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Can Molecular Biological Tools Characterize Methane and Nitrous Oxide Flux in Engineered Environmental Systems?




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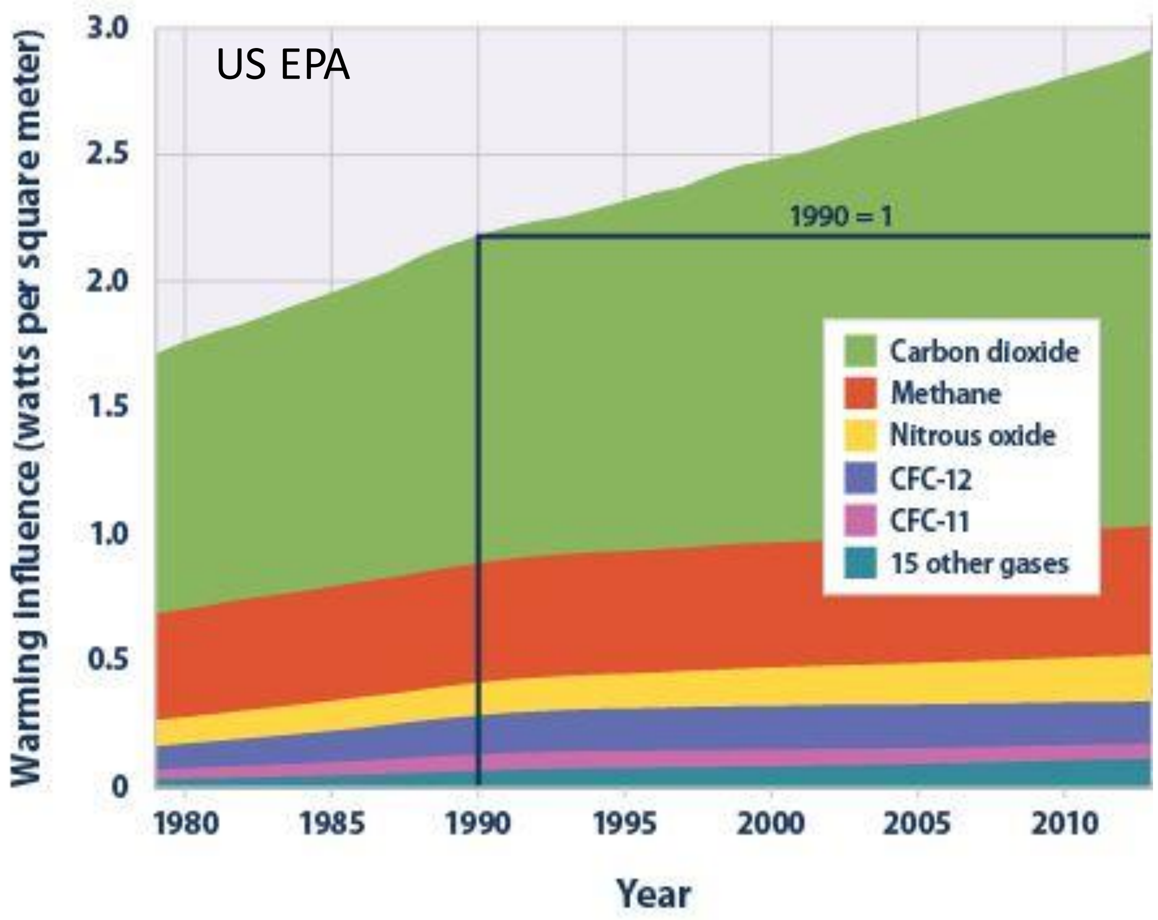
Major Greenhouse Gases



Relative Warming Impact

- Carbon dioxide 74.4%
- Methane 17.3%
- Nitrous Oxide 6.2%
- Other 2.1%

CH_4 and N_2O combined responsible for ~20% of global warming

Source “Our world in Data.org’

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Anthropogenic Sources of Methane and Nitrous Oxide

Sources of Methane (CH_4)

- Livestock (44%)
- Rice cultivation (12%)
- Landfills (5%)-point source
- Biomass burning
- Fossil Fuel Production

Sources of Nitrous Oxide (N_2O)

- Agricultural soils (80%)
- Wastewater treatment (3%)-point source
- Biomass burning

Molecular Biological Tools (MBTs)

Samples

DNA /RNA Extraction

Next Gen Sequencing

qPCR/ RT-qPCR

Quantification of gene copies/ mRNA transcripts

Microbial Community Profiles

Routine

- Bioremediation *Dehalococcoides*
- SARS CoV-2 in wastewater

Microbial GHG Targets

- Nitrification/Denitrification
- Methanogenesis/Methanotrophy

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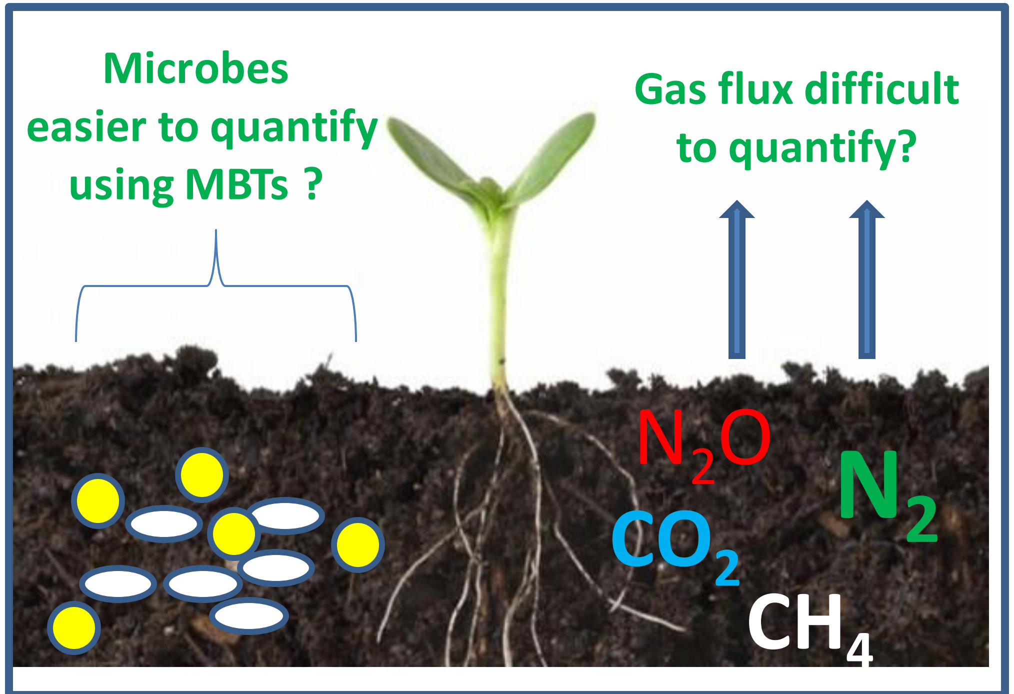
Quantifying Microbial GHG Processes

- **Determining gas flux from soil and water is challenging**

- Low concentrations
- Flux events are highly variable
- End products (N_2/CO_2) can be ubiquitous
- Isotopic methods (e.g., CSIA $^{15}N/^{14}N$) helpful

- **MBTs benefits for quantifying GHG processes**

- Microbes are non-volatile, don't dissipate like gases
- MBTs can detect potential functions, even if not active
- PCR based MBTs are very sensitive
- Tracking populations over time is informative



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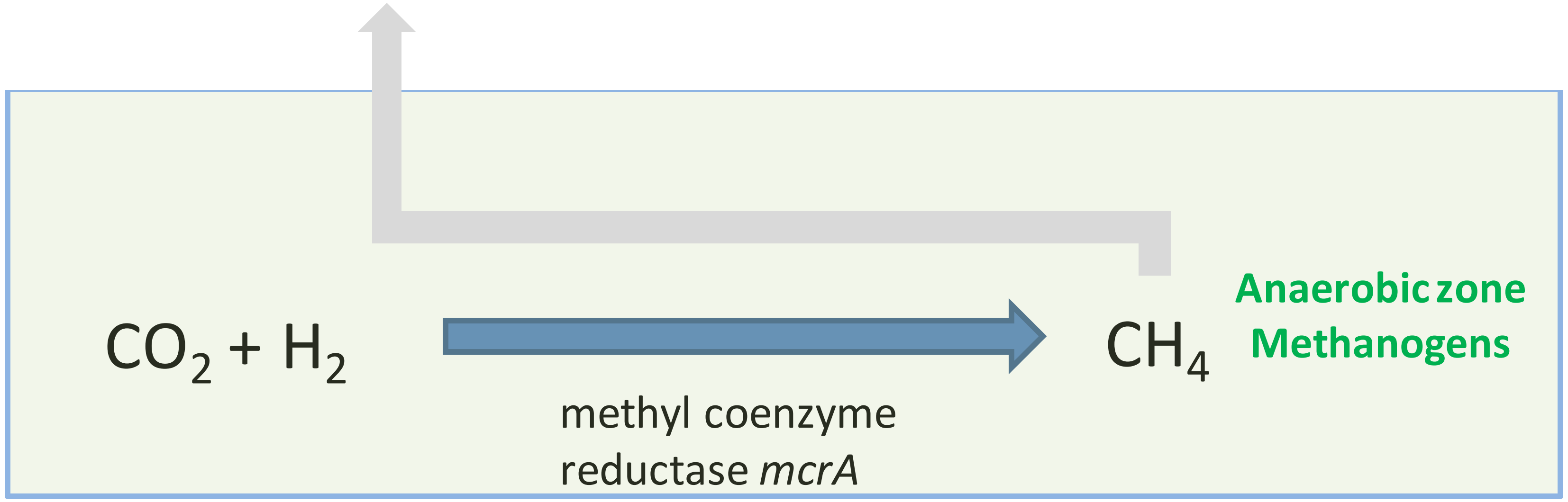
CH_4 Flux Methanogenesis vs. Methanotrophy



- Methanotrophs can attenuate CH_4 impact
- MMO gene can quantify CH_4 degraders e.g., in landfill cover soil
- Maximizing methanotrophy
  - Nutrient addition
  - Moisture control
  - Earthworms
  - Plantings

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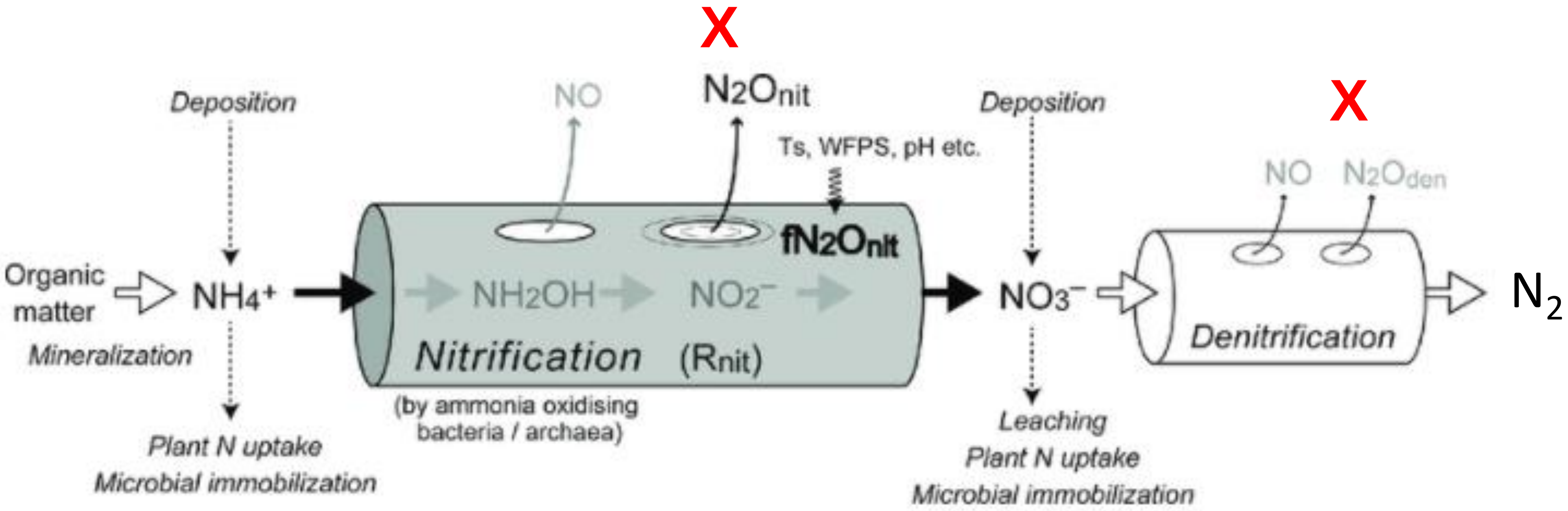
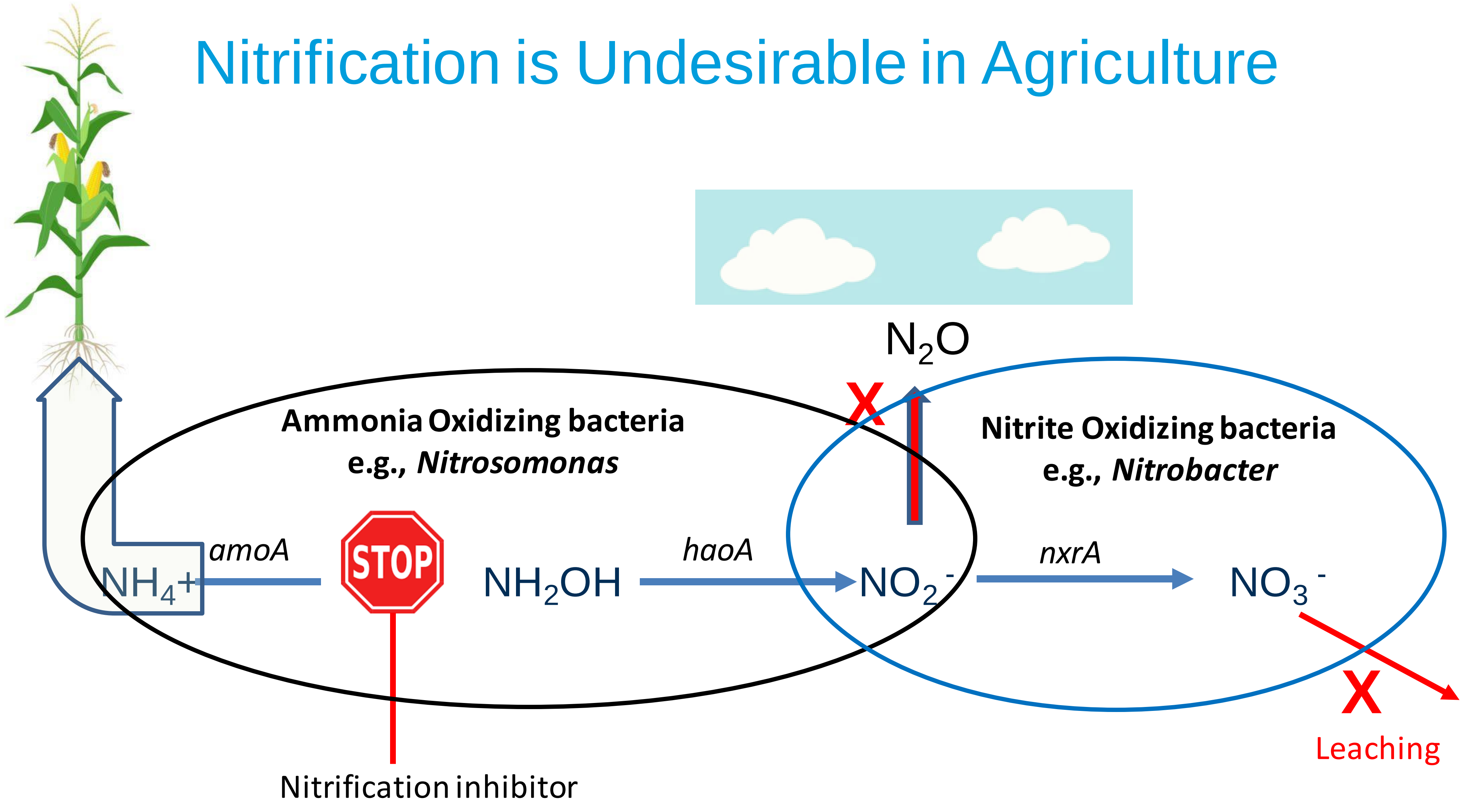
“Holes in a Pipe Concept” Nitrogen Metabolism and N_2O



Inatomi et al., 2019

Nitrification is Undesirable in Agriculture



N loss = ⬆ fertilizer production = ⬆ CO_2 emissions

MBTs could help

- Management of nitrification inhibitors (e.g., nitripyrin) that inhibit *amoA* activity
- Balance of AOB and NOB to minimize N_2O emissions

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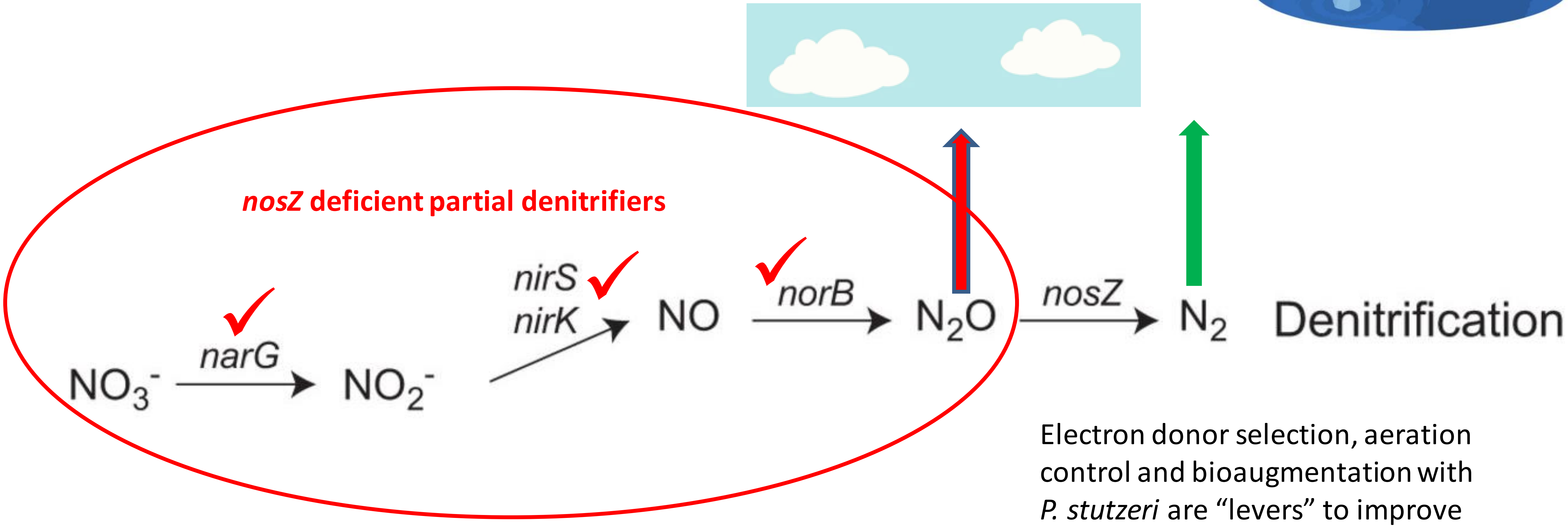
Wastewater Treatment



Electron donor selection, aeration control and bioaugmentation with *P. stutzeri* are "levers" to improve complete denitrification

- In WWT denitrification want full gene set, particularly *nosZ to* prevent N_2O emissions.

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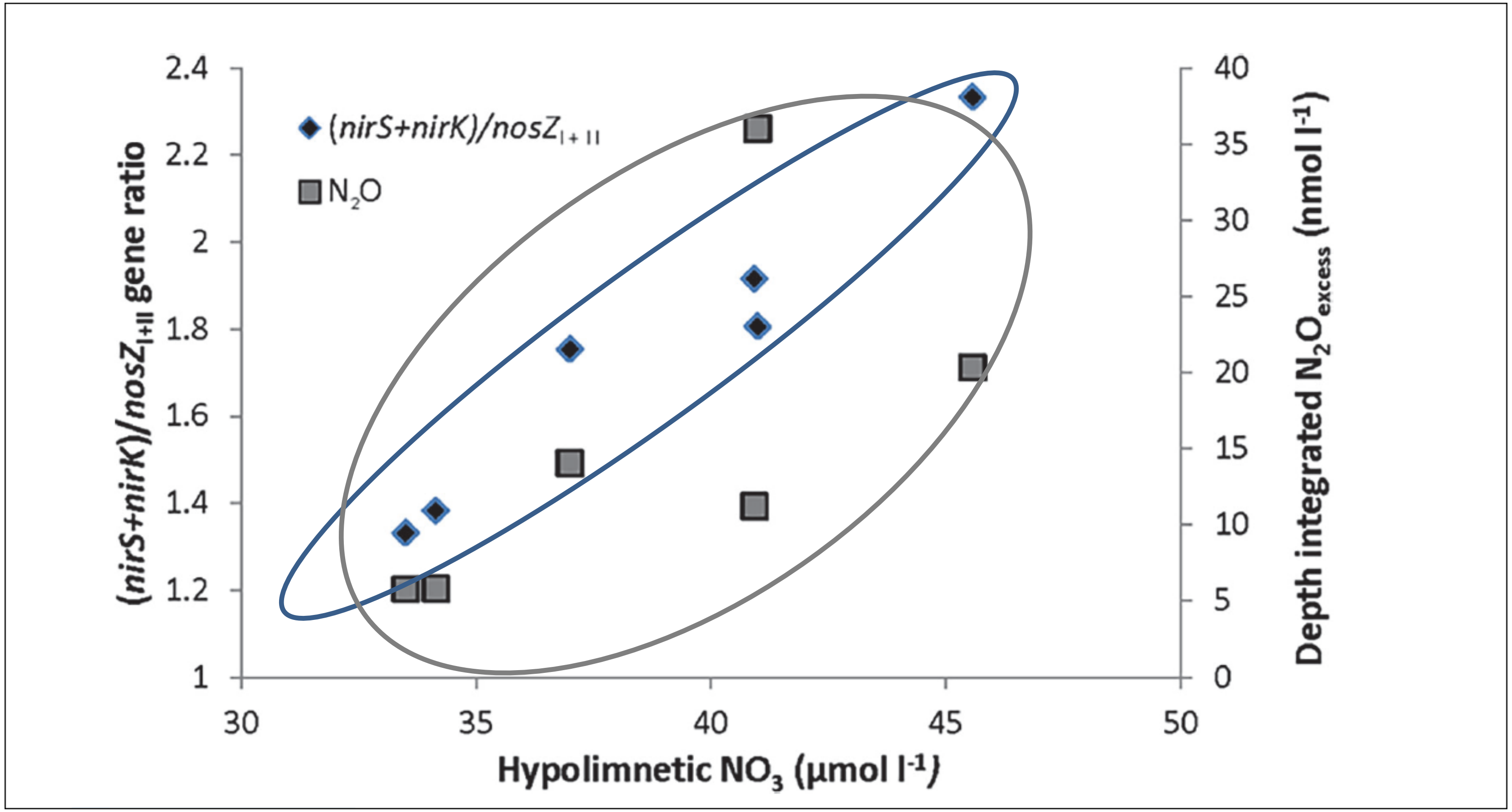
Ratio of denitrification genes corelates with N_2O production



Study in Finnish Lakes (Saarenheimo et al., 2015) indicated that a high (*nirS+nirK*)/*nosZ* gene ratio led to increased N_2O production

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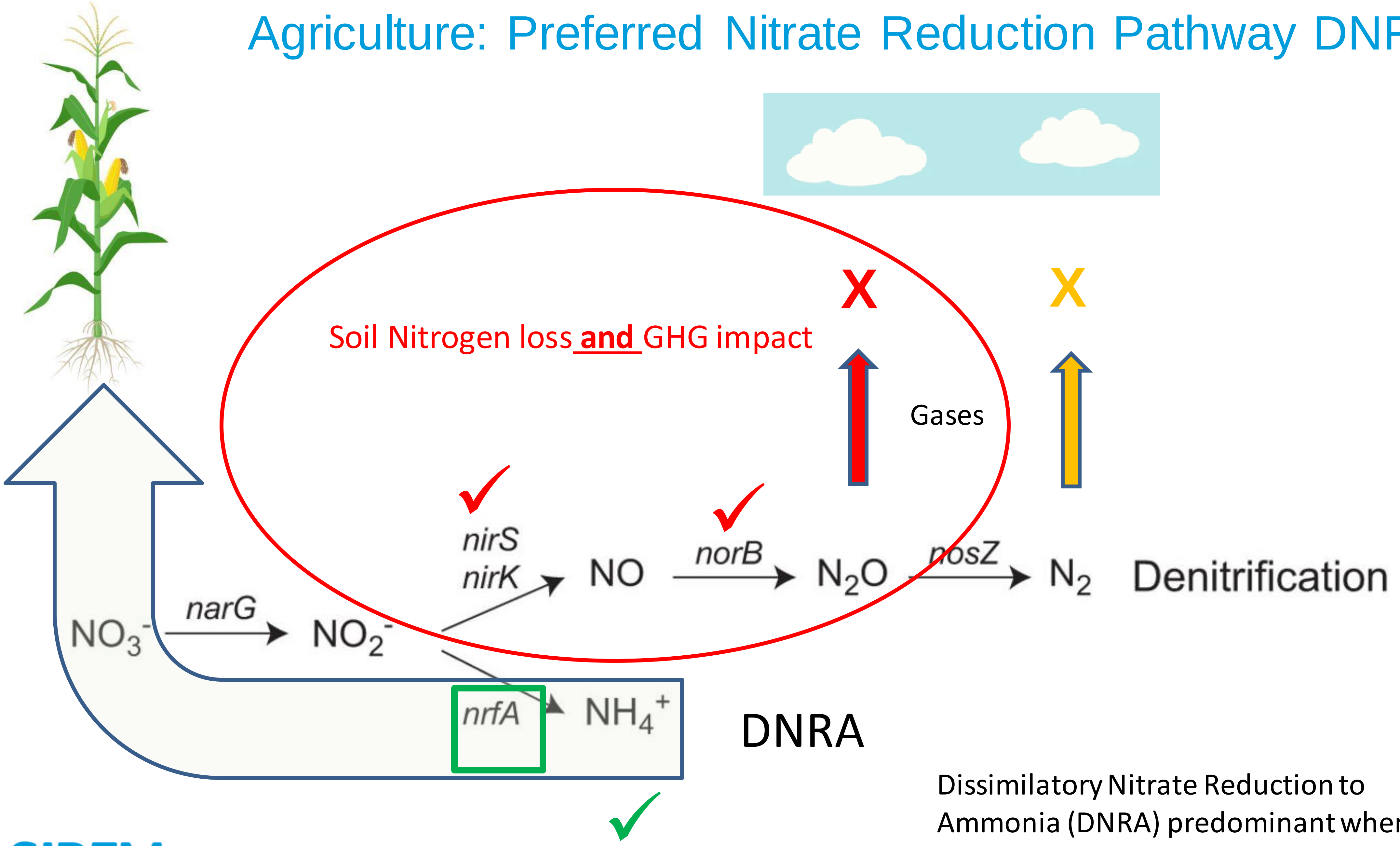
Agriculture: Preferred Nitrate Reduction Pathway DNRA



Dissimilatory Nitrate Reduction to Ammonia (DNRA) predominant when soil organic matter higher

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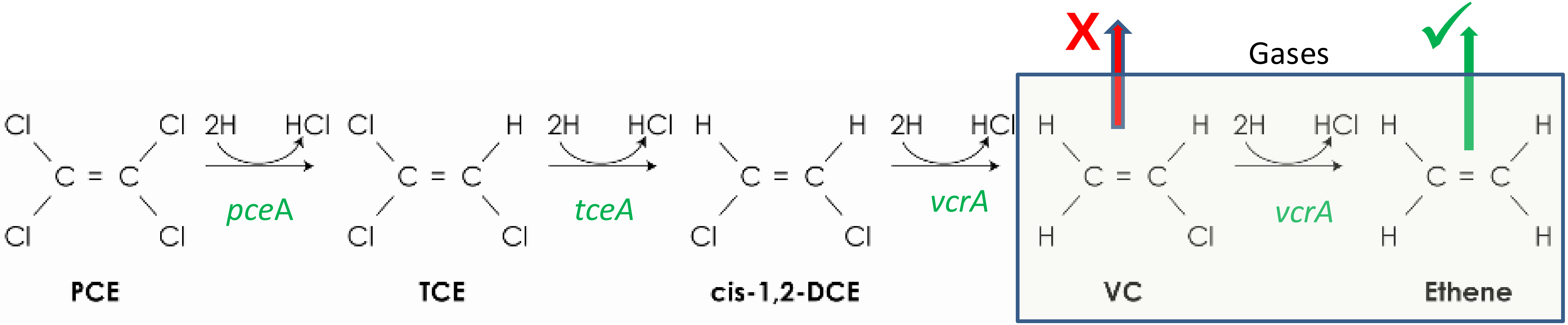
Are use of MBTs in Bioremediation a Model for GHGs ?

- *Dehalococcoides* (*Dhc*) key microbe in chlorinated solvent biodegradation in groundwater
- MBTs for *Dhc* widely used to optimize performance & avoid negative outcomes in bioremediation

- MBTS could play similar role in managing N_2O and CH_4 to reduce GHG impacts



Acknowledgement/References

Acknowledgement

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References

- Ali , Muhammad, et al. 2016 Source identification of nitrous oxide emission pathways from a single-stage nitritation-anammox granular reactor *Water Research* Volume 102, 1 October 2016, Pages 147-157

- Kumaresan, Deepak, Guy C. J. Abell,Levente Bodrossy,Nancy Stralis-Pavese,J. Colin Murrell. 2009 Spatial and temporal diversity of methanotrophs in a landfill cover soil are differentially related to soil abiotic factors *Environmental Microbiology Reports* 1(5) 398-407

- Inatomi, Motoko & Hajima, Tomohiro & Ito, Akihiko. (2019). Fraction of nitrous oxide production in nitrification and its effect on total soil emission: A meta-analysis and global-scale sensitivity analysis using a process-based model. PLOS ONE. 14. e0219159. 10.1371/journal.pone.0219159.

References Continued…

Lu Huijie, Kartik Chandran, David Stensel. Microbial ecology of denitrification in biological wastewater treatment Water Research 64 237-254

Saarenheimo J, Rissanen AJ, Arvola L, Nykänen H, Lehmann MF, et al. (2015) Genetic and Environmental Controls on Nitrous Oxide Accumulation in Lakes. PLOS ONE 10(3): e0121201.

V. Vasilaki , T.M. Massara , P. Stanchev , F. Fatone , E. Katsou. 2019 A decade of nitrous oxide (N2O) monitoring in full-scale wastewater treatment processes: A critical review *Water Research* 161 (2019) 392-412

Thank you for your attention !

Further Information

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