



# Estimating global terrestrial denitrification from measured N<sub>2</sub>O:(N<sub>2</sub>O + N<sub>2</sub>) product ratios<sup>☆</sup>

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The use of nitrogen (N) fertilizers has grown exponentially over the last century, with severe environmental consequences. Most of the anthropogenic reactive nitrogen will ultimately be returned by denitrification to the atmosphere as inert N<sub>2</sub>, but the magnitude of denitrification and the ratio of N<sub>2</sub>O to (N<sub>2</sub>O+N<sub>2</sub>) emitted (R<sub>N<sub>2</sub>O</sub>) is unknown for the vast majority of terrestrial ecosystems. This paper provides estimates of terrestrial denitrification and R<sub>N<sub>2</sub>O</sub> by reviewing existing literature and compiling a N budget for the global land surface. We estimate that terrestrial denitrification has doubled from 80 Tg-N year<sup>-1</sup> in pre-industrial times to 160 Tg-N year<sup>-1</sup> in 2005 and that at the global scale N<sub>2</sub>O comprises approximately 8% (6–11%) of the terrestrial denitrification flux. We conclude that upscaling of R<sub>N<sub>2</sub>O</sub> can provide spatial estimates of terrestrial denitrification when data from acetylene inhibition methods are excluded. Recent advances in methodologies to measure N<sub>2</sub> emissions and R<sub>N<sub>2</sub>O</sub> under field conditions could open the way for more effective management of terrestrial N flows.

## Addresses

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## Introduction

The Green (Agricultural) Revolution has primarily been fuelled by nitrogen (N), with the required reactive N (N<sub>r</sub>) provided mainly by increased fertilizer use but also by increased cultivation of N-fixing crops; both of which have increased the amount of N<sub>r</sub> in the biosphere [1<sup>•</sup>]. As a result, the global N cycle is more severely altered by human activity than the global carbon (C) cycle [2]. While humans have doubled levels of N<sub>r</sub> in circulation, the global C cycle has only been altered by 5–7% due to human activity. The increased accumulation of N<sub>r</sub> in the biosphere is causing serious environmental impacts, including eutrophication, biodiversity loss, human health problems and perturbations of the climate system [3,4<sup>•</sup>,5].

Two microbial processes (nitrification and denitrification) play key roles in the removal of N<sub>r</sub> from soils. Nitrification first oxidizes ammonia/ammonium (NH<sub>3</sub>/NH<sub>4</sub><sup>+</sup>) to nitrate (NO<sub>3</sub><sup>-</sup>), which can then be reduced via denitrification to nitrite (NO<sub>2</sub><sup>-</sup>), nitric oxide (NO), nitrous oxide (N<sub>2</sub>O) and finally dinitrogen (N<sub>2</sub>). Denitrification is the primary sink for N<sub>r</sub> by the conversion to inert N<sub>2</sub> and its release back to the atmosphere, although other processes like anaerobic ammonium oxidation (anammox) may play an important role in the removal of N<sub>r</sub> in aquatic ecosystems [6]. Unfortunately, N<sub>2</sub> fluxes are very difficult to quantify because of the high atmospheric N<sub>2</sub> concentration, substantially hampering our ability to estimate terrestrial N budgets. There are several main N loss pathways from fertilised agro-ecosystems, including NH<sub>3</sub> volatilisation, NO<sub>3</sub><sup>-</sup> leaching, and NO, N<sub>2</sub>O and N<sub>2</sub> emissions. These N losses often represent a substantial loss of applied fertiliser; possibly resulting in less plant N uptake and reducing crop N use efficiency (NUE). Furthermore, these losses create environmental concerns; particularly for N<sub>2</sub>O, which is not only a potent greenhouse gas with a global warming potential 298 times greater than CO<sub>2</sub>, but is also currently the primary contributor to stratospheric ozone depletion [7<sup>•</sup>,8].

Rates of both denitrification and N<sub>2</sub>O emissions have increased with the augmented use of N fertiliser and animal manure and it has been clearly shown that N<sub>2</sub>O

<sup>☆</sup> OECD Disclaimer: The opinions expressed and arguments employed in this publication are the sole responsibility of the authors and do not necessarily reflect those of the OECD or of the governments of its Member countries.

emissions increase exponentially once plant N requirements have been met [9]. Consequently, further intensification of global cropping systems to feed a growing population that includes higher inputs of N fertiliser will result in even higher N<sub>2</sub>O emissions per unit of N fertilizer used, which if abatement action is not taken, could lead to a doubling of N<sub>2</sub>O emissions by 2050 [7•,10]. Therefore, a better understanding of terrestrial denitrification (i.e. the dominant process for N<sub>r</sub> removal and N<sub>2</sub>O emissions) is essential for the development of management strategies that improve crop yields while also mitigating N<sub>r</sub> losses. In spite of its importance, terrestrial denitrification is still one of the least well understood processes in the global N cycle [11••], with the magnitude of total denitrification losses (N<sub>2</sub> + N<sub>2</sub>O) virtually unknown for the vast majority of terrestrial ecosystems [12••].

This paper provides estimates of terrestrial denitrification and the product ratio of N<sub>2</sub>O to (N<sub>2</sub>O + N<sub>2</sub>) emitted (R<sub>N<sub>2</sub>O</sub>), by reviewing existing literature and compiling a N mass balance for the global land surface, where we balanced rates of N<sub>r</sub> creation for the global land surface with outputs to the ocean and atmosphere. Finally, we calculated terrestrial denitrification based on measured R<sub>N<sub>2</sub>O</sub> and assessed how well such an estimate compares with other methods.

### Global estimates of terrestrial N<sub>2</sub>O emissions

Existing information about spatial and temporal patterns of N<sub>2</sub>O exchange between terrestrial ecosystems and the atmosphere at a regional or global scale is uncertain, despite its important role in the Earth's climate system. Currently there are two main approaches for estimating N<sub>2</sub>O emissions from terrestrial ecosystems: bottom-up and top-down approaches. Bottom-up approaches extrapolate site specific emission estimates based on field measurements or modelling results to estimate regional and global N<sub>2</sub>O budgets, while top-down approaches infer N<sub>2</sub>O emissions based on changes in atmospheric N<sub>2</sub>O mixing ratios and an inversion model [7•,13].

The most common bottom-up method used to quantify terrestrial N<sub>2</sub>O emissions is based on emission factors (EFs) that estimate N<sub>2</sub>O emissions as a percentage of soil N inputs. Current inventories of N<sub>2</sub>O emissions from agriculture are mainly based on standard Tier 1 IPCC methodologies, which uses a default emission factor (1% of N inputs in upland systems on mineral soils) and agricultural census data on cropping area and fertilizer use [14]. However, the assumption of a default EF has been questioned from its inception since it does not reflect underlying processes and the impact of management or environmental conditions. A particular concern is that a static EF assumes a linear relationship between N application rates and N<sub>2</sub>O emissions, whereas for many fertilized cropping systems the relationship is non-linear [7•,9]. Another important bottom-up approach is the use

of process-based models and over the last decade, several studies have used different simulation models to estimate N<sub>2</sub>O emissions from terrestrial soils at the global scale [15–19]. Top-down approaches provide an integrated picture of the regional and long-term emissions and a check on the total budget [20]. With improved data precision and modeling capabilities over the recent years, these top-down approaches have been used to determine the spatial and temporal distribution of N<sub>2</sub>O emissions [20–24].

Other recent studies have used a combination of 'top-down' constraints with a bottom-up global inventory approach. This approach compares the observed changes in atmospheric N<sub>2</sub>O mixing ratios over time with the amount of newly fixed N (via Haber–Bosch and biological N fixation in soils) to derive the fraction of newly fixed N that is converted to N<sub>2</sub>O. They estimate that 3–5% of newly fixed N is converted to N<sub>2</sub>O when direct and indirect effects (i.e. cascading effects of fixed N via leaching, runoff, atmospheric deposition, and disposal of human sewage) are considered [7•,26,30].

Both bottom-up and top-down estimates of terrestrial N<sub>2</sub>O emissions on global scale are subject to large uncertainties, but it has been shown that estimates of both methods converge at large scales [13]. Estimates of terrestrial N<sub>2</sub>O emissions derived from previous bottom-up ( $n = 4$ ) and top-down ( $n = 6$ ) studies ranged from 11.1 to 15.2 Tg N year<sup>-1</sup> with mean global terrestrial N<sub>2</sub>O emissions estimated to be  $12.9 \pm 0.4$  Tg N year<sup>-1</sup>. Soils under natural vegetation account for the largest part of terrestrial N<sub>2</sub>O emissions (4.9–9.3 Tg N year<sup>-1</sup>), while agricultural activities contribute 5.0–6.8 Tg N year<sup>-1</sup> to the terrestrial N<sub>2</sub>O budget (Table 1).

### Global estimates of terrestrial denitrification

The main reasons for the uncertainty around terrestrial denitrification rates are that there is not only high spatial and temporal variability, but it is also extremely difficult to measure N<sub>2</sub> emissions against the huge atmospheric N<sub>2</sub> background [31]. As a consequence, only few estimates of denitrification on site, landscape (100 000 m<sup>2</sup>), regional (>100 km<sup>2</sup>) and global scales exist and these come with large uncertainties. Recent research suggests that denitrification may be the dominant N loss pathway from agricultural soils and natural ecosystems alike [32–34], while the importance of anammox as a N<sub>2</sub> producing process remains still largely unquantified for terrestrial systems [6]. Estimates of total global denitrification (i.e. from marine and terrestrial systems combined) vary from 220 to 570 Tg-N year<sup>-1</sup> [35–37,38••], although global denitrification rates in excess of 450 Tg-N year<sup>-1</sup> are difficult to reconcile with current estimates of total global N fixation (fertilizer, biological N fixation, energy processes, lightning), which range from 390 to 440 Tg-N year<sup>-1</sup> [28•,35,36]. This discrepancy may be partially

**Table 1****Estimates of nitrous oxide emissions from terrestrial ecosystems (Tg N<sub>2</sub>O-N year<sup>-1</sup>)**

|                              | Total terrestrial <sup>a</sup> | Soils under natural vegetation | Total agricultural <sup>b</sup> | Other sources <sup>c</sup> | Method   |
|------------------------------|--------------------------------|--------------------------------|---------------------------------|----------------------------|--|
| <b>Bottom up</b>             |                                |                                |                                 |                            |  |
| Mosier <i>et al.</i> [25]    | –                              | –                              | 6.8                             | 1.3                        | Emission factors (IPCC [1998] methodology)   |
| DelGrosso <i>et al.</i> [13] | –                              | –                              | 5.8                             | –                          | Combined DAYCENT model simulations and IPCC [2006] methodology   |
| Davidson [26]                | –                              | –                              | 5.5                             | 0.8                        | Emission factors for manure and fertiliser-N   |
| Syakila and Kroeze [27]      | 13.7                           | 6.5                            | 6.0                             | 1.2                        | Revised emissions factors  |
| Xu-Ri <i>et al.</i> [19]     | –                              | 9.3                            | –                               | –                          | Global vegetation model (DyN-LPJ)  |
| Bouwman <i>et al.</i> [11**] | 13.5                           | 4.9                            | 6.7                             | 1.9                        | Integrated Model to Assess the Global Environment (IMAGE)  |
| Ciais <i>et al.</i> [28*]    | 13.3                           | 6.6                            | 5.4                             | 0.9                        | Upscaling of local flux measurements, emission factors   |
| Tian <i>et al.</i> [29]      | 15.2                           | 8.4                            | 5.5                             | 0.3                        | Meta analysis of bottom up estimates   |
| Tian <i>et al.</i> [18]      | –                              | 6.5                            | –                               | –                          | Ensemble of terrestrial biosphere models   |
| Mean bottom-up               | 13.9 ( <i>n</i> = 4)           | 7.0 ( <i>n</i> = 6)            | 6.0 ( <i>n</i> = 7)             | 1.1 ( <i>n</i> = 6)        |  |
| <b>Top down</b>              |                                |                                |                                 |                            |  |
| Hirsch <i>et al.</i> [21]    | 11.4                           | –                              | –                               | –                          | Atmospheric inversion modelling  |
| DelGrosso <i>et al.</i> [13] | –                              | –                              | 5.6                             | –                          | Changes in atmospheric N <sub>2</sub> O concentration and N <sub>2</sub> O removal rates. Emission factor for newly fixed N <sub>r</sub> |
| Crutzen <i>et al.</i> [30]   | 12.8                           | 6.7                            | 5.1                             | 1.0                        | Changes in atmospheric N <sub>2</sub> O concentration and N <sub>2</sub> O removal rates. Emission factor for newly fixed N <sub>r</sub> |
| Huang <i>et al.</i> [22]     | –                              | 6.7                            | –                               | –                          | Atmospheric inversion modelling  |
| Saikawa <i>et al.</i> [24]   | 12.5                           | 7.7                            | –                               | 0.8                        | Atmospheric inversion modelling  |
| Thompson <i>et al.</i> [23]  | 12.9                           | –                              | –                               | –                          | Atmospheric inversion modelling  |
| Thompson <i>et al.</i> [20]  | 11.1                           | –                              | –                               | –                          | Atmospheric inversion modelling  |
| Tian <i>et al.</i> [29]      | 12.6                           | 7.5                            | 5.0                             | –                          | Meta analysis of atmospheric inversion modelling studies   |
| Mean top-down                | 12.2 ( <i>n</i> = 6)           | 7.1 ( <i>n</i> = 4)            | 5.2 ( <i>n</i> = 3)             | 1.0 ( <i>n</i> = 2)        |  |
| Total mean                   | 12.9 ± 0.4                     | 7.1 ± 0.4                      | 5.7 ± 0.2                       | 1.1 ± 0.2                  |  |

<sup>a</sup> Total emissions from terrestrial ecosystems including fossil fuel combustion and industrial processes.<sup>b</sup> Direct and indirect soil emissions including manure management and biomass burning.<sup>c</sup> Fossil fuel combustion, industrial processes and human sewage.

explained by the high uncertainty in estimates of terrestrial (77–387 Tg-N year<sup>-1</sup>) and marine (75–200 Tg-N year<sup>-1</sup>) biological N fixation (BNF) [6,39], and marine denitrification (100–330 Tg-N year<sup>-1</sup>) [35].

We found 10 studies that estimated denitrification rates for terrestrial ecosystems at the global scale (Table 2). In this paper the term terrestrial denitrification is used for NO<sub>3</sub><sup>-</sup> reduction to N<sub>2</sub>, N<sub>2</sub>O and NO occurring on the global land surface including upland soils, wetlands, riparian zones, and freshwater aquatic ecosystems. Typically, denitrification requires an anoxic environment with a ready supply of NO<sub>3</sub><sup>-</sup> and organic carbon, so terrestrial ecosystems characterized by high water contents (e.g. wetlands, riparian zones and heavily irrigated cropping systems) are considered hot-spots for denitrification [36,40]. However, it has also been shown that anaerobic microsites within soil aggregates can produce hot-spots and hot moments of denitrification activity, resulting in significant contributions to denitrification at the landscape scale from apparently well-drained soils [41,42].

Global estimates of terrestrial denitrification range from 100 to 250 Tg-N year<sup>-1</sup> (Mean 157 ± 27 Tg-N year<sup>-1</sup>), equivalent to about 50% of newly fixed terrestrial N [35]. Soils account for the largest proportion of terrestrial denitrification (67–130 Tg-N year<sup>-1</sup>) with about half (22–87 Tg-N year<sup>-1</sup>) occurring in fertilized agricultural soils, while 24–44 Tg-N year<sup>-1</sup> is denitrified in the groundwater and 37–66 Tg-N year<sup>-1</sup> in lakes, rivers and reservoirs (Table 2). However, there is still a high uncertainty in estimates of denitrification in wetlands and freshwater systems, since only few estimates exist and denitrification in aquatic systems is often confounded with other microbial pathways that potentially remove nitrate, such as reduction of nitrate to ammonium (DNRA), and anammox [6]. In addition, it is hard to distinguish between aquatic and soil denitrification because denitrification often occurs at the interface of aquatic and soil ecosystems (e.g. wetlands, riparian zones) which are key zones in controlling the flow of terrestrial N<sub>r</sub> to rivers and the oceans [11\*\*].

**Table 2****Global estimates of terrestrial denitrification ( $\text{Tg N year}^{-1}$ )**

|  | Soils       | Agricultural soils | Ground-water | Riparian zone | Lakes and rivers | Total terrestrial | Ocean        | Global       |
|--|-------------|--------------------|--------------|---------------|------------------|-------------------|--------------|--------------|
| Galloway <i>et al.</i> [36] (1990s)      | 67          |                    |              |               | 48               | 125               | 278          | 403          |
| Hofstra and Bouwman [43]                 |             | 22–87              |              |               |                  |                   |              |              |
| Seitzinger <i>et al.</i> [38**]          | 124         | 66                 | 44           |               | 66               | 242               | 331          | 565          |
| Gruber and Galloway [37]                 | 127         |                    |              |               | 37               | 164               | 244          | 408          |
| Schlesinger [44**]                       |             | 17                 |              |               |                  | 109               |              |              |
| Canfield <i>et al.</i> [45]              |             |                    |              |               |                  | 100               | 238          | 338          |
| Bouwman <i>et al.</i> [11**] (year 2000) | 79          | 51                 | 24           | 6             |                  | 109               |              |              |
| Fowler <i>et al.</i> [35]                | 113         |                    |              |               |                  |                   | 108–285      | 218–399      |
| Zaehle [46]                              | 115         | 47                 |              |               |                  |                   |              |              |
| Battye <i>et al.</i> [1*]                | 130         |                    |              |               | 120 <sup>a</sup> | 250               | 150          | 400          |
| Mean ( $\pm \text{SE}$ )                 | 108 $\pm$ 9 | 48 $\pm$ 10        | 34 $\pm$ 5   | 6             | 50 $\pm$ 7       | 157 $\pm$ 24      | 233 $\pm$ 30 | 390 $\pm$ 51 |

<sup>a</sup> Estimate for freshwater and groundwater combined, not counted for the mean.

### Estimating total terrestrial denitrification from N balance

Terrestrial denitrification rates were also calculated for the global land surface using a mass balance approach where we juxtapose rates of  $N_r$  creation for the global land surface with outputs to the ocean and atmosphere (Figure 1). For a balanced budget the difference between the input and the output will be either stored in terrestrial ‘sinks’ (e.g. in surface and groundwater, the biosphere or terrestrial soils) or denitrified and released as inert  $N_2$  back to the atmosphere. We estimate that in 2005,  $N_r$  input rates for the global land surface were  $286 \text{ Tg N year}^{-1}$  ( $98 \text{ Tg N year}^{-1}$  as fertilizer via the Haber–Bosch process [28\*, 47, 48];  $93 \text{ Tg N year}^{-1}$  via Biological Nitrogen Fixation (BNF) in natural ecosystems [39, 47, 49];  $60 \text{ Tg N year}^{-1}$  via BNF in agricultural systems [50];  $30 \text{ Tg N year}^{-1}$  via fossil fuel combustion [35, 51]; and  $5 \text{ Tg N year}^{-1}$  via lightning [35]). Of this  $90 \text{ Tg N year}^{-1}$  were transported to the ocean via river flow or atmospheric deposition [52–54],  $36 \text{ Tg N year}^{-1}$  stored in terrestrial or inland aquatic ecosystems [11\*\*, 46], and  $12.9 \text{ Tg N year}^{-1}$  emitted as  $N_2O$  to the atmosphere (Table 1). This provides an estimate of  $147 \text{ Tg N year}^{-1}$  emitted as  $N_2$  from terrestrial ecosystems to the atmosphere. If we assume that the  $12.9 \text{ Tg N}_2\text{O-N year}^{-1}$  emissions mentioned above (Table 1) are produced entirely through denitrification, then  $160 \text{ Tg N year}^{-1}$  are denitrified from the terrestrial land surface with a mean  $R_{N_2O}$  of approximately 0.08. Using the same approach, we estimate that in pre-industrial times terrestrial  $N_r$  creation was  $110 \text{ Tg N year}^{-1}$ , and  $80 \text{ Tg-N year}^{-1}$  denitrified and released to the atmosphere (Figure 1).

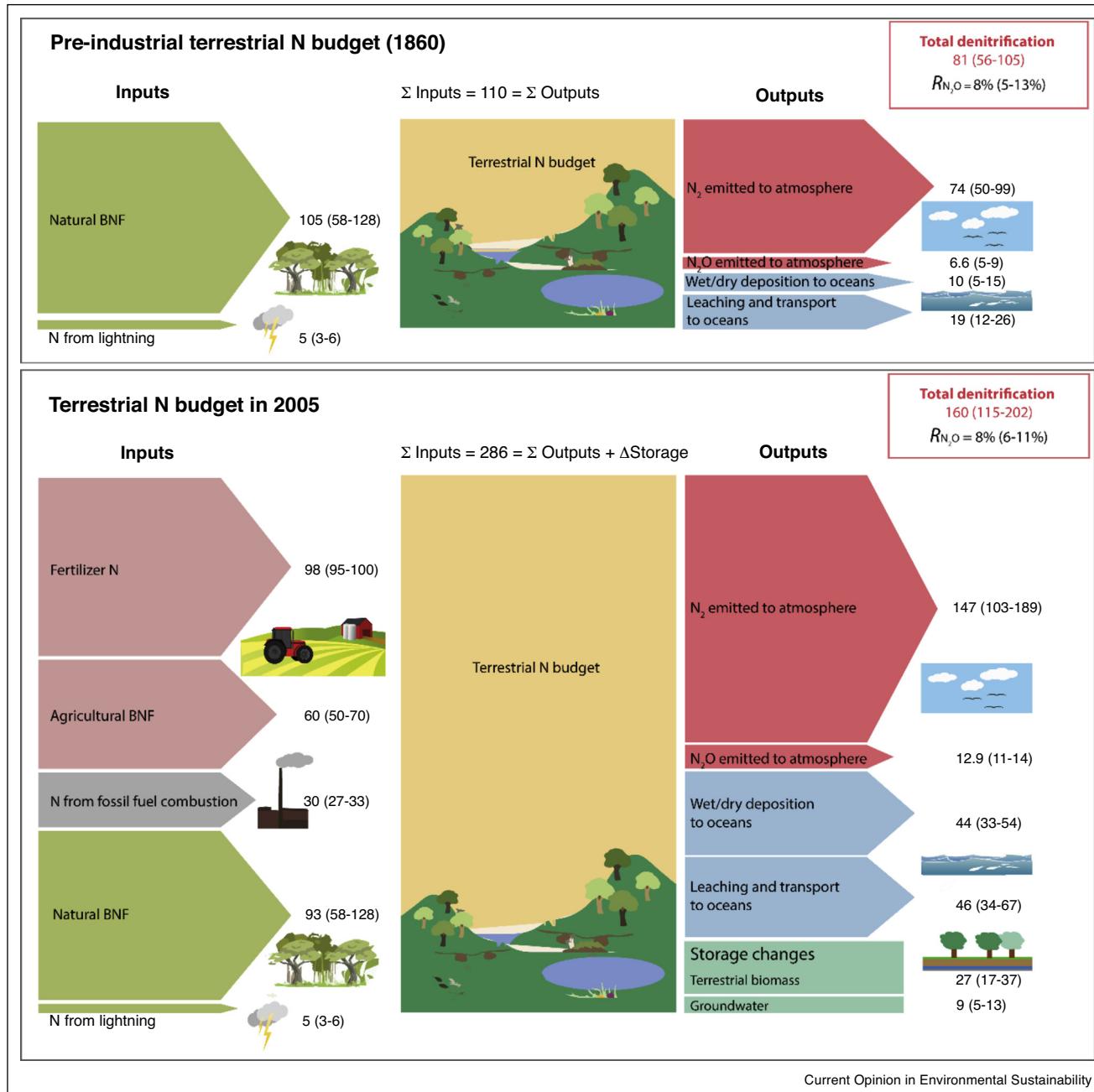
### Estimating terrestrial denitrification from $N_2O$ emissions and $R_{N_2O}$

Another approach to estimate total denitrification rates from terrestrial ecosystems is to use the ratio of  $N_2O$  to the total  $N_2+N_2O$  emitted from denitrification ( $R_{N_2O}$ ) as measured in more controlled, local studies and then

scaling these up to regional or global estimates [44\*\*]. But high uncertainties in  $N_2$  flux estimates exist, caused by the high atmospheric  $N_2$  background. Moreover, many studies on soil denitrification have been conducted in laboratory incubations, with high levels of  $\text{NO}_3^-$  and glucose added to stimulate denitrification. Such studies do most likely not reflect denitrification rates under field conditions. These methodological constraints are further aggravated by the fact that the historically most widely applied method to quantify total denitrification in the field (i.e. the Acetylene Inhibition Technique (AIT)) has been shown to create a systematic but variable, and therefore irreproducible underestimation of denitrification [55–58], and results in a substantial overestimation of  $R_{N_2O}$  [12\*\*]. Therefore, to determine  $R_{N_2O}$  we summarized available datasets where  $N_2$  emissions were measured by  $^{15}\text{N}$ -labelling, the natural N and oxygen isotope approach or with the gas-flow helium incubation method and provide estimates for  $N_2$  as well as  $N_2O$  emissions under field conditions (see supplementary material). Mean  $R_{N_2O}$  for agricultural soils and soils under natural vegetation were  $0.11$  ( $n = 34$ ) and  $0.12$  ( $n = 33$ ), respectively, while the ratio for wetland soils was significantly lower ( $0.02$ ,  $n = 10$ ) (Table 3). These ratios are significantly lower than other studies that estimated  $R_{N_2O}$  using the AIT. For example, Schlesinger [44\*\*] estimated that the mean  $R_{N_2O}$  of denitrification from cropped soils was approximately 0.37, whereas Wang *et al.* [59] came up with a ratio of 0.33. The mean ratios from our summary, however, are in good agreement with the overall  $R_{N_2O}$  for total terrestrial denitrification obtained by our mass balance approach (Figure 1). This corroborates the results of a previous study from Butterbach-Bahl *et al.* [12\*\*] who showed that  $R_{N_2O}$  for soils from different ecosystems are significantly lower if measurements that used AIT are ignored.

To provide an estimate of terrestrial denitrification from soils under natural vegetation, agricultural soils

Figure 1



Global terrestrial N budget for 1860 and in 2005 ( $Tg\ N\ year^{-1}$ ); values in brackets show the 95% confidence interval. Inputs for natural BNF from Refs. [39,47,49]; BNF in agricultural systems from Refs. [50]; Haber-Bosch fertilizer from Refs. [28\*,47,48]; fossil fuel combustion from Refs. [35,51]; lighting from Ref. [35]. Outputs for N transported to the ocean via river flow or atmospheric deposition from Refs. [52–54];  $N_2O$  emitted to the atmosphere (this study, Table 1); storage of N in terrestrial or inland aquatic ecosystems [11\*\*,46].

\*Note: The terrestrial N budget was assumed to be in equilibrium in pre-industrial times, with no net accumulation of N in terrestrial biomass, soils or groundwater. Consequently, there are no storage changes given for the pre-industrial N budget.

and wetlands we used  $N_2O$  emissions estimates from Bouwman *et al.* [11\*\*] for natural and agricultural soils, and wetlands and flooded soils and divided those by the mean  $R_{N_2O}$  (Table 3). The calculated denitrification for agricultural soils is  $656\ Tg\ N\ year^{-1}$  (38–(56–78)),

for soils under natural vegetation  $45\ Tg\ N\ year^{-1}$  (42–37–57) and wetlands  $56\ Tg\ N\ year^{-1}$  (39–40–96). The resulting estimate of total terrestrial denitrification,  $166\ Tg\ N\ year^{-1}$  (131–133 – 231), is thus consistent with the estimate using the mass balance approach

**Table 3****Mean  $R_{N_2O}$ ,  $N_2O$  emissions and calculated rates of  $N_2$  emissions and total denitrification for different terrestrial ecosystems**

| Ecosystem                             | $R_{N_2O}$<br>$N_2O/(N_2+N_2O)$ | $N_2O$ emissions <sup>a</sup><br>(Tg N <sub>2</sub> O-N yr <sup>-1</sup> ) | $N_2$ emissions<br>(Tg N yr <sup>-1</sup> ) | Total denitrification<br>(Tg N yr <sup>-1</sup> ) |
|---------------------------------------|---------------------------------|--|---|---|
| Agricultural soils                    | $0.109 \pm 0.020$               | 6.4  | 59 (50–72)                                  | 65 (56–78)  |
| Soils under natural vegetation        | $0.124 \pm 0.031$               | 4.9  | 40 (32–52)                                  | 45 (37–57)  |
| Freshwater wetlands and flooded soils | $0.020 \pm 0.009$               | 1.1 <sup>b</sup>   | 55 (39–95)                                  | 56 (40–96)  |
| Total terrestrial                     | 0.08 <sup>c</sup>               | 12.4 <sup>d</sup>  | 153 (120–219)                               | 166 (133–231)                                     |

<sup>a</sup> Estimates are taken from Bouwman et al. [11\*\*].<sup>b</sup> Riparian zones and groundwater in Bouwman et al. [11\*\*].<sup>c</sup> Calculated from the sum of estimated  $N_2$  and  $N_2O$  emissions.<sup>d</sup> Sum from Bouwman et al. [11\*\*]. Differs slightly from the value used in Figure 1.

(160 Tg-N year<sup>-1</sup>). However, there are still a number of caveats that should be addressed in future research to allow the presented approach to become more widely applicable:

#### 1 Limited database on non-AIT estimates of $R_{N_2O}$

The magnitude of total denitrification losses ( $N_2 + N_2O$ ) and  $R_{N_2O}$  is still unknown for the vast majority of terrestrial ecosystems. In total, we found 34 studies that reported estimates for  $N_2$  as well as  $N_2O$  emissions under field conditions, 13 for soils under natural vegetation, 14 for fertilized agricultural soils and 8 for wetland or flooded soils. The majority of the studies comes from temperate climate zones, while only 4 studies were carried out in a tropical or subtropical environment. More research on denitrification and  $R_{N_2O}$  for a wide range of ecosystems is urgently required for more robust estimates of terrestrial denitrification.

#### 2 High uncertainty in environmental controls of $R_{N_2O}$

Dynamic, short-term changes in  $R_{N_2O}$  values as driven by changes in soil environmental parameters have been reported in various studies and  $R_{N_2O}$  has been shown to be controlled by soil moisture and temperature [60–62], soil oxygen ( $O_2$ ) availability [63,64], soil texture and pH [43,65,66], or soil nitrate ( $NO_3^-$ ) and carbon (C) concentrations [67,68]. Thus, using a mean  $R_{N_2O}$  for estimating  $N_2$  fluxes from terrestrial ecosystems results in major uncertainties and a recent study showed that parametrization of  $R_{N_2O}$  with environmental controls can improve estimates of denitrification from agricultural ecosystems [69].

#### 3 More accurate attribution of $N_2O$ emissions to different microbial processes

We did our calculations based on the assumption that  $N_2O$  emissions are produced entirely through denitrification, while it is well-known that soil  $N_2O$  emissions are produced via a number of complex processes with several microbial sources involved, of which denitrification may or may not be the most significant [70,71]. Estimates of  $N_2O$  emissions (from field to global scale) are generally not denitrification specific, and therefore the current study is likely to overestimate denitrification derived  $N_2O$  emissions and the  $R_{N_2O}$ . To date it is mainly the  $^{15}N$ -gas flux approach that can reliably be used for measuring denitrification derived  $N_2O$  since the He/ $O_2$  gas-flow incubation method (without  $^{15}N$  tracer used) cannot discriminate the source of  $N_2O$  [72]. Therefore, more field measurements of denitrification, in combination with a more accurate apportionment of  $N_2O$  sources are needed to derive accurate estimates of  $R_{N_2O}$  from denitrification.

#### Conclusion

The amount of  $N_f$  applied to land has increased by 160% over the last 200 years, causing severe environmental disturbances. We used two methods to derive global estimates of  $N_f$  removal by terrestrial denitrification: (i) a mass balance approach with recent estimates of terrestrial N inputs minus N exports via rivers and wet/dry N deposition to oceans (Figure 1), and (ii) a literature review on measured  $R_{N_2O}$  in combination with estimates of  $N_2O$  emissions from soils. Both approaches indicate that at the global scale  $N_2O$  comprises approximately 8% (6–11%) of the terrestrial denitrification flux, which is much lower than previous estimates that also considered studies of denitrification and  $N_2$  formation using the  $C_2H_2$  inhibition technique (AIT) and estimated  $R_{N_2O}$  in the range of 30–40% for terrestrial soils [44\*\*,59]. This global mean  $R_{N_2O}$  of 8% is much more in line with recent estimates that 3–5% of newly fixed N is converted to  $N_2O$  and 50% removed by denitrification in terrestrial ecosystems [7\*,26,30,73], while ratios above 30% are hard to reconcile with these estimates. Our analysis demonstrates that

using measured  $R_{N_2O}$  can provide estimates of terrestrial denitrification that are in good agreement with a global mass balance approach when data from AIT studies are excluded. An advantage of the  $R_{N_2O}$  approach is that it can be used to estimate denitrification rates from specific regions or ecosystems, both with and without fertilizer applications, given that specific data on  $N_2O$  emissions and the product ratio of denitrification is available. This will provide more reliable estimates of denitrification than other methods, and could provide the targeted data required for improving our understanding of this crucial  $N_r$  sink. However, the limited database on non-AIT estimates of  $R_{N_2O}$  still causes high uncertainty, which calls for more research on denitrification and  $R_{N_2O}$  for a wide range of ecosystems. Recent advances in isotopic approaches and analytical methods could pave the way for a significantly improved understanding of N cycling and denitrification in terrestrial agro-ecosystems [32,74–79].

We estimate that terrestrial denitrification has doubled since pre-industrial times and is now in the range of 115–202 Tg-N year<sup>-1</sup>, removing 56% of the newly fixed terrestrial N each year, less than the removal rate of 72% in pre-industrial times. This implies that we are in a phase of accelerated accumulation of  $N_r$  in the biosphere with potentially serious environmental consequences [80]. However, much remains to be learned about how denitrification is affected by increasing levels of  $N_r$  in the environment. Such knowledge is required if we want to understand the fate of  $N_r$  and identify strategies for reducing  $N_r$  creation and its unwanted impact. Our analysis demonstrates that wetlands and flooded soils are important hotspots of denitrification. Despite an area of only 8% of the global land mass, our calculations indicate that approximately one third of the terrestrial denitrification (56 Tg-N year<sup>-1</sup>) occurs in wetlands and flooded soils with a much lower  $R_{N_2O}$ . This highlights that the preservation and restoration of wetlands, or the introduction of constructed wetlands in landscapes, can be a key strategy for removing excess  $N_r$ . In addition to the carbon sequestration potential (0.2–0.8 Gt CO<sub>2</sub>e year<sup>-1</sup>) [81], the restoration of peatlands and wetlands would also help to remove  $N_r$  pollution from agricultural runoff. The needed intensification of crop production for feeding a growing and hungry planet will require a significantly improved N use efficiency in cropping systems and/or even higher inputs of  $N_r$  to terrestrial cropping systems and denitrification will play a key role in maximising the benefits of  $N_r$  while minimizing the environmental impact.

## Conflict of interest statement

Nothing declared.

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