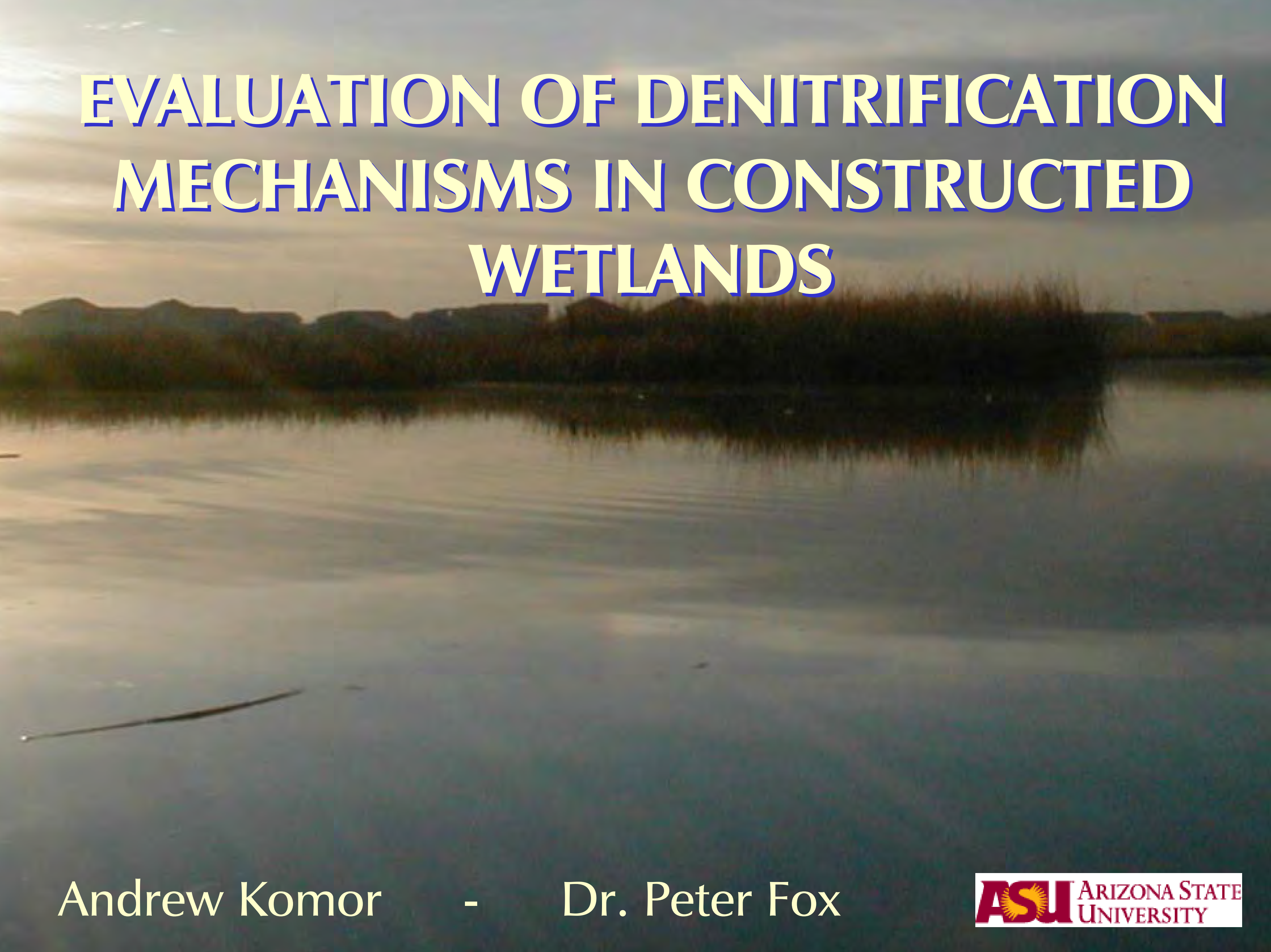

EVALUATION OF DENITRIFICATION MECHANISMS IN CONSTRUCTED WETLANDS

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Arizona State University

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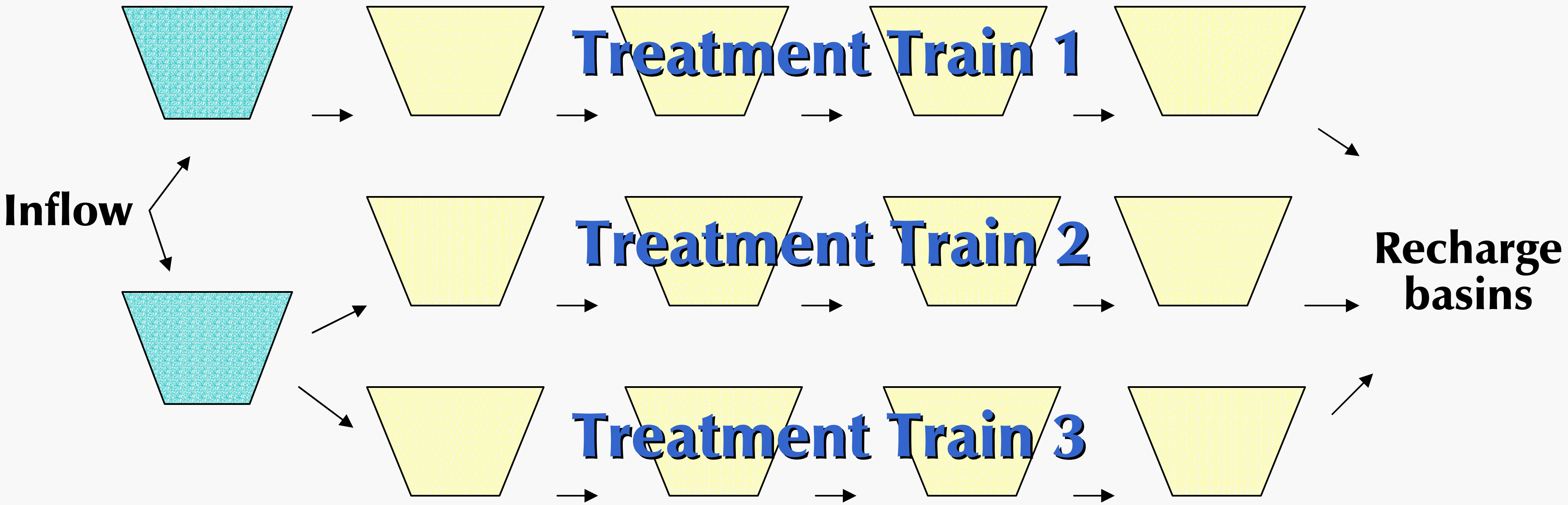
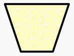
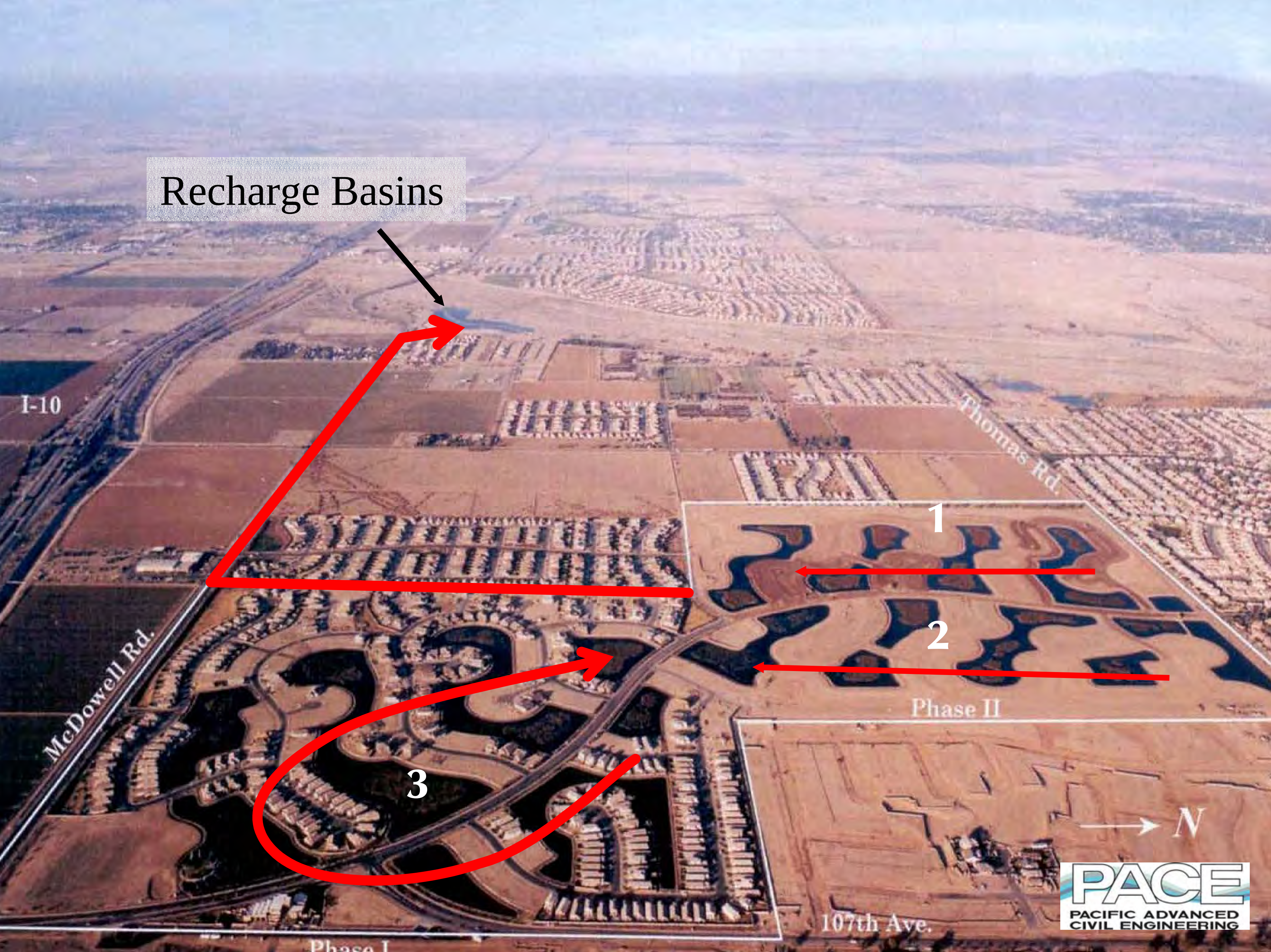
Avondale Treatment Schematic




- 5 acre equalization basin


- 5 acre wetland (2 acres vegetative Island)


Recharge Basins
I-10
Thomas Rd.
1
2
3
McDowell Rd.
Phase II
Phase I
107th Ave.
N
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BIOLOGICAL DENITRIFICATION

Under anoxic conditions, Nitrate (NO_3^-) is used as an electron acceptor for microbial respiration.

Wetland sediments generally contain:

1. Low concentrations of oxygen
2. Large quantities of electron donor

STUDY EXAMINED EFFECTS OF:

1. TYPE OF ELECTRON DONOR
2. SEDIMENT DEPTH
3. OXYGEN & ABIOTIC REACTIONS
4. PLANTS & ATTACHED MICROBES
5. INHIBITORS TO REDUCTION OF NITROUS OXIDE

How were Batch Experiments Performed?

1. Collect 1″ diameter sediment cores
2. Fill 165mL serum bottles with sediment, 80mL wetland water
3. Purge headspace with nitrogen gas

4. Ammend bottles with nitrate and electron donor
5. Mix bottles on shaker table in the dark
6. Monitor ionic concentrations with time

Wetland Microcosms

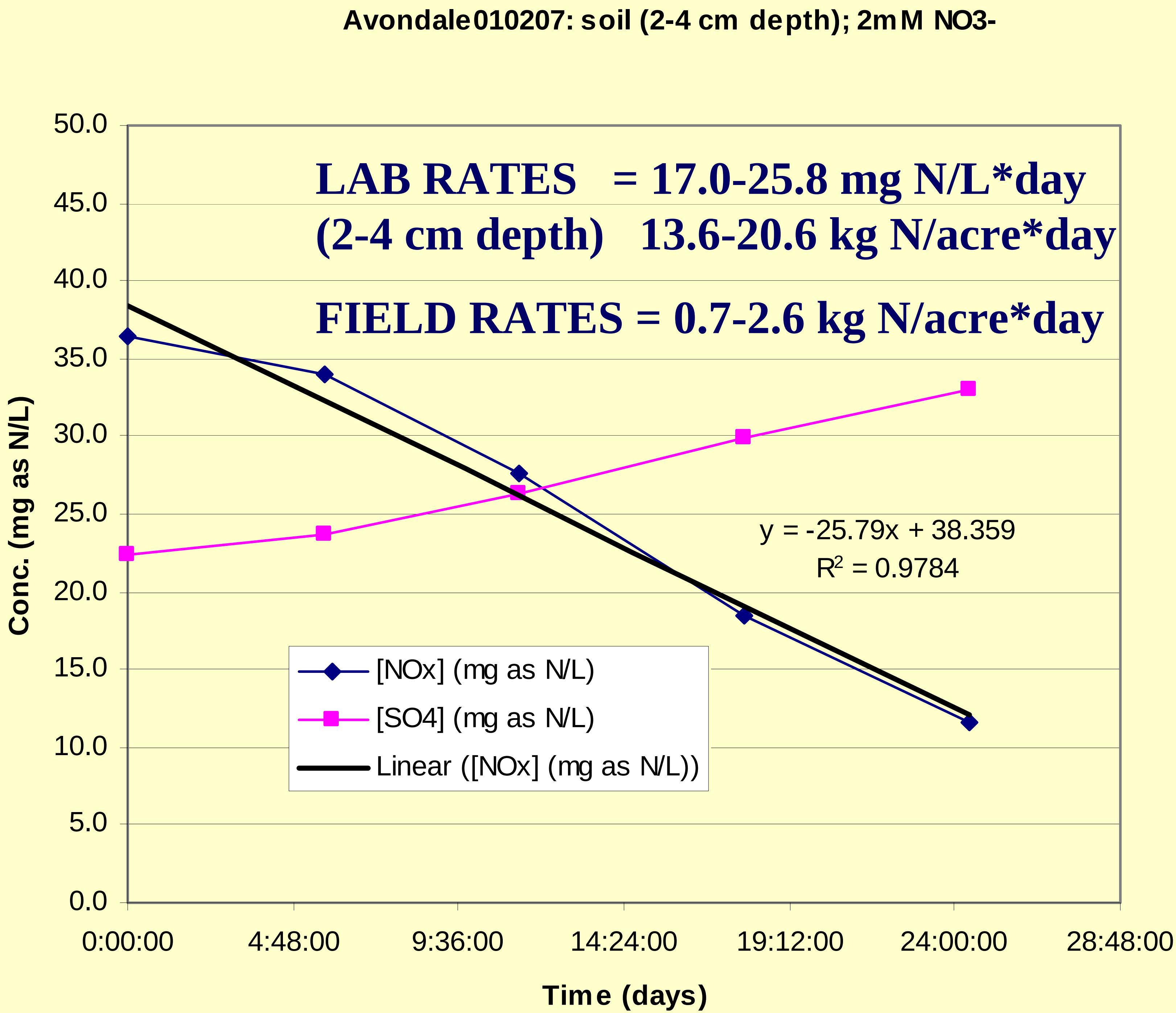
1. Organic/Inorganic Electron Donor

ORGANIC: heterotrophic denitrification

$NO3^{-} + 1.08\ CH_3OH + H^{+} = 0.065\ C_5H_7O_2N +$
$0.47\ N_{2(g)} + 0.76\ CO_2 + 2.4\ H_20$

INORGANIC: autotrophic denitrification

$NO_3^{-} + 0.74\ S^{2-} + 0.1886\ CO_2 = 0.74\ SO_4^{2-} +$
$0.47\ N_{2(g)} + 0.037\ C_5H_7O_2N + 0.1\ H^{+} + 0.37\ H_20$

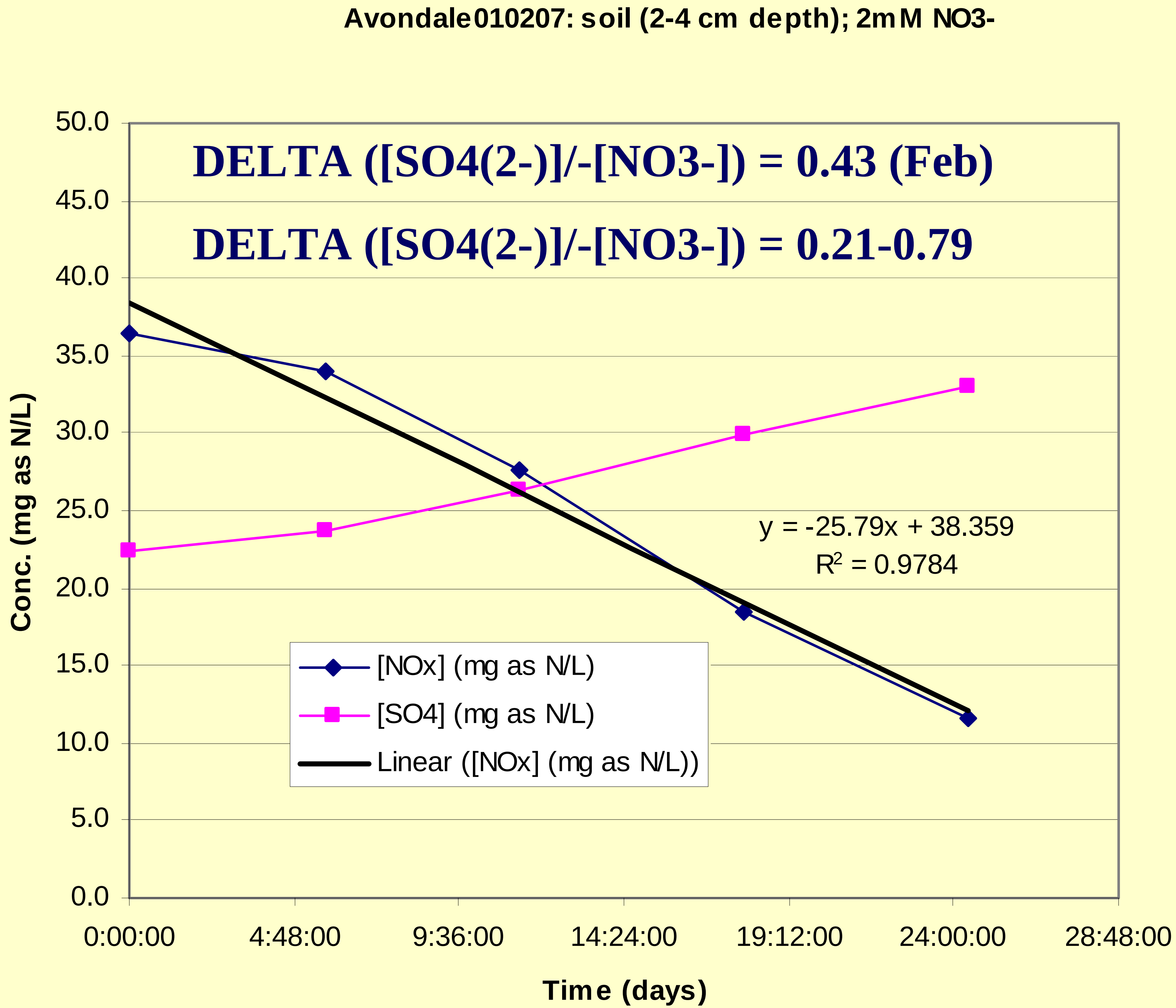

Avondale010207: soil (2-4 cm depth); 2mM NO3-
LAB RATES = 17.0-25.8 mg N/L*day
(2-4 cm depth) 13.6-20.6 kg N/acre*day
FIELD RATES = 0.7-2.6 kg N/acre*day
y = -25.79x + 38.359
R2 = 0.9784
[NOx] (mg as N/L)
[SO4] (mg as N/L)
Linear ([NOx] (mg as N/L))
Conc. (mg as N/L)
Time (days)
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
0:00:00
4:48:00
9:36:00
14:24:00
19:12:00
24:00:00
28:48:00

ORGANIC: heterotrophic denitrification

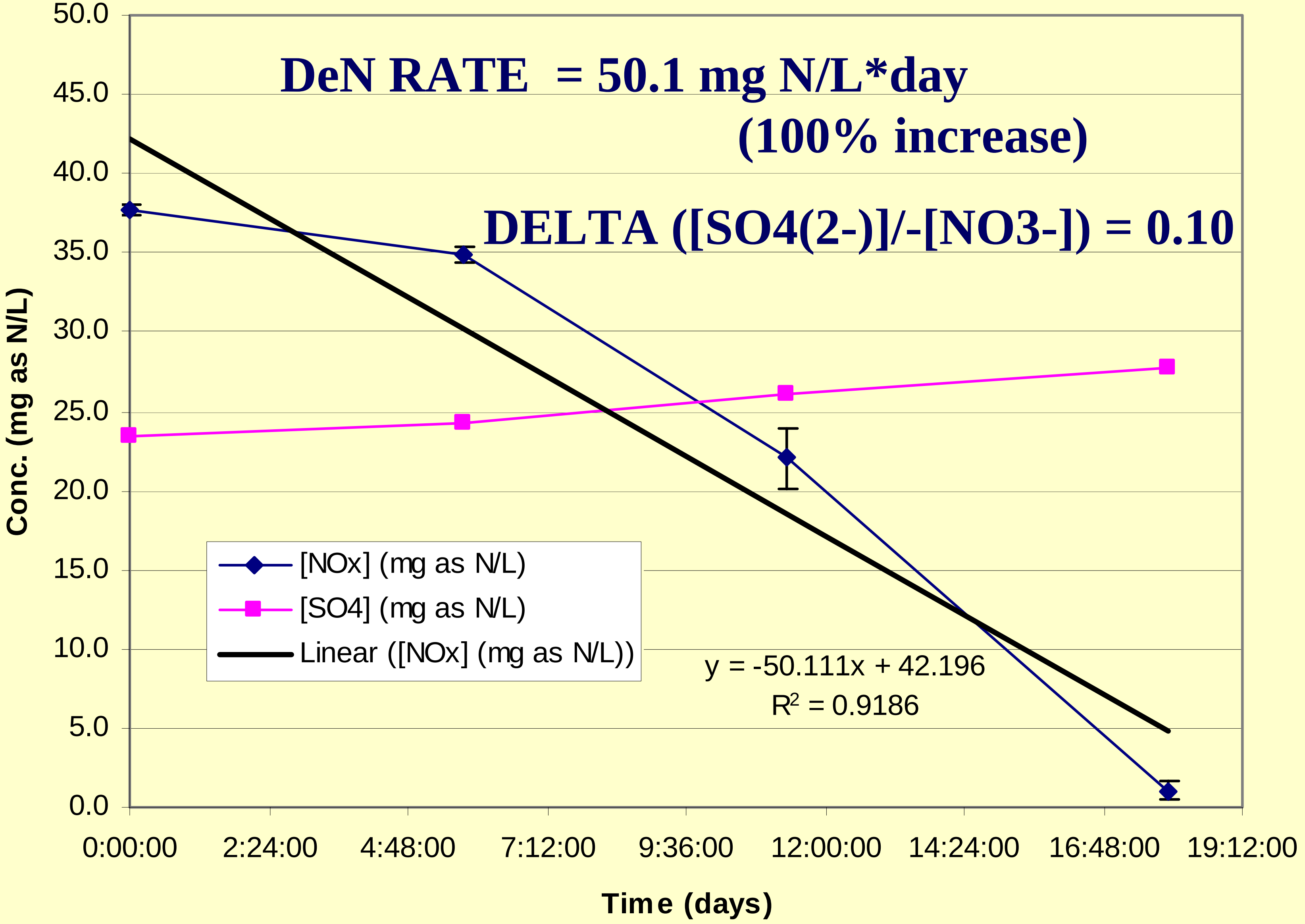
$NO3^- + 1.08\ CH_3OH + H^+ = 0.065\ C_5H_7O_2N + 0.47\ N_{2(g)} + 0.76\ CO_2 + 2.4\ H_20$

INORGANIC: autotrophic denitrification

$NO_3^- + 0.74\ S^{2-} + 0.1886\ CO_2 = 0.74\ SO_4^{2-} + 0.47\ N_{2(g)} + 0.037\ C_5H_7O_2N + 0.1\ H^+ + 0.37\ H_20$


Avondale010207: soil (2-4 cm depth); 2mM NO3-
DELTA ([SO4(2-)]/-[NO3-]) = 0.43 (Feb)
DELTA ([SO4(2-)]/-[NO3-]) = 0.21-0.79
y = -25.79x + 38.359
R^2 = 0.9784
[NOx] (mg as N/L)
[SO4] (mg as N/L)
Linear ([NOx] (mg as N/L))
Conc. (mg as N/L)
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
0:00:00
4:48:00
9:36:00
14:24:00
19:12:00
24:00:00
28:48:00
Time (days)

Avondale010207: soil (2-4 cm), 2mM NO3-, 2g finely chopped fresh plants



Addition of Sulfur:

Increased rates 64% in August

Decreased rates 51% in November

Effect dependent on sediment characteristics

Enriched batch tests **(w/negligible remaining electron donor):**

Autotrophic substrate utilization 68% of heterotrophic rate

2. Sediment Depth

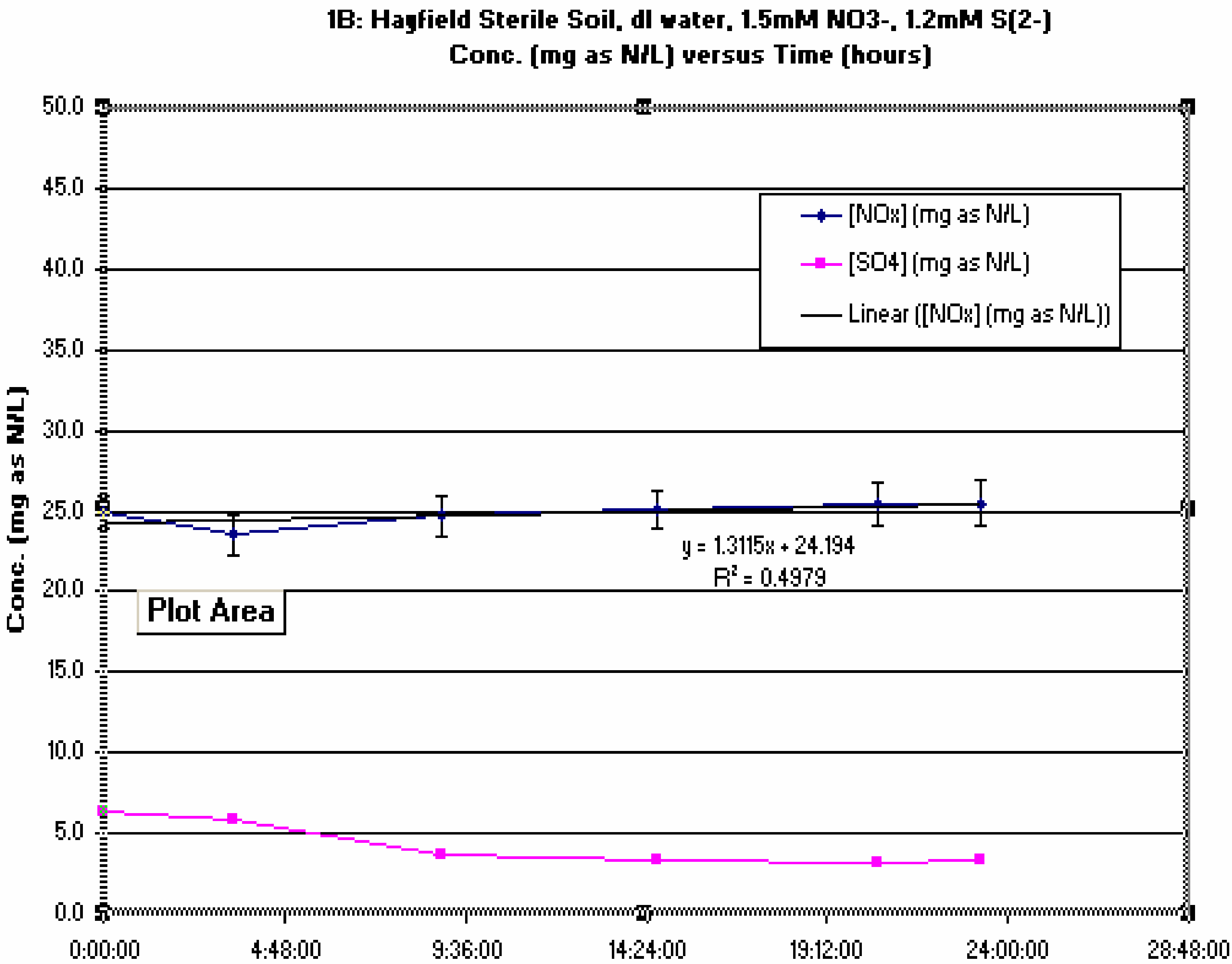
DENITRIFICATION RATE (mg N/L*day)

	NO3- ONLY	NO3- + S(2-)
0-2 cm depth (sandy/leafy)	13.7	21.4
2-4 cm depth (black sediment)	19.4	14.9

3. Oxygen & Abiotic Reactions

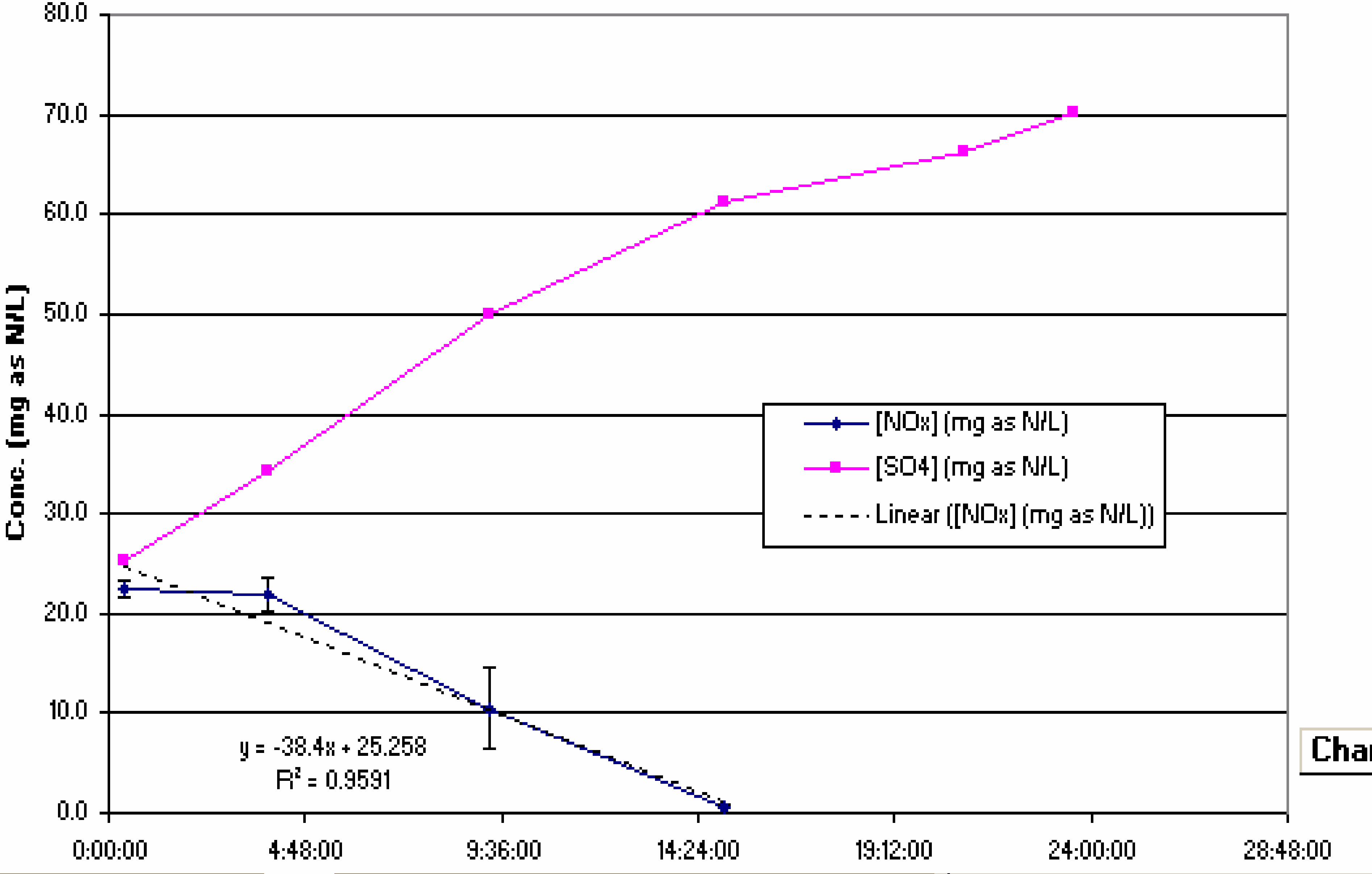
Microcosm Experiments Performed With:

1. Headspace purged w/air instead of nitrogen
2. Sterilized soil and water



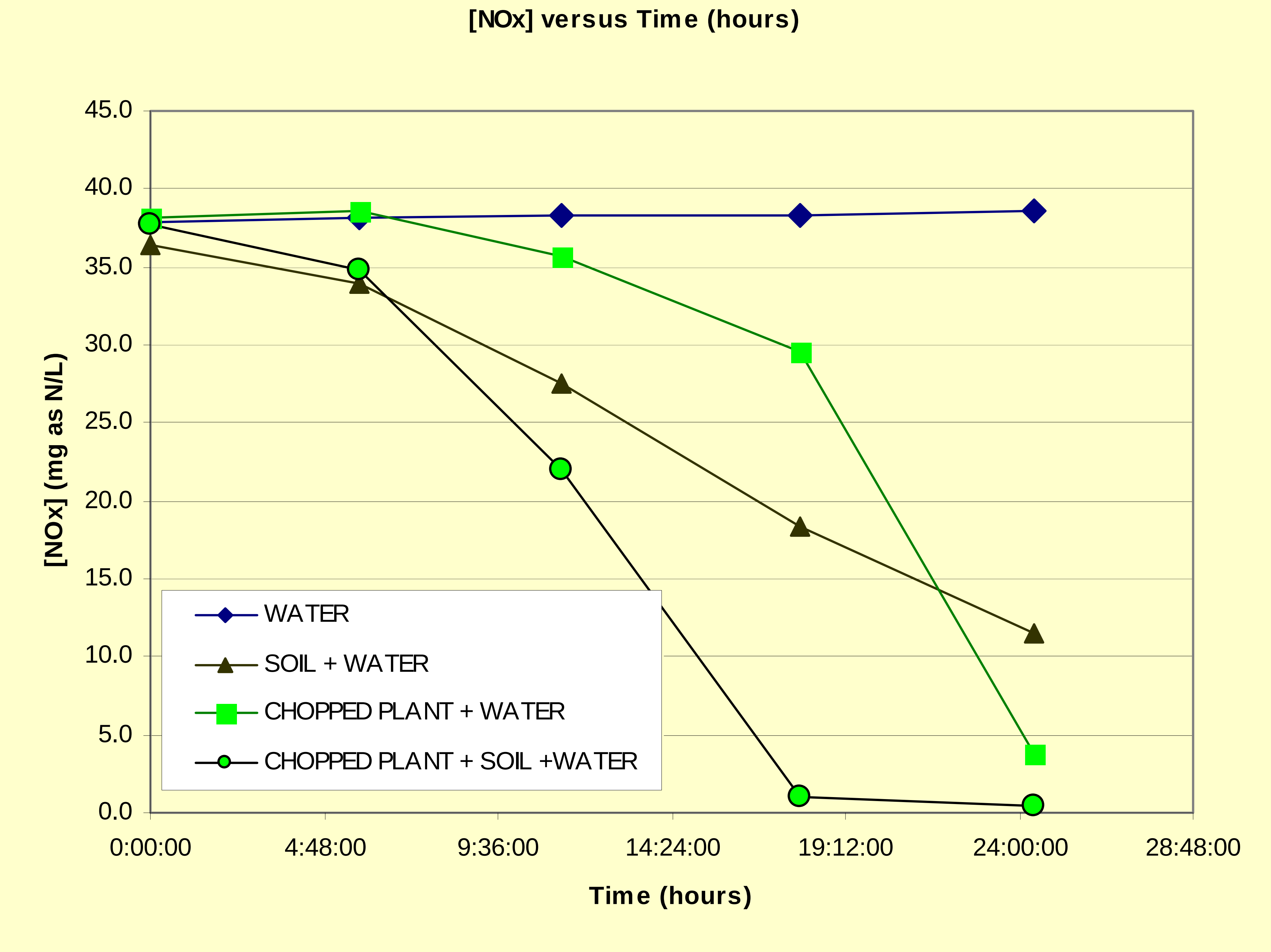
1B: Hagfield Sterile Soil, dI water, 1.5mM NO3-, 1.2mM S(2-)
Conc. (mg as N/L) versus Time (hours)
Conc. (mg as N/L)
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
0:00:00
4:48:00
9:36:00
14:24:00
19:12:00
24:00:00
28:48:00
[NOx] (mg as N/L)
[SO4] (mg as N/L)
Linear ([NOx] (mg as N/L))
y = 1.3115x + 24.194
R² = 0.4979
Plot Area

Figure B1E. (8-7-00) Conc. (mg as N/L) versus Time (hours)
Air Headspace, 20 g Hayfield Soil, 1.5 mM NO3-, 1.2mM S(2-)



4. Plants and Attached Microbes

SAMPLE	[DOC]avg (mg/L)
Water	2
Soil + water	4
Chopped plants + water	55
Cut plants + water	30
Oven-dried plants + water	325

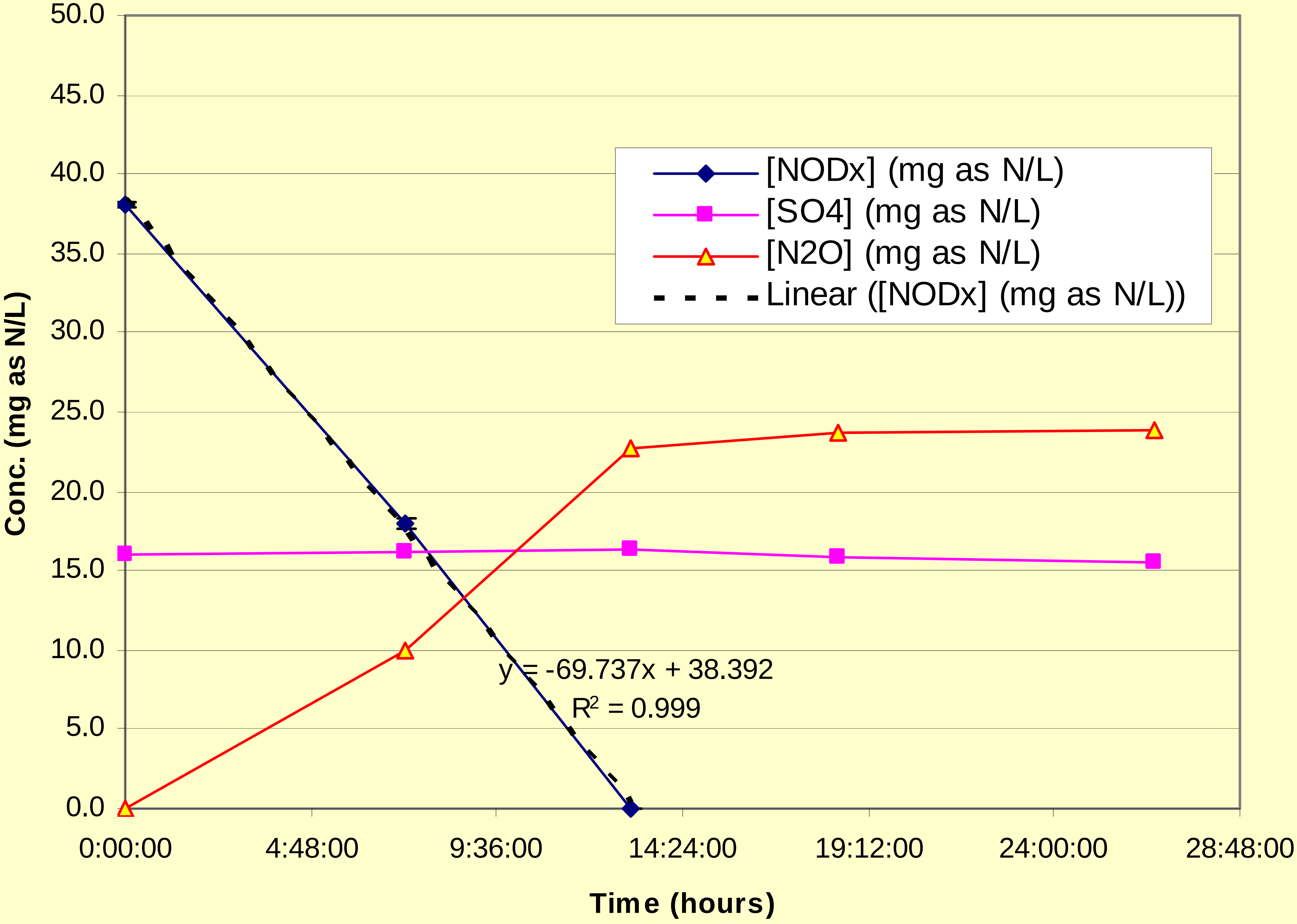

[NOx] versus Time (hours)
[NOx] (mg as N/L)
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
0:00:00
4:48:00
9:36:00
14:24:00
19:12:00
24:00:00
28:48:00
Time (hours)
WATER
SOIL + WATER
CHOPPED PLANT + WATER
CHOPPED PLANT + SOIL +WATER

5. Inhibitors to Reduction of Nitrous Oxide to Nitrogen Gas

ACETYLENE

SULFIDE

Figure B5C. (3-15-01) Conc. (mg as N/L) versus Time (hours)
NdeN Sludge, 2mM NO3-, 1.6mM Glucose, 10% Acetylene Headspace



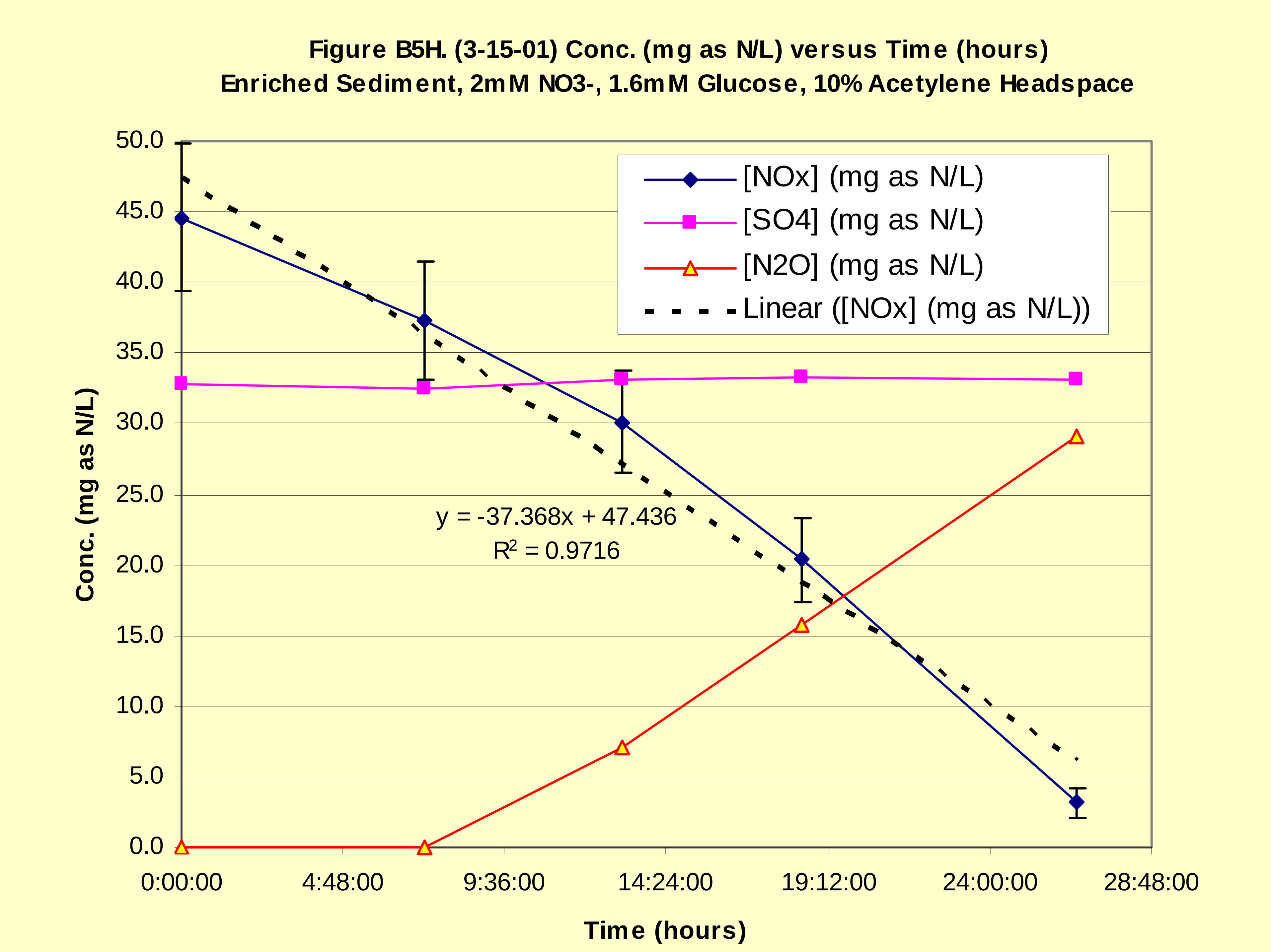


Figure B5H. (3-15-01) Conc. (mg as N/L) versus Time (hours)
Enriched Sediment, 2mM NO3-, 1.6mM Glucose, 10% Acetylene Headspace
[NOx] (mg as N/L)
[SO4] (mg as N/L)
[N2O] (mg as N/L)
Linear ([NOx] (mg as N/L))
y = -37.368x + 47.436
R2 = 0.9716
Conc. (mg as N/L)
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
0:00:00
4:48:00
9:36:00
14:24:00
19:12:00
24:00:00
28:48:00
Time (hours)

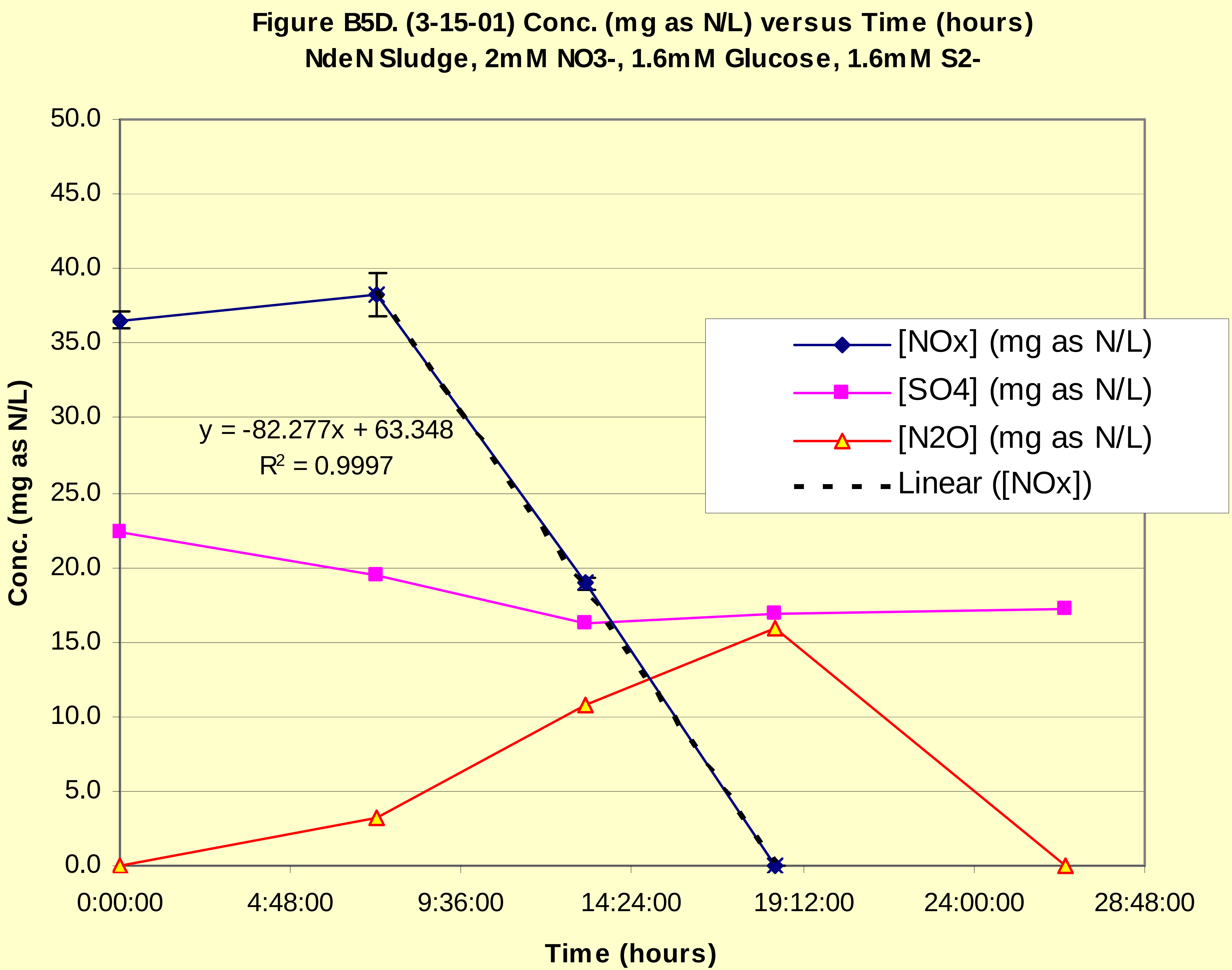


Figure B5D. (3-15-01) Conc. (mg as N/L) versus Time (hours)
NdeN Sludge, 2mM NO3-, 1.6mM Glucose, 1.6mM S2-
Conc. (mg as N/L)
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
y = -82.277x + 63.348
R2 = 0.9997
[NOx] (mg as N/L)
[SO4] (mg as N/L)
[N2O] (mg as N/L)
Linear ([NOx])
0:00:00
4:48:00
9:36:00
14:24:00
19:12:00
24:00:00
28:48:00
Time (hours)

Figure B5I2. (3-15-01) Conc. (mg as N/L) versus Time (hours)
Enriched Sediment, 2mM NO3-, 1.6mM Glucose, 4.8mM S2-

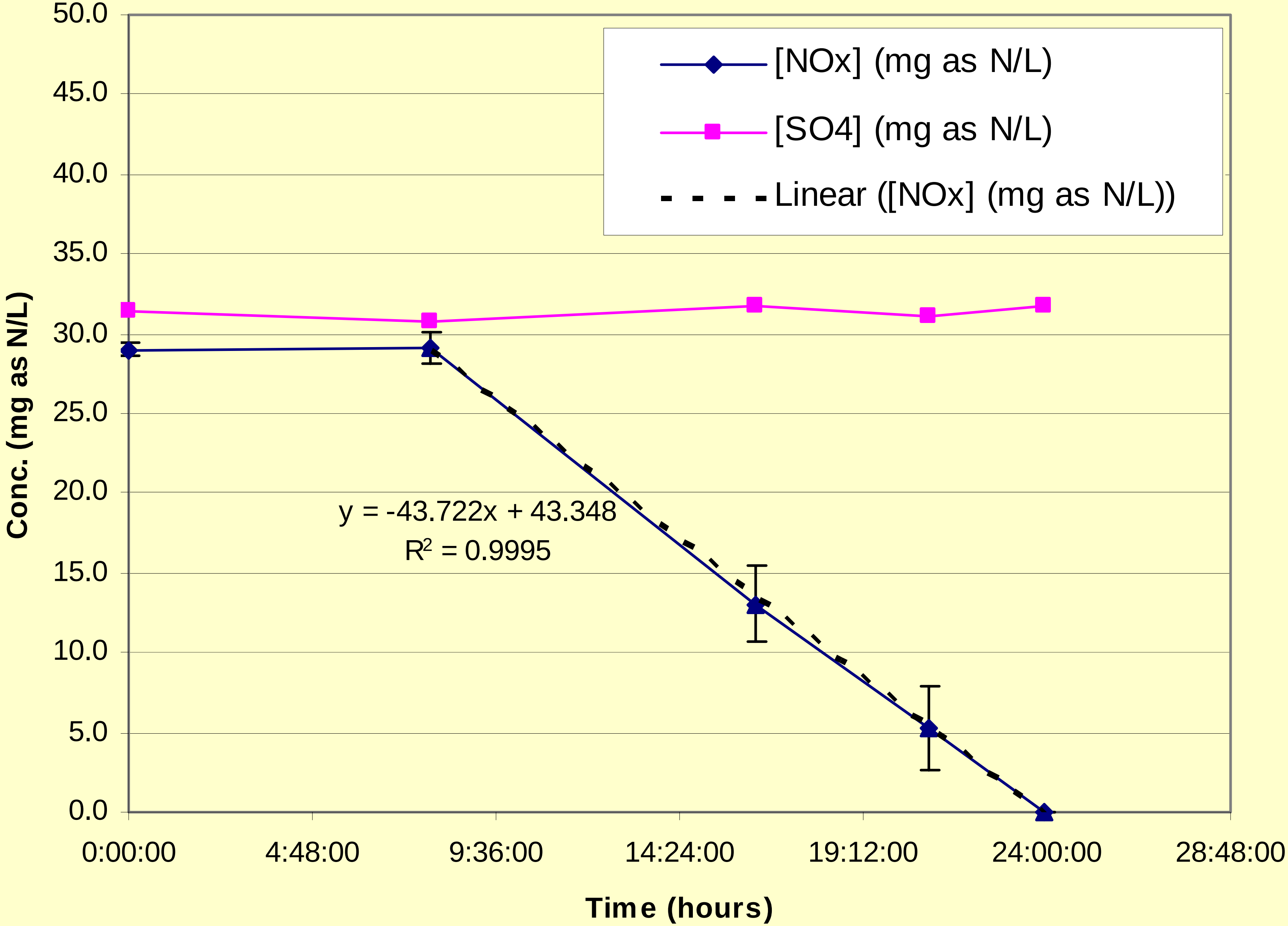
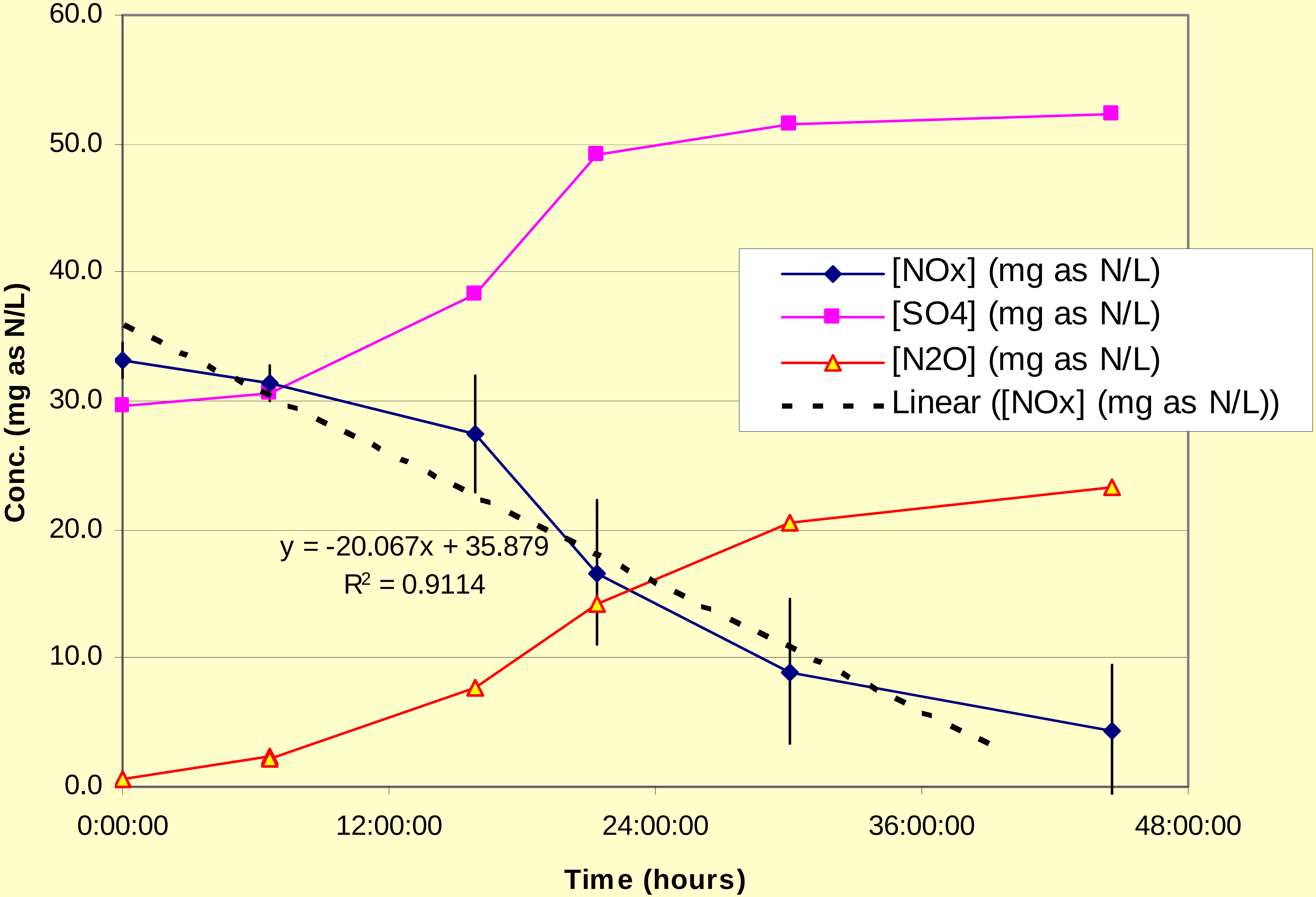


Figure B5E2. (3-15-01) Conc. (mg as N/L) versus Time (hours)
Enriched Sediment 2-4 cm, 2mM NO3-, 1.6mM S(2-), 10% Acetylene Headspace



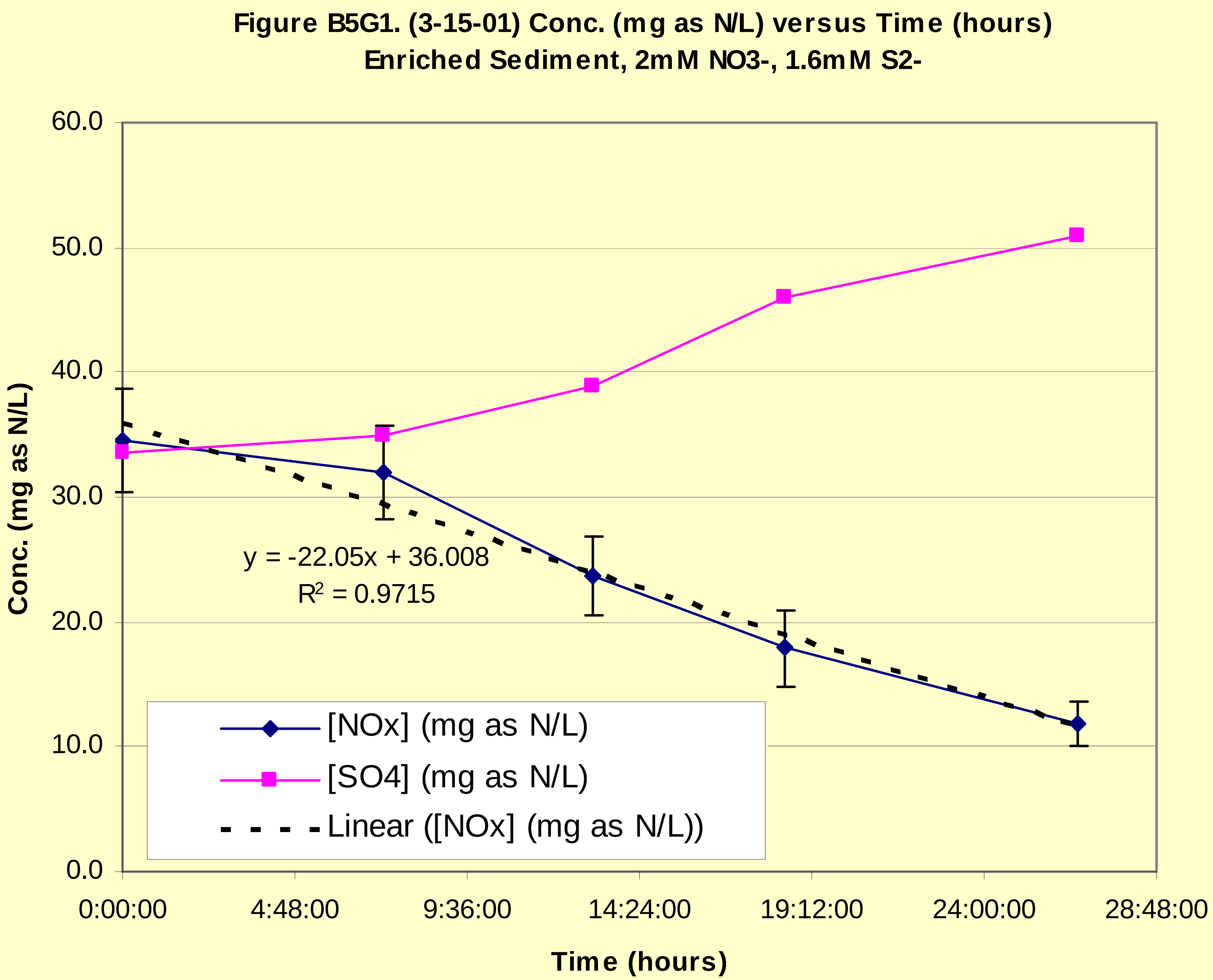


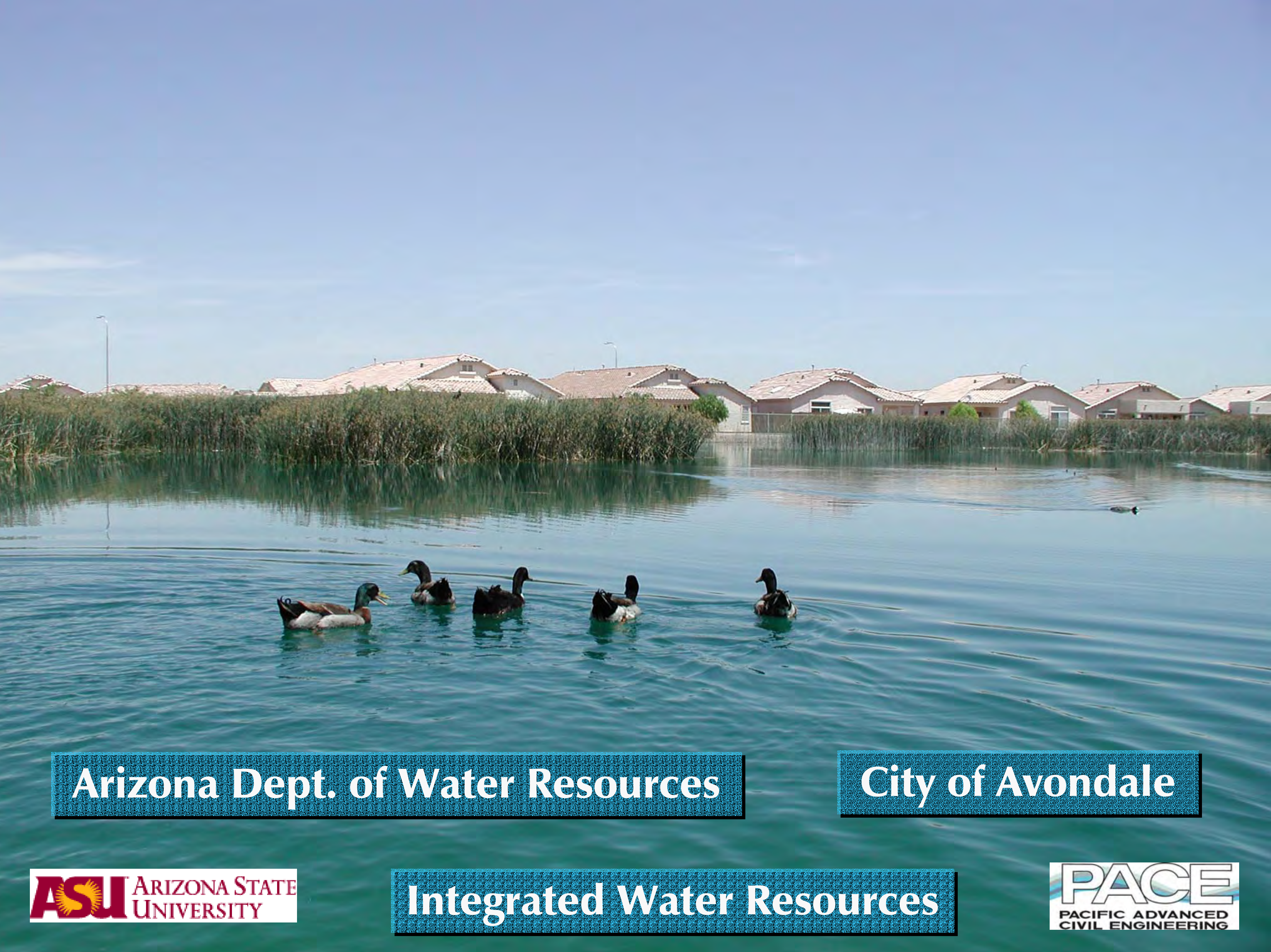
Figure B5G1. (3-15-01) Conc. (mg as N/L) versus Time (hours)
Enriched Sediment, 2mM NO3-, 1.6mM S2-
Conc. (mg as N/L)
60.0
50.0
40.0
30.0
20.0
10.0
0.0
y = -22.05x + 36.008
R2 = 0.9715
[NOx] (mg as N/L)
[SO4] (mg as N/L)
Linear ([NOx] (mg as N/L))
0:00:00
4:48:00
9:36:00
14:24:00
19:12:00
24:00:00
28:48:00
Time (hours)

Conclusions

1. Denitrification via autotrophic & heterotrophic reactions occurred simultaneously , with rates dependent on donor available (seasonal)
2. Similar populations of microbes exist from 0-2 cm as 2-4 cm depth of sediment
3. Non-biological denitrification was insignificant
4. Oxygen did not effect denitrification rates; sulfide oxidation likely scavanged available O2

Conclusions (cont…)

4. Denitrifiers only present in sediment & plants
5. Wetland plants provide attachment for microbes as well as addition of electron donor
6. Acetylene inhibited reduction of N_2O -> N_2, but not the rate of denitrification for both rxn's
7. Sulfide delayed reduction of nitrate & produced some N_2O for heterotrophs, autotrophs inhibited by high pH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Integrated Water Resources
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