

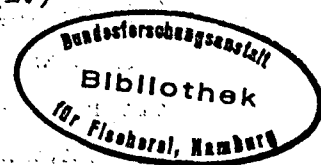
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The effect of algae on the water conditions in fish rearing tanks  
in relation to the growth of juvenile sole, Solea solea (L.)

by

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## INTRODUCTION

The suggestion of a beneficial effect achieved by the presence of algae in tanks in which fish larvae were being reared has been related to some property other than its use as a food either by the larvae or the food organisms on which the larvae were feeding (Jones 1970). Because it is known that algae will utilize ammonia as a source of nitrogen (Corner and Davies 1971) and that ammonia accumulating as an excretory product has an adverse effect on fish growth and survival (Alderson, unpublished results), it was decided to investigate the effect of the presence of unicellular algae both in relation to the level of dissolved ammonia and to the growth of fish held in static conditions.

## EXPERIMENTAL DETAILS

The fish used in the experiments were small Dover sole, Solea solea (L.), of between 4 and 6 g. Twenty-four fish were injected beneath the skin on the ventral surface with coloured latex (Riley 1966) to enable individuals to be identified, and following a recovery period of 7 days all the fish were weighed. Sixteen fish were then selected so that with four fish in each of the four tanks used all the tanks had approximately the same total weight of fish (Table 1). The fish were introduced into the experimental tanks 24 hours after weighing.

The 60 x 30 x 30 cm tanks were of clear Perspex and each contained 40 litres of sea water which had been sterilized by filtration. The four tanks were placed beneath two 1500 watt tungsten halogen tubes which gave an illumination at the surface of between 4000 and 4400 lumens/m<sup>2</sup> (Table 1), and the lights were controlled by a time switch to give a cycle of 12 hours light and 12 hours darkness. Alternate tanks in the row were either left free of algae or were inoculated with the marine flagellate Dunaliella tertiolecta Butcher to give an initial algal density of approximately 150 cells per microlitre. The tanks were maintained without flow for the duration of the experiment, but aeration with compressed air at the same rate in each tank and at a level

sufficient to maintain oxygen at 100% saturation in the tanks without algae was provided by submerged air-diffuser stones.

The fish were presented with an excess level of food over the whole experimental period. The food used was Lumbricillus spp., an enchytraeid worm previously found to give good growth with small juvenile flatfish (Kirk 1972; Kirk and Howell 1972), and in order to measure food consumption a known weight of fresh Lumbricillus was added to the tanks each day after the uneaten food from the previous days' feeding had been removed and weighed. The excess food was removed with a siphon, the worms being retained by a fine-mesh nylon filter and the sea water returned to the tank. As the Lumbricillus did not feed on the algae in the algal tanks, then any difference in the growth of the fish in the two treatments could not be related to a difference in food quality.

Daily measurements of total ammonia nitrogen were made using a phenol-hypochlorite method similar to that of Solorzano (1969), 10 ml portions being taken from a 25 ml sample which had first been centrifuged to remove algal cells and particulate matter. This enabled dissolved ammonia to be measured down to a level of 0.02 mg ammonia N/litre. Temperature and pH were usually measured twice daily; oxygen concentration was monitored at less frequent intervals, using a Mackereth electrode and meter, and routine measurements of algal cell density were also made. All the monitoring of water conditions was normally done during the daytime, but measurements were also made at 2 to 4 hour intervals over a complete 24 hour period in order to obtain information on the changes occurring during the hours of darkness.

The experiment was terminated after 13 days when it became apparent that algae were beginning to develop in the originally algae-free tanks.

## RESULTS

Figure 1 shows the levels of algal cell density, pH and temperature recorded over the experimental period. The temperatures plotted are the means for all the tanks, for although variation between replicates occasionally reached 1.0°C the variation of mean temperature between treatments did not exceed 0.2°C. More detailed temperature records for a short period of the experiment are given in Figure 2, where individual tank temperatures are plotted.

In tanks 1 and 2, pH, after showing an initial decline following the addition of the fish, levelled off at around 8.0 as a balance was achieved between the production of carbon dioxide by the fish and its loss through the water surface by aeration. The increase in pH in these tanks towards the end of the experiment, days 12-14, particularly

evident in tank 1, was caused by the development of algal populations due to contamination from the other tanks. Tanks 3 and 4 were inoculated with algae 3 days before the experiment began, in order to allow the algae to acclimatize to the lower temperatures of the experiment. The master culture from which the algae were taken had been maintained at 21°C. The result of this is evident in Figure 1 from the higher pH levels recorded in these tanks before the addition of the fish. The activity of the algae in tanks 3 and 4 resulted in large daily fluctuations in pH as carbon dioxide was first removed during the illuminated period and then released during the hours of darkness. Towards the end of the experiment, when the algal population was increasing as a result of the addition of a nutrient solution, the consequent increase in carbon dioxide consumption caused a considerable net rise in pH to levels not normally encountered in the open sea. The changes in pH and temperature in both treatments over a complete 24 hour period are shown more clearly in Figure 2.

The original intention had been to try to restrict the algal cell density to a maximum of around 300 cells per microlitre, and on the fourth day of the experiment the cell density was reduced by passing 20 litres of water from each tank through a filter press. However, this action caused a change in the level of dissolved ammonia in tanks 1 and 2 (Figure 3) and so the procedure was not repeated.

The poor development of the algae over days 6 to 9 was thought to be likely to precede a collapse in the populations due to exhaustion of some essential nutrient; 20 ml of the stock enrichment solution and 2 ml of the vitamin stock solution quoted by Walne (1966) for the culture of Isochrysis galbana Parke and other algae were therefore added to all of the experimental tanks, and this was followed by a resumption in algal growth.

Oxygen measurements showed that 100% of saturation was maintained in tanks 1 and 2 throughout the experiment, while in tanks 3 and 4 measurements taken when the algal populations were at their densest showed that 125% of saturation was reached at the end of the light period, this falling to 95-97% of saturation after 12 hours of darkness.

Figure 3 shows that over most of the experimental period the rate of accumulation of ammonia nitrogen in tanks 1 and 2 was fairly constant. The decrease during day 4 resulted from the filtration of 20 litres of water from each tank referred to earlier, and the decrease in rate of accumulation evident, particularly in tank 1, towards the end of the experiment was due to the development of algae. Samples taken for ammonia analysis from tanks 3 and 4 at the same times as those taken

from tanks 1 and 2 gave readings which did not differ significantly from a reagent blank, showing that in those tanks there was complete utilization of all the ammonia nitrogen produced by the fish over the experimental period. Measurements of ammonia nitrogen were also made at the times at which the pH and temperature results shown in Figure 2 were recorded. This was in order to investigate the possibility of an accumulation of ammonia in the tanks containing algae during the dark period. The results once again failed to reveal the presence of ammonia in these tanks at any time during the 24 hour period, while in tanks 1 and 2 ammonia showed a steady accumulation.

Table 1 shows the weights of the fish at the beginning and end of the experiment, together with the weight increases expressed as percentages of the initial weights. A comparison between the replicate means for each treatment showed that the differences were not significant, thus allowing the results from each replicate pair to be pooled for a comparison between the treatment means. This comparison, using a 't' test for small samples, gave a value for 't' of 2.50, which indicates that the difference in growth was significant at the 0.05 level.

The wet weight to wet weight food conversion ratios were calculated from the growth and food consumption data. These are given below, and here too a difference between the two treatments is apparent though this was not found to be significant.

Tank	Food conversion ratio	Mean food conversion ratio	Treatment
1	2.38	2.52	Tanks without algae
2	2.66		
3	2.21	2.26	Tanks with algae
4	2.31		

#### CONCLUSIONS

The results of the experiments clearly show the effectiveness of viable algae in removing dissolved ammonia from static tanks in which fish are maintained. Improvements in water conditions as an aid to larval survival have also been attributed to the presence of algae by Shelbourne (1964), though he maintained the algae Enteromorpha sp., attached to stones, in a separate tank through which the water from his rearing tanks was circulated. In the experiments reported by Jones (1970) it is possible, therefore, that the ability of the Chlorella to

remove metabolites may also have made some contribution to the improvement in larval survival.

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Table 1 Light intensity measurements for the experimental tanks, and growth data for the small juvenile sole in the two treatments

Tank	Light intensity at water surface (lumens/m <sup>2</sup> )	Weight of fish (g)			Weight increase as % of original weight	Mean % increase	Treatment
		Original	Final	Increase			
1	4300	4.16	5.59	1.43	34	47.25	Tanks without algae
		5.10	8.04	2.94	58		
		5.03	7.99	2.96	59		
		5.30	7.91	2.61	49		
		<u>19.59</u>					
2	4000	5.84	8.09	2.25	39	56.75	Tanks with algae
		4.15	6.14	1.99	48		
		4.29	6.10	1.81	42		
		4.63	6.89	2.26	49		
		<u>18.91</u>					
3	4400	4.45	7.25	2.80	63	56.75	Tanks with algae
		5.41	8.02	2.61	48		
		5.10	7.83	2.73	54		
		4.50	6.74	2.24	50		
		<u>19.46</u>					
4	4100	4.70	7.75	3.05	65	56.75	Tanks with algae
		4.67	7.59	2.92	63		
		5.91	9.23	3.32	56		
		4.38	6.80	2.42	55		
		<u>19.66</u>					

Fig1 Records of algal cell density, pH and mean temperature taken over the experimental periods

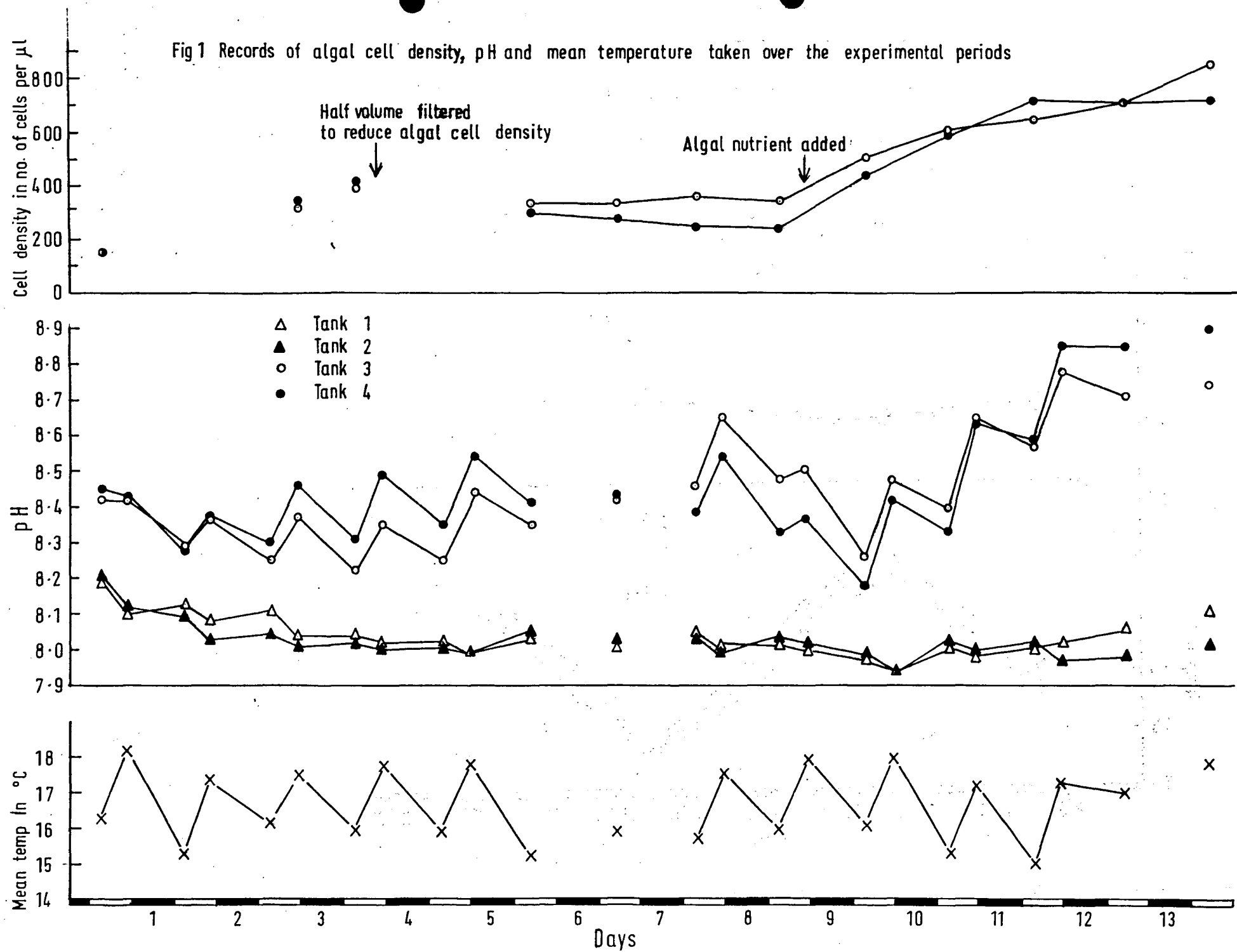
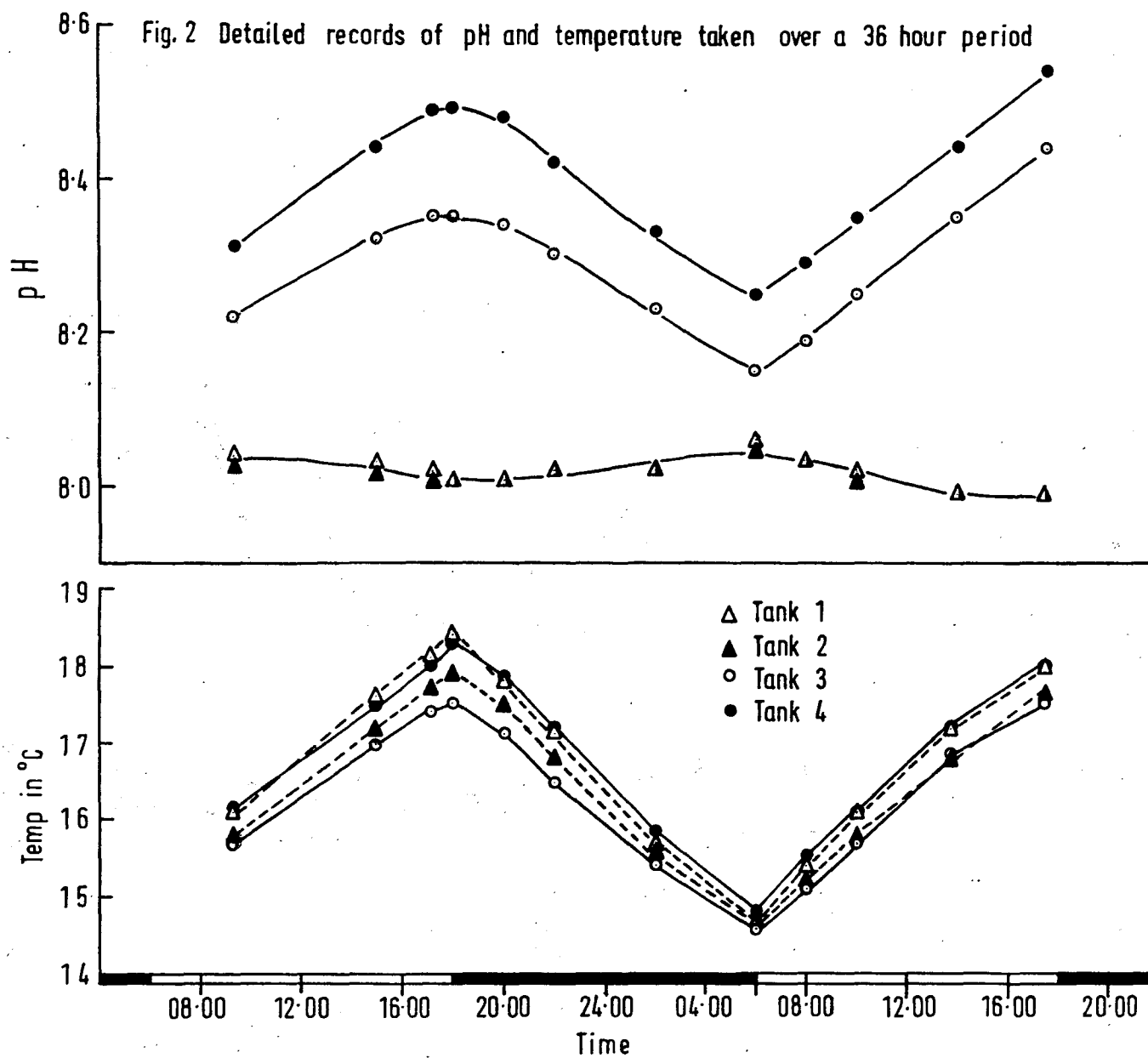


Fig. 2 Detailed records of pH and temperature taken over a 36 hour period





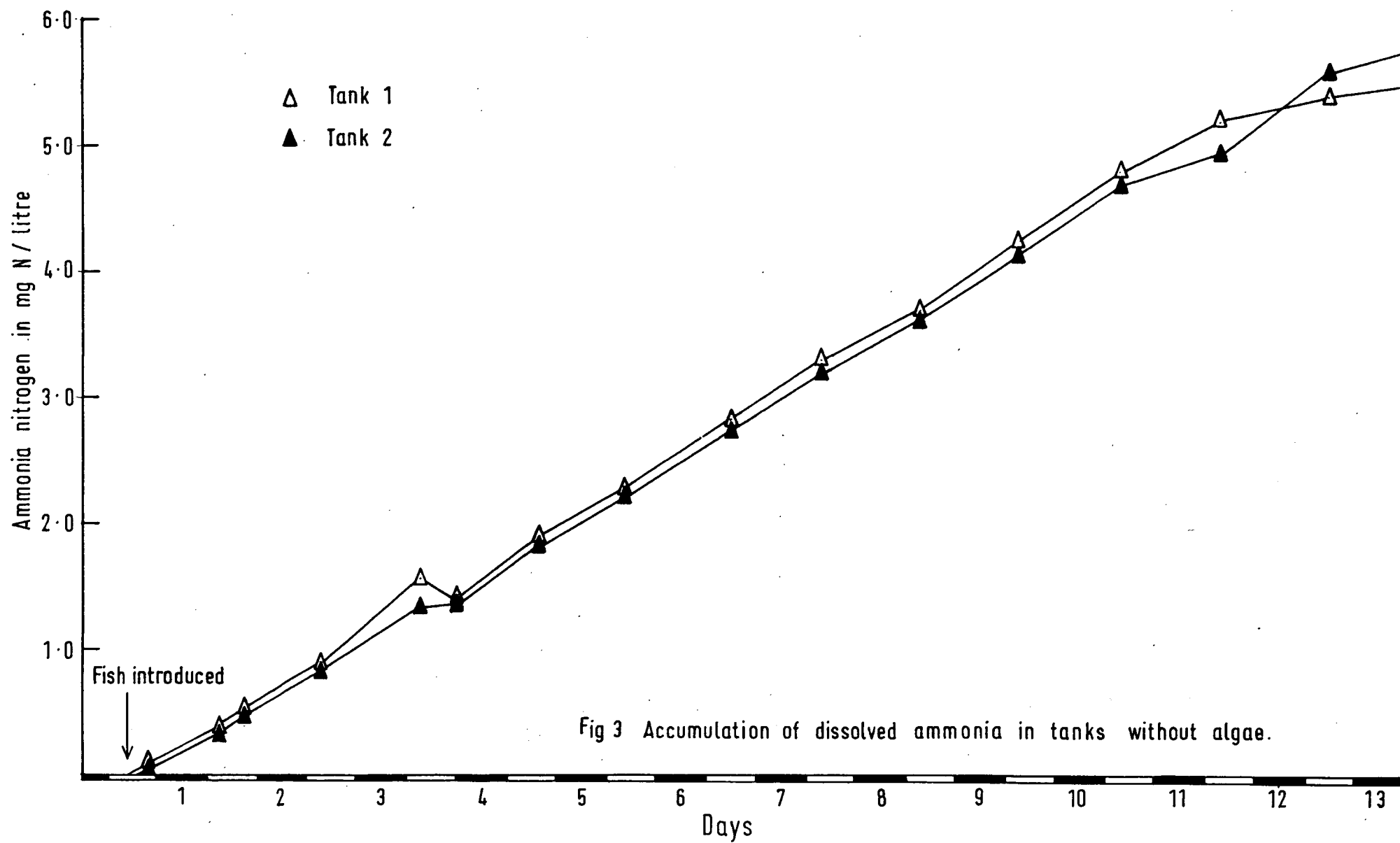


Fig 3 Accumulation of dissolved ammonia in tanks without algae.