Dynamics and isotope effects of denitrification in Lake Lugano

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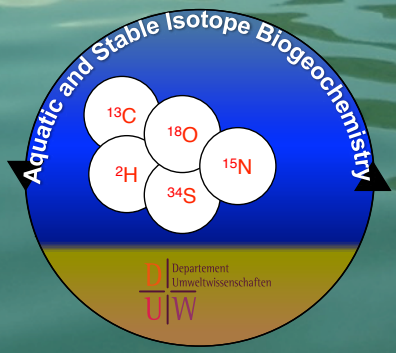


Anthropogenic Impacts on the N cycle

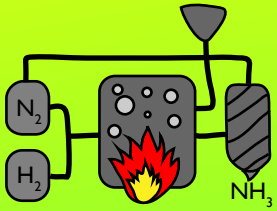
Global warming

Atmosphere

N_2O

N_2

Haber-Bosch Process



N-removal

reactive N

NH_3

Agriculture

Aquatic ecosystems

Run-off

Eutrophication

Treated waste water +
Industrial effluents

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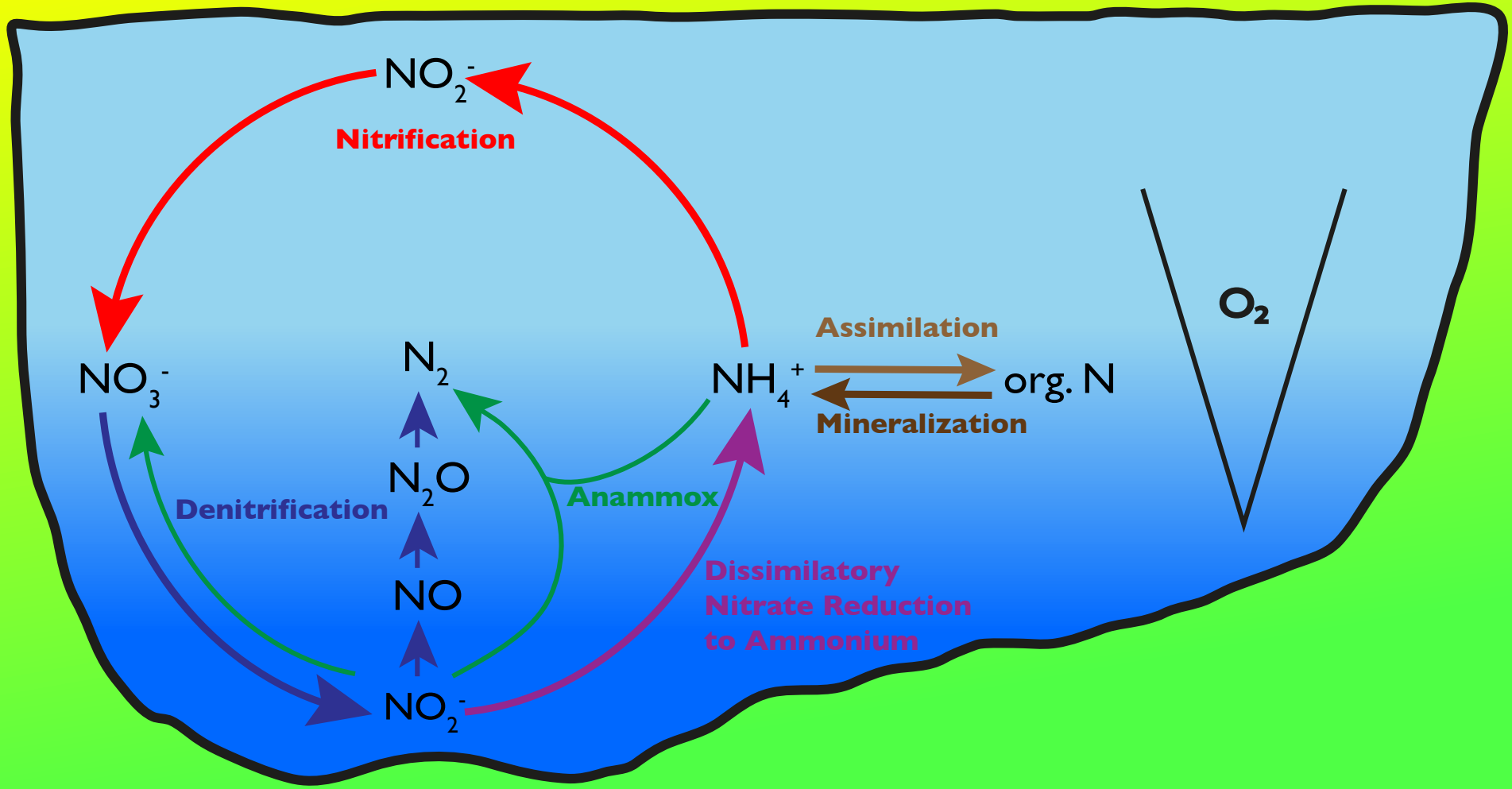
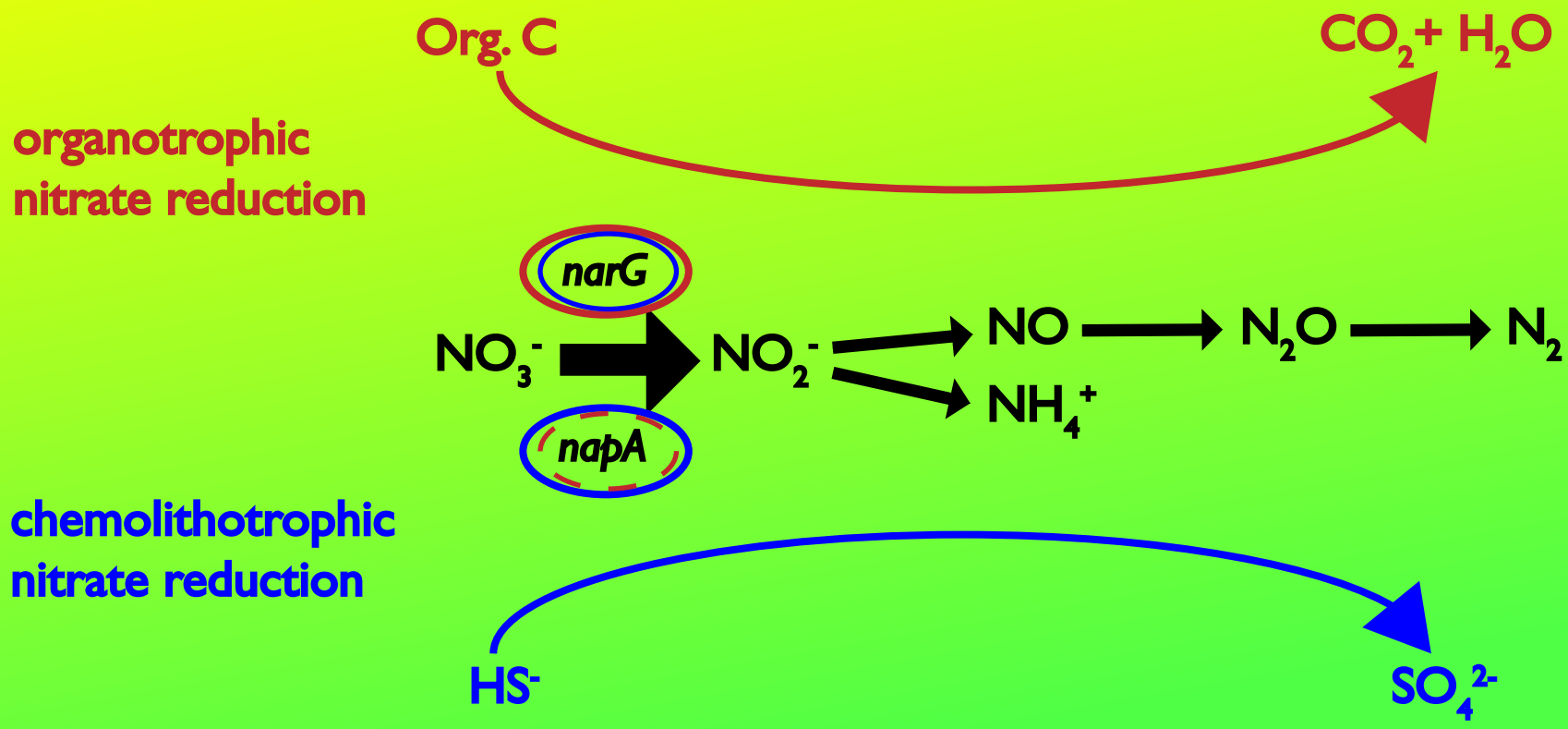
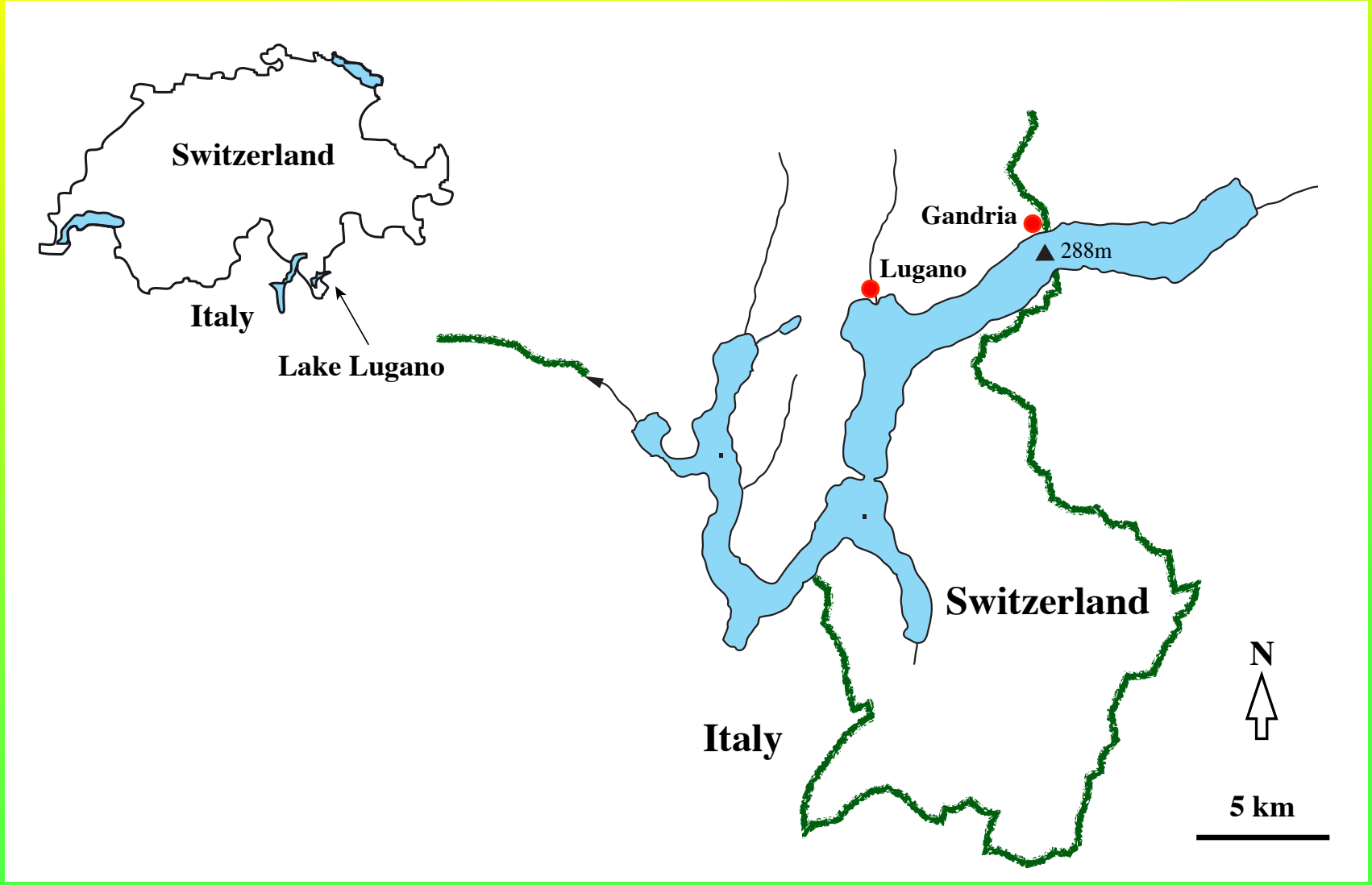
Aquatic N cycle



Types of Nitrate Reduction

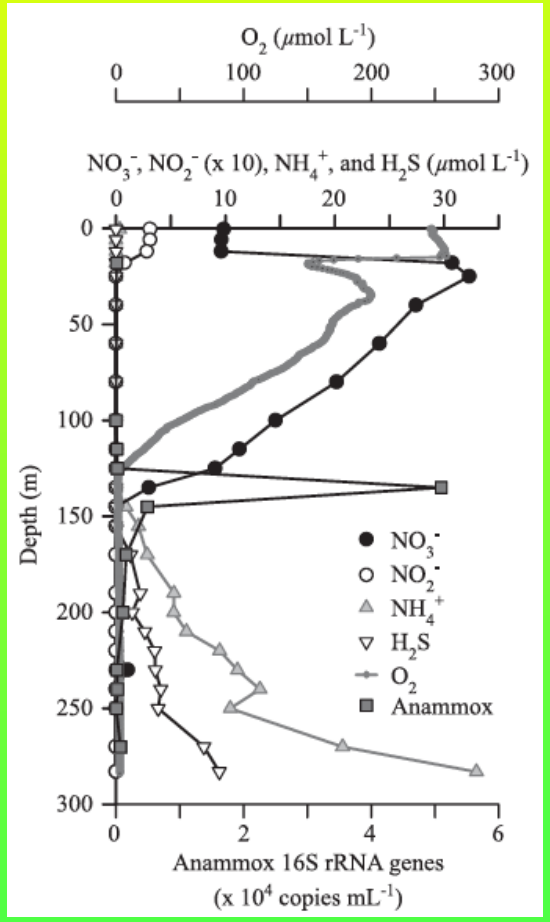


Study site: meromictic North Basin of Lake Lugano



Lake Lugano: Previous Studies

water column profile (October 2009)



known anaerobic N-cycling:

- Sulfur-oxidizing denitrifiers ✔
- Organotrophic denitrifiers ✔
- Anammox ✔

Wenk et al., Limnol. Oceanogr. (2013)

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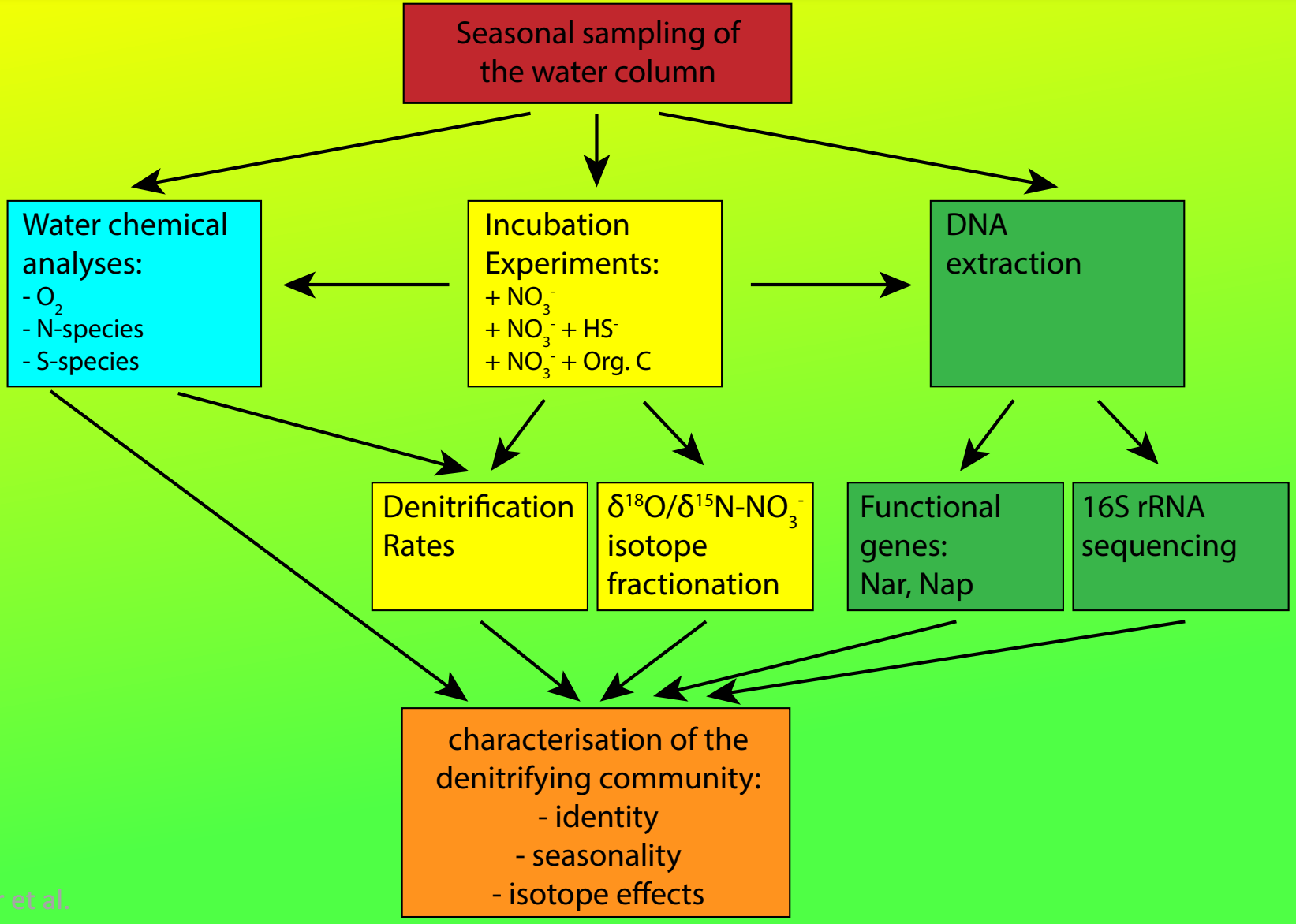
Research questions

Is the dominance of S-dependent denitrification a seasonal feature?

What isotope fractionation effects are associated with denitrification?

Interdisciplinary approach



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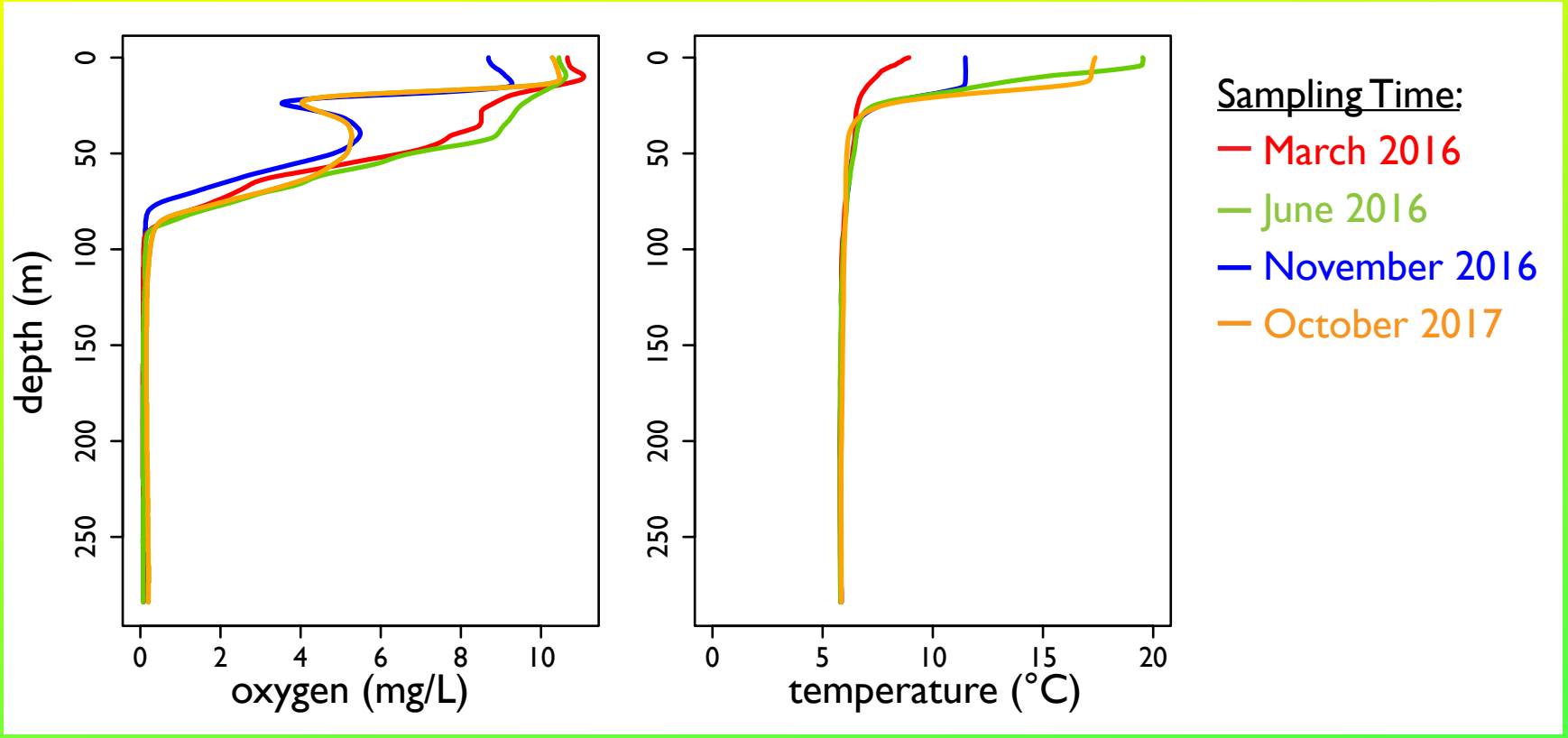
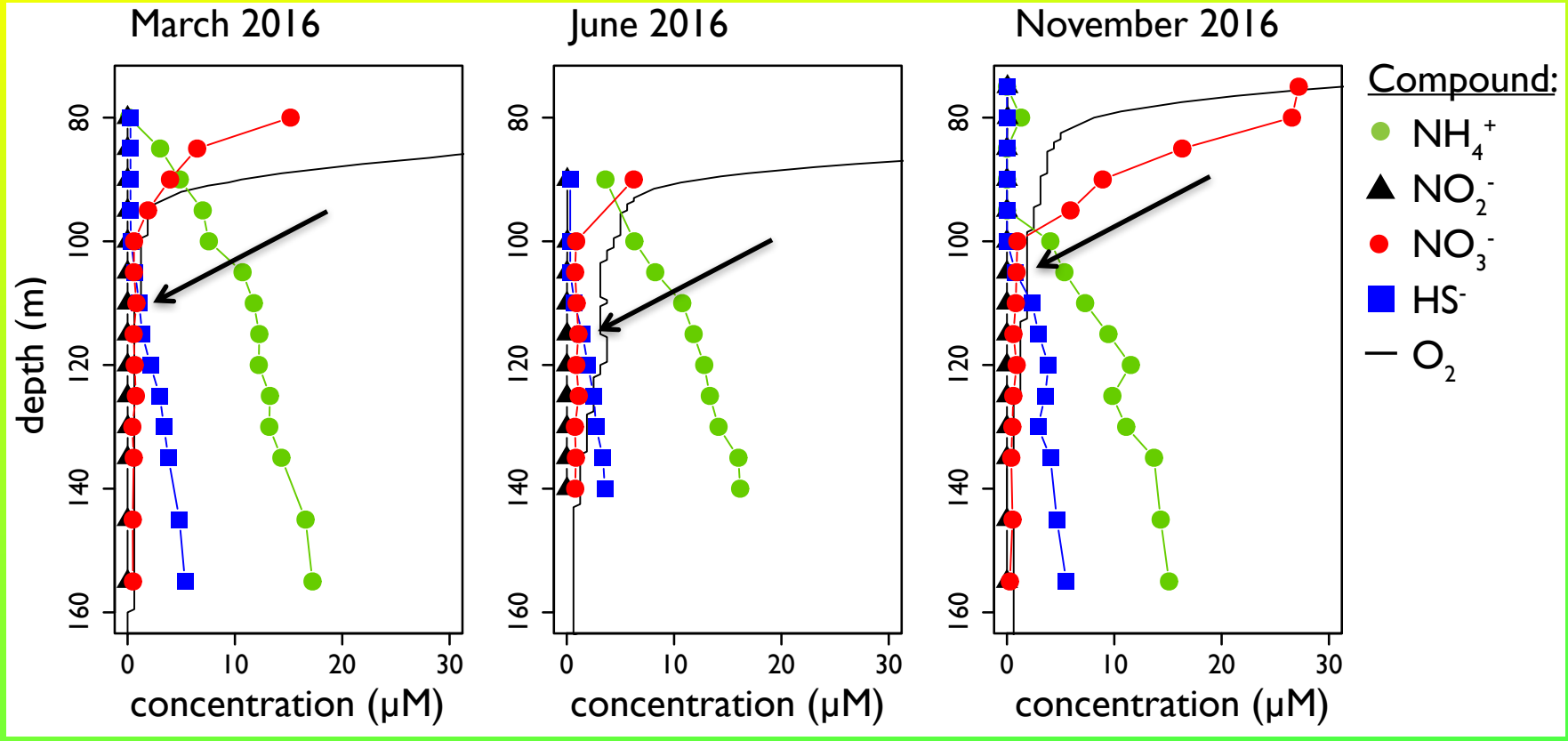
Profiles of the whole water column



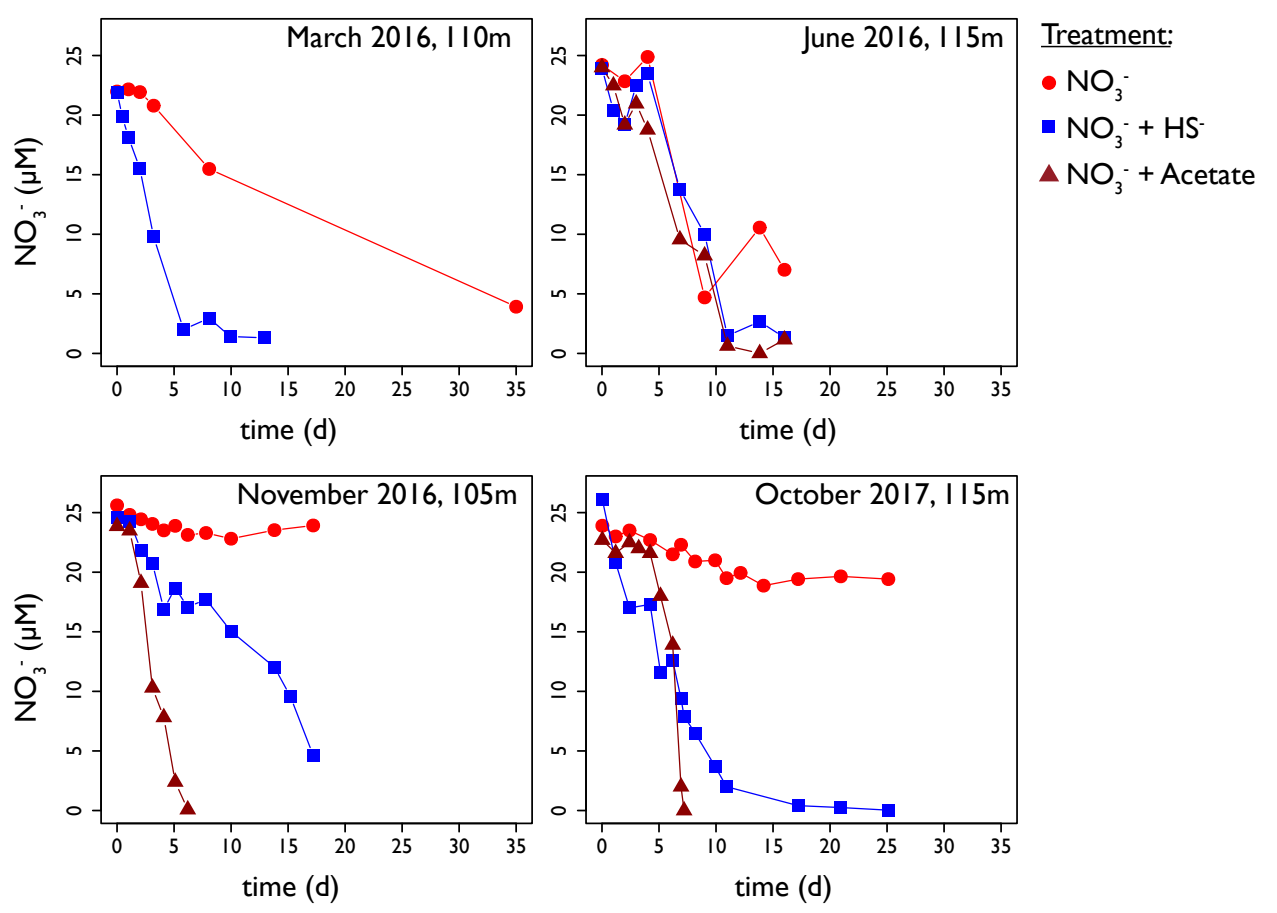
→seasonal water column dynamics

Profiles of the redox transition zone



→ overlapping NO_3^- and HS^- profiles at low concentrations

NO_3^- reduction rates



→ dynamic activity of denitrifying organisms

→ seasonal limitations of organic electron donors

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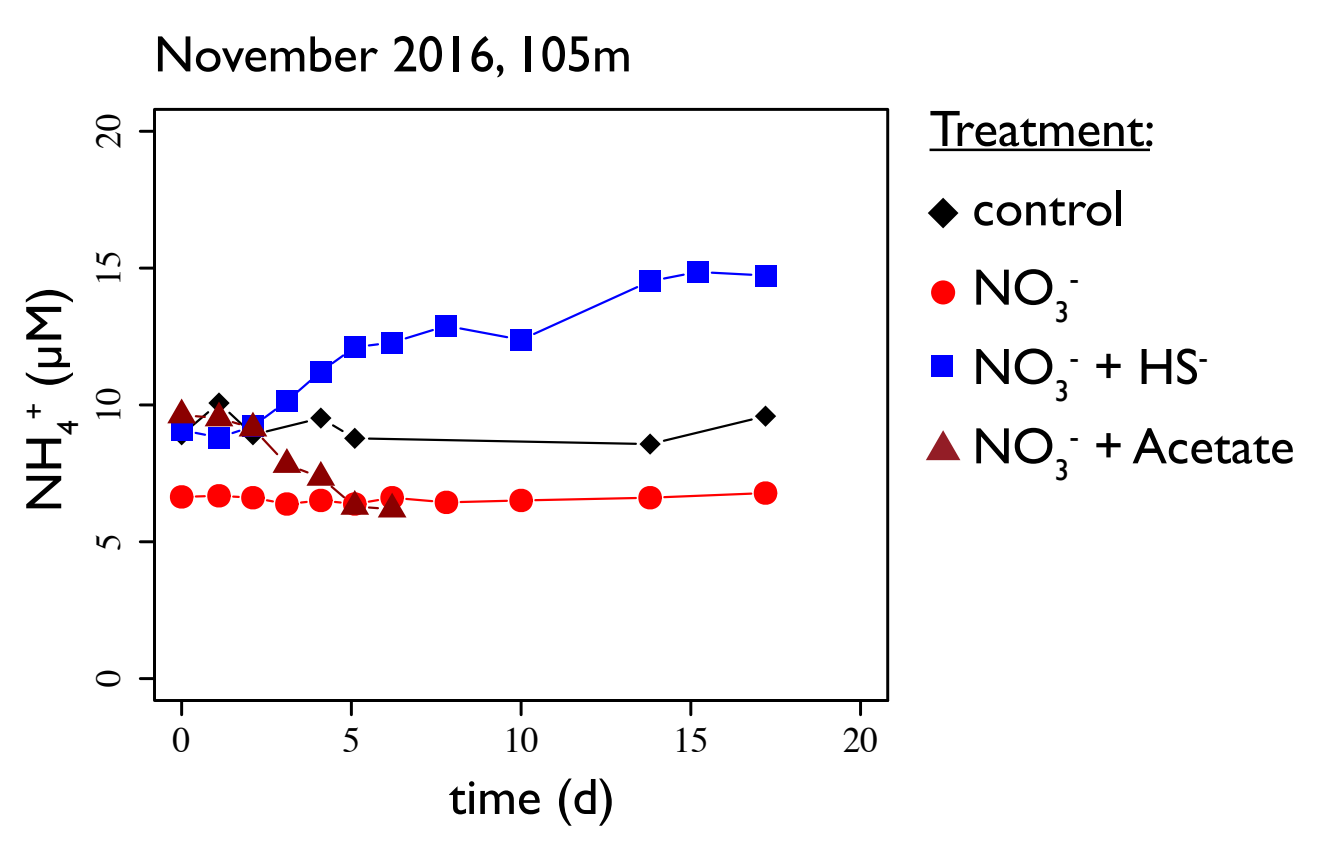
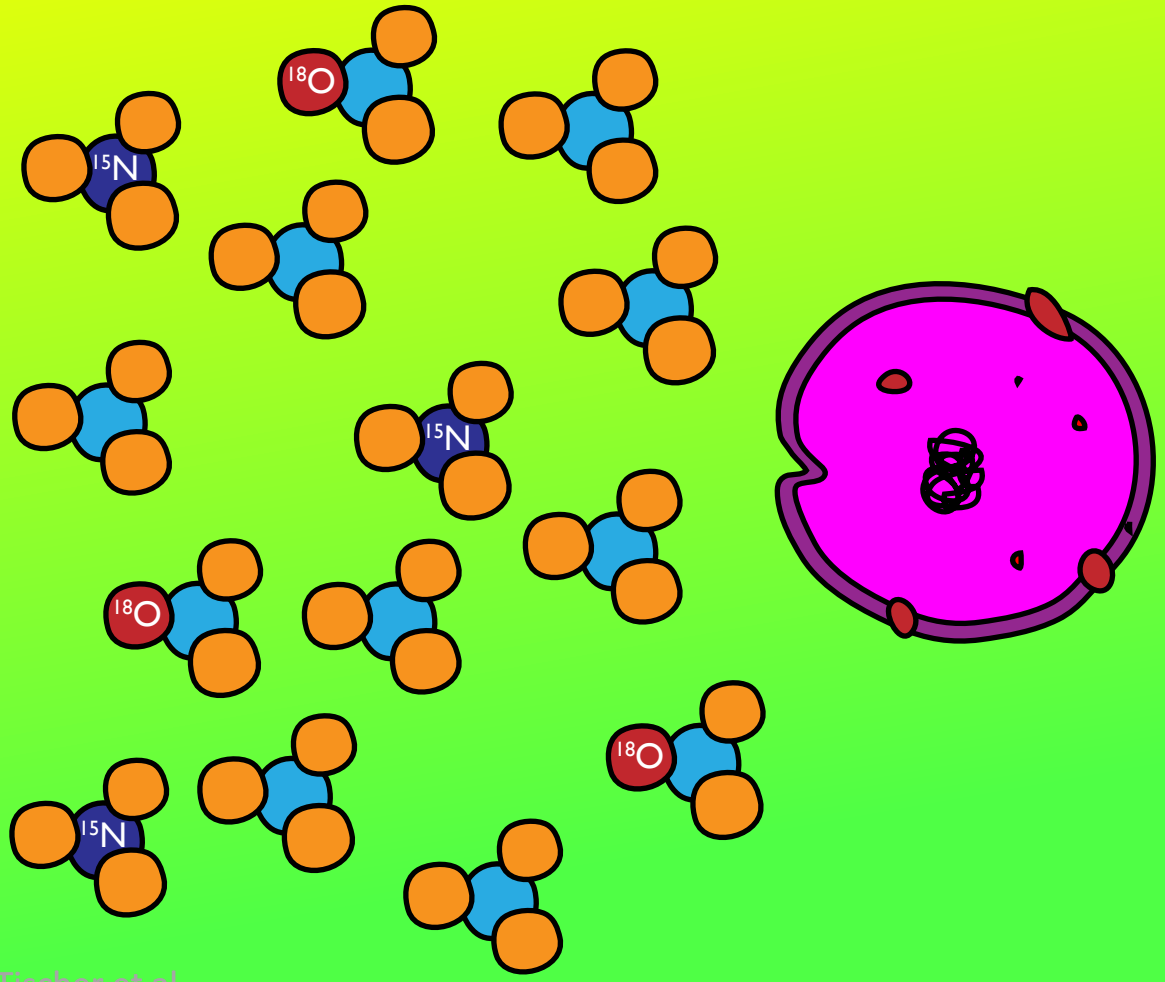
Nitrate reduction pathways



→ nitrate reduction to ammonium by S-oxidizers

Isotope effects of nitrate reduction

nitrate pool ⟶ nitrite pool



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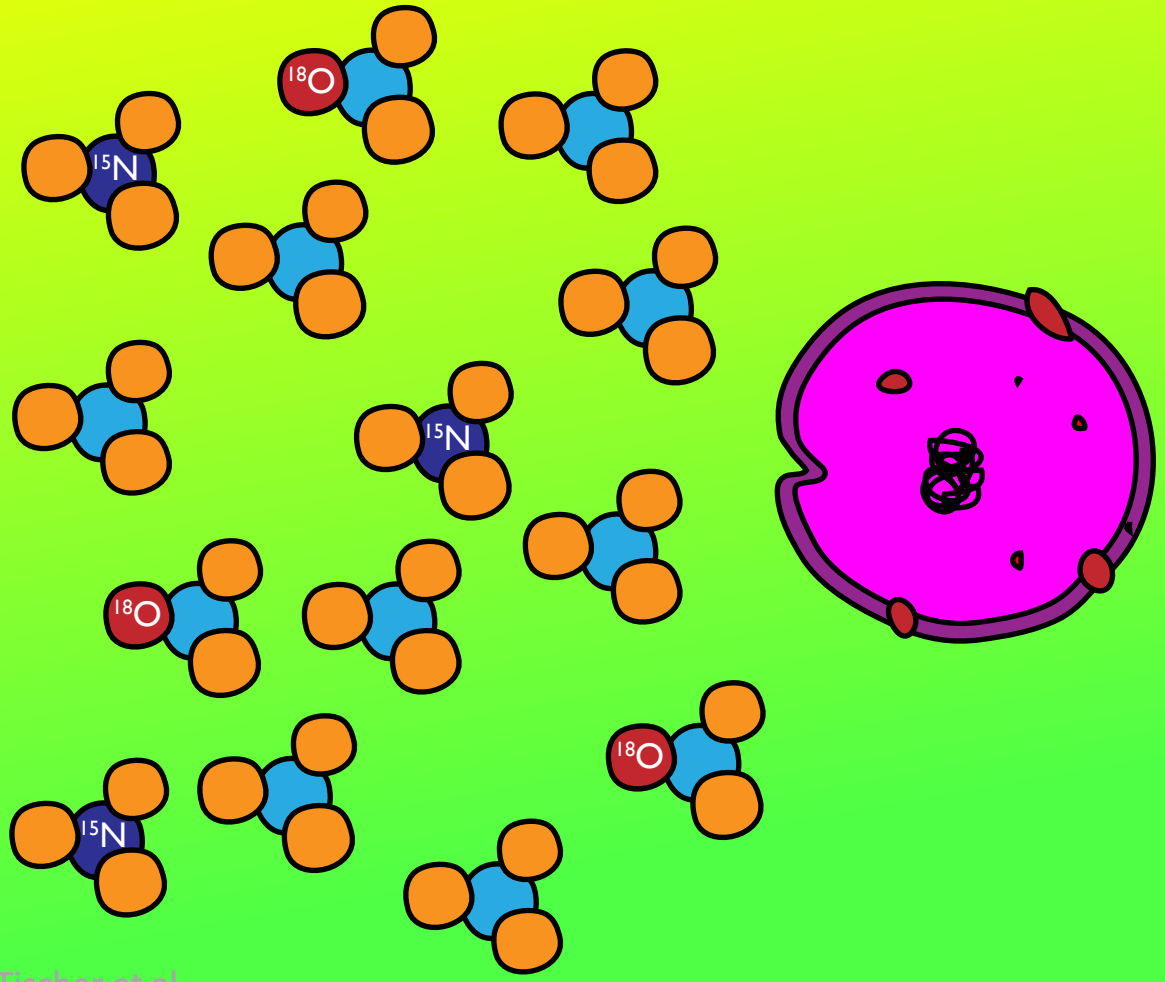
Isotope effects of nitrate reduction

nitrate pool → nitrite pool



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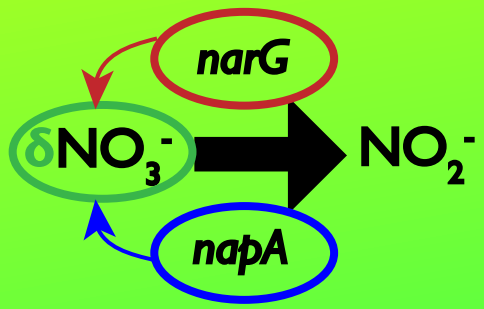
Isotope effects of nitrate reduction

- Isotope fractionation of nitrate during its reduction is variable!

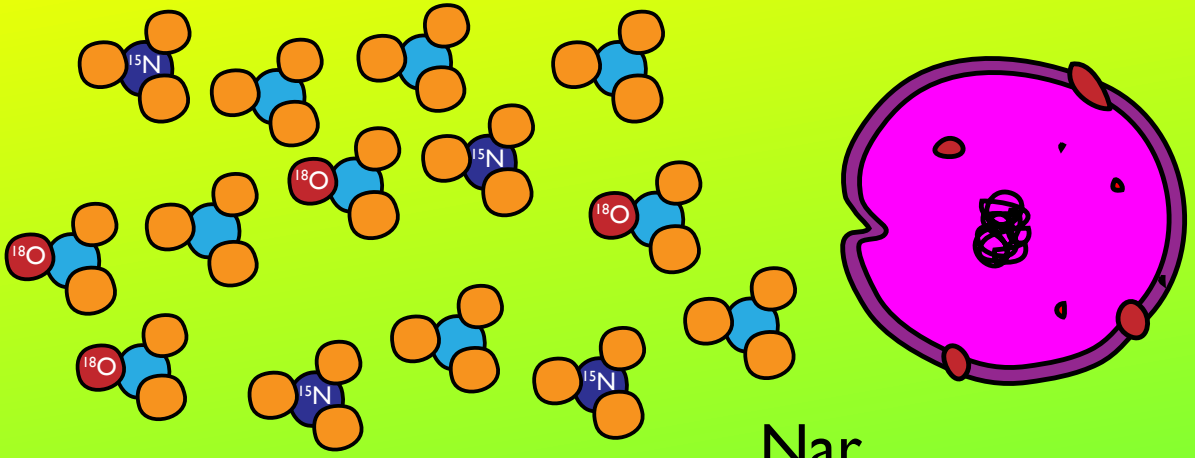
- Possible influencing factors:
  - use of different nitrate reductases Nar and Nap



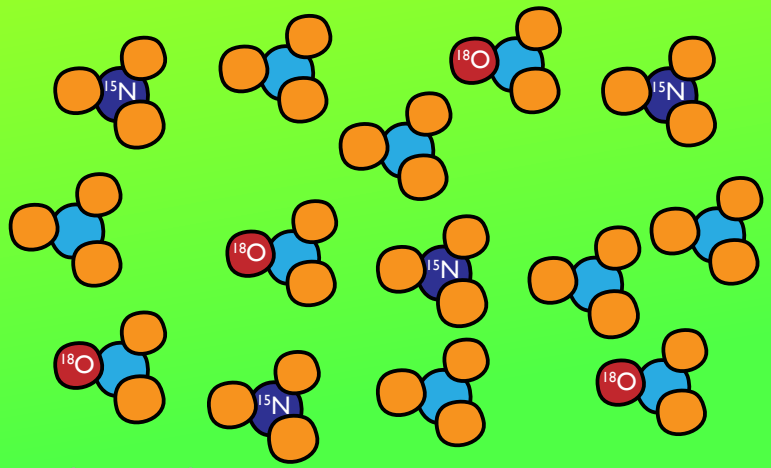
  - simultaneous nitrite oxidation via anammox or nitrification

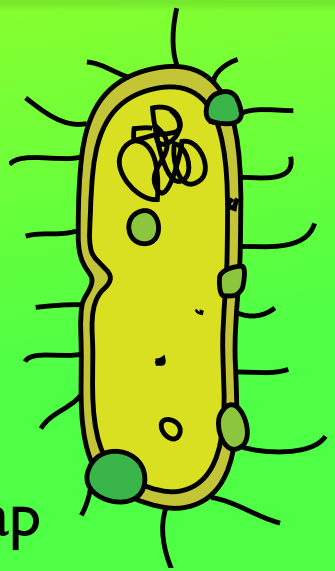
Isotope effects of nitrate reduction: Enzyme effect Nar vs. Nap

nitrate pool → nitrite pool



Nar





Nap

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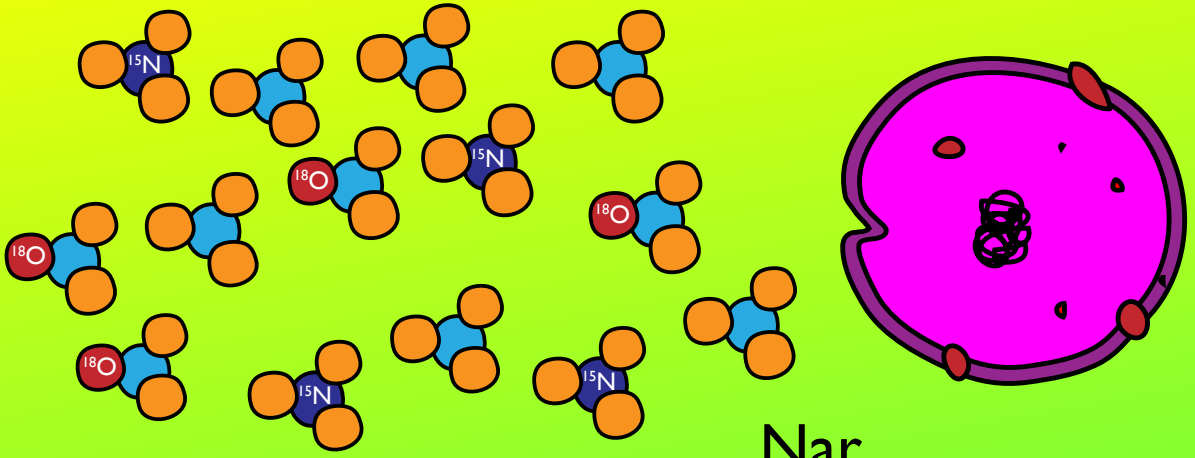
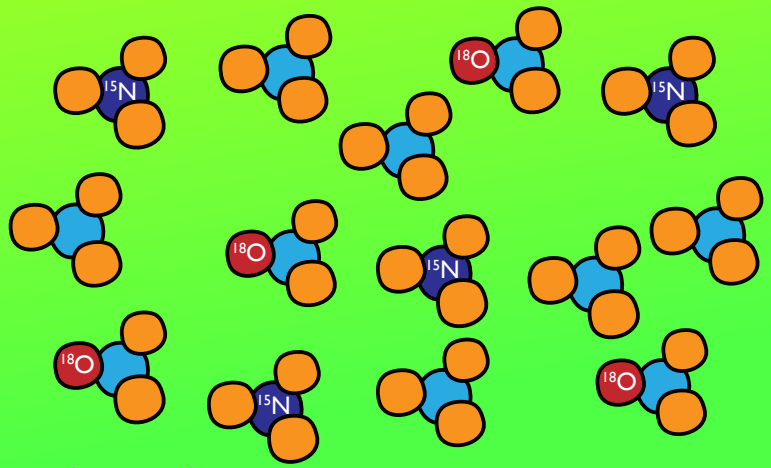
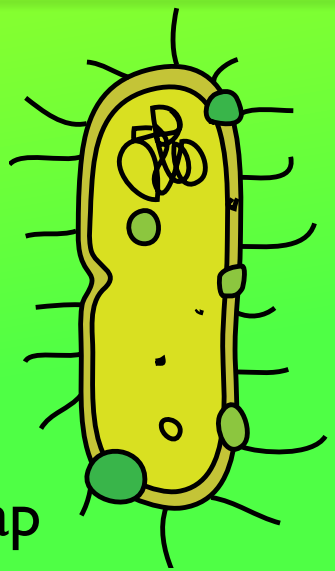
Isotope effects of nitrate reduction: Enzyme effect Nar vs. Nap

nitrate pool → nitrite pool



Nar





Nap

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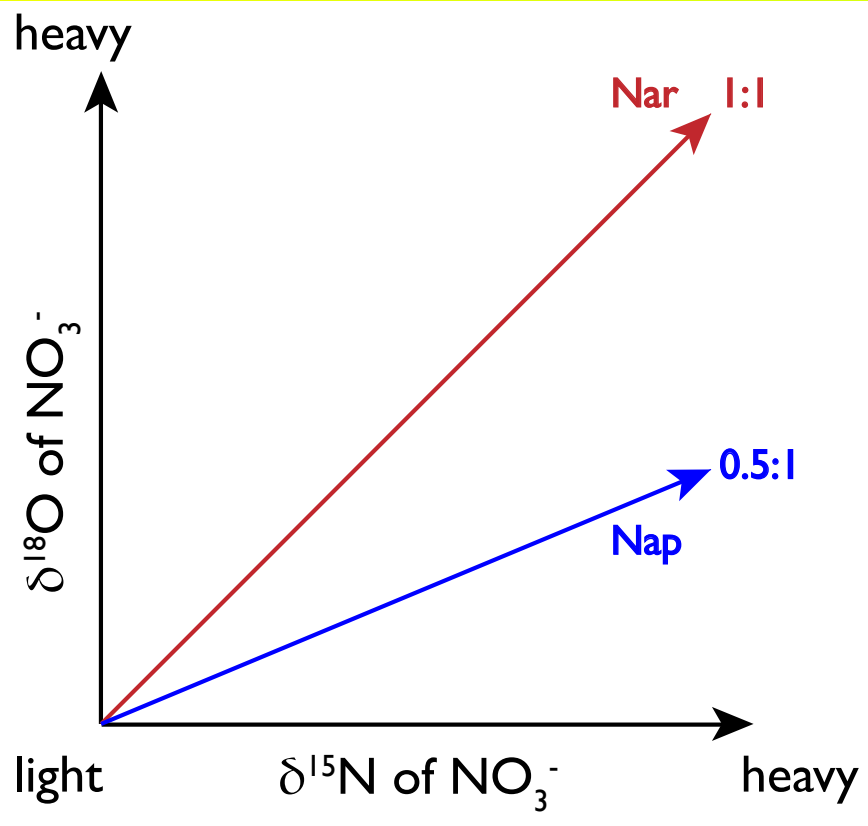
Isotope effects of nitrate reduction: Enzyme effect Nar vs. Nap

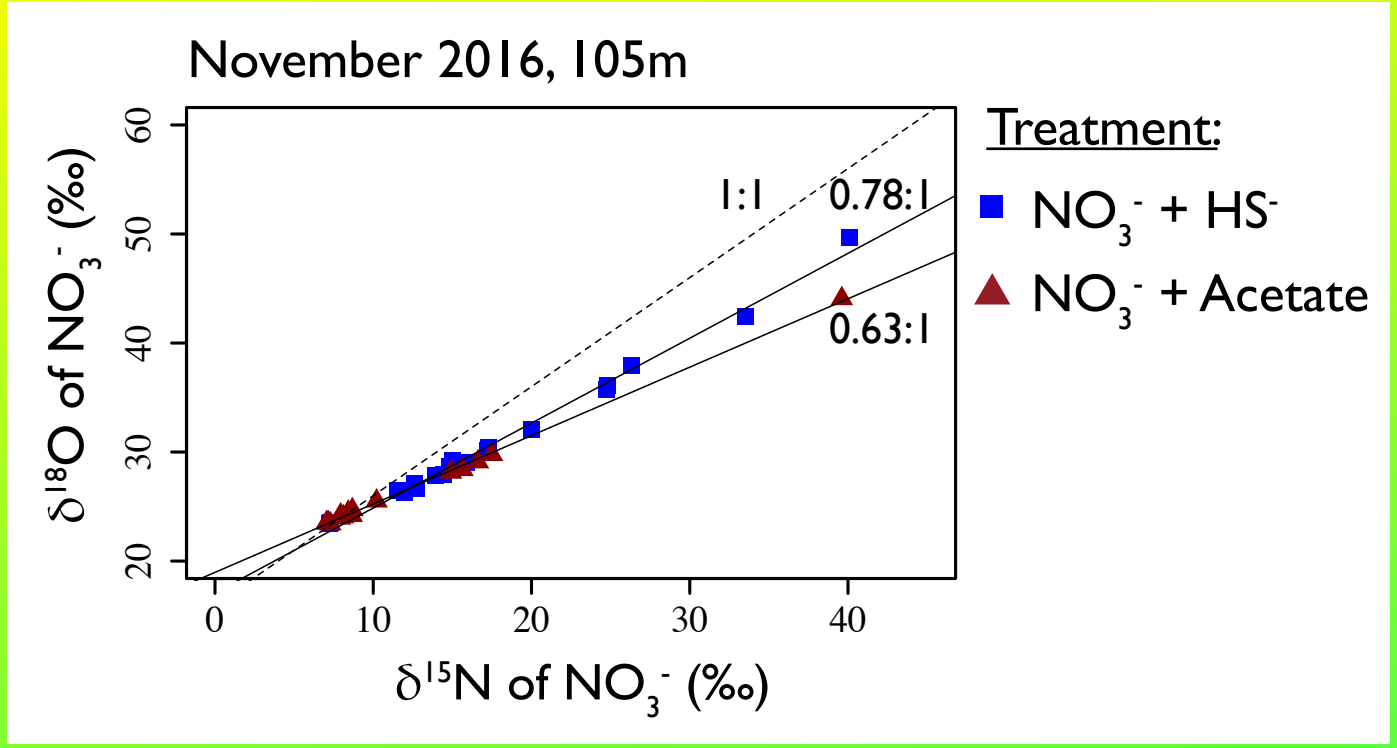


→ Isotope effect diagnostic for specific processes

$\delta^{18}O$-vs-$\delta^{15}N$ ratio during NO_3^- reduction

data from pure culture studies (e.g. Treibergs & Granger, 2017)

Isotope effects:
$\delta^{18}O$-vs-$\delta^{15}N$ ratio in incubation experiments



→ simultaneous use of Nar and Nap by S-oxidizers

→ organotrophic denitrifiers use Nap?

Variation in $\delta^{18}O$:$\delta^{15}N$ ratios

Sampling time and depth	$\delta^{18}O/\delta^{15}N$		
	NO_3^-	NO_3^- + Ac	NO_3^- + HS^-
March 16, 105 m	0.98	-	0.77
March 16, 110 m	0.78	-	0.68
June 16, 115 m	1.09	-	0.70
June 16, 120 m	1.02	0.79	-
November 16, 105 m	0.63	0.63	0.78
November 16, 155 m	0.58	0.60	0.93

→dynamic denitrifying microbial community

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Conclusions

- dynamics of denitrification
  - dynamic and complex denitrifying community
  - seasonal limitations of organic electron donors

- isotope effects
  - probably strong enzyme effect

Outlook

- proof S-dependent nitrate reduction to ammonium
- qPCRs to quantify Nar and Nap
- 16S rRNA sequencing of in-situ samples and after growth stimulation

Acknowledgements

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Thank you!