Measuring river bed denitrification: benefits of using both a laboratory and in situ approach

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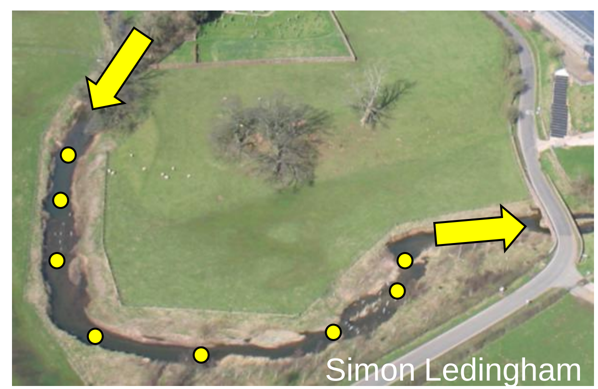
The River Leith (Cumbria, UK)

- Groundwater-fed, gaining reach
- River bed = sand + gravel over s/stone
- Lowland area, agricultural land use
- Groundwater nitrate > surface water nitrate

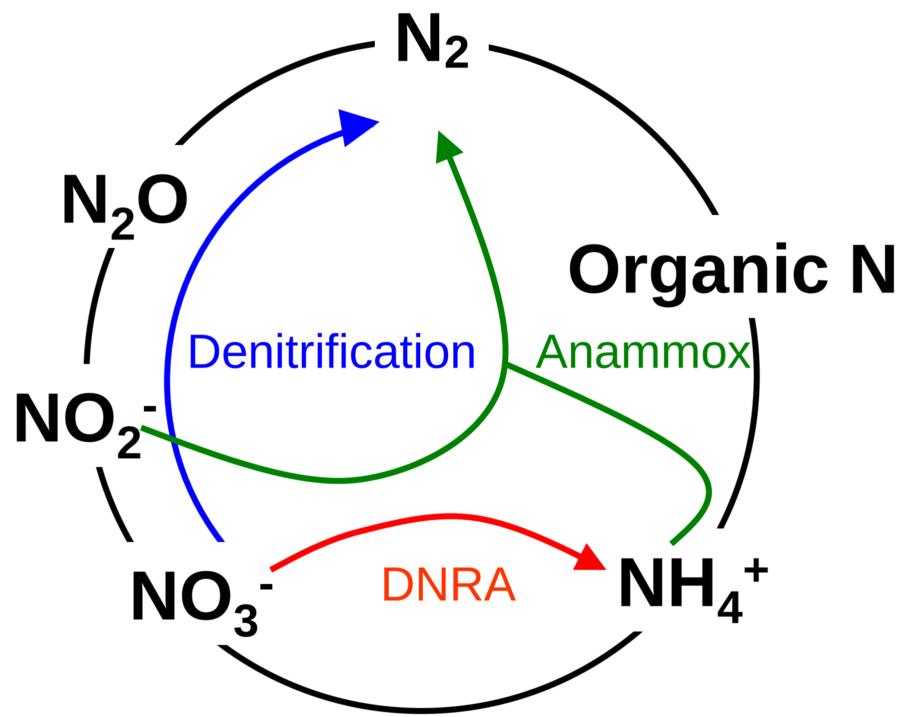
Methods (Field)

- Sediment cores from piezometer installation screened for pathways of dissimilatory nitrate reduction (June 2009)
- 8 sites (4 riffles, 4 pools) within 250m reach
- 5cm slices from 4 depths (5-10cm, 15-20cm, 40-50cm, 60-86cm)



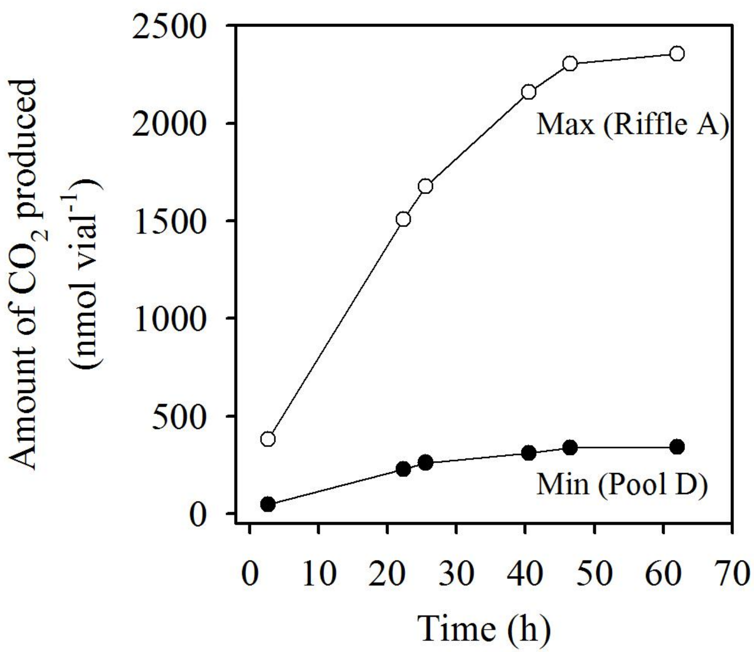
Methods (Laboratory)

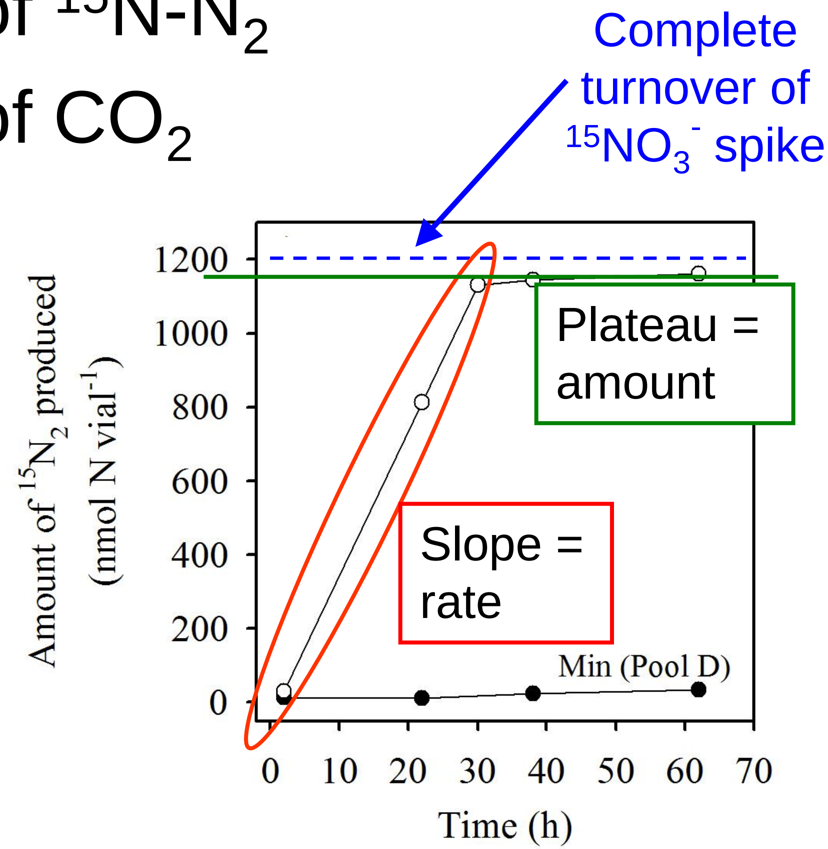
- ^{15}N assay:
  - sediment slurries
  - $^{15}NO_3^-$ spike
  - Measure production of $^{15}N\text{-}N_2$
- “Bioavailable” organic carbon:
  - incubation of sediment
  - Measure production of CO_2



Methods (Laboratory)

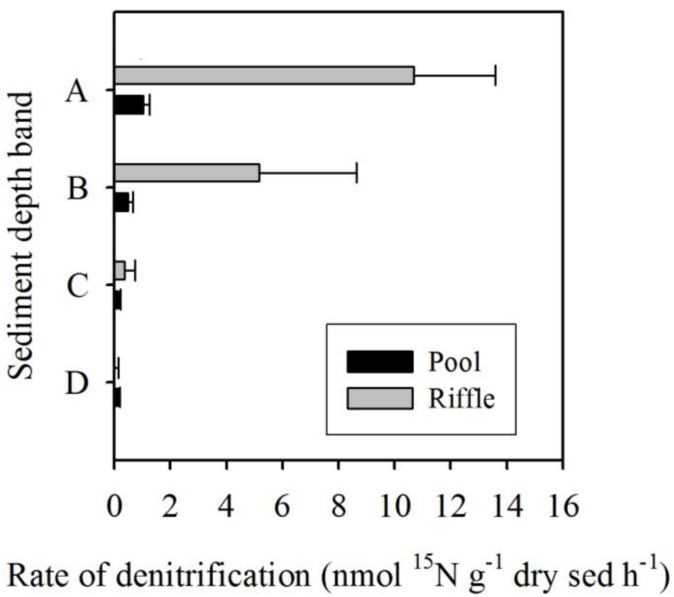
- ^{15}N assay/ "Bioavailable" carbon:
  - Measure production of ^{15}N-N_2
  - Measure production of CO_2

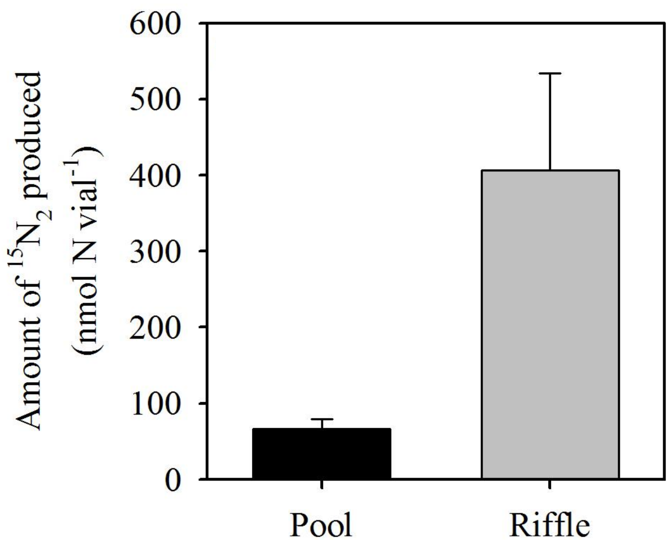




Denitrification potential

- Denitrification potential:
  - decayed with depth
  - was highest in riffle sediments

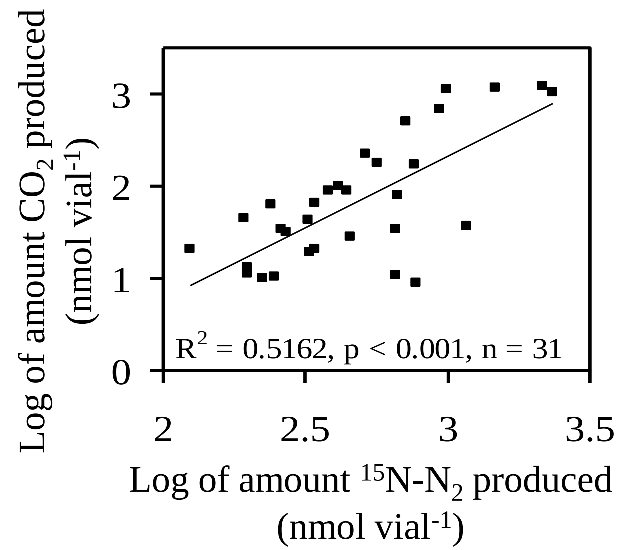




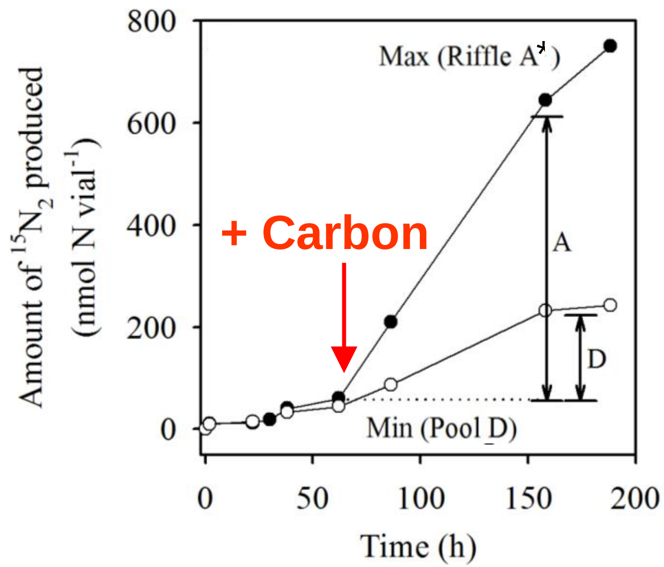
(ANOVA: depth $F_{3,23} = 8.43$, $p < 0.001$, habitat $F_{1,23} = 14.14$, $p = 0.001$, depth x habitat $F_{3,23} = 5.95$, $p = 0.004$)

Denitrification potential

- Denitrification potential:
  - was correlated with CO_2 production



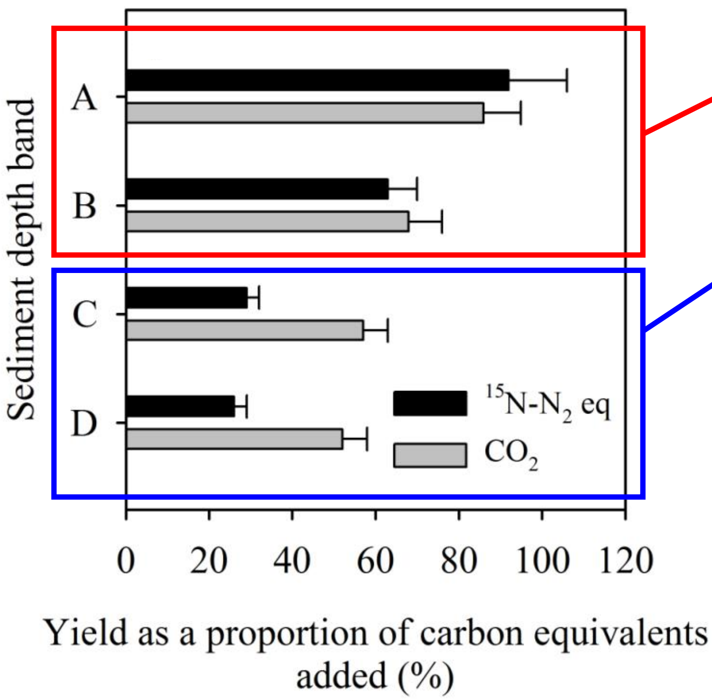
- Carbon addition expmt.
  - Response of denitrification to complex carbon?
  - Carbon source = bacteriological peptone



Denitrification potential

- $5\ \text{"}CH_2O\text{"} + 4NO_3^- \rightarrow 2N_2 + (4HCO_3^-, CO_2) + 3H_2O$
  - 920 nmol C added: 920 nmol CO_2 = 100% yield

    736 nmol N_2 (as N) = 100% yield



Yield $^{15}N\text{-}N_2 \approx$ Yield CO_2, denitrifiers can access all carbon that is bioavailable

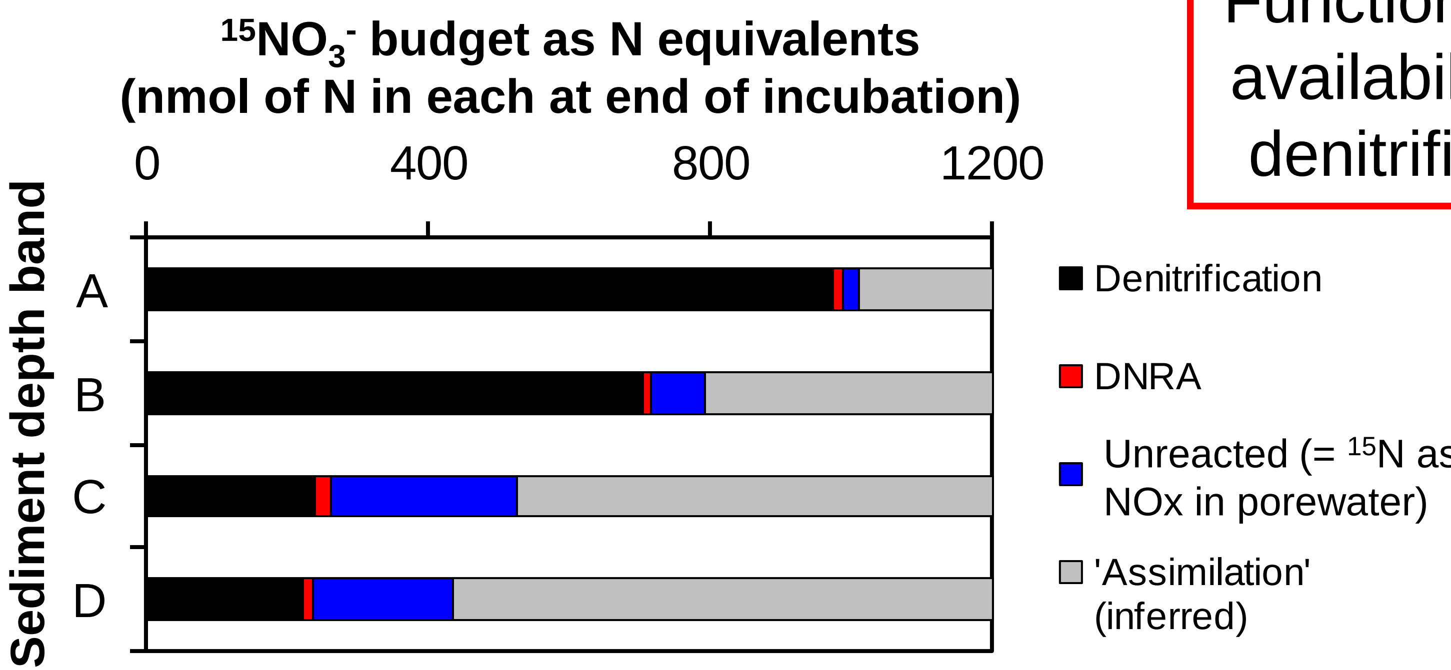
Yield $^{15}N\text{-}N_2 <$ Yield CO_2, denitrifiers **cannot** access all carbon that is bioavailable

At depth in the river bed (> 40cm) denitrification capacity is reduced by inability of denitrifiers to metabolise complex carbon.

Denitrification potential

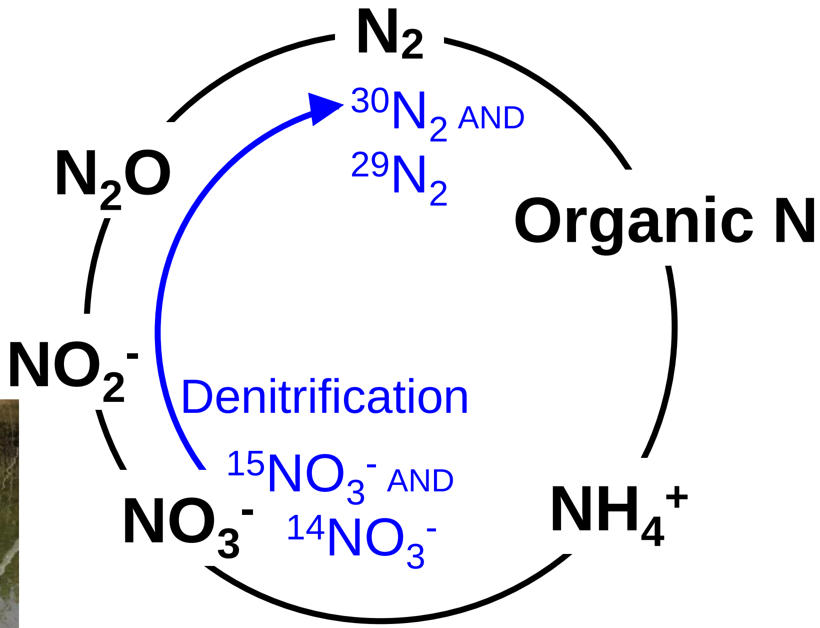
- Fate of nitrate also changes with depth in the sediment profile
  - shallow sediments = denitrification
  - deeper sediments = “assimilation”

Function of C availability to denitrifiers?



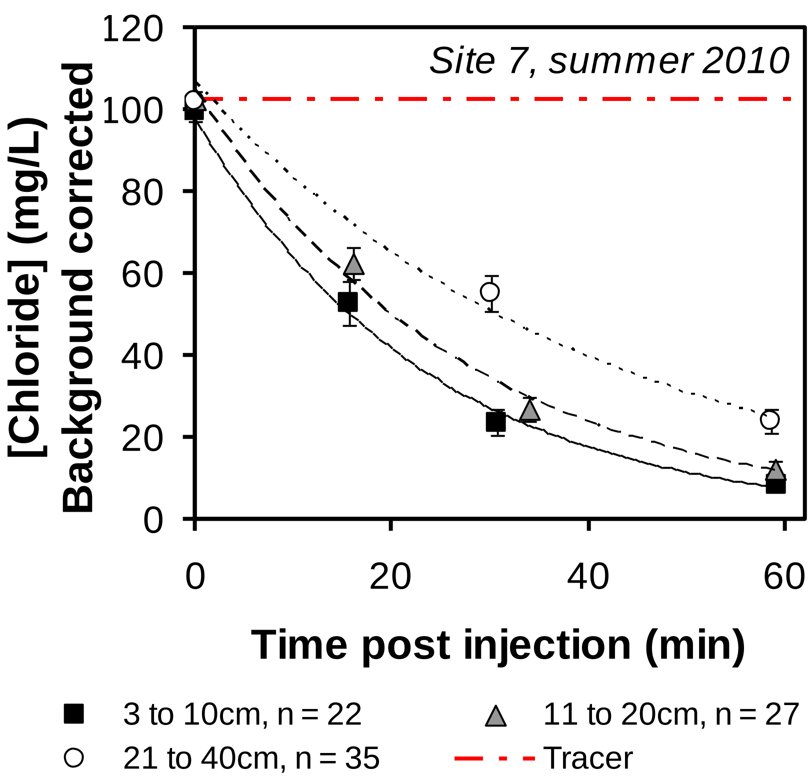
Methods (in situ)

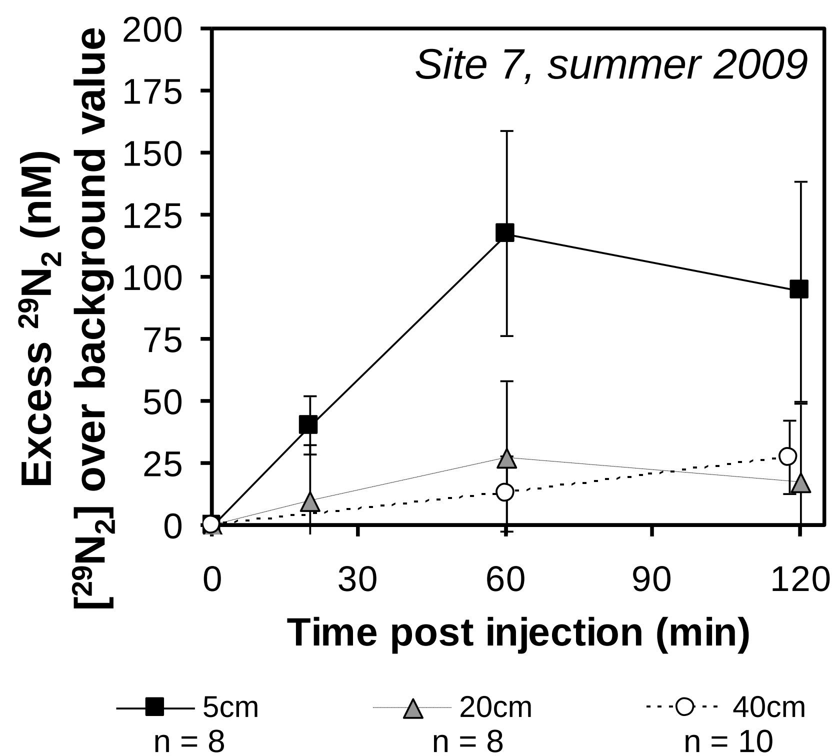
- Inject $^{15}NO_3^-$ into the river bed and collect samples over time
  - – Ambient $[NO_3^-]$
  - – Match [dissolved O_2]
  - – Syn. river water + KCl



Methods (in situ)

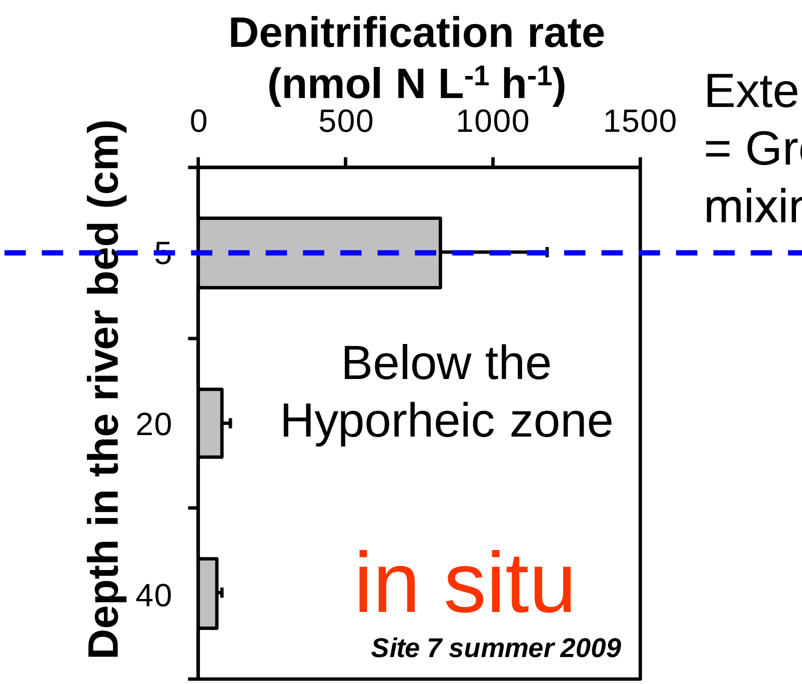
- ^{15}N label was recovered as N_2 however loss of tracer by advection/ dispersion was substantial



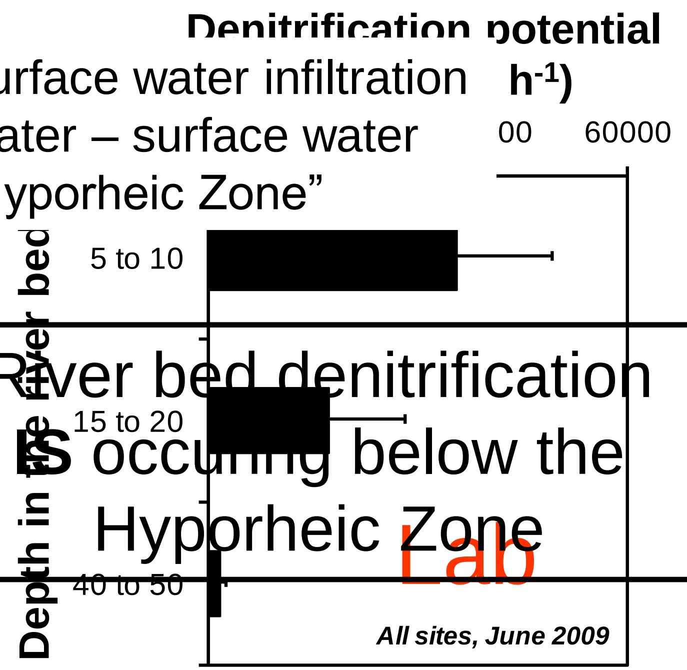


Denitrification: potential vs rate

- Potentials measured in the lab were higher than in situ rates
  - Effect of oxygen?



Extent of surface water infiltration = Groundwater – surface water mixing = "Hyporheic Zone"



River bed denitrification **IS** occurring below the Hyporheic Zone

Conclusions

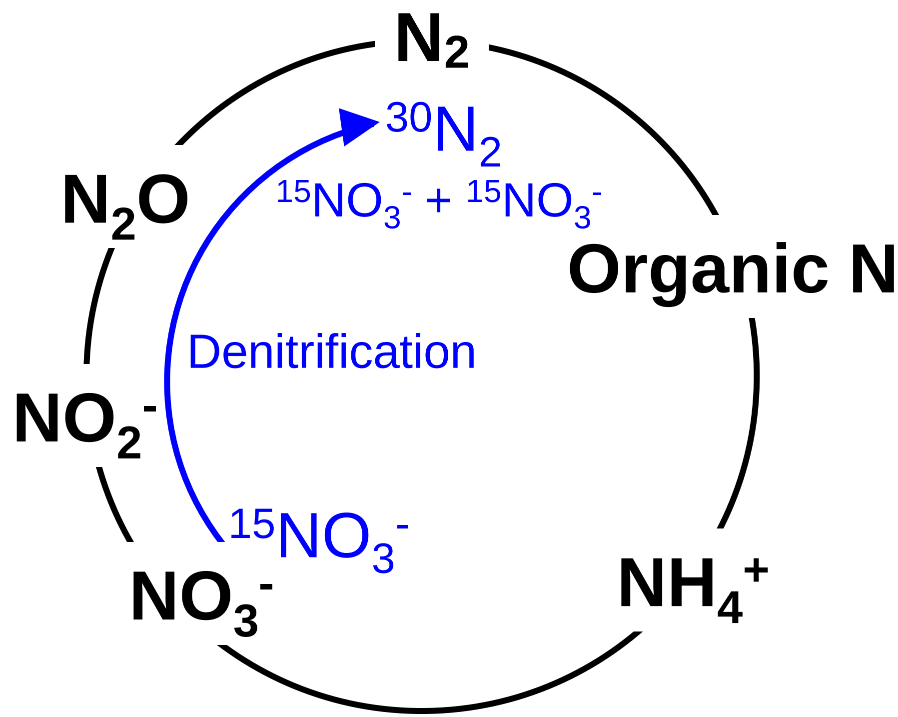
- Denitrification capacity within the river bed is affected by the differential ability of denitrifiers, and the microbial community as a whole, to metabolise an array of organic substrates

- In situ method offers a means by which laboratory findings (and more) can be tested in the river bed

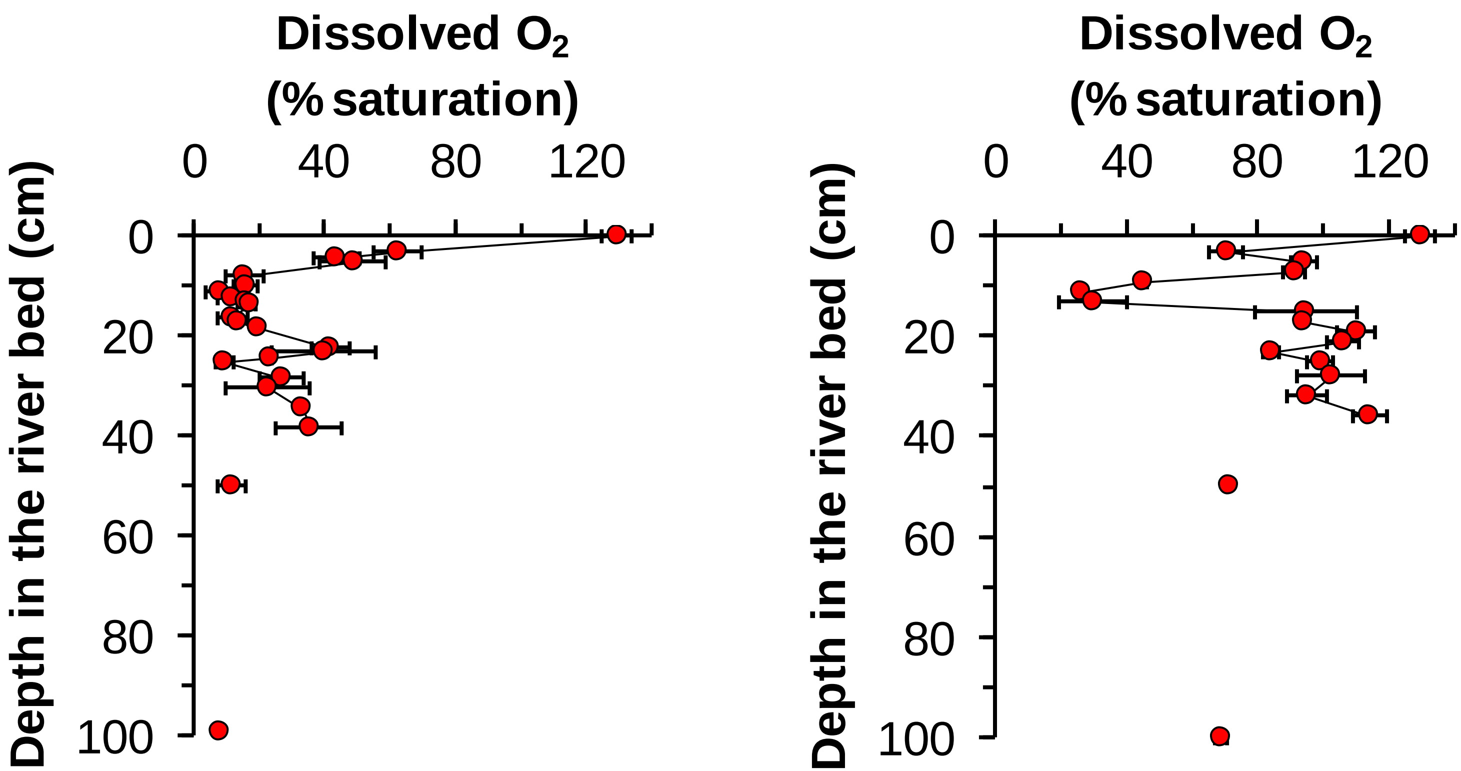
Methods (Laboratory)

- ^{15}N assay:
  - sediment slurries

    (< 2mm sediment + syn. river water)
  - O_2 removed
  - Ambient NO_3^- removed
  - Add $^{15}NO_3^-$ spike
  - Measure production of ^{15}N-N_2
- "Bioavailable" organic carbon:
  - Aerobic incubation of < 2mm sediments
  - Measure production of CO_2



Dissolved oxygen (in situ)



Sediment characteristics

Habitat	Band	n =	Depth (cm)	Substrate	Median grain size (μm)	Organic C (%)*
Pool	A	4	5-10	Sand with gravel	465	0.05 ± 0.01
Pool	B	4	15-20	Sand with silt	283	0.06 ± 0.1
Pool	C	4	45-50	Sand with silt	297	0.02 ± 0.01
Pool	D	4	60-86	Sand with silt	271	0.02 ± 0.01
Riffle	A	4	5-10	Sand and gravel	1589	0.07 ± 0.05
Riffle	B	4	15-20	Sand and gravel	1485	0.05 ± 0.03
Riffle	C	4	40-50	Sand with silt + gravel	439	0.02 ± 0.02
Riffle	D	3	64-85	Sand with silt	300	0.03 ± 0.02

Denitrification potential data

Habitat/ Depth	CO_2 produced (nmol vial^{-1})	CO_2 flux (nmol g^{-1} h^{-1})	^{15}N-N_2 recovered (nmol N vial^{-1})	Denitrification rate * (nmol N g^{-1} h^{-1})
Pool A	719 ± 167	8 ± 2.3	119 ± 34	1 ± 0.2[a]
Pool B	412 ± 111	6 ± 1.4	72 ± 18	0.5 ± 0.16[a]
Pool C	272 ± 32	4 ± 0.5	35 ± 11	0.1 ± 0.08[a]
Pool D	283 ± 36	4 ± 0.3	41 ± 10	0.2 ± 0.05[a]
Riffle A	1676 ± 371	15 ± 3	976 ± 164	11 ± 3[b]
Riffle B	760 ± 125	8 ± 1.2	469 ± 268	5 ± 3.5[c]
Riffle C	422 ± 98	5 ± 1.3	70 ± 53	0.4 ± 0.36[a]
Riffle D	400 ± 190	4 ± 1.9	10 ± 1	0.00 ± 0.04[a]