# Monitoring nitrogen processing in constructed wetlands: two stable isotope approaches.

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## INTRODUCTION:

Constructed wetlands reduce nitrogen (N) loads in storm water runoff in the urban landscape through removal via denitrification (DNF) [1, 2, 3] and temporary storage in plant material via assimilation. We propose two stable isotope approaches that would provide more information and improve the management of N in constructed wetlands.

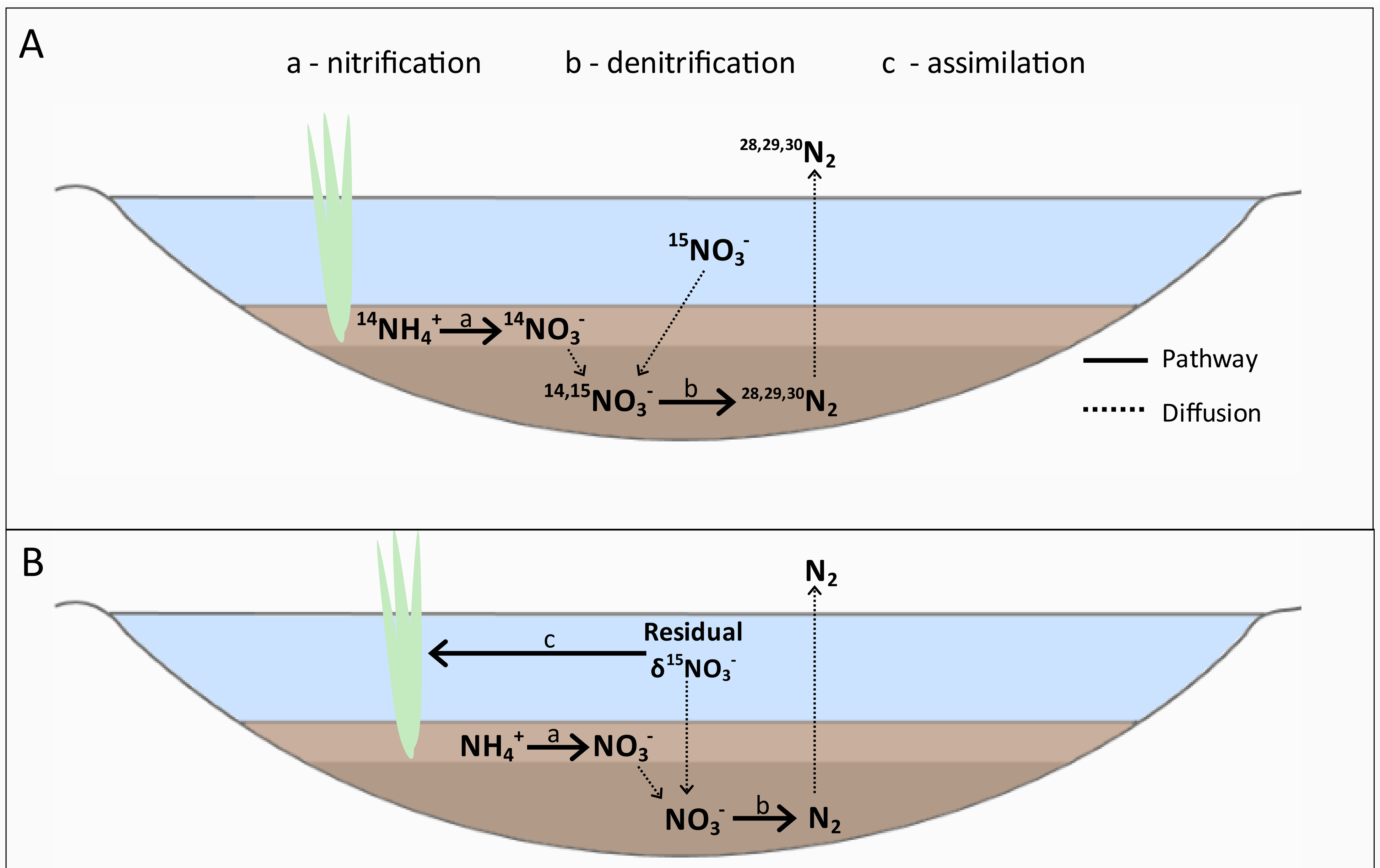
## STABLE ISOTOPE APPROACHES:

### Isotope Pairing Technique (IPT) - Fig 1A

Rates of denitrification were determined using the $^{15}$N-stable isotope pairing technique [4]. Rates are calculated from the accumulation of $^{15}N$-$N_2$ over time. The enriched samples were analysed on an isotope ratio mass spectrometer coupled to a gas chromatograph.

### Dual isotopes of $\delta^{15}N$ & $\delta^{18}O$ in $NO_3^-$ - Fig 1B

For the dual isotopic composition of nitrate at the natural abundance level, $NO_2^-$ was removed with sulfamic acid [5]. Then $NO_3^-$ was converted to $NO_2^-$ through a cadmium reduction step and then to $N_2O$ in a final 1:1 azide : acetic acid step [6]. The samples for $\delta^{15}N$ and $\delta^{18}O$ of $N_2O$ were analysed on an isotope ratio mass spectrometer coupled to a gas chromatograph fitted to a cryoprep system cooled using liquid $N_2$




**Denitrification** $\delta^{15}N$ in residual $NO_3^-$ **increases** by 0 - 2 ‰
**Assimilation** $\delta^{15}N$ in residual $NO_3^-$ **increases** by 5 - 20 ‰
**Nitrification** $\delta^{15}N$ & $\delta^{18}O$ in residual $NO_3^-$ **decreases** up to 40‰ ($\delta^{15}N$) and 10‰ ($\delta^{18}O$)

**Figure 1 A) $^{15}$N-labelled $NO_3^-$ used to measure denitrification and the portion of nitrification-denitrification coupling and B) Dual isotopic composition ($\delta^{15}N$ & $\delta^{18}O$) of $NO_3^-$ a qualitative measure of denitrification, assimilation and nitrification.**

## CASE STUDY

# CASCADES ON CLYDE WETLAND

### Isotope Pairing Technique

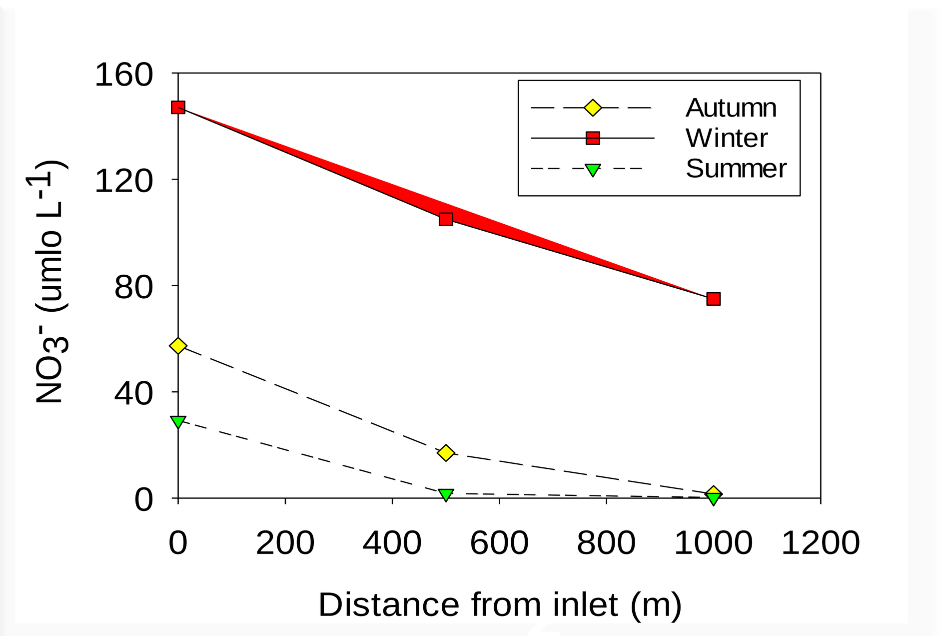



**Figure 2:** Seasonal variability of $NO_3^-$ ($\mu$mol $L^{-1}$) from the inlet to outlet

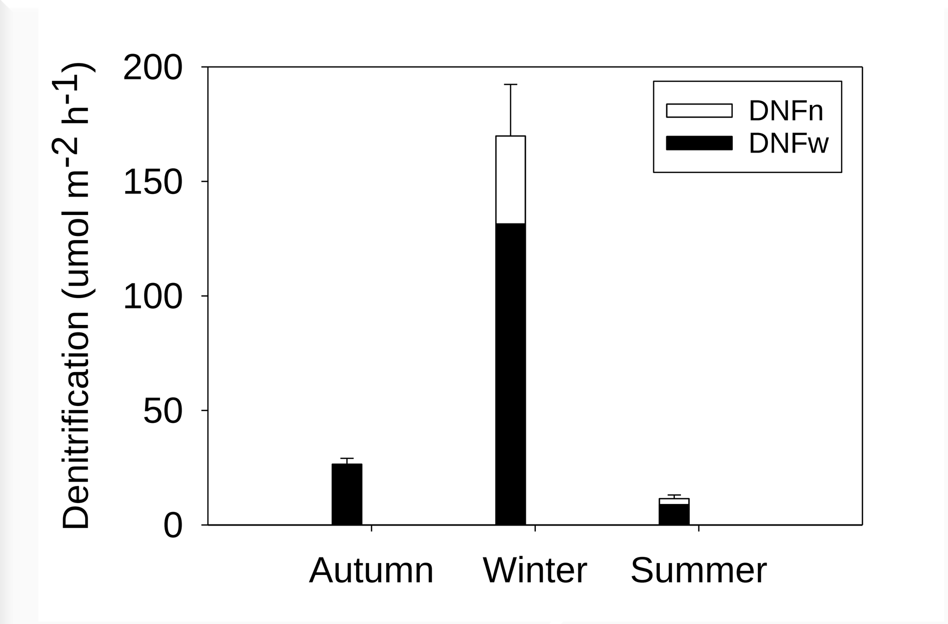



**Figure 3:** Denitrification (DNF) rates per season. Black = water $NO_3^-$ driven DNF, white = coupled nitrification-DNF.

### Denitrification: $^{15}NO_3^- \rightarrow {}^{15}N_2$

- $NO_3^-$ decreased from the inlet to the outlet = **$NO_3^-$ removed** (Fig 2)
- $NO_3^-$ removal ranged from 49% in winter up to 99% in summer (Fig 2)
- Higher $NO_3^-$ in winter led to more $NO_3^-$ removal through **denitrification** in winter (Fig 3).
- Water column $NO_3^-$ was removed by **DENITRIFICATION** in the sediments with some **nitrification-denitrification** coupling occurring.

### Dual isotopes of $\delta^{15}N$ & $\delta^{18}O$ in $NO_3^-$

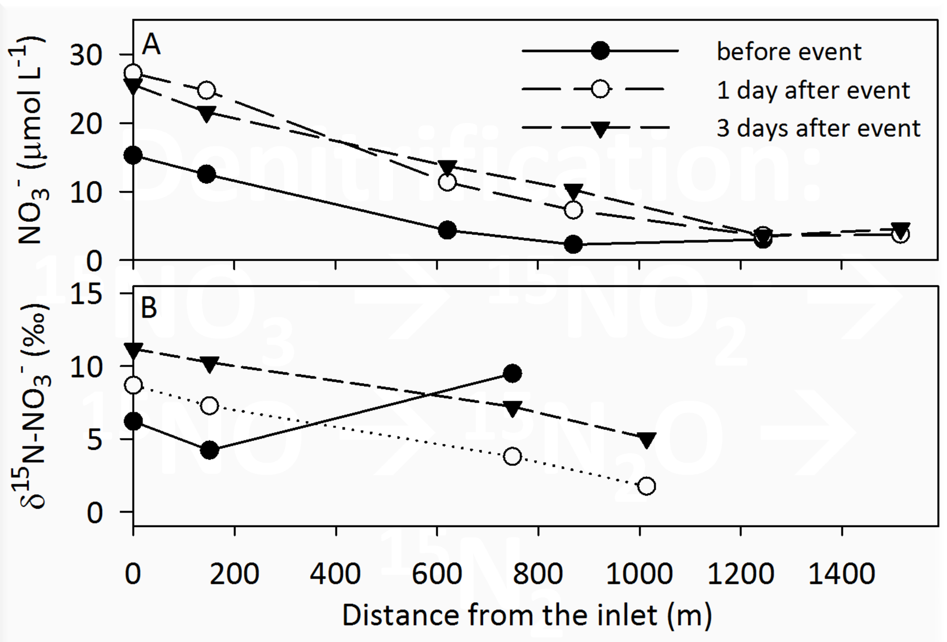



**Figure 4:** $NO_3^-$ ($\mu$mol $L^{-1}$; A) and $\delta^{15}N$-$NO_3^-$ (‰; B) from the inlet to outlet.

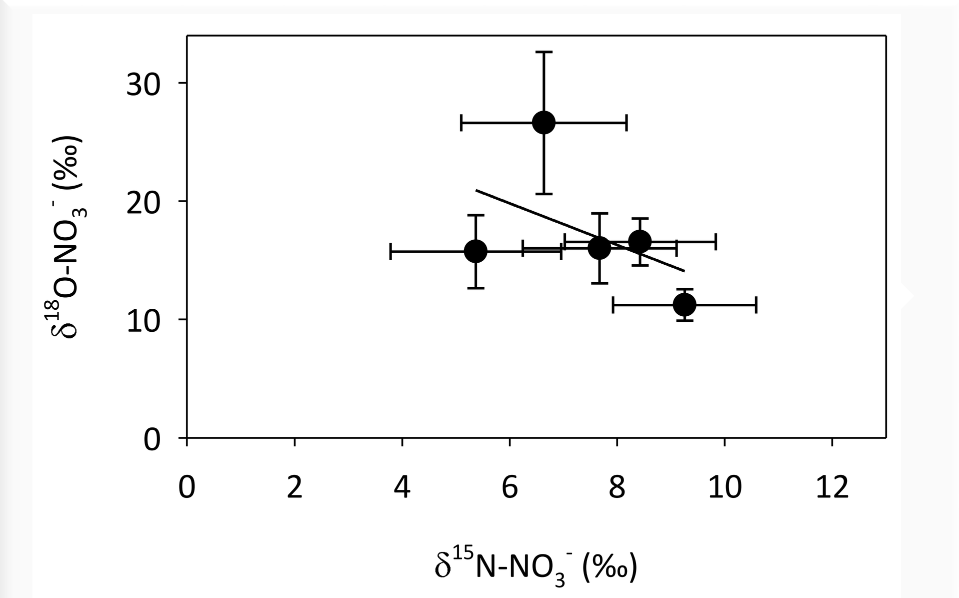



**Figure 5:** $\delta^{18}O$ (‰) vs $\delta^{15}N$ (‰) of $NO_3^-$ in the residual pool
*Error bars represent spatial variability within the wetland on each date.*

- ~80% of input $NO_3^-$ is removed in the wetland (Fig 4A).
- The $\delta^{15}N$-$NO_3^-$ **depletes** throughout the wetland (Fig 4B).
- The dual isotopic composition of $NO_3^-$ supports the **nitrification** hypothesis (Fig 5, Fig 1B).
- $NO_3^-$ is removed (Fig. 4). **DENITRIFICATION** likely removal pathway because it fractionates only a small amount (Fig 1B). **NITRIFICATION** is also an important pathway contributing to the $NO_3^-$ pool.

## CONCLUSION

**Approach 1 (IPT):**

**$NO_3^-$ was removed through DENITRIFICATION.**

**Approach 2 ($\delta^{15}N$-$NO_3^-$):**

**DENITRIFICATION and nitrification were important N pathways.**

**BOTH approaches identified $NO_3^-$ was removed through DENITRIFICATION and nitrification was also a significant pathway.**

**BOTH approaches could be used for monitoring however, the dual isotope technique would be more cost effective and easier to implement.**

**REFERENCES**

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