). The potential harvest from the NE Atlantic has been estimated as 14.6 million tonnes a year (FAO, 1979). The harvest in 1977 was 11.8 million tonnes, a utilisation of 81 per cent.

The potential harvest, however, is unlikely to be met. Energy curves of the catch in weight relative to the cost of time spent in fishing, show that



the energy cost in fishing for the last 2 per cent of the fish in a shoal would be about ten times that for the first 98 per cent (Rawitcher and Mayer, 1977). Furthermore some of the species at present under-utilised are found at low densities (FAO, 1979). Increased fishing efforts may therefore be prohibitively high in energy costs.

There are other reasons why the potential harvest is unlikely to be fully met. The concept of maximum sustainable yield (MSY), may be useful but it has to be carefully interpreted (Roedel, 1975). For example, the estimates of potential harvests are based on assessments of stocks of individual species. Where several species inhabit the same area, interaction between species cause the MSY of all species taken together to be lower than the sum of MSY of the individual species estimated separately (FAO, 1979). Many fish stocks need to be harvested below the MSY to maintain a buffer against adverse conditions, especially for shoaling fish such as herring (FAO, 1979). Lastly the concept assumes that no age class is ever over exploited. With the increased capture and processing of trash fish many more immature stages of commercially exploitable species are included.

Globally it would appear that the actual catches will probably be only 80 to 90 million tonnes (van Cleve, 1978). Assuming the weight of fish used for industrial purposes remained the same (at present, 21 million tonnes) 70 million tonnes would be available for direct human consumption. This may meet the predicted demand for food fish in 1985 but would not meet the demand predicted for the year 2000 by Krone (1979). If consumption of fish is to increase as predicted (Table 1.2), alternative sources of fish will be needed. Fish culture makes an increasingly important contribution to harvests in many parts of the world and to many people it seems a logical choice for development, (Parker, 1979; Krone, 1979; Webber and Riordan, 1979; Bell and Canterbury, 1976; Pillay, 1976 and Bardach et al 1972).

## 1.2 The development of fish culture

Fish culture was apparently practiced in China in 2000 B.C., with the earliest known documents on fish farming dating from the time of the Shou-Jin Dynasty, around 1766 B.C. (Kreuzer, 1974). The earliest records of fish culture in Europe are from the Middle Ages when freshwater fish became important to overcome shortages of meat and for religious "fast days". With monasteries acting as centres of research and innovation, fisheries were established around many large towns (Foster, 1924).

Modern fish culture in Western Europe began with the development of artificial spawning and improved rearing techniques for salmonids in the mid-nineteenth century (Huet, 1972). - Initially this resulted in the culture of European Brown Trout to stock rivers and lakes for sporting purposes. The development of fish culture for food occurred much later with the introduction of Rainbow Trout, a North American species more adaptable to domestication.

Whilst food fish culture in Western Europe is dominated by Rainbow trout, throughout the world a wide variety of animals and plants are cultured. A survey (Reay, 1979) suggests that 314 species of fin fish, 74 crustaceans, 69 molluscs, 43 algae, 13 angiosperms, 12 sponges, 9 amphibians, 4 reptiles, 3 rotifers, 2 annelids, 2 mammals and 1 echinoderm are cultured (excluding aquaria or laboratory species). - Latin and common English names of fish mentioned in the text are listed in Appendix 1.

## 1.3 Production from fish culture

"Fish culture" is often used synonymously with "aquaculture". Technically aquaculture is the husbandry of any aquatic organism, including plant culture, while fish culture may be used loosely to embrace all aquatic animals. In this thesis fish culture refers only to fin fish as distinct from shell fish (molluscs and crustaceans) and other smaller groups. The terms "culture" and "farming" are used as interchangeable synonyms and reflect the similarities between agriculture and aquaculture.

Accurate statistics of fish (fin fish) culture do not exist. Quoted figures often fail to distinguish between fin fish and shell fish and between fish for food and fish used for restocking. The most recent world survey was by Pillay (1976) who estimated world aquaculture production to be over 6 million tonnes, of which two thirds (3.98 million tonnes) was fin fish production, the rest being mainly molluscs and seaweeds. This contributed about 10.5 per cent of the total world fish supplies used directly for human consumption (FAO, 1978). Fish production in Europe contributed only about 5 per cent (212,000 tonnes) of world fin fish production in 1975. Compared to the estimated European harvest from capture fisheries over the same period farmed fish contributed only 2 per cent of fish used directly for human consumption. European culture of shell fish exceeds that of fin fish with about 400,000 tonnes produced in 1975 (Pillay, 1976). The combined culture of fin fish and shell fish is therefore of greater significance in the diet, but still only represents a small proportion of the total food supply (Parker, 1979; McAnuff, 1979).

Since Pillay's survey many countries in Western Europe have experienced rapid expansion in production from more intensive methods of culture. In the United Kingdom there has been a large increase in production, mainly from the development of cage culture and semi-closed systems for salmonids. This growth is expected to continue with a production of 15-20,000 tonnes predicted by 1985, (Fisheries Research and Development Board, 1977).

Similar increases have occurred in other European countries, and as Table 1.3 shows, there has been a 40 per cent increase in table trout production in Europe, from 69,000 tonnes in 1976 to 97,885 in 1981.

Table 2.3 Estimated production of table trout in Western Europe

	Tonnes per annum					% change over 5 years
	1976a	1978b	1979b	1980c	1981c	
Italy	17,000	17,480	18,000	20,400	21,500	+26
France	15,000	18,000	18,000	20,000	21,000	+40
Denmark	15,000	17,500	14,000	17,500	18,000	+20
Germany	10,000	7,500	7,000	11,000	12,000	+20
Spain	6,000	7,800	9,000	10,000	12,000	+100
Finland				4,800	5,400	+13@
Great Britain	2,700^	3,300^	4,650^	4,400	5,200	+96
Norway	1,800	2,200	3,000	3,360	4,000	+122
Austria	1,000	1,300	1,300	1,500	1,600	+60
Switzerland	400	400	400	1,500	1,500	+275
Eire	400	400	400	570	785	+96
Belgium	400	300	300	300	300	-25
Total	69,700	76,540	76,050	90,530*	97,885*	+40

^ Includes values for Northern Ireland

@ Percentage change over two years

\* Excluding values for Finland

Sources: a) Lewis, 1979

b) Lewis, 1980

c) Federation for European Salmonid Culture, 1981

## 1.4 The efficiency of fish culture

A study of the role of fish culture in food production should take into consideration not only output but also efficiency of production. Efficiency can be measured in numerous ways, but perhaps two of the most important are :-

- 1) food conversion efficiency
- 2) efficiency of energy use.

### 1.4.1 Food conversion

Since fish are poikilothermic, the energy requirement for metabolism is less than in warm blooded animals, as they do not have to maintain a constant body temperature. Fish are supported by the water which surrounds them and hence exert relatively little energy to maintain their position in the water, whereas land-based animals must use energy to develop and use a strong skeletal system, and support themselves against gravity. Consequently fish are efficient converters of food to flesh with food conversion ratios frequently in the range of 0.7 to 1.3 as compared to values of 3 or more obtained for poultry and other livestock. However, fish can assimilate and are fed rations with a much higher protein content. For example trout pellets contain 40 to 50 per cent protein compared to 20 per cent for poultry feeds. A large proportion of the protein contained in fish diets is used for energy; Austreng (1976) and Phillips (1969) have estimated that between 50 and 70 per cent of the available energy in trout pellets is supplied by protein. Thus when conversion of diet protein to body protein is compared, the margin in efficiency between fish and other livestock is reduced (Windsor and Cooper, 1977). An increase in the fat content of fish diets appears to have a sparing effect on the protein, increasing the efficiency of protein utilization (Jauncey, 1979).

Most fish culture in Western Europe is based on carnivorous species of fish, typically rainbow trout, fed on high protein diets based on fish meal. Fish meal is produced from low grade industrial species and also from fish wastes and offal produced in processing (Barlow, pers. comm.). This type of fish culture is therefore a highly efficient converter of protein in one form to another of higher quality. However, both in terms of

production costs (Varley, 1977) and support energy (Edwardson, 1976), fish meal based feeds are expensive, and considerable research effort is being directed to find alternative feeds (Halver & Tiews, 1977).

The majority of European fish production takes place in Eastern Europe (Pillay, 1976) where it is based on the culture of cyprinids. These fish are omnivorous and derive much of their food from the natural productivity of ponds and lakes. It is more difficult to examine the efficiency of fish culture based on natural productivity since the inputs are not easily quantified. Production in such systems is often enhanced by fertilisation and/or supplementary feeding. Some gross relationships between yield and fertiliser addition have been calculated for Israel (Wohlfarth and Schroeder, 1979), but none are available for European production. Supplementary feeds are often used in carp culture, and in the Netherlands a conversion ratio of 5:1 (5 Kg of cereal added to a pond increases production by 1 Kg of fish) is used as a rough working estimate (Breteler, pers. comm.).

#### 1.4.2 Support energy use

Table 1.4 compares the support energy use per unit of protein produced in a range of fish culture systems with that in other forms of animal production.

The wide range of values presented by different authors make comparisons difficult, particularly because of different methods of energy accounting (Spedding et al, 1981). In addition the efficiency of support energy use in fish farming varies considerably with the culture methods and climatic conditions (Edwardson, 1976). It is therefore necessary to restrict comparisons to similar cultural methods. Thus the intensive farming of fish as practised in most of Western Europe should be compared to other intensive culture systems, particularly those based on feeds supplied by the farmer. When such comparisons are made it can be seen that pond farming of carp with supplementary feeding and fertilisation has a similar support energy use to egg and broiler production while trout is more comparable to beef and lamb production. Catfish farming in the USA is one of the highest energy users and has closest similarity to feedlot beef production.

Table 1.4 support energy use in animal protein production (MJ/Kg protein)

	Pimentel et al (1975)	White (1975)	Leach (1976)	Spedding et al + (1975)	Rawitcher & Mayer (1977)	Edwardson (1976)
Trout(UKponds)						389
Carp German ponds						250
Japanese recirc- ulating system						3000
Catfish (USA)	580					891
Herring			489*		29	26
Cod			489*		329	309
Flounder			489*		399	
Eggs	220	200	353			
Broiler	369	203	290			
Pork	593	238				
Pig & Poultry			235- 331			
Milk	608	118	208	135		
Beef (rangeland) (feedlot)	169 1300	348		600		
Lamb (rangeland)	271	465	263- 428	397		
Cattle & Sheep			101- 289			

Notes:- + Support energy used per Kg yield of dietary protein  
\* Average value for all UK fishing

The support energy costs of the Japanese recirculating system calculated by Edwardson (1976) are extremely high. The farm studied by Edwardson was for carp rearing. Typically in such a farm the carp are grown in a closely controlled environment with heat used to achieve good hatchery success and high fingerling growth rates. When the fish have passed their most vulnerable stages they are transferred to ponds. Thus when the energy costs over the whole production cycle are averaged, the support energy use is considerably reduced. In the U.K. where carp spawning is unpredictable and hazardous (Bryant, 1981) a similar practice has been found economically viable (Jaffa, pers. comm.).

As indicated in Table 1.4 the energy costs of producing fish to market size entirely in a recirculating system would be high unless the fuel costs could be minimised by the utilisation of a heated effluent (section 1.6). With no heating and low pumping costs the support energy use could approximate that of U.K. trout farming.

Compared with the harvesting of sea fish, fish culture appears on a par with deep sea fishing, but of an order of magnitude greater than inshore fishing. This comparison is of limited value, however, since fishing is more akin to hunting than to farming.

## 1.5 Constraints to development

### 1.5.1 Market limitations

From the data presented in Table 1.3 it can be seen that the largest increases in production came from the smaller producing countries whilst the larger producers experienced more modest growth. One reason for the lower rate of increase of the larger producers is that the home market for the fish produced has largely been filled (Anon, 1981). Any increase in output must be exported or the home market expanded. Of the countries listed in Table 1.3 only Austria, Belgium and Germany consume more trout than they produce. As more fish are produced so competition increases. In the United Kingdom, Needham (1981) has commented that "only the fittest will survive the current trading conditions for long" and calls for



innovation in two areas; better marketing to increase consumer demand and a reduction in costs. In particular he singles out feed costs which are the largest single variable operating costs (Varley, 1979).

### 1.5.2 Water demands

In semi-closed culture systems the density of fish cultured is determined by the flow through the holding facility, that is, the rate at which oxygen is replenished and waste products such as ammonia are removed. The high density of fish typical of intensive trout production depends on large quantities of high quality water. For example, a unit with an assumed production of 100 tonnes of trout would on average require a water flow of 46 million litres a day. This is equivalent to the domestic requirements of a town of 30,000 people (McAnuff, 1979).

Suitable water supplies are limited and their availability is a major constraint to production. The different rates of expansion experienced between countries (Table 1.3) to some extent reflects the number and availability of suitable water supplies for fish culture. Many of the smaller producers are still in an early stage of development, and have suitable water supplies. The larger producers have a longer history of production and most suitable sites for fish culture have already been developed. Continued growth in these countries depends primarily on the expansion of production at existing sites.

In the United Kingdom Lewis (1980) found that a lack of suitable sites for large fish farming operations resulted in the majority of newly established fish farms planning for an annual production of 20 tonnes or less. Similar observations were made by Munro and Waddell (1981) in a survey of Scottish fish farms.

### 1.5.2<sup>3</sup>1 Pollution from fish farm effluents

Where suitable water supplies are available a number of fish farms may develop on adjacent sites. On the River Wylve in England, for example, three fish farms have been built along a 6 Km stretch since 1977/78, (Frake, pers. comm.). In other European countries too, "strings" of fish farms have developed (Alabaster, 1982). Such heavy usage of a single water

resource can pose problems, particularly since the effluent discharged from fish farms can cause numerous changes in the receiving water (Mantle, 1982).

With the growth and development of fish farming, increasing attention is being paid to the control of pollution from fish farm effluents. Table 1.3 shows that in 1979 overall production fell. This was the result of a large fall in production in Denmark and Germany, two of the largest producers. The main reason for this sudden decline was the introduction of anti-pollution legislation governing the quality of effluent discharged by fish farms (Warrer-Hansen, 1979). Subsequently many farms were involved in heavy expenditure on effluent treatment systems for their large volume throughputs. Farms employing recirculation largely avoided this problem because of the treatment already carried out. In addition, with their relatively low volume of waste, conforming to new legislation was easily and cheaply accomplished. With the likelihood of anti-pollution legislation being introduced in other countries, interest in recirculating systems has increased.

### 1.6 Heated effluents

An area of development which at present makes only a minor contribution to fish supplies, but which may become important, is the use of heated effluents. There are numerous sources of warm water available (e.g. power stations, breweries and distilleries) and a growing interest in using these waters for the culture of carp, eels and other warm water species. Hambrey (1980) has estimated that in the UK power stations alone, there is 10,000 m<sup>3</sup>/day of heated effluent available for fish culture.

Most European countries are developing fish culture operations to utilise the large amounts of heated effluents available. Many different species are being cultured, from salmon and trout in Finland and Norway (Kittelsen and Gjearum, 1981 and Sumari and Westman, 1981) to catfish, mullet and shrimp in Italy (Bogese and Smedile, 1981), eels and turbot in Germany (Weinbeck, 1981) and tilapia in Belgium (Melard and Philippart, 1981). In the UK several large companies including RHM, Tomatin Distillers, Blue

Circle Cement, Coats-Paton, the Central Electricity Generating Board and the White Fish Authority are experimenting with warm water fish culture using waste heat.

The use of warm water has several advantages. An increase in temperature up to the optimum for the species will accelerate growth rate and improve feed conversion. This increases the rate of production. For instance raising the temperature from 11°C to 15°C can reduce production time of rainbow trout from 12 to 8 months. This is equivalent to a 50 per cent increase in farm output (Walsingham, 1980). Use of warm water also allows production to take place all year round. This smooths production cycles ensuring continuity of supply and improved marketing opportunities, and hence results in better utilisation of holding capacity (Hambrey, 1980).

In temperate countries the use of warm water is necessary for the spawning of exotic species such as grass carp, a species native to Asia. At present this fish is exported into the UK for use in biological weed control (Solomon et al, 1975). Spawning has been successfully induced by the use of heated water and hormone injections (Stott and Durbin, 1980) and good growth rates achieved for the fry and fingerlings. In Sweden where grass carp are also used in biological weed control, a 15 tonne production unit utilising heated effluent and employing a recirculating system has been established to supply the estimated yearly demand, (Kossman, 1981).

There are a number of problems associated with the use of heated effluents. The output of many power stations fluctuates with power demand and therefore supplies are not assured, (Shearer, 1982). In addition the thermal effluent may suffer marked seasonal (and even daily) variations in temperature, and contain a large amount of suspended solids (Aston et al, 1976). Base load power stations offer the best possibilities for continuous supply (Aston, 1981) and in the United Kingdom most fish farms are associated with this type of power station. Many other industrial processes have supplies of warm water as a by-product, but the water is seldom of suitable quality to be directly used for fish farming. Even where the quality is adequate or when the heat can be exchanged, rarely is it available in sufficient volumes for flow-through circulation, and virtually never is it available on a continuous basis (Shearer, 1982).

Most fish farms utilising heated effluents have overcome these problems through the use of a recirculating system, allowing both water and heat to be conserved and a constant temperature maintained.

### 1.7 Conclusions

Although the farming of fish is still in an early stage of development (McAnuff, 1979) and its contribution to food supplies is negligible, it is expanding rapidly. Fish farming represents an efficient conversion of protein from one form to another of higher quality. The main weakness is its use of high protein feeds based on fish meal. Such feeds are expensive both in terms of support energy (Edwardson, 1976) and production costs (Varley, 1977). Continuing economies should allow production costs to fall thus making farmed fish a highly competitive product (Parker, 1979; McAnuff, 1980). In the long term the development of fish culture could remove uncertainty and instability from domestic fish markets (McAnuff, 1979), increase the variety of fish products available to the consumer (Parker, 1979) and make an increasingly important contribution to the diet. Future development of fish farming may be constrained by market limitations, the availability of suitable water supplies and the introduction of anti-pollution legislation governing the discharge of fish farm effluents. Interest in the use of recirculating systems to overcome these problems has been considerable. McAnuff (1979) for example, feels that "the productive potential of U.K. fish farming could be transformed by the development of fully effective low-cost systems of water recirculation". In addition, recirculating systems offer a number of other benefits, and can help take full advantage of heated effluents.

## **CHAPTER TWO RECIRCULATING SYSTEMS**

### **2.1 Introduction**

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### **2.3 Water Quality**

#### **2.3.1 The effect of fish on culture water**

#### **2.3.2 The effect of culture water on fish**

#### **2.3.3 Treatment processes**

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## 2. RECIRCULATING SYSTEMS

### 2.1 Introduction

Recirculating systems have only been used in commercial aquaculture for some thirty years, although they have been used by aquarists since the last century (Spotte, 1970), with the earliest recorded use being for the U.S. Fish Commission's aquaria at the Columbian World Exposition (Forbes, 1893).

Recirculating systems have become a useful tool in research, enabling a wide range of organisms to be cultured in a closely controlled environment (McCrimmon and Berst, 1966; Guidice, 1966; Wohlfarth and Lahman, 1971; Scott and Gillespie, 1972; Sidall, 1974; Valenti and Aldred, 1975 and Poxton et al, 1980).

The possibilities of employing recirculating systems in commercial aquaculture were first examined in the 1950's by Japanese eel and carp farmers (Bardach et al, 1972). These systems were based on designs developed for public aquaria (Saeki, 1958). This was followed in the 1960's by the use of recirculating systems in U.S. Federal salmon hatcheries (Mayo, 1979).

The documentation of these systems together with other literature which appeared at the time, helped to stimulate the development of recirculating systems that has taken place over the past ten years. Recirculating systems are currently used in a wide variety of applications. Many of these have been reviewed (Mayo, 1980; Berka et al, 1980; Chiba, 1980; and Mayo, 1979).

The diversity of application as well as language difficulties has resulted in the inconsistent use of terms in reporting research and operational data (Coche, 1981). To avoid confusion therefore, definitions of terms commonly encountered are given in the following section.

## 2.2 Definition of Terms

Recirculating system: All or part of the water removed from a fish culture unit is returned for re-use after some form of reconditioning. If the only addition of water to the system is solely to replace losses from evaporation and splashing, the recirculating system may be considered a closed system; (with respect to water). More commonly additional water enters the recirculating system (flush) displacing an equivalent volume for discharge.

Re-use system: A vague term sometimes used synonymously with recirculating system but also used to describe a series of fish culture units served by the same water with some limited treatment, usually just aeration between units.

Treatment: Used to describe any process which improves the quality of the water. Often divided into pre-treatment, reconditioning and post-treatment.

Pre-treatment: Processes applied to incoming water before it is used for fish culture if it is not of sufficiently high quality.

Reconditioning: Processes used to return the used culture water to a condition suitable for re-use. The quality of water in a recirculating system is primarily dependent on the performance of the reconditioning unit. Since some recirculating systems may employ only reconditioning, the term "reconditioning system" has been used synonymously with "recirculating system", although this practice is not endorsed here.

Post-treatment: Post-treatment is applied to the waste waters discharged from a fish culture system to prevent pollution of receiving waters. In a recirculating system it may be the same as the reconditioning treatment, but in a semi-closed system, it may be the only form of treatment employed.

Make-up water: The additional water (new) added to a recirculating system to replace losses from evaporation and splashing, i.e. this does not displace any water from the system.

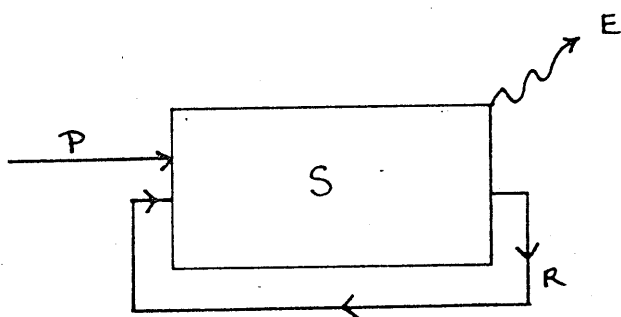
Flush: Water added to a recirculating system above that required for make-up, i.e. displaces an equivalent volume.

Flow: The total volume of water passing through a fish culture unit. In a recirculating system, this is the recirculated volume together with any flush.

The differences between flow, flush and make-up can be seen clearly in Fig. 2.1.

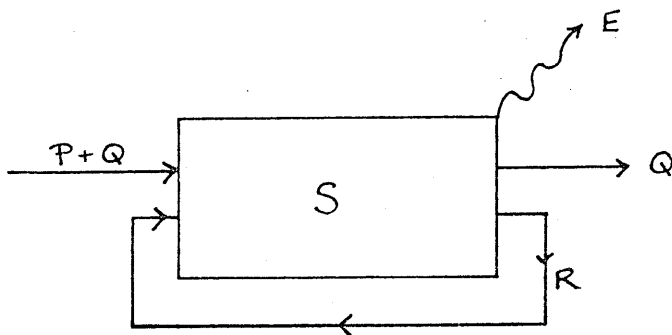
Figure 2.1

Recirculating system with make-up water only



P = make-up  
 R = recirculating  
     volume  
 E = Evaporation  
 P = E  
 S = Total flow through system  
     = R + P

Recirculating system with flush



Q = flush  
 S = R + Q + P

Ammonia: There is some confusion in the terminology used to describe the concentration of ammonia. Throughout this thesis, the terms "ionised" ( $\text{NH}_4^+$ ) and "un-ionised" ( $\text{NH}_3$ ) will be used to describe the two states of ammonia in water, and "ammonia" will refer to the combined concentrations of ionised and un-ionised ammonia ( $\text{NH}_3 + \text{NH}_4^+$ ). Concentrations of ammonia will be expressed as mg  $\text{NH}_3/1$  or as  $\text{NH}_4^+\text{-N}/1$ . In the laboratory total ammonia was measured and is expressed as  $\text{NH}_3/1$ .

Percentage recirculation: Use of this term has been frequently made in



the literature without clear definition. In America the term is generally applied for each turnover, regardless of the time required, while in Europe it is generally applied to the water mass exchanged in the system per day, regardless of the number of cycles during the time period. To avoid confusion in this thesis, the term has been avoided.

Carrying Capacity: The term carrying capacity lacks precise definition but is usually taken as the weight of fish at which water quality deteriorates to an unacceptable level. Here, carrying capacity in a recirculating system is defined as the weight of fish and feeding regime which produces a waste loading greater than can be accommodated by the treatment processes employed.

### 2.3 Water quality

The maximum weight of fish which can be cultured in a given water supply (the carrying capacity) is governed by three factors:-

- a) the concentrations of oxygen and pollutants acceptable during culture,
- b) the rate of oxygen consumption and pollutant production,
- c) the rate of oxygen renewal and removal of pollutants.

In a flow through system oxygen is renewed and pollutants removed by the flow of water through the system. In a recirculating system the rate of removal and reoxygenation is determined by the efficiency of the treatment employed and the rate at which partial water changes are made.

The most important changes in water quality are those which occur rapidly and those to which the fish are most sensitive. The first limiting factor is the concentration of oxygen (Meade, 1974). Addition of oxygen to a system can increase the carrying capacity until ammonia concentrations become limiting. Where ammonia toxicity is reduced by low pH, addition of oxygen can increase carrying capacity up to ten fold before the concentration of suspended solids becomes limiting (Harman, 1977).

The three main functions of the treatment employed in a recirculating system are therefore reoxygenation, ammonia and solids removal. On a

longer time scale, other parameters such as pH and nitrates may become important (experiment 3.3.3.2). Some recirculating systems avoid problems with these by maintaining a constant level of flush or by occasional changes of the water. Achieving a high carrying capacity is usually a major objective in a recirculating system. It is therefore important in operating a recirculating system to have some indication of the many changes in water chemistry that occur (section 2.3.1), the effects these have on the fish (section 2.3.2) and the treatment processes available (section 2.3.3).

Formulae for calculating the carrying capacity of flow through systems have been prepared by a number of authors (Haskell, 1955; Willoughby, 1968 and Piper 1970), mostly based on the levels of oxygen. The formulae have limited application in determining the carrying capacity of recirculating systems since they do not consider the effects of a build up of metabolic products. Equations for determining carrying capacity in recirculating systems have been produced by Liao and Mayo (1972), Speece (1973) and Hirayama (1974), and these form the basis of their design methods (section 2.4).

### 2.3.1 The effect of fish on culture water

Major changes in water chemistry result during the culture of fish. A summary of the quantitative relationships concerning respiration and waste production rates are considered in chapter four.

Respiration: Respiration rates vary during the day (Smart, 1980 and Brett and Zala, 1975), being related to the rate at which metabolic processes take place, temperature and the level of activity. Factors shown to have an effect on respiration are salinity (increasing with increasing salinity) and size of fish (higher for smaller fish of the same species). Free carbon dioxide in the water may depress the affinity of fish blood for oxygen, hence reduced consumption of oxygen occurs with increasing levels of carbon dioxide (Basu, 1954). A number of authors have related oxygen consumption to the quantity of feed given to the fish (Willoughby, 1968; Piper, 1970 and Liao and Mayo, 1972).

Nitrogenous metabolic wastes: Ammonia is excreted through the gills as un-ionised ammonia ( $\text{NH}_3$ ) (Forster and Goldstein, 1969). Other excretory products are urea, creatine and amino acids (Baldwin, 1949). The relative amounts of the various nitrogenous excretory products varies in response to stress (Burrows, 1964; Olson and Fromm, 1971 and Brett and Zala, 1975). Estimates of the total nitrogen excreted as ammonia varies from 25-50 per cent (Saeki, 1958) to 90 per cent (Forster and Goldstein, 1969). Excretory rates follow a fairly regular diurnal pattern (Rosenthal, 1980; Harman, 1977 and Brett and Zala, 1975). Since ammonia is the principle waste product of protein metabolism, its production is influenced by the energy/protein ratio of the diet and the digestibility of the protein. Meade (1974), described the relationship between the production of ammonia to the level of dietary protein, while other authors have related ammonia production with total feed for a variety of species (Westers and Pratt, 1977; Liao and Mayo, 1974 and Speece, 1973).

Other nitrogenous chemicals found in culture water include nitrite and nitrate. These can accumulate in a fish culture system through the biological oxidation of ammonia, although in surveys of flow-through fish farms in the UK Purdom (1982) and Solbe (1982) found that nitrate levels may sometimes decrease during passage through a fish farm.

Solid wastes: These comprise of faeces and uneaten feed (Liao, 1970), although there may be some other proteinaceous material (Spotte, 1970). Relationships between suspended solids, biological oxygen demand (BOD) and chemical oxygen demand (COD) with feed have been produced by a number of authors for different fish culture systems (Queralou, 1981; Liao and Mayo, 1974; Speece, 1973; Willoughby *et al*, 1972 and Murphy and Lipper, 1970). The daily production of faeces depends on the feeding pattern of the fish and the gastric evacuation rates (Fischer, 1979). Management practices have a major influence on the production of solid wastes (Alabaster, 1981), with different levels of food wastage and dust associated with particular feeds (Harman, 1977). The amount of uneaten feed also depends on the species of fish; some species e.g. eels, being "messy" feeders. Wastage of feed is obviously undesirable from the producers point of view, yet studies on fish farm effluents have shown that uneaten feed is a major source of solid wastes (Warrer-Hansen, 1979 and Liao, 1970).

Phosphorus compounds: The main phosphorus compounds found in culture water are phosphates, excreted through the kidneys (Forster and Goldstein, 1969). Other phosphorus compounds may arise from leaching of food and faeces. No data are reported on the toxicity of phosphorus compounds, since concentrations reached in culture water are usually low. Their main effect is as a pollutant, causing eutrophication, particularly in oligotrophic waters (Alabaster, 1981).

Dissolved organics: Some dissolved organic compounds are ubiquitous in natural waters and may include soluble humin or dissolved humic acids of plant origin (Wickins, 1980). Other sources may be pigments found in fish feeds, while the fish themselves excrete organic compounds into the water (Spotte, 1970).

The organic compounds which accumulate in recirculating systems are highly resistant to biological degradation, and increase the COD of the water. They can be broken down by ozone (Otte et al, 1977), although this increases the BOD and may release heavy metals into the water.

### 2.3.2 Effect of culture water on fish

Aquatic animals raised in artificial culture systems may sometimes exhibit what are often regarded as typical symptoms of captivity; osmotic imbalance, thermal shock, partial asphyxiation, stunting and loss of fecundity. These and other ailments are often the tangible results of deteriorating water quality (Spotte, 1970). In fish culture tolerance of water quality may be judged in terms of growth rate, food conversion ratio, disease resistance and flesh quality (Wickins, 1981).

Water quality requirements based on physiological needs and tolerances have been established for a number of freshwater fish and criteria for maintaining freshwater fisheries have been established (Alabaster and Lloyd, 1980). However, the majority of published literature on which these criteria are calculated, are based on acute lethal rather than on the long term chronic effects (Wickins, 1981; Harman, 1977). For many water quality characteristics the minimum levels acceptable in fish farms are higher than those required to maintain natural populations (EIFAC, 1980). Recently water quality criteria have been proposed for intensive

aquaculture by Wickins (1981).

A summary of water quality criteria for freshwater fish culture is presented in Table 2.1. It should be emphasised that these are "safety" levels rather than optimum levels.

Table 2.1 Water quality criteria for freshwater fish culture

Parameter	Max. acceptable level	Min. acceptable level	Ref.
Oxygen	110% saturation (total dissolved gas pressure)	5 mg/l 3 mg/l	Salmonids carp, eels and tilapia a, b
Ammonia	0.0125 mg NH <sub>3</sub> /l 0.1000 mg NH <sub>3</sub> N/l (0.1220 mg NH <sub>3</sub> /l)		c b
Nitrite	0.1 mg NO <sub>2</sub> -N/l	Salmonids	b
Nitrate	100 mg NO <sub>3</sub> -N/l		a, b
pH	8 - 9	6.0 - 6.5	b, d
Carbon dioxide	10 mg CO <sub>2</sub> -C/l (22 mg CO <sub>2</sub> /l)		e
Hardness		25 mg/l as CaCO <sub>3</sub> ;	b
Suspended solids	25 mg/l (high level of protection) 25-80 mg/l (moderate level of protection) 80-400 mg/l (low level of protection)		f
Dissolved organics	no guidelines available		
References	a) Amlacher, 1970 b) Wickins, 1980 c) EIFAC, 1970 d) Alabaster and Lloyd, 1980 e) Smart, 1980 f) Munro, 1978		

Dissolved oxygen: The effects of supersaturated water are not clear, with conflicting evidence concerning the depression of growth (Douderoff and Shumway, 1970; Stewart et al, 1967 and Swift, 1963). Supersaturated levels are unlikely to be lethal except when "gas bubble disease" - where bubbles of nitrogen gas form under the skin - occurs as the partial pressures of gases in water falls rapidly and gases in the blood are freed as bubbles. This may arise with supersaturated levels of dissolved oxygen, since oxygen levels can fall rapidly (Amlacher, 1970). The minimum acceptable levels are those necessary for the maintainance of good growth rates. Lower levels can tolerated, but only for short periods of time.

Ammonia: Only the un-ionised ammonia fraction ( $\text{NH}_3$ ) appears toxic (Forester and Goldstein, 1969). Therefore harmful effects of ammonia depend on pH, temperature, carbon dioxide and alkalinity, since these affect the equilibrium that exists between the ionised and un-ionised fractions (Emerson et al, 1975). The level recommended by EIFAC, (1970) as the maximum tolerable concentration for long term exposure is widely used in fish culture. Recently Wickins (1981) has suggested that this level may be appropriate for natural populations but a level five times greater is acceptable in fish farming.

Nitrite: High levels of nitrite result in methemoglobinaemia. Toxicity is influenced by fish species, size and length of exposure (Russo et al, 1974). There is limited data for non-salmonids. The earlier suggested toxicity levels were much higher; Amlacher (1970) suggests 10-20 mg/l as being toxic.

Nitrate: There is little evidence to suggest that at the levels typically experienced in fish culture operations nitrate is toxic. Levels up to 1,800 mg/l have been reported, without any apparent effect (Naegel et al, 1976). Wickins (1981) and Amlacher (1970) recommend levels should not exceed 100 mg/l.

pH: Most fish are not seriously affected by pH within the range 5-9 (Alabaster and Lloyd, 1980). Very high or very low pH can result in alkalosis or acidosis (Amlacher, 1970). At pH levels below 6.0-6.5 in freshwater problems related to elevated carbon dioxide levels can arise (Wickins, 1981).

Carbon dioxide: High levels of carbon dioxide can cause the condition known as nephrocalcinosis (Smart et al, 1978). Wickins (1981) and Munro (1978) recommend a maximum acceptable level of 6 mg CO<sub>2</sub>-C, while Smart (1980) suggests 10 mg CO<sub>2</sub>-C as tolerable in well aerated water.

Hardness: Although having no direct action on the fish a minimum level is desirable to provide buffering against rapid changes in pH. There is some evidence to suggest that hardness equivalent to 25 mg/l as CaCO<sub>3</sub> provides some protection to salmonids against nitrite poisoning (Wickins, 1981).

Suspended solids: There is little evidence of direct toxicity affects arising from the level of suspended solids, but it is thought that they may have some effect through gill damage and skin and fin erosion, and may be a precursor to gill disease (Harman, 1977). Few studies have been conducted to examine the maximum acceptable levels of organic suspended solids and suggested levels are somewhat arbitrary. Muir (1975) recommended a maximum level of 20 mg/l, while Wickins (1981) recommends a maximum of 15 mg/l. Munro (1978) has recommended a level below 25 mg/l should be maintained for a high level of protection against mechanical gill damage.

Dissolved organics: The effects of a high level of dissolved organic compounds can be two fold. Firstly, a high level encourages the development of heterotrophic bacteria. These may be pathogenic and can reduce the oxygen available to the fish. Secondly, some of the compounds excreted by the fish may be physiologically depressing pheromones (Spotte, 1970). No guidelines have been produced for the maximum acceptable levels in fish culture.

### 2.3.3 Treatment processes

Treatment processes can be classified into the following; mechanical filtration, gravitational separation, biological filtration, chemical filtration, aeration and disinfection (Table 2.2). Reviews of the different treatment processes have been made by a number of different authors (Mantle, 1981, 1980; Berka et al, 1980; Liao, 1981; Muir, 1977 and Wheaton, 1977).

Table 2.2 Treatment processes

Mechanical filtration

screens  
sand filters  
diatomaceous earth filters

Gravitational separation

lagoons  
swirl separators  
sedimentation chambers/tanks

Biological filtration

filters - trickling  
          - submerged  
          horizontal  
activated sludge  
extended aeration  
fluidised bed  
rotating discs/drums  
denitrification

Chemical filtration

ion exchange  
foam fractionation  
activated carbon  
de-gassing  
pH control  
coagulation

Disinfection

ozone  
chlorine  
ultra-violet light  
shocking

Aeration

gravity aerator  
surface aerator  
diffuser aerator  
oxygen injection  
ozone

The treatment employed in a recirculating system depends partly on the level of reconditioning required. In closed systems or systems with a low rate of flush, comprehensive reconditioning is necessary and a number of treatment processes may be used together.

In the majority of recirculating systems, the treatment processes are combined sequentially. This means that the possibly differing optimum conditions for each treatment are not usually achieved, although it is possible to suggest an order for combining treatments to obtain the most effective performance of the individual processes (Muir, 1977 and Wheaton, 1977).

A recent trend has been to have various treatments in parallel, fed directly with water from the culture unit. In this way, each treatment process can be managed to achieve optimum performance (Rosenthal, 1981).



This may overcome any disadvantages of having treatments in series, but some processes benefit from prior treatment. For example, since nitrification requires an adequate supply of oxygen, it benefits from a prior reduction of solids as these increase the BOD.

Through discussions with research workers and operators of recirculating systems, and from the diverse range of designs documented in the literature, it is plain that there is no common agreement on technical or economic grounds, as to the most effective reconditioning treatment for use in intensive fish culture. Many of the treatment processes used have been unsuited to the waste strengths, variability and outlet limitations involved in fish culture. The most suitable treatment processes for use in a recirculating system are those effective at low concentrations and adaptable to the wide variations in input levels typical of many fish culture operations. The reconditioning unit should be simple in operation and easily managed and maintained. It should also be easily modified by the operator to optimise performance, and have low capital and running costs. Finally, the system should be stable, not subject to sudden changes in performance, and it should be reliable, since a single malfunction could prove fatal to the fish. At present no recirculating system employs treatment which fulfills all of these requirements.

#### 2.4 The design of recirculating systems

The design of the best system for any purpose should be carried out rationally. An aid to this process is the methodology of de Neufville and Stafford (1971). The methodology has five stages:-

- 1) definition of objectives,
- 2) formation of measures of effectiveness,
- 3) generation of alternatives,
- 4) evaluation of alternatives,
- 5) selection.

The discussion here centres on the first part of the methodology.

Definition of objectives: Associated with any system with a defined purpose is a set of objectives. The relative importance of each will vary

with the overall purpose of the system, as shown in Table 2.3, and some objectives may conflict.

The aim of the methodology is to allow the selection of a design which best achieves the set of objectives. It is therefore necessary to assess the effectiveness of alternative designs, using agreed measures of effectiveness.

Table 2.3 Primary objectives of recirculating systems

Objectives	Recirculating systems			
	A	B	C	D
Increase production/water use	+	+		
Reduce energy consumption	+	+	+	
Increase environmental control			+	+
Increase growth rate	+			
Increase stability of water quality			+	+
Increase control of disease	+	+	+	+
Ensure production all year round	+	+		

Key      A = intensive food producer  
           B = fry and fingerling producer  
           C = public aquaria  
           D = holding facility for experimental fish

Measures of effectiveness: These can frequently be expressed in terms of efficiency. That is, as a ratio between an output and an input of a system (Spedding, 1973). For example, a measure of effectiveness of water use in a recirculating system might be the weight of fish produced per unit volume of water used in their production.

The choice of measures of effectiveness is important since it determines to a great extent the design finally selected. For example if an objective is to conserve water, a suitable measure of effectiveness might be the volume of flush per day (usually expressed as a percentage of the system volume per day). A design, however, that minimises flush does not necessarily use less water per weight of fish produced.

The development of designs to optimise the measures of effectiveness (the "generation of alternatives"), requires an understanding of the relationships between the variable factors of recirculating systems. There is little guidance available on the importance of each factor, or of the relationships between them. As a result the development of a design for a recirculating system is often rather haphazard. Frequently the selection of a seemingly appropriate design for a new recirculating system is made simply after a review of the literature (Csavas and Varadi, 1981). Although such procedures are unsatisfactory, they have often been adopted because of the lack of suitable guidelines; indeed at the EIFAC symposium on recirculating systems this was recognised as a major barrier to the effective application of recirculating system technology (Coche, 1981).

Most systems have been designed by balancing the purifying action of the reconditioning unit (usually a filter) with the rate of metabolite production. For example, Speece (1973) produced a series of graphs for estimating all the basic parameters necessary to design a recirculating system for trout culture simply from trout size and water temperature. However, the graphs require the use of a "hatchery constant", i.e. the monthly increase in length of the trout fingerlings at a given temperature and under local rearing conditions. In addition the assumptions made in developing the graphs were considerable:-

- i) temperature between 4.4 and 15.6°C
- ii) growth rate is 1.52 cm/month at 10°C
- iii) feed conversion ratio is 1.5:1
- iv) maximum allowable ammonia concentration is 0.5 mg NH<sub>3</sub>-N/l
- v) filter media has the same relationship of specific surface area to volume and particle diameter.

Also, the value determined for filter volume from the graphs requires correction according to the particular features of the design being

prepared, since Speece's relationship linking the nitrification capacity per specific filter area with temperature was based upon only one measurement of nitrification in his experimental system. Adjustment can be made to the method to correct for the circumstances of a new design, but some changes would make the entire method questionable or invalid.

The design procedure of Liao and Mayo (1974) is similar to that of Speece, being developed for freshwater culture of salmonids using biological filters for reconditioning. From experiments conducted at the U.S. Federal hatchery at Bozeman, Liao and Mayo (1972) were able to develop relationships for metabolite production and filter performance. These were used to design a number of recirculating systems for other Federal salmonid hatcheries (Mayo, 1972). Like Speece's method, their method has a number of assumptions, both explicit and built-in, and therefore has limited application. A major limitation in both these design methods is that they are based on salmonid culture at temperatures less than 16°C, while many recirculating systems are used for warm water culture.

Design methods for marine recirculating systems are also limited, resulting mainly from their use as public aquaria (Hirayama, 1974 and Spotte, 1970). Hirayama (1974) working with aquarium fish and sand filters in columns, was able to develop a single equation for determining the carrying capacity of small sand filters; where the left-hand side of the equation describes the oxidising capacity of the filter(s) and the right-hand side describes the rate of production of metabolites in the system. Metabolite production was related to the amount of food fed as in the methods of Speece (1977) and Liao and Mayo (1974). The equation takes little account of the consumption of oxygen by heterotrophic bacteria, and assumes a constant impurity level being sustained by a large aquarium so that the influent concentration is not affected by the return from the biological filter; conditions unlikely to be met in intensive fish culture (Poxton et al, 1980).

The conclusion drawn from this examination of current design methods is that those available are too inflexible for general use. The EIFAC symposium on recirculating systems recognised this when it recommended (Recommendation 80/11) that EIFAC and ICES (International Council for the

Exploration of the Seas) should promote the preparation of "a technical manual on bio-engineering criteria for the design of recirculation systems" (Tiews, 1981).

## 2.5 Research needs

Recirculating systems can be used to advantage in a variety of circumstances but as Mayo (1979) has pointed out many recirculating systems have proved to be partial or total failures, and "successful systems are not the rule". Hambrey (1980) has argued that few schemes employing recirculation have been "genuinely economic". Caution is necessary in considering such statements, since much depends on the criteria by which success is measured. While some recirculating systems may have been simply unsuitable for the application in which they were used it is clear that there are still numerous difficulties to be overcome.

Recirculating systems are complex and their design and operation requires a high level of understanding, and a detailed knowledge of the interactions and processes in the whole system. Much of the research which has been conducted into recirculating systems has been of detailed studies of parts of the system. Although these are helpful in understanding individual aspects, they do not assist in understanding the complex interactions amongst various inputs, or the synergistic effects of these inputs into the system, (Neal and Mock, 1979). It would seem therefore, that the logical progression towards establishing the feasibility of recirculating systems would be studies directed at the whole system, (Orth, 1980).

The problems of studying the whole system can be tackled in a number of ways, such as by systems analysis, operations research and simulation modelling. These have been successfully applied in ecology and natural resource management, (Jeffers, 1973, Watt, 1966 and 1968), fisheries science (Patten, 1969) and agriculture (Spedding, 1979; Dent and Blackie, 1979 and Brockington, 1979), but there has been limited application in aquaculture.

The literature does provide some examples: systems analysis has been applied to lobster culture (Allen and Johnson, 1976), while Price (1978) and Epifanio et al (1973) have reported on the interdisciplinary studies in aquaculture conducted at the University of Delaware. In addition Botsford et al (1974) have described the application of systems analysis and optimisation theory to the economics of aquaculture.

Few simulation models have been developed for use in aquaculture. Hammond and Lackey (1976) produced a model for analysing alternative management strategies for catchable trout fisheries, while Huang et al (1976) developed a simulation model of prawn culture in ponds, for predicting numbers, sex and size of prawns at each harvest. Earlier Powers (1973, 1974) used simulation modelling to describe the dynamics of a salmon culture pond. No models of recirculating systems have been built.

The development of a simulation model of a recirculating system offers a number of advantages. By providing a framework for understanding behaviour and performance, a model could be of considerable value in the development of guidelines for the design and operation of a recirculating system (and aid in the interpretation of operational data). In addition, if a method such as that of de Neufville and Stafford (see Section 2.4), is used in the design of a recirculating system the construction of a model is a key step in the "evaluation of alternatives" (de Neufville and Stafford, 1971).

The development of a simulation model usually follows a period of observation and experimentation (Ford et al, 1982; Biddlescombe et al, 1981; Teng, Blackie and Close, 1980 and Dent and Blackie, 1979), since the selection of an appropriate modelling technique and the actual construction of the model requires knowledge of the nature of the system to be modelled (Mihram, 1972).

A research programme with two phases was therefore initiated.

#### I Observation and experimentation on the system

- a) Design and construct a recirculating system
- b) Operate the system, monitor and evaluate its performance

- c) Examine variables affecting performance and list all the variables and causal pathways of potential importance in determining the functioning of the system. Use this as the basis for detailed experimental work
- d) Describe relationships between variables, evaluating their relative importance and indicating conflicts and interdependence

## II Simulation modelling

- e) Construct a model based on the important variables
- f) Model implementation
- g) Model verification and sensitivity testing
- h) Model validation
- i) Use of model

## CHAPTER THREE EXPERIMENTAL PROGRAMME

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**3.3.3.2 Salt Accumulation in a Closed System**

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**3.3.5 Conclusions**

### 3. EXPERIMENTAL PROGRAMME

#### 3.1 Nature and scope of the investigation

The first phase of the research programme involved observation and experimentation on a recirculating system. Since none of the recirculating systems or design methods described in the literature were suitable, it was necessary to design and construct a suitable experimental system, an activity carried out jointly with Mr. S. Lawson. Six similar systems were built for use in the laboratory of the Applied Biosystems Research Group.

The first part of the experimental programme involved monitoring a number of biological and environmental variables to assess system performance. A second series of experiments focused on the three areas identified as important during experimental series I; the characterisation of wastes, filter performance and analysis of growth and feed conversion.

## 3.2 Experimental series I

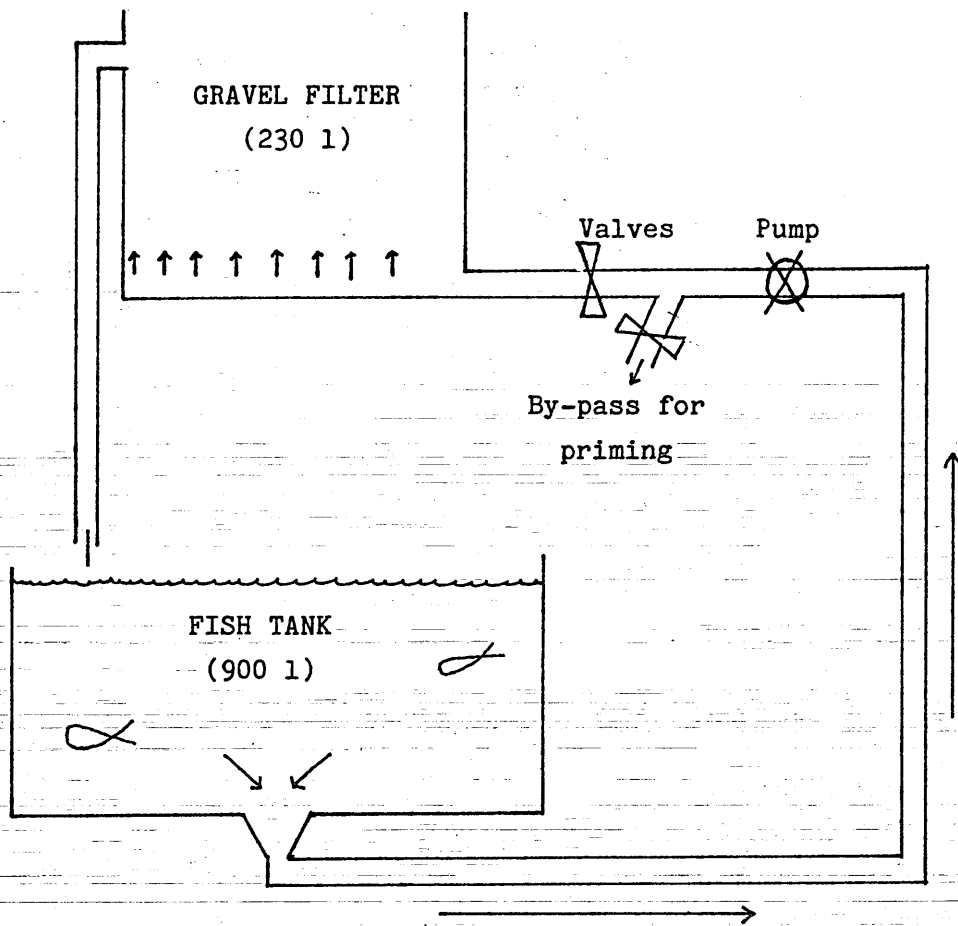
### 3.2.1 Introduction

The recirculating systems designed for use in the laboratory were entirely experimental and there could be no guarantee of their suitability for fish culture. Therefore the primary objective of experimental series I was to monitor a number of biological and environmental variables to assess the performance of the system. This presented a further difficulty that in many cases the only guidelines in the literature concerning which variables should be monitored were vague. Thus new guidelines had to be developed. As a preliminary step towards more detailed experimentation a note was made of variables and causal pathways of potential importance.

### 3.2.2 System design

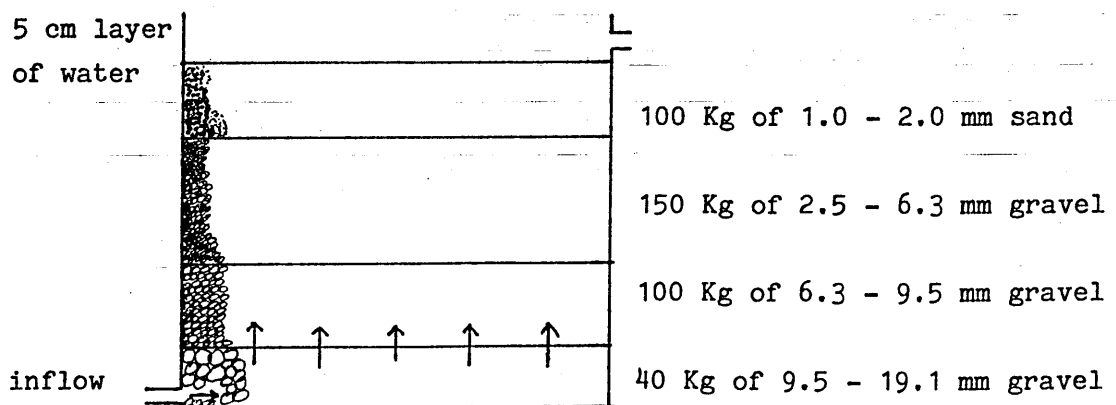
The laboratory system consisted of six fibreglass fish tanks of 900 litres. Each tank was served by an upward flow gravel filter in a 230 litre plastic tank. Culture water was pumped up through the filter and drained back into the fish tank under gravity (Fig. 3.1).

Figure 3.1 Basic flow pattern



Each filter bed was filled with washed gravel of different sizes to form a graded bed (Fig. 3.2).

Figure 3.2 Schematic cross-section through the filter



Inside the filter four 2 cm ABS pipes were laid to distribute the incoming water (Fig. 3.3). These pipes had 6 mm holes drilled at intervals to effect even flow (Fig. 3.4).

Figure 3.3 Pipe layout in base of filter bed

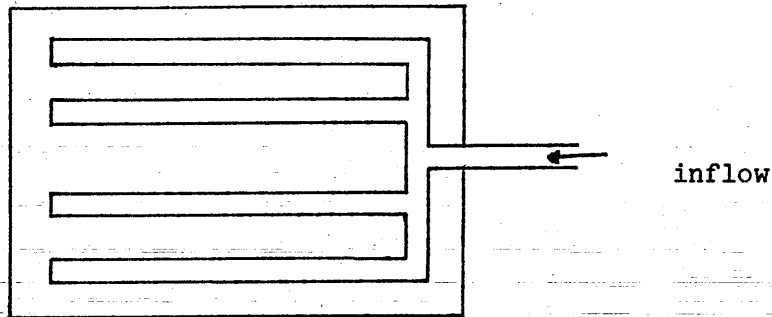
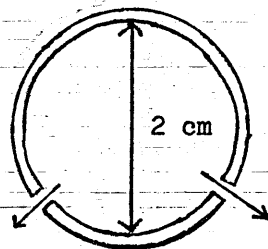


Figure 3.4 Cross-section through drilled pipe



Filter bed base

The water flowed from the filter back into the fish tank via a 6.5 cm hose mounted 5 cm above the top of the sand. The layer of water above the filter bed had a reduced flow rate which allowed considerable final sedimentation to take place.

The filter beds and pumps were mounted above the fish tanks on a wooden platform above "Triclamp" scaffolding. The pipework used throughout was 2 cm reinforced clear plastic pipe. A by-pass and shut-off valves were included to aid priming and maintenance, (Fig. 3.1). The valves could not be used to control the flow rate accurately since this was influenced by clogging of the pipes and the filter bed.

Aeration for the six tanks was provided by the use of a compressor and porous plastic pipe. Each tank was heated by "tube" aquarium heaters (5 x 200 w per tank) and the temperature was controlled by independent thermostats. Skylights gave natural lighting for most of the year but during working hours throughout winter fluorescent tubes were used. The filter bed platform shaded approximately two-thirds of each tank. Make-up water was supplied from a single mains outlet via taps over each fish tank.

### 3.2.3 Experimental methods

#### 3.2.3.1 Fish production

Five tanks were used during experimental series I, with a further tank kept on standby for use in an emergency. All five systems were initially used for the culture of common carp, but tanks 1 and 5 were later used for the culture of grass carp. The common carp used in experimental series I were supplied by Cotswold Carp Farm after fish from other sources were rejected because of their poor condition on arrival. The grass carp were supplied by the Southern Water Authority.

The fish were fed with Baker's "Omega Carp Pellets" throughout the experiment. The composition of the pellets is given in Table 3.1. The weight of feed (dry weight) was calculated as a percentage of the total fish biomass (wet weight), the percentage being known as "ration size". In order to maintain the same ration size throughout an experiment the weight of feed was increased daily in proportion to an assumed increase in fish weight resulting from the previous feed. The assumed increase in fish weight was calculated using an assumed food conversion efficiency (FCE) - see below. The FCE used for feed calculation was taken from the results of the previous growing period. Initially a value of 0.4 was used.

Invariably the FCE used for feed calculation did not agree with the actual FCE for that period, therefore the mean actual ration size was calculated by the equation

$$\text{Mean Ration Size (\%)} = \frac{\text{Average weight of feed/day}}{\text{Average fish biomass}} \times 100$$

where

$$\text{Average weight of feed/day} = \frac{\text{Total weight of feed}}{\text{No. of feeding days}} \quad (\text{g})$$

and

$$\text{Average fish biomass} = \frac{\text{Initial biomass} + \text{Final biomass} - \text{Initial biomass}}{2}$$

Initially the fish were hand fed in the morning only but by the end of the experiment two equal feeds were given, morning and late afternoon/evening. Stocking densities and ration size were experimentally varied and are described with the results for each system.

The fish were weighed by one of two methods depending on whether individual or total fish weights were required (Appendix 2). Specific growth rates (SGR) were calculated according to the formula by Brown (1947) where

$$\text{SGR} \quad (\text{G}_w \text{ \% day}^{-1}) = \frac{\log_e \text{final weight} - \log_e \text{initial weight}}{\text{Period of growth (days)}} \times 100$$

Food conversion efficiencies (FCE) were calculated from

$$\text{FCE} = \frac{\text{Wet weight increase in fish (g)}}{\text{Dry weight of food given (g)}}$$

The ratio of food utilisation, i.e. the food conversion ratio (FCR) is the reciprocal of FCE.

Regular examinations for disease were made and any fish displaying an abnormality was isolated in an aquarium for further observation and/or possible treatment.

Table 3.1 Composition of carp pellets

Calculated analysis

Protein	%	35.0
Oil	%	5.0
Fibre	%	4.5 (declared as 5.5)
Ash	%	10.5
Moisture	%	9.0
Carbohydrate (Nitrogen free extract)	%	35.0
Vitamin A	18,000	international units/Kg
Vitamin D	2,000	international units/Kg
Vitamin E	120	international units/Kg
Selenium	0.1	mg/Kg

This food also contains an anti-oxidant.

The pellets are manufactured from marine fish meal, which is a blend comprising mainly of white fish & herring, soya, meat meal and distillers' solubles. The fibre content comes from wheat millings and meal. The oil content is from the fish and wheat. This is a low temperature extruded pellet.

### 3.2.3.2 Conditioning

The procedure adopted for conditioning was experimentally varied and is described with the results for each system.

### 3.2.3.3 Water Quality

Checks of water quality were made daily, with the formation of foam, cloudiness, odour and colour being noted. Measurement of pH, temperature, oxygen, nitrite and total dissolved solid was made frequently during the start-up period with occasional measurements made once the system was fully conditioned. Ammonia (as total ammonia) was measured on a daily or more frequent basis throughout. Analytical methods employed throughout are



described in Appendix 2.

### 3.2.3.4 Operation

The tanks were filled with mains water and heated by "tube" aquarium heaters. The thermostats were set at 25°C. The systems were normally run fully closed, with make-up water from the mains added approximately once every 1-2 weeks. Analysis of the mains water was performed by the Anglian Water Authority (Table 3.2). The rate of water recirculation and the rate of flush were measured by timing the filling of a container of known volume. Five measurements were taken for all readings and the average value recorded.

Table 3.2 Analysis of the make-up water

Analysis by	Anglian Water Authority	
Date	29/11/1978	
Sampling point	Open University (Systems Department)	
pH	7.57	
Electrical conductivity (Rec. megohms)	1010	
P.V. (4 hrs. at 27°C)	0.91	
Ammonia F. and S. (as N)	0.05	mg/l
Ammonia Alb. (as N)	<0.01	"
Nitrate (as N)	5.2	"
Alkalinity (as CaCO <sub>3</sub> )	204	"
Total hardness (as CaCO <sub>3</sub> )	306	"
Calcium	107	"
Magnesium	9.2	"
Sodium	100	"
Potassium	9.4	"
Phosphate soluble (as P)	0.01	"
Fluoride	0.28	"
Chloride	59	"
Sulphate (as SO <sub>4</sub> <sup>2-</sup> )	192	"
Silica (as SiO <sub>2</sub> )	3.3	"
Iron (Total)	0.03	"
Manganese (Total)	0.01	"
Zinc	0.10	"
Copper	0.01	"
Cadmium	<0.005	"
Mercury	<0.0005	"
Lead	<0.01	"
Arsenic	<0.05	"

For experimental series I, water quality criteria were based on Amlacher (1970) and EIFAC (1970) and not those presented in Table 2.1. Thus the following maximum values were adopted:-

Ammonia	0.025 mg NH <sub>3</sub> /l (unionised)	EIFAC 1970
Nitrite	10 - 20 mg/l	Amlacher 1970
pH	6 - 8 (lethal at 10.8)	Amlacher 1970

When toxic conditions occurred no more food was given until levels returned to safe concentrations. If levels were found to be so high as to be lethal the taps over the tanks were opened and the system flushed. This caused fluctuation in pH and temperature (Fig. 3.5) and to avoid increased stress to the fish this option was avoided if possible.

### 3.2.4 Results

#### Tank one

Commissioning: The tank was seeded with a suspension made from 125 g of well composted soil, 18/5/1979. Plate culture (study by Biology Dept., O.U.) showed the following were present; Proteus, micrococci, pseudomonas and aerobacter. There were remarkably few bacteria, particularly bacilli, present. The tank was therefore re-seeded on 21/5/1979 with a suspension from 125 g of soil of a different source.

Fish production: See Table 3.3.

Water quality: Results for the culture of common carp during the initial period, 16/5 - 29/7/1979 are plotted in Fig. 3.5. The range of water quality values, excluding ammonia, are given in Table 3.4. Prefeed ammonia concentrations during grass carp culture 7/8 - 8/11/1979 are plotted in Figs. 3.6, 3.7 and 3.8. Diurnal fluctuations of ammonia concentration in the fish tank are shown in Fig. 3.9.

Filter performance: During the monitoring of diurnal variations of ammonia, measurements were made of the influent and effluent concentrations of ammonia, see Fig. 3.10. The influent ammonia concentration to the filter has been plotted against effluent concentration in Fig. 3.11.

## Tank Two

Commissioning: The tank was seeded on 16/7/1979 with a suspension from the filter of tank one. Fourteen days later the tank was spiked with ammonium chloride to give a total ammonia concentration of 5.0 mg/l. The concentration of ammonia was monitored (Fig. 3.12). When the ammonia level reached 0.24 mg/l it was presumed that the bacterial population was active and so the tank was stocked.

Fish production: See Table 3.5.

Water quality: Results for the initial culture period 16/5 - 22/6/1979 are plotted in Fig. 3.5. The range of water quality values, excluding ammonia, are given in Table 3.6. Prefeed ammonia concentrations from 1/8 to 15/10/1979 are plotted in Figs. 3.6, 7 and 8 whilst daily variations in ammonia concentrations are shown in Fig. 3.9

Filter performance: Monitoring of influent and effluent ammonia concentrations was carried out as for tank one. Results are given in Figs. 3.10 and 11.

## Tank Three

Commissioning: The tank was not seeded or spiked, but run on closed circulation to examine the effects of such action.

Fish production: See Table 3.7.

Water quality: Measurements made during the experimental period are shown in Figs. 3.6, 7 and 8 and Table 3.8.

Filter performance: Readings of influent and effluent ammonia concentrations were presented in Fig. 3.11.

## Tank Four

This tank was kept as the spare for emergency use.

## Tank Five

Commissioning: The tank was not seeded or spiked prior to the culture of common carp. Before grass carp culture, the system was seeded with an inoculum taken from tank six, (17/7/1979). Fourteen days were allowed for

the bacteria to establish before the system was spiked with ammonium chloride to give a concentration of 1.8 mg/l. The level of ammonia was monitored, (Fig. 3.12). By 3/8/1979 the ammonia concentration had fallen to 0.08 mg/l and on 7/8/1979 grass carp were stocked.

Fish Production: See Table 3.9.

Water Quality: The results of water quality analysis during common carp culture are shown in Fig. 3.5 and Table 3.10. Figs. 3.6, 7 and 8 give the prefeed ammonia concentrations 10/8 - 15/10/79, whilst the diurnal variations of ammonia concentrations are plotted in Fig. 3.9.

Filter Performance: Both influent and effluent ammonia concentrations showed marked diurnal variation (see Fig. 3.10) and are plotted against each other in Fig. 3.11.

#### Tank Six

Commissioning: As tank one.

Fish Production: See Table 3.11.

Water Quality: The measurements made during the initial part of the experiment are shown in Fig. 3.5. Table 3.12 gives the measurements made during the whole experimental period. Prefeed ammonia concentrations are plotted in Fig. 3.6.

Table 3.3 Tank 1 Fish Production

Date 1979	Species	No. Days	Ini. No. Fish	Mort. %	Ini. Wt. (g)	Wt. Gain (g)	Wt. Feed (g)	FCE	SGR (G <sub>w</sub> % day <sup>-1</sup> )
16/5- 21/6	C.C.	36	197	21	1024	599		+	1.94
22/6- 30/7	C.C.	38	226	17	2017	439		+	1.00
7/8- 23/10	G.C.	77	50	2	1178	2540	5021	0.51	1.51
23/10- 8/11	G.C.	16	41	0	3295	652	2136	0.31	1.13

+ Because of mortalities FCE could not be calculated

Table 3.4 Tank 1 Range of water quality measurements (tank readings)

Date 1979	Species	O <sub>2</sub> Sat. (%)	pH	Total Diss. Solids (mg/l)	Temp. °c	Nitrite (mg/l)
16/5 27/7	C.C.	95-102	8.0-8.5	0.6-0.8	17-23	-
7/8- 8/11	G.C.	80*	7.6-7.7	1.3*	26-28	0.1-0.15

\* 1 measurement only

Table 3.5 Tank 2 Fish Production

Date 1979	Species	No. Days	Ini. No. Fish	Mort. %	Ini. Wt. (g)	Wt. Gain (g)	Wt. Feed (g)	FCE	SGR ( $G_w\%$ day <sup>-1</sup> )
16/5- 22/6	C.C.	37	99	28	517	123			0.16
31/7- 24/9	C.C.	55	188	3	2456	4357	8352	0.51	1.90
24/9- 15/10	C.C.	21	183	0	6813	991	2694	0.37	0.65

Table 3.6 Tank 2 Range of water quality measurements (tank readings)

Date 1979	Species	O <sub>2</sub> Sat. (%)	pH	Total Diss. Solids (mg/l)	Temp. °c	Nitrite (mg/l)
25/5- 10/6	C.C.	83-97	8.0-8.4	0.60-0.75	13.5-23	0.1-0.5
7/8- 8/11	C.C.	94*	7.2*	0.85*	28	0.1-0.5

\* 1 measurement only

Table 3.7 Tank 3 Fish Production

Date 1979	Species	No. Days	Ini. No. Fish	Mort. %	Ini. Wt. (g)	Wt. Gain (g)	Wt. Feed (g)	SGR ( $G_w\%$ day <sup>-1</sup> )
8/8- 15/10	C.C.	68	5	20	3856	288	6619	0.43

Table 3.8 Tank 3 Range of water quality measurements (tank readings)

Date 1979	Species	O <sub>2</sub> Sat. (%)	pH	Total Diss. Solids (mg/l)	Temp. °c	Nitrite (mg/l)
8/8- 15/10	C.C.	80-100	7.7-7.1	1.1	22-26	0-12

Table 3.9 Tank 5 Fish production

Date 1979	Species	No. Days	Ini. No. Fish	Mort. %	Ini. Wt. (g)	Wt. Gain (g)	Wt. Feed (g)	FCE	SGR (G <sub>w</sub> % day <sup>-1</sup> )
25/5- 23/6	C.C.	29	99	46	545	-229			0.27
7/8- 24/10	G.C.	78	50	2	1276	3212	8361	0.39	1.64

Table 3.10 Tank 5 Range of water quality measurements (tank readings)

Date 1979	Species	O <sub>2</sub> Sat. (%)	pH	Total Diss. Solids (mg/l)	Temp. °c	Nitrite (mg/l)
25/5- 23/6	C.C.	94-97	8.0-8.4	6.0-7.5	14.5-22.5	
7/8- 24/10	G.C.	78	6.8-7.7		25.0-28.0	0.02-1

Table 3.11 Tank 6 Fish production

Date 1979	Species	No. Days	Ini. No. Fish	Mort. %	Ini. Wt. (g)	SGR (G <sub>w</sub> % day <sup>-1</sup> )
16/5- 23/6	C.C.	38	143	27	997	1.64
23/6- 17/8	C.C.	55	196	16*	1694	1.11

\* 25 fish died overnight as the result of a power failure

Table 3.12 Tank 6 Range of water quality measurements (tank readings)

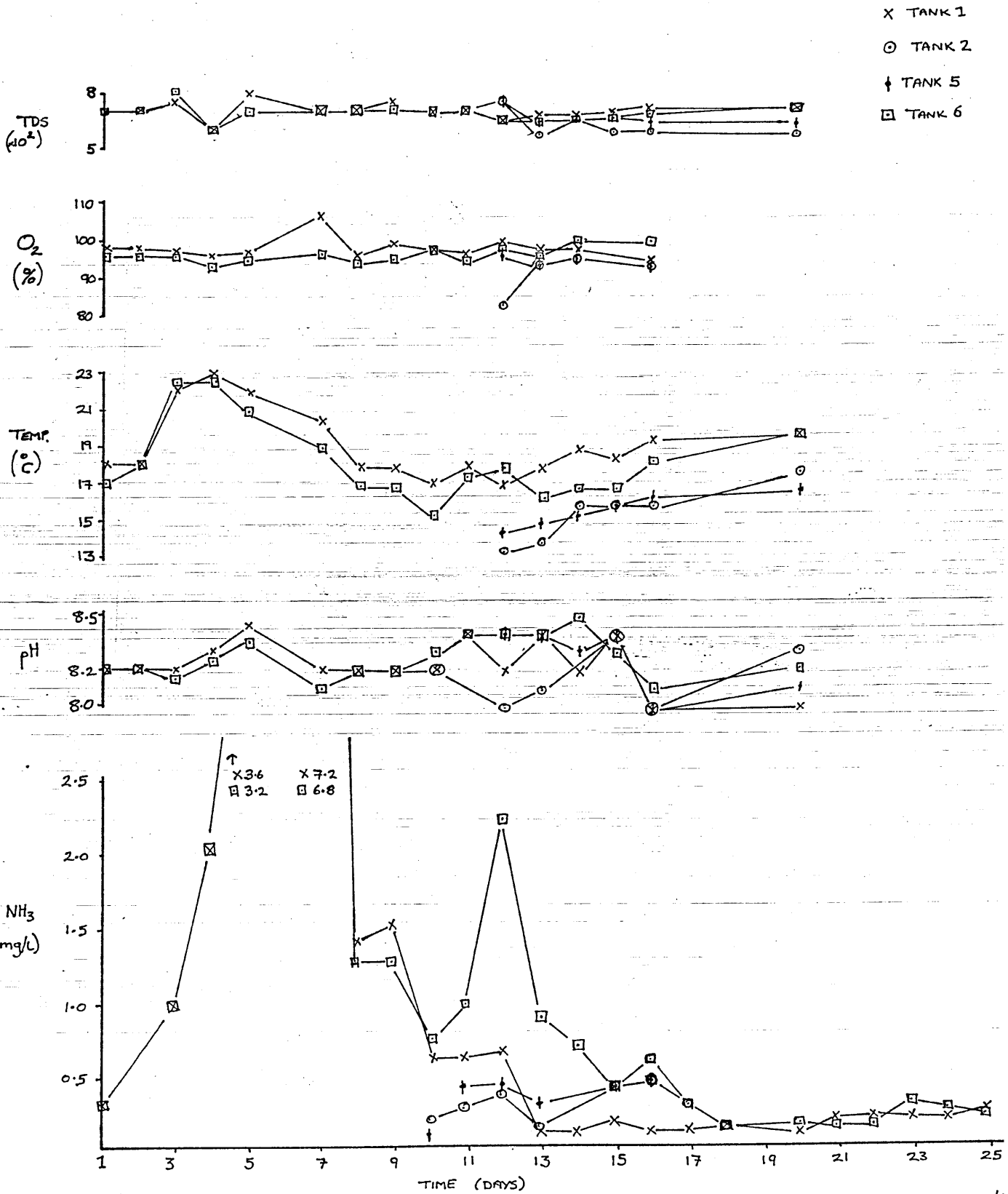
Date 1979	Species	O <sub>2</sub> Sat. (%)	pH	Total Diss. Solids (mg/l)	Temp. °c	Nitrite (mg/l)
16/5- 13/6	C.C.	95-100	8.1-8.7	6.0-8.0	15-20	0.42*
23/6- 17/8	C.C.	57*+	6.7-7.6		27-28	0.26*

\* 1 measurement only

+ pump failure overnight, 25 fish died



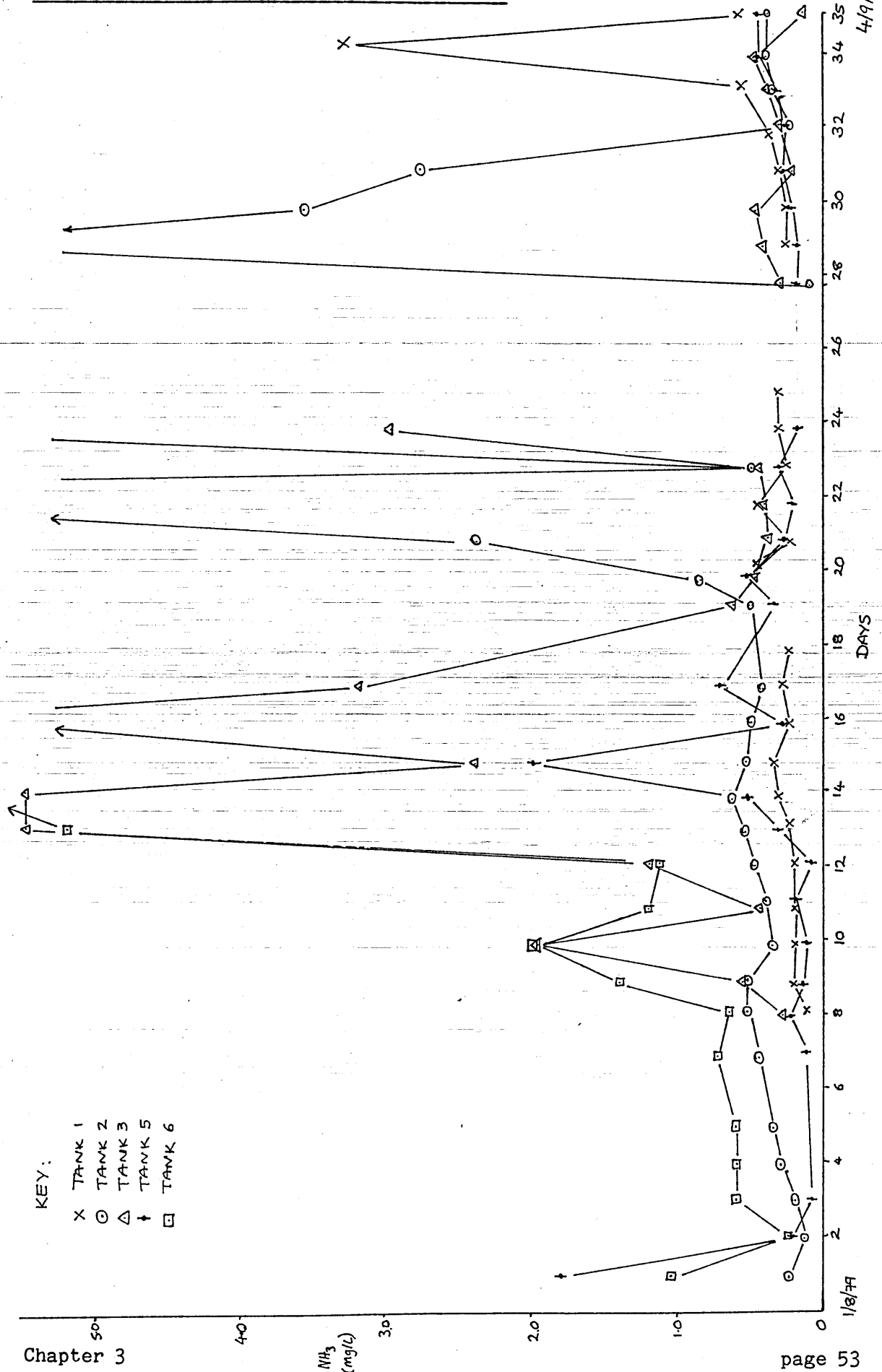
Figure 3.5 Water quality results for tanks 1, 2, 5 and 6



16/5/79

9/6

Figure 3.6 Pre-feed ammonia concentrations



KEY:

- X TANK 1
- O TANK 2
- Δ TANK 3
- † TANK 4
- TANK 5

Figure 3.7 Pre-feed ammonia concentrations

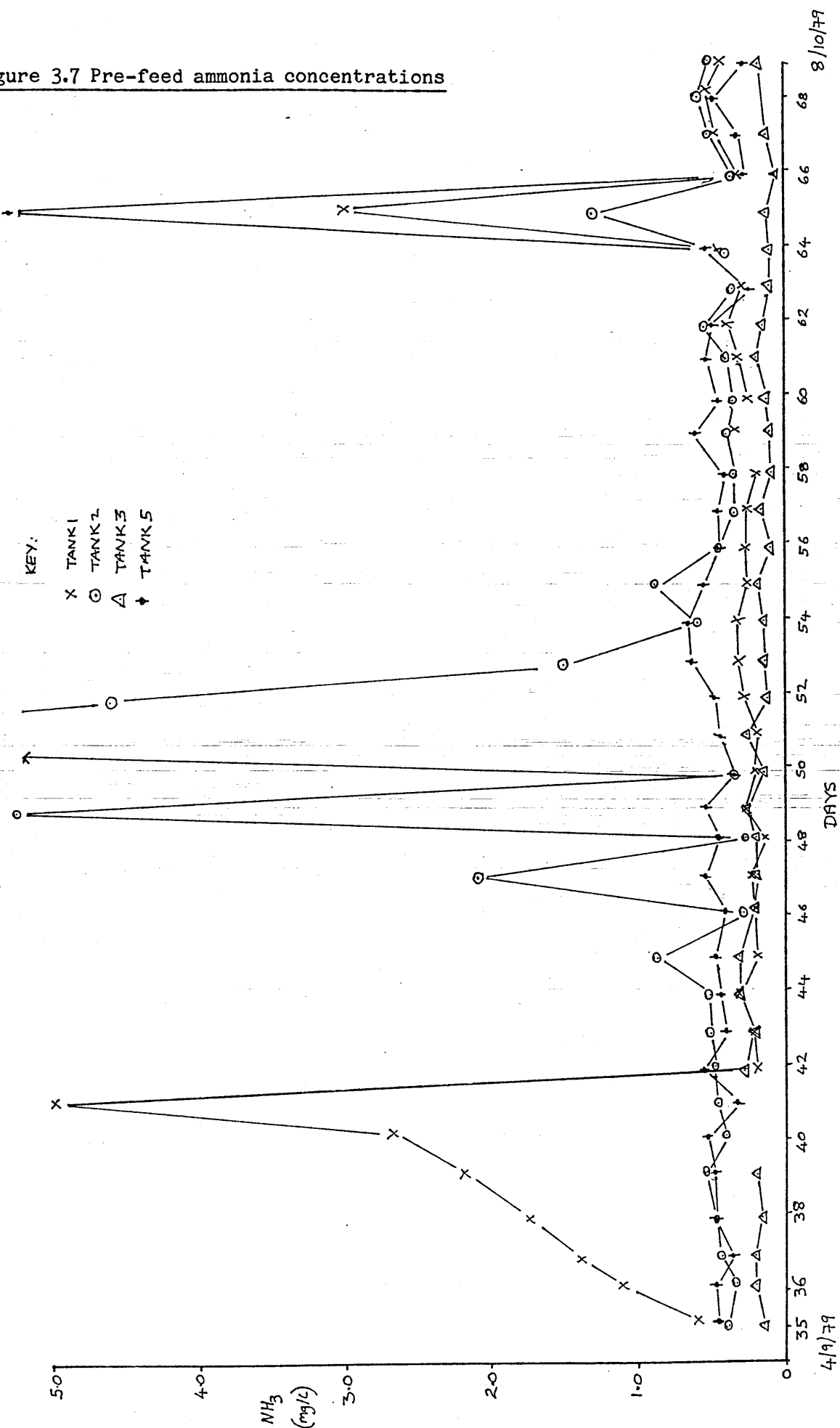


Figure 3.8 Pre-feed ammonia concentrations

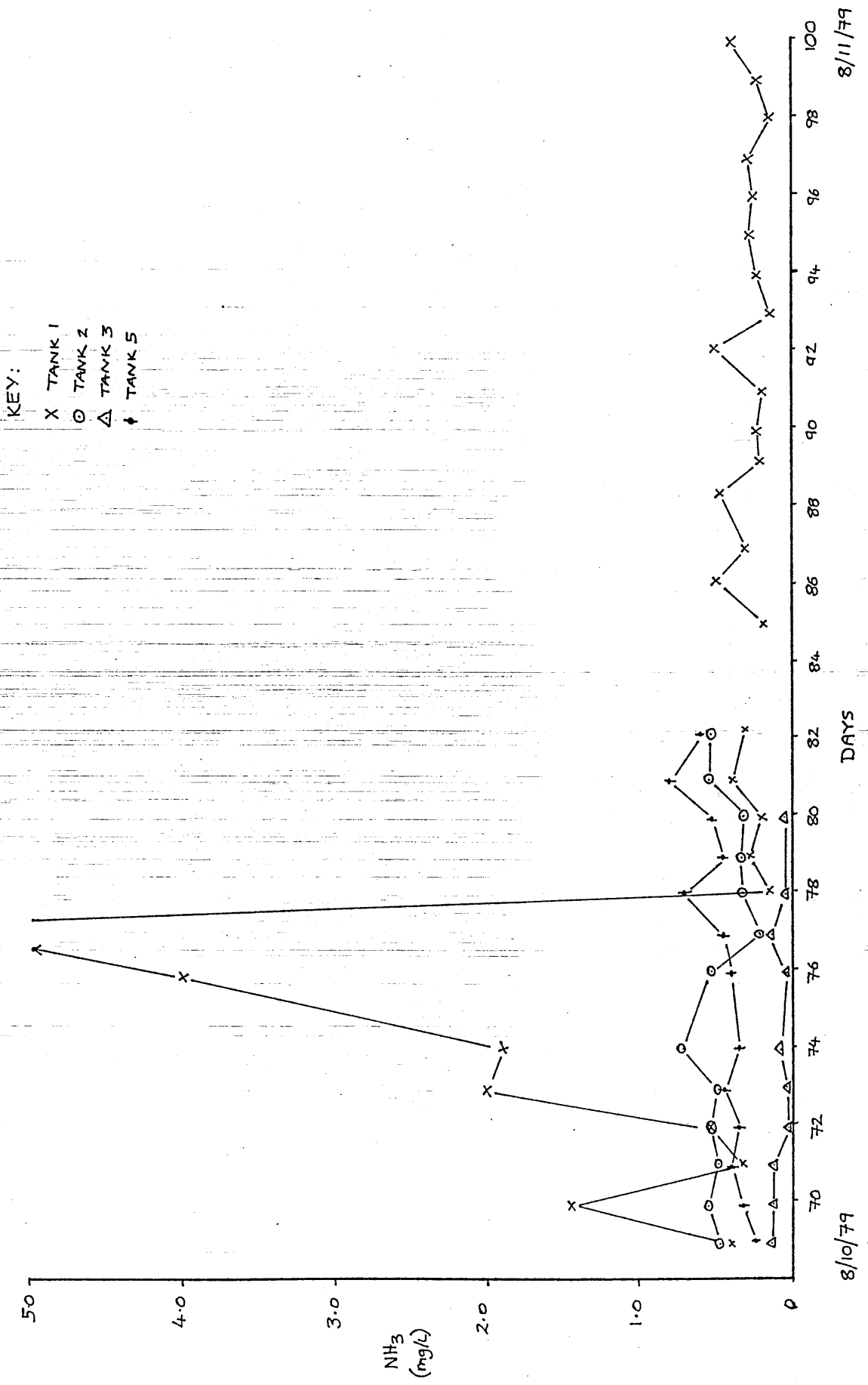


Figure 3.9 Diurnal fluctuations of ammonia concentrations

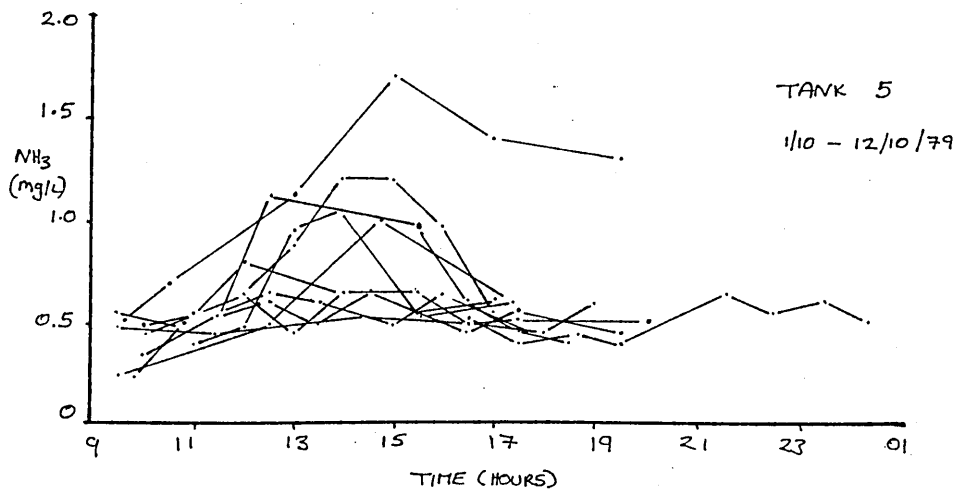
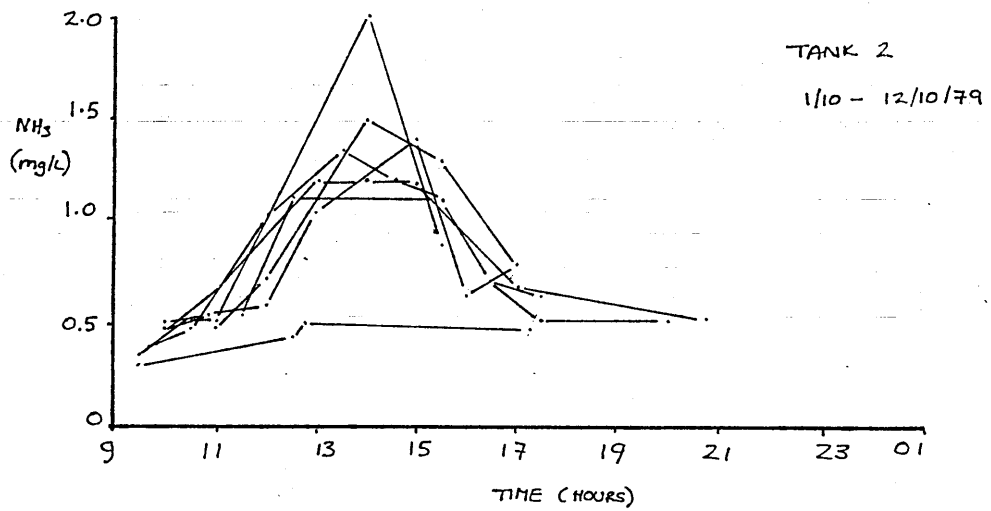
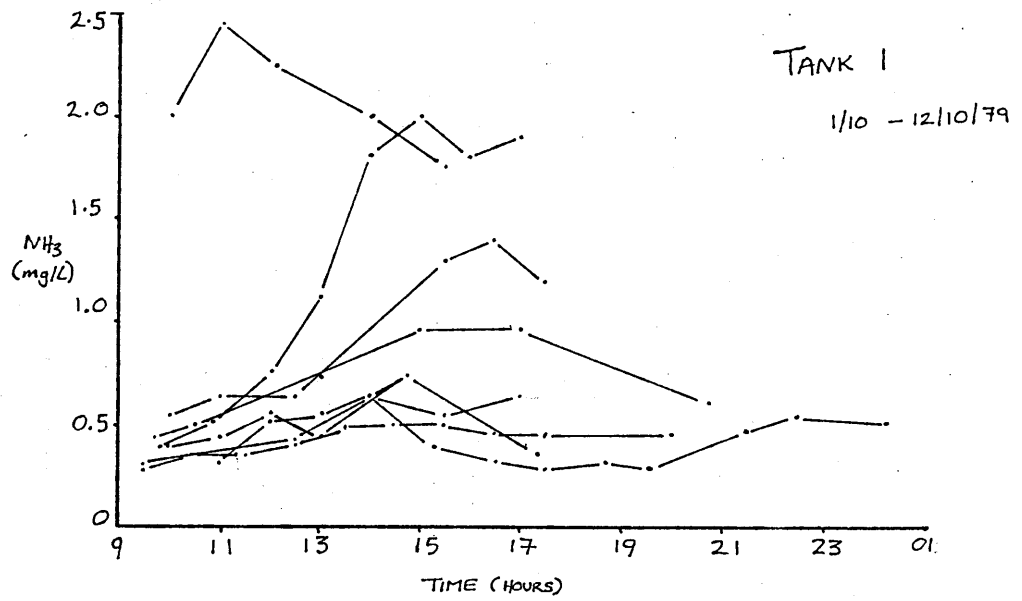


Figure 3.10 Influent and effluent concentrations of ammonia

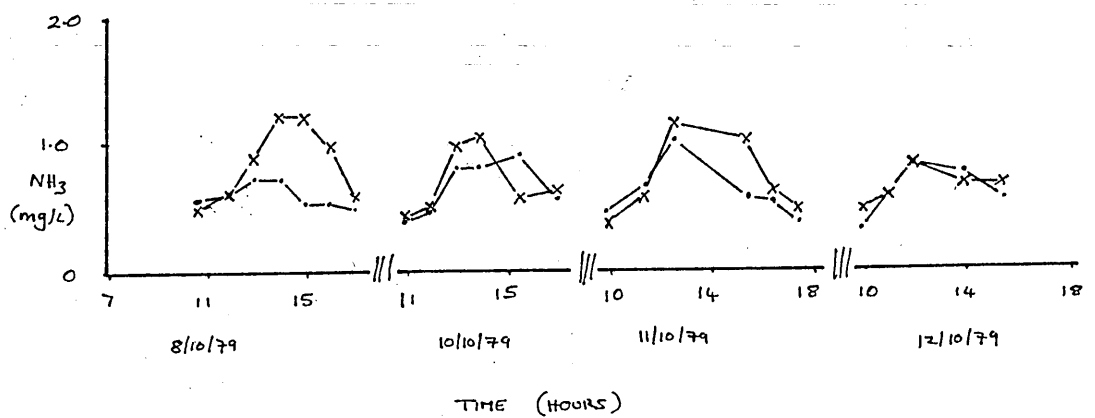
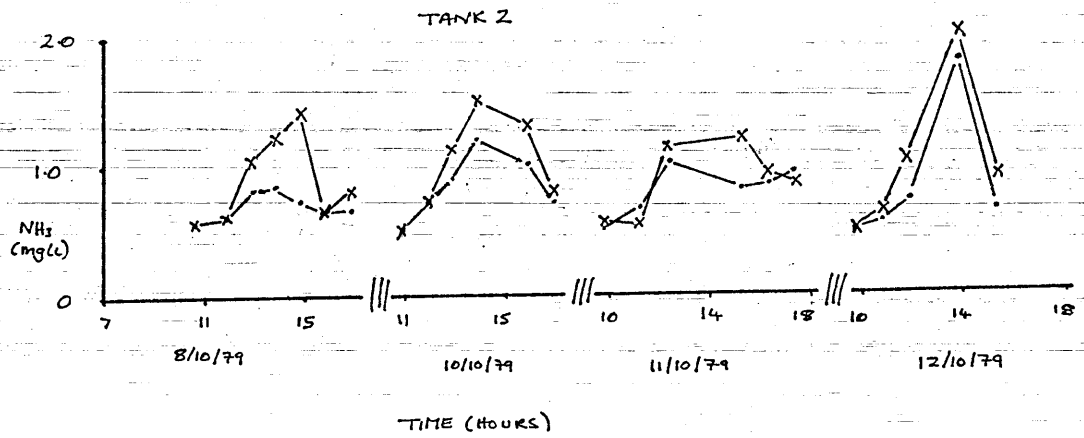
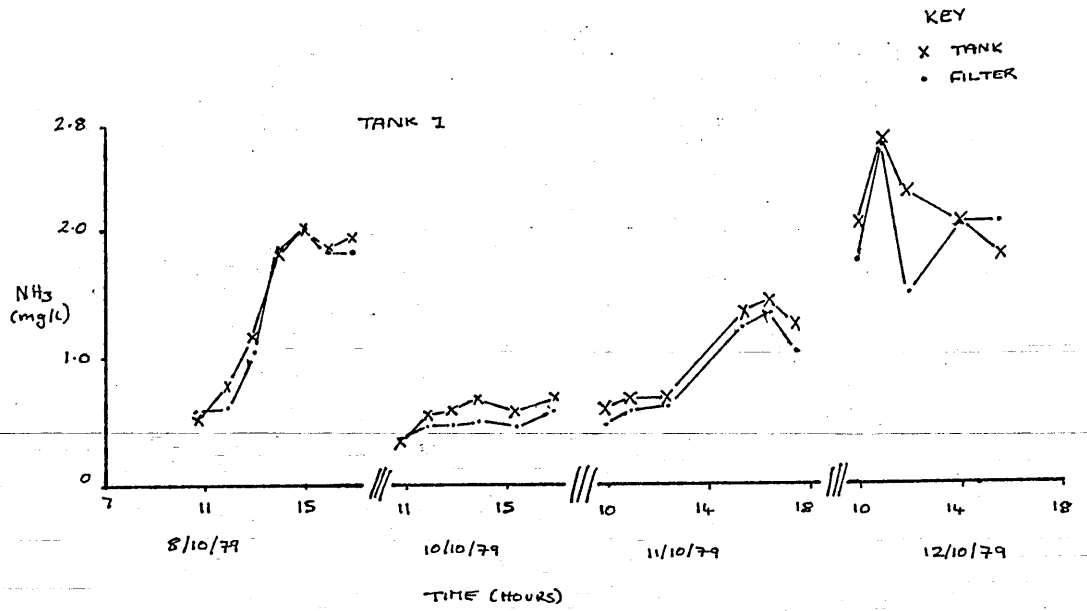


Figure 3.11 Influent/effluent concentrations of ammonia

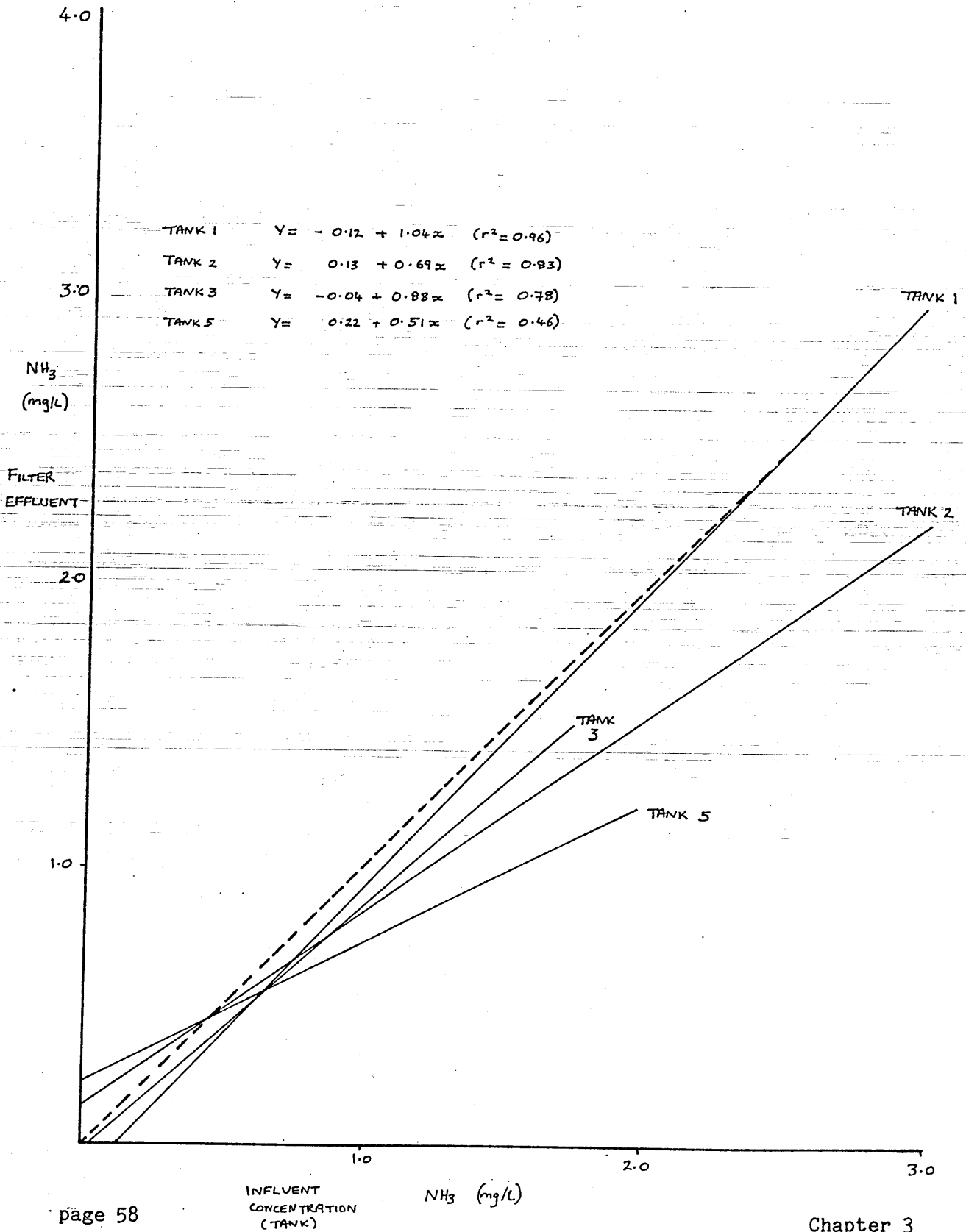
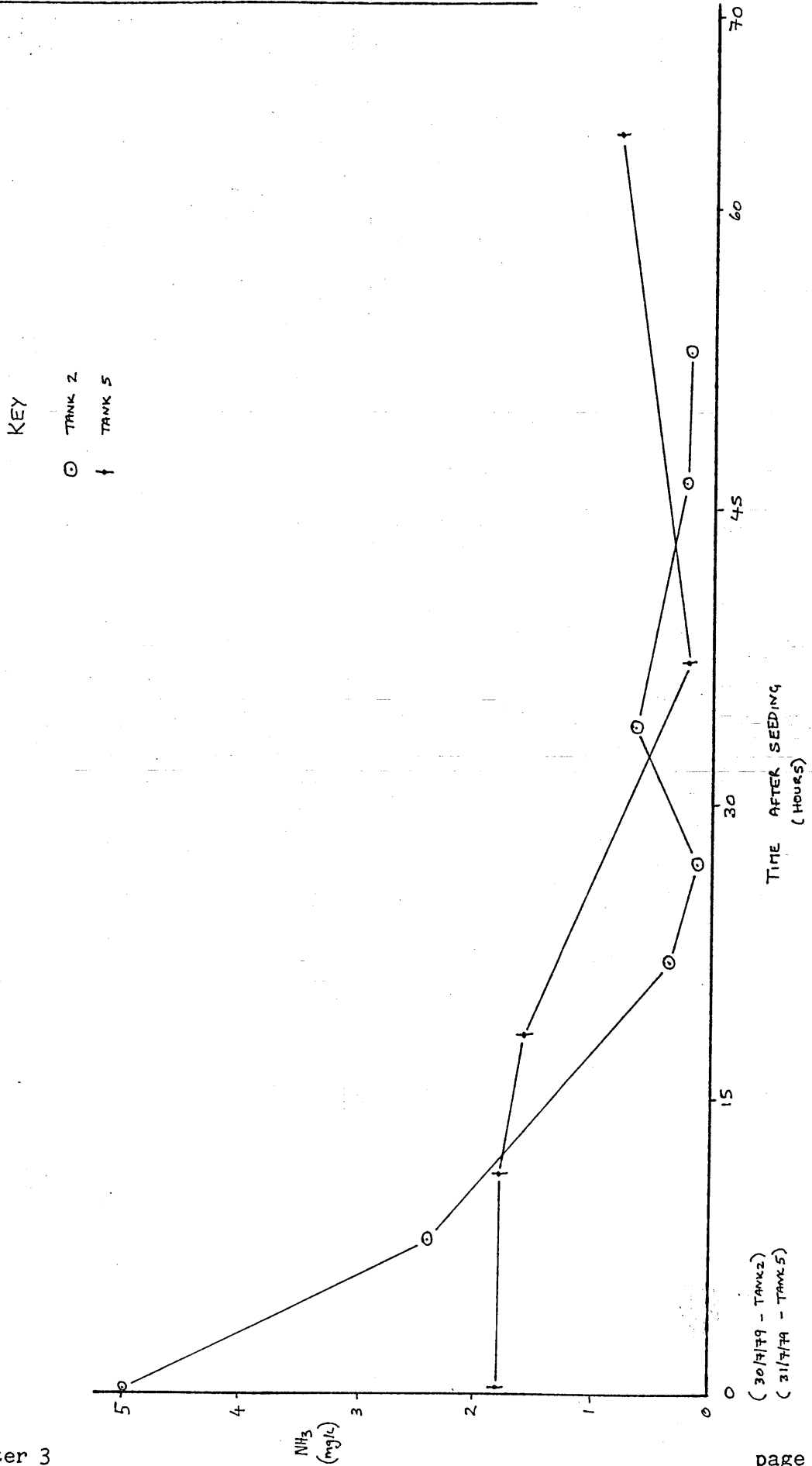


Figure 3.12 Ammonia concentrations after seeding





### 3.2.5 Discussion

#### Fish Production

##### 3.2.5.1 Mortalities

During the early stages of the experiment high mortality rates were experienced in tanks 1, 2, 5 and 6. Examination of the dead fish revealed one or more of the following: gill haemorrhaging, ciliated protozoans in gills, skin pale and/or green with occasional grey areas and ruptured anus. Before death the fish displayed a strange corkscrew-like swimming motion and were found lying on the bottom of the tank still alive. Fish were sent to Dr. R. Sweeting of Thames Water Authority for examination, but no disease or infection was found on any of the specimens.

A possible explanation is that the mortalities were caused by toxic levels of nitrite. The water quality criteria adopted for experimental series I were based on Amlacher (1970) who suggested 10 - 20 mg/l as the maximum acceptable concentration in culture water. A much lower value of 0.1 mg/l however, is recommended by Wickins (1981) and Munro (1978) as the maximum acceptable concentration. Since nitrite was considered less toxic than ammonia, its concentration was recorded less frequently. The most frequent readings of nitrite concentration were taken in tank 3 where levels rose to 12 mg/l. No mortalities occurred in tank 3. This can perhaps be explained by the lower susceptibility to nitrite poisoning of larger fish.

The main toxic action of nitrite is to reduce the effectiveness of the blood for oxygen transfer, a condition known as Methaemoglobinaemia. Small fish with high growth rates and increased oxygen demand are therefore more susceptible to nitrite poisoning.

The concentration of nitrite at which mortality will occur is also dependent on the presence of chloride ions in the water (Perrone and Meade, 1977). Wickins (1981) has suggested that a chloride ion concentration of 25 mg/l affords some protection. Although no measurements of chlorine were made, the make-up water contained a chloride concentration of 59 mg/l (Table 3.2). Since it has been shown that chloride ions accumulate in a closed system (see Section 3.3.3.2 and Table 3.27), it

can therefore be assumed that some protection was provided to the fish in tank 3.

The value suggested by Wickins (1981) and Munro (1978) as the maximum acceptable is based on the minimum recorded concentration at which nitrite oxidises haemoglobin to methaemoglobin in trout. Under cultural conditions higher levels can be tolerated with only minimal effect on the fish. Trout are generally more susceptible to toxic effect than other culture species. For example, the 96 hour LC<sub>50</sub> values for trout range from 0.19 to 0.55 mg NO<sub>2</sub>-N/L (Spotte, 1979), while Konikoff (1975) found the 96 hour LC<sub>50</sub> for catfish to be 7.55mg NO<sub>2</sub>-N/L. Wickins (pers. comm.) suggested that a level of 0.50 mg NO<sub>2</sub>-N/l might be more appropriate as the maximum desirable concentration for carp in culture, and therefore this value was adopted during experimental series II.

#### 3.2.5.2 Growth rates

The majority of references made in the literature to the growth of common carp do not refer to intensive culture or controlled conditions. Only Huisman (1974, 1976, 1978), Kausch and Ballion-Cusmano (1976) and Hambrey (1980) have published data on the growth of common carp under a range of temperatures, ration conditions and fish weights. The data of Kausch and Ballion-Cusmano (1976) and Hambrey (1980) are, however, limited, and incompatible with that of Huisman. Similarly, few allusions to the growth of grass carp refer to intensive culture or controlled conditions. Only Huisman and Valentijn (1981) have published data on the growth of grass carp fed with compound feeds similar to those used during the experimental programme, and for a range of ration conditions. Comparisons of growth rates obtained during the experimental programme are therefore usually based on the data of Huisman.

#### Common Carp

Specific growth rates (SGR) during experimental series I varied considerably, both between systems and over time, ranging from 0.16 to 1.94 (G<sub>w</sub>% day<sup>-1</sup>). The differences in SGR can be largely attributed to environmental factors, such as temperature, ration size and feeding frequency, although biotic factors such as stocking density, stress and

individual fish weight may also have had an effect.

The SGR of the common carp in tank 2 during the period 31/7/79 - 18/10/79 can be compared with the data of Huisman (1978) and is presented in Table 3.13.

Table 3.13 Comparison of common carp specific growth rates Tank two

<u>31/7 - 15/10</u>				
Date	Food ration (% of biomass)	Temp.	SGR	SGR estimated from Huisman's data
31/7- 24/9	2.23	13-23	1.90	1.50 small carp) 1.75 fingerlings) 23°C
24/9 15/10	1.76	28	0.65	0.90 small carp 27°C

From the Table above it can be seen that SGR's for the first period were larger than those estimated from Huisman's data, while during the second period the SGR's were lower than estimated.

The reduction in SGR during the two growing periods can be largely explained by the differences in temperature, fish size and ration level. As indicated in Table 3.13, Huisman (1978) found that the SGR's of fingerlings (<20g) was higher than that of small fish (30-100g) fed the same ration size. The fish used in the present experiment increased from 13 to 37g in the first experimental period and from 37 to 42g in the later period, and therefore some reduction in growth rates could be expected.

During the first growing period, water temperature increased slowly towards the desired temperature of 25°C. Flushing with mains water resulted in considerable variations and prevented the desired temperature from being achieved, although temperatures of 23°C were recorded. Huisman (1978) found that growth of small carp (30-100g) was greater at 23°C than at 17 or 27°C. Extrapolation of his data suggests that growth at 28°C is comparable to that at 18°C. This suggests that the increased growth during the first growing period may have been promoted by the differences in temperature. The specific growth rate estimated from Huisman's data for

the second growing period may be a little high since it is based on observations made at 27°C compared to the measured water temperature of 28°C. The addition of mains water was much less frequent in the second growing period than in the first period and changes in water temperature were probably too small to have much effect. Some of the difference between the actual and predicted SGR may therefore be the result of the lower temperature used in Huisman's experiment.

During the first growing period, high ammonia levels resulted in the fish being fed on only 49 of the 55 days (89%) and on 14 of the 21 days of the second growing period (67%). In addition, during the first period, it was necessary to reduce ration size from 5 per cent of body weight a day, fed for the first 23 days, to 2.5 per cent. For the final four feeds it was necessary to split the daily ration into two portions of 1.25 per cent, fed in the morning and in late afternoon. For the first four days of the second growing period a single feed of 2.5 per cent was given, but after this it was necessary to split the feed as before, into two portions of 1.25 per cent.

In a study of the effects of short-term food deprivation on channel catfish, Randolph and Clemens (1978) found that for each day of missed feeding it took two days for the previous rate of growth to be resumed. In addition, after one day's missed feed the appetite of the fish on the following day was increased. After longer periods of deprivation however, appetite was suppressed and the return to predeprivation levels of feeding required one day for each day of missed feeding. If a similar response is assumed for the carp in the present experiment, one would expect the actual growth rates achieved to be less than those predicted by Huisman, whose fish were fed daily. This was more pronounced in the later growing period where the percentage of missed feeds was greater. As discussed later (Section 3.3.3.1), there is some doubt concerning the methods of Randolph and Clemens (1978) and their results should be viewed with caution.

A further factor contributing to the differences in SGR is stress. Stress can develop from a number of causes such as poor water quality or handling during stock management (Pickering et al, 1982). Smart (1980) has shown

that in intensive fish culture stress can cause reductions in appetite and this increases the amount of food lost to waste and increases metabolic rate, so reducing the amount of assimilated food available for growth. It is not possible to quantify the level or effect of stress during experimental series I, although it is perhaps significant that during the first growing period the fish were undisturbed for longer intervals than in the second growing period as weighing was less frequent.

### Grass Carp

The growth rates of the grass carp can be compared with values taken from Huisman and Valentijn (1981):

Table 3.14 Comparison of grass carp specific growth rates

Tank	Date	Feed Ration	Temp. °c	SGR	SGR Predicted from Huisman & Valentijn's data
1	7/8 - 23/10	1.76	26-28	1.51	0.70
1	23/10- 8/11	3.38	26-28	1.13	1.75
5	7/8 - 24/10	1.28	25-28	1.64	0.40

As can be seen in Table 3.14, over the second growing period the growth of grass carp in tank 1 was depressed. Growth during the first period was similar in tanks 1 and 5. The slight differences in SGR's experienced in tanks 1 and 5 may be a reflection of the different feed rations. However, the data from Huisman and Valentijn (1981) suggests that the differences in feed ration should have the opposite effect. Although tank 1 had the higher feed ration, 7 per cent more feeds were missed than in tank 5. It is possible then, that the missed feeds had a greater effect on growth than the differences in feed ration. Temperature ranges experienced in both tanks were similar and it is unlikely that this had much effect.

As with the common carp in tank 2, the growth rate during the second growing period of the grass carp in tank 1 was 25 per cent lower than the previous SGR. Unlike the common carp the grass carp had almost twice the

feed ration. A main contributory factor may have been stress. The second growing period for the grass carp was shorter than that for the common carp and therefore stress from handling was possibly greater. In addition, grass carp are difficult fish to culture because of their easily stressed nature. During weighing they may leap into the sides of the fish tank, even out of the tank, inflicting damage upon themselves. Similar "jumping" behaviour has been noticed by Ellis (1974). In comparing the SGR's achieved during experimental series I with those of Huisman and Valentijn (Table 3.14), a number of differences can be seen. These differences are greater than those found when comparing SGR's of the common carp with data from Huisman (1978). Whilst similar explanations for the differences apply, two other factors may also have had an effect in the case of the grass carp.

Because of their timid behaviour the grass carp were somewhat reluctant to feed during the day, particularly when people were present in the laboratory. As a result, a larger percentage of the feed was probably wasted.

Secondly, the pelleted feed used throughout the experimental period was developed by 'Edward Baker' to meet the dietary needs of common carp. The feed formulated by Huisman and Valentijn (1981) for use in intensive culture of the grass carp had a similar protein content to the 'Edward Baker' feed, but contained more carbohydrate, with less fibre and slightly more oil. If the feed used in this experiment resulted in a dietary deficiency this may have affected the growth rate. Any effects would probably be gradual, becoming more marked with time. Since SGR represents the average growth rate over the time period, any changes in growth rate towards the end of the first growing period would be less apparent when averaged with earlier growth rates. Over the second growing period, a change in growth rate would be more apparent.

In absolute terms, the growth rates achieved during the experimental period were not high. Huisman and Valentijn (1981) claim a SGR of 10 per cent body weight/day for grass carp under normal production conditions using their compound feed, and comment on the lower SGR of grass carp achieved by Dabrowski (1977) and Dabrowski and Kozak (1979), who did not achieve more than 3 per cent body weight/day. However, Huisman &

Valentijn's growth rates refer to grass carp fry only. The maximum SGR of small grass carp (48g) found by Huisman & Valentijn (1981) was 3.5 per cent. Maximum SGRs of 2.87 to 3.65 per cent were also found by Shireman et al (1977), who fed 46 - 73g grass carp to satiation with a diet based on duckweed at 25°C.

### 3.2.5.3 Food Conversion

The variations in food conversion generally match the variations in growth rate, with more efficient food conversion by both the grass carp and common carp during the first growing period (Table 3.15)

Table 3.15 Food conversion during experimental series I

Date	Species	Tank No.	Weight gain(g)	Weight of feed(g)	Ration size	Food conversion efficiency	SGR
7/8							
23/10	G.C.	1	2540	5021	1.76	0.51	1.51
23/10							
8/11	G.C.	1	250	2136	3.38	0.31	1.13
7/8							
24/10	G.C.	5	3238	8361	1.28	0.39	1.64
31/7-							
24/9	C.C.	2	4423	8352	2.23	0.51	1.90
24/9-							
15/10	C.C.	2	991	2694	1.76	0.37	0.65

G.C. = Grass Carp, C.C. = Common Carp.

The decrease in efficiency of food conversion by the grass carp in tank 1 over the two growing periods may reflect a loss of appetite and increased metabolism arising from an increase in stress (see 3.2.5.2). The grass carp in tank 5, although growing faster than the grass carp in tank 1, were less efficient at food conversion over the same period. It is difficult to explain this difference, since although food conversion is affected by water temperature, fish weight and ration size, over the ranges considered here their effect on food conversion is the same as on growth. The common carp in tank 2 showed a marked decrease in food conversion efficiency

during the second growing period. This corresponded with a decrease in specific growth rates. Reasons for the reductions have been discussed above (3.2.5.2).

#### 3.2.5.4 Conditioning

Newly established biological filters usually have a population of nitrifying bacteria inadequate to maintain ammonia and nitrite at appropriately low concentrations. Carmignani and Bennett (1977) showed that seeding not only reduced start-up time, but also considerably reduced the maximum concentrations of ammonia and nitrite achieved prior to the full filter activation. Sources of nitrifying bacteria used for seed have varied; Scott and Gillespie (1972) added soil nitrifiers, while Meade (1974) recommended using nitrifiers from garden soil or stream sediment, and also spiking the water with nutrients. Siddall (1973) recommended spiking the water with 3 - 6 mg  $\text{NH}_4^+$ /l while Forster (1974) used 1 mg  $\text{NH}_4^+$ /l, and Poxton et al (1980) used 3 mg  $\text{NH}_4^+$ /l. Carmignani and Bennett (1977) seeded with gravel from an established bed, while Bower and Turner (1981) found seeding with a wet filtrant from an established system with similar environmental conditions to be the most effective method, compared with the use of culture water from an established system, a dry filtrant or commercial additives containing nitrifying bacteria.

From the limited data gathered during experimental series I it is difficult to assess the effectiveness of the two strategies used to aid conditioning, but some observations are possible.

The start-up period in tanks 1 and 6 was marked by a rapid rise in ammonia levels, reaching 7 mg/l in tank 1 (Fig. 3.5) and both systems needed flushing. By day 13 both systems appeared to be conditioned. The ammonia concentrations recorded in tanks 2 and 5 were similar to each other, (Fig. 3.5), and generally lower and more stable than in tanks 1 and 6. Whether this was because of the reduced loading in tanks 2 and 5 or the result of the different start-up procedures is uncertain.

Tank 3 received no seeding or spiking and showed a rapid rise in ammonia concentration similar to that in tanks 1 and 6, and again some flush was necessary. The time taken for levels in tank 3 to become low and stable on



closed circuit was longer. Whether the variation in the start-up time was due to differences in the prior treatments of the systems or whether it was a reflection of the different fish populations is uncertain. Measurements of the nitrite concentrations in tank 3 showed an increase when the ammonia levels had fallen, similar to observations made by a number of other authors (Bower and Turner, 1981; Carmignani and Bennett, 1977; Liao and Mayo, 1974; Hirayama, 1974, and Spotte, 1970). Since nitrite was not measured as frequently in the commissioning of tanks 1, 2, 5 and 6 it is uncertain whether a rise in the concentration of nitrite occurred when ammonia levels fell. In view of the fish mortality and peaks of ammonia experienced in tanks 1 and 6, it would seem likely that a rise in nitrite level did take place. Tanks 2 and 5 did not have high ammonia concentrations, but they did have the highest mortality rate, and it is likely that they suffered from nitrite peaks also.

#### 3.2.5.5 Water Quality

Monitoring strategy: The literature provided little guidance as to the most appropriate monitoring strategy to adopt. Dwivedy (1974) for example, monitored at unspecified intervals, and while Anderson (1974) monitored ammonia, nitrite and oxygen levels daily, Scott and Gillespie (1974) and Wickins (1976) monitored once a week. Hirayama (1976) over many years of study did not monitor at a fixed interval but recorded at one, two and three week intervals.

None of the above authors comment on the time of day when measurements were made, and a number of different analytical techniques were endorsed for the measurement of water quality. It was therefore necessary to develop a suitable monitoring strategy empirically.

Although it was desirable to monitor a wide range of variables, the frequency, precision and accuracy of these measurements must be balanced with available time, facilities and usefulness of the results. Thus while a number of variables were initially monitored, assessment of water quality eventually focused on the concentration of ammonia. However, in recognition of the more recent evidence concerning the toxicity of nitrite (Wickins, 1981 and Munro, 1978) in the later part of the experiment monitoring of water quality included regular recording of nitrite

concentration.

In view of the importance of diurnal variations in water quality, hourly measurements appear desirable. In practice this is prohibitively expensive and time consuming, and most authors measure water quality on a weekly or daily basis. It is therefore appropriate to consider the need for more frequent monitoring.

It can be argued that diurnal variations in water quality are only important if they pose a threat to fish health or if maximum utilization of the filter is required at all times. Many recirculating systems operate at less than maximum loading in order to provide a margin of safety. In such circumstances diurnal variations are important only if the peaks have a deleterious effect on health. Rather than requiring frequent monitoring safety margins should be incorporated at the design stage, with the employment of a filter of a size sufficient to provide some buffering to rapid changes. In most cases therefore, monitoring more frequently than once a day would appear unjustified.

The time when measurements are made should be determined with regard to the feeding strategy. Where peaks are potentially damaging it may be advantageous to monitor at the time when the peaks are at their highest, in these experiments this was between 14 - 16.00 hours (Fig. 3.10). Damaging levels can then be corrected by flushing if necessary. It is however, probably better to monitor prior to feeding when damaging levels can be prevented by withholding the feed.

Monitoring less frequently than daily may involve risk depending on the loading of the system and the built-in safety margin. In experimental series I the filter did not prove to be reliable over a long time scale and it was therefore concluded that during experimental series II monitoring would be carried out on daily basis prior to feeding.

Oxygen: The compressor provided a reliable service throughout the experimental period. Oxygen levels were maintained between 80-100% saturation in the fish tank except during a power failure. The porous plastic pipe presented no problems if it was given a clean under hot water once a month.

pH: The production of hydrogen ions during nitrification resulted in a fall in pH during periods of closed circulation. This was buffered by the addition of alkaline make-up water (Table 3.2). As indicated in Table 2.1 carp are tolerant of a wider range of pH values. While the preferred pH for nitrifying bacteria is in the range 7 - 8.5 (Painter, 1970) the percentage of total ammonia as un-ionised ammonia decreases with decreasing pH (Emerson et al, 1975). The measured pH values achieved a compromise between these constraints and were considered satisfactory.

Temperature: The initial increase in temperature using the in-tank heaters was slow, and once the desired temperature was reached temperature was maintained only within + or - 3°C. This was partially due to a number of heaters failing because of faulty seals. The fault was rectified for experimental series II. To overcome the variations in temperature which occurred with the addition of mains make-up water, an on-line 7.2 kw heater was installed for the second experimental series. This raised the temperature of the incoming water by 10°C.

Total dissolved solids: Measurements made during the initial period of operation indicated that during periods of closed circulation there was an accumulation of dissolved solids. Flushing reduced the levels and since the levels recorded were not considered hazardous monitoring was discontinued. It was recognised though, that during long periods of closed circulation, accumulation could become hazardous to the fish and/or nitrifying bacteria, and a more detailed investigation would be necessary.

Ammonia: During the initial period of closed circulation following commissioning the ammonia levels were low and stable (Fig. 3.5). In tank 1 a gradual increase in the pre-feed ammonia concentration was recorded. This appeared to be correlated with the weight of food fed to the fish the previous day. The pre-feed concentration (PFC) or "baseline" concentration depends principally on the balance between nitrifying activity and ammonia production. Since a number of authors have shown a clear relationship between metabolic and faecal wastes and the weight of food (see Section 4.5.1), this relationship can be taken as an indication of the nitrifying activity of the filter. The weight of food fed to the fish each day was increased to allow for the increase in fish weight. This would have increased the production of ammonia. The increase in baseline

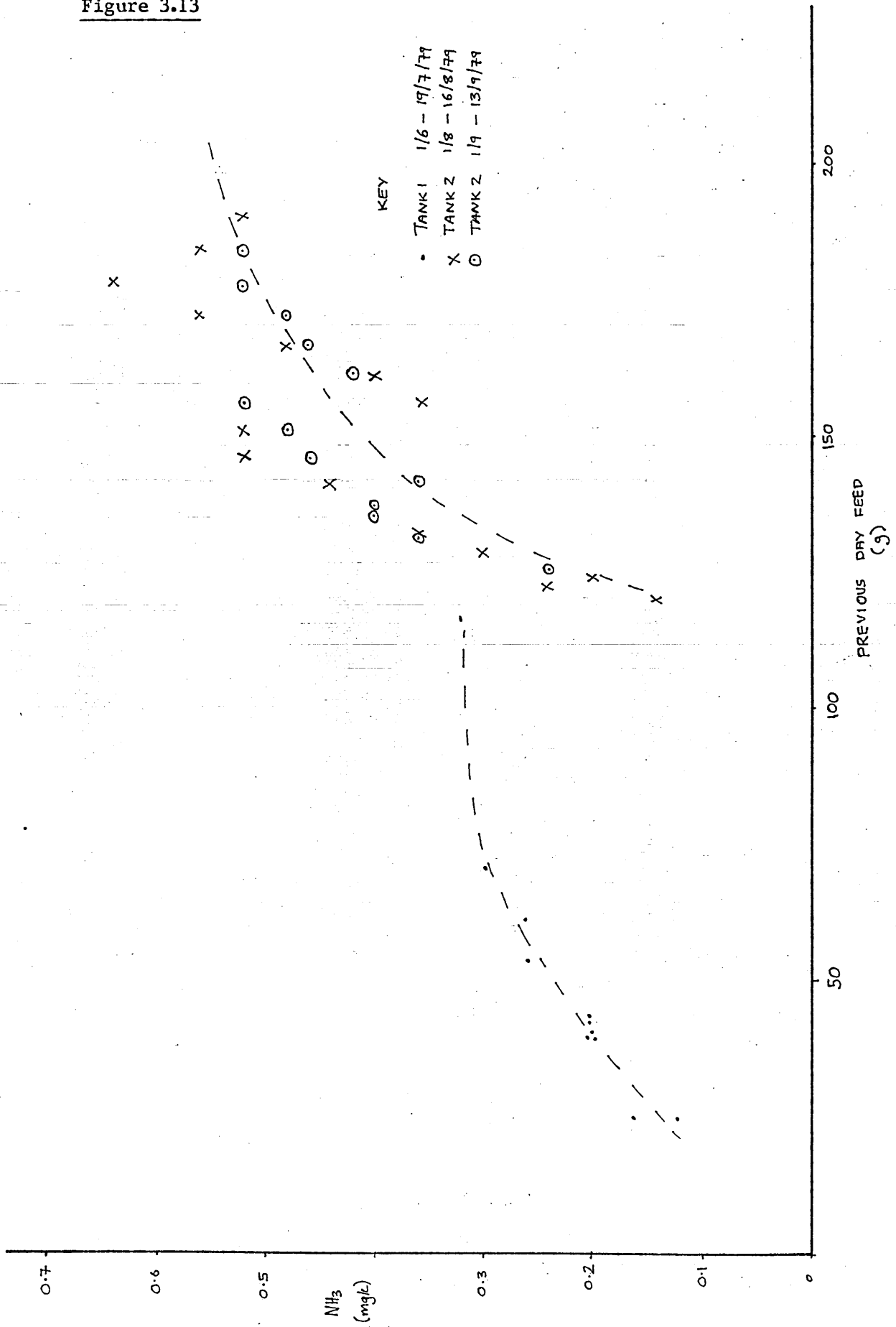
concentration suggests that the increases in loading were more rapid than increases in nitrifying activity. Similar increases in pre-feed ammonia concentrations were also found in tank 2 during 1/8 - 16/8/1979 and 1/9 - 13/9/1979. When the results from tanks 1 and 2 are plotted (Fig. 3.13) they appear to follow a curvilinear relationship, (dashed lines) suggesting an increase in nitrifying activity by the filter in response to the increased loading. On more careful inspection however, the results from tank 2 can be seen to show a steady increase in concentration terminated by a sharp fall followed by a further steady increase, thus the ammonia concentration oscillates about an upward curvilinear trend.

A possible explanation for these oscillations is that they represent an increase in ammonia resulting from increasing weight of feed, as was similarly found in tank 1. Compared to heterotrophic bacteria, nitrifiers are slow growing (Painter, 1970). The sudden fall in concentration may represent the exponential growth phase of an expanding nitrifying population, with the period between these falls representing their generation time. The larger daily increases in baseline concentration recorded for the first period in tank 2 are probably a reflection of the larger feed ration; 5 per cent of biomass per day compared to 2.5 per cent during the second period. The data of Huisman (1978) suggests that the optimum ration size for small carp is 3 per cent. At ration levels above this there will be greater excretion of ammonia due to an increase in protein metabolism.

Regular monitoring of ammonia indicated that changes in concentration over a period of days could be both rapid and extreme, (Fig. 3.6, 7 and 8). For the majority of readings ammonia levels were low, but occasionally the concentration rose rapidly and then flushing was often necessary to avoid mortalities. In tank 1 over a 74 day period, 90 per cent of the pre-feed ammonia levels were below 0.6 mg/l while the other 10 per cent were above 1.4 mg/l. Similar patterns were observed in the other systems.

The increasing repetition of high pre-feed morning ammonia concentrations resulted in a greater number of missed feeds. Eventually, to overcome this problem, the weight of food fed to the fish was halved, i.e. 2.5 per cent of body weight instead of 5 per cent of body weight per day. This allowed

Figure 3.13



regular daily feeding to resume with ammonia concentrations remaining low and stable (e.g. Fig. 3.7) until the levels once more rose rapidly. The occurrence of these peaks appeared to be influenced by the weight of feed. For example, in tank 2 peaks occurred when the weight of food reached 220g, 190g and 170g on 21/8, 14/9 and 1/10/1979 respectively. The reduction in weight of food at which these peaks occurred may indicate a decrease in the efficiency of the filter with time. As considered later this was thought to be in response to an accumulation of solids in the filter.

It became clear that the time of day when the ammonia concentration was measured had some influence on the levels recorded. Attention was therefore focused on daily changes in concentration. As Fig. 3.9 shows, marked diurnal variations were recorded. Although it is well known that certain water quality parameters may fluctuate during the day (due to changes in fish metabolic rates), little information is available on the extent of variations and the factors affecting them. Such information requires detailed monitoring and is very labour intensive. Only Rosenthal et al (1980), Murray (pers. comm.) and Goodson (pers. comm.) have examined the daily variations in water quality during the operation of a recirculating system. Their results show that the short term variability in water quality is affected by fish size, stocking density, water replacement rates and reconditioning rates. It is interesting to note that the data of Rosenthal et al (1980) and the results of this present study show maximum ammonia concentrations to occur at 14-16:00 hours. The size of the peaks recorded in this experiment were strongly influenced by the amount of feed.

Feeding the fish with a single feed of 2.5 per cent of body weight per day, eventually resulted in unacceptably high peak concentrations. The daily ration was therefore split, with half the ration fed in the morning and the other half fed when the ammonia levels had returned to an acceptable level after the peak following the first feed, usually by the late afternoon. By splitting the feed, the size of the peaks decreased. Intensive monitoring during the day continued until the end of experimental series I. Based on these observations it was proposed that in experimental series II, feeding should be spaced throughout the day.

Fluctuations in ammonia concentration are undesirable since they can result in short term exposure of fish to potentially toxic levels resulting in depression of growth. In addition the effect on the bacterial populations in the filter is uncertain.

Diurnal variations in the concentration of ammonia in a recirculating system could arise for a number of reasons: the rate of ammonia production is greater than the rate of removal of ammonia from the fish tank, as determined by the flow rate; the rate of production is greater than the rate of removal by the filter; or it may be the result of a lag between the response of the nitrifying bacteria and the change in ammonia concentration. In some circumstances fluctuations may result from all three. The design of the system will clearly have an effect. In systems where the circulation rate is sufficient to prevent accumulation of ammonia in the fish tank, fluctuations may still occur, particularly where the system is operated close to its carrying capacity. Increasing the size of the filter may overcome difficulties where ammonia production is greater than removal, but still some fluctuations will arise from a lag in the response of the bacteria to changes in the loading. Enlarging the filter to cope with peaks is undesirable, since for the majority of the time it will be under-utilised and thus increase the time taken to respond to changes in loading.

#### 3.2.5.6 Filter Performance

Solids removal: Solids were successfully removed by the filters in all five systems. The accumulation of solids in the pipework and filter bed did give rise to some problems. Pipes leading to the filters developed thick films, which considerably reduced flow rate. In the filter bed, short circuiting of flow through the bed occurred and it is probable that anaerobic areas developed. Backwashing by reversal of flow was insufficient to remove more than a small amount of accumulated solids. Agitation of the bed increased the effectiveness of backwashing, but may have had a detrimental effect on the nitrifying bacterial population. Some fine solids passed through the filter, but these settled out onto the top of the filter bed and could be removed by siphoning.

Ammonia removal: Towards the end of the experimental period, ammonia

removal by the filters was assessed. The results are plotted in Fig. 3.10. Over the period of observation the filters used to treat the water from tanks 2 and 5 were more effective at ammonia removal than in tank 1. In filters 2 and 5 the greatest removal of ammonia appeared to take place during the afternoons, returning the ammonia concentration in the tank to pre-feed levels by late afternoon. In tank 1 the concentration of ammonia in the water leaving the filter was similar to that entering. However, although the tank concentration had not fallen to the pre-feed levels by late afternoon, it had by the following day.

As a measure of the efficiency of ammonia removal in each system, the concentration of ammonia entering the filter (influent) has been plotted against the concentration of ammonia leaving the filter (effluent) and the line of best fit calculated. The value of the slope provides an indication of the efficiency of ammonia removal in each filter. A line of  $45^\circ$  passing through the origin (shown as a dashed line) represents no ammonia removal.

The relationships produced for the filters serving tanks 2,3 and 5 are similar in showing an increase in total ammonia removal with increasing influent concentration. Ammonia removal was greatest in tank 5, but with an  $r^2$  value of only 0.46 little confidence can be placed in such comparisons. The relationship described for the tank 1 filter shows a decrease in the amount of ammonia removed with increasing influent concentration.

As noted earlier the performance of the filters was not constant but showed some adaptation to increasing loads (Fig. 3.13). Beyond a certain weight of food, unacceptable peaks in ammonia occurred (Fig. 3.14). The weight of fish and feed at which this occurred was taken as the carrying capacity of the system. Carrying capacity appeared to be affected by the reduction in filter volume that occurred through clogging and short circuiting. This suggests that a considerable increase in efficiency and thus carrying capacity would be possible if short circuiting and clogging could be overcome. The efficiency of ammonia removal was probably affected by backwashing but any immediate effect went unobserved.

Chlorine has a detrimental effect on nitrifying bacteria (Rosenthal, 1980), but no appreciable differences in filter performance were noticed



following the addition of the chlorine rich make-up water. Flushing probably had a greater effect, not only because of the increased contact with chlorine, but also because of the change in temperature that occurred. As discussed earlier in section 3.2.6.1, some chlorine in the culture water is beneficial, offering protection against high nitrite concentrations. The concentrations at which protection is afforded is less than that contained by the water used to supply the system. Since it is unlikely that chlorine is removed by biological filtration, for experimental series II further additions of chlorine were prevented by the inclusion of an activated charcoal filter into the mains water supply pipe.

### 3.2.5.7 Operation and Maintenance of the system

The difficulties of maintaining water quality and filter performance became more apparent as the weight of fish held by each system increased. Under light loading the system appeared to operate satisfactorily.

In running the systems no major operational difficulties were encountered. Selection of an appropriate pump was found to be important and some problems with shaft seals were experienced. These problems were aggravated by increased back pressure caused by blocked pipework. Careful mounting of the pumps and more frequent maintenance of the pumps helped to reduce the problems.

Clogging of the filter bed and pipes during operation reduced the flow rate. Reversal of flow removed some solids, but approximately every two months the pipes needed cleaning with a brush. At the same time, the filter bed was agitated, usually with mains pressure water, and backflushed. Solids removed in this way were discarded as waste from the system. Solids settling on the top of the filter were removed periodically by siphoning to waste. Accumulation of solids in the fish tank sumps and bases of the standpipes were removed by lowering the standpipe. Efficient operation of the aeration system was ensured by periodically cleaning the porous plastic pipe in hot soapy water.

Fish cultured in recirculating systems are totally dependent on mechanical life-support systems to provide acceptable environmental conditions. Failure of any part of the life-support systems places the

fish at risk. In experimental series I an overnight power failure resulted in a rapid decrease in oxygen concentration and an increase in ammonia concentration to toxic levels. The importance of developing back-up facilities in the design of any recirculating system was recognised, and further failures were avoided by the introduction of an emergency electrical supply system.

Although no detrimental effects of backwashing were observed during the course of the experiment, as a result of encouragement by other workers a policy of reducing backwashing to a minimum was adopted.

### 3.2.6 Conclusions

The first experimental series had two objectives; to evaluate the performance of the recirculating system through the monitoring of a number of biological and environmental variables, and as a preliminary to more detailed experimentation, to note variables and causal pathways of potential importance in determining the functioning of the system.

Whilst it was possible to observe and note phenomena which were important to the operation of the system, its complexity made the identification of causal pathways difficult. Some apparent relationships were noted and these are recorded in the text. Lack of replication was recognised as a limitation, but within the constraints of the laboratory facilities, further replication was not possible.

From the results presented in section 3.2.4 it is clear that although the system could be used for closed-circulation fish culture, there were a number of difficulties:-

1. Growth and food conversion: The initial fish mortality rate was unacceptably high, emphasizing the need for care during commissioning. Growth rates were not high and were probably affected by irregular feeding and high ammonia levels. While the efficiency of food conversion was initially acceptable there was a marked deterioration.
2. Water Quality: Fluctuating levels of ammonia prevented regular feeding and diurnal variations in concentration after feeding may have had an adverse effect on the fish. The size of diurnal variation was reduced by split feeding. High nitrite concentrations during

commissioning may have been responsible for the high early mortality rate, and regular recording of nitrite was identified as important in future monitoring.

3. Filter performance: Whilst the filter successfully removed solids from the culture water, maintaining a low turbidity, backwashing was necessary to prevent clogging and short-circuiting of flow. The efficiency of ammonia removal varied considerably with a number of periods of instability recorded.

The difficulties in maintaining water quality and filter performance increased as the weight of fish held by each system increased. Under light loading the system appeared to operate satisfactorily.

### 3.3 Experimental series II

#### 3.3.1 Introduction

During experimental series I a number of difficulties were experienced concerning food conversion and growth by the fish and the maintenance of water quality. In the second series of experiments these areas were considered further. In particular, the production of solid wastes, salt accumulation and filter performance were examined, together with an analysis of growth and food conversion in new populations of common and grass carp.

Following the first experimental series, a 7.2 kw online heater and an activated charcoal filter were installed to pre-treat mains water in order to reduce changes in water temperature and any effects of chlorine on the nitrifying bacteria.

#### 3.3.2 Analysis of growth and food conversion

This section reviews experiments concerning;

1. Growth and food conversion in common and grass carp.
2. Weight loss in common carp.
3. The relationship between fish length and weight.

##### 3.3.2.1 Growth and food conversion in common and grass carp

Object: During experimental series I, the growth rate of both common and grass carp was not high, and showed considerable variation, generally worsening with time. This was reflected in the efficiency of food conversion, which although initially acceptable showed a marked deterioration.

The object of this experiment was to consider growth and food conversion efficiency in these two species, using a new batch of fish cultured in a fully conditioned system where problems of intermittent feeding caused by high ammonia levels would be minimal. Since three of the laboratory recirculating systems were available, one tank was used for the culture of

grass carp and one for common carp. In the third tank a combination of grass and common carp was cultured and examined for competition or enhancement of growth and food conversion.

Method: The three recirculating systems were stocked as follows:-

Tank 1 30 common carp (4018 g) + 30 grass carp (2772 g)

Tank 2 60 common carp (7537 g)

Tank 3 60 grass carp (5883 g)

The common carp and grass carp used in this experiment were supplied by the North-West Water Authority. The grass carp were from the same batch of fish imported into this Country from Yugoslavia as those supplied for experimental series I by the Southern Water Authority (Pritchett, pers. comm.) The common carp were spawned by the North-West Water Authority from fish collected from the wild, and contained a mixture of fully scaled fish and fish with a reduced number of scales - a strain referred to as Mirror carp. No evidence was available to suggest differences between the growth rates and food conversion efficiencies of the two strains, and for the purpose of this experiment they are considered together under the term common carp.

The fish in each tank were fed by hand in small quantities throughout the day (09:00 - 17:00), except at weekends when a single daily feed was usual. The same number of feeds were given to each tank throughout the experimental period. Baker's Omega carp pellets (Composition - Table 3.1) were used throughout. Periodical weighing and counting of the fish yielded data from which SGR and FCE were calculated.

Similar environmental conditions were maintained in each system with temperatures set at 25°C. Daily measurements were made of ammonia and nitrite concentrations, with less frequent measurements made of oxygen, temperature and pH.

Results: Tabulated data for all three tanks are presented in Appendix 3. Changes in average fish weight with time are plotted in Fig. 3.14 with mean specific growth rates compared in Table 3.16 and mean food conversion efficiencies in Table 3.17. The relationships between specific growth rate and ration size are plotted for common carp (Fig. 3.15) and grass carp (Fig. 3.16). Water quality data is summarised in Table 3.18.

Table 3.16 Comparison of Mean specific growth rates

	Mean specific growth rate	
	Common carp	Grass carp
Mono culture	0.51 +/- 0.34	0.16 +/- 0.34
Mixed culture	0.71 +/- 0.40	0.07 +/- 0.28

Indices of competition (c)

$$C_1 = \frac{0.71}{0.51} = 1.39$$

$$C_2 = \frac{0.07}{0.16} = 0.44$$

$$C_1 + C_2 = 1.83$$

Table 3.17 Comparison of mean food conversion efficiencies

Tank	Species	Mean food conversion efficiencies	
1	CC + GC	0.34 +/-	0.26
2	CC	0.31 +/-	0.20
3	GC	0.08 +/-	0.19

Table 3.18 Summary of water quality data

Tank	Ammonia (mg/l)	Nitrite (mg/l)	Measurement Oxygen (% Sat.)	Temperature (°C)	pH
1	0.69+/-1.02	0.27+/-0.77	85.47+/-11.90	24.47+/-2.21	7.29+/-0.34
2	1.39+/-1.67	0.40+/-1.01	83.21+/- 9.08	25.80+/-2.02	7.39+/-0.19
3	0.33+/-0.22	0.11+/-0.04	91.15+/- 7.61	24.65+/-1.64	7.32+/-0.19

Discussion: Difficulties were experienced in the operation of all three recirculating systems. Pump seals wore out with increasing rapidity, causing reduced flow rates and entrainment of air. Bubbles of air entered the filter bed, and although this probably increased the availability of oxygen in the filter, it also resulted in an increase in turbidity through the resuspension of solids, and movement of the filter media. Abrasion

Figure 3.14

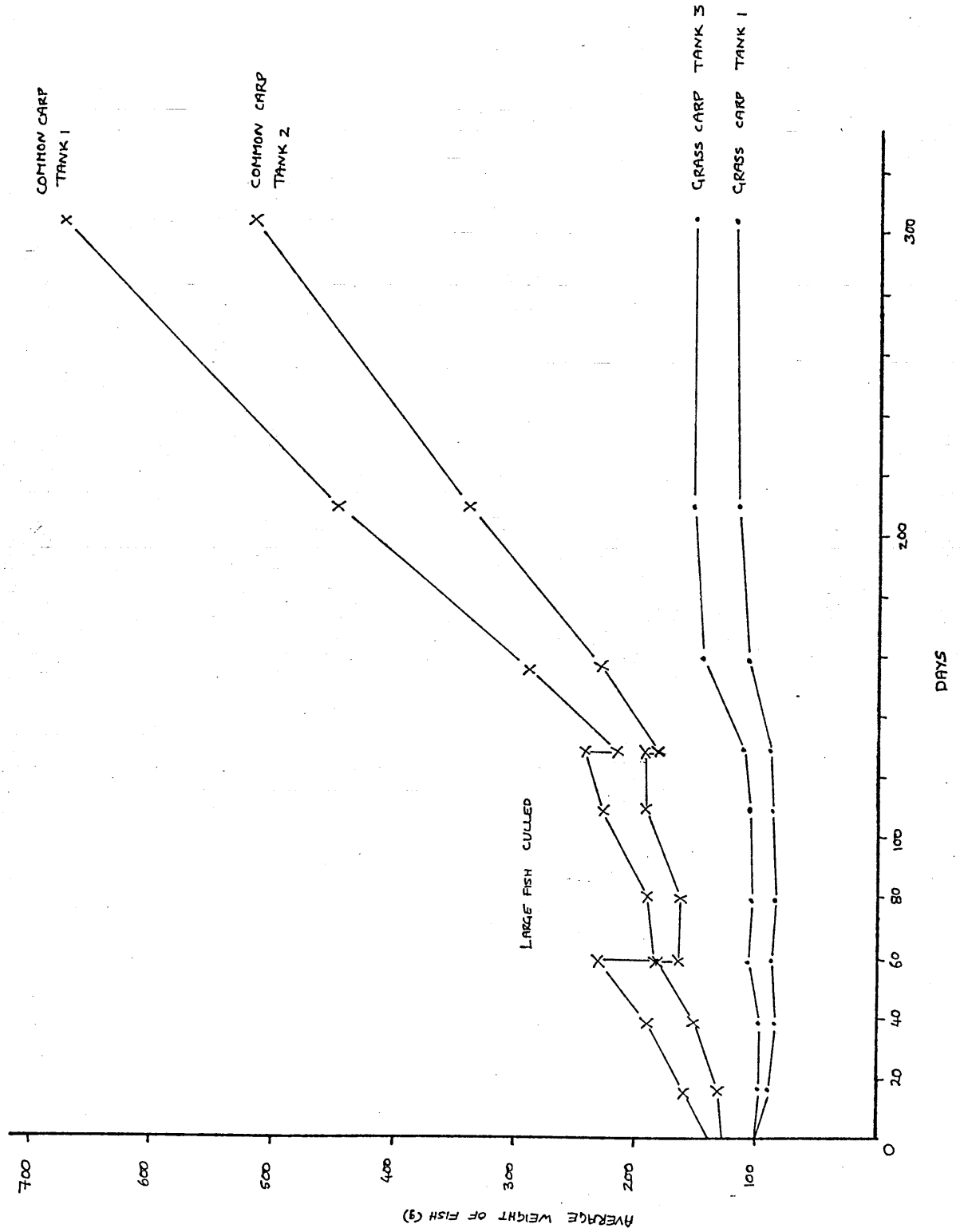


Figure 3.15 Specific growth rate vs ration size in common carp

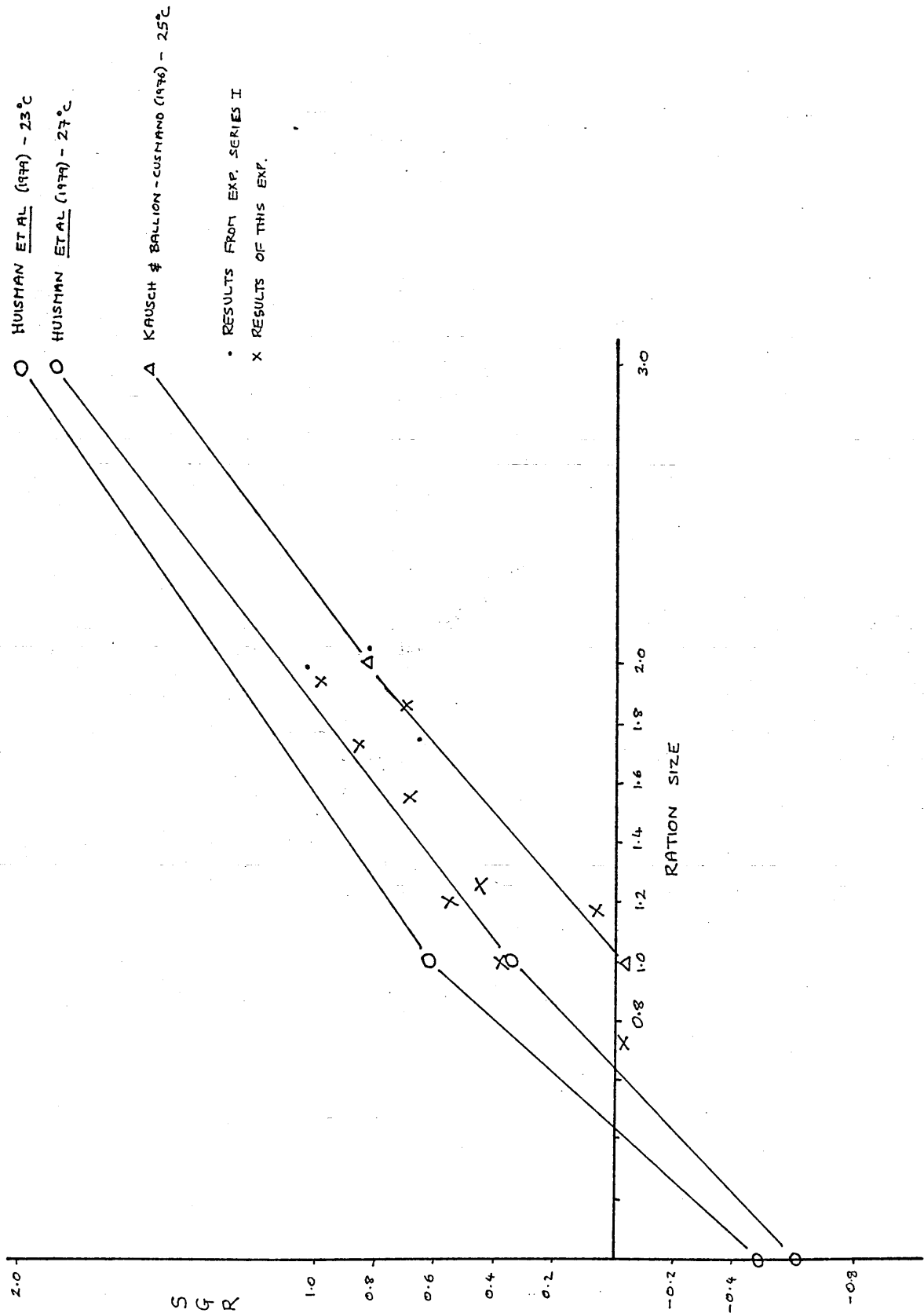
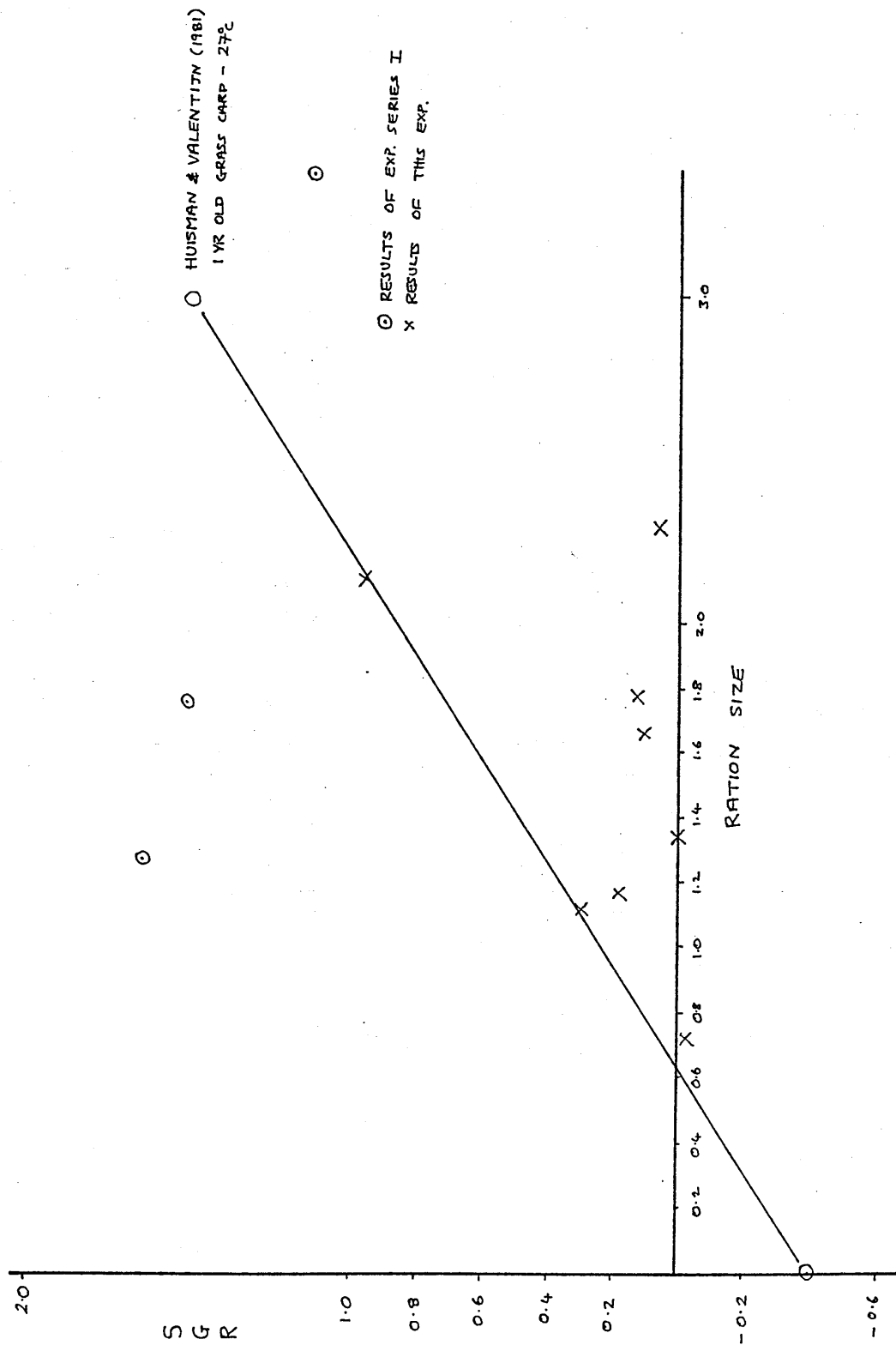




Figure 3.16 Specific growth rate vs ration size in grass carp



caused by the movement of the filter media may have had an effect on the microbial film covering each particle. Entrainment of air also induced a short circuiting of flow through the filter bed. Some difficulties were also caused by total pump failure, the causes of which were uncertain.

These difficulties were partially responsible for the variable performance of the recirculating system throughout this experiment. After periods of apparently stable operation elevated levels of ammonia and/or nitrite were recorded, (Table 3.18). In this respect, the behaviour of the recirculating systems during this experiment was similar to that recorded in experimental series I. During experimental series I, the incidence of elevated ammonia levels was reduced by splitting the daily feed ration into a morning and evening ration. For this experiment the daily ration was supplied throughout the course of the day, except at weekends when one daily feed was usual, (no differences being detected in the occurrence of elevated levels as the result of the weekend feeding pattern). Difficulties with elevated ammonia and nitrite levels were noticeably greater in tank 2. At first it was thought that this was because tank 2 always held the largest weight of fish, yet similar ammonia and nitrite levels were not experienced in tanks 1 and 3, when the weight of fish reached that previously held in tank 2.

The higher average water temperature in tank 2 should have resulted in an increase in the rate of nitrification (Wheaton, 1977), although it would also have reduced the solubility of oxygen. The net effect of an increase in temperature was examined with the aid of the simulation model (5.2.2.3). This indicated that an increase in temperature would reduce the amount of oxygen available in the filter, but that ammonia concentrations would show a decrease.

A possible explanation for the poor performance of tank 2 is that for a given weight of food, ammonia production by common carp was higher than by grass carp. It has been shown that nitrogen excretion in fish is directly proportional to the consumption of protein (Savitz, 1971). If the grass carp consumed only a proportion of their daily feed (see below) then the production of ammonia would be less. In feeding fish a large proportion of dietary protein is used as an energy source (Jauncey, 1979). Huisman and

Valentijn (1981) found the metabolic rate of grass carp to be less than that of common carp. It is possible therefore, that of the protein consumed by grass carp less was metabolised for energy, with a resultant reduction in the excretion of ammonia. Thus one would expect the loading in tank 3 containing grass carp only to be less than in tank 1 containing 50 per cent grass carp (by number) which in turn would be less than in tank 2 containing only common carp. This agrees with the results obtained (Table 3.18).

To maintain similar ammonia and nitrite levels, an increase in nitrification corresponding to the increased ammonia production would be necessary. Together with the increased respiratory demand of feeding fish, this would result in a reduction in oxygen concentration. As Table 3.18 shows, the lowest mean oxygen concentration was accordingly found in tank 2 followed by that in tank 1.

There were no mortalities during this experiment, although the nitrite levels were on average above that recommended by Wickins (1981) and Munro (1978) as the maximum acceptable (Table 2.1). As discussed previously (3.2.5.1) under cultural conditions, 0.1 mg  $\text{NO}_2\text{-N/l}$  is too low and during this experiment 0.5 mg  $\text{NO}_2\text{-N/l}$  was taken as the maximum acceptable concentration, above which feed was withheld.

To enable comparisons to be made between growth and food conversion efficiencies in the three tanks, ration size and number of feeds were kept similar. When it was necessary to withhold food from one tank because of poor water quality, food was withheld from all three. As indicated by the mean specific growth rates in Table 3.16 the growth of the common carp in tank 2 was over three times that of the grass carp in tank 3. The growth of the grass carp was so poor that for four months the fish showed no growth and/or lost weight. The grass carp grown with the common carp in tank 1 showed even worse growth with an average specific growth rate less than half that of the grass carp in tank 3. The common carp in tank 1 however, had higher specific growth rates than the fish in tank 2. This is reflected by the indices of competition (Table 3.16). From the addition of the indices ( $C_1 + C_2$ ) it can be seen that the enhanced growth of the common carp in tank 1 was insufficient to compensate for the reduced growth of

the grass carp, and overall, production was reduced by 8.5 per cent.

As noted in the first experimental series, grass carp are shy and disinclined to feed during the day, whereas common carp are voracious feeders and consume the feed as soon as it enters the tank. It is likely that most of the feed allocated to tank 1 was consumed by the common carp. The suppressed growth of the grass carp in tank 1 may therefore have been partly caused by a lack of food.

The common carp in both tanks 1 and tank 2 did not grow uniformly, with some fish increasing in weight at a faster rate than others. At feeding, these larger fish were more aggressive and appeared to consume more food than the smaller, more timid fish. As indicated in Fig. 3.14, on two occasions it was necessary to remove the larger common carp from both tanks.

The mean food conversion efficiencies for each tank (Table 3.17) show that the most efficient conversion of food was in tank 1. The efficiency of food conversion in tank 2 was on average similar, but showed greater variation. The small weight gains (if any) of the grass carp in tank 3 resulted in very small FCE's, except for one period between days 115 and 160 when the FCE was 0.45 and the SGR was at its highest at 0.97 g%/day, (Fig. 3.14) No explanation is available for this sudden improvement.

If it can be assumed that the food conversion efficiency shown by the grass carp in tank 1 was comparable with that of the grass carp in tank 3, then the common carp in tank 1 were not only growing faster but also converting food more efficiently. This would indicate that the food ration supplied for the common carp in tank 1 (on average 1.90 - 1.32 depending on the proportion eaten by the grass carp) and in tank 2, (1.39 per cent of body weight) was less than optimum. This is in agreement with Huisman (1974, 1978) who found that at 27°C the optimum ration, i.e. the ration for maximum efficiency of food conversion was between 2 - 3 per cent of body weight for fish of 50 - 700 g.

The relationship between SGR and ration size in the common carp held in tank 2 (see Fig. 3.15) is similar to that found for common carp by Kausch and Ballion-Cusmano (1976) but is less similar to that found by Huisman et

al (1979). The differences between the published results can be attributed neither to temperature nor to fish size. Huisman used fish ranging in weight from 56 -111g, while Kausch and Ballion-Cusmano used fish from 16 - 100 g. Better growth by smaller fish should have enhanced the results of the latter authors. During the present experiment fish varied in weight from 18 - 80g. As found by Kausch and Ballion-Cusmano (1976) there was greater variation in growth at lower ration levels. Since in this experiment the lower ration levels reflect a greater number of missed feeds, the growth rates at these lower levels are probably influenced by the short term effect of food deprivation (Randolph & Clemens, 1978). In addition, the results of this experiment were obtained sequentially, and it was not possible to isolate and remove any historical effects on SGR.

The SGR of the common carp in tank 2 during the first experimental series is also indicated in Fig. 3.15. This shows that the SGR during the first growing period in experimental series I was similar to the results of the present experiment but as thought earlier, during the second period it was depressed.

From Fig. 3.16 the growth of the grass carp in tank three appears to be independent of ration size, with little growth recorded at all. This is in marked contrast to the results of the first experimental series (also shown on the graph) where the growth of the grass carp at the lower rations exceeded that predicted from Huisman's data (plotted). A possible cause of the poor growth and food conversion recorded in this experiment may have been the diet. The fish received from the North-West Water Authority were larger than those received from the Southern Water Authority for experimental series I. It is possible that the larger grass carp were less able to adapt to an entirely pelleted diet. Prior to being used in this experiment, the fish from the North-West Water Authority were being grown-on, with a diet of bread, lettuce and pellets. As considered earlier (3.2.5.2) the pelleted diet may have been nutritionally inadequate. The examination of nutritional aspects of grass carp culture was considered to be outside the scope of this investigation, and further use of grass carp as an experimental fish was not considered feasible.

In culture systems where production is based on natural productivity, polyculture enhances production through the utilization of different food resources by the different species. For example, common carp and grass carp are frequently cultured together, since not only are their nutritional habits different, but it is also claimed that common carp derive nutritional benefit from the consumption of grass carp faeces. The results of this experiment indicate that in an intensive system where all the nutritional requirements are supplied by pelleted feeds, mixed culture does not offer any advantages; the enhanced growth of the common carp probably just reflecting the increased availability of feed.

In a similar experiment run concurrently with this one, Lawson (1982) fed grass carp and common carp, grown singly and in polyculture, a diet of minced canteen wastes. As in this experiment, better growth rates were experienced with common carp grown in polyculture while grass carp grew less well. On average food conversion was most efficient in polyculture while grass carp grown on their own had poor feed conversion efficiency. Unfortunately, due to high mortality rates, Mr. Lawson's experiment was prematurely terminated. From the results available no clear indication was given as to whether polyculture enhanced or diminished tank productivity.

Wilson and Hilton (1981) added tilapia to tanks containing channel catfish. With daily rations based on the weight of catfish only, they found that as the density of tilapia increased, so aggressive behaviour and competition for food increased. Although tanks containing tilapia at densities less than 25 per cent of the weight of catfish showed an initial net gain in production from the same feed, this advantage was later lost and as in this study they concluded that mixed cultures were not practical. In commercial fish farming the culture of more than one species in a tank may have an added disadvantage of sorting the fish and thus increase the labour costs of harvesting.

### 3.3.2.2 Weight loss in common carp

Object: In any form of fish culture it is occasionally necessary to withhold feed from the fish. This may be due to poor water quality and/or quantity, preparation for shipment, treatment or lack of staff

(Fredenburgh, 1969). The effect on growing fish is two-fold. Firstly there is no weight gain as no food is given (theoretical loss), and secondly there is weight loss when food is withheld (actual loss). The object of this experiment was to estimate the potential weight loss (the sum of the theoretical and actual loss) in unfed common carp.

Method: The method employed was similar to that of Huisman *et al* (1979). Three recirculating systems were stocked with c. 11 kg of common carp (average weight = 123 g). The fish were grown on at 25°C for 10 days with a daily ration of 3 per cent of body weight. At the end of this 10 day period the fish were starved for 36 hours to clear the gut and reweighed. The fish were then returned to each tank and food withheld for seven days. The fish were then reweighed and weight loss calculated. At each weighing water temperature was recorded.

Results: From the increases in fish weight recorded during the first 10 days, specific growth rates and food conversion efficiencies were calculated (Table 3.19). Weight loss in the unfed fish is recorded in Table 3.20, and daily weight losses calculated (Table 3.21).

Table 3.19 Growth and food conversion in common carp

Tank No.	Initial weight (g)	Final weight (g)	Ration size (%)	SGR (%/day)	FCE	Water temp. (°C)
1	11,100	11,700	3.00	0.53	0.29	24.5
2	11,100	11,340	2.38	0.21	0.17	25.3
3	11,130	11,870	3.09	0.64	0.30	23.6
<u>Huisman et al (1979)</u>			3.00	2.11	0.88	23.0
			3.00	1.91	0.81	27.0
<u>Kausch &amp; Ballion-Cusmano (1976)</u>			3.00	1.58	-	25.0

Discussion: Like previous experiments, some difficulties were experienced in the operation of the recirculating systems and during the initial ten day period some feeds were withheld because of poor water quality. The specific growth rates and food conversion efficiencies

Table 3.20 Weight loss in unfed carp

Tank No.	Initial weight (g)	Final weight (g)	SGR (%/day)	Water temp (°c)
1	11,700	11,280	- 0.52	24.5 - 27.0
2	11,340	10,730	- 0.79	24.5 - 27.5
3	11,870	11,490	- 0.46	23.5 - 26.0
Huisman <u>et al</u> (1979)			- 0.50	23.0
			- 0.62	27.0
Kausch & Ballion-Cusmano (1976)			- 0.68	25.0

Table 3.21 Daily weight loss (%/day)

Tank no.	Theoretical loss	actual loss	potential loss
1	0.53	0.52	1.05
2	0.21	0.79	1.00
3	0.64	0.46	1.10
Huisman	2.11	0.50	2.61
<u>et al</u> (1979)	1.91	0.62	2.53
Kausch & Ballion-Cusmano (1976)	1.58	0.68	2.26

achieved were, however, comparable to the mean SGR and FCE recorded for common carp in the previous experiment (Tables 3.16, 17). In comparison with the SGR and FCE of Huisman et al (1979) and Kausch & Ballion-Cusmano (1976) however, much lower values were recorded.

Withholding food from the fish for a period of seven days had a marked effect on growth, with specific growth rates falling to between -0.46 and -0.79. The weight losses recorded by Huisman et al (1979) and Kausch and Ballion-Cusmano (1976) were similar to those recorded in this experiment with values lying within the variation recorded here (Table 3.20). Differences in the water temperature of each tank may have been responsible for some of the variations in weight loss between tanks. When potential weight loss is calculated (Table 3.21) variation between tanks is reduced. From these results it appears that the higher the growth rate before feed is withheld, the lower the actual weight loss.



Huisman et al (1979) examined changes in the body composition of growing and starved carp, and found that the higher the growth rate, the greater the increase in the proportion of fats in the body. When the fish were starved there was a greater decrease in the proportion of body fat compared with body protein. This supports the hypothesis of Storer (1967) that the energy requirements of starved fish are met first by the utilization of glucose and the mobilization of glycogen, followed by the oxidation of fats and finally amino acid breakdown.

A possible explanation for the results of this experiment is that fish with a lower growth rate, prior to starvation had a lower percentage of body fat. To make the same amount of energy available during starvation as fish with a larger percentage of body fat, a greater proportion of protein must be broken down. Since the energy released by 1g of fat is 1.7 times that released by 1g of protein (Brody, 1945) fish with a greater percentage of their energy needs supplied by protein metabolism will show a greater actual weight loss. Studies of the protein sparing effect of fats have been limited to feeding fish and the identification of a similar mechanism in starved fish should be investigated.

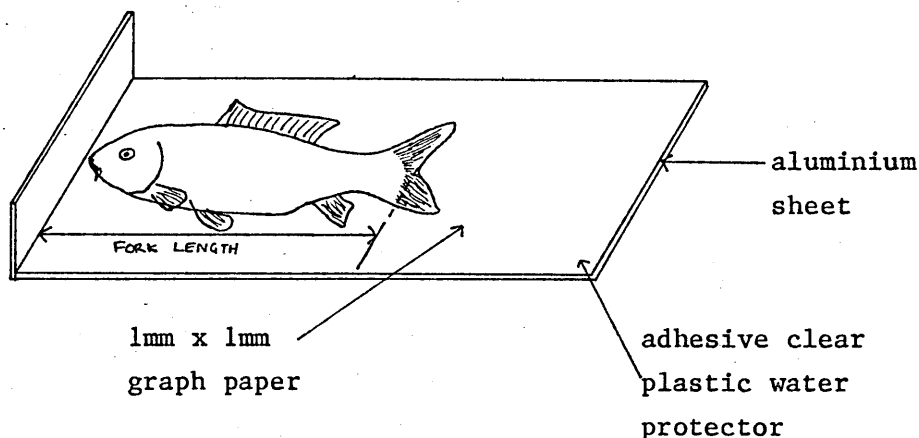
### 3.3.2.3 The relationship between fish length and weight

Object: Increases in fish length have been used by a number of authors as a measure of growth. For example, formulae for determining the amount of feed needed for trout in hatcheries using historical records of feed conversion and length increases were developed by Haskell (1959), Buterbaugh and Willoughby (1967) and Piper (1970). These formulae were later used by Speece (1973) in his method for the "rational design of nitrification facilities for water re-use in hatcheries". Measurement of fish length however, is more difficult and time consuming than weighing and probably induces more stress in the fish. Although a formula for transforming weight into length would be useful, such relationships are not available in the literature. The object of this experiment was therefore to describe the relationships between length and weight for mirror carp, common carp and grass carp.

Method: Common, mirror and grass carp were anaesthetised with benzocaine (Laird and Oswald, 1975) weighed on a top pan balance (Appendix 2) and

measured on a specially constructed board (Fig. 3.17). Length was measured from the snout to the centre of the tail-fork, (fork length - Mann, 1973). Regression analysis was performed on the logarithmically transformed data, and the slopes of the regression equations were compared (Appendix 4).

Figure 3.17 Fish Measuring Board



Results: The results have been plotted (Fig. 3.19, 18 and 20) and a detailed analysis of the length-weight relationships is presented in Table 3.22. The slope of the regression equation was significantly greater ( $d=100$ ,  $df=764$ ,  $P<0.001$ ) for common carp than for mirror carp and in terms of length/weight relationships these should be considered separately. The slope of the regression equation for grass carp was significantly greater than for common carp ( $d=1404$ ,  $df=206$ ,  $P<0.001$ ) and mirror carp ( $d=95$ ,  $df=858$ ,  $P<0.001$ ).

Table 3.22 Regression analysis of logarithmically transformed length/weight data for grass carp (G.C.), common carp (C.C.) and mirror carp (M.C.)

Species	Sample size	$r^2$	Standard error of estimate	Y cm	Equation
G.C.	158	0.99	0.05	45.70	$Y=-2.27 + 3.43x$
C.C.	58	0.99	0.05	8.64	$Y=-1.79 + 3.15x$
M.C.	710	0.99	0.04	8.02	$Y=-1.70 + 3.04x$

where  $Y = \log \text{ weight (g)}$  and  $x = \log \text{ length (cm)}$

In view of the lack of published data, some unpublished data were later made available for comparisons, (Table 3.23, Fig. 3.21).

Table 3.23 Length-weight relations (log transformed data)

Species	Size Range		Equation	r <sup>2</sup>	n	Source
	w(g)	l(cm)				
G.C.	1-1000	4-45	Y=-1.8116+3.014x	0.997	250	Russell
C.C.	78-1230	13.5-36.3	Y=-1.54 +2.99x	0.98	-	Grady

Sources - Pers. Comm.

Discussion: Table 3.22 provides a useful description of the morphology of the laboratory fish. From Fig. 3.21 it is clear that body shape in grass carp can vary considerably with environmental and nutritional factors. The fish cultured in this experiment were considerably plumper than those measured by Frake or Russell. All the fish represented in Fig. 3.21 were imported from the same hatchery in Yugoslavia and it is likely that they were from the same genetic stock (Stott, pers. comm.). It would appear therefore, that the variations in grass carp morphology are the result of nutritional and/or environmental differences.

The shape of common carp has been considerably influenced by fish culture over the centuries. In China the practice of netting ponds and leaving escapees to breed has selected for sleek fully-scaled fish. In Europe, where ponds are usually drained at harvest, the selection of larger and hence faster-growing, deeper-bodied fish for breeding was possible. These tended to be fish with fewer scales - the strain known as mirror carp. Over the years strains have been bred specifically for cultivation, such as the Dor C90's and the Dinklesbuhl carp.

The common carp measured in this experiment were the offspring of wild carp captured from a population of carp imported from China in 1927 (Pritchett, pers. comm.). The mirror carp measured in this experiment were the offspring of mirror carp also collected from the wild (Pritchett, pers. comm.). It is probable that these fish were naturally occurring and had not been influenced by selective breeding as they were captured from an

Figure 3.18 Length-weight relation for common carp

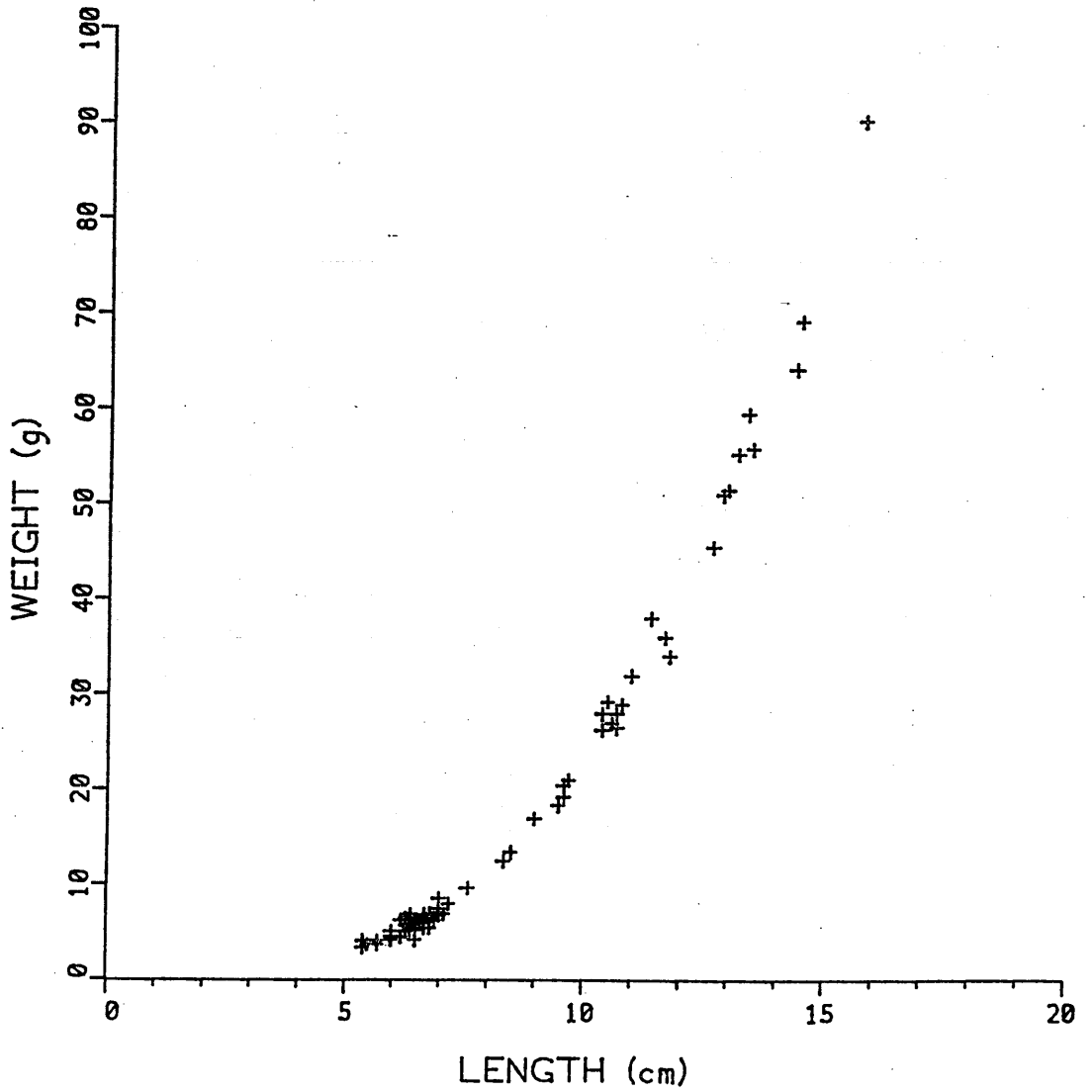


Figure 3.19 Length-weight relation for mirror carp

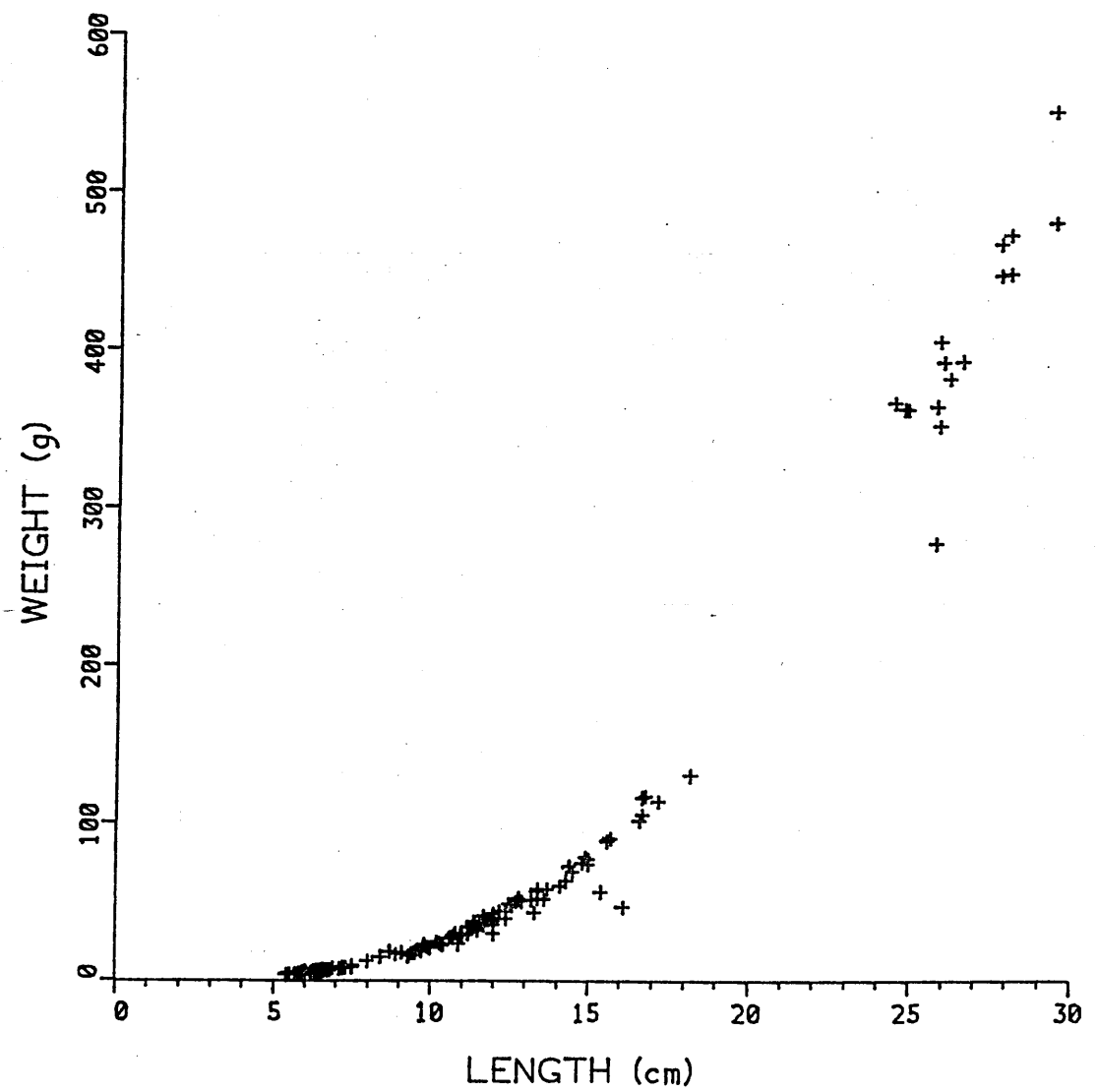


Figure 3.20 Length-weight relation for grass carp

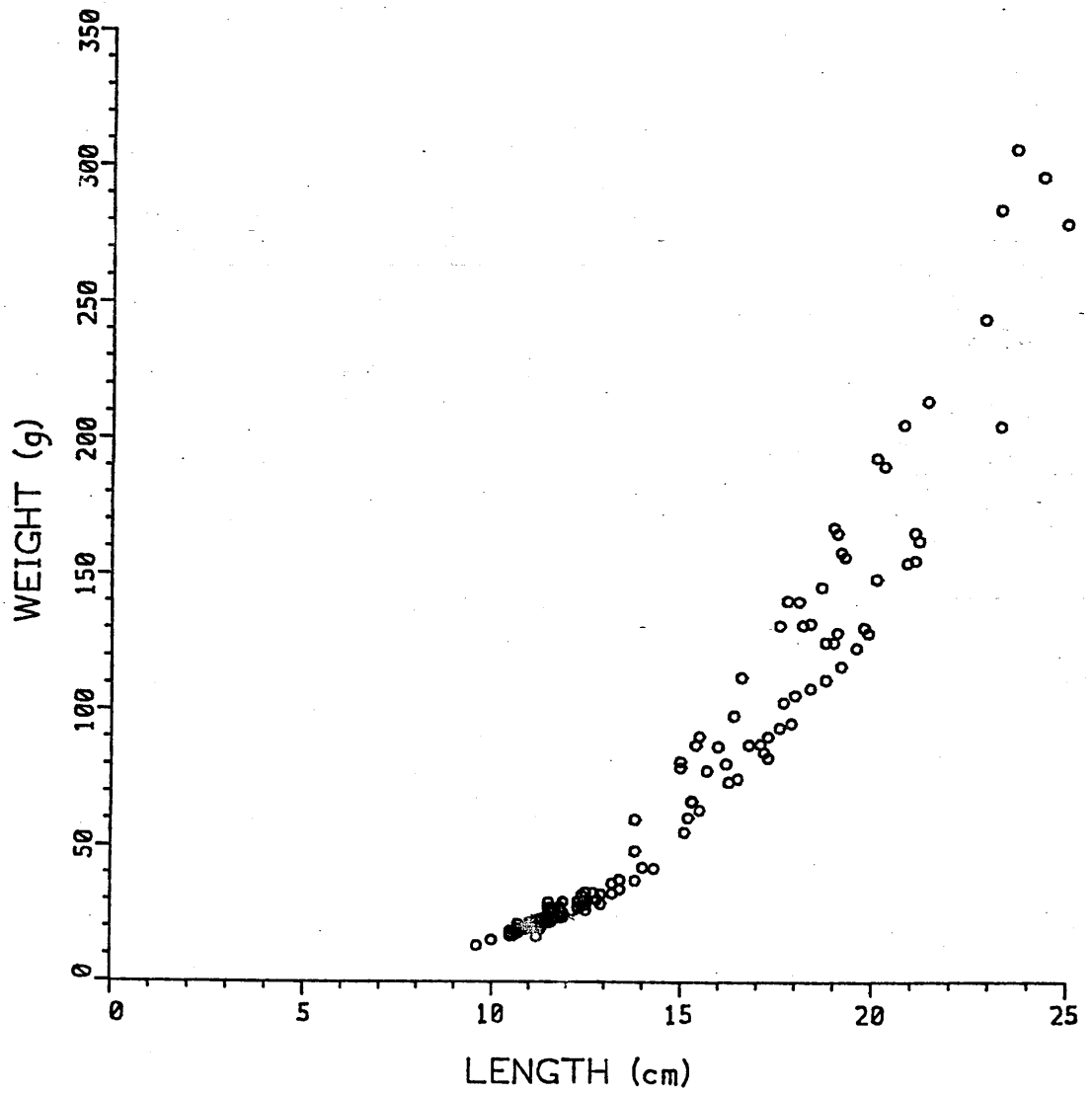
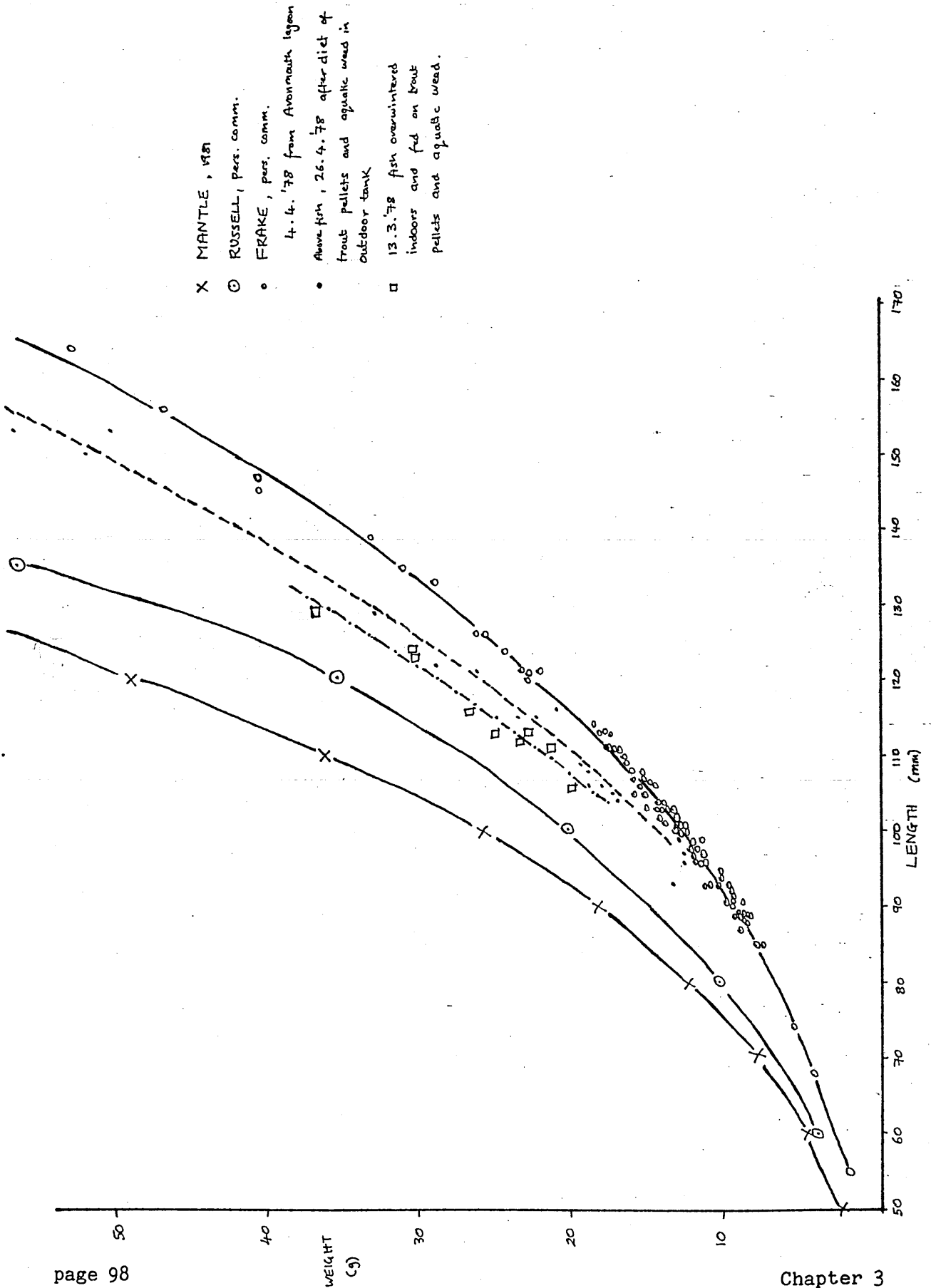


Figure 3.21 Length-weight relationships for grass carp



isolated lake (Pritchett, pers. comm.). Without further information regarding the genetic history of fish measured in this experiment and fish measured by Grady, it is not possible to indicate whether the differences in body shape are largely genetic or environmental.

Since there was a significant difference in the body shape of the mirror and common carp cultured during experimental series II, it is possible that these fish differed in their efficiency of food conversion and growth rate. However, if the mirror carp were not influenced by selective breeding, then any differences would have been small. In addition, although the shape of the mirror carp was significantly different from the shape of the common carp, in absolute terms the difference was small, and by just looking at the fish no difference was distinguishable. Therefore to consider the two strains as one with regard to growth and food conversion (as in 3.3.2.1, and 3.3.2.2) was considered acceptable.

In view of the variations in length-weight relationship found in comparisons of the experimental fish and the fish measured by other authors, it is concluded that the relationships can only be used for fish of the same genetic stock and maintained in the same nutritional and environmental conditions.

### 3.3.3 The production of wastes

This section reviews the following experiments:-

- 1) Solids production by common carp
- 2) Salt accumulation in a closed system

#### 3.3.3.1 Solids production by Common Carp

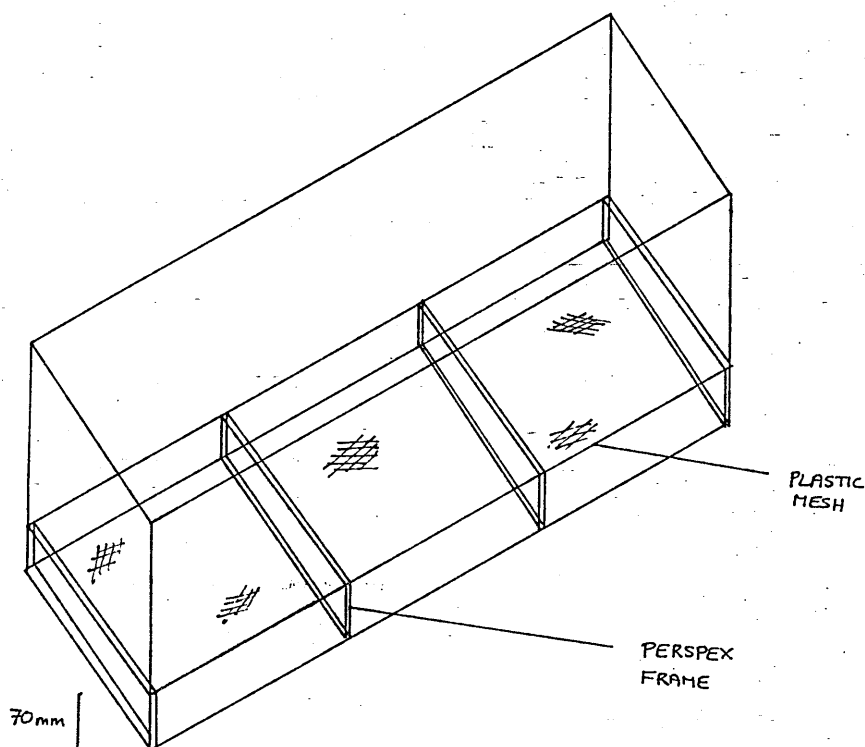
Object: The levels of solids in waste waters can have a significant effect on the efficiency of filters for nitrification (Liao and Mayo, 1974). A high solids loading encourages the growth of heterotrophic bacteria. These have a faster growth rate than nitrifying bacteria (Painter, 1970) and as a result increase the need for backwashing and other maintenance. In addition they reduce the area of media surface available to nitrifying bacteria and have a high oxygen demand. The object of this experiment was to estimate the production of solid wastes by



common carp, information currently unavailable in the literature but necessary in developing design criteria.

Method: An aquarium equipped with a plastic mesh false bottom was employed (Fig. 3.22).

Figure 3.22 Simplified diagram of aquarium showing false-bottom



A perspex frame 70 mm high supported the 5mm mesh through which the solids settled. Resuspension of the solids was prevented by the mesh which reduced water velocity and prevented fish disturbance. The aquarium was stocked with 47 common carp. These fish were fed by hand with Baker's Omega carp pellets (Composition - see Table 3.1), the food being given between 09:00 and 17:00 hours, except at weekends when a single daily feed was usual. The food ration was varied experimentally between a nominal 2.5 and 5.0 per cent of body weight per day. Water quality was maintained by four upflow gravel filters described elsewhere (3.3.4.1). Temperature was maintained at  $25 \pm 1^\circ\text{C}$ . Every one to two weeks the fish were group weighed (Appendix 2) and the settled solids removed by siphoning. Inevitably with siphoning a considerable volume of water was also collected, so after removal the solids were allowed to settle for a day and

the supernatant drained off. The remaining solids and water were then oven dried at 80°C to a constant weight. As it was not possible to separate un-utilised food from faeces, the collected solids represented not only faeces, but also waste food and undigested food. There was very little dust associated with the feed, and observations made at feeding showed that all the food was consumed within 5 minutes of being administered, so the proportion of un-utilised food was considered insignificant. Changes in the concentration of dissolved solids were not measured during the experiment as they were considered negligible.

Results: Data collected during the experimental period are shown in Table 3.24. Where enough data were available the efficiency of assimilation ( $A_E$ ) was calculated by the formula;

$$A_E = \frac{\text{weight of food} - \text{weight of faeces}}{\text{weight of food}} \times 100\%$$

After Solomon and Brafield (1972).

Relationships between weight of faeces and weight of food (Fig. 3.23), weight of faeces and ration size (Fig. 3.24) and assimilation efficiency and ration size (Fig. 3.25) have been plotted. No clear relationship between food conversion efficiency and ration size could be determined and the results of this comparison are not included.

Table 3.24 Results of Solids Production Experiment

Start	Date	Finish	Wt. Initial	Wt. gain (g)	Food (g)	Faeces % of Food (x)	Feed/ Days	FCE	Ration Size (%)	SGR	Age $\frac{\text{Food-Faeces}}{\text{Food}} \times 100\%$
26.11.79	3.12.79	1619	174	135.18	26.14	3/8	1.29	0.94	1.28	73.86	
4.12.79	16.12.79	1793	-128	274.33	20.58	6/13	-0.47	1.15	-0.57	79.42	
17.12.79	3.1.80	1540	20	47.35	56.77	1/18	0.42	0.14	0.07	43.23	
4.1.80	14.1.80	1560	-15	153.6	15.39	4/11	-0.10	1.00	-0.09	84.61	
15.1.80	24.1.80	1545	115	293.44	14.45	7/8	0.39	2.19	0.72	85.55	
24.1.80	3.2.80	1660	135	445.42	16.87	10/11	0.30	2.27	0.71	83.15	
4.2.80	11.2.80	1795	242	515.41	15.85	6/8	0.47	3.75	1.58	84.15	
12.2.80	20.2.80	2037	156	536.13	18.28	5/9	0.29	2.78	0.82	81.72	
21.2.80	29.2.80	2193	129	450.17	21.26	4/9	0.29	2.22	0.64	78.74	
1.3.80	7.3.80	2322	78	352.56	-	3/7	0.22	2.13	0.47	-	
7.3.80	10.3.80	2400	-	-	-	0/4	-	-	-	-	
11.3.80	18.3.80	1794	-34	273.63	17.83	3/8	-0.12	1.88	-0.24	82.17	
19.3.80	27.3.80	1760	110	269.51	19.72	6/9	0.41	1.88	0.67	80.28	
28.3.80	9.4.80	1870	130	383.95	17.92	8/13	0.34	1.67	0.52	82.08	
10.4.80	25.4.80	2000	170	269.39	14.08	12/6	0.27	2.00	0.51	85.92	
25.4.80	13.5.80	2170	400	913.36	14.29	16/19	0.44	2.22	0.89	85.71	

$\bar{x} = 20.67$   $s = 10.90$   
 or  $(\bar{x} = 17.90$   $s = 3.43$  if 56.77% excl.)

Figure 3.23 Relationship between weight of faeces and food

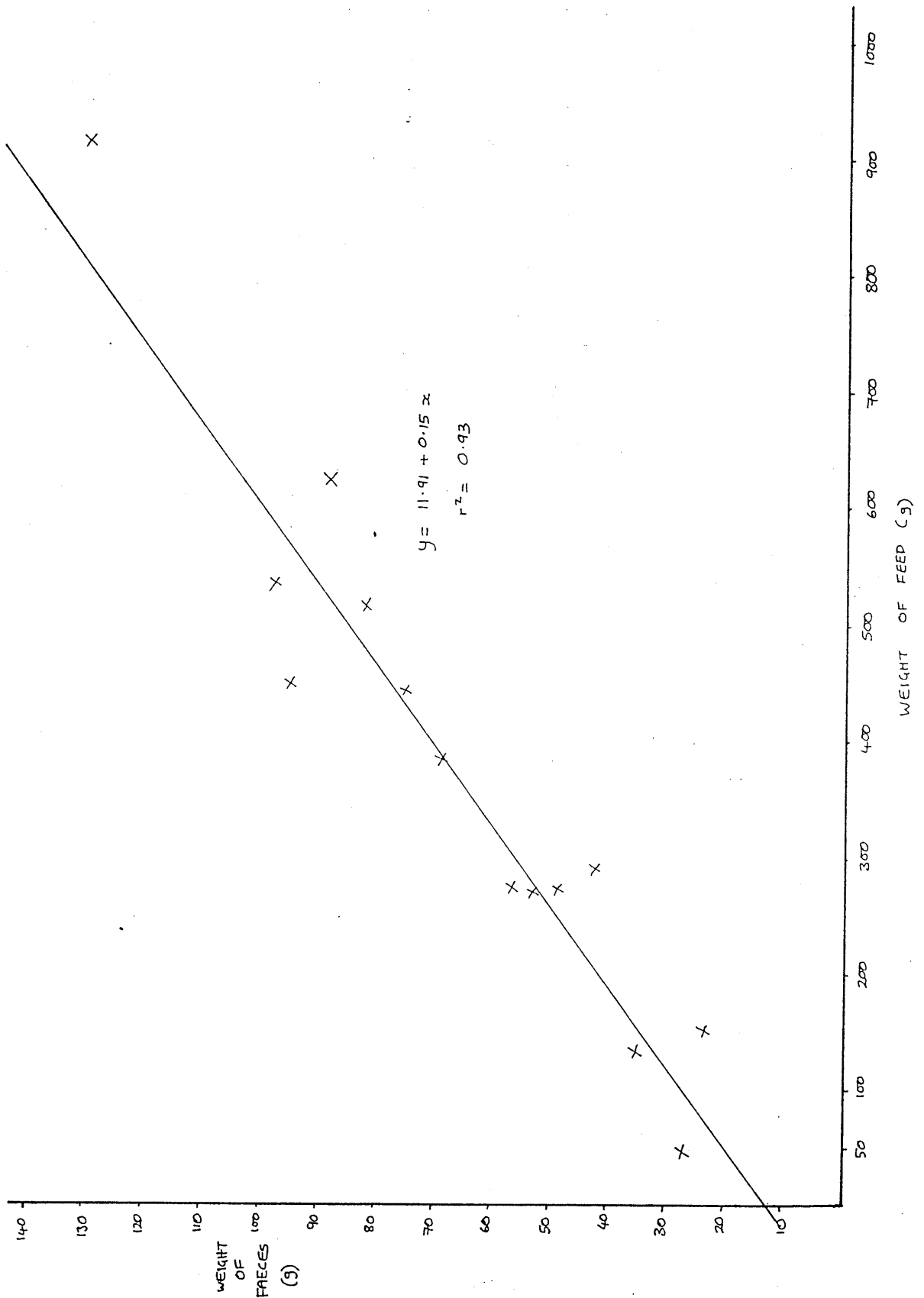


Figure 3.24 Relationship between weight of faeces and ration size

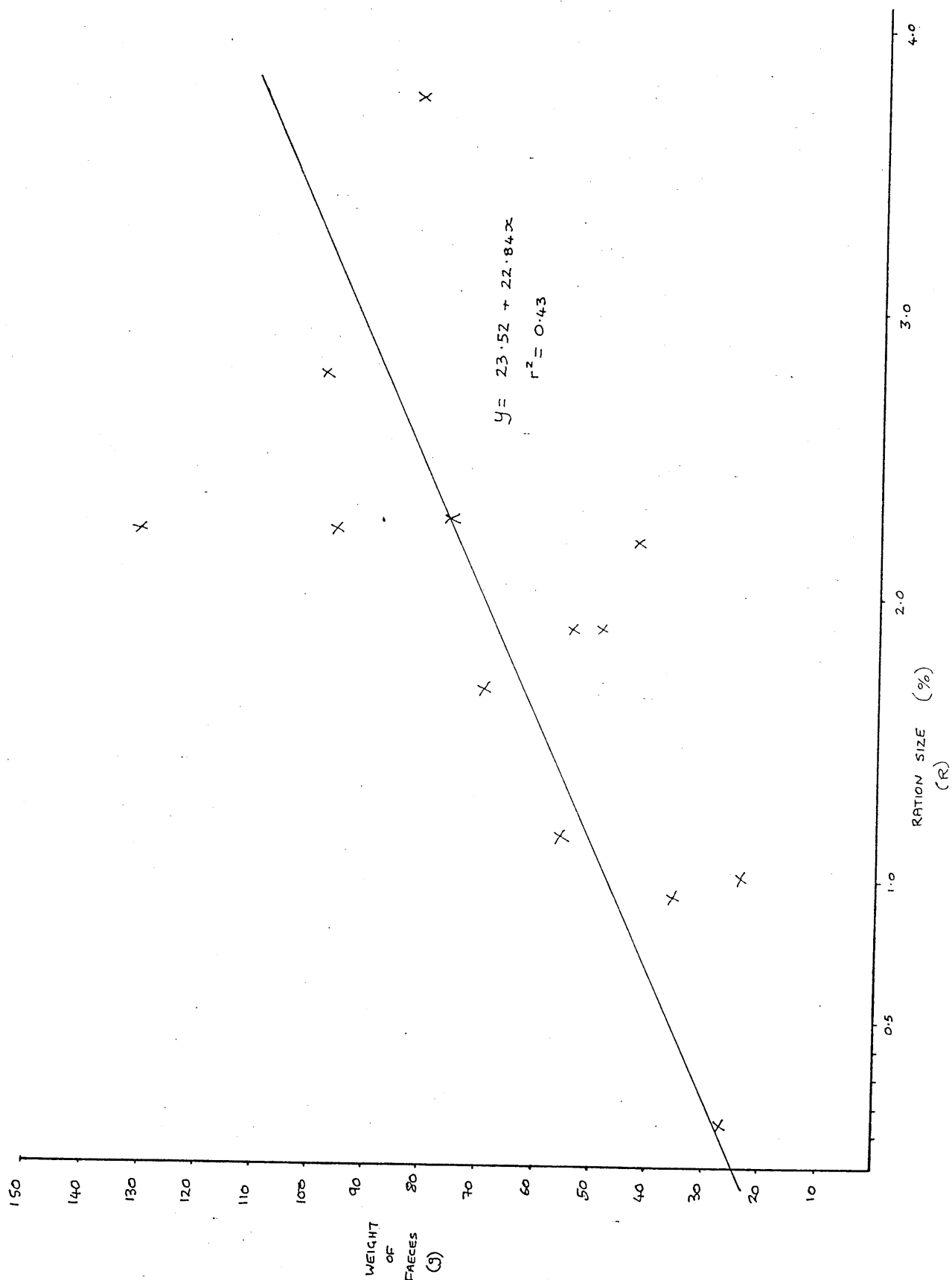
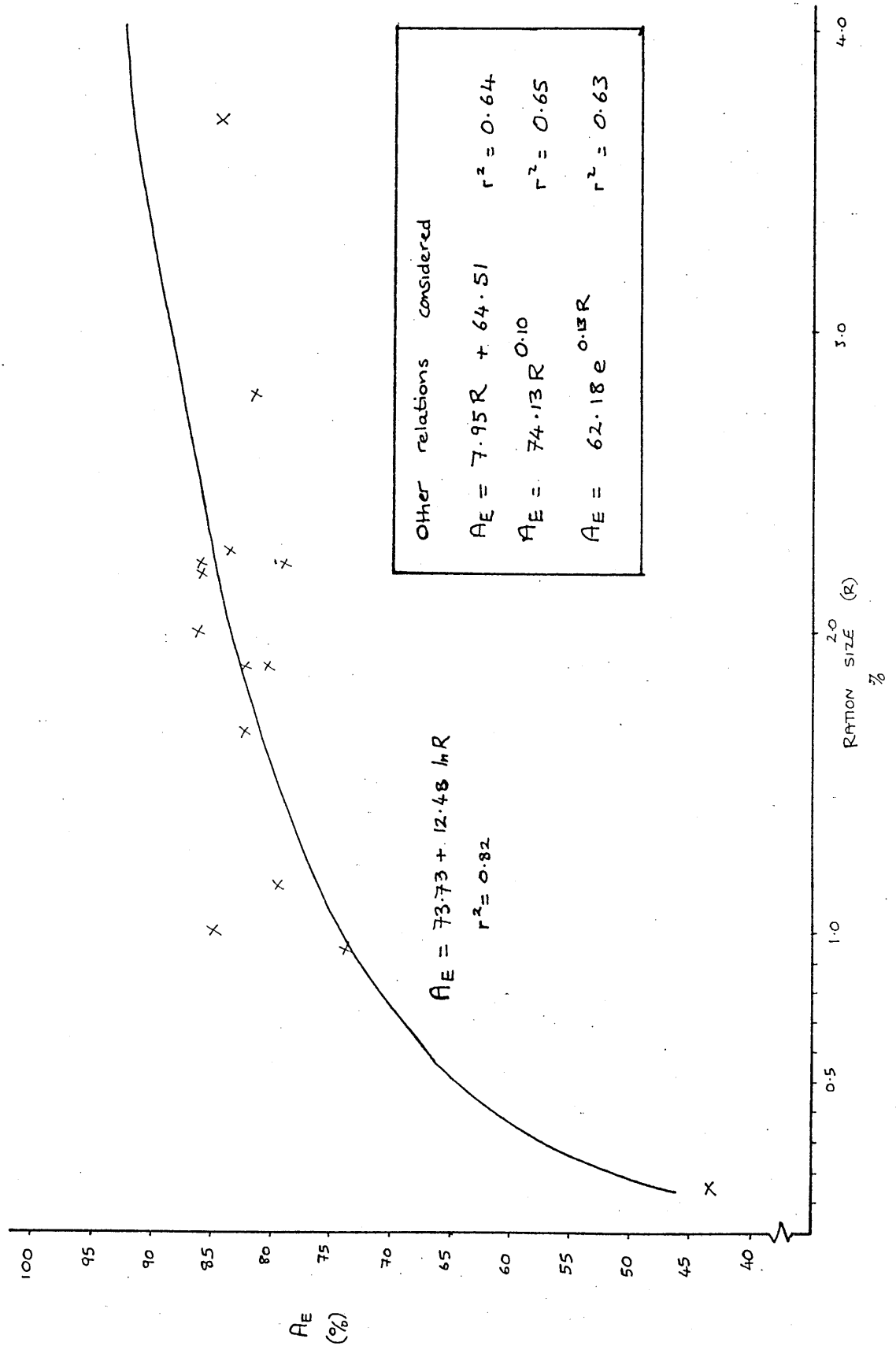


Figure 3.25 Relationship between assimilation efficiency and ration size



Discussion: Fig. 3.23 indicates a strong positive relationship between the weight of feed and the weight of faeces produced. The positive value of the 'Y' intercept probably represents a real phenomenon, with the 'faecal' matter produced with no feed, including material sloughed from the lining of the gut and secretory products such as mucus (Elliott, 1976). The slope of the line of best fit indicates that although the total weight of faeces increases with increasing weight of feed, the weight of faeces as a percentage of the feed decreases. Faecal production from a given weight of food depends on several factors, such as feeding frequency, ration size, temperature and age, weight and condition of fish.

Elliott (1976) found that when weight of feed is expressed in terms of ration level, variations from different fish weights became insignificant. Fig. 3.24, therefore shows the weight of faeces plotted against ration size. This gives a relationship similar to that found in Fig. 3.23, although there is greater scatter around the line of best fit, indicating a limited effect of changes in fish weight on faecal production. Throughout the experimental period, water temperature was maintained within 2°C and variations in faecal production from this source were probably insignificant.

During the experimental period ammonia levels in the culture water intermittently prohibited feeding (Table 3.24). Two possible effects of this on faecal production were to increase the proportion of the faecal material derived from secretions and sloughings from the gut, and to change the efficiency of assimilation. Randolph and Clemens (1978) found that one day's deprivation of food caused the appetite of channel catfish to increase, but that it required two days for the previous growth rate to be resumed. Since reduced ration levels reflect a greater number of missed feeds, this would tend to show a reduction in the efficiency of assimilation at lower ration levels. The effect on faecal production and on the efficiency of assimilation from food being withheld for more than one day are not clear, since the results of Randolph and Clemens (1978) show a suppression of both growth and appetite. There is some uncertainty, whether the results of Randolph and Clemens represent a real phenomenon. In their experiment feeding was based on the use of pendulum feeders in a pond. Their results may thus reflect a learning response by the fish when

no food was provided together with increased respiratory losses from increased food seeking activity. Some caution is therefore necessary in the interpretation of their results.

The underestimation of faecal production by the inefficient collection of faeces during this experiment would also give an apparent effect of increasing assimilation efficiency with increasing ration size. A constant weight of faeces missed would represent a different percentage of the faeces at different feeding levels, representing a greater proportion of the feed at lower rations.

When assimilation efficiency ( $A_E$ ) is plotted against ration size (R) (Fig. 3.25) it can be seen that  $A_E$  increases with increasing ration size, the relationship best represented by the expression;

$$A_E = 73.87 + 12.48 \ln R \quad (r^2 = 0.82)$$

(As indicated on Fig. 3.25 other expressions considered were linear, exponential and power. In all cases a better fit was obtained with the outlying value of  $A_E = 43.23$  included).

An indication of the true nature of this relationship can be found by consideration of the energetically balanced equation;

$$C = \Delta B + R + F + U$$

where; C = consumption,  $\Delta B$  = growth, R = respiration, F = faeces and U = excretion.

The formula used to calculate assimilation efficiency ( $A_E$ ) is a simplified version used where only faeces have been taken into consideration in the determination of waste production (Fischer, 1979). A more rigorous expression where the components are expressed in units of energy is:-

$$\text{Assimilation efficiency (U}_1^{-1}\%) = \frac{C - (F+U)}{C} = \frac{\Delta B + R}{C} \quad \text{Kleouski \& Duncan (1975)}$$



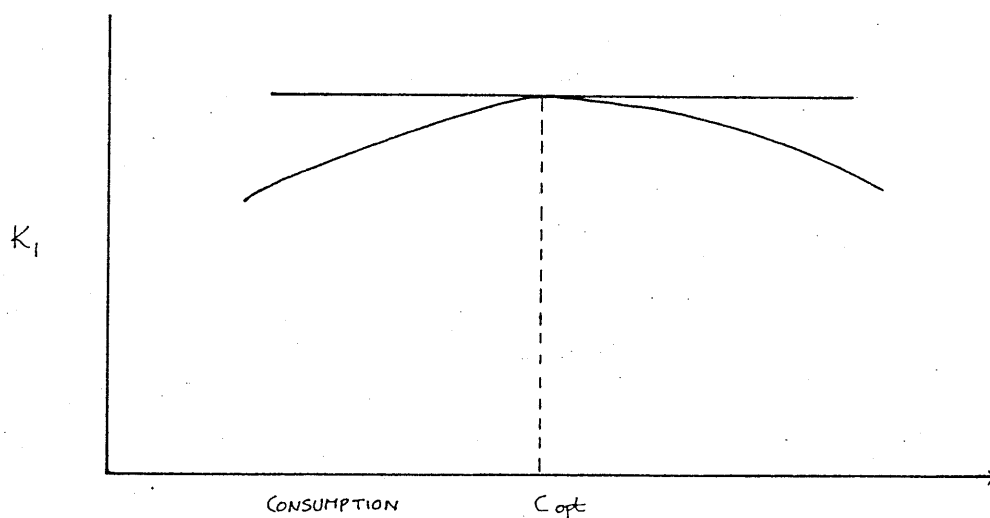
Taking the expression  $\Delta B+R/C$ , if respiration remains constant with respect to  $C$ , then the efficiency of assimilation will be dependent on the relationship of  $\Delta B/C$ . There is some evidence in the literature that the percentage of consumed energy lost in respiration remains fairly constant. Whilst respiration is affected by many factors including fish size, stress, water temperature, feeding and activity of the fish, during the experimental period the effects of variations in temperature and fish size would be minimal. The energy required for the processes of digestion, movement and deposition of food materials (specific dynamic action) is often difficult to distinguish from energy used for activity in feeding fish, and the two are often combined in the component known as apparent specific dynamic action (ASDA).

Muir and Niimi (1972) found that energy losses from ASDA were approximately 16 per cent of food energy irrespective of ration size, and Hambrey (1980) has suggested that ASDA probably amounts to approximately 15 per cent of total food energy over 'a normal range of feeding levels'. Hambrey has also suggested that the relationship between ASDA and energy intake is the basis of the empirical linear relationships generally adopted in fish culture between oxygen consumption and weight of feed (eg Willoughby, 1968; Liao, 1970; Kramer, Chin & Mayo, 1972 and Huisman 1974, see section 4.6.1).

The ratio  $\Delta B/C$  is often known by the coefficient  $K_1$  (Fisher, 1979), and indicates how much energy consumed is utilised for growth. It is therefore, roughly equivalent to the term 'food conversion efficiency'. The generalised form of the relationship between  $K_1$  and consumption is shown in Fig. 3.26. This shows that if respiration is constant at rations less than  $C_{opt}$ , an increase in consumption results in an increase in  $K_1$ , and thus an increase in assimilation efficiency ( $U_1^{-1}\%$ ). At rations larger than  $C_{opt}$ , increases in ration result in a reduction in assimilation efficiency.

Kausch and Ballion-Cusmano (1976) found that in carp fed at 25°C,  $K_1$  was greatest at a ration of 3-3.5 percent, while Huisman et al (1979) found that at both 23 and 27°C,  $K_1$  was greater with a ration size of 3 per cent than at 1 or 5 per cent. Rations during the experiment were less than

Figure 3.26 Generalised relationship between  $K_1$  and consumption



optimum, hence the increase in assimilation efficiency with increased ration (Fig. 3.25).

Kinne (1960) also found that assimilation efficiency ( $A_E$ ) increased with ration size, but Elliott (1976) and Solomon and Brafield (1972) found a decrease with increasing ration. From the discussion above it would appear that these authors were working with rations larger than  $C_{opt}$ , but examination of their results reveals that a wide range of rations were fed. Solomon and Brafield (1972) question their own results, and suggest that even a small constant loss of faeces in collection would give the apparent relationship from a constant true assimilation efficiency. No explanation is available for the results of Elliott. The efficiency of assimilation ( $A_E$ ) calculated by Solomon and Brafield (1972) did not include the energy losses in excretion. When these were included, assimilation ( $U_1^{-1}\%$ ) is found to be almost independent of ration size. Elliott (1976) also found that when the proportion of energy lost in faeces and excretion are combined, variations in ration size and temperature appear to have little effect. Excretion was not measured in this experiment and the coefficient  $U_1^{-1}\%$  cannot be calculated. Further examination of the effects of ration size on faecal losses is seen as an important area for investigation.

### 3.3.3.2 Salt accumulation in a closed system

Object: Long term operation of a recirculating system where the treatment employed is aimed at reoxygenation together with ammonia and solids removal may result in the accumulation of potentially toxic chemicals, (Rosenthal, 1980). The object of this experiment was to monitor the accumulation of inorganic salts in a mature closed system.

Method: Water samples were taken from three recirculating systems being used for the culture of common and grass carp (3.3.3.1). Samples were taken from the fish tanks at the start and finish of two growing periods during which no additional water entered the system and water losses were recorded. Analysis of the water samples was carried out by the Agricultural Development and Advisory Service (ADAS).

Results: ADAS analysis sheets are presented in Appendix 3. From these, changes in concentration have been tabulated (Table 3.25). For selected variables mass changes have been calculated assuming that 50 percent of water losses from the system were evaporative (Table 3.26). These changes can be expressed as a proportion of the weight of food fed, (Table 3.27).

Table 3.25 Changes in concentration

Date	Tank	Weight of fish (g)	Total weight of food(g)	pH	T.D.S. %	NO <sub>3</sub> -N	P	K	Mg	Na	Cl	B	Ca	SO <sub>4</sub> -S	S.S.
6/3	1	8,544-	1238	-0.2	268	14.8	2.00	-1.0	6.0	37.0	28.0	-0.05	-10	-63	0.007
-27/3		8,265													
	2	9,844-	1523		334	-0.03	0.50	23.0	10.0	48.0	53.0	0.06	-12	-92	-0.016
		9,780													
	3	5,950-	897		181	1.2	4.0	10.0	8.0	39.0	41.0		-34	-121	-0.010
		5,920													
25/4	1	9,329-	2181	-0.8	352	34.5	3.75	35.5	7.5	27.0	42.0	0.18	12	14	
-15/5		9,800													
	2	11,101-	2676	-0.2	312	11.6	0.41	32.1	7.5	32.0	44.0	0.21	11	42	
		11,240													
	3	6,250-	1431	-1.3	279	28.3	2.73	37.2	8.0	25.0	35.0	0.17		-12	
		6,570													

No changes were recorded in NH<sub>4</sub>-N, Fe, Mn, Zn and Cu

Table 3.26 Mass changes in concentration

Date	Tank	NO <sub>3</sub> -N	P	K	Mg (mg)	Mg	Cl	Food (g)
6/3-	1	12,761.15	1,724.48	-862.24	5,173.44	31,902.88	24,142.72	1238
27/3	2	-230.36	383.93	17,660.55	7,678.50	36,856.80	40,696.05	1523
	3	955.14	3,183.80	7,959.50	6,367.60	31,042.05	32,633.95	897
25/4-	1	27,460.28	2,984.81	28,256.23	5,969.63	21,490.65	33,429.90	2181
15/5	2	9,664.08	341.58	26,742.83	6,248.33	26,659.52	36,656.84	2676
	3	23,577.01	2,274.39	30,996.69	6,664.88	20,827.75	29,158.85	1431
	x	12,364.55	1,815.50	18,458.09	6,350.40	28,129.94	32,786.39	1,658
	s	11,400.50	1,239.37	12,695.38	825.07	6,299.81	5,759.38	653

Table 3.27 Mass concentration changes as a proportion of weight of food

mg change/100g food +/- S.D.

NO <sub>3</sub> -N	731.66	+/- 677.11
P	138.00	+/- 123.21
K	1072.99	+/- 720.64
Mg	434.15	+/- 172.16
Na	1982.45	+/- 997.45
Cl	2200.10	+/- 838.36

Discussion: Most recirculating systems operate with at least some degree of flush. As a result the accumulation of salts in closed systems has not been measured. Only Rosenthal and Otte (1979) and Otte and Rosenthal (1979) have considered accumulation in closed systems, monitoring the rise in Total Organic Carbon (TOC), Chemical Oxygen Demand (COD) and Biochemical Oxygen Demand (BOD). They found that whilst BOD remained fairly constant, COD and TOC increased. The inclusion of ozonation in the reconditioning process reduced the levels of TOC and COD, breaking many of the long-chain organic molecules not susceptible to biological degradation. They also found that the accumulation of a yellow substance ('Gelbisch'), was a good indicator of the levels of non-biodegradable compounds. The development of a yellow colour in culture water has been noted by a number of authors (Muir, 1977), and was observed during this experiment.

The levels to which salt accumulated varied considerably, both between tanks, and between experimental periods (Table 3.25). Ammonia together with four other compounds showed no change in concentration, whilst only phosphate, magnesium, sodium and chlorine did not show a decrease in concentration in at least one of the tanks. Much of the variability comes from the diversity of sources from which the salts arise. However, it is reasonable to suppose that ultimately these salts are derived from the feed. Therefore the changes in six of the salts have been tabulated together with the weight of food fed to each tank during the experiment

(Table 3.26). Without additional data, it is only possible to present these results in terms of the average increase per weight of feed (Table 3.27).

The accumulation of salts recorded during this experiment were not hazardous to fish health. However, if the system was run with only make-up water added over a long time period, then it is possible that accumulations could present a hazard. The largest increases were of sodium, potassium and chlorine. The increased conductance of the water reflects the increase in salinity (Table 3.25). Increased salinity is not problematic for carp since they can adapt to salinities of 20 parts per thousand (Bardach et al, 1972).

During this experiment increases in the level of both total dissolved solids and non-biodegradable substances (as reflected by an increase in Gelbish) were experienced. It would appear that using biological filtration only, salt accumulation cannot be avoided. Therefore it seems necessary to replace a certain amount of water continuously, or to introduce additional treatment. As previously discussed ozone effectively reduces the level of long-chain organic molecules. Reduction of T.D.S. can be more difficult. Traditionally dissolved inorganic solids are removed by physico-chemical processes such as ion exchange, electrodialysis or reverse osmosis. Of these only ion exchange would appear to be feasible in fish culture (KCM,1972). Some of the dissolved inorganic solids are potential plant nutrients and in a number of recirculating systems accumulation of nitrates and phosphates in particular have been prevented by the use of hydroponic plant culture (Naegel, 1977, Lewis et al, 1978, 1980, and van Toever and Mackay, 1980). To date only limited consideration has been given to biological reduction of inorganic solids in waste waters and this is seen as an important area for further study.

#### 3.3.4 Filter Performance

This section contains only one experiment.

##### 3.3.4.1 The effect of grain size and flow rate on nitrification

Object: The efficiency of nitrification in a biological filter bed is dependent on retention time, hydraulic load, media specific surface area

and the waste strength and composition (Forster, 1974; Liao and Mayo 1974; Hirayama, 1974; Speece, 1973 and Kramer, Chin and Mayo, 1972). There is some confusion in the literature as to the importance of some of these factors in the design of a recirculating system. For example, Forster (1974), showed that as the hydraulic load on a filter increased, so the efficiency of ammonia removal decreased. Liao and Mayo (1974) on the other hand found that although at first hydraulic load appeared to influence the efficiency of ammonia removal, their later results did not confirm this, and ammonia removal efficiency seemed to be independent of hydraulic load. Liao and Mayo did find that nitrification was directly related to retention time. This result is surprising since retention time is the reciprocal of hydraulic load (although in practice it is usually modified by the configuration and packing of the bed and by flow characteristics).

Hirayama (1974) showed that the oxygen consumption of a biological filter (an index of nitrification and carbonaceous oxidation) was directly proportional to the specific surface area of the filter media employed. Thus the selection of an appropriate filter medium to match the production of pollutants forms an important part of his design method. The design of Speece (1973) also balances the production of pollutants with the size of filter medium, by the calculation of specific surface area required per 100 lbs of trout as a function of fish length and temperature. However, Liao and Mayo (1974) found that unit ammonia removal was independent of the size of media employed.

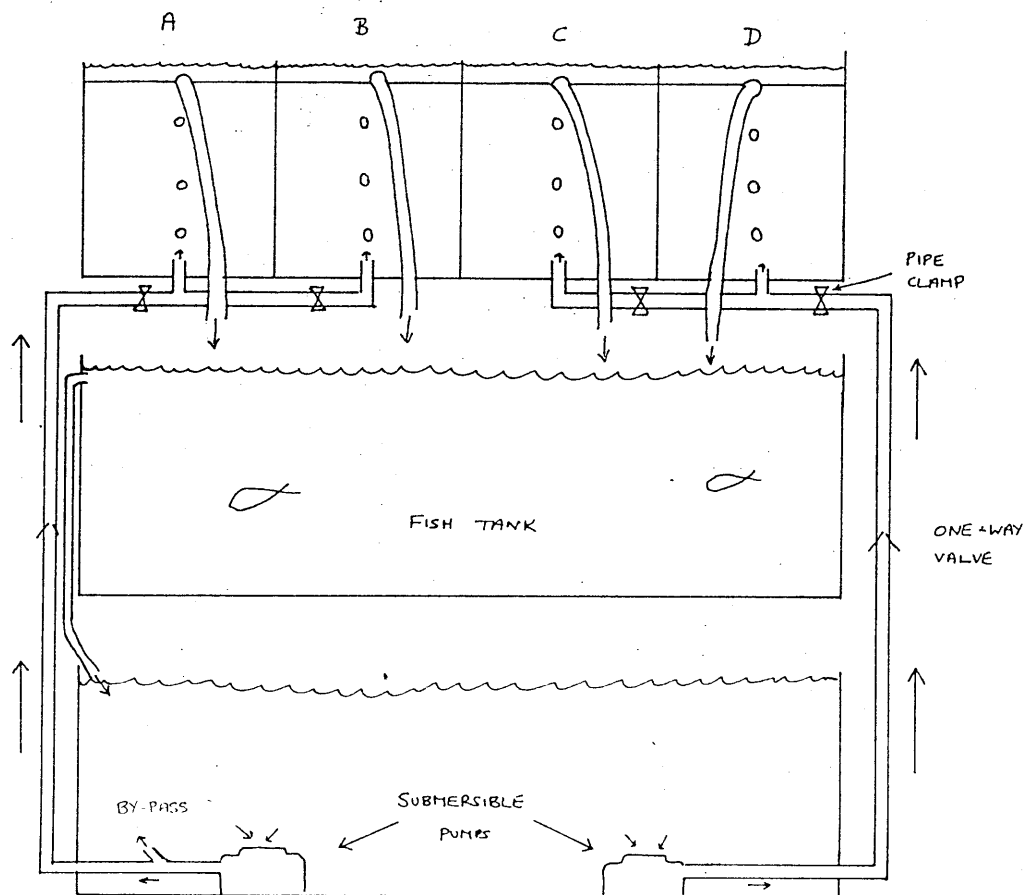
In view of this confusion it was decided to investigate the effect of grain size and flow rate on ammonia removal in an upflow filter.

Method: Two gravels, similar to those used in the main laboratory filters were used in this experiment. Filter beds (A) and (C) contained 2.5 - 6.3mm diameter gravel, and beds (B) and (D) contained 1 - 2mm diameter sand. To achieve two different flow rates two submersible pumps were employed. The first pump serviced filters (A) and (B) and was fitted with a by-pass for flow control, whilst a second pump without a by-pass served filters (C) and (D). Clamps on the pipes entering the filters allowed fine adjustment of flow rate. Failure of electricity supply on one occasion resulted in the filter draining back through the pumps and sand prevented the



restarting of the pumps. To prevent recurrence on-line one way valves were fitted. The experimental rig is shown in Fig. 3.27.

Figure 3.27 The experimental rig



This experiment was run concurrently with the measurements of faecal production (3.3.2.1) as a preliminary to a more rigorous period of experimentation following the termination of the waste production experiment. In this experiment therefore, the filter was fully conditioned and supporting a population of mirror carp. The initial conditions of operation are indicated in Table 3.28.

Results: Ammonia concentration was measured each day in the fish aquarium prior to feeding. For the purposes of this experiment additional measurements of ammonia concentration were made on a number of occasions in the influent water (samples taken from just above the inflow to the pumps), and in the outflow from each of the filters (Appendix 6). These results are summarised in table 3.29. Flow rates were determined from the

time taken to fill a vessel of known capacity. Five measurements were taken and the mean recorded (Table 3.30).

Table 3.28 Initial Operation Conditions

	Grain size (mm diameter)	Filter volume (l)	Flow rate (l/hr)	Hydraulic load (l/l/hr)
Filter A	2.5 - 6.3	34.3	129.30	3.77
B	1.0 - 2.0	31.9	45.30	1.42
C	2.5 - 6.3	31.9	174.90	5.48
D	1.0 - 2.0	34.3	42.00	1.22

Temp = 25°C      Fish Mirror carp = 47  
 O<sub>2</sub>% = 72%                      weight = 1545 g  
 Feed Baker's 'Omega' pellets, 2.5% of body wt/day

Table 3.29 Ammonia Removal by Filters (mg/l)

	Influent ammonia concentration	Effluent concentrations			
		A	B	C	D
$\bar{X}$	0.29	0.26	0.37	0.27	0.31
S	0.02	0.01	0.16	0.02	0.07

Table 3.30 Flow Rates through the Filter Beds

Date	Filter (l/hr)			
	A	B	C	D
19/12	164.23	84.37	247.08	78.14
16/1	129.30	45.30	174.90	42.00
5/2	94.20	6.00	102.60	6.00
12/2	54.60	3.60	76.20	3.00
7/3	20.18	0.08	11.40	0.43

Discussion: As shown in Table 3.30, control of flow rates proved very difficult. Coating of the pipework and pumps by filamentous growths gradually reduced flow rates to all filters. Perhaps more importantly, differences in the permeability of the sand and gravel were difficult to overcome by use of simple pipe clamps. These difficulties were accentuated

by the more effective removal of solids from the culture water by the sand filters. Thus over a 79 day period (19/12 - 7/3), the flow rates in filters A, B, C and D were reduced by 87.7, 99.9, 95.4 and 99.5 per cent respectively (Table 3.30). In a submerged filter where all the oxygen requirements are met by the influent water, reduced flow rates limit the amount of oxygen available for nitrification and carbonaceous oxidation. Therefore, since the conditions under which each filter operated could not be controlled, comparisons between the performance of each filter are not valid.

The filtration of solids from the culture water, while at first causing differential reduction of flow rates within the four filters eventually resulted in clogging of interstitial spaces. This changed the flow patterns within the filters, leading to short-circuiting and the development of "dark zones". These "dark zones" probably represent areas in the filter bed with little through-flow and low oxygen concentrations. The largest number and area of dark zones developed in the sand filters. Some dark zones did develop in the gravel filters, but the principal visible effect was the accumulation of solids around the inflow to the filters.

The measurements of ammonia removal (Table 3.29) indicate that the efficiency of all four filters was poor. On average only the gravel filters A and C had a lower ammonia concentration in the effluent, although if the last two measurements are excluded in the calculation of means, all four filters are similar. However; examination of Table 3.29 reveals that by the end of the experiment, the concentration of ammonia in the filter effluents were frequently higher than the influent concentrations, (although generally not in all four filters at the same time). Since the influent and effluent ammonia concentrations were recorded at the same time, they are not truly comparable. A better measure of ammonia removal would be obtained if the ammonia concentration in the effluent was made after the measurement of the influent concentration, the time lag between measurements depending on the flow characteristics of the filter. This, however, would only account for elevated concentrations if the ammonia levels in the influent water were falling rapidly.

The main cause of the increased ammonia concentration in the effluents is likely to be the secondary production of ammonia by ammonification in the areas of low oxygen concentration within the filters. The sand filters (B and D) had the lowest flow rates and the largest "dark zones". It is not surprising then that filters B and D showed the largest increases in ammonia concentration (Table 3.29). It is possible that the "dark zones" were the main areas for the secondary production of ammonia, but no evidence is available to support this. The sudden rise in ammonia concentration in the effluents from filters B and D at the end of this experiment were associated with an almost complete clogging of the filters and consequent loss of flow. To ensure continuity of the faecal production experiment some flush was necessary and filters B and D were mechanically agitated and thoroughly backwashed. As a result this present experiment was terminated.

The filtration of solids by the filters not only affects the efficiency of ammonia removal by reducing the effective volume, but it also encourages the growth of heterotrophic bacteria. The growth of heterotrophs can reduce the efficiency of ammonia removal by competing with the nitrifying bacteria for oxygen and reducing the area of media surface available to nitrifiers for attachment, (see 4.5.2.2).

Many of the difficulties encountered in this experiment were the result of accumulating solids. Backwashing was not possible since this would severely disrupt the bacterial populations. For this reason a number of authors have based their studies of nitrification on "artificial" culture water which does not contain any organic matter, (Forster, 1974; Haug and McCarty, 1972). The validity of such work when applied to fish culture has been questioned (Wheaton, 1977).

In practical terms some differences in ammonia removal can be seen with different grain sizes. From the results presented here, it seems that under fairly intensive culture conditions the theoretical advantages of increased specific surface area per unit volume are outweighed by the disadvantages of clogging, short-circuiting and increased head loss through the system. Plastic media in the 2.5 to 7.5 cm diameter range appear to avoid the clogging problems while providing reasonable surface

area per unit volume (Wheaton, 1977 and Liao and Mayo, 1974). Wheaton (1977) suggests that rock media should be at least 2cm in diameter and preferably larger, to prevent clogging while Spotte (1970) suggests 2-4 cm diameter gravel for filter media. Although the particle sizes used in this experiment are too small for commercial culture systems because of the high levels of organic loading they may be appropriate for small aquaria. Indeed under low solids loading it appears that smaller grain sizes provide an advantage (Hirayama, 1974).

### 3.3.5 Conclusions

The second experimental series focused on three areas identified as important during experimental series I, with the objective of describing relationships between variables, evaluating their relative importance and indicating conflicts and interdependence (2.5). Within the limitations of this thesis, to fulfill this objective in all three areas proved impossible. The experiments conducted provided 'first estimates', and in more conventional research would be a preliminary to further detailed investigation, being used to show the range of results expected and changes to experimental design. In this thesis, however, the second phase of more detailed experimentation is replaced by simulation modelling, with the results of the observation and experimentation of the first phase used to aid model construction.

The conclusions drawn from the second series of experiments were:

1. Common carp were better suited to an all pellet diet than grass carp and in intensive culture systems based on pelleted feed, the combined culture of these species does not offer any advantages over single species culture. Good growth rates and food conversion can be achieved with common carp by regular feeding and ration levels between 2-3 percent. Withholding feed because of poor water quality, and reduced ration levels result in poor growth rates and low food conversion efficiency.
2. Weight loss in starved fish held at 25°C is approximately 0.5%/day. Actual weight loss is dependent on growth rate prior to starvation; a high SGR results in less weight loss due to increased availability of fat for metabolism. Potential weight

loss shows less variation.

3. Precise relationships between fish length and weight can be calculated but their usefulness is limited to fish of the same genetic stock and maintained in similar nutritional and environmental conditions. Relationships for grass, common and mirror carp were found to be significantly different ( $p < 0.001$ ), but differences between mirror and common carp were too small to be discerned by eye.
4. The weight of faeces produced by common carp on a pelleted diet was directly proportional to the weight of feed and ration size. The relationship between assimilation efficiency and ration size follows a parabola, with efficiency increasing with increased ration size up to an optimum ration at which feed conversion efficiency is maximal. For common carp optimum ration size was thought to be approximately 3 per cent.
5. Without flush, there was an accumulation of salt within the system. Although theoretically the rate of accumulation should be related to the rate of feeding in each tank, with the limited data available there was considerable variation when expressed in this manner. Nevertheless the relationships produced provide a general guide to the changes occurring in the quality of the water during a period of closed-circulation.
6. The advantages of increased specific surface area with small grain filter media are offset by the increased rate at which the filter becomes clogged, with subsequent reduction of flow, short-circuiting and eventual failure as a biological or mechanical filter.

## CHAPTER FOUR SIMULATION MODELLING

### 4.1 Introduction

### 4.2 Model Construction

### 4.3 Growth Sub-model (.WEIGHT)

### 4.4 Sub-model .SIMPLE

### 4.5 Ammonia Sub-model (.AMMO)

#### 4.5.1 Ammonia production

#### 4.5.2 Ammonia removal

##### 4.5.2.1 Nitrification

##### 4.5.2.2 The production of organic wastes

### 4.6 Oxygen Sub-model (.OSIM)

#### 4.6.1 Removal of Oxygen

##### 4.6.1.1 Oxygen removed by fish respiration

##### 4.6.1.2 Oxygen consumed by oxidative processes

#### 4.6.2 Addition of Oxygen

### 4.7 Assembly of the model

### 4.8 Operation of the model

#### 4.8.1 Running the model

#### 4.8.2 Model output

## 4. SIMULATION MODELLING

### 4.1 Introduction

There were three distinct aims in building the model;

- (1) To indicate areas of inadequate knowledge
- (2) To understand the behaviour of the laboratory recirculating systems
- (3) To determine carrying capacity and optimum design of filters for recirculating systems

The dynamics of the laboratory recirculating systems were modelled using a computer programme written in Dynamo (Forrester, 1968 and Pugh, 1970) and run on the Cambridge University IBM 370/165. Dynamo was chosen since it is a relatively simple high level language with which the author had some familiarity and because many of the commonly occurring features of dynamic systems are easily represented. Dynamo was written to represent the diagramming convention known as 'System Dynamics' (Forrester, 1968). Throughout this thesis, diagrams of the model have been drawn according to this convention. A key to the symbols used is found in Fig. 4.2 and again for convenience on the loose insert in Appendix 12.

The model was built using a multistage approach (Shannon, 1975; Mihram, 1972). This is an iterative process with major changes made to the model particularly as a result of verification and validation. The model presented below represents a final version arrived at after two earlier versions. Differences between these three versions are detailed in section 5.2.1.



## 4.2 Model Construction

The first stage was to produce a comprehensive list of all the variables and pathways that were considered relevant. With a view to keeping the model as simple as possible, this list was then shortened to those variables which the experimental programme showed to be most important. The list is presented in appendix 7.

A diagram was produced to show the relationships between the relevant variables. This was later used as the basis for the full network diagram of the System Dynamics model (Fig. 4.13).

The development of a System Dynamics model requires the selection of 'levels' and 'rates'. A level can be defined as a 'state variable' (Forrester, 1968), that is a variable whose value describes the state or condition of the system at any given time. Rates describe the flow between the levels. The choice of levels should already have been partly made, through the earlier stage of determining appropriate measures of the system's performance (i.e. measures of effectiveness - 2.4). In this model the state or condition of the system is described by the variables:

Weight of fish	(W)
Ammonia concentration in fish tank	(AT)
Ammonia concentration in filter	(AF)
Oxygen level in tank	(OT)
Oxygen level in filter	(OF)

The model was based on these five levels. It proved simpler to divide the model into four basic parts or sub-models. These sub-models were:

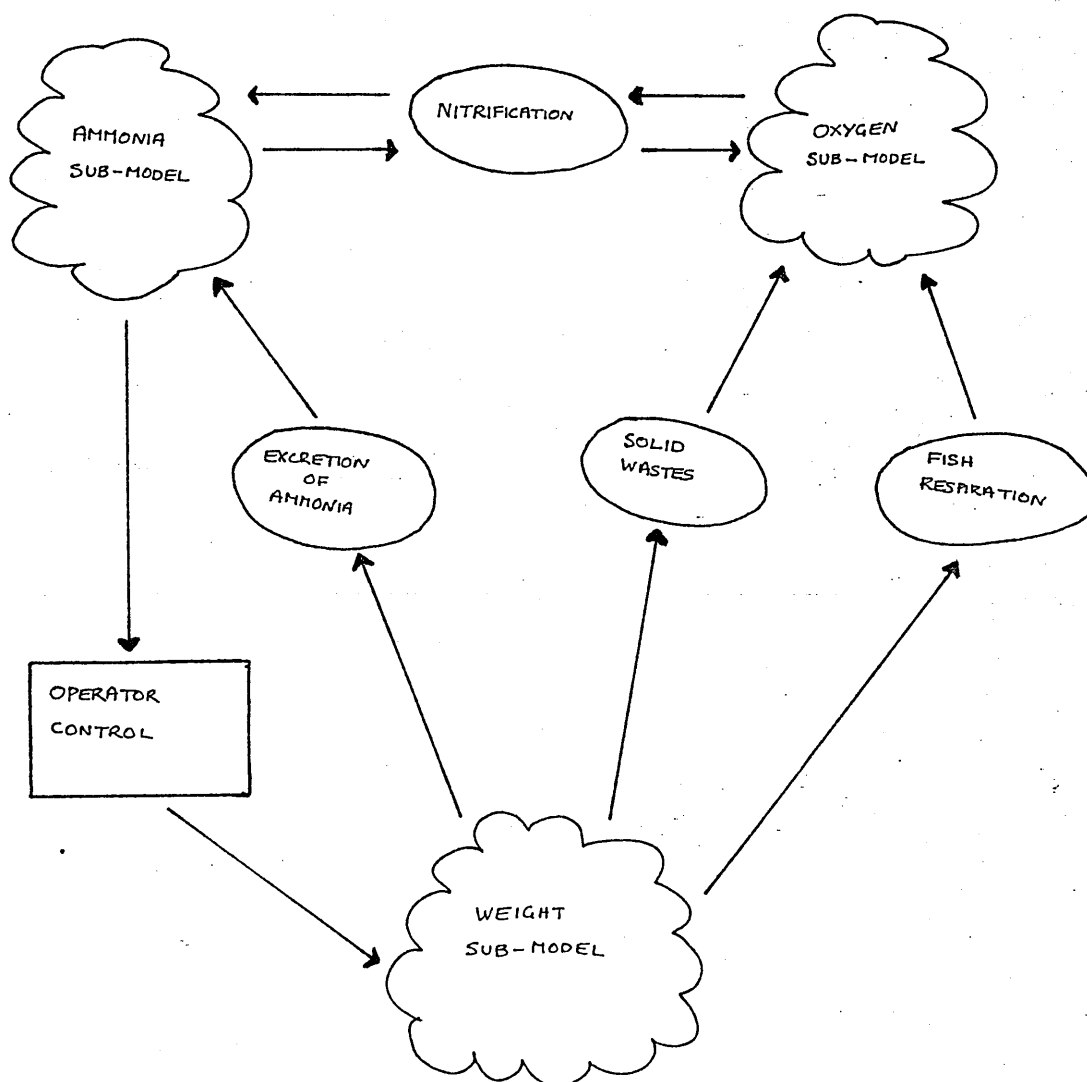
- .WEIGHT concerned with the level of fish biomass and the feeding regime employed. It incorporates a simple growth model.
  
- .SIMPLE is a structure common to both .AMMO and .OSIM; it is a closed loop containing two levels joined by a circulation rate.

.AMMO deals with the levels of ammonia in the system, its production and removal.

.OSIM models the levels of oxygen, its addition and removal within the system.

Each sub-model was constructed and tested before being brought together to form the final model. The relationships between the sub-models (except .SIMPLE which is incorporated into .AMMO and .OSIM) are shown in Fig. 4.1.

Figure 4.1 Simplified Model Structure



### 4.3 Growth Sub-model (.WEIGHT)

This part of the model deals with the biomass of fish in the tank (level W) the feed given (F) and the rate at which weight increases (WIR) and decreases (WDR). The increase in fish weight (W) from feeding is calculated as

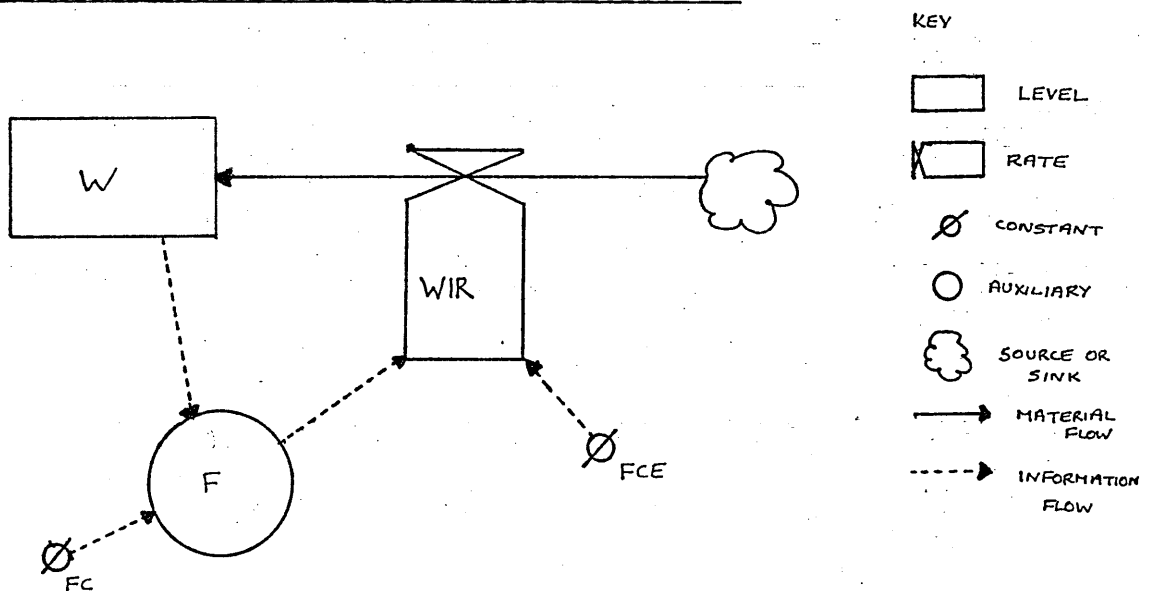
$$\begin{array}{l} \text{Rate of weight} \\ \text{increase (WIR)} \end{array} = \begin{array}{l} \text{Food given} \\ (F) \end{array} \times \begin{array}{l} \text{Food conversion efficiency} \\ (FCE) \end{array}$$

where the food given is a percentage of the total weight of fish (FC) calculated from

$$F = W \times FC$$

This is represented diagrammatically in Fig. 4.2.

Figure 4.2 Mechanism for increase in fish weight



A key to this and other System Dynamics diagrams are presented for convenience on the loose insert found in Appendix 12.

In the model the fish are fed once every 24 hours and the weight increase is assumed to occur discretely every 24 hours. This is achieved by the use of an information sub-routine (appendix 8). During experimental series I, single daily feeds resulted in large diurnal variations in ammonia concentration. These were potentially hazardous to the fish and more

frequent feeding was adopted during the second experimental series. This latter feeding pattern could have been approximated in the model by a subroutine, but because of its complexity this was not considered desirable. In addition, it was thought that since an aim of the model was to increase understanding of the behaviour of the system, information obtained from the model would be more clearly understood if there was only a single input of food each day.

Whilst it was recognised that growth is not discrete but probably nearer to being continuous, no data on patterns of growth over 24 hours were available. The effect on the model will be small, resulting in discrete increases in the values for oxygen removal through fish respiration since this is calculated from the weight of fish.

Food conversion efficiency is described in the model by a constant. In practice, it is unlikely to remain constant, with its value influenced by variations in ration size, fish size, temperature, oxygen concentration and food deprivation. However, insufficient information was available to incorporate this in the model. If the pre-feed ammonia concentration predicted by the model is potentially hazardous, the daily food ration is withheld. The resulting loss in weight is represented in the model by the rate WDR. The rate is calculated as a percentage (0.5 per cent) of the weight of fish, subtracted whenever a feed is missed. The percentage value is based on the results of the experimental programme (3.3.2.2).

The decision whether or not to feed is simulated by consideration of the ammonia concentration. If the concentration of ammonia in the tank is greater than or equal to 1 mg/l when food is about to be given, no food is given and WDR operates (see appendix 9). As discussed earlier (2.3.2) only the un-ionized ammonia fraction is toxic. At 25°C (the temperature at which the model is based) the percentage of total ammonia that is in the un-ionized form varies from 0.566 at pH 7.0, to 5.380 per cent at pH 8.0. Therefore, at 25°C, the 1 mg/l total ammonia threshold is equivalent to 0.0125 mg/l un-ionized ammonia at pH 7.4, 0.006 mg/l at pH 7.0, and 0.054 mg/l at pH 8.0. Given that pH is not specified, and in the reference system varies between 6.7 and 8.4 (3.2.5) this was thought to be a representative value. Should the ammonia concentration in the model ever reach 20 mg/l

then the model has a catastrophe state where the fish are instantly "killed".

This is a simple growth model. Attempts have been made to produce more rigorous equations for predicting the growth of carp, derived from the data of Huisman by Hambrey (1980). However, as Hambrey (pers. comm.) has pointed out, since the data of Huisman are very limited, and no other data are available in the literature, the equations he derived are unsatisfactory in many respects. Huisman (pers. comm.) agreed with Hambrey's conclusions and supported the use of the simple growth model described here.

#### 4.4 Sub-Model .SIMPLE

The structure of .SIMPLE is common to both the oxygen and ammonia sub-models, and represents the circulation of water in the system. It comprises two levels (L1 and L2) representing the filter and fish tank respectively. These are linked by a material flow (water) from one to the other (R1 and R2). The rate of flow is determined in both cases by the circulation rate (the constant 'CR'). Although the levels of oxygen and ammonia in the tanks and filters are expressed in terms of their total weights, to calculate the rates of flow (R1 and R2) between the levels L1 and L2, it was necessary to use concentrations. Thus:

$$\text{Rate 1} = \frac{\text{Total quantity (of ammonia or oxygen) in level 2}}{\text{Volume of level 2}} \times \text{CR}$$

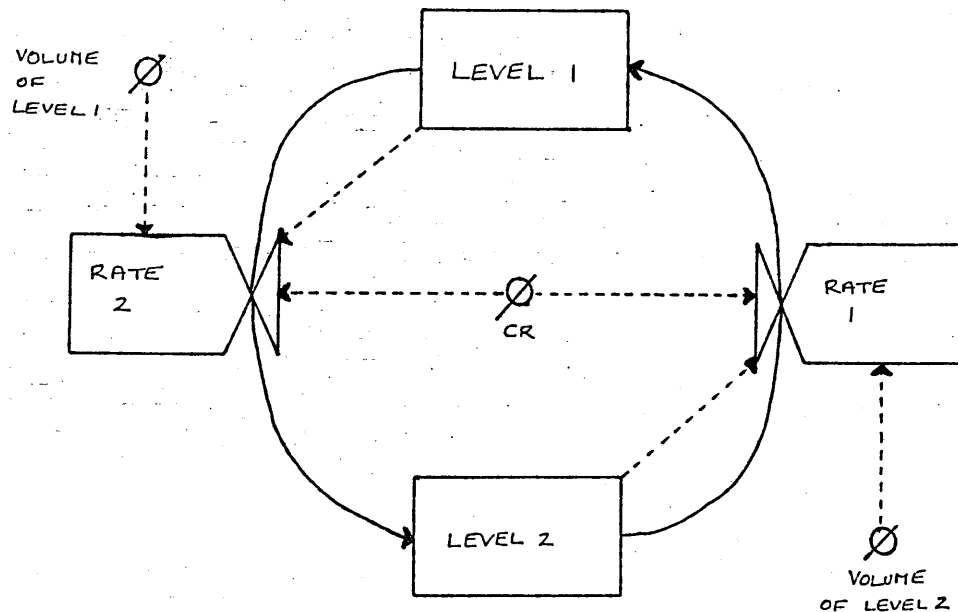
$$\text{Rate 2} = \frac{\text{Total quantity in Level 1}}{\text{Volume of level 1}} \times \text{CR}$$

.SIMPLE is represented diagrammatically in Fig. 4.3.

A key to this and other System Dynamics diagrams is presented for convenience on the card found in Appendix 12.

Three properties of .SIMPLE were examined in detail, the effect of changing the value of the time interval (DT) at which the equations are solved, the

Figure 4.3 System Dynamics network of .SIMPLE



effect of changing the initial values attributed to levels 1 and 2 and the effect of changing the circulation rate. The sub-model .SIMPLE was run on the computer with the tests based on the levels and flow of ammonia. The results of these tests can be summarised:

1. Values of DT larger than 0.15 hours gave inconsistent results with the development of unstable oscillations in the values of levels 1 and 2. The value of DT was linked to circulation rate ; if the rate of circulation increased, the size of DT needed to be reduced accordingly. It was concluded that with a flow of 900 l/hr, a DT value of 0.125 hours was satisfactory.
2. Changing the initial values given to levels 1 and 2 without any input or output simply changed the length of time over which .SIMPLE needed to run before the levels equilibrated. The time taken to reach equilibrium also depended on the circulation rate.

#### 4.5 Ammonia Sub-model (.AMMO)

In the ammonia sub-model, two levels of ammonia are considered, that in the filter (AF) and that in the fish tank (AT). Ammonia is taken from the fish tank to the filter and vice versa by the circulation of water. This basic structure with no input of ammonia into, or loss of ammonia from, the system has been described under .SIMPLE (4.4). In addition to this basic circulation .AMMO also models both the input of ammonia from the fish following feeding and the removal of ammonia by nitrification.

##### 4.5.1 Ammonia Production

The rate at which ammonia is produced and enters a system is determined by the amount of food fed to the fish, and by the fish themselves. One of the difficulties encountered in building the model was finding an adequate relationship to describe ammonia production. Experimentation to determine ammonia production is difficult, since there is a negative feedback process involved, where the rate of excretion of ammonia by the fish decreases as the concentration in the water increases, and this takes effect at very low concentration. Excretion is therefore not often measured but calculated by difference.

There are a few studies in which excretory rates have been determined (Solomon and Brafield, 1972, Elliot 1976) but none of them have been conducted on carp, and they are therefore not directly applicable. Furakawa and Ogasawara (1955), in the course of investigating the digestibility of protein in carp diets, measured the rate of nitrogenous excretion. They found that under normal conditions, the main excretory product was  $\text{NH}_3\text{-N}$  and for 20-30g carp at 16-25°C,  $\text{NH}_3\text{-N}$  excretion was 10-20 mg/110g per 24 hours. Under stress conditions they found an increase in the Urea-Nitrogen excreted. Other workers have confirmed that a relationship exists between ammonia, urea and stress (Burrow, 1964; Olson and Fromm 1971 and Brett and Zala, 1975).

Saeki (1958), citing Baldwin (1949), states that the nitrogen compounds excreted from fish contain 25-50 per cent of ammonia, and the rest can be divided into urea, creatine, and amino acids. Collecting several workers'

results together (including Furakawa and Ogasawara, 1955) Saeki generalises that, except for thin or flat fish, ammonia excretion at 20-25°C is 25 mg per day per 100g of fish weight, and the total amount of nitrogen is 50 mg per 100 g.

Kaushik (1980), working at 16-18°C with carp of 350 g, examined the excretion of nitrogenous wastes in relation to four different feeding strategies; under fast, a fixed ration fed at a single meal, one meal fed to satiation, and two meals fed to satiation. He found that the number of meals did not affect total nitrogenous excretion, and that the maximum excretion with a fixed meal occurred six hours after onset of feeding. Under fast, ammonia excretion was found to be 51.8 mg NH<sub>3</sub>-N/kg/day. Excretion was shown to be related to nitrogen intake, according to the relationship:

$$\log Y = 1.73 + 0.62X$$

where Y = nitrogenous excretion  
(mg N/Kg Body wt/day)

X = nitrogen intake  
(g/Kg Body wt/day)

Hambrey (1980) found, experimentally, that:

$$\text{NH}_3(\text{mg/Kg/Min}) = 0.169 + 0.6887 \text{ SGR}$$

where SGR = specific growth rate (expressed as percentage weight per day).

There are a number of difficulties involved in using any of these studies in the model to simulate ammonia production. As Elliott (1976) showed, excretion varies with temperature. Since the model is based on a temperature of 25°C, simulating laboratory conditions, the work of Kaushik is not directly applicable. Similar work over a range of temperatures is required, in order to determine the influence of temperature. Furakawa and Ogasawara (1955) did not relate their rate of ammonia production to temperature or ration level. Saeki, in his comparison of various work on nitrogenous excretion, decided that a value of twice the Furakawa and



Ogasawara lowest value was representative, and asserted that ammonia was 50 per cent of the total nitrogenous excretion. Most workers, however, have found that in non-stressed fish, ammonia forms 80 - 90 per cent of the nitrogen in the excretory products (Solomon and Brafield, 1972; Brett and Zala, 1975; Elliott 1976).

Furakawa and Ogasawara (1955) found ammonia to be the dominant form of excretory product, with urea varying from trace levels to 23 per cent under non-stress conditions, and to 42 per cent under stress conditions. Clearly there is a great deal of disagreement in these laboratory measurements of fish excretory rates.

Hambrey's relationship depends on the use of specific growth rate and therefore can only be used retrospectively. Since none of the other workers report on the specific growth rate of their fish, this equation cannot be compared.

If an example is taken from the author's laboratory records however, and the different equations are used to predict ammonia production, a comparison can be made.

From the laboratory records

Number of fish	=	49
Initial weight of fish	=	1178g
Weight of fish after		
77 days	=	3697g
Specific growth rate	=	1.48% growth per day
Feeding rate	=	1.76% body weight/day
Food contains 35% protein.		
If it is assumed that 16% of protein is nitrogen, then		
100g of food	=	5.6 g nitrogen.

Author Total excretion predicted for  
24 hr period for whole tank

Kaushik 975 mg NH<sub>3</sub>/day

Furakawa and Ogasawara 409 - 818 mg NH<sub>3</sub>/day

Saeki 924 -1848 mg NH<sub>3</sub>/day

Hambrey 6324 mg NH<sub>3</sub>/day

The calculations for these are presented in Appendix 10

In the literature another type of relationship is described, where ammonia produced is expressed as a percentage of the food fed, e.g:

Speece (1973) 2.6 - 3.2%

Liao and Mayo (1974) 3.1 - 40.4%

Westers and Pratt  
(1977) 2.6 - 3.4%

These data were obtained by measuring the increase in the concentration of ammonia from the head to the tail of raceways, culturing many different species of fish, at different temperatures, and relating this to feed. Liao (1970) produced a relationship:

Ammonia production rate (APR) =  $xF$ , where  $F$  = food fed (mg)  
(mg NH<sub>3</sub>-N) and  $x \in (0.0067, 0.179)$

From the range, Liao (pers. comm.) recommended that for carp culture, a value of 0.03 would be a good approximation.

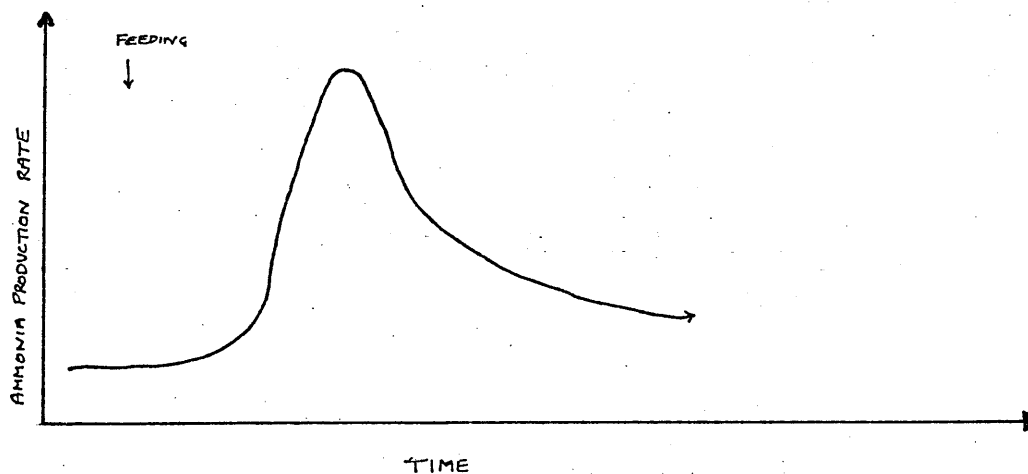
If this value is used in the same example as the relationships of Hambrey, Kaushik, Furakawa and Ogasawara, and Saeki, then:

Food given to 3697 g of fish = 65.06 g  
then APR = 0.03 x 65060  
= 1952mg

This compares well with the value derived from Saeki for total nitrogenous excretion but is higher than Kaushik, and Furakawa and Ogasawara and lower than Hambrey. In view of the difficulties involved in producing a precise relationship in .AMMO, Liao's relationship was used. This was partly because the values it produces lie within the range predicted by other workers, partly because of the simplicity in building this into the model, and partly because its level of accuracy based on field data is comparable to other relationships used in the model.

As described earlier (5.3) in the model the fish are fed once a day. This means that the ammonia enters the system in one load and is not spread out over the day. However it is now generally accepted (Rosenthal, 1980) that after feeding ammonia production takes the form of Fig. 4.4.

Figure 4.4 Ammonia production following feeding (After Brett and Zala, 1975)

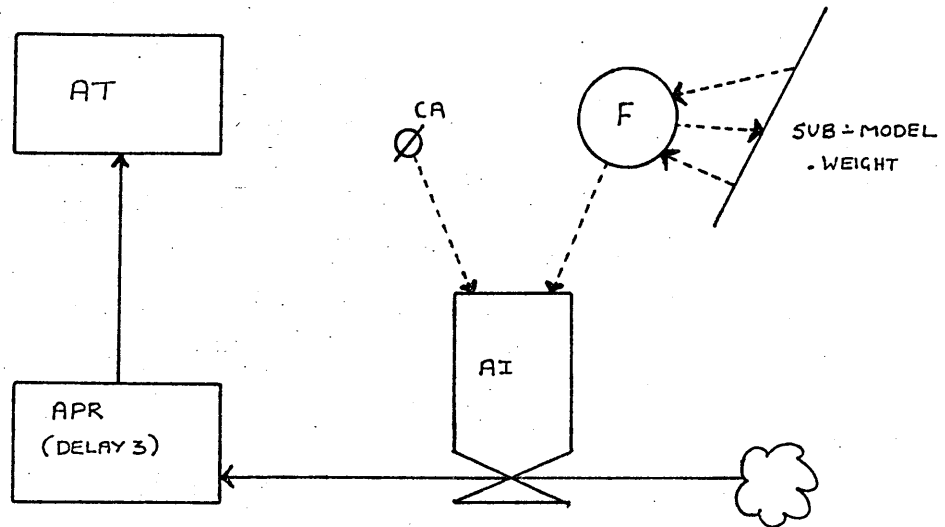


In order to create an input of ammonia in the model which corresponds to this, a delay function was added. After testing, DELAY 3 with a delay period of 6 hours appeared closest to the curve produced by Brett and Zala

(1975) and agrees with the results of Kaushik (1980).

The production of ammonia can be shown diagrammatically (Fig. 4.5)

Figure 4.5 System Dynamics network diagram describing ammonia production



#### 4.5.2 Ammonia Removal

The reference system employs a single filter for the removal of ammonia and organic wastes (3.2). The ammonia is removed principally by chemoautotrophic bacteria of the genus Nitrosomonas and Nitrobacter, which oxidise the ammonia to nitrite and then to nitrate respectively (Nitrification). In the model the rate of ammonia removal (ARR) is determined by the rate of nitrification (AN). Organic wastes (OROG) are removed by the action of heterotrophic bacteria. The removal of both ammonia and organic wastes within the same filter results in a number of interactions. The principal effect of this concerns the availability of oxygen for nitrification. Because of the influence of organic wastes on ammonia removal this aspect is included here.

##### 4.5.2.1 Nitrification

If there are no limiting variables, the rates of oxidation of ammonia and nitrite are proportional to the rates of growth of the bacteria Nitrosomonas and Nitrobacter respectively (Downing and Knowles, 1970).

Therefore it is important in modelling nitrification to understand the phenomenon of bacterial growth and those factors affecting growth.

In 1942 Monod successfully applied Michaelis-Menten enzyme-substrate kinetics to bacteria growth. He established that the relationship between the specific rate of growth of bacteria and their maximum specific growth rate was similar to the hyperbolic relationship describing the rate of substrate removal by an enzyme to its maximum rate of removal. Therefore, bacterial growth ( $\mu$ ) can be described by the equation:

$$\mu = \mu_{\max} \cdot \left( \frac{S}{K_s + S} \right)$$

and  $\mu = \frac{1}{x} \cdot \frac{dx}{dt}$

where  $\mu$  = specific bacterial growth rate  
 $\mu$  = increase in cell mass per unit cell  
 and  $\mu_{\max}$  = max specific growth rate  
 $S$  = substrate concentration, mg/l  
 $K_s$  = saturation constant  
 $x$  = bacterial concentration, mg/l

Thus by substitution:

$$\mu = \frac{1}{x} \cdot \frac{dx}{dt} = \mu_{\max} \cdot \left( \frac{S}{K_s + S} \right)$$

or  $\frac{dx}{dt} = \mu_{\max} \cdot x \cdot \left( \frac{S}{K_s + S} \right)$

which expresses the growth rate of bacteria ( $dx/dt$ ) in terms of the concentration of substrate. The saturation constant is important, since when  $S \ll K_s$ , the rate of bacterial growth is directly proportional to the substrate concentration, and  $dx/dt \propto xS$ , which is a first order differential equation. When  $S \gg K_s$ , the rate of bacterial growth becomes independent of the substrate concentration, and  $dx/dt$  becomes a zero order differential equation. The saturation constant varies with temperature. For ammonia at 25°C  $K_s = 3.5$  mgN/l (Ulken, 1963). Since the rate of nitrification is proportional to bacterial growth, the rate of nitrification at 25°C is dependent on the substrate concentration if the

concentration is significantly less than 3.5 mgN/l.

An equation to describe this relationship between nitrification and bacterial growth can be produced by considering the rate of substrate removal and the yield of cells from the substrate:

$$-\frac{dS}{dt} = \frac{\text{max. } x}{y} \cdot \frac{(S)}{K_s+S}$$

where  $y$  = yield coefficient  
=  $\frac{-dx}{dt}$   
=  $\frac{\text{wt of bacteria formed}}{\text{wt of substrate used}}$

Downing and workers (Knowles, et al, 1965; and Downing 1968), applied these Monod-type equations to describe nitrification in activated sludge plants. Jones and Paskins (1980) have developed this further with a mathematical model for the removal of a substrate in activated sludge, which included both a Monod term for substrate utilisation by growing bacteria and a Michaelis-Menten term for the consumption of a substrate by non-viable bacteria. There are, however, many difficulties in the estimation of nitrifier concentration for determination of specific growth rate, yield coefficient and the saturation constant (Sharma and Ahlert, 1977).

If it is assumed that the reference filter is in equilibrium and that there is no change in growth, then  $(\text{max. } x/y)$  can be assumed constant.

Therefore,

$$\frac{-dS}{dt} = K \cdot \left( \frac{S}{K_s+S} \right) \text{ where } K = \text{a constant}$$

These models were based on studies with activated sludge. In nitrification in filters mass-transfer limitations can result (Sharma and Ahlert, 1977). It will be recalled that at low substrate concentrations ( $S \ll K_s$ ), the Monod equation reduces to a first order expression. In the reference system the concentration of ammonia is usually below 3.5 mgN/l, the value of the saturation constant, and during the experimental programme ammonia

removal was found to be dependent on influent concentration (see Fig. 3.11). First order rate equations were therefore sought to model nitrification.

Before considering these equations it is necessary to examine those factors which may influence the rate of nitrification. The main factors which affect the nitrification process in freshwater are temperature, pH, dissolved oxygen concentration, nitrifying bacterial population, and retention time (see 3.3.4).

Nitrification reactions follow the Van't Hoff-Arrhenius law up to 30°C (Metcalf and Eddy, 1973). Therefore, if temperature is increased, the rate of nitrification increases. Equations to describe this relationship are discussed below. Optimum temperatures reported range from 28°C to 42°C (Painter, 1970). pH appears to have minimal effect over a wide range of values, but above and below certain values the rate of nitrification drops sharply (Kramer, Chin and Mayo, Inc., 1972). The values at which inhibition is reported to occur vary widely but it is generally held that optimum levels occur within pH 7-9 (Painter, 1970). During the second experimental series there was little variation in pH (Table 3.18) and it remained within the optimum range indicated above. pH was therefore considered as a constant in the model.

If the dissolved oxygen concentration falls below 2 mg/l, nitrification is retarded, but it is maintained that for efficient nitrification, the dissolved oxygen concentration must be above 4 mg/l (Kramer, Chin and Mayo 1972; and Knowles, Downing and Barrett, 1965). The significance of this is discussed later.

When a filter is first brought into operation, the nitrifying bacterial population is not yet established. Since the growth of these autotrophic bacteria is slow, there is a period during which nitrification is limited by the bacterial population. This slow growth rate is important, since if there are sudden rises in the amount of ammonia to be oxidised, then there is a lag before the filter fully responds. Similarly, if there is a period when no ammonia is present, some bacteria may die, and again there will be a lag before the filter fully responds when ammonia is reintroduced (see

#### 3.2.6.4.)

The efficiency of nitrification has been shown to be related to the time that the water containing the ammonia is retained within the filter (retention time,  $T_M$ ). As retention time increases so the amount of ammonia nitrified increases (Liao and Mayo 1972; Haug and McCarty, 1972). If, however, there is no diffusion of oxygen into the water whilst it is retained in the filter (as in a submerged filter), then the benefit of increased retention time will be reduced through lack of sufficient oxygen, and anaerobic zones may develop (see 3.3.4).

The literature contains a number of references which provide first order relationships describing nitrification yet there are few references where the relationship described is adequate. Haug and McCarty (1972) produced a widely quoted description of nitrification, formulating the relationship:

$$\frac{ds}{dt} = (0.11.T - 0.20) \frac{(S)}{10} 1.2$$

where  $T$  = temperature ( $^{\circ}\text{C}$ ) and  $S$  = influent ammonia concentration (mg/l)

This is a first order equation, similar to the reduced Monod expression, but its use is restricted, since it is not related in any way to the size of the bacterial population, and therefore cannot be related to the filter employed in any system. This equation is specific to the reaction chamber used in their experiments. This cannot be generalised (Kramer, Chin and Mayo, Inc., 1972), since their research was conducted using a synthetic waste containing no organic matter, an oxygen concentration 300 per cent higher than normal saturation levels, and an influent ammonia concentration of 10 mg/l, higher than is generally found in recirculating systems (Wheaton, 1977), and larger than the Michaelis constant. A zero order equation might therefore have been expected at these concentrations (Painter, 1980, pers. comm.).

Speece (1973) realised the limitations of Haug and McCarty's relationship and, using very limited experimental data of his own, produced a new relationship relating the nitrification capacity per specific filter area with temperature. Since this relationship is based upon only one



measurement of nitrification at a given temperature and filter area, its application to this present study is somewhat dubious.

Srna and Baggaley (1975), working on a seawater submerged filter, produced an equation of the form:

$$Kt = \ln \left( \frac{C_0}{C_0 - X} \right)$$

in which

$C_0$  = initial concentration of nutrient substrate

$X$  = amount of nutrient substrate reacted after time  $t$

$K$  = first order rate constant

They produced a range of first order rate constants according to pH for the oxidation of both ammonia to nitrite and nitrite to nitrate:

$$K_{\text{NH}_3} = \text{pH} (0.230) - 1.60 \quad (\text{pH range} = 7.4 - 7.8)$$

$$K_{\text{NO}_2^-} = \text{pH} (-0.053) + 0.521 \quad (\text{pH range} = 6.6 - 8.6)$$

In conducting their experiments Srna and Baggaley used a synthetic waste comprising ammonium chloride and sodium nitrite and supplied no organic matter to the filter system. The rate constants produced were found to be up to an order of magnitude higher than those reported by Knowles et al (1965) and Carlucci and Strickland (1968). Their conclusions are valid only for higher nutrient concentrations; concentrations less than 3.5 mg/l as N were not studied. In view of this, and that this work was conducted in seawater and not freshwater, the relationships for nitrification were not used to simulate nitrification in the model.

Liao and Mayo (1972) also produced a first order equation describing nitrification, relating ammonia removal to retention time for a variety of loading rates.

$$N_{AR} = 0.96 A_L t_m$$

where  $N_{AR}$  = ammonia removal rate of filter at 10-15°C  
 (KgNH<sub>4</sub> - N/m<sup>2</sup>day)  
 (m<sup>2</sup> is square metre of specific medium  
 surface area)

$A_L$  = ammonia loading rate (KgNH<sub>4</sub>-N/m<sup>2</sup>day)

$t_m$  = media retention time (hrs).

In order to balance this equation it is assumed that the constant 0.96 has dimensions of reciprocal time.

Since  $N_{AR}$  and  $A_L$  are the same units, this equation can be modified so that if  $A_L$  is expressed in mg NH<sub>3</sub>-N, then  $N_{AR}$  is expressed in mg NH<sub>3</sub>-N. The equation has a number of limitations, however, which include:

1. Media retention time between 12.6 and 27.6 mins.
2. Hydraulic loading less than 100 l/min/m<sup>2</sup>.
3. Ammonia concentration about 1 mgNH<sub>4</sub>-N/l or less.
4. Ammonia loading less than 4.07 kgNH<sub>4</sub>-N/m<sup>2</sup>/hr.
5. Water temperature within the range 10°C to 15°C.
6. No oxygen limitation.

Accepting these limitations, this relationship has been used in the model to simulate nitrification. It is possible to extend the temperature range of the equation by modifying the rate constant 0.96 by the use of a relationship between the nitrification constant and temperature. Such a relationship was produced by Haug and McCarty (1972) and by Downing (1968), and Downing and Knowles (1970). For a discussion of the limitations of these various relationships see Wheaton (1977). In .AMMO, the relationship developed by Haug and McCarty has been used so that the relationship becomes:

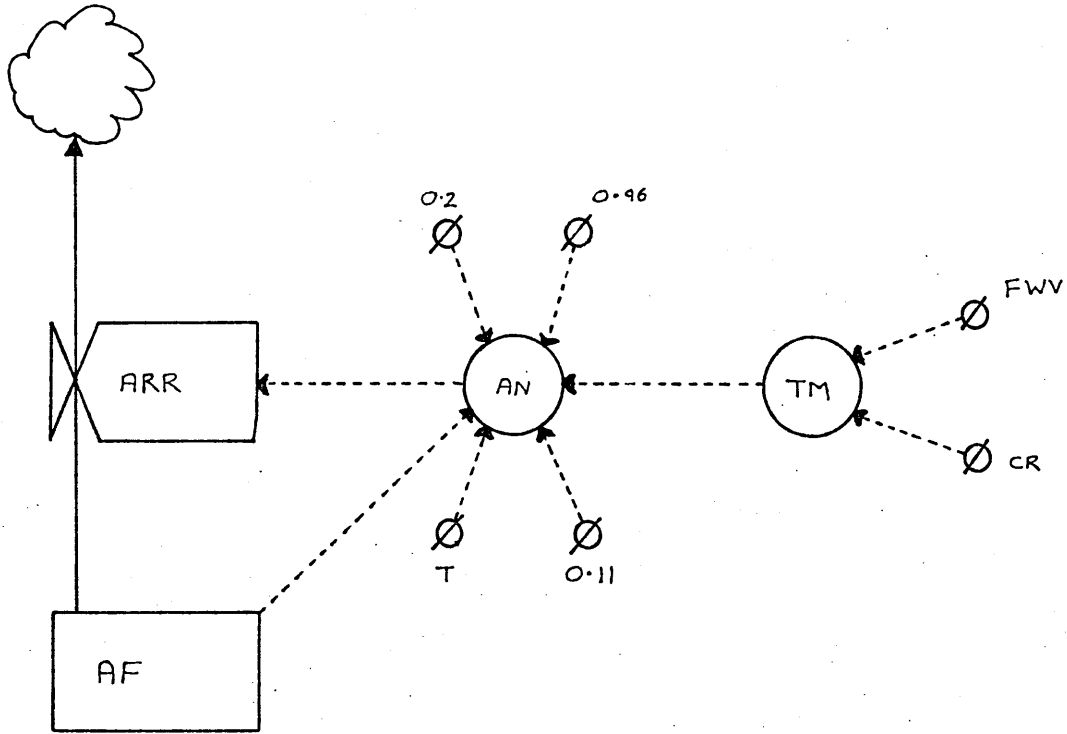
$$N_{AR} = (0.11T-0.2) \times 0.96 \times t_m \times A_L$$

or, in the terminology of .AMMO

$$AN = (0.11T - 0.2) \times 0.96 \times TM \times AF$$

This is represented diagrammatically (Fig. 4.6)

Figure 4.6 System Dynamics network diagram of nitrification



As indicated earlier the rate of nitrification is affected by the availability of oxygen. The oxygen available for nitrification in the filter will depend on a number of factors. Firstly it will depend on the oxygen contained in the water and the flow rate. This is discussed further under .OSIM (4.6). It will also depend on the amount of oxygen used in the filter by the heterotrophic bacteria breaking down the organic matter. The saturation constant ( $K_s$ ) of oxygen for the heterotrophic bacteria is lower than that of the autotrophic nitrifying bacteria. Consequently heterotrophic bacteria are favoured at low oxygen concentrations. It is therefore important to examine the production of organic wastes in the system and how this affects nitrification.

#### 4.5.2.2 The production of organic wastes

The organic wastes in the reference system arise from uneaten food and faecal wastes. They are almost entirely organic carbon based, although there may be some proteinaceous material (Spotte, 1970).

Equations have been developed by a number of workers to describe the relationship between suspended solids (SS), biological oxygen demand (BOD) and chemical oxygen demand (COD) with the feeding rate. These relationships are summarised below:

Liao and Mayo (1972) SS = 0.52F Kg SS/100 kg fish/day at 10-14°C  
(salmonids) BOD = 0.60F Kg BOD/100 kg fish/day at 10-14°C  
COD = 1.89F Kg COD/100 kg fish/day at 10-14°C

where F = Feeding rate (Kg food/100 kg fish/day)

Murphy and Lipper BOD = 0.25F Kg/day

(1970) where F = Kg food/day (- 2% body wt. fed).

Speece (1973) SS = 0.4F

(catfish) where SS = (Dry faecal mater suspended solids)

and F = Kg of feed

Willoughby, Larsen BOD (1b) = 0.34F

and Bowen (1972) SS (1b) = 0.39F

where F = lb feed

Speece (1973) also found that 1 kg of dry faecal material suspended solids exerts approximately 1 kg of ultimate BOD. Accordingly, for each kilogram of feed 0.4 kg of ultimate BOD was produced.

The production of faecal material by carp at 25°C was investigated by the author (3.3.3.1). The production of dry faecal material suspended solids was found to vary in proportion to the amount of feed according to the relationship

$$y = 11.91 + 0.15x$$

where y = weight of faeces (g)

and x = weight of food (g)

In view of the uncertainty of the relationship (see 3.3.3.1) the model uses the more approximate relationship of:

$$SS = 0.207F$$

where 0.207 = average proportion of food lost as faeces  
(taken from Table 3.24)

and F = weight of feed (g)

Together with Speece's observation that 1 kg of dry faecal material suspended solids exerts approximately 1 kg of ultimate BOD, this approximates to:

$$\begin{aligned} BOD_u &= 0.2F \\ (\text{Kg } O_2) &= (\text{kg food}) \end{aligned}$$

The production of organic wastes over twenty-four hours will depend on the gastric evacuation rate (GER) of the fish. Three types of GER have been described (Fischer, 1979), according to three patterns of feeding; continuous feeders, discontinuous periodic feeders, and discontinuous aperiodic feeders. Carp can be classified as discontinuous periodic feeders (Fischer, 1979). As there is no detailed information available on the production of faeces over twenty-four hours on which to model the production of organic wastes, the model assumes a continuous production of faeces, based on the weight of food fed per twenty-four hours.

In the model the oxygen that will be consumed by the organic wastes entering the filter (OROG) is subtracted from the oxygen before it enters the filter (OFIR). The oxygen that remains after this subtraction is made is then assumed available for nitrification. This has a limitation in that it does not allow for build up of organic wastes in the filter, but assumes that organic oxidation takes place instantly and completely. This does not seem realistic, except for equilibrium conditions when the rate of removal equals the rate of production exactly. This rarely occurs in the reference system, or in any other recirculating system where some sludge wastage occurs.

At lower concentrations of dissolved oxygen, nitrification becomes limited. Table 4.1 presents some of the reported limiting levels of dissolved oxygen.

Table 4.1 After Sharma and Ahlert, 1977

Dissolved Oxygen Concen. mg/l	Observation	Reference
Below 2	Limiting for <u>Nitrosomonas</u> growth	Knowles, Downing and Barrett (1965)
Below 4	Limiting for <u>Nitrobacter</u> growth	Barrett (1965)
Below 1-1.5 0.1	Limiting for growth Nitrification occurs	Carlucci and McNally (1969)
0.5-0.7	Critical	Downing, Boon and Bagley (1962: cited in Huang, 1973)
1.0	Limiting	Metcalf and Eddy (1973)
0.6-0.7	Limiting	Forster (1974)

From Table 4.1 it would seem that below approximately 4 mg/l nitrification becomes inhibited, and below 0.7 mg/l nitrification effectively stops. In order to allow this cessation of ammonia removal to be included in the model, the equation for nitrification (AN) includes a multiplier (AL). This is a 'clip' function, which examines the level of dissolved oxygen in the filter. If the level is below 0.7 mg/l, then no nitrification is assumed to occur, and AL = zero. If there is greater than 0.7 mg O<sub>2</sub>/l in the filter, then AL = 1.

Thus:

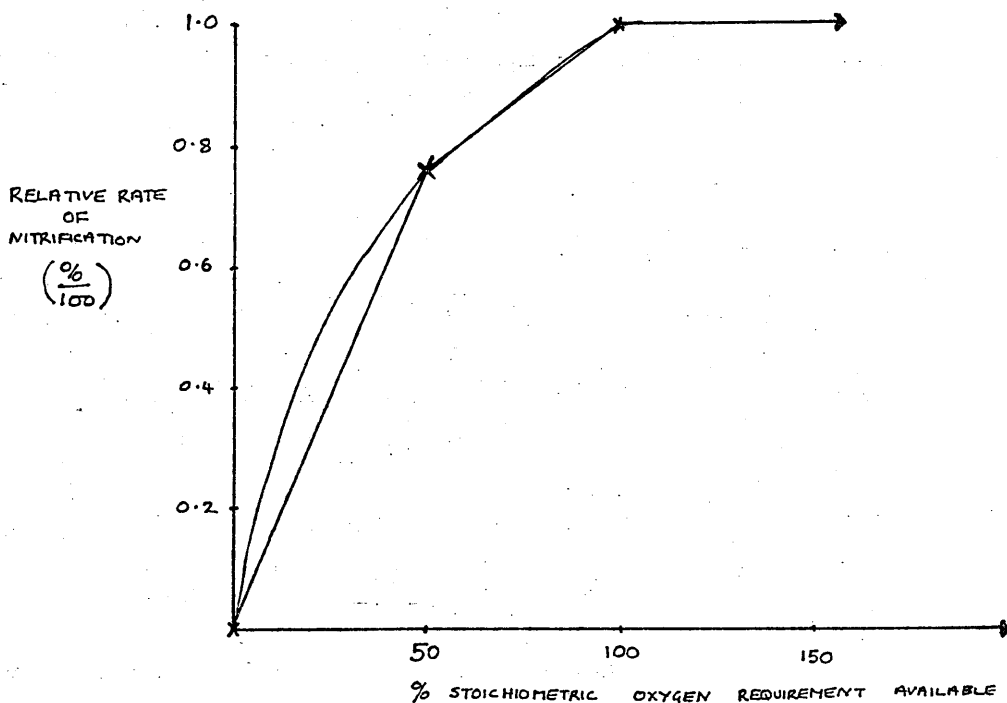
$$AN = (0.11T - 0.2) * 0.96 * TM * AF * AL$$

where AL = CLIP (1, 0, OF, 93.8) (mg O<sub>2</sub>)

Although at concentrations of oxygen above 0.7 mg/l nitrification takes place, it does so at a reduced rate. To prevent inhibition of nitrification

through lack of oxygen the 'stoichiometric' oxygen requirement for nitrification must be met (Haug and McCarty, 1972). That is, every 1 mg of ammonia which is oxidised to nitrate requires 4.57 mg of oxygen. Haug and McCarty (1972) showed that when the stoichiometric requirement was met there was no inhibition of nitrification. As the percentage of the stoichiometric requirements was reduced, so the rate of nitrification was reduced. The relationship produced can be approximated by three points on a graph, (Fig. 4.7).

Figure 4.7 Relative rate of nitrification



Haug and McCarty's stoichiometric oxygen requirement of 4.57 mg O<sub>2</sub> / mg NH<sub>3</sub> is the theoretical oxygen requirement. Experimentally a value of 4.33 mg has been found (Wezernak and Gannon, 1967; Montgomery and Bourne, 1966). This difference is attributed to the production of oxygen during the process of protoplasm synthesis. In the model a value of 4.33 mg has been used. This is done by the inclusion of a multiplier (OM) in the equation defining the rate of nitrification (AN). If there is enough oxygen the value of OM is 1.0 and it has no effect. If the stoichiometric requirements are not met the value of OM drops below 1 and the rate of nitrification is reduced. The value of OM is equivalent to the relative

rate of nitrification in Haug and McCarty's relationship (see Fig. 4.7).

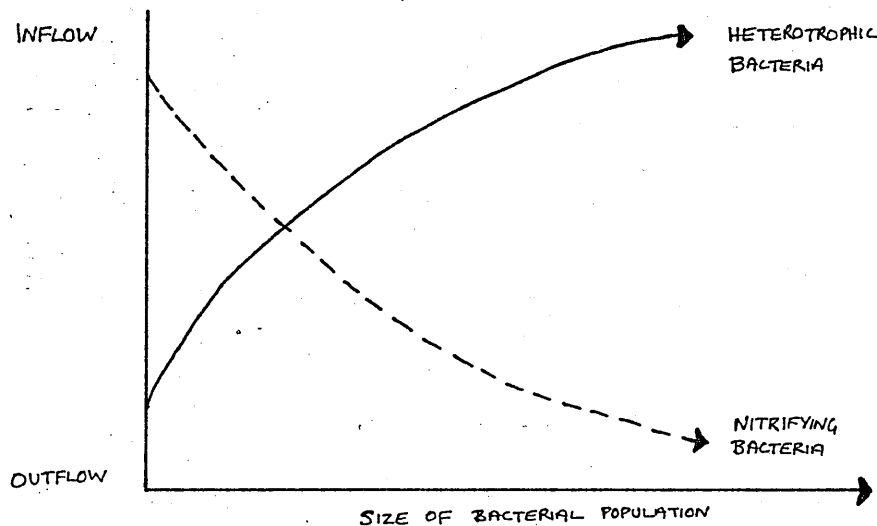
OM is determined by the use of a 'TABHL' function, which given the specification of Haug and McCarty's graph, calculates the value of OM for each measure of oxygen availability. The percentage of stoichiometric oxygen required that is available is determined in the model by dividing the oxygen within the filter (OF) by the ammonia within the filter (AF).

Thus, the equation for nitrification becomes:

$$AN = (0.11T - 0.2) * 0.96 * TM * AF * OM * AL$$

The organic matter that enters the filter is also important, since during its breakdown by heterotrophic bacteria some secondary ammonia is produced (Spotte, 1970). Biological filters receiving an effluent containing a low organic load show a characteristic profile (Kawai, 1965; Spotte, 1970), (Fig. 4.8).

Figure 4.8 Filter profile



With this profile, any secondary ammonia produced by the heterotrophs is available to the nitrifying bacteria deeper in the filter. In the reference system, however, there is a high solids loading, and it can be assumed that there is a uniform concentration of heterotrophic and nitrifying bacteria throughout. This means in practice that it is impossible to achieve from the filter an effluent which is completely free

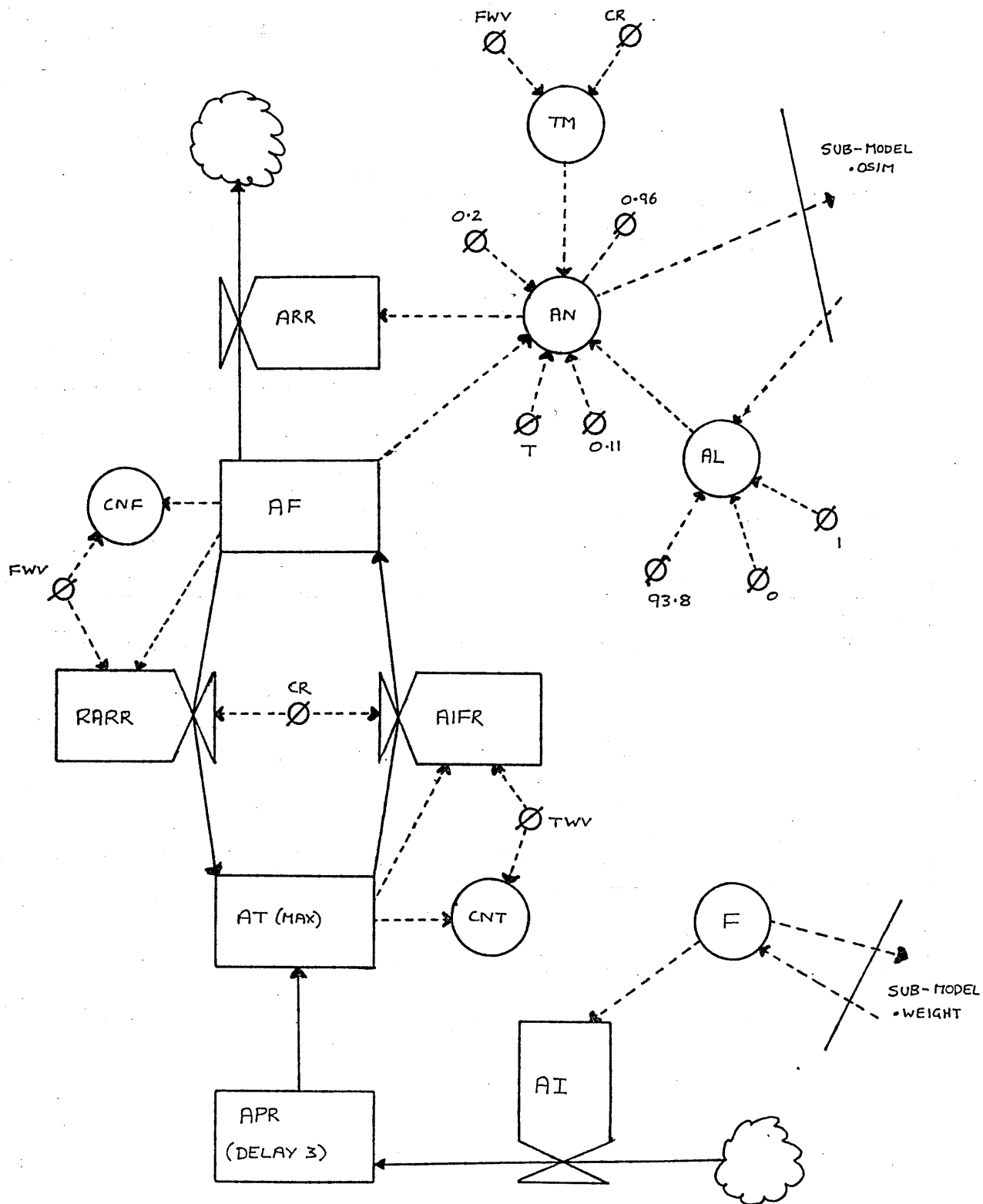


of ammonia. In addition, the development of anaerobic zones within the filter also leads to secondary ammonia production by ammonification.

Experience suggests that in the reference system, the baseline ammonia concentration leaving the filter is 0.2 mg/l. To simulate this, the level equation for the ammonia in the tank uses a 'MAX' function. This says that the ammonia level within the tank is the greater of the values - either 0.2 mg  $\text{NH}_3\text{-N/l}$  or the calculated value of AT.

The System Dynamics network diagram for the complete ammonia sub-model is shown below in Fig. 4.9

Figure 4.9 Complete ammonia sub-model



## 4.6 The oxygen sub-model .OSIM

Like the ammonia sub-model, .OSIM is based upon the framework of .SIMPLE (4.4) with two levels of oxygen considered, that in the fish tank (OT) and that in the filter (OF), joined by a continuous circulation of water containing oxygen. The oxygen levels are modified by:

- a) removal (by fish respiration in the tank and oxidative processes in the filter)
- b) replacement of oxygen (aeration in the tanks).

### 4.6.1 Removal of oxygen

#### 4.6.1.1 Oxygen removed by fish respiration (ORFR)

The oxygen removed from the water by fish depends on a number of factors. These are summarised in section 2.3.1.

As with estimates of ammonia production, two types of relationship describing oxygen consumption by the fish are to be found in the literature.

The first is the type based upon measurements made in the field, often in raceway culture, which looks at the depletion of oxygen along the length of the raceway. Usually this is then related to the feed given to the fish held in the raceway. For example, Willoughby (1968) stated that 100 g of oxygen was required to metabolise one pound of trout pellets by trout in raceway culture, giving : 22g oxygen/100g pellets (temp = 4.5 - 15.5°C). The relationship was later used by Speece (1973) as a basis for his design criteria for a recirculating culture system.

In a study for the development of fish hatchery water treatment systems by Kramer, Chin and Mayo, Inc. (1972), the relationship  $O_c = 0.54F$  was found, where:

F = Feeding rate, g food/100g fish-day

and  $O_c$  = Oxygen consumption, g/100g fish-day

This relationship was established with the following constraints:

1. fish size - 7.5 to 28 cm trout;
2. water temperature 10 - 14.4°C
3. water velocity 0.152 - 0.305 m/S
4. feed rate according to the Buterbaugh and Willoughby formula (1967).

The other types of relationship described in the literature are the more detailed estimates of oxygen removal, and are based on studies of fish respiration carried out by researchers studying fish energetics.

It is generally accepted that the metabolic rate of a fish (R) varies in direct proportion to some power function of weight (W) (Winberg, 1956):

$$R = \alpha W^b$$

where,  $\alpha$  = level of metabolism

b = weight exponent.

The weight exponent, usually takes a value between 0.5 and 1 (Hambrey, 1980). This is generally explained by the theory that metabolic rate is determined not only by the weight of fish but also by its surface area. Since the surface area increases in proportion to weight raised to the power 2/3, it would be expected that the weight exponent b should lie between 2/3 and 1.

In fact for most fish it comes very close to 0.8.

Therefore the equation becomes:

$$R = \alpha W^{0.8}$$

A number of workers have produced similar equations to the above to describe the respiration of carp. These relationships are summarised below

Respiration (R) = ml O<sub>2</sub>/hr/individual, at 20°C

Weight (W) = g

R	= 0.45 W <sup>0.81</sup>	Ivlev, 1939
R	= 0.39 W <sup>0.81</sup>	for all species (in Winberg, 1956)
R	= 0.343 W <sup>0.85</sup>	Winberg, 1956
R	= 0.60 W <sup>0.98</sup>	(carp fry) Winberg and Khartova, 1953
R	= 0.64 W <sup>0.97</sup>	(carp fry) Kamler, 1976
R	= 1.271 W <sup>0.80</sup>	Kamler, 1972
R	= 0.595 W <sup>0.98</sup>	(carp fry) Kamler, 1972
R	= 1.148 W <sup>0.816</sup>	Kausch, 1968
R	= 0.29 W <sup>0.81</sup>	(starving) Huisman, 1974
R	= 0.67 W <sup>0.77</sup>	Huisman, 1974
R	= 35 mg/g/hr	standard oxygen Beamish, 1964

Huisman (1974; 1978), in conducting research into optimal rearing conditions for carp, examined consumption of oxygen of growing and fasting carp of different weights. For fasting carp the relationship between oxygen consumption (T, in ml/hr) and fish weight (W in grams) was described:

$$T = 0.29 W^{0.81} \quad \text{at } 20^{\circ}\text{C}$$

For growing carp the relationship between oxygen consumption and feeding level was established, showing that oxygen consumption increased with increased feeding level, up to what Huisman describes as "the production optimal feeding level". At this level the relationship could be described:

$$T = 0.67 W^{0.77} \quad \text{at } 20^{\circ}\text{C}$$

At "P-0 Levels" the amount of oxygen consumed for each Kg of food was 147 litres, independent of fish weight and water temperature.

Since 224 litres of oxygen weighs 1 mole = 32g, the oxygen consumption can therefore be described:

$$T \text{ (mg O}_2\text{)} = 0.21 F, \text{ where } F = \text{food fed (mg)}$$

Winberg (1956) produced his relationship  $R = 0.343 W^{0.85}$  after considering results produced by many different authors, at different temperatures, by different methods, and using a very wide range in the size of carp considered. Despite this, all these results could be plotted along a straight line, with a correlation coefficient of  $R = 0.997$ , on a graph of weight versus respiration.

Clearly there is a wide range of relationships that could be used in .OSIM.

It is hoped that sensitivity analysis will show how important this relationship is in the model. As a first estimate of the relationship .OSIM uses a relationship whose value lies between the earlier work of Ivlev and Winberg, and the later work of Kamler.

$$R = 0.5 W^{0.81} \text{ ml/hr/ind at } 20^{\circ}\text{c}$$

The units in which this relationship is expressed are not compatible with the rest of the model, and therefore need to be changed.

Firstly, this relationship is based at  $20^{\circ}\text{c}$ , whilst the model works at  $25^{\circ}\text{c}$ . This can be corrected for by using a 'q' factor (Winberg, 1956), based on the 'normal curve':

Thus,

$$R = 0.76 W^{0.81} \text{ ml/hr/ind (g) at } 25^{\circ}\text{c}$$

or in preferred units;

$$R = 1.09 W^{0.81} \text{ mg/hr/ind}$$

where  $W = \text{Weight of individual fish (g)}$

Secondly, in the model only the total weight of fish in the system is considered, whereas this relationship is based on individual fish. An allowance for this is made by taking the total weight of fish in the system ( $W$ ) and dividing this by the number of fish ( $N$ ) to give an average weight ( $AV$ ).  $W$  is expressed in the model in mg, but for use in the above equation it should be expressed in g.

Therefore,

$$Av = (W / 1000) / N$$

This average weight is then used to compute the oxygen consumption for an individual, and this is multiplied by the number of fish to give the total 'average' consumption.

So,

$$ORFR = (1.09 Av^{0.81}) \times N$$

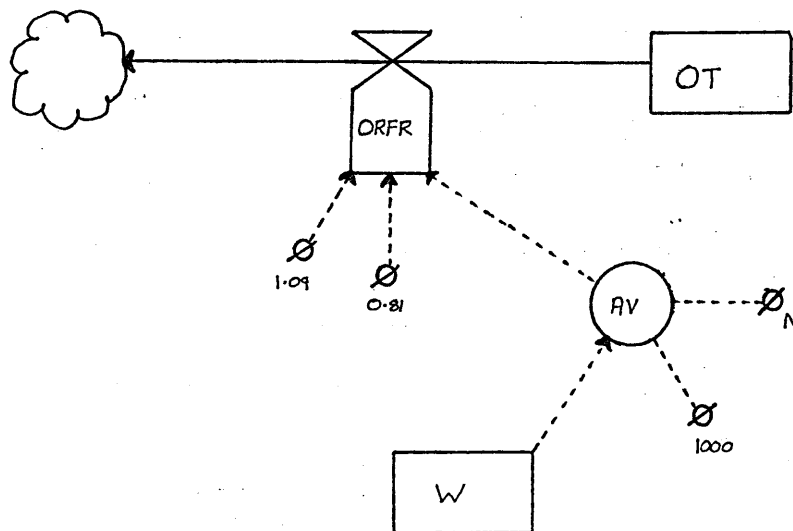
To make this acceptable to the computer the power relationship must be represented by  $A(\text{EXP}(B)\text{LOGN})$ .

So ORFR becomes:

$$ORFR = 1.09 (\text{EXP}[0.81 * \text{LOGN}(AV)]) * N$$

This is represented diagrammatically (Fig. 4.10).

Figure 4.10 System dynamics diagram of oxygen removal by fish respiration



#### 4.6.1.2 Oxygen consumed by oxidative processes

There are two oxidative processes considered in the model, (i) oxygen consumed in nitrification, and (ii) the oxidation of organic wastes.

(i) oxygen consumed through nitrification (ORNA).

For every mg of ammonia that is nitrified (AN) 4.33 mg of oxygen is consumed (see section 4.5.2.2).

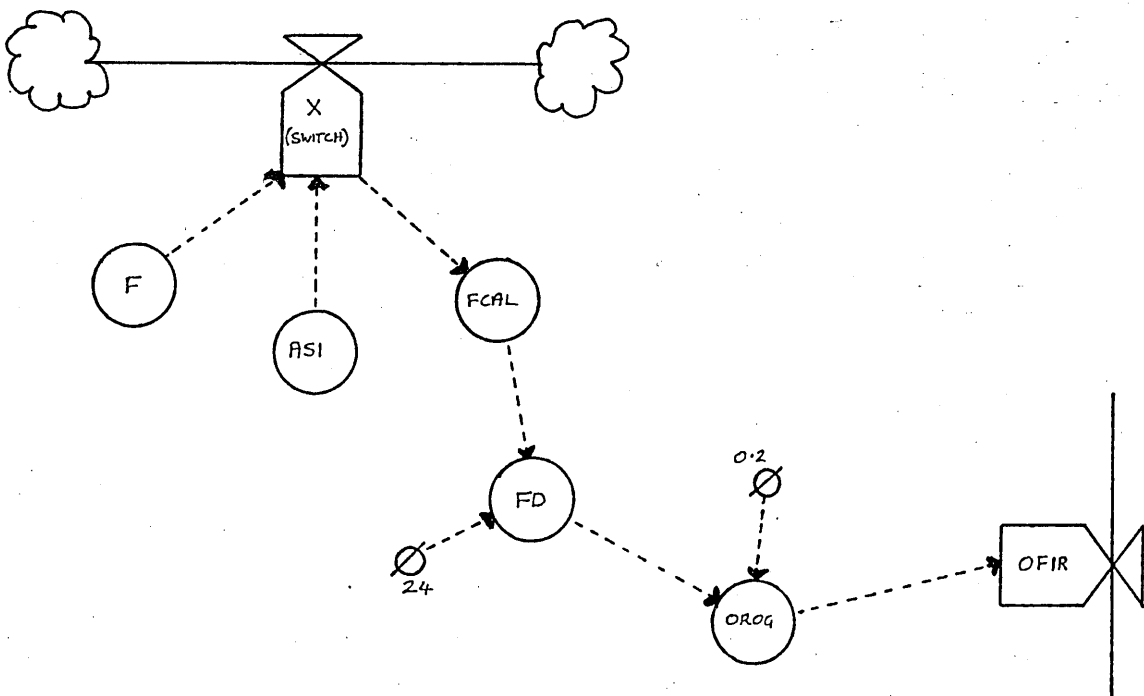
$$\begin{aligned} \text{Therefore ORNA} &= \text{AN} \times \text{ONC} \\ \text{where ONC} &= \text{oxygen consumed in nitrification (a constant)} \\ &= 4.33\text{mg} \end{aligned}$$

(ii) Oxygen consumed during oxidation of organic wastes (OROG).  
As discussed earlier (4.5.2.2) the model assumes that;

$$\begin{aligned} \text{OROG} &= 0.2 \times F \quad \text{where } F = \text{food given per hour in mg.} \\ &(\text{mg O}_2) \end{aligned}$$

It will be recalled that in the model the fish are fed once a day. It was therefore necessary to use two auxiliary variables (FCAL and FD) and a rate (X) to provide the auxiliary variable OROG with the weight of food per hour (Fig. 4.12). A switch is included to ensure that no organic matter is produced if the fish are not fed (see Appendix 11).

Figure 4.11 System Dynamics network diagram of OROG pathway





#### 4.6.2 Addition of Oxygen to the System (OR)

In the reference system oxygen is added by the use of a compressor and porous pipe. The efficiency of aeration depends not only on the aerating system employed, but also on the condition of the water. One of the most important factors is the degree to which the water is already saturated. The gas transfer equation shown below indicates a reduction in the rate of gas transfer as the actual dissolved oxygen in the water increases:

$$dc/dt = K(C_{\text{Sat}} - C_{\text{Act}})$$

where  $dc/dt$  = rate of gas transfer

$K$  = aeration coefficient

$C_{\text{Sat}}$  = Saturation dissolved oxygen

$C_{\text{Act}}$  = Actual level of dissolved oxygen.

from Speece (1980).

For any given water, it is difficult to estimate the aeration coefficient ( $K$ ) and in consequence, the efficiency of any aeration system. Even if the performance of the aeration system is known, the quantity of oxygen entering the water will vary with the variation of the concentration gradient. In a well maintained system it should be possible to maintain the water in the tank at, or near, 100 per cent saturation at all times, although this will depend on stocking density and whether oxygen or air is used.

Therefore, rather than specifying the addition of oxygen to the system, .OSIM predicts the rate of oxygenation necessary to maintain the tank at 100 per cent saturation and then uses this value in calculating the changes in the oxygen levels. There are two advantages in this. Firstly the level of oxygen calculated for the tank will not exceed 100 per cent saturation. (This could happen in the model if there was a constant rate of aeration and at some time this exceeded the rate of oxygen removal), and secondly, it ensures that the level of oxygen in the tank is never negative (if in the model, the rate of depletion exceeded the rate of aeration then this could occur).

The addition of oxygen to the system is calculated by first adding to the system the difference between the theoretical maximum value of oxygen contained in 900 litres of water (Reoxygenation constant ROC) and the actual level of oxygen in the tank. That is:

$$\begin{aligned} \text{at } 25^{\circ}\text{c, } 100\% \text{ saturation} &= 8.25 \text{ mgO}_2/\text{l} \\ \text{therefore 900 litres can contain} &8.25 \times 900 \\ \text{then ROC} &= 7425 \text{ mgO}_2 \\ \text{So, OR} &= \text{ROC} - \text{OT} \end{aligned}$$

However, when .OSIM was run with this relationship, the level of oxygen in the tank became negative. This was because the oxygen removed by the fish respiration (ORFR) was taken from the tank level (OT) after the value for OR was calculated, and therefore, since the oxygen consumption by the fish over the period DT exceeded the level of oxygen that the tank could hold at any one time, the levels became negative.

This was overcome by adjusting the value of OR to include ORFR. i.e.;

$$\text{OR} = (\text{ROC} - \text{OT}) + \text{ORFR}$$

The level of oxygen in the filter is determined by the balance between the rate of oxygen removal by nitrification (ORNA) and by the rate of oxygen entering the filter (OIFR).

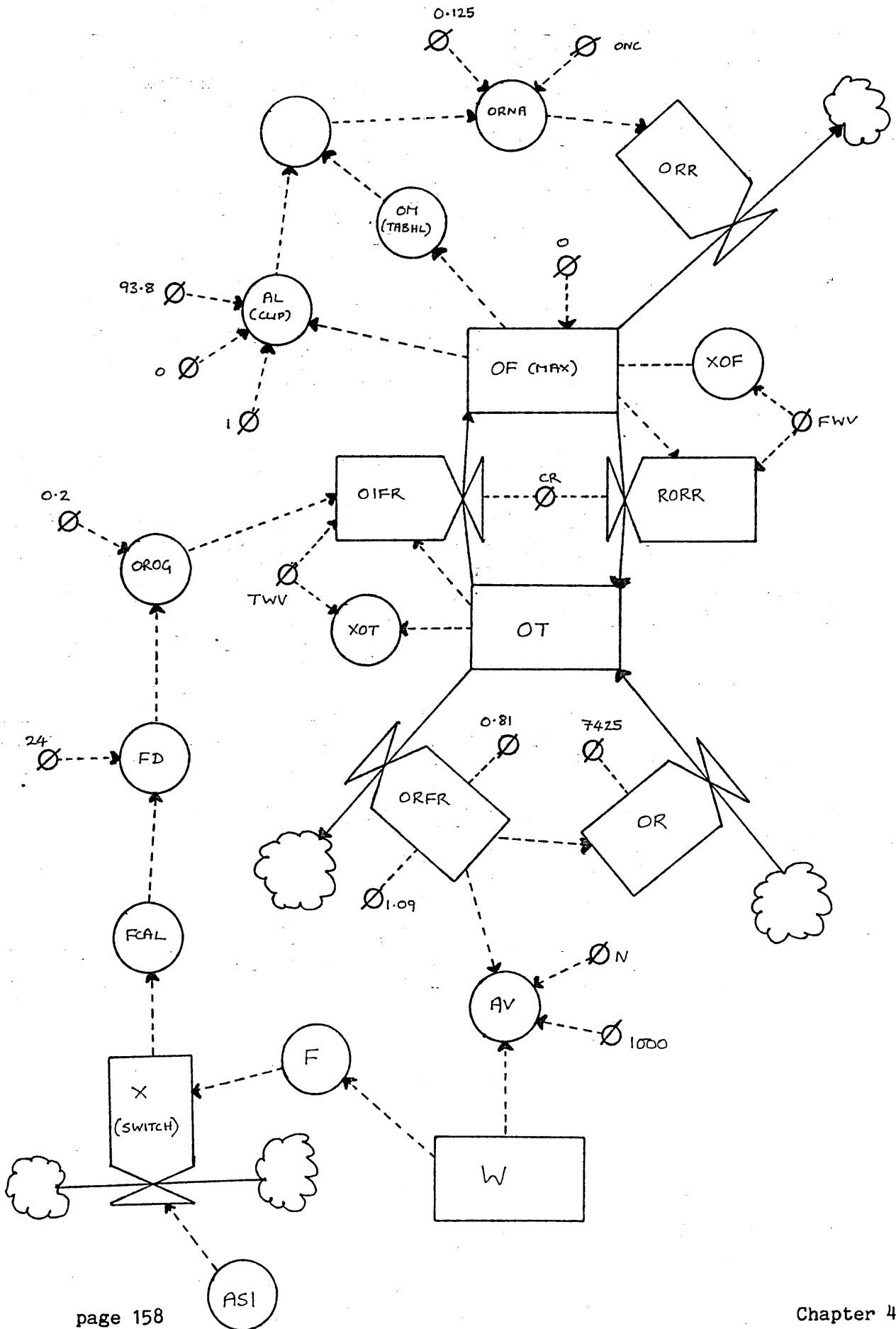
It is unnecessary to safeguard the level of oxygen in the filter against exceeding 100 per cent saturation, since there are no additions of oxygen to the filter, and the water entering the filter can only be at 100 per cent saturation. It is necessary to ensure that the level never becomes negative, but is only allowed to fall to a value of zero. This can be achieved by the use of a 'maximum' function:

So,

$$\text{OF} = \text{MAX} (0, \text{OF} + \text{DT}*(\text{OFIR} - \text{ORR} - \text{RORR} ))$$

The diagram to represent .OSIM is given below;

Figure 4.12 System Dynamics network diagram of .OSIM



#### 4.7 Assembly of Model

When all four sub-models had been constructed and tested they were assembled together. The three sub-models, .AMMO, .OSIM and .WEIGHT were linked in the following way:

- .WEIGHT was connected to .AMMO via AI;
- .WEIGHT was connected to .OSIM via AV and OROG;
- and .OSIM and .AMMO were connected together via AL and AN.

This integration necessitated a thorough checking to ensure no replication or incompatibility of time periods.

The full network diagram of the model is given in Fig. 4.13 and the programme listing is presented in Table 4.2.



Table 4.2 Key to Figure 4.13

AF	=	Ammonia in Filter
AT	=	Ammonia in Tank
OF	=	Oxygen in Filter
OT	=	Oxygen in Tank
W	=	Weight of Fish
A	=	Information level
ARR	=	Ammonia Removal
AIFR	=	Ammonia entering into the Filter
RARR	=	Residual Ammonia returning to Tank
APR	=	Ammonia production
AI	=	Ammonia input
ORR	=	Oxygen removal
RORR	=	Residual Oxygen returning to Tank
OFIR	=	Oxygen entering into Filter
OR	=	Oxygenation
ORFR	=	Oxygen removed by Fish Respiration
WDR	=	Weight decrease of Fish
WIR	=	Weight increase of Fish
ORNA	=	Oxygen removed by the nitrification of ammonia
OROG	=	Oxygen removed by oxidation of organic matter
ASI	=	Information equation used to determine a 24 hour cycle
ASIF	=	Information equation used to override ASI if high ammonia levels occur
CHECK	=	Information equation checking if ammonia level in tank exceeds limit
KILL	=	Information equation checking if ammonia level in tank reaches toxic limit
HI	}	Information equations used to operate WDR if no food fed
HS		
SLOW)		
TM	=	Retention time
AL	=	A multiplier used in AN to stop nitrification when oxygen in the filter falls

AN = Nitrification equation  
OM = Multiplier used in AN  
FCAL = Food to be fed for whole day (theoretical information)  
FD = Food to be fed per hour (theoretical information)  
F = Food fed to Fish  
AV = Average weight of individual Fish  
ONC = mg oxygen removed per mg ammonia nitrified  
N = Number of Fish  
FWW = Filter water volume  
TWV = Tank water volume  
CR = Circulation rate  
CA = Ammonia produced as a percentage of food fed  
T = Temperature of water  
FCE = Food conversion efficiency  
X = Switch to provide information on feeding for simulation of  
organic waste production.  
XOT = Concentration of Oxygen in Tank  
XOF = Concentration of Oxygen in Filter  
CNT = Concentration of Ammonia in Tank  
CNF = Concentration of Ammonia in Filter

Other constants shown in Figure 4.13 and not listed above appear as numbers used within equations.

#### Dynamo 'Macro' Functions Used

MAX = Maximum value chosen ( max = P if  $P > Q$   
( max = Q if  $P < Q$ )  
EXP = Exponential  
LOGN =  $\text{Log}_e$   
CLIP (P, Q, CRIT, REF) CLIP = P if CRIT > REF, CLIP = Q if CRIT < REF  
DELAY<sub>3</sub> = Third order delay function  
SWITCH (P, Q, CRIT) SWITCH = P if CRIT = 0, SWITCH = Q if CRIT = 0  
TABHL = Linear interpolation between points in a table. If values higher  
than that used in table then assume the last value of table.

Table 4.3 Programme listing for standard run

```
L OF.K=MAX(0,OF.J+DT*(OFIR.JK-ORR.JK-RORR.JK))
L OT.K=OT.J+DT*(RORR.JK+OR.JK-ORFR.JK-OFIR.JK)
R ORR.KL=ORNA.K
A ORNA.K=(AN.K*ONC)/0.125
C ONC=4.33
R OR.KL=(ROC-OT.K)+ORFR.JK
C ROC=7425
C N=60
A AV.K=(W.K/1000)/N
R ORFR.KL=1.09*(EXP(0.81*LOGN(AV.K)))*N
R RORR.KL=(OF.K/FWV)*CR
C SC=0.2
A OROG.K=SC*FD.K
S XOT.K=OT.K/TWV
S XOF.K=OF.K/FWV
R OFIR.KL=((OT.K/TWV)*CR)-OROG.K
C FWV=134
C TWV=900
C CR=900
N AF=26.8
N AT=180
L AF.K=AF.J+(DT*(AIFR.JK-ARR.JK-RARR.JK))
L AT.K=MAX(180,AT.J+DT*(APR.JK-AIFR.JK+RARR.JK))
N OF=1107
N OT=7425
L A.K=A.J*(1-ASI.J)+DT
A ASI.K=CLIP(1,0,A.K,24)
N A=0
R APR.KL=DELAY3(AI.JK,6.0)
A ASIF.K=CLIP(0,1,CHECK.K,ASI.K)
A CHECK.K=CLIP(1,0,AT.K,900)
A KILL.K=CLIP(1,0,AT.K,18000)
A HI.K=SWITCH(2,1,ASI.K)
A HS.K=CHECK.K-HI.K
```



```

A SLOW.K=SWITCH(1,0,HS.K)
R AI.KL=F.K*CA
C CA=0.03
R AIFR.KL=(AT.K/TWV)*CR
R ARR.KL=AN.K
C T=25
A TM.K=FWV/CR
R RARR.KL=(AF.K/FWV)*CR
A AL.K=CLIP(1,0,OF.K,93.8)
A AN.K=(0.11*T-0.2)*0.96*TM.K*AF.K*OM.K*AL.K
A OM.K=TABHL(OMOMT,(OF.K/AF.K),0,4.33,2.165)
T OMOMT=0/0.75/1
S CNT.K=AT.K/TWV
L W.K=MAX(0.1,W.J+(DT/.125)(WIR.JK-(WDR.JK*SLOW.J))-W.J*KILL.J)
R WDR.KL=W.K*0.005
S CNF.K=AF.K/FWV
R WIR.KL=F.K*FCR
A FCAL.K=X.JK
N X=0
R X.KL=SWITCH(X.JK,F.K,ASI.K)
A FD.K=FCAL.K/24
A F.K=W.K*FC*ASIF.K
N FC=0.025
C PLTPER=24
C PRTPER=24
C LENGTH=8760
C DT=0.125
N W=5000000
C FCR=0.4
PLOT W=W/CNT=T/CNF=F/XOT=X/XOF=Z
PRINT W,F,APR,AIFR,OROG,RARR,AN,OM,OT,OF,AT,AF,RORR,OFIR
RUN
C PLTPER=1
C PRTPER=1
C LENGTH=500
RUN

```

## 4.8 Operation of the Model

### 4.8.1 Running the Model

The following input data are required for the operation of the model:-

- a) The initial values of the state variables (levels). For the standard run these were set as follows:

Ammonia in tank (0.2 mg/l)	AT = 180.00
Ammonia in filter (0.2 mg/l)	AF = 26.8
Oxygen in tank (8.25 mg/l)	OT = 7425.0
Oxygen in filter (8.25 mg/l)	OF = 1107
Weight of fish (5 kg)	W = 5,000,000

- b) A description of the operating condition for the run; temperature (T), number of fish (N), circulation rate (CR) food conversion efficiency (FCE) and ration level (FC). In addition it is necessary to specify the reoxygenation constant (ROC) which is dependent on temperature.
- c) The total number of hours the model is to run (LENGTH), the variables to be plotted and the symbols used, the variables to be printed and the frequency at which the results are plotted (PLTPER) and printed (PRTPER).

### 4.8.2 Model Output

Within Dynamo, any variable value can be output, at intervals specified by the user. In the 'standard' runs for this model the results are printed at hourly intervals (PRTPER = 1) over a 500 hour period, and once every 24 hours for a one year period (LENGTH = 8760).

A plotting facility is available in Dynamo, allowing a maximum of fourteen variables in any one plot. To aid clarity in the standard run only the state variables are plotted.

Interpretation of the results is made easier if the levels of oxygen and ammonia are expressed as concentrations. Supplementary variables labelled S in the programme listing, (Table 4.3) were introduced into the programme

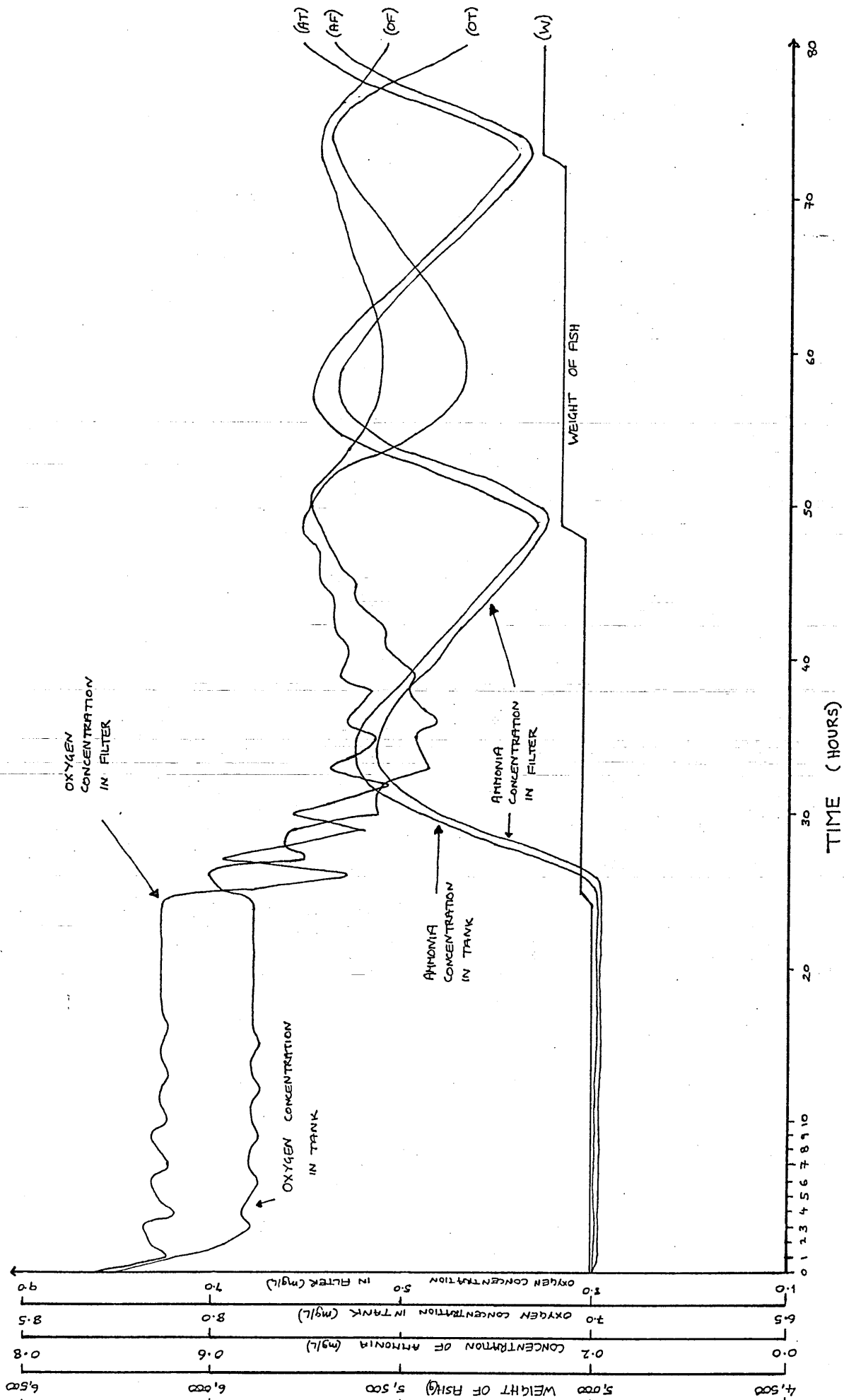
for this purpose. The variables CNT, CNF, XOT and XOF represent respectively the concentrations of ammonia in the tank and filter and oxygen concentrations in the tank and filter, and at any given time are equivalent to AT, AF, OT and OF.

Plots of the state variables at one hour and twenty four hour intervals are presented in Figs. 4.14, 4.15 and 4.16 respectively.

Examinations of the hourly plots reveal that for the model system:

1. Equilibrium was established seventeen hours after initialization.
2. Feeding resulted in a discrete increase in fish weight.
3. Ammonia concentration in the tank rose smoothly to a maximum of 0.46 mg/l ten hours after feeding, falling more gradually to a minimum (baseline) concentration of 0.27 mg/l one hour after the second feed. After each successive feed the baseline concentration showed an increase.
4. The ammonia concentration in the filter was consistently lower than in the tank, but followed the same curve.
5. After feeding the oxygen concentration decreased sharply, falling to 7.42 mg/l in the tank and 5.28 mg/l in the filter. Both levels fluctuated erratically, with the largest variations exhibited in the filter concentration.
6. The oxygen concentration in the tank returned to a maximum of 7.78 mg/l (94% saturation) twenty six hours after feeding. A maximum oxygen concentration of 6.11 mg/l (74% saturation) was reached in the filter twenty four hours after feeding.
7. Following the second and subsequent feeds the variations in oxygen concentration were regular. After the second feed the concentration in the tank fell smoothly to a minimum of 7.36 mg/l eleven hours after feeding rising to a maximum (baseline concentration) of 7.72 mg/l two hours after the next feed.
8. The oxygen concentration in the filter after the second feed fell to a minimum of 5.29 mg/l eleven hours after feeding, rising smoothly again to a maximum of 6.00 mg/l after 25 hours.

FIGURE 4.14



The twenty-four hourly plots revealed.

1. A steady increase in the weight of fish, although during the 80 day period from day 80 to 160, the increase was 'checked' twenty eight times because of feed being withheld.
2. The ammonia concentration in both tank and filter, (the baseline concentration) rose over the first 82 days, with the rate of increase becoming more rapid with each successive feed until a level of 1.04 mg/l was reached on day 83. With the concentration over 1 mg/l the fish were not fed and the first check to fish growth occurred. The weight of fish at this point was 11.194 kg and the feed was 277g.
3. Following the withholding of feed on day 83, the ammonia concentration on day 84 had fallen to 0.35 mg/l. Feeding was resumed with the baseline concentration increasing again to a concentration of 1.11 mg/l on day 88 when the next feed was missed. A pattern of missed feeds was established with the number of days between each missed feed decreasing as the increase in baseline concentration became more rapid with each successive feed.
4. As the run continued the level to which the ammonia concentration fell after each missed feed began to rise and the maximum baseline concentration also increased.
5. The ammonia concentrations in both tank and filter were similar throughout the run, and because of the scales used are occasionally shown as a single plot.
6. The decrease in the baseline concentration of oxygen in both tank and filter was rapid for the first three days, but then declined more slowly until day 82. Initially similar, after four days the oxygen concentration in the filter was 22 per cent less in the tank. By day 82, the increased loading resulted in an oxygen concentration in the filter 61 per cent less than in the tank, i.e. 2.62 mg/l compared to 6.71 mg/l in the tank.

FIGURE 4.15.

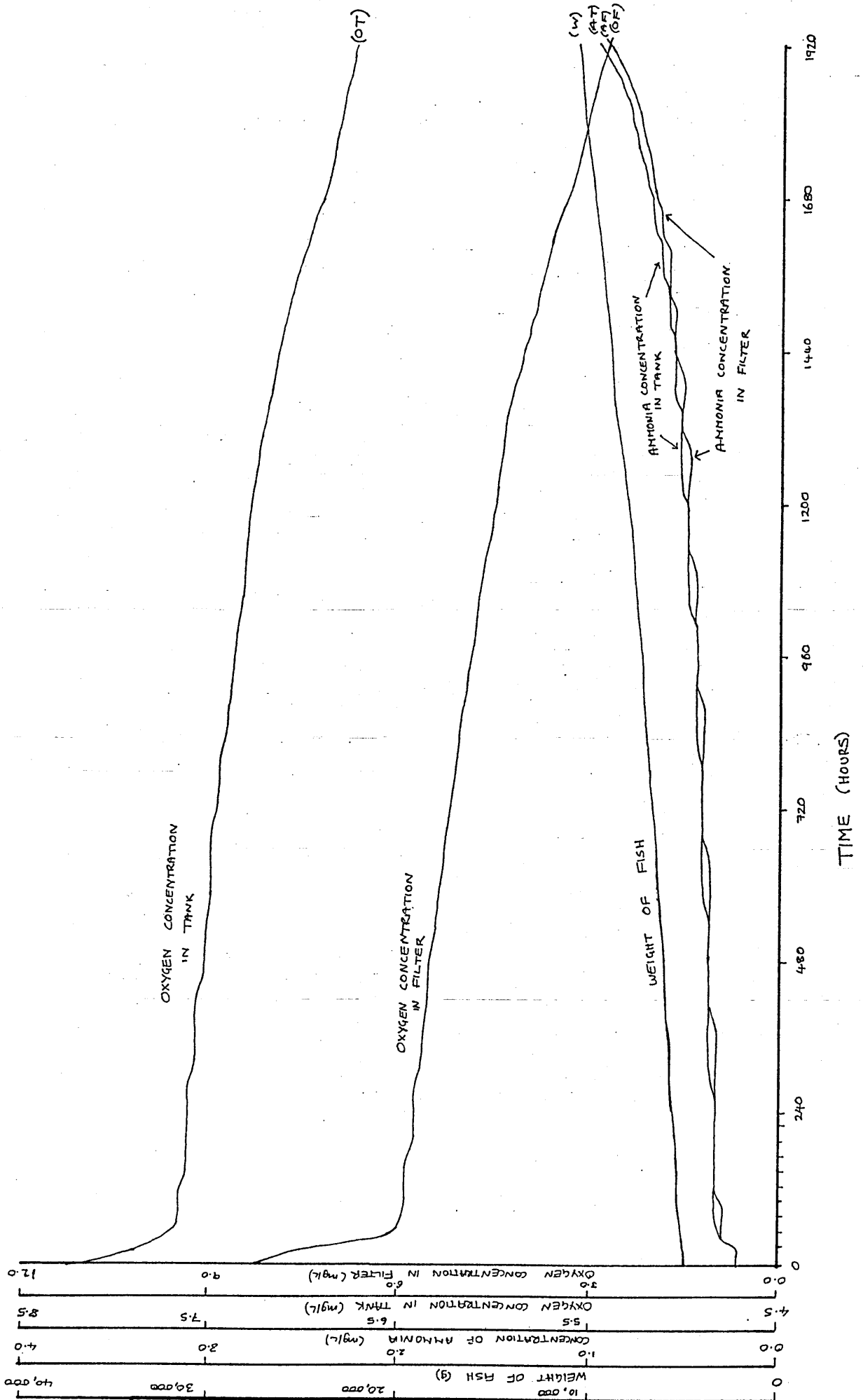
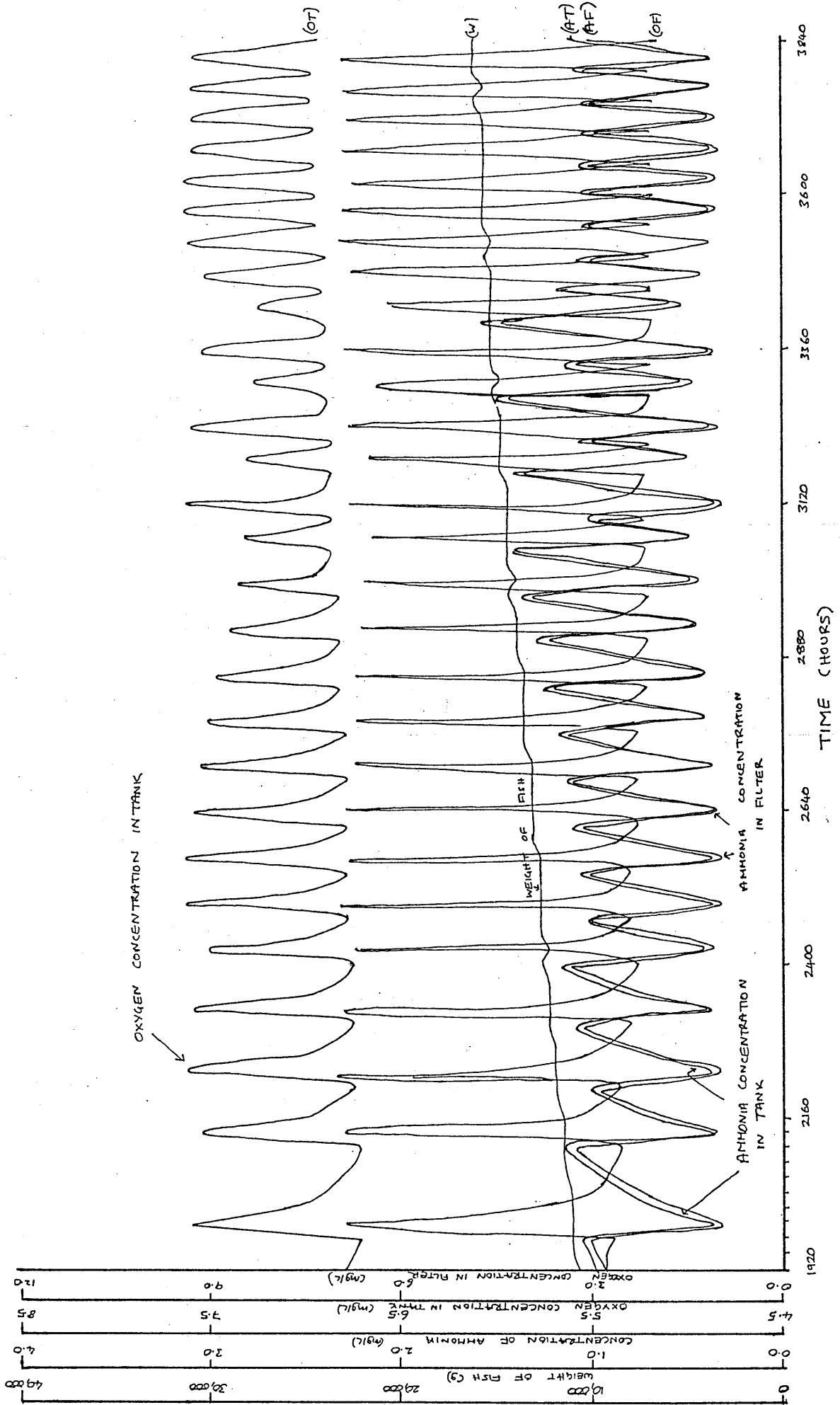


FIGURE 4.16.



7. As a result of the variations in ammonia production arising from the missed feeds, the oxygen concentration in both tank and filter showed considerable variation. As the pattern of feeding/missed feeds became established, so the pattern of variations in oxygen became more regular. The minimum concentration reached during the oscillations decreased in the filter from 2.46 mg/l to 1.97 mg/l while in the tank the minimum level increased from 6.70 mg/l to 6.95 mg/l.

Although no serious difficulties were encountered, the standard runs did reveal a difference between the expected and actual behaviour of the model with regard to the oxygen concentration in the tank. The model was constructed with the intention that the tank concentration would be maintained at 100% saturation. To achieve this the aeration of the tank was modelled by adding to the tank the amount of oxygen consumed in respiration (ORFR) together with the difference between the theoretical maximum concentration contained in the tank (ROC) and the actual level in the tank (OT), that is;

$$OR = (ROC - OT) + ORFR$$

The results of the standard run clearly showed that the tank concentration was not maintained at 100 per cent saturation. This is because of the constraints of the time notation required by Dynamo, and the order in which Dynamo solves the equations of the model (5.3.4). As can be seen from the concentrations of oxygen measured during experiment 3.3.2.1 (Table 3.3.3) 100 per cent saturation in the tank was not maintained in the reference system either. The structure of the model was therefore left unchanged.



## CHAPTER FIVE EVALUATION OF THE MODEL

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  - 5.2.1. Development of the model
  - 5.2.2. Analysis of the final model
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  - 5.2.3. Conclusions.
  
- 5.3 Validation
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  - 5.3.3. Discussion
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## 5. EVALUATION OF THE MODEL

### 5.1 Introduction

The process of model evaluation can be divided into verification and validation (Penning de Vries, 1977). Whilst Naylor and Finger (1967) have treated these terms as synonyms meaning "to prove the model to be true", this presents a number of philosophical difficulties. In practical terms verification can be considered as ensuring that the model behaves as the modeller intended, and validation as the testing for agreement between the models behaviour and the reference system (Fishman and Kiviatt, 1967; Mihram, 1972).

### 5.2 Verification

Since an aim of the model was to increase understanding of the way in which the system behaves, it was essential to ensure that there were no inconsistencies in the model's internal structure and behaviour, and that the detailed structure agreed with the original specification. In addition, verification is particularly important if the model has predictive results or is to be used in a predictive manner.

At present there are no systematic methods for the verification of simulation models (Brockington, 1979). In many models verification does not extend beyond checking the computer programme for errors in the numerical data, and for consistency of equation dimensions. Stone (1982) has developed a suite of computer programmes for the analysis of system dynamics models based on:-

- a) Analysis of model structure through use of graph theory
- b) Inspection of a standard run for maximum, minimum, mean and variation.
- c) Sensitivity testing.

These programmes were made available by Stone for the evaluation of the model, and assistance in running the programmes and interpretation of the output was provided. A report containing the results of the initial analyses was presented to the author by Stone (1981) and where appropriate figures and tables have been reproduced and presented in the text.

#### 5.2.1 Development of the model

Analysis of the initial model revealed inconsistencies in the internal structure and behaviour that required alteration. These changes to the model's structure were sufficient to necessitate re-analysis of the whole model. The model described in chapter four represents the final version of this iterative process and differs from the initial model in the following respects:-

- a) The sub-model .AMMO, .OSIM and .WEIGHT were joined in the initial model with paths through AN, AI and AV with A and ASI providing the feeding switch mechanism (Fig. 5.1). This was revised with additional switching mechanisms, AL - to prevent nitrification when the oxygen level in the filter falls below 0.7 mg/l; ASIF, CHECK, HS and SLOW - to control feeding when ammonia levels exceed 1 mg/l and to bring into operation the equations for reducing fish weight and KILL - to operate the catastrophe state where all the fish die from excess ammonia. In addition a separate path was introduced for the removal of oxygen through the oxidation of organic wastes (OROG) with a switching mechanism (X) making the path inoperative when the fish were not fed.
- b) In the initial model nitrification in the filter was calculated using the incoming ammonia concentration. This was revised so that the rate of nitrification would be more sensitive to changing concentrations of ammonia within the filter (the replacement of AT by AF in the appropriate equation).
- c) Analysis of the initial model revealed that the weight of fish changed by only a small amount during a standard run. Careful inspection of the programme revealed that only a fraction of the food fed was converted into weight. This resulted from the use of DT in the level equation. Weight increase occurs in response to feeding and the use of switching mechanisms to control

feeding means that weight increase occurs over one time step only. To allow the full weight of food to be available for conversion into fish weight, DT was divided by its numerical value.

- d) The constant used in the clip AL to represent the concentration of oxygen at which nitrification ceases was wrongly calculated; the concentration 0.7 mg/l was multiplied by the weight of oxygen in the tank and not in the filter. This was detected during validation of the model, and its correction required reconfiguring the model and re-analysis of the results. It was also necessary to re-examine the model using the inspection programme and repeat the sensitivity analysis.

Figure 5.1 Initial model

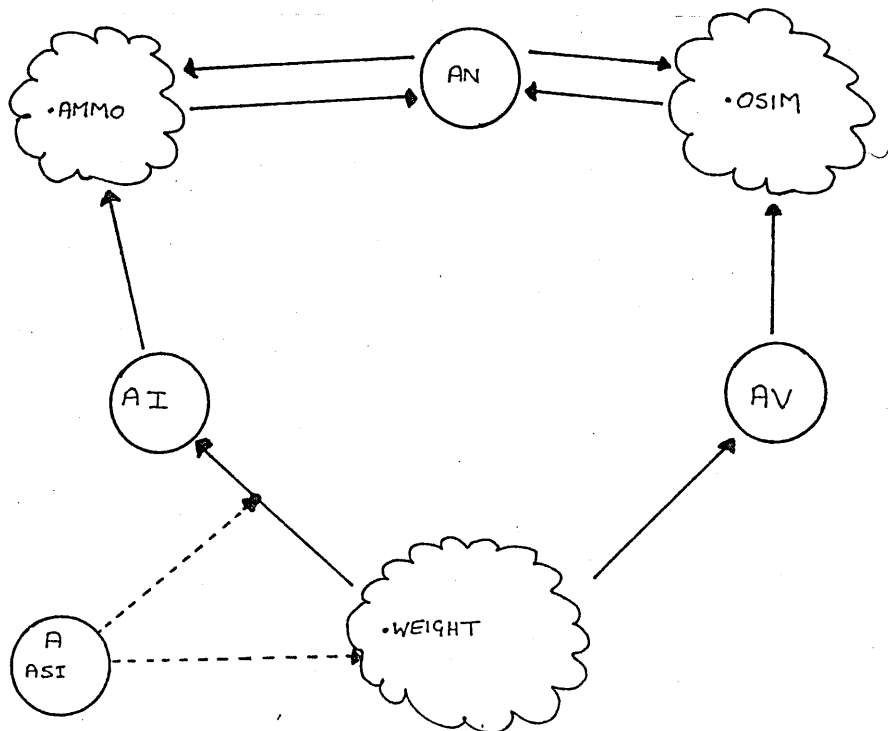
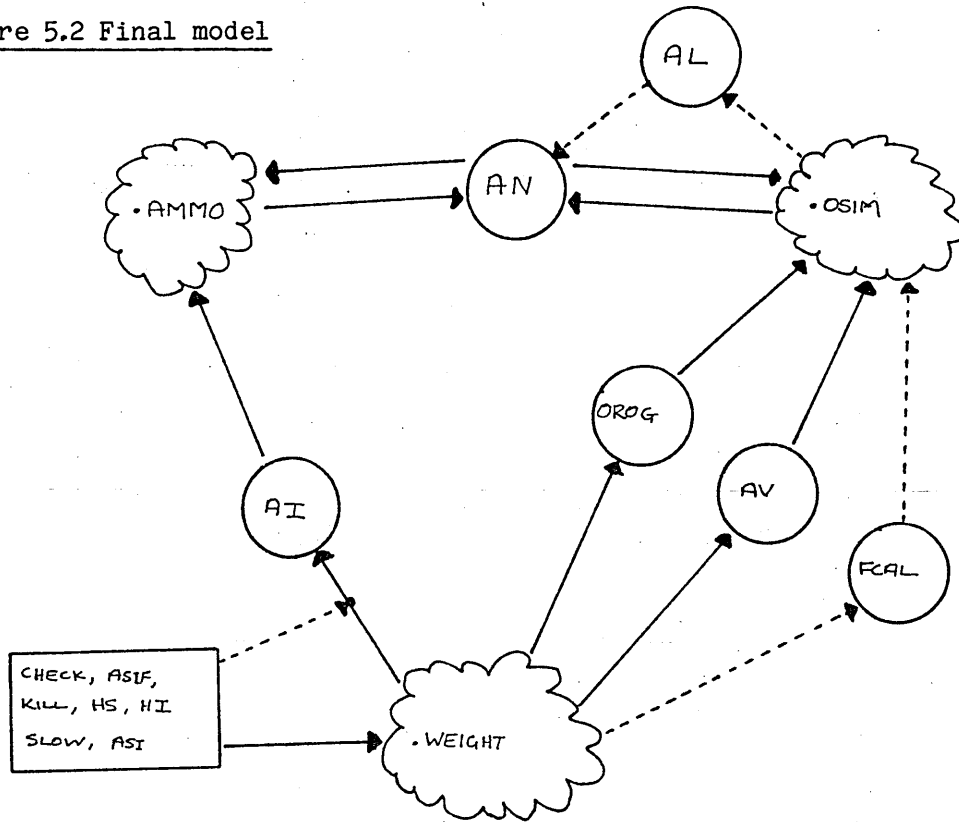


Figure 5.2 Final model



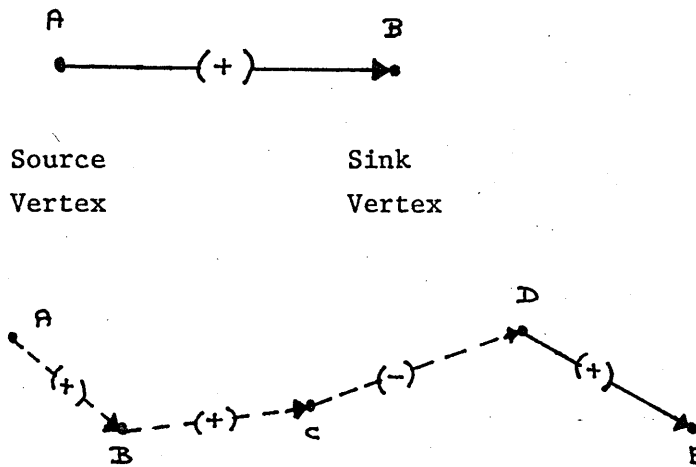
5.2.2 Analysis of the final model

5.2.2.1 Examination of model structure using signed digraphs

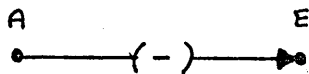
A signed digraph is a "map" of the model and is similar to a network diagram in System Dynamics. The main difference is in the treatment of the lines joining rates and levels (Stone, 1981). In a digraph each variable is called a vertex and the lines joining vertices are called arcs. Normally vertices are represented in a digraph by the same symbol, but to maintain some continuity with System Dynamics the vertices are represented according to that convention. However the arcs joining the vertices do not represent the flows of material and information but indicate the temporal relationship between vertices. Each digraph contains both time-sliced arcs (where the value of B at time t is calculated from the numerical value of A at time t-1) and event-sequenced arcs (where the numerical value of B at time t is calculated from the numerical value of A at time t). Arcs between switches are shown as dotted lines. The signs attached to each arc provide a qualitative assessment of the effect a change in the source vertex will

have on the sink vertex (Fig. 5.3). For example, if the arc is signed with a (+) this means that when A is numerically increased this will always cause a numerical increase in B. If the sign is negative (-) a numerical increase in A will cause a numerical decrease in B. If the change in B can be either positive or negative, then the arc is signed with a query (?).

Figure 5.3 Guide to terms used in the Graph Theory



An event sequenced path with a time-sliced arc



Same path in time-slice of full digraph

where  $(-) = [(+) * (+) * (-) * (+)]$

Results: The examination of signed digraphs constructed for earlier versions of the model resulted in the replacement of an incorrect arc and the introduction of a number of switches (section 5.2.1 (a)+(b)). The effect of these switches was to create five modes of behaviour. One mode is the catastrophe state in which all the fish are killed when the ammonia concentration in the tank exceeds 20 mg/l. This mode was not analysed.

The other four modes are:-

Mode 1 Fish are fed ( $AT < 1 \text{ mg/l}$ ), nitrification occurs ( $OF > 0.7 \text{ mg/l}$ )

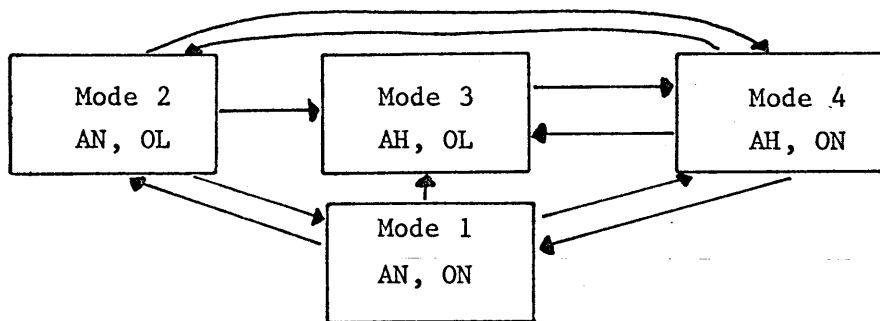
Mode 2 Fish are fed ( $AT < 1 \text{ mg/l}$ ), no nitrification ( $OF < 0.7 \text{ mg/l}$ )

Mode 3 Fish not fed ( $AT > 1 \text{ mg/l}$ ), no nitrification ( $OF < 0.7 \text{ mg/l}$ )

Mode 4 Fish not fed ( $AT > 1 \text{ mg/l}$ ), nitrification occurs ( $OF > 0.7 \text{ mg/l}$ )

As indicated, the change from one mode to another is governed by the level of ammonia in the tank (AT) and the level of oxygen in the filter (OF). An indication of how the modes can change is given in Fig.5.4.

Figure 5.4 Based on Stone (1981)



Key

Ammonia	AN	(Normal operating condition, $AT < 1 \text{ mg/l}$ )
in tank	AH	(Ammonia high, $AT > 1 \text{ mg/l}$ )
Oxygen	ON	(Normal operating condition, $OF > 0.7 \text{ mg/l}$ )
in filter	OL	(Oxygen low, $OF < 0.7 \text{ mg/l}$ )

It is only possible to go from mode 3 to mode 4 because no reduction in ammonia level can occur with insufficient oxygen. The links between modes 2 and 4, and between modes 1 and 3 have the least probability of occurring, because there is a requirement that both levels cross the threshold values at the same time.

The most likely loop sequences are mode 1 followed by mode 4, then either, mode 3, mode 4, mode 1 or back to mode 1 directly. This occurs if the ammonia concentration in the tank rises above  $1 \text{ mg/l}$  without the oxygen level in the filter falling below  $0.7 \text{ mg/l}$ . If the oxygen level falls without the ammonia level rising above  $1 \text{ mg/l}$  the most likely loop sequence is mode 1, followed by mode 2, then either back to mode 1 directly,

or via mode 3 and mode 4. The passage from mode 3 to mode 4 in both cases marks the recovery of the system with the resumption of nitrification. In modes 3 and 4 the fish are not fed. In commercial fish culture this would not be acceptable, and entry into either of these modes can be used to signify that the carrying capacity of the system has been reached (see also 2.2).

Each of the modes has itself two modes of behaviour, one associated with the proposed feeding time (sub-mode a), and one for the following 24 hours (sub-mode b). The principal difference between these sub-modes is that in sub-mode (b) the weight of fish ( $W$ ) remains constant and acts as a source vertex, and the rate  $AI$  is zero. The DELAY also acts as a source vertex, but tends to zero by the end of the 24 hour period. The analysis of the modes was based on the proposed feeding time (sub-mode a) as this was the basis for the following cycle. Digraphs for the four modes are presented in Figs. 5.5 - 5.8 while Fig. 5.9 is the digraph of the complete model. On the digraphs all vertices are included, but only the arcs that are not zero. A key to the digraphs is presented in Fig. 5.5.

Figure 5.5 Mode 1

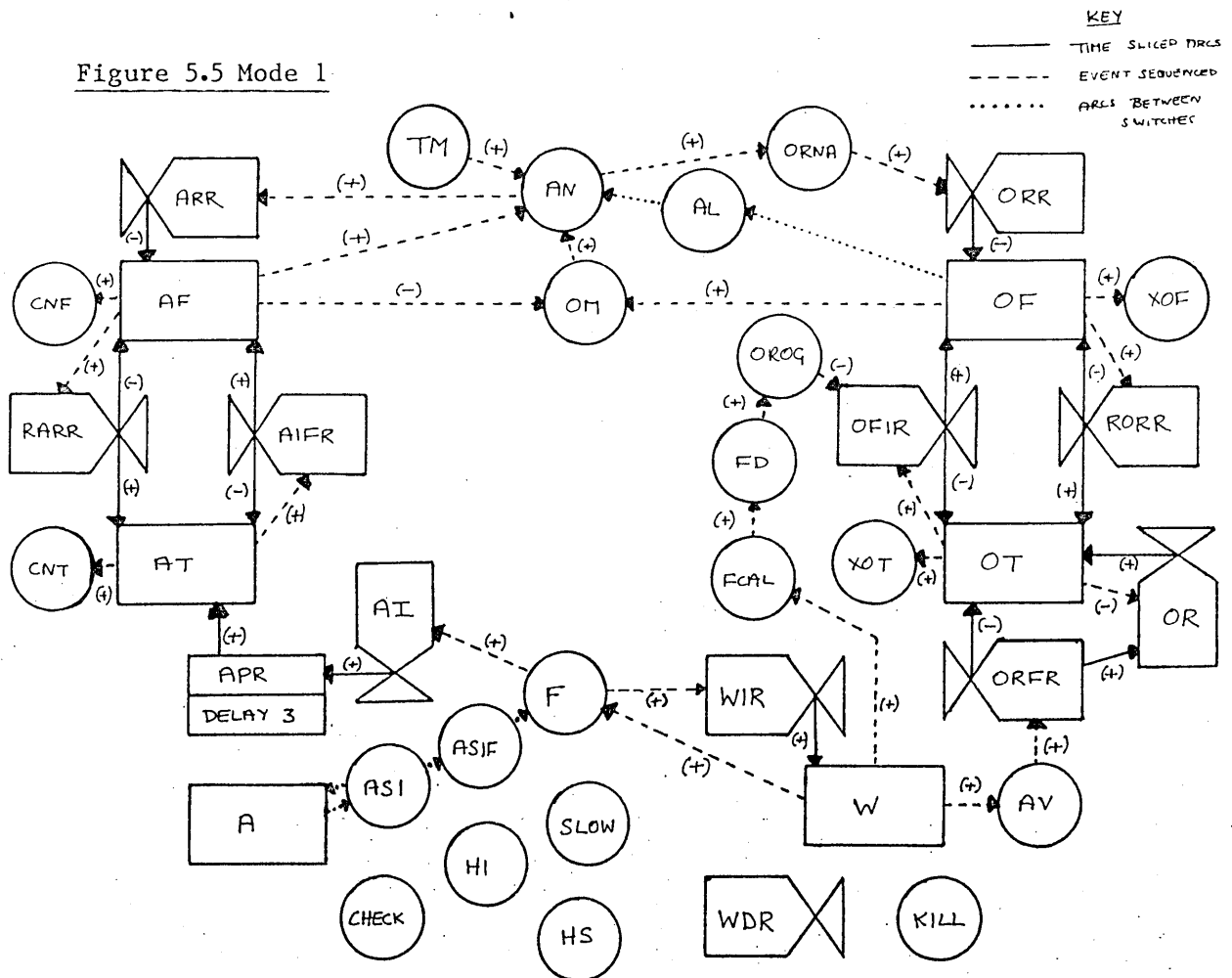




Figure 5.6 Mode 2

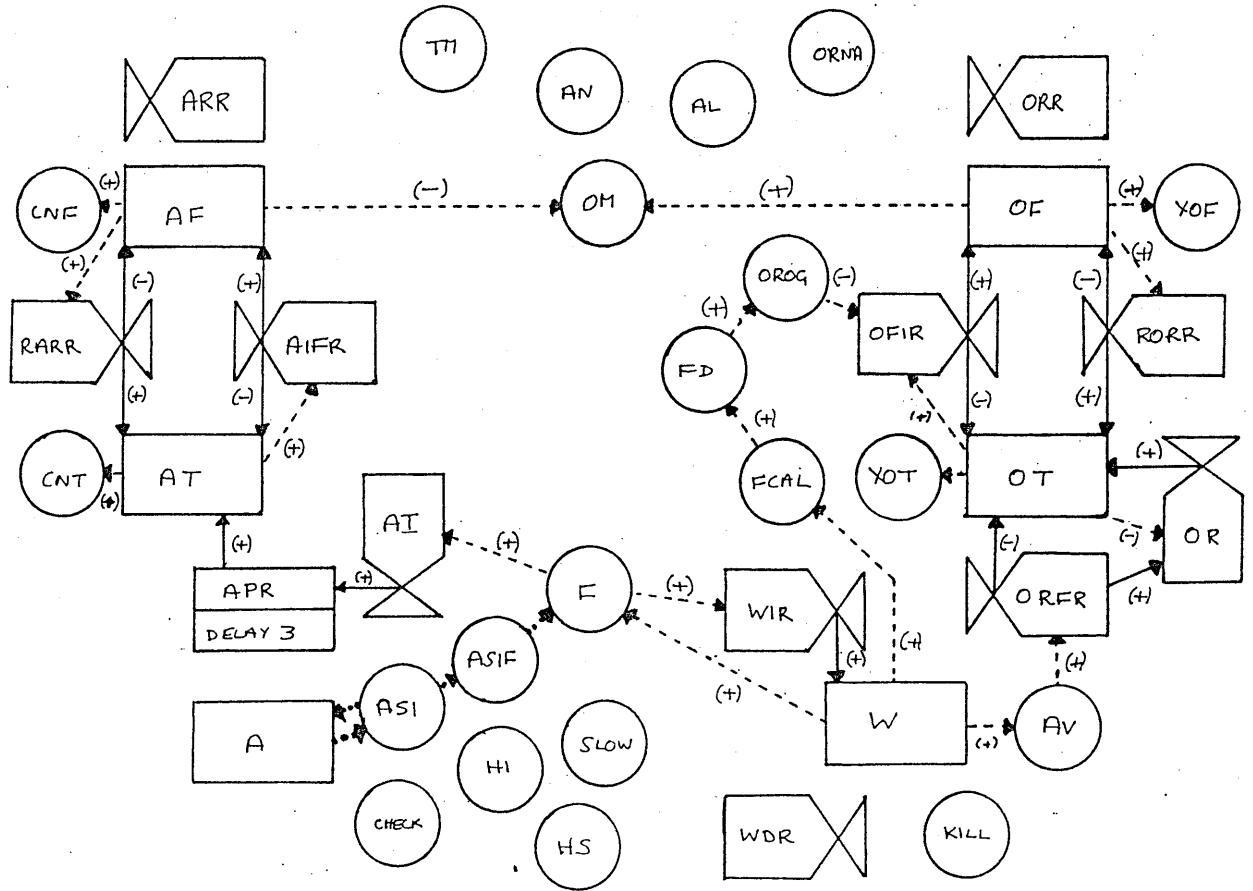


Figure 5.7 Mode 3

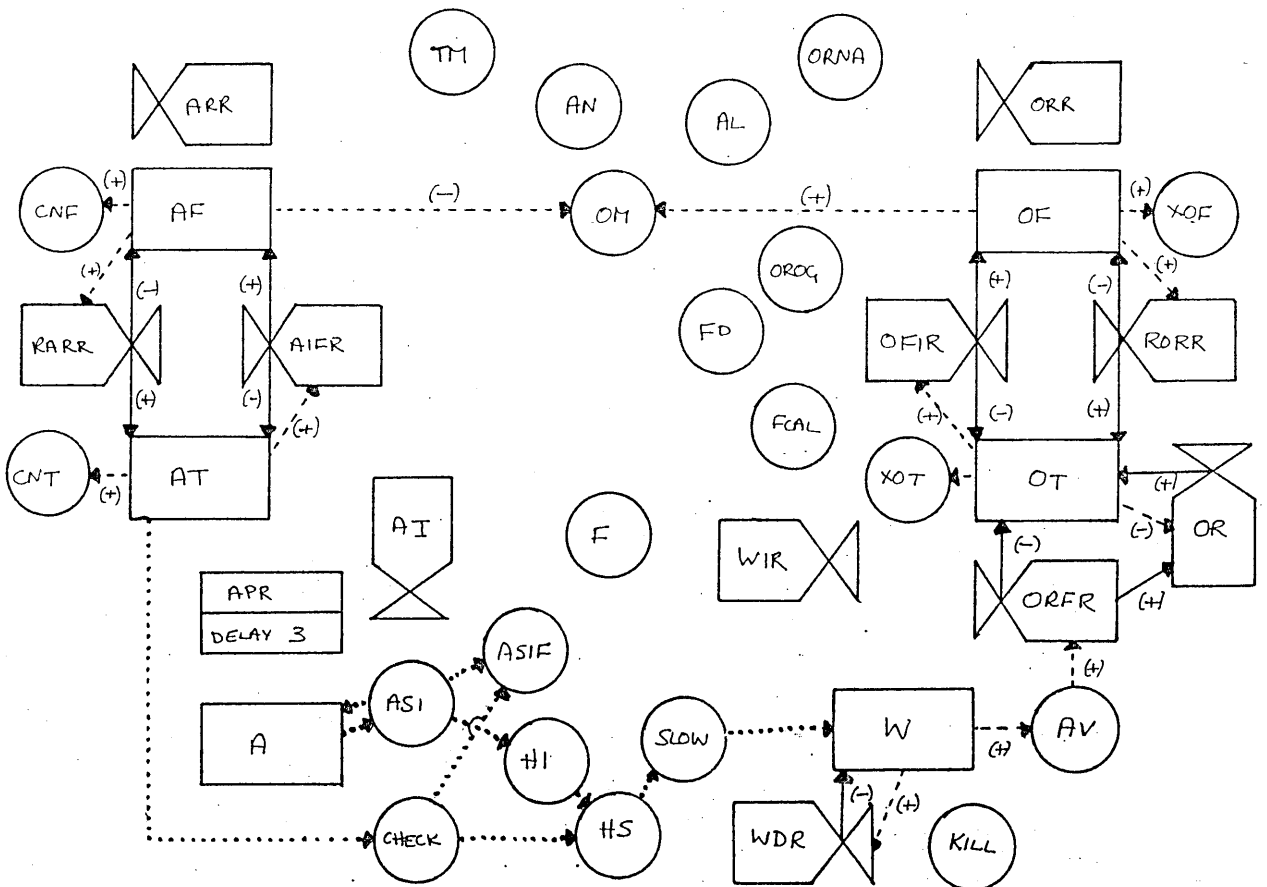


Figure 5.8 Mode 4

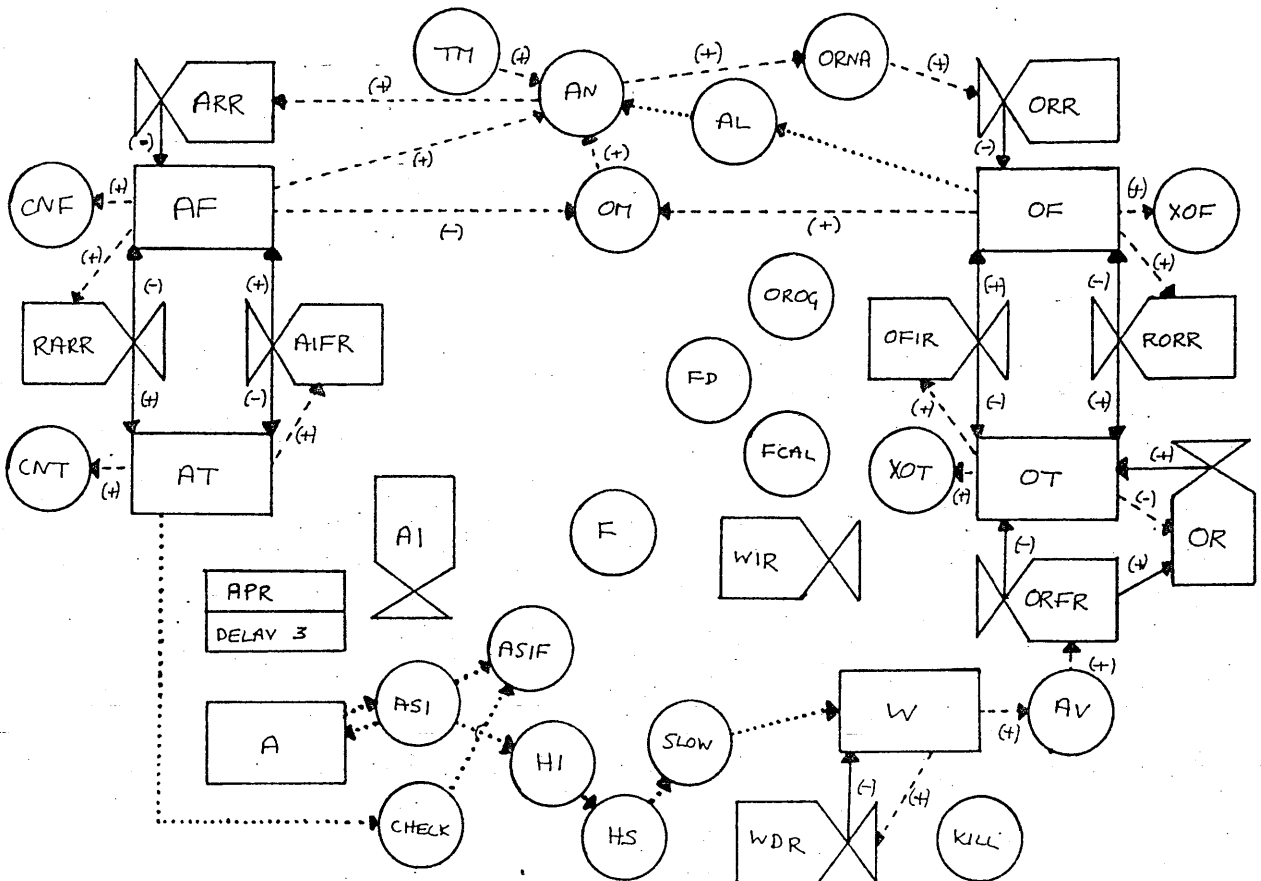
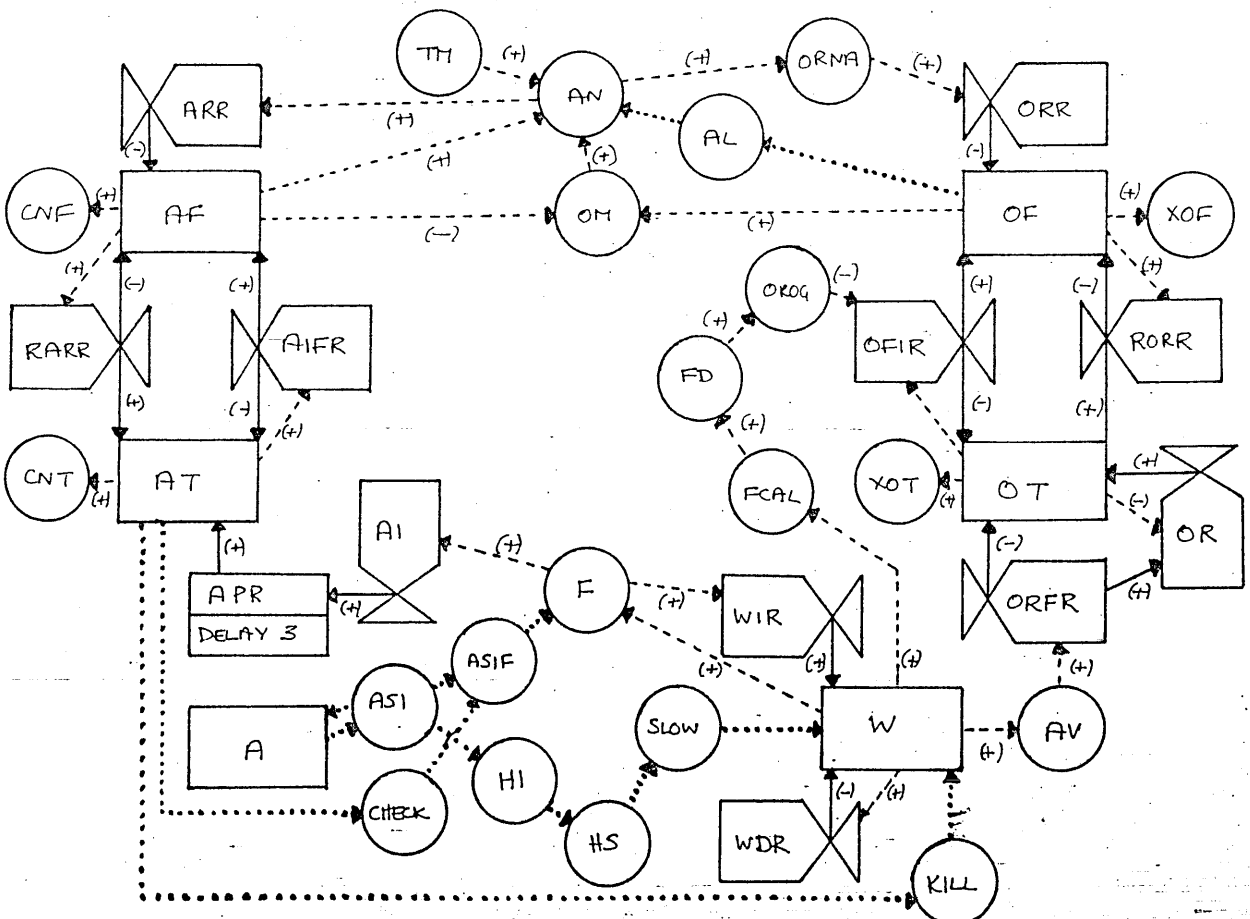


Figure 5.9 All paths



Discussion: With Dynamo it is difficult to follow the effects and interaction of the switches in creating different modes of behaviour. Use of signed digraphs to indicate the action of the switches in isolating variables was therefore of considerable value. The digraphs presented in Figs. 5.5 - 5.9 clearly indicate which variables were acting as source and sink vertices. In mode 1 (Fig. 5.5) WDR is isolated (switches CHECK, SLOW and KILL are at zero), TM is a source vertex and XOT, XOF, CNF and CNT are sink vertices. In mode 2 (Fig. 5.6) AL is switched to zero isolating ARR, TM, ORNA and ORR. Other switches are as in mode 1, thus still isolating WDR. OM has become a sink vertex. In mode 3 (Fig. 5.7) ASIF and X are now zero and CHECK and SLOW are unity. Thus WDR operates and F and WIR become isolates, also isolating AI, APR, DELAY, FCAL, FD and OROG. In mode 4 (Fig. 5.8) AL is unity so that OM is no longer a sink vertex and ARR, AN, ORNA and TM are not isolated.

The results of this analysis emphasise the major changes to the structure and behaviour of the model which can occur as the result of changes in two key variables; AT and OF. How clearly this corresponds to the reference system is considered later. Differences in the source, sink and isolate vertices shown by the four digraphs (Figs. 5.5 - 5.8) confirm the four modes of behaviour identified in Fig. 5.4. The changes in the model's structure and behaviour which result from changes in mode were found by the author to be an acceptable representation of the reference system.

The digraphs drawn for each mode provide a means of examining the behaviour of the model when it is not dependent on a set of parameter values. In establishing the credibility of the model it was necessary to check the arcs and signs of each digraph and thus ensure that the structure and internal behaviour of the model under various conditions was as intended, or at least acceptable to the author. Apart from an incorrect arc detected in the initial version of the model, the structure and internal behaviour of the model represented by the signed digraphs was found to be as intended. Although no other errors were detected by further checking of arcs and signs, the author considered this technique to be of value, particularly since it required a comparison to be made between two alternative representations of the model and forced a careful examination of the model's structure to be made.

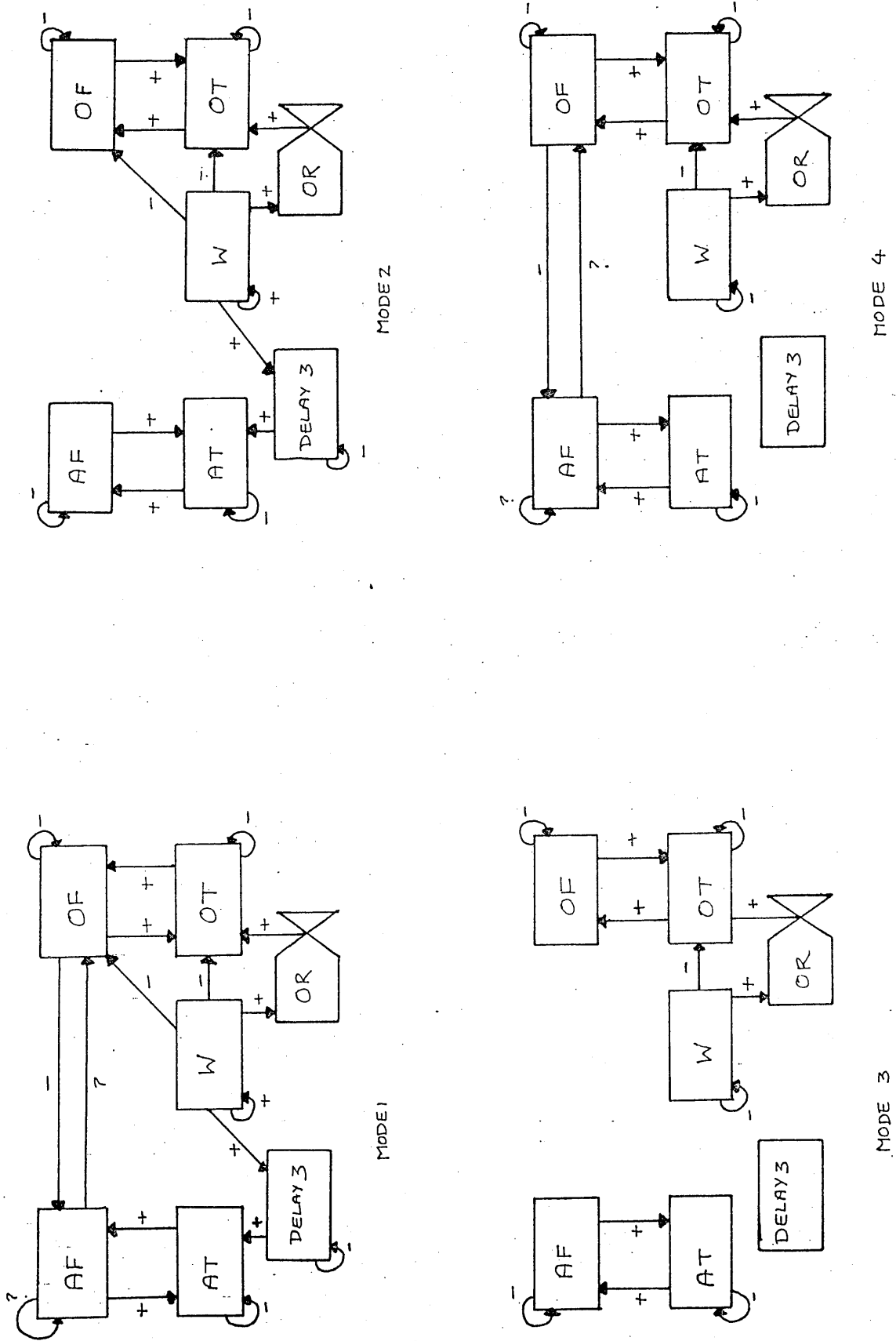
An additional benefit of the signed digraph is that it allows state variable dependence on state variable to be seen. For a better appreciation, the information presented in the digraphs 5.5 - 5.8 can be summarised. This is achieved by combining the signs of the event-sequenced arcs with the appropriate adjacent time-sliced arcs (Fig. 5.10). In this digraph DELAY 3 has been drawn as a single level. If a change in part of the model is to be traced through the whole model it must be remembered that there are two additional time steps in DELAY 3.

From Fig. 5.10 it can be seen that the weight of fish (W) is the source of the influences on the other state variables. In mode 1 an increase in W will cause a decrease in the oxygen in the filter (OF) and tank (OT), but it will also increase oxygenation (OR), thus restoring the oxygen levels in OT. The value of W at one time point affects OT at the next two time points. This is true of all modes. This temporal relationship was not intended by the author but arose from the notation required in Dynamo. After considering the reference system the two-time step influence of W on OT was thought acceptable.

In mode 2 the arc between OF and AF is lost, showing the oxygen and ammonia sub-models to be acting as sink vertices. This is because mode 2 represents a low oxygen concentration in the filter and no nitrification taking place. In mode 3 the ammonia sub-model becomes isolated, as the fish are no longer fed, and the path from W to OF via OFIR ceases so that no oxygen is removed prior to it passing into the filter, allowing the filter to recover more quickly. The sign of the self-loop to W has changed as the fish now lose weight with WDR replacing WIR. In mode 4 the recovery of the system is seen with the reforming of the arc between AF and OF. The fish are still losing weight and thus the paths between W and OF, and W and AT have not been reformed, with DELAY remaining isolated.

This analysis confirmed the action of the switches shown by the earlier digraphs, and provide a clear indication of the influence of one state variable (W) on the others. No errors in the model's internal structure and behaviour were detected, and the model was thus seen to be operating as intended by the author.

Figure 5.10 Time-slice digraph of the complete model



### 5.2.2.2 Inspection of the Standard Run

The inspection programme tabulated maximum, minimum, mean and percentage error (a measure of variation) for each of the variables used in the model during a standard run.

The maximum and minimum values indicate the range of the variable, while an indication of the symmetry of the variation is given by the mean. If the mean falls half way between the maximum and minimum then the values recorded during the run are symmetrical about the mean. The closer the mean to either the maximum or minimum, the more asymmetric the behaviour of the variable. At the extreme such behaviour is similar to pulses introduced into otherwise constant behaviour.

The percentage error also gives an indication of symmetry. If the percentage error is greater than 100 per cent, and the maximum and minimum values have the same sign, then pulse-like behaviour is indicated as possible. The greater the percentage error, the sharper the pulse may be. For percentage errors less than 100, symmetry is found using the mean. A percentage error of 100 indicates that either the maximum or minimum value is zero and symmetry is again examined using the mean. Percentage error was calculated from:

$$\left\{ \begin{array}{l} \text{Maximum} \\ \text{Minimum} \end{array} \left\{ \frac{\text{Max-Mean}}{\text{Mean}}, \frac{\text{Min-Mean}}{\text{Mean}} \right\} \right\} \times 100$$

The results of the inspection programme for the 500 and 8760 hour runs have been tabulated and are presented in Tables 5.1 and 5.2. The variables have been grouped according to their percentage error, allowing a simple study of the behaviour of the variables. To provide an indication of which switches operated during the standard run, switch variables were included in the inspection programme and are grouped separately.

Results:

Table 5.1 Results of Inspection Programme for 500 hour run, sampled hourly

Variable	MIN	MAX	MEAN	%ERROR
I (TM	0.15	0.15	0.15	0.00
I (OM	1.00	1.00	1.00	0.00
(ORFR	2352.00	2763.40	2550.31	8.36
II (W	5000.00	6100.90	5527.00	10.37
II (AV	83.33	101.68	92.12	10.37
(WDR	25.00	30.51	27.64	10.38
(OT	6424.40	7425.00	6714.25	10.59
(OR	2352.00	3764.00	3260.66	27.87
III (OFIR	5165.90	7425.00	5625.22	31.99
(RORR	4165.00	7435.10	4914.32	51.29
(OF	620.20	1107.00	731.45	51.34
(AF	25.42	79.87	56.47	54.98
(AN	9.27	29.11	20.58	54.98
IV (ARR	9.27	29.11	20.58	54.98
(RARR	170.73	536.43	379.26	54.98
(AT	180.00	565.94	399.89	54.99
(AIFR	180.00	565.93	399.89	54.99
(ORR	320.90	1008.40	712.95	54.99
(ORNA	320.90	1008.40	712.95	54.99
(OROG	0.00	1258.40	1088.68	100.00
V (FCAL	0.00	151.01	130.64	100.00
(FD	0.00	6292.20	5443.19	100.00
VI (APR	0.00	79.16	20.62	283.89
VII (F	0.00	181.01	5.49	2405.00
(AI	0.00	4530.40	104.81	2648.83
VIII (WIR	0.00	60.40	2.20	2648.81
(CHECK	0.00	0.00	0.00	0.00
(SLOW	0.00	0.00	0.00	0.00
(AL	1.00	1.00	1.00	0.00
IX (HI	1.00	2.00	1.96	48.98
(HS	-2.00	-1.00	-1.96	-48.98
(ASI	0.00	1.00	0.04	2405.00
(ASIF	0.00	1.00	0.04	2405.00

Table 5.2 Results of Inspection programme for 8760 hour run, sampled 24 hourly

Variable	MIN	MAX	MEAN	%ERROR
I TM	0.15	0.15	0.15	0.00
II OT	5618.60	7425.00	6434.77	15.39
(ORFR	2352.00	8335.10	5983.47	60.69
(OFIR	1797.90	7425.00	4612.48	61.02
III (OR	2.35	10.10	6.97	66.27
(W	5.00	23.84	16.05	68.84
(AV	83.33	397.36	267.43	68.84
(WDR	25.00	119.21	80.23	68.84
(OM	0.17	1.00	0.79	78.39
(ORR	320.90	1771.10	959.00	84.68
IV (ORNA	320.90	1771.10	959.00	84.68
(ARR	9.27	51.13	27.68	84.68
(AN	9.27	51.13	27.68	84.68
(OF	162.00	1107.00	543.30	103.76
V (RORR	1079.70	7435.10	3642.61	104.11
(AT	180.00	2246.30	860.74	160.97
(AIFR	180.00	2246.30	860.74	160.97
(AF	25.42	231.55	124.55	166.19
(RARR	170.70	2226.80	836.55	166.19
(OROG	0.00	4917.80	1819.37	170.30
VI (APR	0.00	0.40	0.15	
(AI	0.00	17.70	6.55	170.30
(WIR	0.00	236.05	87.33	170.30
(FCAL	0.00	590.13	218.32	170.30
(FD	0.00	24.59	9.10	170.30
(F	0.00	590.13	218.32	170.30
(AL	1.00	1.00	1.00	0.00
(HI	1.00	2.00	1.00	99.46
(ASI	0.00	1.00	-1.00	100.00
VII (ASIF	0.00	1.00	0.62	100.00
(CHECK	0.00	1.00	0.38	163.31
(SLOW	0.00	1.00	0.38	163.31
(X	0.00	590.13	218.32	170.30
(HS	-2.00	0.00	-0.62	-221.05

Discussion: Under certain circumstances it may be desirable to simplify model structure through the use of constants to replace variables showing limited variation, for example with a percentage error less than 15. From Table 5.1 it can be seen that during a standard run of 500 hours, eight variables had percentage errors of 15 or less. This reduced to two variables, OT and TM, for the longer run of 8760 hours.



WDR and W progressively increased in value during each run. During the shorter run, the daily increases compared to the initial value were small, giving a low percentage error. The increases in WDR and W over the longer run were much larger compared with the initial value, giving a correspondingly larger percentage error. The values of AV and ORFR were calculated using W, and therefore for the same reasons, showed similar increases in percentage error in the longer run. OM was used to limit the rate of nitrification according to the availability of oxygen. Its constancy during the shorter run indicates that although the oxygen concentration in the filter, ranged from 8.25 to 4.63 mg/l, this was always sufficient to meet the oxygen demands of nitrification. In the longer run oxygen concentrations in the filter fell to 1.21 mg/l which was not sufficient to meet the increased range of ammonia concentrations. OM therefore operated, with a consequent increase in its percentage error.

Although an auxiliary variable in the model, TM was used as a constant giving the zero percentage error in both runs. The 'topping-up' process of re-oxygenation (4.6.2) ensured minimal variation in the concentration of oxygen in the tank. As noted below (5.2.2.3), the model was insensitive to 1 per cent changes in the initial value of OT. A possible simplification of the model could be made by making OT a constant. With OT constant the time-sliced digraph of the complete model would change, with the removal of OT and OR. This would isolate W in modes 3 and 4, but the effect of this would be small as W only has a real effect on the behaviour of the model when it is increasing (modes 1 and 2).

The switching variables CHECK and SLOW operate when the concentration of ammonia in the tank reaches 1 mg/l or more. During the run of 500 hours the maximum value of AT was 0.63 mg/l (Table 5.1) and thus these switches did not operate. The ammonia concentration in the tank during the longer run reached 2.5 mg/l (Table 5.2), hence the use of these switches and larger percentage error. AL switches when the concentration of oxygen in the filter falls below 93.8 mg (0.7 mg/l). Since AL was not used in either run, modes 2 and 3 were not entered. These results indicate that during the shorter run the model operated in mode 1 only, while in the longer run mode 4 was occasionally entered, returning directly to mode 1.

During both standard runs, asymmetrical behaviour was found in many variables as a result of the switching mechanisms. In the 500 hour run, pulse-like behaviour was clearly displayed by groups V, VI, VII and VIII, and was related to the feeding mechanism. Group V were also affected by the switching mechanism of X, which operates when a feed is missed. The influence of the feeding mechanism was found in group IV, with a diminishing influence on groups III and II.

In the 8760 hour run other switch mechanisms operated reducing the influence of the feeding mechanism on the percentage error of all the variables. The results show that during the run HS and ASIF were on more often than off, whereas CHECK, SLOW, X and HI were off more often than on.

The symmetrical behaviour shown by variables in the groups I, II, and III reflected the 'topping-up' process of re-oxygenation and the constancy of ORFR as an oxygen sink.

By using the inspection programme a number of structural and behavioural patterns were identified in the standard runs. Grouping the variables by percentage error prompted an examination of the type of behaviour exhibited by each group and was of value in checking for anomalies. None were found. In addition it resulted in a thorough check of the computer programme for numerical errors and for consistency of equation dimension, and thus the programming error in W was detected as described earlier in 'The Development of the model' (5.2.1.(c)).

#### 5.2.2.3 Sensitivity analysis

The third stage of the analysis was that of examining state variable sensitivity to parameter change. During the two standard runs of 500 and 8760 hours the absolute maximum percentage change in the state variables was examined as each parameter was successively changed by one per cent. The definition of 'sensitive' used here is that of a percentage change greater than 1 per cent, implying that the change in state variable is proportionately greater than that in the parameter.

Results: Switching between modes made the use of sensitivity testing difficult. For example, if at time t during a standard run, AT is 899.9 the

fish will be fed; if with a change in parameter value the value of AT at time t is 900.1 the fish will not be fed. With a change in AT of only 0.2 the mode of behaviour of the model changes. A change in mode could result in a considerable change in the ammonia levels. Thus the sensitivity could be primarily connected with the change in modes and not with the behaviour within the modes themselves.

The maximum absolute sensitivity values are given in Tables 5.3 and 5.4. In both runs the maximum sensitivity to all parameters was found at the end of the run.

Table 5.3 Maximum absolute sensitivity of model over 500 hours  
(% change/1% change in variable)

	W	AT	AF	OT	OF
W	1.0	1.0	1.0	0.1	0.6
AT					
AF					
OT					
OF					
AL					
CA		1.0	1.0	0.11	0.3
CHECK					
CR		1.0	1.1	0.0	0.4
FC	0.2	1.2	1.2	0.1	0.7
FCE	0.2	0.2	0.2		0.1
FWV		2.2	1.3		1.1
OMOM					
ONC				0.1	0.3
ROC				1.1	1.6
T		1.2	1.2		0.3
TWV		1.1	0.1		1.4
0.005					
1.09					
0.11		1.2	1.2		
0.81					
0.96		1.1	1.1		
0.2 (AN)		0.1	0.1		
0.2 (OROG)					

Table 5.4 Maximum absolute sensitivity of model over 8760 hours  
 (% change/1% change in variable)

	W	AT	AF	OT	OF
W	-0.5	66.4	66.4	20.3	58.9
AT					
AF					
OT					
OF					
AL					
CA		219.1	227.1	119.1	71.4
CHECK		65.2	65.3	19.1	57.7
CR		1.2	1.4	0.4	3.3
FC	0.8	64.7	64.8	19.7	57.2
FCE	0.7	4.7	4.8	0.7	0.5
FWV	1.5	82.6	90.9	19.5	72.6
OMOM		0.8	0.9	0.1	0.5
ONC		1.0	1.0	0.4	0.1
ROC	1.5	120.3	128.3	20.5	71.4
T		66.2	66.3	20.1	58.4
TWV		120.6	125.5	19.1	-62.9
0.005	0.7	66.6	66.7	20.4	59.1
1.09					
0.11		66.2	66.3	20.1	58.4
0.81					
0.96		66.2	66.3	20.1	58.3
0.2 (AN)		0.1	0.1		
0.2 (OROG)		2.4	2.5	0.4	0.1

Table 5.5 Maximum absolute sensitivity to combined parameters  
 over 500 hours (% change/1% change in variable)

	W	AT	AF	OT	OF
Com 1					
(TWV,ROC,OT,AT)		1.1	0.1	1.1	0.1
Com 2					
(FWV,AF,OF)		2.2	1.3	0.0	1.1
Com 3					
(ONC,OMOM)				0.1	0.3

Table 5.6 Maximum absolute sensitivity to combined parameters  
over 8760 hours (% change/1% change in variable)

	W	AT	AF	OT	OF
Com 1 (TWV,ROC,OT,AT)		0.4	0.6	1.4	0.1
Com 2 (FWV,AF,OF)	1.5	82.6	90.9	19.5	72.6
Com 3 (ONC,OMOM)		1.9	1.9	0.4	0.1

Discussion: The first impression gained from a comparison of the state variable sensitivity over 500 and 8760 hours is the considerable increase in sensitivity over the longer run. This is due partly to the increased length of run and partly to increased switching between modes during the longer run.

From Table 5.3 it can be seen that during a run of 500 hours, W was insensitive to all parameters except its own initial value. During the longer run (Table 5.4) it became sensitive to FWV and ROC and less sensitive to its initial value.

The ammonia levels in tank and filter were sensitive to more variables. During the run of 500 hours, AF was sensitive to CA, CR, FC, FWV, T, W and the constants 0.11 and 0.96 used in the equation for AN. AT was additionally sensitive to TWV. Over 8760 hours, the sensitivity to these variables increases dramatically in the case of CA, FC, FWV, T, TWV, W and the constants 0.11 and 0.96. AF became very sensitive to TWV, and both AF and AT became sensitive to FCE, ONC, ROC, the constants 0.005 and 0.2 (OROG), and the value of CHECK (1 mg/l) used to control feeding.

The oxygen levels were less sensitive; over 500 hours OT was sensitive only to ROC while OF was sensitive to CA, FWV, OT and TWV. Sensitivity increased considerably over 8760 hours with both OT and OF becoming sensitive to FC, FWV, ROC, T, TWV, W, the constants 0.005, 0.11, 0.96 and the value of CHECK. OF was additionally sensitive to CR.

During the run of 500 hours the state variables were completely insensitive to one per cent changes in AF, AL, AT, CHECK, OF, OMOM, OT, and

the constants 0.005, 0.2 (OROG), 1.09 and 0.81. In the longer run the state variables were still completely insensitive to AF, AL, AT, OF, OT, and the constants 1.09 and 0.81. Except for the initial weight of fish the model is reasonably autonomous. As discussed earlier (5.2.2.6) AL is a switching value to cease nitrification in the filter which is not used. The constants 1.09 and 0.81 are used in the equation to calculate the consumption of oxygen by fish respiration, indicating perhaps that a simpler expression could be used (see 4.6.1.1).

In the sensitivity analysis it has so far, been assumed that all the parameters are independent. This is not correct, TWV, ROC and the initial values of OT and AT are all mutually dependent. Two other groups are FWV and the initial values of AF and OF, and ONC and OMOM. The sensitivity to the individual parameters is then only of academic interest. The sensitivity of the three combinations over 500 and 8760 hours are given in Tables 5.5 and 5.6 respectively.

Over 500 hours the combination of TWV/ROC/OT/AT had a moderating effect on the sensitivity of OF but the effect on AT, AF and OT was the same as by the individual parameters TWV and ROC. The moderating effect of the combination over 8760 hours on AT, AF, OT and OF is striking, reducing the sensitivity of the former two state variables by two orders of magnitude. The sensitivity of the second combination of variables over both 500 and 8760 hours are the same as that to FWV. This is due to the insensitivity of the variables to AF and OF. The sensitivity of the third combination over 500 hours is the same as that to ONC. Over 8760 hours, the sensitivity of AT and AF to the combination is greater than to either ONC or OMOM, but the sensitivity of OT and OF to the combination is the same.

The results of the sensitivity testing show that for short runs the sensitivity of the model to small (1%) changes in parameter values was not high. By showing parameters or initial values with a sensitivity greater than 1 per cent, areas for further investigation were noted.

The sensitivity of AT and AF over 500 hours indicates the importance of ammonia production rates, the equation used to describe nitrification and the size of filter, and these are also seen as important areas for further

study. The weight of fish is insensitive to most parameters and in any further development of the model a better growth model should be pursued.

### 5.2.3 Conclusions

The techniques developed by Stone for the analysis of System Dynamics models proved useful in ensuring that there were no inconsistencies in the internal structure and behaviour of the model. The modes and pulse-like behaviour identified by the analysis presents no difficulty with regard to the reference system. The main area of difficulty concerns the sensitivity of the model to changes in mode. Much of the sensitivity comes from the need to use precise values in the switching mechanism. The use of transitions rather than precise values would perhaps reduce sensitivity and be a better representation of the reference system. Sensitivity over short runs was not high where there was limited switching of modes. The higher sensitivities over longer runs would present difficulties if the model was used for prediction. For the use of the model as an aid to understanding the behaviour of the reference system this does not present a problem.

### 5.3 Validation

Van Horn (1969) defined validation as "the process of building up an acceptable level of confidence that an inference about a (similar) process is a correct or valid inference for the actual process". The determination of an acceptable level of confidence is subjective; validation cannot prove a model to be correct and very rarely shows a model to be incorrect. In most cases validation serves to indicate the "degree of validity" of the model (Shannon, 1975). One of the most frequently adopted methods for increasing confidence placed in the predictions of system behaviour by a model is to compare the output of the model with the actual behaviour of the system using, if possible, identical inputs. This method was adopted here. Data was collected from three recirculating systems run with a range of fish weights and ration levels. Numerical and graphical comparisons were then made between simulated and measured results.

### 5.3.1 Method

The most informative comparison between the model and the reference system can be made when the system approaches its carrying capacity. In the model this is reflected by a change of mode and an increase in sensitivity. Stocking density and ration level in each run was therefore influenced by the occurrence of missed feeds and the levels of oxygen and ammonia in the preceding runs. Ration level and weight of fish stocked in the first run were based on experimental series II when carrying capacity was reached with 9.1 kg of fish fed approximately 200g/day.

Table 5.7 shows the number and weight of fish, ration level, temperature, circulation rate and the length of each experimental run. For calculation of feed an FCE of 0.4 was assumed throughout. In each system the tank concentrations of ammonia and oxygen were measured daily, and for the first four runs, a weekly measurement was made of the ammonia and oxygen levels every 1-2 hours. Circulation rate and pH were recorded each week.

The recirculating systems used were the same as those described earlier (3.2.2) but because of difficulties with pump maintenance, new pumps had been installed with maximum flow rates twice those of the original pumps. Prior to the start of the experiments, all three systems were cleaned and fully conditioned.

Table 5.7 Conditions for each run

Trial	Run	Tank	Weight of fish (g),	No. of Fish	Ration (%)	CR (l/h)(°C)	Temp.	Length of run (days)
1	1	1	9010	126	2	1710	25.0	29
2	1	2	9000	139	2	1710	23.2	29
3	2	1	11980	138	2	1656	24.3	20
4	3	3	750	19	12	1728	23.4	29
5	4	2	7070	77	3	1617	25.1	20
6	4	3	7110	71	3	1628	24.8	20
7	5	1	11100	92	3	1638	24.5	10
8	5	2	11100	88	3	1628	25.3	10
9	5	3	11130	103	3	1704	23.0	10



Simulation runs to predict concentrations of ammonia and oxygen in each system were initialised using data presented in Table 5.7. Initial values for the levels of ammonia and oxygen were taken from the first measurements in each run.

Analysis of the measured and simulated results using statistical tests of "goodness of fit" were considered but rejected because of possible autocorrelation. Spectral analysis does not depend on the statistical independence of the generated points and has been proposed as a means of objective comparison of time series data generated by a computer model with observed time data. However, the data should be Gaussian and have no trend in the mean or variance of the series (covariance stationery) and a large number of observations are required. Because of these constraints, spectral analysis has only limited use with simulation models (Van Horn, 1969; Shannon, 1975). Analysis of the results was therefore, confined to visual and numerical comparisons.

### 5.3.2 Results

A visual comparison was made between the levels of ammonia and oxygen recorded in the laboratory and those predicted by the model (Fig. 5.11 to 5.19). Due to scaling in some places it is difficult to distinguish between the time intervals for the observed results. A key to the graphs is presented in Table 5.8. The maximum, minimum and mean concentrations of ammonia and oxygen in each run have been calculated and are presented in Table 5.9.

Table 5.8 Key to the symbols and colours used in Figs 5.11 - 5.19

Variable	Colour	Symbol (measured result)	Line (predicted result)
Ammonia in tank	Black	◻	_____
Ammonia in filter	Blue	○	_____
Oxygen in tank	Red	△	_____
Oxygen in filter	Green	◇	_____

Because of the scales used, the predicted levels of ammonia in tank and filter appear as the same line, although the slight differences between them are indicated in Table 5.9.

#### 5.3.2.1 Ammonia

The range of values predicted by the model generally lay within the range measured in the laboratory. The ammonia levels predicted by the model showed stable and regular diurnal variations in both tank and filter. After initialisation of each run, the ammonia concentration rose from the initial value recorded in the laboratory to the level of 0.2 mg/l as determined by the 'MAX' function incorporated into the equation for AT.

The pre-feed concentrations (PFC's) of ammonia predicted by the model were in general higher than those recorded in the laboratory (Table 5.9), but the predicted diurnal variations were over an order of magnitude lower and a different shape. The height of peak reached after feeding increased during each run. For example in run 3 the predicted daily range in concentration increased from an initial 0.08, to 0.15 mg/l at the end of the run. The measured range was 2 to 10 times greater, increasing from 0.15 to 1.6 mg/l. In the laboratory the maximum concentration was reached 6 to 7 hours after feeding, falling almost to the PFC by 12 hours. The model predicted a maximum concentration approximately 9 hours after feeding and the PFC almost reached by 24 hours. Failure to reach the PFC after 24 hours resulted in a daily increase in PFC.

A daily increase in PFC was predicted by the model in all five runs. Following the first feeds in each run, the model predicted a rapid rise in PFC. In runs 1, 3, 4 and 5 this gave way to a more gradual upward trend. In run 5 this upward trend resulted in a PFC greater than 1 mg/l in the tank and food was withheld (Figs. 5.17 - 5.19). In run 4 the initial daily increase in ammonia suggested that a PFC greater than 1 mg/l would be reached after 10 days operation, but because of the decline in the daily increase in PFC, carrying capacity was not reached by day 20 (Figs. 5.15 and 5.16). In run 2 the daily increase in PFC started to decline but a PFC greater than 1 mg/l was reached after 6 days and food was withheld. Subsequent to this the daily rise in PFC (in run 2) increased as the run progressed, resulting in a changing pattern of feeding/not feeding (Fig.

5.13). In the laboratory the tank ammonia levels showed a steady increase in PFC's, except in run 5 where carrying capacity was exceeded.

The daily increases in PFC recorded in the laboratory were less regular than predicted, being in part influenced by errors in measurement (Appendix 2). A consideration of the errors in the model's predictions is presented later in section 5.4.3.1.

The concentrations of ammonia measured in the filter effluent were generally lower than those in the tank, but as found in the first experimental series (Fig. 3.10) the filter concentration occasionally showed a more rapid increase after feeding than the tank concentration (runs 2, 3 and 4). The maximum concentration reached in the filter effluent was never as great as that in the tank and tended to level off rather than peak. The model predicted a slightly lower concentration in the filter than in the tank, with both tank and filter following the same pattern of diurnal variation.

When more than one tank was used per run, it was not possible to ensure that the conditions in each tank were identical (Table 5.7). In run 4, where the weight of fish, water temperature and circulation rate differed between tanks by approximately 1 per cent, no difference was predicted in the ammonia levels for tank 2 and 3 (Table 5.9). Differences between tanks in the weight of fish, temperature and circulation rate were larger in runs 1 and 5 (up to 9 per cent) with the result that the model predicted small differences in ammonia levels (Table 5.9). In run 5 this had a particularly marked effect, since the differences resulted in different rates of increase in PFC in the three tanks. This led to feed being withheld in tank 1 after 7 days, after 10 days in tank 2 and on days 6 and 10 in tank 3 (Figs. 5.17 - 5.19).

In run 2 the model predicted that carrying capacity would be reached after 6 days. As noted earlier, this had a marked effect on subsequent predicted ammonia levels. Although a concentration of 0.96 mg/l was recorded in the laboratory, the PFC did not reach 1 mg/l and therefore, unlike in the model, no feeds were withheld.

Whilst carrying capacity was not reached in the laboratory during run 2, the increased weight of fish did increase the range of ammonia concentration recorded compared to that in run 1 (Table 5.9). As predicted by the model, carrying capacity was reached in run 5, although the agreement between simulated and measured results was not good. This was in part due to the effect of feeds being withheld after different time intervals which put the rest of the results "out of step".

### 5.3.3 Oxygen

The range of values predicted by the model generally lay within the range recorded in the laboratory. The oxygen levels predicted by the model showed stable and regular diurnal variations in both tank and filter, with the variations in the tank generally 2/3rds of those in the filter. After initialisation neither tank nor filter concentrations regained 100 per cent saturation, and in runs 1, 3 and 4 a steady decrease with time was predicted. In runs 2 and 5 this steady decline was interrupted by a sharp increase in concentration when food was withheld, followed by a sharp decrease when feeding was resumed. In addition, the amplitude of the diurnal variations predicted in runs 2 and 5 decreased with time, whereas in runs, 1, 3 and 4 the amplitude of the diurnal variations increased.

In all runs a consistently lower concentration was predicted in the filter compared to that in the tank, because of oxygen consumed in nitrification (ORNA). As the weight of fish increased during each run, so the differences between tank and filter concentrations increased.

Compared to the predicted oxygen concentrations, those recorded in the laboratory showed greater variation. This was partly due to variations in temperature in the laboratory (Table 5.7) which affected oxygen solubility. Except in run 4, no general trends were discerned by eye in the concentrations of oxygen recorded in the tank. Oscillation in the measured concentrations of oxygen in the tank were noticed in all runs, and were pronounced in run 1, where the periodicity of the oscillations appeared to be 4 - 7 days.

As predicted the diurnal variation in oxygen concentration measured was greater in the filter than in the tank, although the range predicted was

considerably less than that measured. For example, in run 4 the diurnal ranges of concentrations predicted were approximately 0.5 and 0.8 mg/l in tank and filter respectively, while the ranges measured in the tank and filter were 2.4-3.3 and 2.4-5.2 mg/l (tanks 2 and 3 respectively). During run 4 the oxygen concentration in the filter fell to 0.83 and 0.73 mg/l in tanks 2 and 3 respectively. Although this was higher than the value of the switches used in AL (0.7 mg/l) it is likely that some inhibition of nitrification was taking place. A similarly low concentration was also recorded in run 2. This was not predicted by the model.

Differences in weight of fish, circulation rate and temperature between tanks in run 1, 4 and 5 influenced the range of concentrations predicted in tanks and filters (Table 5.9). Temperature differences had a particularly marked effect by influencing the solubility of oxygen. In the measured results it is not possible to distinguish this from other sources of variation.

Table 5.9 A comparison of maxima, minima and means

	AT			AF			OT			OF		
	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean
Run 1 S	0.72	0.20	0.52	0.71	0.20	0.52	7.88	6.92	7.24	7.67	5.14	5.76
Tank 1 O	4.20	0.08	1.10	3.40	0.10	1.71	9.08	4.67	6.88	6.40	1.50	3.40
Run 1 S	0.81	0.20	0.58	0.79	0.20	0.58	8.19	7.16	7.50	8.00	5.31	5.98
Tank 2 O	3.40	0.06	0.71	3.20	0.06	1.06	8.99	6.11	7.67	7.01	2.92	4.95
Run 2 S	1.28	0.20	0.85	1.26	0.20	0.84	8.19	6.18	6.77	7.77	3.17	4.89
Tank 1 O	4.80	0.10	1.49	4.00	0.44	2.76	8.08	4.27	6.83	3.80	0.74	2.35
Run 3 S	0.52	0.20	0.34	0.51	0.20	0.34	8.14	7.59	7.87	7.94	6.33	6.95
Tank 3 O	2.00	0.06	0.47	1.76	0.06	0.70	9.08	6.92	7.97	7.27	2.23	5.55
Run 4 S	0.93	0.20	0.64	0.92	0.20	0.63	7.84	6.36	6.94	7.61	3.83	4.97
Tank 2 O	7.50	0.14	2.23	5.00	1.40	3.30	7.53	4.30	6.16	2.58	0.83	1.83
Run 4 S	0.93	0.20	0.64	0.92	0.20	0.63	7.89	6.42	6.99	7.66	3.94	5.05
Tank 3 O	5.20	0.14	1.27	2.60	0.36	1.74	7.96	4.62	6.79	5.94	0.73	2.98
Run 5 S	1.27	0.24	0.85	1.25	0.23	0.84	7.94	6.30	6.75	6.62	3.39	4.24
Tank 1 O	3.20	0.16	0.93				7.97	6.23	6.80			
Run 5 S	1.20	0.29	0.90	1.19	0.28	0.89	7.69	6.18	6.50	5.86	3.28	3.80
Tank 2 O	14.00	0.20	4.75				7.09	5.73	6.29			
Run 5 S	1.06	0.24	0.82	1.04	0.23	0.81	8.20	6.73	7.07	6.85	4.08	5.30
Tank 3 O	14.00	0.16	4.52				7.93	6.77	7.41			

S = simulated  
O = observed

Fig. 5.11 RUN ONE : TANK ONE

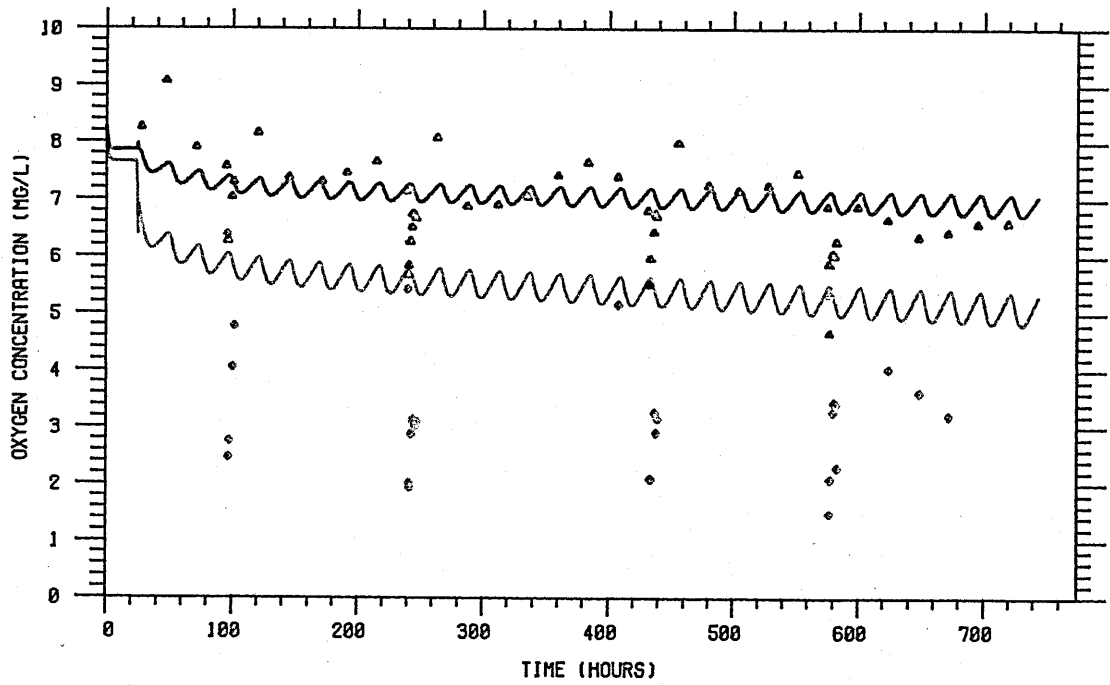
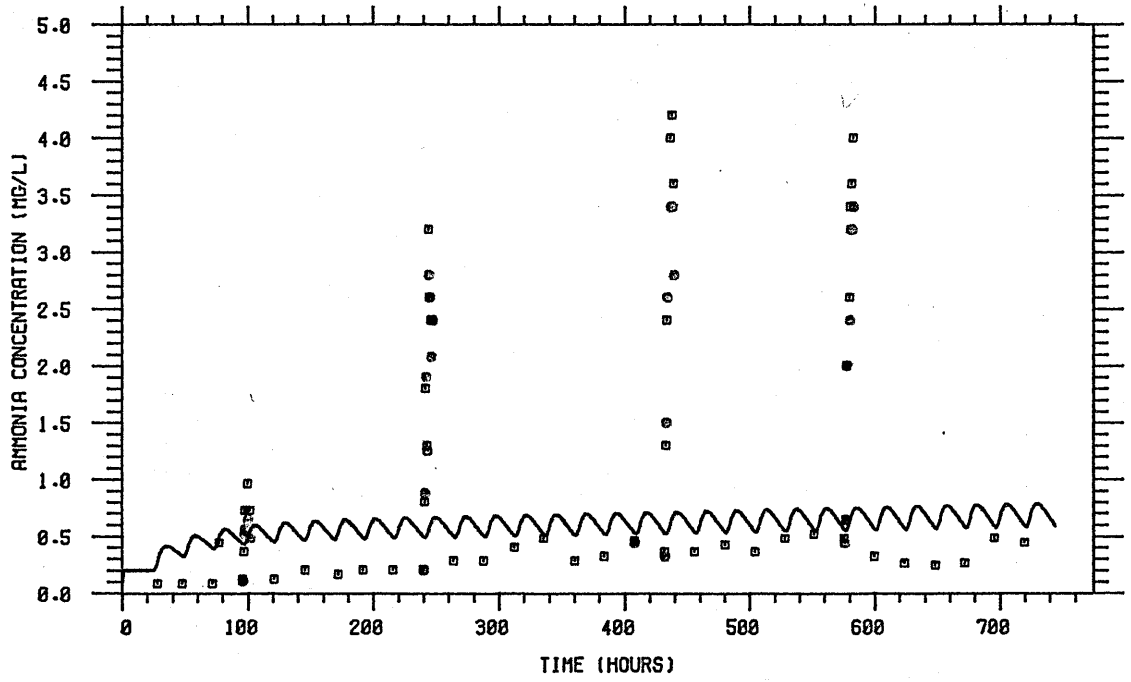


FIG. 5.12 RUN ONE : TANK TWO

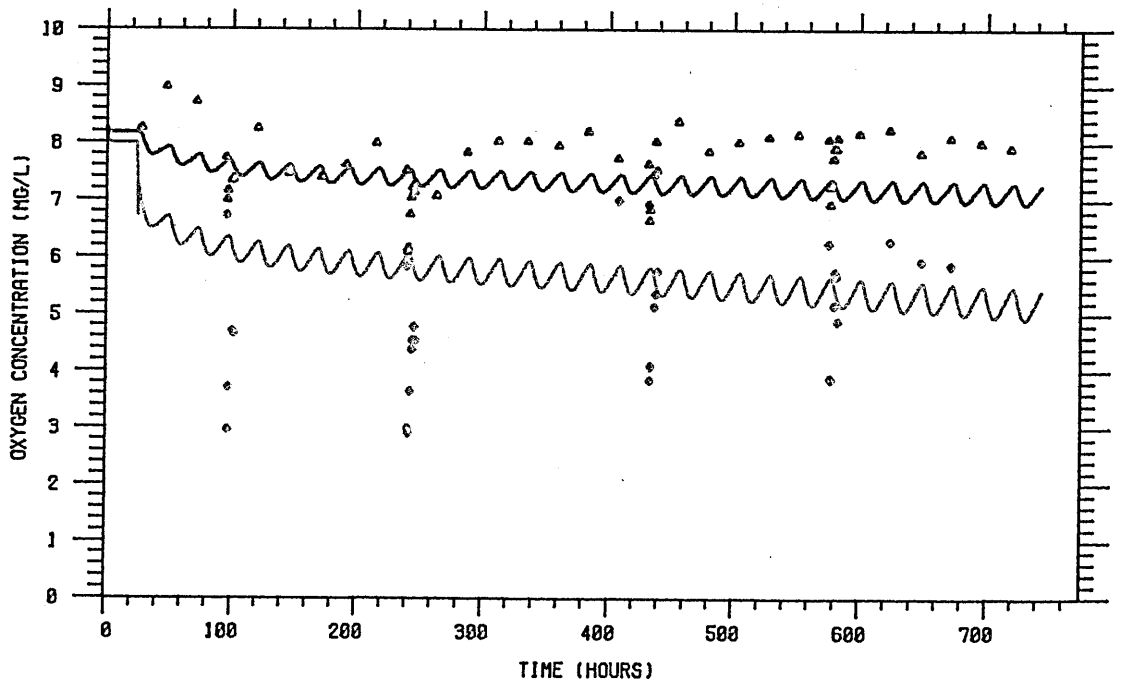
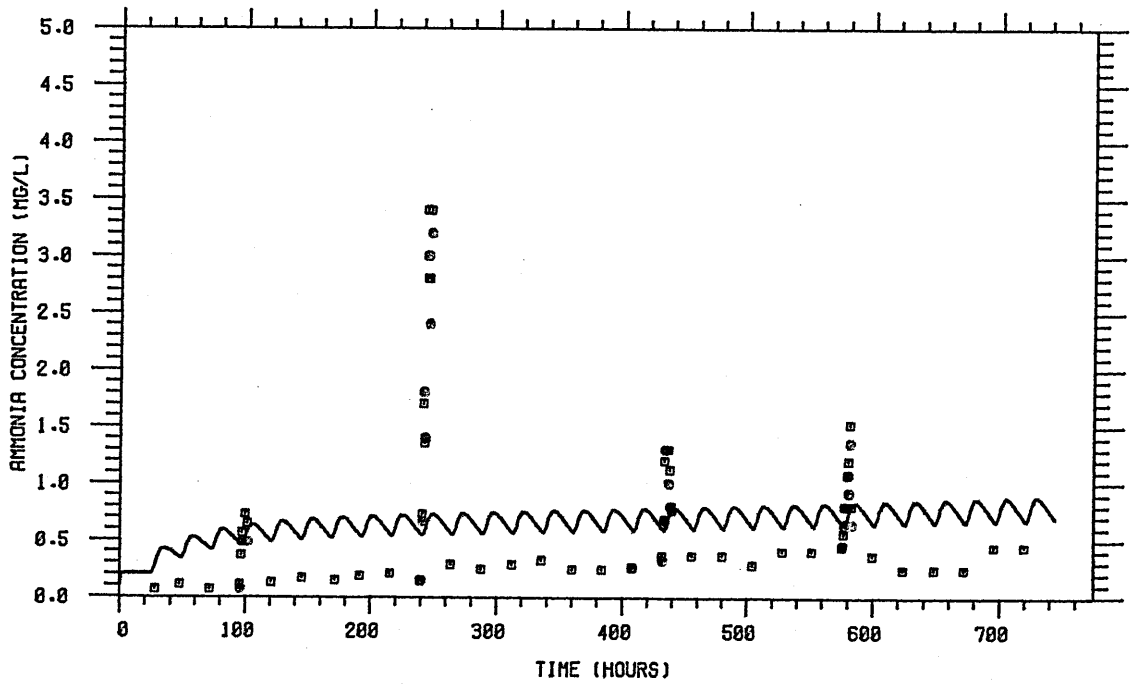




FIG 5.13 RUN TWO: TANK ONE

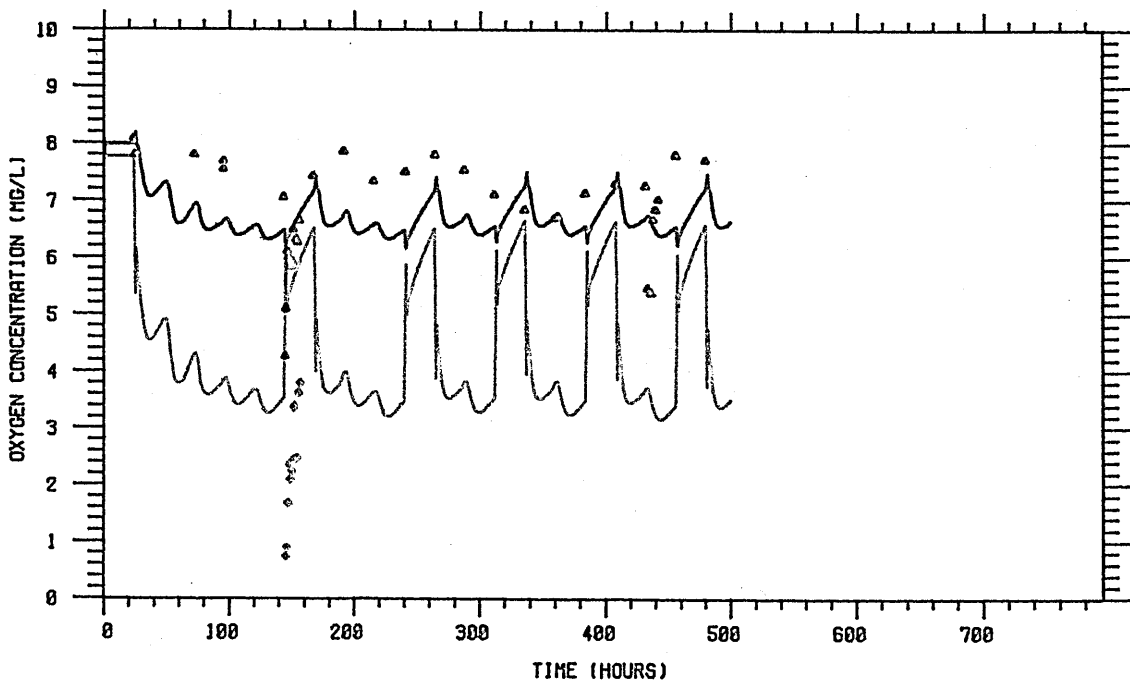
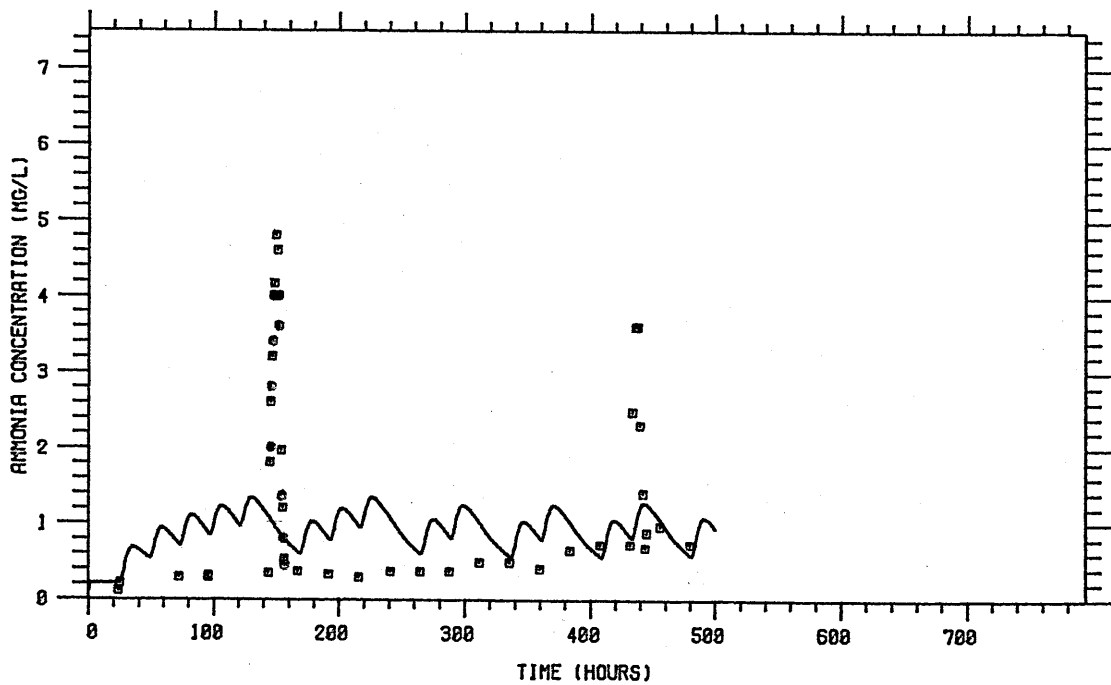


Fig 5.14 RUN THREE: TANK THREE

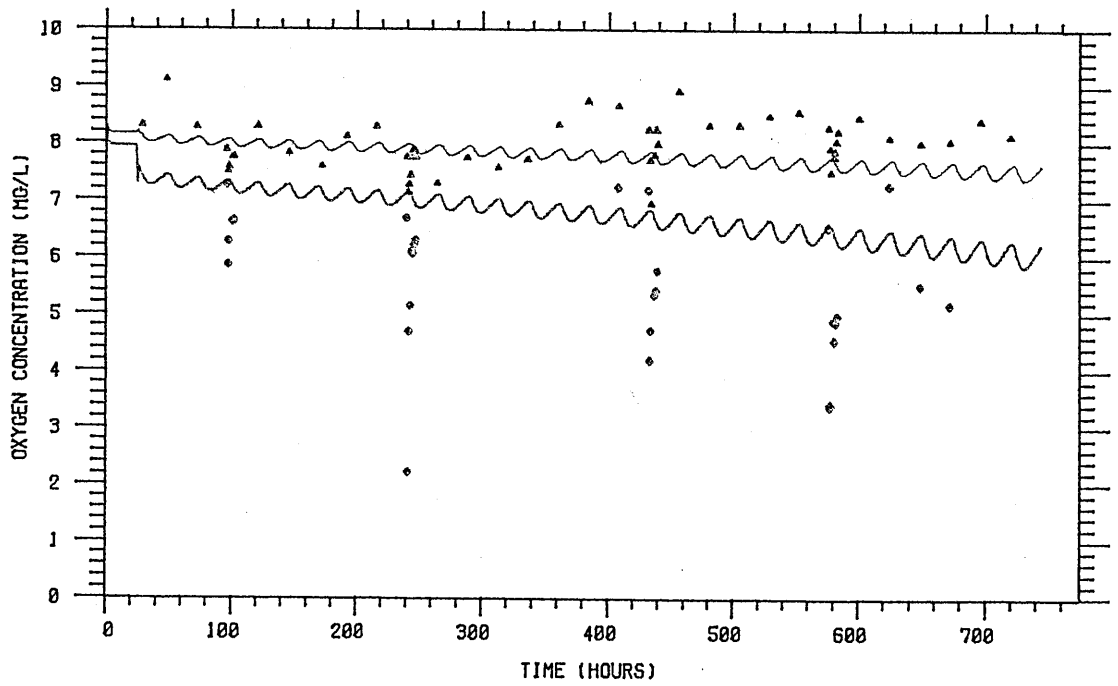
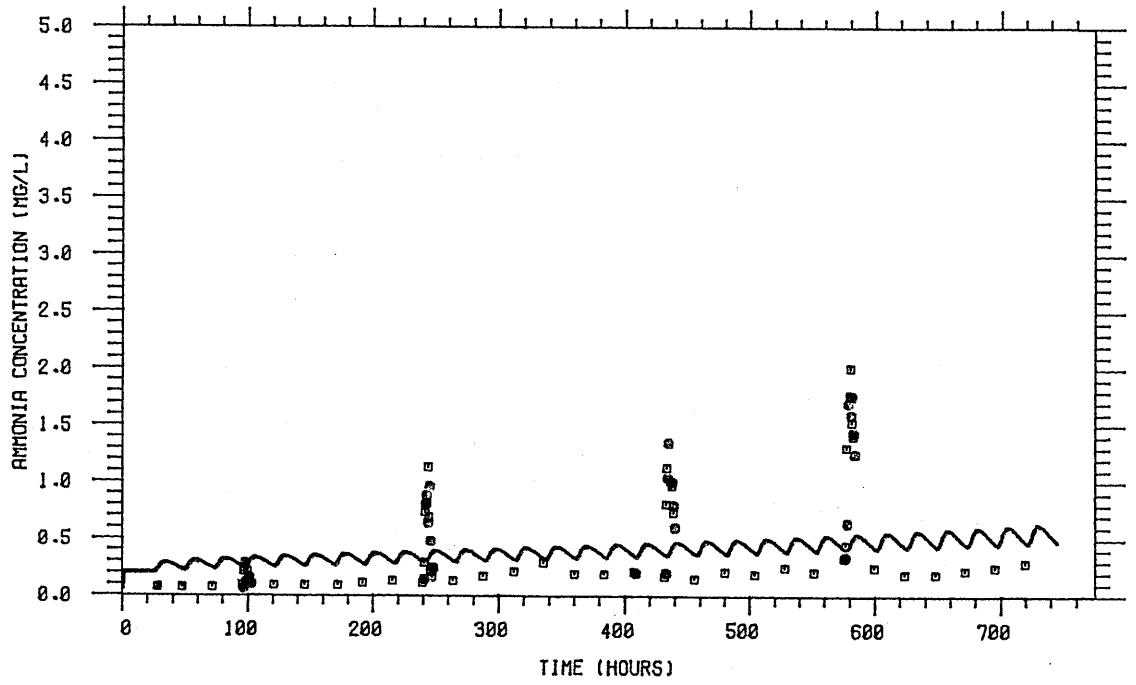


Fig 5.15 RUN FOUR : TANK TWO

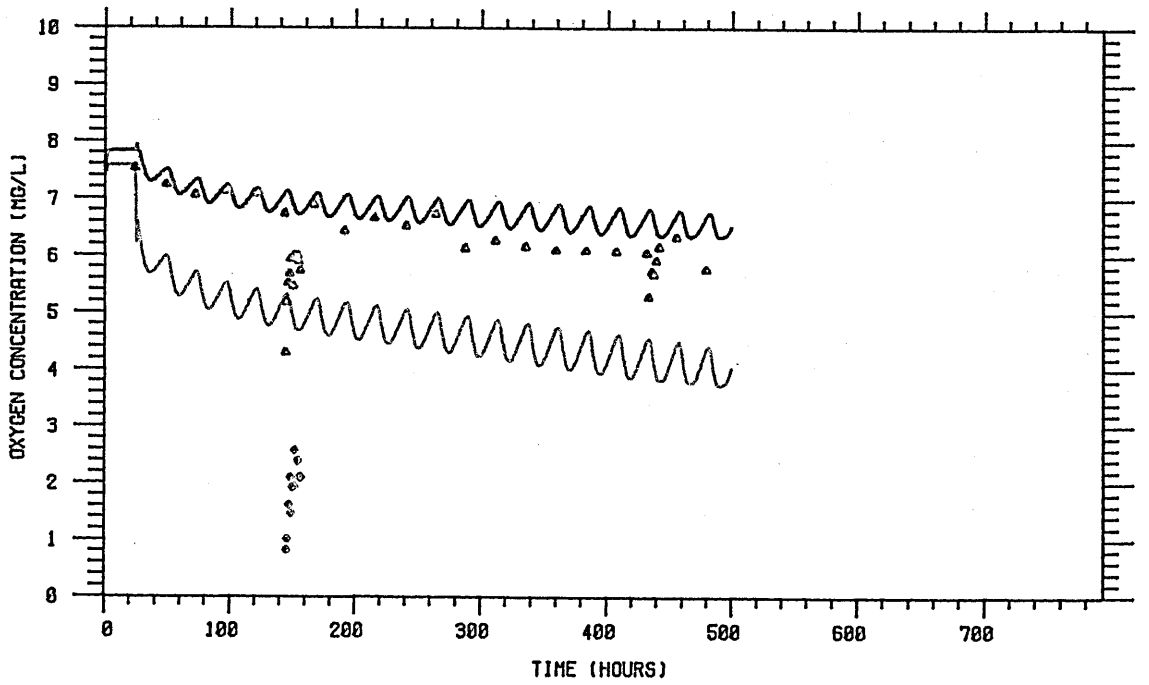
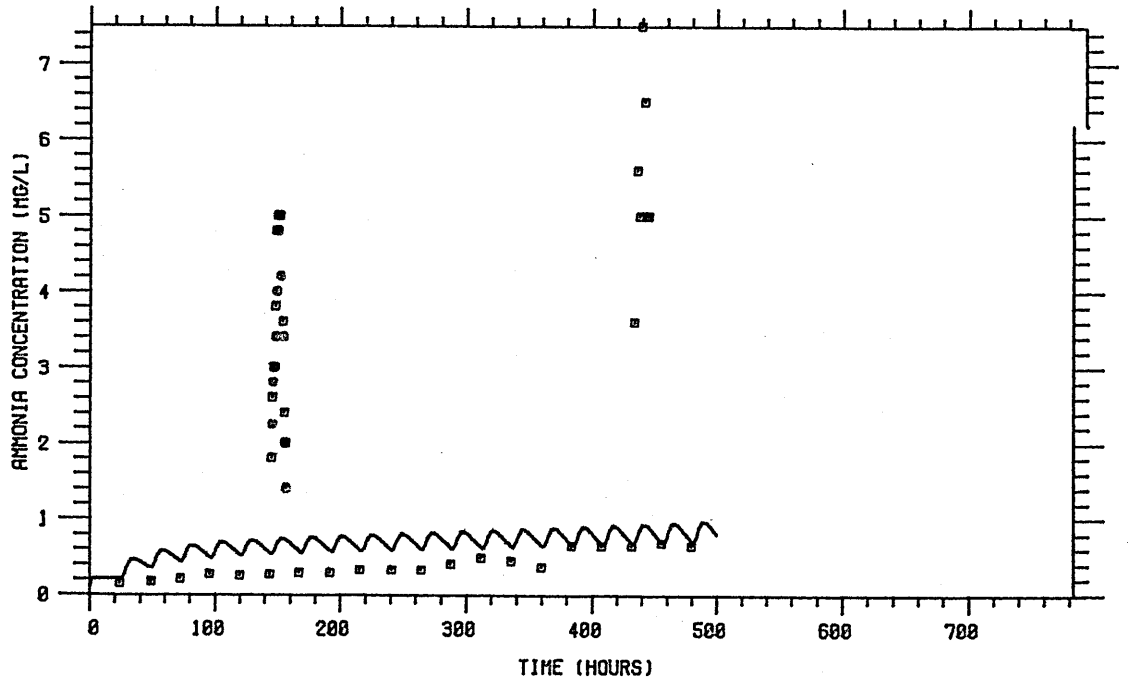


FIG 5.16 RUN FOUR: TANK THREE

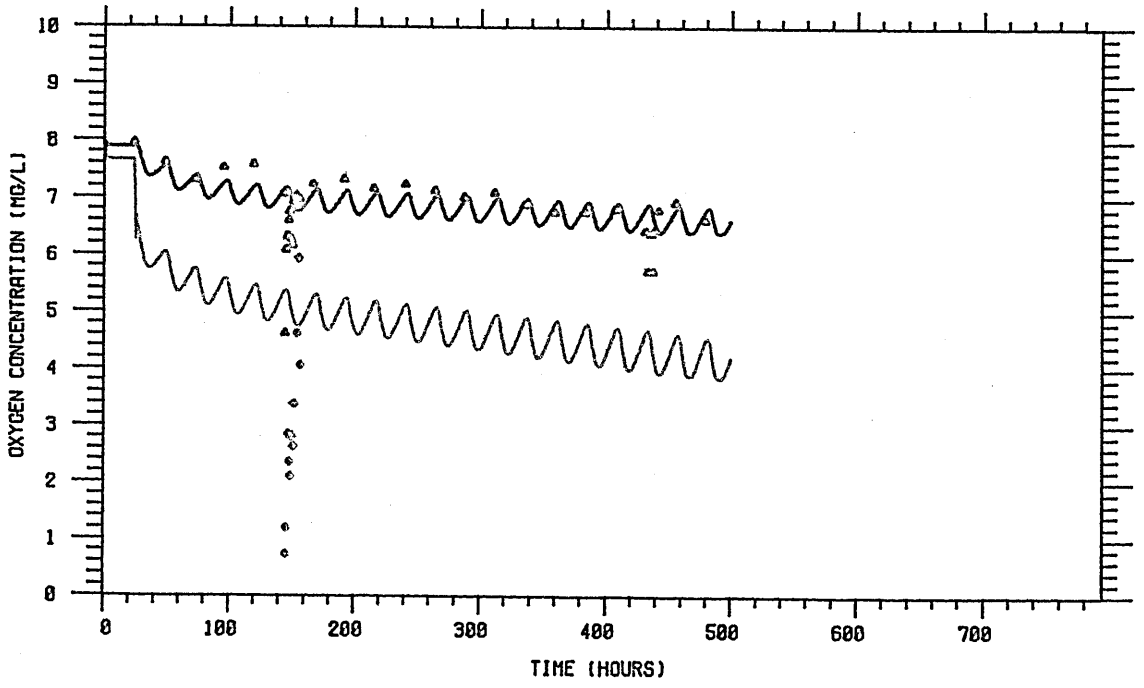
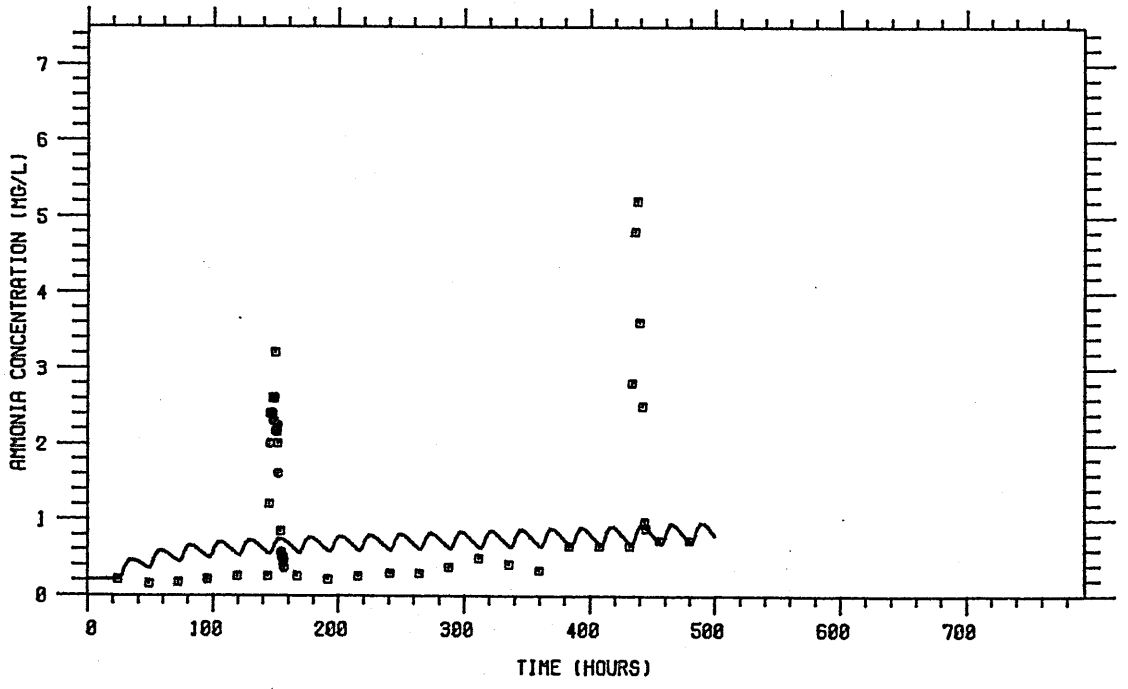


FIG. 5.17 RUN FIVE : TANK ONE

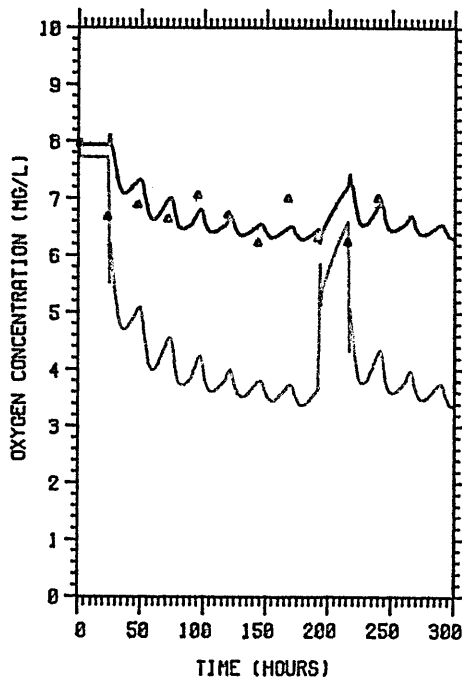
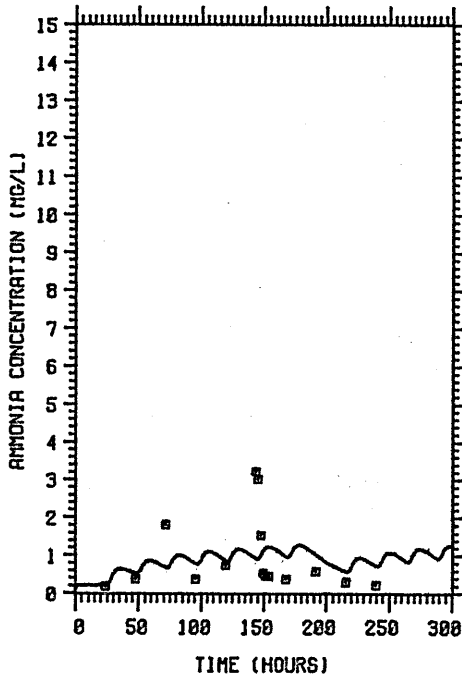


FIG. 5.18

RUN FIVE: TANK TWO

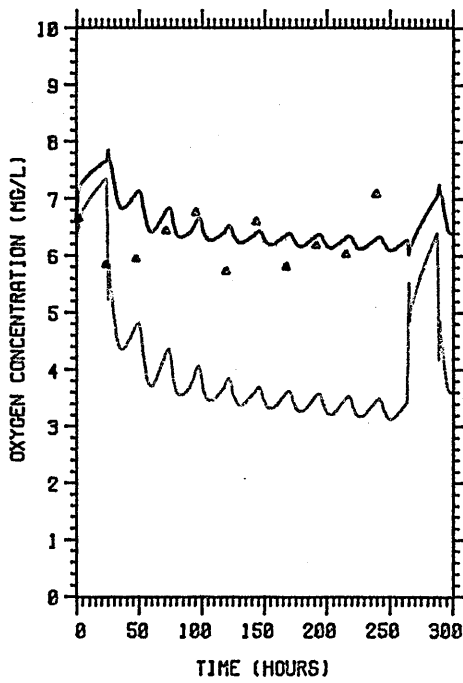
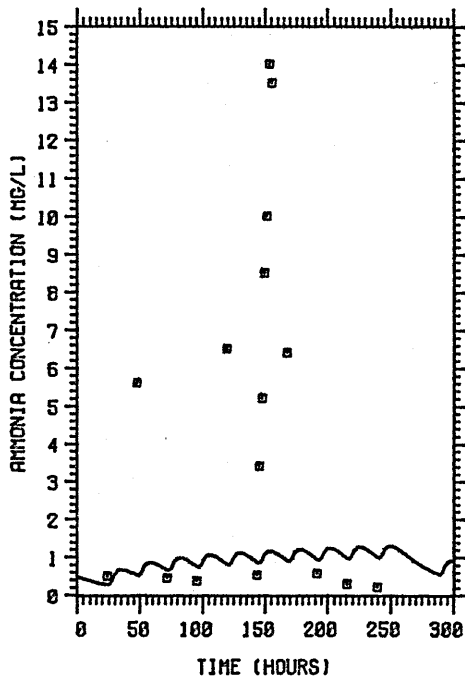
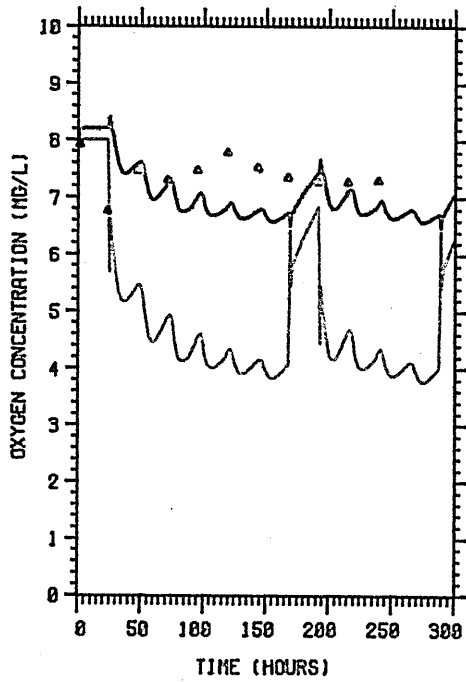
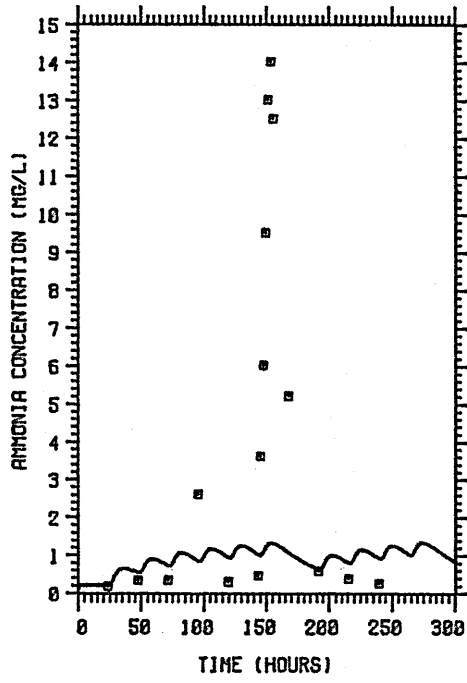


Fig. 5.19 RUN FIVE: TANK THREE



#### 5.3.4 Discussion

Sensitivity analysis indicated an increase in the sensitivity of the model when changes in mode occurred (5.2.2.3). For a satisfactory evaluation of the model, it was important to compare the predictions by the model with the actual behaviour of the system over a range of initial conditions sufficiently wide for the effects of mode change to be examined. Since changes in mode occur as the loading on the system reaches carrying capacity, each run was designed to reach carrying capacity.

Only in the fifth run did PFC's of ammonia exceed 1 mg/l. Carrying capacity was predicted by the model to be reached in runs 2 and 5, but in run 2, although a PFC of 0.96 mg/l was recorded in the laboratory, no food was withheld. As indicated in Appendix 2, the error attached to an ammonia measurement of 0.96 mg/l is approximately + or - 0.09 mg/l. It is therefore possible that the concentration in the tank was greater than 1 mg/l. This emphasises the conclusion drawn in section 6.2.3, that one of the limitations of the model is the use of discrete switching values as these increase the sensitivity of the model and make validation more difficult. The accuracy of the predicted results is considered further in section 5.4.3.1.

The sensitivity of the model to variations in circulation (CR) rate and temperature (T) was reflected in the difference in predicted concentrations where more than one tank was used to a run. The results presented here differ from the sensitivity analysis in that small differences in circulation rate and temperature (as in run 4) affected only the predicted oxygen concentrations, while with larger differences (as in runs 1 and 5), both ammonia and oxygen levels were affected. Sensitivity analysis indicated that ammonia levels were more sensitive to changes in CR and T than were the oxygen levels (Table 5.3). No explanation is available for this disparity apart from the possibility that the effect of the combined differences in CR, T, W and FC differed from the effects of these factors considered separately as in the sensitivity analysis. In the laboratory, variations in the concentrations of oxygen and ammonia recorded during each run were too large for differences between tanks to be distinguished.



In the model runs carrying capacity was largely determined by the weight of feed and therefore weight of fish (W) and ration level (FC). Carrying capacity in the laboratory appeared to be influenced not only by weight of feed but also by the "previous history" of the system, i.e., for how long the system has been operated previously and under what conditions. For example, in all three tanks in run 5, the last three measurements of ammonia showed a decrease, compared to the increases and sharp peaks experienced at the start of the run. In run 2, although carrying capacity was predicted by the model, the pre-feed ammonia concentrations in the reference system did not exceed 1 mg/l. One explanation could be that since run 2 followed run 1 almost immediately, the filter was more fully established and better able to adapt to the increases in loading. It also appears that when the loading on a filter is increased slowly, a higher level of loading can be accommodated than when the increases in loading are more rapid. However, problems of clogging may be increased. Since the model is autonomous, previous history cannot be taken into account, unless a function is included in the model to simulate adaption by the filter according to the rate at which loading increases. The possible form of such a function is unclear, since results do not indicate which mechanisms are involved. At present the model only responds to changes in rate of load increase in so far as this affects the weight of feed. This is clearly an important area for further investigation.

Further support for adaptation to loading is found in the measurements of oxygen and ammonia. During both experimental series I (Figs. 3.14 and 3.15) and the validation experiments (particularly runs 1, 2, 3 and 4) the pre-feed concentrations of oxygen and ammonia appeared to oscillate rather than follow a clear trend. One explanation is that these oscillations reflect the adaptation of the filter to a continuing increase in load. Whilst it is uncertain which biological phenomenon they could represent, one possibility is bacterial growth, with the variations in ammonia concentration reflecting growth of the nitrifying bacteria and variations in oxygen concentration reflecting growth of both nitrifying and heterotrophic bacteria. The longer regeneration period and slower growth of nitrifying bacteria could explain the longer frequency of oscillation in the ammonia concentrations as compared with those in the oxygen concentrations.

Similar oscillations have been observed by Forster (pers. comm.) and were found to be the result of physical disturbance of the filter population during backwashing. During the validation runs the laboratory recirculating systems were not physically disturbed, but the environmental conditions were subject to great changes. It is uncertain what effect the regular diurnal variations had on the bacterial populations. One possibility is that a population develops which is sufficient to treat high inputs, and is maintained by lower levels. This could account for the lag in response to ammonia production.

In general, the agreement between the measured and predicted pre-feed concentrations of ammonia in tank and filter was good. It is probable that the agreement could be improved by calibration of the model, that is, adapting weak or unknown parameters or relations to reach the best overall agreement (van Keulen, 1976). For example, the model uses a 'MAX' function in the equation for AT (the level of ammonia in the tank) to set the minimum concentration of AT to 0.2 mg/l. This function was included since daily measurements of ammonia during experimental series I and II indicated that in an established system, the concentration at its lowest did not fall below 0.2 mg/l. Prior to the validation experiments, the three recirculating systems were given a thorough clean to remove any accumulated solids. The measurements made during the validation runs suggest that as a result of cleaning, the use of 0.2 mg/l as a minimum was too high. This would only have affected the PFC's at the start of each run, since in all runs the PFC quickly rose with feeding to above 0.2 mg/l.

The agreement between the measured and predicted pre-feed concentration of oxygen in the tank was also good. When the model was originally built it was intended to maintain the tank concentration of oxygen at 100 per cent saturation through the use of OR. However, although the arcs from ORFR to OT and OR, and from OR to OT are time-sliced, the arc from OT to OR is event sequenced (Fig. 5.9). This means that the replenishment of oxygen in the tank occurs one time step after it has been removed for nitrification (ORR), oxidation of organic matter (OROG) and fish respiration (ORFR). This appears to be a reasonably adequate representation of the reference system. Agreement between the predicted and measured oxygen levels in the filter was not as good, and this was noted as an area for future

investigation.

A sharper decrease in oxygen concentration after feeding was recorded in the laboratory than was predicted by the model. This may have been partially responsible for the more rapid increase in the ammonia in the filter than in the tank, occasionally recorded during runs 2, 3 and 4. A lowered oxygen concentration in the filter may have resulted in some secondary ammonia production. The measurements made in the laboratory, indicated that after the initial sharp decrease in oxygen concentration, the levels steadily rose. As the fish excreted ammonia as a result of feeding, the oxygen in the filter would have risen for nitrification to take place, thus preventing the ammonia concentration in the filter reaching the same level as in the tank.

The agreement between the predicted and measured diurnal variations in oxygen and ammonia concentration was not good, with the model predicting (less skewed) peaks and of an amplitude often an order of magnitude less than those measured. There are a number of possible reasons for these dissimilarities.

In order to model diurnal variations in oxygen and ammonia it is necessary to have data concerning changes which occur over 24 hours for all the associated variables. During construction of the model only data concerning ammonia production was available. The growth of fish and oxygen consumed in fish respiration were modelled as step changes, while the production and oxidation of organic wastes were modelled as a continuous process. The data concerning ammonia production were based on Salmonids. It was uncertain whether this should be used, as carp digestive tracts are quite dissimilar, and carp may have different assimilation rates and excretion patterns but no other information was available. In the model ammonia production rate is maximal 4 hours after feeding, 2 hours earlier than found by Brett and Zala (1975) and Kaushik (1980). The maximum concentration predicted in the tank was at 9 hours after feeding. In the laboratory the maximum tank concentration was recorded after 6 hours. This suggests a maximum ammonia production rate in the laboratory less than 6 hours, perhaps less than 4 hours, after feeding.

The diurnal variation in oxygen concentration predicted by the model is almost a mirror image of that predicted in ammonia concentration. Thus in the model the minimum oxygen concentration is reached 9 hours after feeding, while in the laboratory a minimum concentration occurs in the tank and filter 1-3 hours after feeding. In the laboratory it is clear that the respiration rate is not continuous, but increases with activity at feeding time, reducing to a basic rate during the night.

No information is currently available concerning diurnal variations in faecal production rates. Some solids arise from uneaten food and the movement of these solids into the filter following feeding likewise needs to be investigated.

The maximum and minimum concentrations of ammonia and oxygen predicted by the model differed by up to an order of magnitude from those recorded in the laboratory. Since the oxygen concentration predicted by the model mirrored the ammonia concentration, the production and removal of ammonia was further investigated.

The model was operated for run 1, tank 1 with no removal of ammonia taking place. The concentration of ammonia increased by 0.33 mg/l per feed at the start of the run. In the laboratory the concentration increased to a maximum of 1 mg/l after a single feed (4 days after the run was started) and later in the run to a maximum concentration of 3-4 mg/l. It was therefore concluded that the model predicted too little ammonia production. The amount of ammonia produced was determined as a percentage of the weight of food. Obviously this is a simplification with ammonia production not constant.

Ammonia production was increased from 3 per cent of the weight of food to 30 per cent. When the model was run with no removal of ammonia, the tank concentration rose to a maximum of 3.46 mg/l. When run 1, tank 1 was run with CA = 0.3, it almost immediately resulted in missed feeds and ammonia increased from 0.2 to 3.02 mg/l after 38 hours decreasing to 2.80 mg/l after 48 hours, 1.98 after 72 hours, 1.18 after 98 hours to 0.72 mg/l after 120 hours, at which point feeding of the fish recommenced, and with it an increase in ammonia. The agreement between predicted and measured diurnal

variations was therefore improved by this simple modification to the constant CA.

The model assumes that in the reference system the water flows evenly through the filter bed and thus, dividing the filter water volume by the circulation rate, gives the retention time of water in the filter. In practice, however, channelling of flow occurs, with considerably reduced flow in certain zones of the filter. According to Liao and Mayo's (1974) equation describing nitrification in a filter (4.5.2.1), this will effect the efficiency of ammonia removal since nitrification increases with longer retention times. Their equation was developed for retention times between 0.21 and 0.46 hours. In the standard version of the model (4.8) retention time was approximately 0.15 hours, which although outside the range of Liao and Mayo, was not considered sufficiently different to invalidate use of the equation (Liao, pers. comm.). In the validation experiments circulation rate was considerably higher, with retention time reduced to 0.08 hours. With short circuiting through the filter, retention time of water passing along well defined channels will be considerably less. For example, if 80 per cent of the flow passed through 20 per cent of the bed, retention time would be only 0.02 hours, while in the rest of the bed retention time would be 0.31 hours. Whilst the latter value falls within the range of Liao and Mayo's equation the former value is much lower than their minimum value and it is uncertain whether the equation would still apply.

Another difficulty with the equation used for simulating ammonia removal is that it was developed using data from systems where the concentration of ammonia was 1 mg/l or less (Kramer, Chin and Mayo, 1972). During the validation experiments, measurements of diurnal variations indicated that concentrations over 1 mg/l were reached just a few hours after each feed. In addition as the ammonia concentration nears the value of the saturation constant ( $K_s$ ) ammonia removal becomes less dependent on influent concentration. At 25°C  $K_s = 3.5\text{mg/l}$  (Ulken, 1963). Thus ammonia removal in the laboratory at higher concentration is lower than that predicted by the model. This again questions the suitability of using Liao and Mayo's equation to simulate nitrification under these experimental conditions.

Short-circuiting in the filter may have had an effect on the diurnal variations. Since the main flow passed directly through the filter, there would be little ammonia removal and therefore little buffering against the sharp rise in ammonia production which follows feeding (see 4.5.1). (This would in part account for the sharp rise in ammonia concentration measured in the reference systems). The slower flow through the main part of the filter would result in high nitrification rates, but because of the longer retention time, high ammonia concentrations could have developed before any effects of this treated water would become noticeable. In addition, once high concentrations had developed, oxygen may have become limiting since in the slower flowing zones of the filter there was less frequent oxygen replacement, this being dependent on water flow. Once the concentration started to fall, the rate of decline would increase, perhaps one reason why the observed diurnal variations showed a more rapid decrease in concentration than predicted by the model.

In order to ascertain whether short-circuiting in the filter was responsible for the increased diurnal variation and lower baseline concentrations measured in the reference systems (as compared to the model) modifications were made to the model's structure (5.3.5).

#### 5.3.5 Modification of model structure

The structure of the model was modified in order to simulate short circuiting through the filter. The filter was divided into two sections (A and B), with the proportion of the total filter water volume represented by section B determined by the constant FVB. Section B represents the reduced area through which most of the flow is concentrated. The proportion of the total flow passing through section B is determined by the constant CRB, and the proportion of the flow through section A by the constant CRA. The retention time of water in section A is determined by the new auxiliary function.

$$TMT.K = (FWV * FVA) / (CR * CRA)$$

The previous equation for retention time was modified to represent section B.

$$TM.K = (FWV * FVB) / (CR * CRB)$$

Nitrification in section A of the filter was simulated by the new auxiliary function ANT.K, while nitrification in section B was simulated by modifying the previous equation for nitrification.

$$ANT.K = (0.11 * T - 0.2) * 0.96 * TM.K * AF.K * OM.K * AL.K * FVB * FF$$

$$AN.K = (0.11 * T - 0.2) * 0.96 * TMT.K * AF.K * OM.K * AL.K * FVA * FF$$

In modifying the structure of the model it was necessary to use the constants FVA and FVB in the equations describing nitrification, in order to partition the loading received by each section of the filter. One difficulty in using two equations to describe nitrification is that it is necessary to use the variable AF in both equations. This has the effect of doubling the weight of ammonia to be removed. The introduction of a 'fudge factor' (FF) with a value of 0.5 into the equations describing nitrification, halves the weight of ammonia and overcomes this problem.

The rate of ammonia removal from the system is determined by the variable ARR. In the standard version of the model this is equivalent to the value of AN. This was modified to incorporate the new variable ANT. The effects of ammonia removal in section A of the filter is delayed because of the longer time the water is retained in this section. In section B the retention time is considerably less and ammonia removal has effect more quickly. The difference is simulated in the model by the use of a delay function for ANT. The modified equation of ammonia removal is therefore:

$$ARR.KL = AN.K + DELAY 3 (ANT.K, TMT.K)$$

where the length of the delay is equal to the retention time TMT. This is a very crude representation of the reference system, since the removal of ammonia will also be affected by the time taken by the nitrifying bacteria to respond to the input of ammonia. In addition, since the input of ammonia is not constant, the different retention times in sections A and B will

result in different ammonia concentrations being received by the two sections. Insufficient information was available to model this.

### 5.3.6 Comparison between simulated and measured results using modified model

To ascertain the effects of these modifications on the behaviour of the model, data corresponding to run 1, tank 1 were used to initialise the model. The model was re-run with different values attributed to the constants FVA, FVB, CRA and CRB, (Table 5.10). The results were analysed in terms of the variation in ammonia concentration in the tank, both diurnally and throughout the length of the run, and are presented in Table 5.11.

#### 5.3.6.1 Results

Table 5.10 Values attributed to constants FVA, FVB, CRA and CRB during re-runs

Variable	Number of Run						
	0	1	2	3	4	5	6
FVA	-	.8	.9	.9	.8	.85	.8
FVB	-	.2	.1	.1	.2	.15	.2
CRA	-	.2	.1	.2	.1	.2	.15
CRB	-	.8	.9	.8	.9	.8	.85

Table 5.11 Results of simulated runs using modified model

Run	Range of ammonia concentrations (mg/l) after time t hours				
	t= 25-33	193-201	385-393	601-609	721-729
0	.20-.41	.48-.65	.52-.70	.56-.75	.58-.79
1	.20-.38	.27-.44	.29-.47	.31-.51	.32-.53
2	.20-.29	.20-.30	.20-.31	.20-.33	.20-.34
3	.20-.36	.20-.37	.21-.40	.23-.43	.24-.44
4	.20-.31	.20-.32	.20-.34	.20-.36	.20-.37
5	.20-.37	.23-.40	.25-.43	.27-.46	.28-.48
6	.20-.35	.20-.37	.20-.38	.21-.41	.22-.43



### 5.3.6.2 Discussion

The results show that whilst having a marked effect on the baseline concentrations, the modifications to the model had only a minor influence on diurnal variations.

Table 5.11 shows increased short circuiting to result in increased efficiency of ammonia removal. Nitrification is dependent on retention time (Liao and Mayo, 1974), and with the modifications, the reduction in nitrification by the concentration of flow through a reduced area is over compensated by the increased ammonia removal in the large area of filter with a reduced flow. In the reference system, however, short circuiting is associated with a reduction in the efficiency of ammonia removal. This is caused by the factors which lead to the short circuiting and also in response to the short circuiting itself.

In the reference system short circuiting results primarily from the clogging of the filter media. This creates zones which receive so little flow that they become anaerobic (see section 3.3.3.1). As a result of clogging the flow is concentrated into certain areas of the filter, where it creates channels along lines of least resistance. This leads to slower flow in areas not clogged. These areas are the places of high biological activity and thus may suffer from shortage of oxygen. In addition there may be other forms of competition between heterotrophic and nitrifying bacteria.

The model simulates the effects of a reduced oxygen supply by the functions AL and OM (4.5.2.2). It does not include a function to simulate the reduction in media surface available to nitrifying bacteria nor is any distinction made between the effectiveness of nitrification in the different parts of filter. Further modification to the model could be made, although any functions developed would of necessity be purely empirical.

As a final modification the improved ammonia removal effected by short circuiting was combined with increases in ammonia production rate (Table 5.12). The results of these runs are presented in Table 5.13.

Table 5.12 Values attributed to constants CA, FVA, FVB, CRA and CRB during re-runs

Run	1	2	3	4	5
CA	0.30	0.30	0.30	0.20	0.10
FVA	0.80	0.90	0.95	0.95	0.95
FVB	0.20	0.10	0.05	0.05	0.05
CRA	0.20	0.10	0.05	0.05	0.05
CRB	0.80	0.90	0.95	0.95	0.95

Table 5.13 Results of runs using further modified model

Run	AT	AF	OT	OF
	Max (hour)		mg/l	Min (hour)
1	2.80 (38)	2.79 (38)	5.40 (32-48)	2.87 (31-48)
2	2.65 (37)	2.61 (37)	4.48 (32-48)	1.40 (30-48)
3	2.54 (37)	2.51 (37)	4.00 (29-48)	0.73 (29-48)
4	1.53 (36)	1.50 (36)	4.00 (29-55+)	0.73 (29-55+)
5	0.59 (33)	0.56 (33)	4.00 (31-41)	0.73 (31-39)

As can be seen from Table 5.13, the largest ammonia concentration occurred with the highest ammonia production rate and least degree of short circuiting (run 1), whereas the lowest oxygen concentration occurred with the greatest degree of short circuiting (runs 3, 4 and 5). Only in run 5 was it unnecessary to withhold feeds. In run 1 the second and third feeds were withheld, in runs 2 and 3 the second feed was withheld while in run 4 it was the third feed that was missed. The time of the maximum concentration reached in the tank also varied between runs, from fourteen hours in run 1 to 9 hours in run 5. In none of the five runs was the rise and fall in ammonia and oxygen concentration as rapid as recorded in the laboratory.

The results presented here show better agreement with measured diurnal variations than the results produced by the unmodified model. No one run gave the best all round agreement. While run 1 has results similar to the diurnal variations measured on day 10 (240 hours) in the laboratory, a lower rate of ammonia production gives a better fit to the ammonia

concentration measured on day 4, but not as good an agreement for oxygen.

Further calibration of the model by additional changes to the constant CA and the degree of short circuiting may have resulted in closer agreement between predicted and measured diurnal variation in water quality. However, this was not considered worthwhile since the results would not be based on knowledge of the underlying structure. For a substantial improvement in the predictive value of the model a return to the system analysis stage would be necessary. At this point the model was considered to have been fully developed with available data, and in the next section the use of the model is considered.

#### 5.4 Use of the Model

There were three distinct aims in building the model:

- (1) To indicate areas of inadequate knowledge.
- (2) To understand the behaviour of the laboratory recirculating systems.
- (3) To develop guidelines for determining carrying capacity and optimum design of filters for recirculating systems.

The discussion below focuses on how well these aims can be or have been satisfied.

##### 5.4.1 Indicating area of inadequate knowledge

During the construction and evaluation of the model several areas for further investigation were noted.

1. Daily changes in production of ammonia, faecal solids, oxygen consumption and fish growth should be investigated if diurnal variations are to be modelled satisfactorily.
2. The equations incorporated into the model concerning:
  - (a) Growth - the insensitivity of W confirmed the need for a better growth model which allows the prediction of growth and responds to changes in biological and environmental factors.
  - (b) Ammonia production - this was sensitive to a 1 per cent change in the constant CA. The relationships considered for inclusion in

the model varied by 1400 per cent (4.5.1). An accurate relationship for the species and temperature considered, needs to be developed. Effect of diet should also be considered.

- (c) Ammonia removal - The equation of Liao and Mayo (1974) was used outside the range of conditions for which it was intended, i.e. retention time lower and influent ammonia concentration higher. In addition the model was sensitive to this equation. Its use in the model therefore requires further consideration.
  - (d) Solids production and removal - Since some solids accumulate in the reference system (3.2.5.7) the representation of solids removal by their immediate oxidation on entering the filter was considered inadequate. A relationship which takes into account accumulation of solids and their long term degradation should be investigated. This may increase the sensitivity of the model to the consumption of oxygen in the degradation of organic wastes, particularly if a better relationship could be described concerning the consumption of oxygen during a 24 hour period.
3. The ammonia concentration used in the clip function controlling the withholding of food was considered by Forster (pers. comm.) to be too low and suggested a value of 0.1 mg  $\text{NH}_3\text{N/L}$  (Table 2.1). It is uncertain if this value was used and not 1 mg/l total ammonia, whether acute or chronic toxicity could develop during daily variations. Other possible long term effects are also uncertain. Variations in pH were not modelled, being considered unimportant with regard to nitrification. Their inclusion to allow calculation of un-ionised ammonia from total ammonia needs to be considered.
4. The representation of interaction between filter bacteria was superficial and requires further study. In particular the following aspects were noted for consideration:
- (a) Profile of nitrifying activity through the filter.
  - (b) Competition for oxygen and space between nitrifying and heterotrophic bacteria.
  - (c) Secondary ammonia production.
  - (d) Bacterial growth and oscillations in PFC's.
  - (e) The adaptive response to changes in ammonia and solid concentrations.

5. No account is taken in the model of variations in fish size. When calculating the oxygen consumed by fish respiration an average fish size is taken. The effect of variations in fish size on the sensitivity of the model to respiration should be considered. Variations in fish size may also influence growth rates and waste production rates.
6. The use of discrete values for switch and clip functions increased sensitivity. Perhaps a better representation of the reference system could be achieved by transitions and therefore the rates of transitions should be examined.

#### 5.4.2 Understanding the behaviour of the laboratory recirculating systems

There were many ways in which the modelling process helped increase understanding:

1. It necessitated the collation and evaluation of data from a wide range of studies, for example, from water sanitary engineering and bacterial kinetics to studies of fish energetics.
2. To develop the model's structure it was necessary to identify the important variables and the relationships between them. With this structure it was possible to investigate the effects of a change in one variable on other parts of the system. Such experiments were not possible in the laboratory. In a similar way, by considering the model predictions as a control, it was possible to identify adaptation by the filters to increase in load.
3. It clearly showed the large effects on the behaviour of the system caused by management decisions such as whether to give or withhold food because of high ammonia levels.
4. In evaluating model output a comparison was made with a mental prediction of what one would expect given the inputs to the model. Through checking whether this prediction matched up to the output and why, one's understanding of the system increased.
5. Use of graph theory in the verification of the model presented the author with many questions regarding the relationships between variables. Understanding was developed in answering these questions, particularly where this involved examination of the results of the earlier experiments. In addition, the construction of signed digraphs

- provided the author with an alternative means of viewing the system.
6. Use of the inspection programme identified groups of variables with similar patterns of behaviour. It also identified different types of behaviour, such as pulses. To ensure the credibility of the model it was necessary to check these against those of the reference system.
  7. Through sensitivity analysis, the relative importance of each variable was identified and checked, and hence those factors having a dominant effect on the behaviour of the system could be located.

In the future, the increased understanding promoted by the simulation model may be of value in managing recirculating systems, indicating effective action against possible system events which effect the economic or biological efficiency of the system. In addition the model may allow a transition from closed-loop (feedback) process control to open-loop control. For example, the decision whether or not to feed is determined by the PFC of ammonia. Thus monitoring the water quality is necessary on a daily basis, and hence labour intensive. The model could be used to predict PFC's of ammonia for perhaps 1 week and then check these predictions with a weekly monitoring. This requires that sufficient confidence is established in the predictions of the model. Whilst agreement between predicted and measured PFC's of ammonia was good, a level of confidence sufficient for the model to be used in prediction has not yet been established (see for example 5.4.1.2 (b)).

#### 5.4.3 The development of guidelines for carrying capacity and optimum design of filters for recirculating systems

It was earlier suggested (2.4) that to design the best system for a particular purpose the design method of de Neufville and Stafford (1971) would be of value. After the definition of objectives and the formulation of measures of effectiveness, the method requires a number of alternative designs to be generated and evaluated in terms of the measures of effectiveness. On the basis of this, the design which best achieves the stated purpose can be selected. In order to discover how the measures of effectiveness change with different designs, de Neufville & Stafford propose the use of a model. Following further development, the simulation

model developed here could be used in this manner.

The uses to which the predictions by the model may be put depends on the degree of confidence which has been established in their reliability (Brockington, 1979). Confidence is built up by knowledge of the accuracy of the predictions (considered below), and the degree of validity shown in the evaluation of the model.

#### 5.4.3.1 Accuracy of the predictions

In a stochastic model the accuracy of the predictions may be indicated by placing confidence intervals about the results; in a deterministic model this is not possible and a full error analysis is necessary. This is a complex process requiring error bounds to be attached to all parameters and initial values. As suggested by Stone (pers. comm.) this can be simplified to initial values and parameters with a maximum sensitivity greater than a 1 per cent. To determine the potential range of predicted results and hence accuracy, the values of the model should be changed in the direction of their sensitivity sign. The model should then be run, so that the worst combination of errors is encountered, for each state variable in turn.

In the case of this model, full error analysis was considered unnecessary since it had already been concluded (5.2.3) that the high sensitivities recorded during the longer run would present difficulties if the model was used for prediction. Even over shorter runs the sensitivity of the model to certain variables results in a level of inaccuracy in the model output too high for the model to be used in prediction. For example, sensitivity analysis showed that 1 per cent change in circulation rate resulted in a 1.0 and 1.1 per cent change in the concentration of ammonia in tank and filter respectively. During the operation of the laboratory systems, circulation rate showed variations of up to 6.5 per cent. Thus the potential range of values of AT and AF resulting from variations in circulation rate alone would be high.

Whilst it is clear that the model cannot be used to develop quantitative relationships concerning carrying capacity and optimum design filters, the results of modelling can be used in a qualitative sense. In particular, the

following points emerged concerning the design and carrying capacity of the laboratory systems.

1. Feeding the fish once a day leads to large fluctuations in oxygen and ammonia levels. Feeding throughout the day may be more labour intensive but can lead to increased carrying capacity.
2. The filter is complex in that it does both biological and mechanical filtration. The latter varies in efficiency with time and at high loads may require frequent backwashing, thus causing nitrogen and carbon cycling among the bacterial populations as they become physically disturbed (Forster, pers. comm.). Competition between the heterotrophic and nitrifying bacteria results in a decrease in the efficiency of ammonia removal. Improvement to the design of the laboratory system would include the separation of the solids removal and nitrification stages, with re-oxygenation between them.
3. The efficiency of ammonia removal is markedly influenced by retention time. Whilst increased retention time increases ammonia removed, in a submerged filter it also results in decreased oxygen availability.

In a closed recirculating system the effect of retention time is further complicated, since although a higher circulation rate may decrease retention time, the frequency with which the culture water re-enters the filter is increased. The investigation into the effects of retention time in experiment 3.3.4(a) was unsuccessful. The model suggests that nitrification increases with an increase in circulation rate to a maximum beyond which the rate of ammonia removal decreases rapidly. The circulation rate at which ammonia removal is maximal may vary according to the values used in the initialization of a run.



## 6. CONCLUSIONS

This thesis attempted to develop a set of guidelines for the design and operation of a recirculating system. The lack of such guidelines was seen as a major limitation in establishing the feasibility of recirculating systems for use in commercial fish culture.

A study of the behaviour and performance of a recirculating system was seen as a key factor in the development of design and operational guidelines. This required that the system be studied as a whole, and a research programme leading to the construction of a simulation model was initiated.

The first phase of the research programme involved the design, construction and operation of a number of similar experimental recirculating systems. The performance of the systems in the culture of common and grass carp was evaluated by monitoring a number of biological and environmental variables. After an initially high mortality rate, the systems operated satisfactorily with acceptable specific growth rates and food conversion efficiencies. However, in the latter part of the experiment, growth and food conversion deteriorated, and difficulties were encountered concerning water quality and filter performance. Therefore experiments to consider the production of solid wastes by common carp, salt accumulation in a closed system and the effects of grain size and flow rate on filter performance were undertaken, together with an analysis of growth and food conversion in new populations of common and grass carp.

An equation describing the relationship between the production of solid wastes and weight of feed was developed, and the results of this experiment were incorporated in the simulation model. Considerable variation in the rates of salt accumulation were experienced, but it became clear that the largest increases were of sodium, potassium and chlorine ions. Because of difficulties in controlling flow rate and clogging of the filter bed, the experiment to consider the effects of grain size and flow rate on filter performance was abandoned. From the results obtained it was clear that the advantages of increased specific

surface area with small grain filter media were offset by the increased rate at which the bed clogged with subsequent reduction of flow, short-circuiting and eventual failure as a biological or mechanical filter.

Good growth and food conversion rates were achieved with the new population of common carp. As at the end of the first experimental period, the grass carp showed little if any growth. Dietary difficulties were suspected but not confirmed, and further culture of grass carp was discontinued. The mixed culture of grass carp and common carp in the same tank offered no advantages, but indicated that ration levels for the common carp were less than optimum i.e. growth and food conversion efficiency increased with an increase in weight of feed. Withholding food for a week resulted in a marked loss in weight. The rates of weight loss were similar to those found by other authors and varied according to the SGR prior to starvation. This was thought to reflect differences in the availability of body fat for metabolism. An average rate of weight loss was calculated and this was incorporated in the model. Relationships between fish weight and length were calculated for grass carp, mirror carp and common carp and significant differences were found between them. The difference between common and mirror carp, however, was slight compared to differences obtained from comparisons with some unpublished data. The relationships developed were therefore considered to be of use only for fish of the same genetic stock and maintained in similar nutritional and environmental conditions.

Based on the understanding gained from the experimental programme a computer model was constructed. This focused on the simulation of oxygen and ammonia concentrations in both fish tank and filter, and used a simple growth model. The model was verified using graph theory, by inspection of standard runs for maximum, minimum, mean and variation and by sensitivity testing. Some inconsistencies in the internal structure and behaviour of the model were found and corrected. Four modes of behaviour were identified and analysed, and pulse like behaviour was found in a number of variables. This presented no difficulties with regard to the reference system, but changes in mode made the use of sensitivity testing difficult. Over short runs sensitivity values were not high, but over longer runs the model became highly sensitive. Thus the model was not considered suitable

for prediction.

The model was validated by the comparison of model output with the actual behaviour of the system using a range of stocking densities and ration levels. Agreement between predicted and actual pre-feed concentrations of ammonia and oxygen in the tank was good, but there was often an order of magnitude difference between predicted and actual diurnal variations of ammonia and oxygen in both tank and filter. Modifications to ammonia production, and simulating short-circuiting in the filter were considered, but while some improvement in agreement between diurnal variations was achieved, such calibration was considered of limited value since it was not helpful in understanding the underlying structure.

The use of the model was considered. It was concluded that the model had been successful in indicating areas of inadequate knowledge. Whilst not providing a complete explanation for observed behaviour, the model did provide a framework for evaluating system behaviour and offered some insights into the possible causes of this behaviour. Use of the model to develop guidelines for determining carrying capacity and optimum design of filters was primarily restricted by the sensitivity of the model to changes in mode and some variables, and by the lack of data to model diurnal variations satisfactorily. Refinements to the model and increased accuracy with which variables may be controlled and monitored may allow the model to be used in a more predictive manner.

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## APPENDICES

- 1 List of Fish mentioned in the Text
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- 3 Analysis of Growth and Food Conversion
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APPENDIX 1. LIST OF FISH MENTIONED IN THE TEXT

<u>Common English Name</u>	<u>Latin Name</u>
Catfish, Channel	Ictalurus punctatus Raf.
Carp, Common	Cyprinus carpio L.
Mirror	Cyprinus carpio L.
Grass	Ctenopharyngodon idella Val.
Cod	Gadua morhua L.
Eel	Anguilla anguilla L.
Flounder	Platichthys flesus L.
Herring	Clupea harengus L.
Mullet	Mugil cephalus L.
Salmon, Atlantic	Salmo salar L.
Shrimp	Penaeus spp.
Tilapia	Tilapia spp.
Trout, Brown	Salmo trutta L.
Rainbow	Salmo gairdneri R.
Turbot	Scophthalmus maximus L.

## APPENDIX 2. ANALYTICAL METHODS

### A2.1 Fish Production

Two methods were used for estimating the weight of fish, depending on whether individual or total fish weights were required. In both methods the fish were starved for 24 - 36 hours before weighing to empty the gut.

#### A2.1.1 Weighing Individuals

The fish tank was drained to facilitate a complete netting of the fish. A dustbin half filled with well aerated water from the tank was used to hold the fish. A dosing tank containing 0.05 mg/l of Benzocaine was prepared (Laird and Oswald, 1975) for anaesthetising. About five fish at a time were taken from the dustbin and placed in the tank until subdued. The fish were then lifted out and drained before being individually weighed on a top pan balance. The weighed fish were measured (length +/- 5 mm) and placed into a well aerated tank to recover before being returned to the main fish tank. Difficulties with this method were that it:-

- a) Requires near complete draining of fish tank.
- b) Causes a state of high metabolic activity and stress during netting and handling.
- c) Gives a variable weight depending on the amount of water which is measured with the fish.

Repeated weighings of individual fish, however, showed that errors were within 1 per cent.

#### A2.1.2 Group Weighing

Where individual weights were not required a less time consuming method was to bulk weigh the fish using a spring balance and net. As in the previous method the fish were first netted. By the time this method was first employed the design of the fish tanks was modified to facilitate complete netting without draining the tank. Netted fish were held in a

dustbin of well aerated water before being counted into the weighing net. After weighing the fish were returned directly to the main tank. Errors in weighing arose from fish activity causing fluctuations in balance readings and from variable quantities of water covering the fish. The weight of water decreased the longer the fish were suspended on the balance as water drained away. To reduce these errors readings were only made with a still net of fish from which water had stopped flowing but continued to drip. Repeated weighings of nets of different weights of fish reimmersed in water showed errors to be +/- 30 grams. Since the lightest net-full of fish weighed over 1 kg this was less than 5 per cent.

## A2.2 Chemical Analysis

In the course of the investigation, it was necessary to monitor the following variables:

Ammonia	pH
Nitrite	Conductivity
Dissolved Oxygen	Temperature

An initial survey was made of available analytical methods to select those for large sample numbers and least affected by the presence of interfering ions (e.g. Hydrogen Sulphite in ammonia determinations) and suspended solids. In view of the lack of analytical facilities, simple methods were preferred, occasionally at the expense of accuracy. In all cases analyses were made immediately after sampling because of the large number of samples taken and limited storage.

### Ammonia

Initial determinations were made spectrophotometrically using the Direct Nesslerisation method (HMSO, 1972). Many samples, however, were turbid and difficulties were experienced in obtaining stable readings. Addition of potassium sodium tartrate reagent or cupric sulphate solution with caustic soda, as recommended by BDH Chemicals Ltd., caused changes in colour intensity and results were unsatisfactory. Muir (1977) recommends an autoanalysis procedure based on a phenol- hypochlorite method. Due to the limited facilities available this method could not be adopted here.

A Colorimetric chemical method was used in the determination of ammonia in fish culture water by Lewis and Buynak (1976), Konikoff (1974) and by Solomon and Brafield (1972). Colorimetric analysis requires three separate procedures to determine the amount of particular chemical present in a sample:-

1. Isolation of the chemical from interfering materials in the sample.
2. Production of a colour by the action of a chemical reagent on the isolated chemical.
3. Measurement of colour so produced.

(Thomas and Chamberlain, 1974).

The colour produced in the measurement of ammonia was assessed by Konikoff (1974), Solomon and Brafield (1972) and here by visually matching the test colour in a comparator, against a range of permanent glass standards. A number of different comparators are available, the most popular model being manufactured by Hach. Cuenco and Stickney (1980) found a maximum error of 20 per cent of the mean (as determined by the standard method) using the Hach Kit to measure total ammoniacal nitrogen. A similar result was obtained by Boyd (1980), who evaluated four comparator methods for the determination of ammonia. He concluded that while this method was not suitable where a high degree of accuracy and precision was necessary, it could be considered reliable, giving consistent results.

The BDH Lovibond comparator was selected for use since it is as simple to operate as the Hach Kit, provides a greater range of glass standards, and uses an optical path of 113 cm. With pale coloured solutions the use of a longer optical path facilitates discrimination. It is possible to interpolate between standards and thus achieve an accuracy of better than 0.02 mg/50 ml sample at lower concentrations. Solomon and Brafield (1972) found this method to be "objective and repeatable".

The method adopted for the production of a measureable colour was based on the use of Nessler's reagent (a strongly alkaline solution of potassium mercuric iodide) following the procedures given in the Colorimetric Chemical Analytical Methods manual (Thomas and Chamberlain 1974). Commercial Nessler's reagent (AR) which was compatible with the glass colour standards was used throughout.

This method allows the determination of total ammonia by driving the equation;  $\text{NH}_3 + \text{H}^+ \rightleftharpoons \text{NH}_4^+$  - to the left, and measuring unionised ( $\text{NH}_3$ ) ammonia.

### Nitrite

The standard sulphanilic acid method (HMSO, 1972) was initially used. The wide variations in the nitrite concentrations recorded necessitated a large range of dilutions to be prepared for a suitable calibration curve to be produced. In view of the preparation of fresh standards necessary each day and the large number of samples to be analysed, this method was found to be impractical. Later, when nitrite determinations were found essential as an aid to water quality control, a highly simplified method was used. This involved the addition of seven drops (approximately 0.2 ml per drop) of each of two reagents to a 5 ml sample contained in a cuvette. The resulting colour change was visually compared with a colour chart of four intensities of colour representing nitrite concentrations of less than 0.1 mg/l, up to 0.15 mg/l up to 0.5 mg/l and greater than 1.0 mg/l.

### Dissolved Oxygen and Temperature

Throughout the experimental period these were measured in situ with a pHox model 62 TE combined oxygen and temperature meter with a mackereth type probe. Standardisation of the meter was by exposure of the membrane to fully saturated clean water for at least fifteen minutes. Immersion in a sodium sulphate solution gave a zero response for oxygen. Calibration was necessary for isolated determinations, while a daily check was found adequate during continuous use.

Temperature was recorded by a thermocouple in the probe. The accuracy of the thermocouple was checked periodically with a mercury thermometer. No differences were detected.

### pH

A pHox model 42E pH meter was used for in situ measurements. This was calibrated with buffer solutions in pH 9.2 and 4.8. Temperature compensation was manual.

## Conductivity

This was measured by a pHox model 52E conductivity meter.

## A2.3 Errors in Data Collection

### Fish Production

All recorded weights have been rounded up to the nearest gram, although the top pan balance was accurate to one hundredth of a gram. A major source of variance in fish weight was the water weighed with each fish. One technique to standardise this, is to towel-dry each fish prior to weighing. This removes some mucus which could increase the possibility of disease and also may further stress the fish by increased handling. After some experimentation a drilled dish was used which allowed excess water on the fish to drain away before weighing. Repeated weighings of individual fish showed that errors with this method were within 1 per cent. Bulk weighing was less stressful to the fish and errors for a whole tank were within 5 per cent. Therefore this method was preferred.

### Water Quality

As discussed earlier (4.2.5.5) the time scale for changes in water chemistry may be much smaller than the frequency of monitoring. Thus the concentration on any day may be at any point within the range of diurnal variations. This increases the inaccuracy of the measurement and makes data interpretation difficult, particularly with a limited number of samples.

The majority of published papers presenting data collected from recirculating systems do not give an indication of the errors attached to any of the measurements made. This also makes the interpretation of results difficult. One of the reasons for this is, as in this experimental programme, is that many of the readings are isolated records. Ideally each reading would represent the mean of a number of measurements all taken at the same time, and an indication of its accuracy given through some measure of variance. Such procedure, however, requires an inordinate amount



of time particularly when monitoring may be continued over a number of months. Additionally, the precision of the measurement should be appropriate for the level of investigation. Most research in recirculating systems is still at the level of the first approximation. As a result, errors are usually based on the techniques employed and many of the published papers estimate errors as a certain percentage of the measured value (e.g. 10, 20 per cent). In the present study the errors of the method were considerably larger than errors of measurement. An indication of the method errors for ammonia determination are given in Table a2.1.

Table A2.1 Accuracy of ammonia determinations

Concentration of sample mg/l	Expected Accuracy of determination mg/l
0.02 - 0.16	+/- 0.02
0.16 - 0.56	+/- 0.04
0.56 - 1.20	+/- 0.08
1.20 - 2.00	+/- 0.10
2.00 - 4.00	+/- 0.20
4.00 - 8.00	+/- 0.80

Percentage errors for volumetric standards,

0.02 - 2.00	+/- 1%
> 2.00	+/- 4%

APPENDIX 3. ANALYSIS OF GROWTH AND FOOD CONVERSION

The following tables contain the results of experiment 3.3.2, the analysis of growth and food conversion.

Table A3.1 Mixed culture in tank one

Date	Grass Carp		Common Carp		SGR	Wt. of food (g)	Total wt. gain (g)	FCE	Ration size	
	Init. wt. (g)	Wt. gain (g)	Init. wt. (g)	Wt. gain (g)					(1) (%)	(2)
8/1-23/1	2772	-118	4018	727	1.11	1038	609	0.59	0.97	1.61
23/1-14/2	2654	-75	4745	971	0.85	2785	896	0.32	1.62	2.43
14/2-6/3	2579	51	5716	1219	0.92	3169	1270	0.40	1.69	2.39
6/3-27/3	2630	-50	5914	-229	-0.19	1238	-279	-0.23	0.70	1.08
27/3-25/4	2580	80	5685	1085	0.60	1831	1165	0.64	0.71	1.01
25/4-15/5	2559	11	6770	460	0.73	2181	471	0.22	1.14	1.56
15/5-12/6	2570	580	5356	1890	1.08	5073	2470	0.49	1.98	2.87
12/6-6/8	3071	229	7246	3936	0.79	12800	4165	0.33	1.88	2.53
6/8-6/11	3064	136	11058	5782	0.46	20508	5918	0.29	1.31	1.60
		$\bar{x}$			0.71			0.34		
		s			0.40			0.26	1.32	1.90

(1) Ration size calculated on total weight of fish

(2) Ration size calculated on weight of common carp only

Table A3.2 Common carp culture in tank two

Date	Initial weight (g)	Weight gain (g)	Weight of food (g)	Ration size (%)	SGR	FCE
8/1-23/1	7537	430	1161	1.00	0.37	0.37
23/1-14/2	7834	1264	2891	1.55	0.68	0.44
14/2-6/3	9098	1791	3655	1.74	0.86	0.49
6/3-27/3	9844	-60	1523	0.74	-0.03	-0.04
27/3-25/4	9713	1667	3688	1.21	0.55	0.45
25/4-15/5	11101	139	2647	1.18	0.06	0.05
15/5-12/6	9330	2980	5920	1.95	0.99	0.50
12/6-6/8	12078	5692	15366	1.87	0.70	0.37
6/8-6/11	17770	4040	25848	1.26	0.45	0.16
					$\bar{x}$ 0.51	0.31
					s 0.34	0.20

Table A3.3 Grass carp culture in tank three

Date	Initial weight (g)	Weight gain (g)	Weight of food (g)	Ration size (%)	SGR	FCE
8/1-23/1	5883	-232	897	1.04	-0.27	-0.26
23/1-14/2	5651	133	2089	1.66	0.11	0.06
14/2-6/3	5784	166	2193	1.78	0.13	0.08
6/3-27/3	5950	-30	897	0.72	-0.02	-0.03
27/3-25/4	5920	330	2104	1.17	0.19	0.16
25/4-15/5	6250	320	1431	1.12	0.25	0.22
15/5-12/6	6570	2050	4553	2.14	0.97	0.45
12/6-6/8	8620	340	10874	2.25	0.07	0.03
6/8-6/11	8847	3	11027	1.35	0.00	0.00
					$\bar{x}$ 0.16	0.08
					s 0.34	0.19

APPENDIX 4. WEIGHT/LENGTH RELATIONSHIPS

Table A4.1 Further results

Species	Sample size	r <sup>2</sup>	Standard error of estimate	Y	Equation (y = a + bx)
G.C.	152	0.9899	0.0522	45.6985	Y = -2.2679 + 3.4325x
C.C.	58	0.9897	0.0443	8.6417	Y = -1.7941 + 3.1501x
M.C.	710	0.9905	0.0432	8.0205	Y = -1.6953 + 3.0352x

where Y = log weight (g)  
 and x = log length (cm)  
 G.C. = Grass Carp  
 C.C. = Common Carp  
 M.C. = Mirror Carp

Slopes of the simple linear regressions of log weight on log length were compared (Bailey, 1959) using the statistic d, where

$$d = \frac{b_1 - b_2}{\sqrt{\frac{S_1^2}{1(x-\bar{x}_1)^2} + \frac{S_2^2}{2(x-\bar{x}_2)^2}}}$$

$$df = n_1 + n_2 - 4$$

$$S.E. = \frac{s}{\sqrt{(x-\bar{x})^2}}$$

The regression equations for mirror and common carp showed a significant difference, the slope of the regression equation was significantly greater for common carp than for mirror carp. The slope of the regression equation was significantly greater for grass carp than for mirror or common carp.

Comparison of common carp and mirror carp

$$d = \frac{3.1501 + 3.0352}{\sqrt{(0.0443)^2 + (0.0432)^2}}$$

= 99.9616    df = 764    p<0.001

Common carp and grass carp

d = 1404.3389    df = 206    p<0.001

Mirror carp and grass carp

d = 95.4536    df = 858    p<0.001

APPENDIX 5. WATER QUALITY ANALYSES

Table A5.1 Water quality results

Analysis carried out by:

ADAS

Sample taken 27.3.80

Soil Science Department

Date analysed 1.4.80

Government Buildings

Coley park

Reading

Analysis	Concentration		
	Tank 1	Tank 2	Tank 3
pH	7.5	7.6	7.6
cf	1309	1619	1520
NO <sub>3</sub> -N (mg/l)	21	6.5	7.1
NH <sub>4</sub> -N "	<0.2	<0.2	<0.2
P "	4	4	9
K "	32	68	70
Mg "	19	27	28
Na "	117	158	165
SO <sub>4</sub> -S "	237	292	304
Cl "	130	193	206
Fe "	<0.1	<0.1	<0.1
Mn "	<0.1	<0.1	<0.1
Zn "	<0.1	<0.1	<0.1
Cu "	<0.05	<0.05	<0.05
B "	0.40	0.56	0.57
Ca "	132	140	104
S.S. (g/l)	0.030	0.016	0.024

Table A5.2 water quality results

Analysis carried out by:

AJAS

Soil Science Department

Government Buildings

Coley park

Reading

Sample taken 6.3.80

Date analysed 11.3.80

Analysis	Concentration		
	Tank 1	Tank 2	Tank 3
pH	7.7	7.6	7.6
cf	1041	1285	1339
NO <sub>3</sub> -N (mg/l)	6.2	6.8	5.9
NH <sub>4</sub> -N "	<0.2	<0.2	<0.2
P "	2.0	3.5	5.0
K "	33	45	60
Mg "	13	17	20
Na "	80	110	126
SO <sub>4</sub> -S "	300	384	425
Cl "	102	140	165
Fe "	<0.1	<0.1	<0.1
Mn "	<0.1	<0.1	<0.1
Zn "	<0.1	<0.1	<0.1
Cu "	<0.05	<0.05	<0.05
B "	0.45	0.50	0.57
Ca "	142	152	138
S.S. (g/l)	0.023	0.032	0.034





Table A5.4 Water quality results

Analysis carried out by:

ADAS  
 Soil Science Department  
 Government Buildings  
 Coley Park  
 Reading

Sample taken 15.5.80  
 Date analysed 16.5.80

Analysis	Concentration		
	Tank 1	Tank 2	Tank 3
pH	7.1	7.9	6.8
cf	1285	1252	1126
NO <sub>3</sub> -N (mg/l)	44	19	37
NH <sub>4</sub> -N "	<0.2	<0.2	<0.2
P "	4.00	1.18	3.23
K "	52	51	47
Mg "	18	18	17
Na "	100	107	88
SO <sub>4</sub> -S "	245	272	219
Cl "	113	120	95
Fe "	<0.1	<0.1	<0.1
Mn "	<0.1	<0.1	<0.1
Zn "	<0.1	<0.1	<0.1
Cu "	<0.05	<0.05	<0.05
B "	0.54	0.57	0.53
Ca "	136	131	115

**APPENDIX 6. AMMONIA REMOVAL BY FILTERS**

Table A6.1 Ammonia removal by filters (mg/l)

Date	Time	Influent ammonia concentration	Effluent concentration			
			A	B	C	D
4.1.	09:30	0.08	0.08	0.08	0.10	0.10
4.1.	11:30	0.20	0.18	0.20	0.10	0.16
4.1.	13:30	0.26	0.24	0.26	0.26	0.26
4.1.	15:30	0.52	0.52	0.40	0.42	0.40
15.1.	09:30	0.14	0.14	0.14	0.14	0.14
16.1.	09:30	0.14	0.14	0.14	0.14	0.14
17.1.	09:30	0.24	0.20	0.14	0.20	0.14
18.1.	09:30	0.24	0.20	0.14	0.20	0.14
21.1.	09:30	0.28	0.16	0.12	0.16	0.12
22.1.	10:30	0.20	0.20	0.24	0.24	0.24
22.1.	12:30	0.48	0.28	0.24	0.32	0.24
22.1.	14:30	0.60	0.48	0.36	0.44	0.20
22.1.	16:30	0.38	0.34	0.26	0.34	0.28
22.1.	18:30	0.32	0.20	0.20	0.28	0.20
22.1.	21:00	0.28	0.28	0.24	0.28	0.20
28.1.	09:30	0.20	0.24	0.24	0.24	0.24
29.1.	09:30	0.28	0.30	0.36	0.24	0.28
30.1.	09:30	0.34	0.28	0.36	0.26	0.24
31.1.	09:30	0.32	0.40	0.32	0.28	0.32
1.2.	09:30	0.40	0.40	0.32	0.28	0.36
11.3.	10:00	0.16	0.10	0.20	0.14	0.12
11.3.	14:00	0.24	0.24	0.52	0.44	0.48
12.3.	09:30	0.22	0.48	0.52	0.80	0.56
13.3.	09:30	0.32	0.20	2.00	0.18	1.00
16.3.	09:30	0.40	0.24	1.20	0.28	1.20
	$\bar{x}$	0.29	0.26	0.37	0.27	0.31
	s	0.02	0.01	0.16	0.02	0.07

APPENDIX 7. VARIABLES CONSIDERED IN THE MODEL

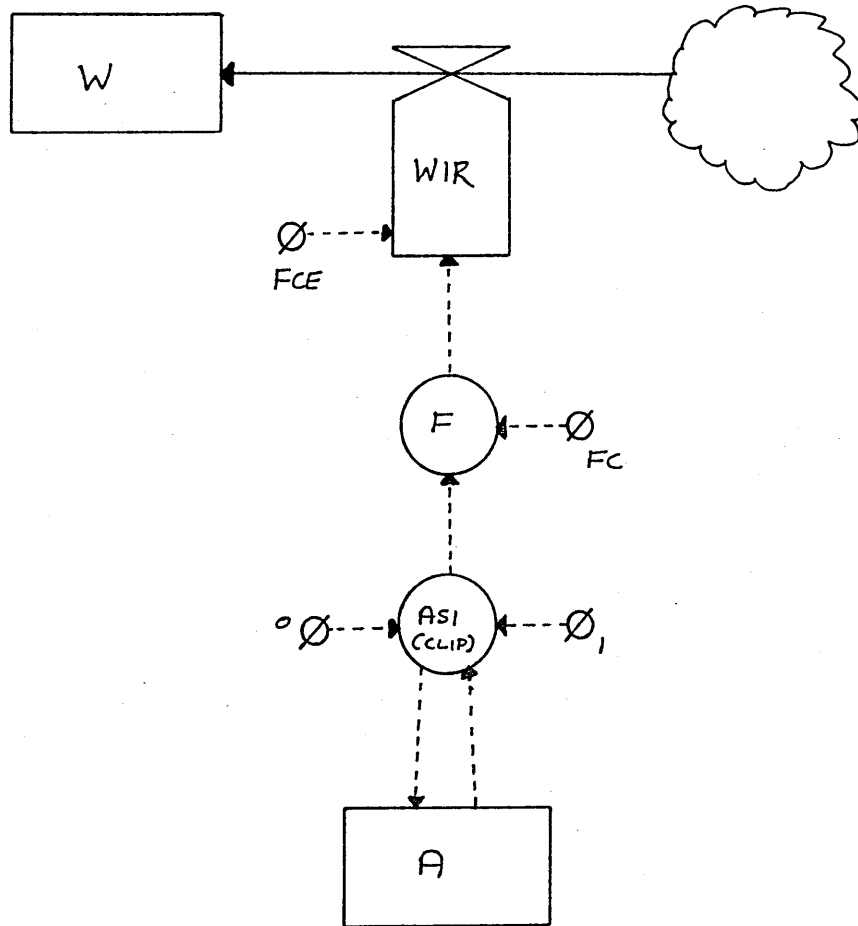
Table A7.1 Variables considered in the model

	Symbol used in Model	Unit	Dependent (D) or Controllable (C) Variable
Oxygen level in filter	OF	mg	D
Oxygen level in tank	OT	mg	D
Rate of oxygen removed from filter by nitrification	ORNA	mg/t	D
Oxygenation of the water by aeration	OR	mg/t	C
Oxygen removed by fish respiration	ORFR	mg/t	D
Number of fish	N		C
Weight of fish (biomass)	W	mg	C
Filter water volume	FWV	l	C
Tank water volume	TWV	l	C
Circulation rate	CR	l/t	C
Ammonia level in filter	AF	mg	D
Ammonia level in tank	AT	mg	D
Rate of production of ammonia	APR	mg/t	D
Wt of food fed	F	mg	C
% body weight to feed	FC	-	C
% of food fed going to ammonia	CA	-	D
Temperature	T	°c	C
Retention time	TM	t	C
Rate of ammonia removal by filter	ARR	mg/t	D
Nitrification equation	AN	mg/t	D
Food conversion efficiency	FCE		D
Weight increase rate	WIR	mg/t	D

## APPENDIX 8. THE INFORMATION SUB-ROUTINE

In Dynamo all equations are solved every DT. However in the model not all the processes are continuous. For example, the fish are fed only once every twenty-four hours, and the growth is assumed only once every twenty-four hours. To get round this the model contains an information sub-routine which effectively acts as a timer. The basic time unit in the model is the hour. Therefore DT represents an hourly fraction. The timer works by the use of a level (A), which sums the values of DT. When this value reaches 24, a clip-function operates, which changes the value of an auxiliary function (ASI) from zero to one. Processes multiplied by this function can now take place. At the next DT, the level A has reset itself by the use of ASI and the value of the function ASI has changed back to zero by the use of the clip. Processes multiplied by ASI now effectively stop working. Both the equations for W and for F are multiplied by ASI.

Figure A8.1 The information sub-routine



## APPENDIX 9. FEEDING SUBROUTINE

The management decision of whether to feed or not is simulated by consideration of the ammonia concentration. An auxiliary clip function (CHECK) monitors the ammonia concentration, and if it is  $> 1$  mg/l changes from 0 (zero) to 1. Another clip function ASIF, monitors the values of the functions ASI and CHECK. If  $ASI = 1$ , and  $CHECK = 1$ , then  $ASIF = 0$ . ASIF is used in the equation for calculating the food to be fed. If  $ASI = 1$  and  $CHECK = 0$ , then  $ASIF = 1$ . Therefore the fish are fed if  $ASI = 1$  and the ammonia concentration in the tank is less than 1 mg/l. If the concentration is  $> 1$  mg/l and  $ASI = 1$ , then no food is fed since  $ASIF = 0$ .

The weight decrease rate (WDR) is present in the equation for calculating the change in the level (W). It is only required to operate when  $ASIF = 0$ , i.e. when no food is fed. This is achieved by multiplying it with the auxiliary clip function (SLOW). The switch function, HI, assumes the value 2 if  $ASI = 0$  and the value 1 if  $ASI = 1$ . Another auxiliary (HS) subtracts the value of HI from the value of the function CHECK. So, if  $ASI = 0$ ,  $HI = 2$ . Therefore  $HS = CHECK - 2$ . If  $ASI = 1$ ,  $HI = 1$ , so  $HS = CHECK - 1$ . If  $CHECK = 0$  (ammonia concentration  $< 1$  ppm), then  $HS =$  either  $-2$  or  $-1$ . If  $CHECK = 1$  (ammonia concentration  $> 1$  ppm) then  $HS =$  either  $-1$  or  $0$ .

If  $HS = 0$ , then the switch function SLOW has the value 1. If  $HS \neq 0$ , then  $SLOW = 0$ . WDR is multiplied by SLOW. Therefore WDR only operates if the ammonia concentration is  $> 1$  mg/l, and  $ASI = 1$ .

## APPENDIX 10. OROG SUBROUTINE

In the reference system the production of organic matter (OROG) is largely dependent on the fish being fed. Although the fish are fed once a day, production of organic matter is thought to occur continuously. In the model this is simulated by the use of two auxiliary variables (FCAL and FD) and a rate, X. The value of the rate, X, is determined by a switch which selects between the previous value of X or the value of F depending on the value of ASI. The variable FD takes the value of FCAL (which equals F when the fish are fed, via X) and divides it by 24, to provide the auxiliary variable OROG with the weight of food per hour.

APPENDIX 11. AMMONIA CALCULATIONS FROM LABORATORY RECORDS

From Laboratory records

Number of fish = 49

Initial weight of fish = 1178 g

Weight of fish after 77 days = 3697 g

Specific Growth Rate = 1.48 % growth/day

Feeding Rate = 1.76 % body weight/day

Food contains 35 % protein

If it is assumed that 16 %  
of protein is nitrogen,  
then 100 g of food = 5.6 g nitrogen

Kaushik (1980)

Average weight of fish = 75.44 g

Fed over 24 hour period,  
1.76 % of body weight = 1.33 g

If 100 g of food contains  
5.6 g N, then 1.33 g of  
food contains = 0.074 g N

Nitrogen intake (X) = 0.074 g N/75.44 g fish/day



therefore X = 0.987 g N/Kg fish/day

log Y = 1.73 + 0.62 X Kaushik, 1980

log Y = 1.73 + (0.62 x 0.987)

= 2.34

thus Y = 219.84 mg N/Kg body wt/day

Since 3.637 Kg of fish are,  
in tank, total N excreted = 219.84 x 3.637 mg/N/tank/day

= 812.76 mg/N/tank/day

To convert mg N to mg NH<sub>3</sub> multiply by 1.19924  
(Thomas and Chamberlain, 1974)

= 974.70 mg NH<sub>3</sub>/tank/day

Furakawa and Ogasawara, (1955)

For 3697g of fish excreting ammonia at the rate of  
10 mg NH<sub>3</sub>-N/110g /24 hours

Total excretion = 336 mg NH<sub>3</sub>-N/day

= 404 mg NH<sub>3</sub>/day

At the rate of 20 mg NH<sub>3</sub>-N/110g /24 hours

Total excretion = 672 mg NH<sub>3</sub>-N/day

= 818 mg NH<sub>3</sub>/day

To convert mg NH<sub>3</sub>-N/1 to mg NH<sub>3</sub>/1 multiply by 1.21589

Saeki (1958)

NH<sub>3</sub> excretion = 25 mg/100 g fish; for 3697 g of fish  
For 3697g of fish = 924 mg NH<sub>3</sub>/day

Total N excretion = 50 mg/100 g wt; for 3697 g of fish  
For 3697g of fish = 1848 mg NH<sub>3</sub>/day

Hambrey (1980)

NH<sub>3</sub> excretion = 0.69 + 0.68875 SGR  
= 1.19 mg/Kg/min  
= 6325 mg/tank/day

## APPENDIX 12. KEY TO SYSTEM DYNAMICS DIAGRAMS

### Key to symbols



Level



Constant



Rate



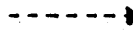
Source or Sink



Auxiliary



Material Flow



Information Flow

### Key to abbreviations

AF	=	Ammonia in Filter
AT	=	Ammonia in Tank
OF	=	Oxygen in Filter
OT	=	Oxygen in Tank
W	=	Weight of Fish
A	=	Information level
ARR	=	Ammonia Removal
AIFR	=	Ammonia entering into the Filter
RARR	=	Residual Ammonia returning to Tank
APR	=	Ammonia production
AI	=	Ammonia input
ORR	=	Oxygen removal
RORR	=	Residual Oxygen returning to Tank
OFIR	=	Oxygen entering into Filter
OR	=	Oxygenation
ORFR	=	Oxygen removed by Fish Respiration
WDR	=	Weight decrease of Fish
WIR	=	Weight increase of Fish
ORNA	=	Oxygen removed by the nitrification of ammonia
OROG	=	Oxygen removed by oxidation of organic matter
ASI	=	Information equation used to determine a 24 hour cycle
ASIF	=	Information equation used to override ASI if high ammonia levels occur

CHECK= Information equation checking if ammonia level in tank exceeds limit

KILL = Information equation checking if ammonia level in tank reaches toxic limit

HI }  
 HS } = Information equations used to operate WDR if no food fed  
 SLOW }

TM = Retention time

AL = A multiplier used in AN to stop nitrification when oxygen in the filter falls

An = Nitrification equation

OM = Multiplier used in AN

FCAL = Food to be fed for whole day (theoretical information)

FD = Food to be fed per hour (theoretical information)

F = Food fed to Fish

AV = Average weight of individual Fish

ONC = mg Oxygen removed per mg ammonia nitrified

N = Number of Fish

FWW = Filter water volume

TWV = Tank water volume

CR = Circulation rate

CA = Ammonia produced as a percentage of food fed

T = Temperature of water

FCE = Food conversion efficiency

X = Switch to provide information on feeding for simulation of organic waste production.

XOT = Concentration of Oxygen in Tank

XOF = Concentration of Oxygen in Filter

CNT = Concentration of Ammonia in Tank

CNF = Concentration of Ammonia in Filter

Other constants shown in Figure 4.13 and not listed above appear as numbers used within equations.

#### Dynamo 'Macro' Functions Used

MAX = Maximum value chosen ( max = P if P > Q )  
 ( max = Q if P < Q )

EXP = Exponential

LOGN = Log<sub>e</sub>

CLIP (P, Q, CRIT, REF) CLIP = P if CRIT > REF, CLIP = Q if CRIT < REF

DELAY<sub>3</sub> = Third order delay function

SWITCH (P, Q, CRIT) SWITCH = P if CRIT = 0, SWITCH = Q if CRIT = 0

TABHL = Linear interpolation between points in a table. If values higher than that used in table then assume the last value of table.