Quantifying isotopic signatures of N₂O using quantum cascade laser absorption spectroscopy

Kristýna Kantnerová[a]*, Béla Tuzson[a], Lukas Emmenegger[a], Stefano M. Bernasconi[b] and Joachim Mohn[a]*

_____

*Correspondence: Dr. J. Mohn

E-mail: joachim.mohn@empa.ch

[a]Laboratory for Air Pollution / Environmental Technology

Empa – Swiss Federal Laboratories for Materials Science and Technology

Überlandstrasse 129, CH-8600 Dübendorf, Switzerland

[b]Geological Institute, Department of Earth Sciences, ETH Zürich

Sonneggstrasse 5, CH-8092 Zürich, Switzerland

Abstract: Nitrous oxide, N_2O, is the environmentally most relevant constituent of the biogeochemical nitrogen cycle. Human activities, e.g. the agricultural use of mineral fertilizers, accelerate nitrogen transformations, leading to higher emissions of this strong greenhouse gas. Investigating the stable isotopic composition of N_2O provides a better understanding of formation mechanisms to disentagle its highly variable source and sink processes. Mid-infrared (mid-IR) laser spectroscopy is a highly attractive technique to analyze N_2O isotopocules based on their specific ro-vibrational absorption characteristics. Specifically, quantum cascade laser absorption spectroscopy (QCLAS) in combination with preconcentration has shown to be powerful for simultaneous and high-precision analysis of the main N_2O isotopocules. Recently, in the scope of my PhD project, we have been advancing this analytical technique for the analysis of the very rare doubly substituted N_2O isotopic species $^{15}N^{14}N^{18}O$, $^{14}N^{15}N^{18}O$, and $^{15}N^{15}N^{16}O$, also known as clumped isotopes. Currently, we are investigating the potential of these novel isotopic tracers to track the complex N_2O production and consumption pathways. Improved understanding of the nitrogen cycle will be a major step towards N_2O emission reduction.

2

Kristýna Kantnerová was born in the Czech Republic in 1991. In 2016, she obtained her Master's degree in Analytical Chemistry and Quality Engineering from University of Chemistry and Technology, Prague with honours. She conducted her Master's thesis in the Molecular Electrochemistry group at the J. Heyrovsky Institute of Physical Chemistry. Since 2016, she is in the group of Dr. Joachim Mohn at Empa Dübendorf and in the group of Prof. Stefano Bernasconi at ETH Zürich. Her PhD work is focused on developing a quantum cascade laser based analytical technique for the analysis of the doubly substituted ("clumped") isotopic species of nitrous oxide and exploring their usage.

1. Introduction

Mitigating anthropogenic emmisions of greenhouse gases (GHG) into the Earth's atmosphere is one of the biggest challenges that the scientific community and the society in general is currently facing. Emissions of nitrous oxide, N_2O, contribute to global warming by 6 %[1] due to its strong global warming potential, which is 300 times higher than that of CO_2.[2] In addition, N_2O is the most important anthropogenically emitted substance responsible for the depletion of stratospheric ozone.[3] Its atmospheric abundance has increased from around 270 ppb in the preindustrial era (before the year 1750)[4] to 329.9 ± 0.1 ppb in year 2017,[5] and the increase steadily continues. Developing mitigation options for this trace gas requires a profound understanding of its biogeochemical cycle and control parameters.

Although the global N_2O budget and the main source areas – soils under natural vegetation, agricultural land use, and oceans – are well known, fluxes exhibit very high spatial and temporal variability, which makes predictions and mitigation very challenging. In addition, the specific biogeochemical production pathways of N_2O display a high complexity, with many potential microbial and abiotic transformations. A detailed description of processes known to lead to N_2O formation or consumption and of challenges in their characterization can be found in a number of review articles. [6,7] Currently, the most important pathways in soils are considered to be: bacterial heterotrophic denitrification and nitrifier-denitrification, nitrification, and chemodenitrification. However, as some of these processes start from the same substrates (e.g. nitrates or nitrites), and even exhibit the same reaction intermediates, quantification of their individual contributions is still not possible.

In this context, natural abundance stable isotope signatures can be used as fingerprints to trace transformations of substances present in the environment. Likewise, the stable isotopic composition of GHG, such as N_2O, provides

important information about source and sink processes.[8,9] The natural variations in stable isotope abundances are due to the fact that in many biotic and abiotic processes, the reaction rates differ between isotopic species, e.g. reduction of $^{15}NO_2^-$ versus $^{14}NO_2^-$, leading to a so called isotopic fractionation.[10] As the isotopic fractionation is distinct for certain reaction pathways, this information can be used to differentiate source processes from each other. The N_2O molecule (N=N=O) involves in total 12 distinct "isotopically substituted molecules" or "isotopocules", representing all possible combinations of the nitrogen isotopes ^{14}N and ^{15}N and the oxygen isotopes ^{16}O, ^{17}O, and ^{18}O. A particular feature for the asymmetric N_2O molecule is that the central (α, N^*NO) and the end (β, $^*NNO$) N atom are distinguishable, which has particular potential for source attribution as discussed below. A list of all N_2O isotopocules together with their average abundance and molecular mass can be found in Table 1. The abundance of isotopocules is usually reported in the δ notation and the deviation from a reference standard expressed in ‰ values:

$$\delta X = \left(\frac{R_{sample}}{R_{reference}} - 1 \right) \qquad (1)$$

$$SP = \delta^{15}N^{\alpha} - \delta^{15}N^{\beta} \qquad (2)$$

where X denotes e.g. $^{15}N^{\alpha}$, $^{15}N^{\beta}$, $^{15}N^{bulk}$ (for average ^{15}N content), and R is the ratio between the rare isotopic species and the most abundant species $^{14}N^{14}N^{16}O$ for the sample and the reference gas.[11] The difference between ^{15}N substitution in α and β position, defined according to Eq. (2), is denoted as site preference (SP). Reference standards provide the link to the international scale for isotope ratios, which is atmospheric N_2 (Air-N_2) for nitrogen isotopes and Vienna Standard Mean Ocean Water (VSMOW) for oxygen isotopes.[12]

Table 1. The N_2O isotopocules, their natural average abundance, and molecular masses. For the multiply substituted isotopic species, random (stochastic) distribution of isotopes among all isotopocules is assumed.

isotopocule	relative abundance	mass [u]
$^{14}N^{14}N^{16}O$	$9.903 \cdot 10^{-1}$	44.0011
$^{14}N^{15}N^{16}O$	$3.641 \cdot 10^{-3}$	44.9981
$^{15}N^{14}N^{16}O$	$3.641 \cdot 10^{-3}$	44.9981
$^{14}N^{14}N^{18}O$	$1.986 \cdot 10^{-3}$	46.0053

$^{14}N^{14}N^{17}O$	$3.762 \cdot 10^{-4}$	45.0053
$^{15}N^{15}N^{16}O$	$1.339 \cdot 10^{-5}$	45.9951
$^{14}N^{15}N^{18}O$	$7.300 \cdot 10^{-6}$	47.0023
$^{15}N^{14}N^{18}O$	$7.300 \cdot 10^{-6}$	47.0023
$^{14}N^{15}N^{17}O$	$1.383 \cdot 10^{-6}$	46.0023
$^{15}N^{14}N^{17}O$	$1.383 \cdot 10^{-6}$	46.0023
$^{15}N^{15}N^{18}O$	$2.684 \cdot 10^{-8}$	47.9994
$^{15}N^{15}N^{17}O$	$5.085 \cdot 10^{-9}$	46.9993

In the dual N_2O isotopocule mapping approach, isotope signatures are combined to allocate source processes (Fig. 1). The bulk isotopic composition of N_2O ($\delta^{15}N^{bulk}$, $\delta^{18}O$) is influenced not only by the reaction through which it is produced but also by the composition of the substrate. In contrast, the difference between ^{15}N substitution in α and β position, SP, is independent from the isotopic composition of the precursor and therefore of particular interest to differentiate biogeochemical reaction pathways in the nitrogen cycle.[13] Generally, there is a stronger affinity of ^{15}N towards the central than towards the end N position in the N_2O molecule, due to a lower zero-point energy of $^{14}N^{15}N^{1x}O$ as compared to $^{15}N^{14}N^{1x}O$.[14] While isotopocule mapping has shown potential to study individual reactions, in particular if additional supporting parameters or modelling approaches are available, overlapping isotopocule signatures often prevent a quantitative assignment.
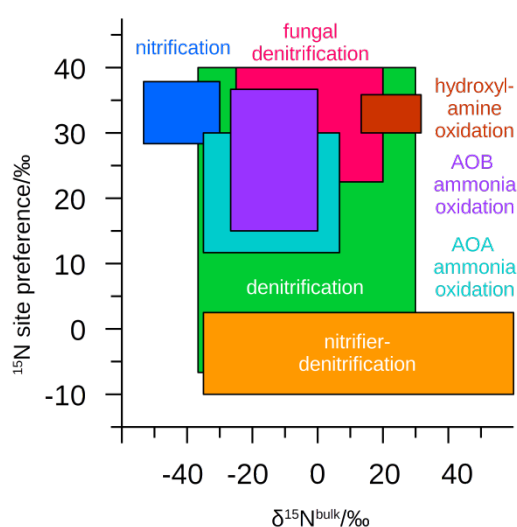

Fig. 1. Dual isotopocule mapping approach with isotopic signatures of several biotic and abiotic sources of N_2O (adapted from literature[13,15]).

Theoretical predictions indicate that multiply substituted, so called "clumped" isotopocules, provide additional constraints on reaction pathways. Under equilibrium conditions, clumping of heavier isotopes in a chemical bond is thermodynamically favorable because "heavier" chemical bonds are more stable.[16] The extent of this phenomenon is temperature-dependent, meaning that the abundance of "clumped isotopes" in a sample bears information about the equilibrium formation temperature and the reversibility of a process.[17–19] In carbonate paleothermometry, this formalism is used to study past climate on Earth.[20–23] Under non-equilibrium kinetic conditions, the clumped signature provides information on the reversibility of a reaction and thus additional dimensionality for interpretation.[24] Apart from their use in source studies, clumped N_2O isotopes have been suggested as powerful tracers for the main sink process, stratospheric photolysis.[25] Concentrations of clumped isotopic species are given in the Δ notation:

$$\Delta 556 = \left[\left(\frac{R556_{sample}}{R556_{stochastic}} - 1\right) - \left(\frac{R456_{sample}}{R456_{stochastic}} - 1\right) - \left(\frac{R546_{sample}}{R546_{stochastic}} - 1\right)\right] \qquad (3)$$

where R556, R456, R546 are the abundance ratios of the isotopocules $^{15}N^{15}N^{16}O$, $^{14}N^{15}N^{16}O$, and $^{15}N^{14}N^{16}O$ relative to $^{14}N^{14}N^{16}O$, in the sample and in a reference gas with stochastic (random) isotopic composition. SP[18] is defined as the difference between $\Delta 458$ and $\Delta 548$.

Traditionally, isotopocules of N_2O are analyzed by isotope-ratio mass spectrometry (IRMS).[11] For the analysis of clumped isotopocules, subtle mass differences have to be resolved, which requires cutting-edge ultra high-resolution IRMS.[26] Although precision and sensitivity of mass spectrometric measurements are usually very high,[27] the technique is only suitable for laboratory-based experiments. In addition, "isotopic isomers" (isotopomers), e.g. $^{14}N^{15}N^{16}O$ and $^{15}N^{14}N^{16}O$, or $^{14}N^{15}N^{18}O$ and $^{15}N^{14}N^{18}O$, cannot be resolved by mass but require a complex analysis of molecular N_2O^+ and fragment NO^+ ions involving correction for isotopic scrambling in the ion source.[11,28]

In recent years, laser spectroscopic techniques for analysis of the singly substituted N_2O isotopocules in the mid-IR have been developed using quantum-cascade laser absorption spectroscopy (QCLAS)[29–33] and cavity ring-down spectroscopy (CRDS).[34,35] They are selective for individual isotopomers, offer precision levels comparable to IRMS, and, in contrast to the latter, are also field-deployable, which is a frequent requirement for environmental applications. Recently, we have adopted the QCLAS technique for the analysis of clumped N_2O isotopocules $^{14}N^{15}N^{18}O$, $^{15}N^{14}N^{18}O$, and $^{15}N^{15}N^{16}O$. Being a completely new and exciting field of research, analysis of clumped N_2O isotopes offers a broad range of prospective applications, in particular on N_2O

produced by microbial, fungal and abiotic sources processes. In a larger perspective, this new technique may also be applied in other research areas such as stratospheric chemistry or industrial catalytic processes. With respect to biogeochemical N_2O cycles, clumped N_2O is expected to significantly advance our understanding by providing a new class of approaches and molecular-scale insights.

2. Principle of QCLAS for isotopic analysis

The mid-infrared (mid-IR) range is a highly attractive spectral region as it provides access to strong fundamental vibrations of many GHG and therefore enables high-precision analysis. Isotope-selective analysis is inherently feasible as the spectral characteristics depend not only on molecular mass, but also on the symmetry of the molecule which are changed upon isotopic substitution. Fig. 2 displays ro-vibrational spectra of the fundamental asymmetric stretching vibration v_3 between 2100 and 2300 cm^{-1} for the ten most abundant N_2O isotopocules. The most adequate spectral windows for laser spectroscopic analysis are indicated by shaded lines. These regions were selected considering the narrow tuning of distributed feedback (DFB) lasers, maximum sensitivity for the low abundant N_2O isotopocules (e.g. $^{15}N^{14}N^{18}O$ with a relative abundance of $7.3\cdot10^{-6}$), comparable intensity for the most abundant isotopic species ($^{14}N^{14}N^{16}O$ with $9.903\cdot10^{-1}$), and minimal spectral interference from other atmospheric trace gases, e.g. CO_2 and CO.
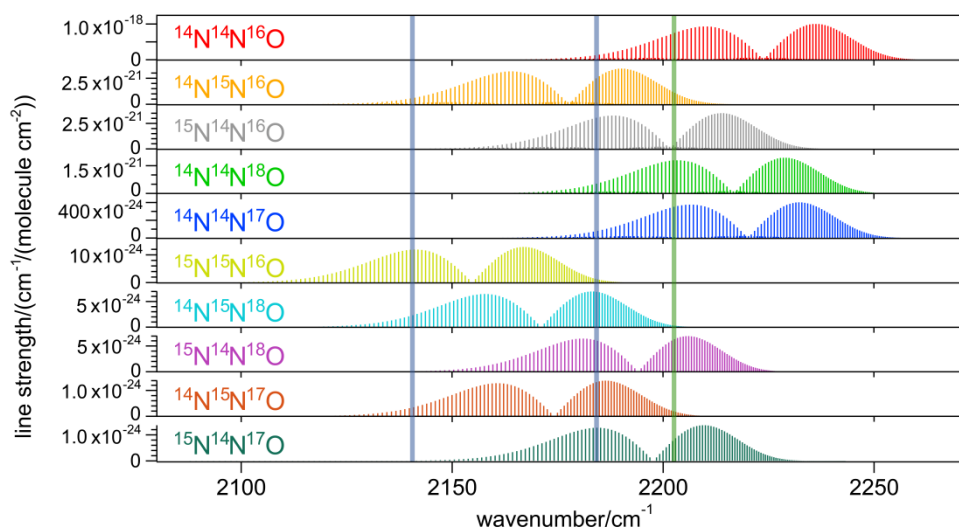

Fig. 2. Absorption bands for the fundamental asymmetric stretching vibration v_3 of the ten most abundant isotopocules of N_2O. The shaded lines indicate spectral regions which are used for analysis of singly (green) and doubly substituted N_2O isotopocules (blue). Linestrength data for $^{14}N^{14}N^{16}O$ and all the singly substituted isotopocules were retrieved from the HITRAN 2012 database.[36] Data for doubly substituted isotopocules were simulated in the software PGOPHER[37] using spectroscopic parameters available in literature.[38–40]

In principle, laser spectroscopy is an absolute method to determine concentrations of individual isotopocules and calculate isotope ratios. In practice, however, the obtained concentration values are affected by a variety of parameters such as uncertainty in gas temperature and pressure, gas matrix, fluctuations in laser temperature and current, optical fringes, etc. To account for these effects and assure long-term stability, a calibration scheme has to be implemented (see Fig. 3b). Thereby, sample gas measurements are bracketed by analyses of calibration gases, and a second calibration gas with different isotopic composition is analysed regularly for two-point calibration of the δ values. In addition, a target gas (compressed air) is measured every few hours in between samples to monitor the data quality of the complete analytical technique. The full analytical routine is automated by a custom-developed LabVIEW software and is robust enough for unattended field campaigns.

Important criteria for data quality in laser spectroscopy are precision and drift/stability of the measured signal as characterized using the two-sample standard deviation (Allan deviation),[41] which describes how the variance evolves in time when the signal is integrated. In Fig. 3a, a representative Allan plot for the $\delta^{14}N^{15}N^{18}O$ is given, where an optimal integration time of around 6 minutes yields an Allan precision of 0.06 ‰. Beyond this time, various drift effects hinder a further increase in precision.
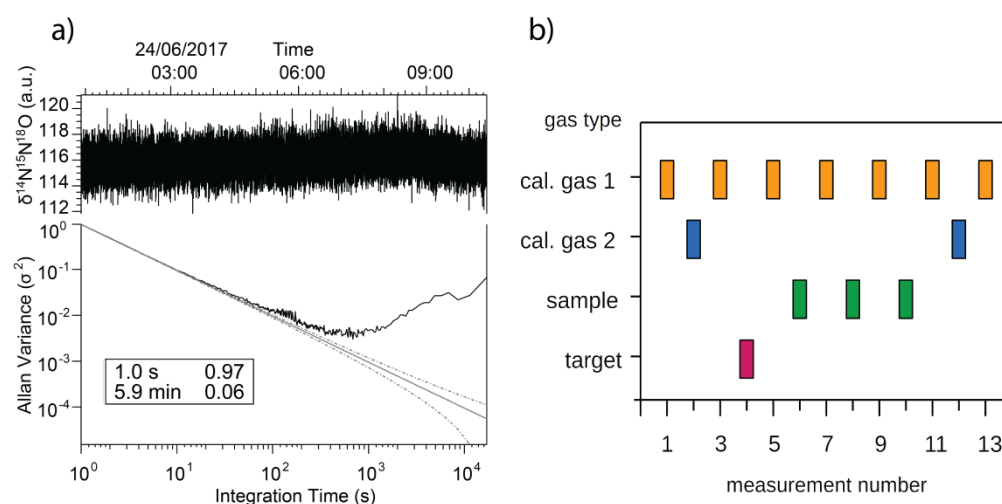

Fig. 3. a) A representative Allan variance plot for $\delta^{14}N^{15}N^{18}O$, indicating a precision of 0.06 ‰ that is achieved after 6 min averaging. The intersect at 1 s shows a precision level of 1 ‰. b) A full measurement cycle implemented for QCLAS analysis. Sample gas measurements are bracketed by analysis of calibration gas 1. For two-point δ-calibration, a second calibration gas is measured regularly, while data quality is monitored by analysis of N_2O in air target gas.

The high-precision analysis of N_2O isotopocules by QCLAS requires enhanced concentration levels and thus preconcentration from ambient air.[42,43] Fig. 4 shows a fully-automated preconcentration device developed at Empa, called TRace gas EXractor (TREX), that has the capability to adsorb N_2O of dried ambient air on a HayeSepD filled adsorption trap. After trapping is finished, N_2O is separated from co-adsorbed gases by a step-wise increase of the trap temperature and flushed into the astigmatic Herriott multi-pass absorption cell of the laser spectrometer using a carrier gas, usually high-purity synthetic air or N_2. Concentrations of individual N_2O isotopocules are analyzed at low cell pressure, to reduce pressure broadening of the absorption lines, using a laser spectrometer from Aerodyne Research Inc. (USA) equipped with appropriate QCLs. Laser scanning, signal processing, and quantitative spectral fitting is fully automated through the software TDL Wintel (Aerodyne Research Inc., USA).
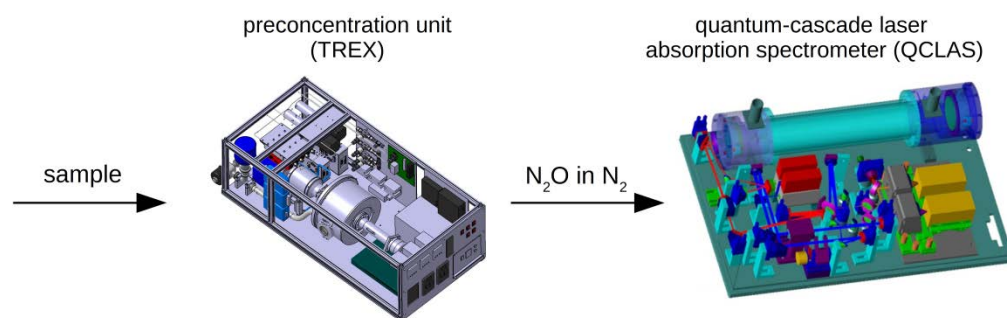

Fig. 4. Scheme of the analytical technique TREX-QCLAS.

3. Applications

Our laboratory pioneers the analysis of novel N_2O isotopocules by laser spectroscopy with a main focus on applications to trace transformation pathways. The first spectrometer developed in 2008[30] was based on a pulsed QCL (2188 cm^{-1}) with the capability for $\delta^{15}N^{\alpha}$ and $\delta^{15}N^{\beta}$ analysis and precision levels of 0.5 ‰ at 90 ppm N_2O. In 2013, we implemented cw-QCL (continuous wave; 2203 cm^{-1}) offering a five-fold increase in precision and the capability for additional analysis of $\delta^{18}O$.[44] Within my PhD thesis, since 2016, we established the first dual cw-QCL spectrometer (2143 and 2183 cm^{-1}) for doubly substituted N_2O isotopocules. This

spectrometer enables specific and high-precision analysis of five singly substituted ($^{14}N^{14}N^{16}O$, $^{14}N^{15}N^{16}O$, $^{15}N^{14}N^{16}O$, $^{14}N^{14}N^{18}O$, $^{14}N^{14}N^{17}O$) and three clumped N$_2$O isotopocules ($^{14}N^{15}N^{18}O$, $^{15}N^{14}N^{18}O$, $^{15}N^{15}N^{16}O$). N$_2$O isotope research, in particular by laser spectroscopy, is a relatively immature technique. Therefore, it is crucial to establish reliable and traceable reference materials. We collaborate with other research groups on their development and on the compatibility of measurements[45], e.g. within the European research project Stable Isotope Reference Standards (SIRS).[46]

Recently, we exploited the capability of QCLAS for real-time measurements of N$_2$O isotopocules in a number of laboratory[44,47] and pilot scale incubation studies.[48] Fig. 5 displays an example of the chemical reaction of hydroxylamine (NH$_2$OH) with nitrite yielding N$_2$O.[44] Our measurements indicate that the produced N$_2$O for all reaction conditions shows a very constant SP value, which is indicative for this process. This finding further implies that the abiotic production of N$_2$O from hydroxylamine proceeds via a defined transition state, which controls the isotopic identity of product N$_2$O. The identified isotopic signature can be used to pinpoint the process while studying the N$_2$O biogeochemical cycle.
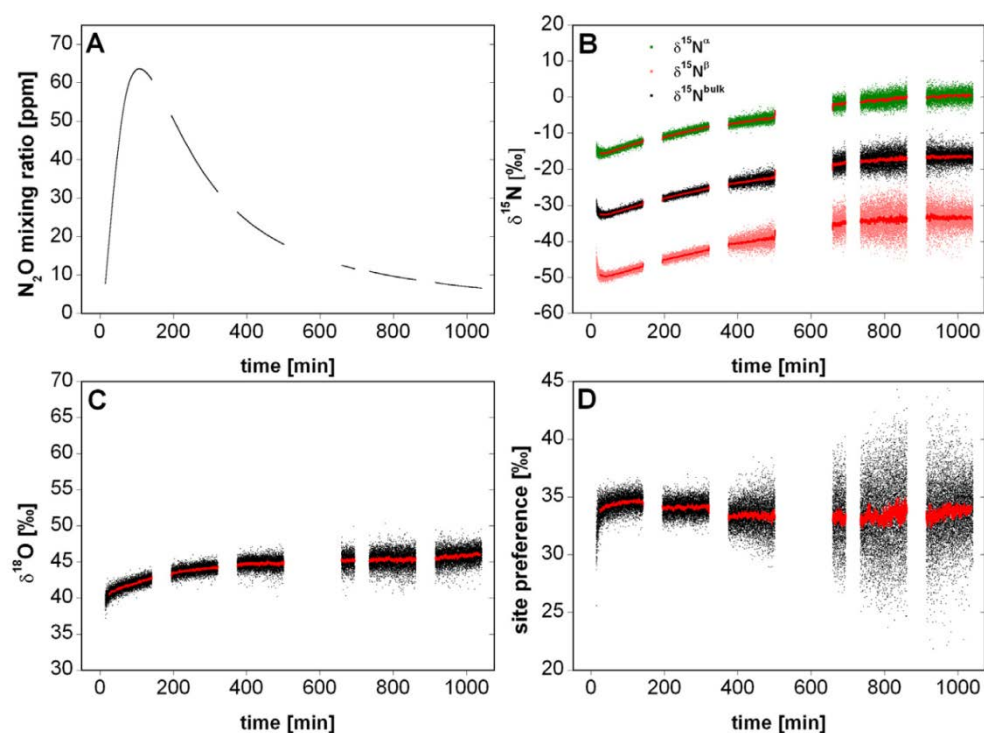
Fig. 5. Mixing ratios (A) and isotopic composition (B – D) of N$_2$O produced by the abiotic reaction between 1 mM NH$_2$OH and 1 mM NO$_2^-$, buffered at pH 4 (citrate–phosphate buffer). Results show that: $\delta^{15}N$ bulk, $\delta^{15}N^{\alpha}$, and $\delta^{15}N^{\beta}$ (B) and $\delta^{18}O$ of N$_2$O (C) display isotopic fractionation, while the N$_2$O site preference (SP) (D) remains constant, indicative of the reaction transition state. All values are 1 Hz data, red lines show the 60 s moving average. Figure reproduced with permission from Elsevier.[44]

In combination with automated preconcentration, TREX-QCLAS allows highly precise real-time measurements of singly substituted N_2O isotopocules even at ambient concentrations around 330 ppb.[32,43] Deviations of N_2O concentrations and isotopic composition from background conditions are interpreted with respect to pollution events and their isotopic signatures determined by using a mixing model, e.g. Keeling or Miller-Tans plots. In two exemplary studies on N_2O source processes in grassland,[29,49] emissions were mainly attributed to microbial denitrification with a variable fraction of N_2O being reduced to N_2. In an extended study at a sub-urban site (Dübendorf, Switzerland), changes in N_2O concentration and derived source signatures were interpreted using an atmospheric modelling approach.[50] In the future, isotopic information is expected to provide additional constraints to validate emission inventories not only for overall source strength but also for individual source categories.

Seasonal variability of N_2O isotopic composition in the atmosphere has been identified in a bi-weekly monitoring project at the high Alpine research station Sphinx, Jungfraujoch since April 2014. The ongoing field observations have clearly shown an increasing trend of atmospheric N_2O mixing ratios by 1.2 ppb·y^{-1}. The high-precision of our analytical technique allowed revealing seasonal variability of the SP, i.e. lowest SP during summer period and highest SP in winter period (Fig. 6). This can be explained by stronger contribution of denitrification, which produces N_2O with generally low SP, in the summer period and vice versa.
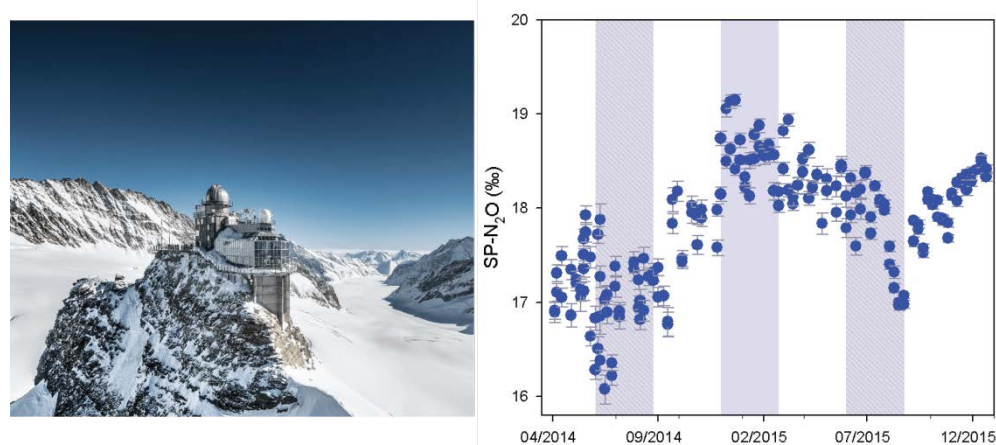

Fig. 6. Seasonal variability of SP in N_2O (right) from the air sampled at the high-altitude research station Sphinx located at Jungraujoch in Switzerland (left).

During my PhD project in the Laboratory for Air Pollution / Environmetal Technology at Empa, I got well acquainted with the field of N_2O isotopocule studies by QCLAS. In the first phase of my thesis, I established and optimized a novel laser spectrometer (Aerodyne Research, Inc. USA) with two QCL light sources emitting at

2143 and 2183 cm^{-1}, for direct, site-specific and high-precision analysis of rare, so far unaccessible "clumped isotopes" of N_2O, $^{14}N^{15}N^{18}O$, $^{15}N^{14}N^{18}O$, and $^{15}N^{15}N^{16}O$. In addition, five singly substituted isotopocules are analysed in the selected spectral region to reference measurements to bulk isotopic composition. Thereby, the instrument provides simultaneous high precision data on eight N_2O isotopocules, which is so far not achievable with any other analyser (neither using laser spectroscopy nor IRMS).

Due to the novelty of this field of research, no reference gases for the studied clumped N_2O isotopocules are available. Therefore, my research is currently focused on the establishment of a new reference frame. Two methods are evaluated: a) linking N_2O composition to stochastic distribution[16] by heating N_2O gases of different isotopic composition over activated Al_2O_3 at two different temperatures, 100 and 200 °C; and b) calibration of individual isotopocules using gravimetrically prepared mixtures of N_2O in N_2. Using two independent approaches will enhance the reliability of the data and support future applications.

In a third work package, I will explore the use of clumped N_2O isotopocules as novel tracers for the N_2O biogeochemical cycle. Part of this work will be done in spring 2019 during an exchange stay in the group of Prof. Naohiro Yoshida at Tokyo Institute of Technology, funded by the Bilateral Japanese-Swiss Science and Technology Program. In short, clumped isotopic signatures of N_2O, in particular SP[18] (difference in abundance of $^{14}N^{15}N^{18}O$ and $^{15}N^{14}N^{18}O$), produced by pure microbial cultures of ammonium oxidizer *Nitrosococcus oceani* and bacterial denitrifiers *Pseudomonas aureofaciens* and *Pseudomonas stutzeri*[51–54] will be tested for their potential to resolve processes that cannot be distinguished with the classical SP approach.

4. Conclusion and outlook

QCLAS has opened a new field of research in the N_2O biogeochemical cycle due to its capability for real-time analysis of isotopic N_2O species in both laboratory and field applications. In combination with automated preconcentration (TREX), laser spectroscopy not only reaches precision levels comparable with state-of-the-art IRMS, but also has inherent selectivity for isotopic isomers. The combined TREX-QCLAS technique has demonstrated its potential to trace changes in the N_2O isotopic composition in various studies, on conventional and novel waste-water treatment processes, snow-covered soils, agriculturally managed grasslands, as well as in chemical and combustion sources.

Within my PhD project, the application of QCLAS has been extended for the analysis of very rare, so called "clumped isotopes" of N_2O. The novel analytical technique offers the unique opportunity for simultaneous high-precision analysis of eight N_2O isotopocules. Theoretical predictions and analogies with clumped CO_2 and CH_4 indicate the potential of clumped N_2O isotopocules as novel reservoir-insensitive tracers for the biogeochemical nitrogen cycle.

[1]   P. Forster, V. Ramaswamy, P. Artaxo, T. Berntsen, R. Betts, D. Fahey, J. Haywood, J. Lean, D. Lowe, G. Myhre, J. Nganga, R. Prinn, G. Raga, M. Schulz, R. V. Dorland, 'Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change', Eds. S. Solomon, D. Qin, M. Manning, Z. Chen, M. Marquis, K. B. Averyt, M.Tignor, H. L. Miller, Cambridge University Press, **2007**.

[2]   B. C. Bates, Z. W. Kundzewicz, S. Wu, J. P. Palutikof, 'Climate Change and Water. Technical Paper of the Intergovernmental Panel on Climate Change' IPCC Secretariat, **2008**.

[3]   A. R. Ravishankara, J. S. Daniel, R. W. Portmann, *Science* **2009**, *326*, 123.

[4]   T. F. Stocker, D. Qin, G.-K. Plattner, M. Tignor, S. K. Allen, J. Boschung, A. Nauels, Y. Xia, V. Bex, P. M. Midgley, 'IPCC, 2013: Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change', Cambridge University Press, **2014**.

[5]   World Meteorological Organization, Global Atmosphere Watch, 'WMO Greenhouse Gas Bulletin (GHG Bulletin) - No. 14', **2018**.

[6]   K. Butterbach-Bahl, E. M. Baggs, M. Dannenmann, R. Kiese, S. Zechmeister-Boltenstern, *Philos. Trans. R. Soc. B Biol. Sci.* **2013**, *368*, 1.

[7]   M. M. M Kuypers, H. K. Marchant, B. Kartal, *Nat. Rev. Microbiol.* **2018**, *16*, 263.

[8]   K. A. Spokas, 'Encyclopedia of Agrophysics', Eds. J. Gliński, J. Horabik, J. Lipiec, Springer Netherlands, **2011**.

[9]   L. B. Flanagan, J. R. Ehleringer, D. E. Pataki, 'Stable Isotopes and Biosphere - Atmosphere Interactions' Academic Press, **2005**.

[10]   T. R. A. Denk, J. Mohn, C. Decock, D. Lewicka-Szczebak, E. Harris, K. Butterbach-Bahl, R. Kiese, B. Wolf, *Soil Biol. Biochem.* **2017**, *105*, 121.

[11]   S. Toyoda, N. Yoshida, *Anal. Chem.* **1999**, *71*, 4711.

[12]   R. A. Werner, W. A. Brand, *Rapid Commun. Mass Spectrom.* **2001**, *15*, 501.

[13]   S. Toyoda, N. Yoshida, K. Koba, *Mass Spectrom. Rev.* **2017**, *36*, 135.

[14]   P. Richet, 'The Physical Basis of Thermodynamics With Applications to Chemistry' Springer Science+Business Media, **2001**.

[15]   H.-W. Hu, D. Chen, J.-Z. He, *FEMS Microbiol. Rev.* **2015**, *39*, 729.

[16]   Z. Wang, E. A. Schauble, J. M. Eiler, *Geochim. Cosmochim. Acta* **2004**, *68*, 4779.

[17]   J. M. Eiler, *Earth Planet. Sci. Lett.* **2007**, *262*, 309.

[18]   J. M. Eiler, *Annu. Rev. Earth Planet. Sci.* **2013**, *41*, 411.

[19]   J. M. Eiler, B. Bergquist, I. Bourg, P. Cartigny, J. Farquhar, A. Gagnon, W. Guo, I. Halevy, A. Hofmann, T. E. Larson, N. Levin, E. A. Schauble, D. Stolper, *Chem. Geol.* **2014**, *372*, 119.

[20]   A. L. Grauel, T. W. Schmid, B. Hu, C. Bergami, L. Capotondi, L. Zhou, S. M. Bernasconi, *Geochim. Cosmochim. Acta* **2013**, *108*, 125.

[21]   K. W. Huntington, J. M. Eiler, H. P. Affek, W. Guo, M. Bonifacie, L. Y. Yeung, N. Thiagarajan, B. Passey, A. Tripati, M. Daëron, R. Came, *J. Mass Spectrom.* **2009**, *44*, 1318.

[22]   S. Kele, S. F. M. Breitenbach, E. Capezzuoli, A. N. Meckler, M. Ziegler, I. M. Millan, T. Kluge, J. Deák, K. Hanselmann, C. M. John, H. Yan, Z. Liu, S. M. Bernasconi, *Geochim. Cosmochim. Acta* **2015**, *168*, 172.

[23]   A. N. Meckler, S. Affolter, Y. V. Dublyansky, Y. Krüger, N. Vogel, S. M. Bernasconi, M. Frenz, R. Kipfer, M. Leuenberger, C. Spötl, S. Carolin, K. M. Cobb, J. Moerman, J. F. Adkins, D. Fleitmann, *Quat. Sci. Rev.* **2015**, *127*, 90.

[24]   D. T. Wang, D. S. Gruen, B. S. Lollar, K. Hinrichs, L. C. Stewart, J. F. Holden, A. N. Hristov, J. W. Pohlman, P. L. Morrill, M. Könneke, B. Delwiche, E. P. Reeves, C. N. Sutcliffe, D. J. Ritter, J. S. Seewald, K. B. Delwiche, E. P. Reeves, C. N. Sutcliffe, D. J. Ritter, J. S. Seewald, J. C. McIntosh, H. F. Hemond, M. D. Kubo, D. Cardace, T. M.

Hoehler, S. Ono, *Science* **2015**, *348*, 428.

[25]   J. A. Schmidt, M. S. Johnson, *Geophys. Res. Lett.* **2015**, *42*, 3546.

[26]   P. M. Magyar, V. J. Orphan, J. M. Eiler, *Rapid Commun. Mass Spectrom.* **2016**, *30*, 1923.

[27]   S. Toyoda, N. Yoshida, *Atmos. Meas. Tech.* **2016**, *9*, 2093.

[28]   T. Röckmann, J. Kaiser, C. A. M. Brenninkmeijer, W. A. Brand, *Rapid Commun. Mass Spectrom.* **2003**, *17*, 1897.

[29]   B. Wolf, L. Merbold, C. Decock, B. Tuzson, E. Harris, J. Six, L. Emmenegger, J. Mohn, *Biogeosciences* **2015**, *12*, 2517.

[30]   H. Waechter, J. Mohn, B. Tuzson, L. Emmenegger, M. W. Sigrist, *Opt. Express* **2008**, *16*, 9239.

[31]   B. Tuzson, J. Mohn, M. J. Zeeman, R. A. Werner, W. Eugster, M. S. Zahniser, D. D. Nelson, J. B. McManus, L. Emmenegger, *Appl. Phys. B Lasers Opt.* **2008**, *92*, 451.

[32]   J. Mohn, B. Tuzson, A. Manninen, N. Yoshida, S. Toyoda, W. A. Brand, L. Emmenegger, *Atmos. Meas. Tech.* **2012**, *5*, 1601.

[33]   P. Sturm, B. Tuzson, S. Henne, L. Emmenegger, *Atmos. Meas. Tech.* **2013**, *6*, 1659.

[34]   D. V. Erler, T. M. Duncan, R. Murray, D. T. Maher, I. R. Santos, J. R. Gatland, P. Mangion, B. D. Eyre, *Limnol. Oceanogr. Methods* **2015**, *13*, 391.

[35]   P. Gupta, D. Noone, J. Galewsky, C. Sweeney, B. H. Vaughn, *Rapid Commun. Mass Spectrom.* **2009**, *23*, 2534.

[36]   L. S. Rothman, I. E. Gordon, Y. Babikov, A. Barbe, D. Chris Benner, P. F. Bernath, M. Birk, L. Bizzocchi, V. Boudon, L. R. Brown, A. Campargue, K. Chance, E. A. Cohen, L. H. Coudert, V. M. Devi, B. J. Drouin, A. Fayt, J.-M. Flaud, R. R. Gamache, J. J. Harrison, J.-M. Hartmann, C. Hill, J. T. Hodges, D. Jacquemart, A. Jolly, J. Lamouroux, R. J. Le Roy, G. Li, D. A. Long, O. M. Lyulin, C. J. Mackie, S. T. Massie, S. Mikhailenko, H. S. P. Müller, O. V. Naumenko, A. V. Nikitin, J. Orphal, V. Perevalov, A. Perrin, E. R. Polovtseva, C. Richard, M. A. H. Smith, E. Starikova, K. Sung, S. Tashkun, J. Tennyson, G. C. Toon, V. G. Tyuterev, G. Wagner, *J. Quant. Spectrosc. Radiat. Transf.* **2013**, *130*, 4.

[37]   C. M. Western, *J. Quant. Spectrosc. Radiat. Transf.* **2017**, *186*, 221.

[38]   R. A. Toth, *Appl. Opt.* **1991**, *30*, 5289.

[39]   J. Du, A. Liu, V. I. Perevalov, S. A. Tashkun, S. Hu, *Chinese J. Chem. Phys.* **2011**, *24*, 611.

[40] C. Y. Wang, A. W. Liu, V. I. Perevalov, S. A. Tashkun, K. F. Song, S. M. Hu, *J. Mol. Spectrosc.* **2009**, *257*, 94.

[41] P. Werle, R. Mücke, F. Slemr, *Appl. Phys. B Photophysics Laser Chem.* **1993**, *57*, 131.

[42] J. Mohn, C. Guggenheim, B. Tuzson, M. K. Vollmer, S. Toyoda, N. Yoshida, L. Emmenegger, *Atmos. Meas. Tech.* **2010**, *3*, 609.

[43] E. Ibraim, E. Harris, S. Eyer, B. Tuzson, L. Emmenegger, J. Six, J. Mohn, *Isotopes Environ. Health Stud.* **2017**, *54*, 1.

[44] J. Heil, B. Wolf, N. Brüggemann, L. Emmenegger, B. Tuzson, H. Vereecken, J. Mohn, *Geochim. Cosmochim. Acta* **2014**, *139*, 72.

[45] J. Mohn, B. Wolf, S. Toyoda, C. T. Lin, M. C. Liang, N. Brüggemann, H. Wissel, A. E. Steiker, J. Dyckmans, L. Szwec, N. E. Ostrom, K. L. Casciotti, M. Forbes, A. Giesemann, R. Well, R. R. Doucett, C. T. Yarnes, A. R. Ridley, J. Kaiser, N. Yoshida, *Rapid Commun. Mass Spectrom.* **2014**, *28*, 1995.

[46] N. E. Ostrom, H. Gandhi, T. B. Coplen, S. Toyoda, J. K. Böhlke, W. A. Brand, K. L. Casciotti, J. Dyckmans, A. Giesemann, J. Mohn, R. Well, L. Yu, N. Yoshida, *Rapid Commun. Mass Spectrom.* **2018**, *32*, 1207.

[47] J. R. Köster, R. Well, B. Tuzson, R. Bol, K. Dittert, A. Giesemann, L. Emmenegger, A. Manninen, L. Cárdenas, J. Mohn, *Rapid Commun. Mass Spectrom.* **2013**, *27*, 216.

[48] E. Harris, A. Joss, L. Emmenegger, M. Kipf, B. Wolf, J. Mohn, P. Wunderlin, *Water Res.* **2015**, *83*, 258.

[49] E. Ibraim, B. Wolf, E. Harris, R. Gasche, J. Wei, L. Yu, R. Kiese, S. Eggleston, K. Butterbach-Bahl, M. Zeeman, B. Tuzson, L. Emmenegger, J. Six, S. Henne, J. Mohn, *Biogeosciences Discuss.* **2018**, *in review*.

[50] E. Harris, S. Henne, C. Hüglin, C. Zellweger, B. Tuzson, E. Ibraim, L. Emmenegger, J. Mohn, *J. Geophys. Res. Atmos.* **2017**, *122*, 1850.

[51] T. Yamazaki, T. Hozuki, K. Arai, S. Toyoda, K. Koba, T. Fujiwara, N. Yoshida, *Biogeosciences* **2014**, *11*, 2679.

[52] R. Well, H. Flessa, *Rapid Commun. Mass Spectrom.* **2009**, *23*, 2996.

[53] S. Toyoda, H. Mutobe, H. Yamagishi, N. Yoshida, Y. Tanji, *Soil Biol. Biochem.* **2005**, *37*, 1535.

[54] S. Hattori, J. Savarino, K. Kamezaki, S. Ishino, J. Dyckmans, T. Fujinawa, N. Caillon, A. Barbero, A. Mukotaka, S. Toyoda, R. Well, N. Yoshida, *Rapid Commun. Mass Spectrom.* **2016**, *30*, 2635.