



BIOACTIVATION:

**ESTABLISHMENT OF FULL NITRIFICATION OF FILTER
MEDIA**

WEBS PROJECT NUMBER 821607/0014

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GLP COMPLIANCE STATEMENT

The work described in this report was performed in a facility working to high technical standards and Good Laboratory Practice as defined in UK law, which is itself compatible with the OECD Principles of GLP (ENV/MC/CHEM(98)17).

Adherence to proper technical procedure within the test facility is regularly assessed by the internal Quality Assurance Manager, but the conduct of this study may not have been the specific subject of this inspection. The report has been reviewed for technical accuracy.

This report fully and accurately reflects the procedures used and data generated.

..... DATE:.....

Francis Saunders
Study Director.

1. Method of Analysis

1.1 Introduction

The test programme was conducted with the specific objective of determining the relative rates at which full biological nitrification of ammonia to complete oxidation to nitrate could be established using commercially available forms of biofilter support media.

Within the fish culture industry the presence of ammonia ($\text{NH}_4\text{-N}$) and the partially oxidised nitrite (NO_2) are considered highly undesirable being particularly toxic to fish. It is therefore the practice to oxidise such materials through the process of biological oxidation to convert them to the much less harmful form of nitrate ($\text{NO}_3\text{-N}$).

Such a procedure requires the establishment and maintenance of a biological culture, which is normally achieved using a biofilter, by passing the water through filter media where the bacteria, which carry out the conversion of the toxic substances into non-toxic materials, grow on the surface of the media.

The provision of an appropriate and effective form of media is a critical part of good practice within the industry.

1.2 Test Substrates

The media submitted for testing were:

Kaldnes K1
New Springflo Ribbon

1.3 Preparation

The method employed for the test was to commission two identical bioreactor vessels each with a working volume of 5 litres.

The reactors were placed within a temperature-controlled laboratory where the temperature was maintained in the range 23.7 – 25.1 °C throughout the duration of the test.

Reactors were randomly selected for allocation to the testing of one specific media.

Both reactors were loaded with equivalent quantities of media based on the specified surface area information and recommendations provided by the suppliers.

A single reservoir of feed-substrate containing a dilute solution of ammonium bicarbonate and 0.25 mg/l sodium triphosphate, pH adjusted to 7.80 was provided. The feed substrate to the individual reactors was delivered via Watson and Marlow

multi-head peristaltic pump to ensure that each reactor received identical loadings of ammonia solution.

Aeration to the reactors was supplied from a common source via a Charles Austin blower, delivering air through a multi-head splitter manifold, to the base of the reactors where the air was discharged into the aqueous phase through triple fine bubble diffusers.

At all times throughout the test period the dissolved oxygen levels within the reactors were maintained at or above 8.0 mg/l.

Operation of the reactors throughout the entire test period was on a continuous flow basis.

1.4 Procedure

The tests commenced on Day 1 when each of the reactors was prepared with the requisite quantity of the specified media and then filled to the TWL (nominal 5 litres) using tap water. No specific conditioning of the tap water was carried out.

The aeration was turned on and the reactors permitted to stabilise over night. After over night stabilisation the feed to the reactors was switched on (Day 2) and 10g of NS5000 product, a commercially available nitrifying culture supplied to the fish culture trade by AHL, was added to each reactor.

At all times throughout the test period the dissolved oxygen levels within the reactors was maintained at or above 8.0 mg/l.

Thereafter the feed to the reactors was maintained and steady state operational conditions through to the end of the test period.

2. Monitoring and Analysis

2.1 Parameters

The determination of the effectiveness of the respective media under test for the establishment and maintenance of full biological nitrification was made through the monitoring of the following parameters:

i)	dissolved oxygen.	:	WTW Oxi 340i
ii)	pH.	:	GM017/ TM013
iii)	NH ₄ -N (ammonia) of feed.	:	LCK303 TM001
iv)	NH ₄ -N (ammonia) of outlet.	:	LCK303 TM001
v)	NO ₃ -N (nitrate) of outlet.	:	LCK339 TM003
vi)	NO ₂ -N (nitrite) of outlet.	:	LCK341 TM004
vii)	Temperature of reactors.	:	WEB070 TM019

All analysis was carried out in accordance with methods specified in Controlled Documents UKAS Accredited Testing Laboratory No. 2608 (Initial Accreditation 07 August 2004) Test Methods Manual.

3 Results

See accompanying tables.

3. Discussion

The monitoring system was found to have acceptable levels of compliance. The analytical procedures were effective in accurately determining the respective values for the parameters specified.

Following the initial seeding of the reactors with the NS500 nitrifying culture the bio-oxidation of the ammonia present in the feed to each of the reactors was evident in both reactors.

Thereafter, over duration of the test it was clear that the rate at which full biological nitrification was established and maintained was significantly faster in the reactor No. 2 which contained the New Springflo Ribbon.

Reactor No 1, which contained the Kaldnes K1 media, was much less effective at enabling and supporting the establishment of an effective biomass able to fully oxidise the ammonia in the feed to the reactor.

The results indicate that the New Springflo media was able to reach stable and safe residual concentrations of both ammonia and nitrite, under the operating conditions described, within approximately 20 days. The Kaldnes K1 media was unable to reach these levels of removal even after 30 days of testing.

It is not clear why the Kaldnes K1 media should be less effective at supporting a biological film able to oxidise ammonia and nitrite. The internal surfaces of the reactor vessel provided a more effective surface for a biofilm than did the media itself. As a result the levels of both ammonia and nitrite in the outlet from this reactor were consistently higher than from any of the other media types tested.

On the evidence of this test it is concluded that over the period of the test the New Springflo Ribbon media performed most effectively as a biofilter media for the support of a biofilm capable of fully oxidising ammonia, and the intermediate oxidation product, nitrite, through to the much less harmful nitrate.

SPRINGFLO TRIALS - SPIREX AQUATEC

Ammonia Out

Date	Day	Kaldnes	New Springflo	Feed N-NH4
20.05	2			5.58
24.05	5	1.53	0.88	5.58
25.05	6	1.22	0.52	5.58
30.05	10	2.00	1.33	5.58
31.05	11	3.19	2.06	6.21
2.06	13	3.37	0.49	6.21
6.06	17	0.83	0.36	6.21
8.06	19	2.41	1.38	8.86
10.06	21	1.81	0.09	8.86
14.06	25	0.37	0.06	8.86
17.06	28	0.88	0.04	9.06
20.06	31	2.34	0.04	9.06
22.06	33	1.04	0.04	9.06

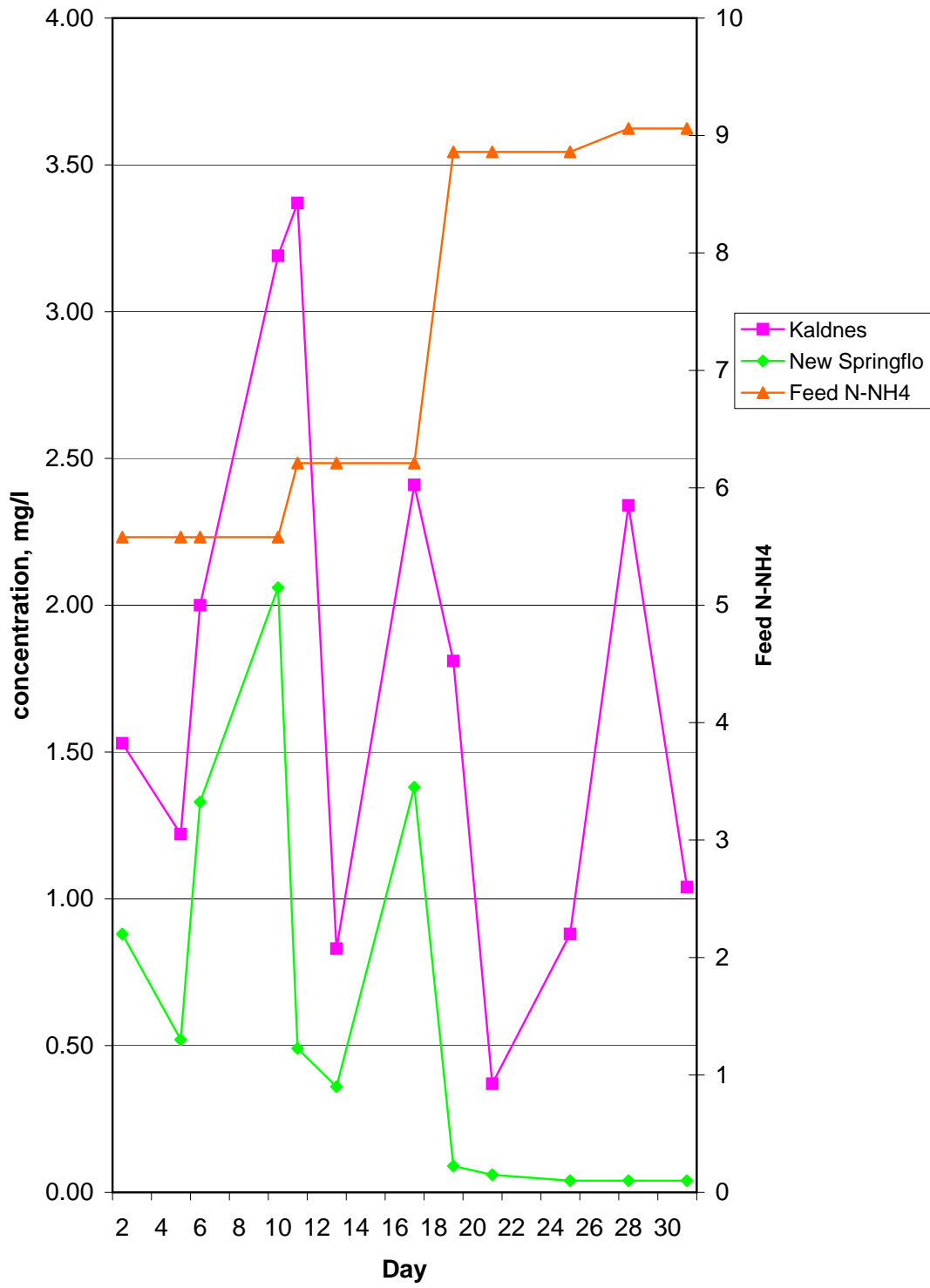
Nitrate Concentration

Date	Day	Kaldnes	New Springflo	Feed N-NH4
20.05	2			5.58
24.05	5	9.55	8.78	5.58
25.05	6	8.36	7.92	5.58
30.05	10	5.37	9.74	5.58
31.05	11	5.08	9.08	6.21
2.06	13	5.17	11.60	6.21
6.06	17	6.74	12.50	6.21
8.06	19	7.44	12.50	8.86
10.06	21	7.78	17.00	8.86
14.06	25	11.50	21.40	8.86
17.06	28	12.10	23.90	9.06
20.06	31	16.40	24.20	9.06
22.06	33	17.83	25.60	9.06

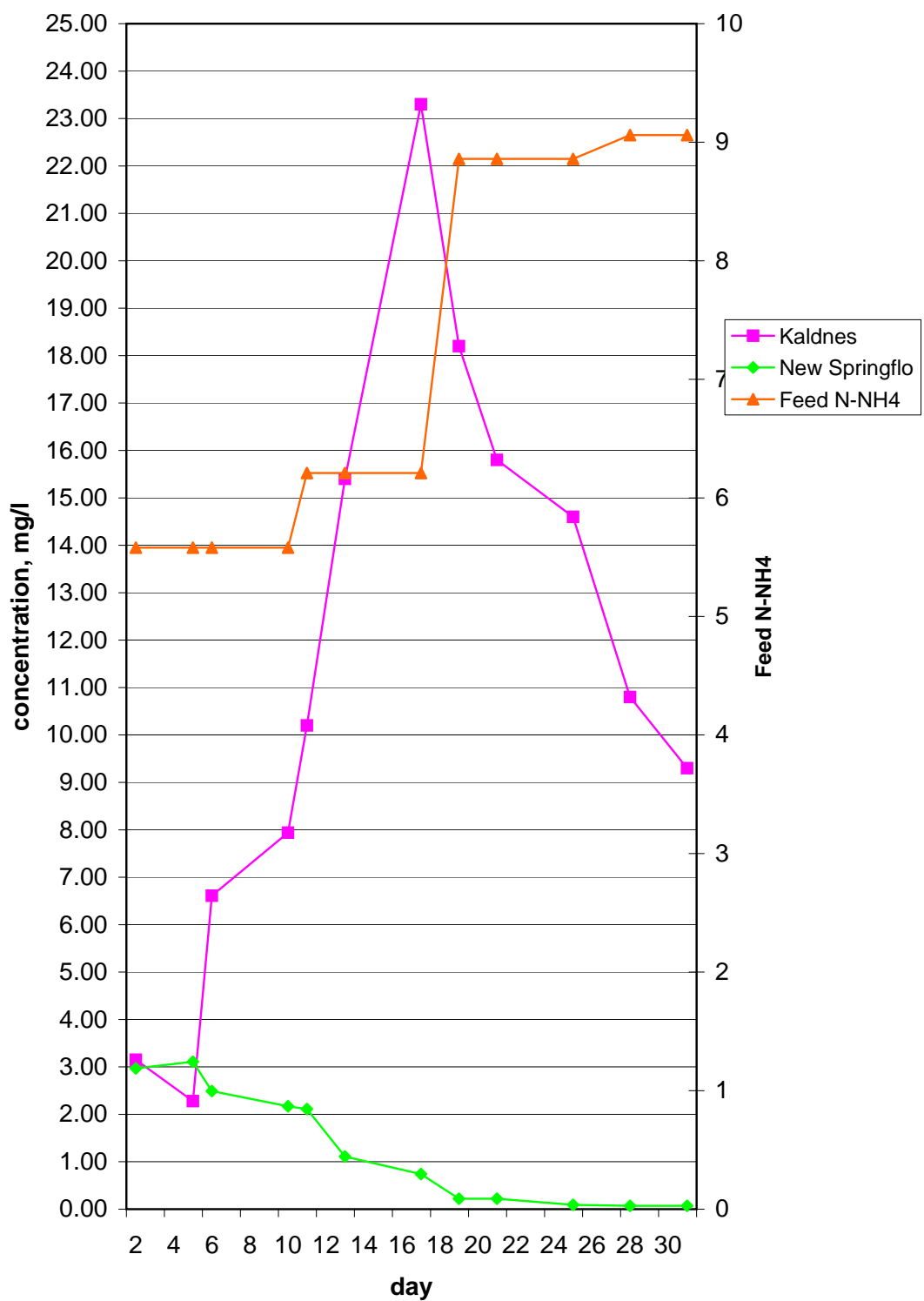
Nitrite Concentration

Date	Day	Kaldnes	New Springflo	Feed N-NH4
20.05.	2			5.58
24.05	5	3.15	2.97	5.58
25.05	6	2.28	3.11	5.58
30.05	10	6.61	2.49	5.58
31.05	11	7.94	2.17	6.21
2.06	13	10.20	2.11	6.21
6.06	17	15.40	1.11	6.21
8.06	19	23.30	0.74	8.86
10.06	21	18.20	0.22	8.86
14.06	25	15.80	0.22	8.86
17.06	28	14.60	0.09	9.06
20.06	31	10.80	0.07	9.06
22.06	33	9.30	0.07	9.06

Ammonia Out



Nitrite Out



Nitrate

