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A. van den Pol-van Dasselaar 7 **M.L. van Beusichem** 7 **O. Oenema**

Effects of nitrogen input and grazing on methane fluxes of extensively and intensively managed grasslands in the Netherlands

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Abstract Generally, grasslands are considered as sinks for atmospheric $CH₄$, and N input as a factor which reduces $CH₄$ uptake by soils. We aimed to assess the short- and long-term effects of a wide range of N inputs, and of grazing versus mowing, on net $CH₄$ emissions of grasslands in the Netherlands. These grasslands are mostly intensively managed with a total N input via fertilisation and atmospheric deposition in the range of 300–500 kg N ha⁻¹ year⁻¹. Net CH₄ emissions were measured with vented, closed flux chambers at four contrasting sites, which were chosen to represent a range of N inputs. There were no significant effects of grazing versus mowing, stocking density, and withholding N fertilisation for $3-9$ years, on net CH₄ emissions. When the ground-water level was close to the soil surface, the injection of cattle slurry resulted in a significant net CH_4 production. The highest atmospheric CH_4 uptake was found at the site with the lowest N input and the lowest ground-water level, with an annual $CH₄$ uptake of 1.1 kg CH_4 ha⁻¹ year⁻¹. This is assumed to be the upper limit of CH_4 uptake by grasslands in the Netherlands. We conclude that grasslands in the Netherlands are a net sink of $CH₄$, with an estimated $CH₄$ uptake of 0.5 Gg CH₄ year⁻¹. At the current rates of total N input, the overall effect of N fertilisation on net $CH₄$ emissions from grasslands is thought to be small or negligible.

Key words Grassland · Methane · Mowing versus $grazing \cdot Nitrogen$ fertilisation \cdot Nitrogen input

Tel.: $+31-317-482339$, Fax: $+31-317-483766$

Introduction

The atmospheric $CH₄$ concentration is increasing, which is of concern as it may contribute to global warming. $CH₄$ is the most important greenhouse gas next to $CO₂$. It has been reported to contribute about 20% to the enhanced greenhouse effect (IPCC 1995). $CH₄$ can be formed in soils whenever organic matter is degraded by micro organisms under anaerobic conditions. Under aerobic conditions, the soil may act as a sink where methanotrophic micro organisms oxidise either atmospheric CH_4 , or CH_4 that has been produced in anaerobic parts of the soil. On a global scale, soils contribute 3–9% to the total sink strength for atmospheric CH₄ (IPCC 1995).

Grasslands are generally considered as sinks for CH4. The net uptake rate depends on environmental conditions, like the ground-water level, soil moisture content, temperature, and grassland management (Czepiel et al. 1995; Dunfield et al. 1995; Van den Pol-van Dasselaar et al. 1998). The C and N dynamics of grazed grasslands differ from those of mown grasslands, but whether this difference causes a difference in $CH₄$ flux has not yet been demonstrated. Several authors have observed that N fertilisation decreases $CH₄$ uptake by soil. This is caused either by an immediate inhibition of methanotrophy (short-term effect) or by a change in the composition and size of the microbial community due to repeated fertiliser N application (long-term effect). The decrease in the $CH₄$ uptake of a soil may be associated with (1) NH₄⁺ (Hütsch et al. 1994; King and Schnell 1994; Dunfield and Knowles 1995; Willison et al. 1995), (2) $NO₂⁻$ (King and Schnell 1994; Dunfield and Knowles 1995), and (3) high N turnover rates (i.e. mineralisation and nitrification; Mosier et al. 1991; Hütsch et al. 1993). Our understanding of the mechanisms of decreasing $CH₄$ consumption capacity due to N fertilisation is limited, because our knowledge of the underlying microbiology is poor (Roslev et al. 1997).

Most grasslands in the Netherlands are intensively managed with a total N input via fertilisation and atmo-

A. van den Pol-van Dasselaar (\boxtimes) · M.L. van Beusichem O. Oenema

Department of Environmental Sciences, Sub-department Soil Science and Plant Nutrition, Wageningen Agricultural University, P.O. Box 8005, 6700 EC Wageningen, The Netherlands

spheric deposition in the range of 300–500 kg N ha⁻¹ year–1. High N inputs commenced after 1950 following a rapid intensification of livestock farming and industry. Unfertilised and extensively managed grasslands have a total N input of about 30–50 kg N ha⁻¹ year⁻¹ via atmospheric deposition of NH_v and NO_x (Erisman and Draaijers 1995).

We aimed to assess the short- and long-term effects of a wide range of N inputs, and of grazing versus mowing, on CH_4 uptake by grasslands in the Netherlands. We investigated the effects of withholding N fertilisation, application of different types of mineral N fertilisers, and application of cattle slurry. Net $CH₄$ emissions were measured at four contrasting sites, which were chosen to represent a range of N inputs. We hypothesised that: (1) N fertilisation reduces net $CH₄$ uptake in the short-, and (2) the long-term, causing differences in net $CH₄$ uptake between sites with different N inputs, (3) that withholding N fertilisation increases the net $CH₄$ uptake, and (4) that grazing decreases the net $CH₄$ uptake compared to mowing, as the annual amount of C and N applied to the soil is larger under grazing than under mowing (Van den Pol-van Dasselaar and Lantinga 1995).

Materials and methods

Site description

Field experiments were carried out at four sites in the Netherlands (characteristics of these sites are shown in Table 1):

1. Wolfheze. The site is a heather grassland on dry sandy soil, situated in a nature reserve. The vegetation is dominated by grasses, heather and mosses. The ground water is at least 3 m below the surface. The site has not been fertilised for several centuries. In summer, a few cattle graze the area to maintain the grassland. The N input via atmospheric deposition is estimated to be 40 kg N ha⁻¹ year⁻¹ (Erisman and Draaijers 1995).

2. Bovenbuurtse Weilanden. This site is a poorly drained grassland on sandy soil, and the vegetation is dominated by grasses. The N input via atmospheric deposition is estimated to be 45 kg N ha⁻¹ year⁻¹ (Erisman and Draaijers 1995). Between 1950 and 1972, the site received about 200 kg N ha^{-1} year⁻¹ via fertilisation. Different treatments were established in 1972: grazing with a low stocking density of about 3 animals ha⁻¹, and no mineral N application (G_{low}); grazing with a high stocking density of about 4.5 animals ha⁻¹, and no mineral N application (G_{high}); mowing twice a year, with a N application of $\overline{50}$ kg N ha⁻¹ year⁻¹ as calcium ammonium nitrate in the period 1972–1985, and no N application since 1986 (M–); mowing twice a year, with a N application of 50 kg N ha⁻¹ year⁻¹ as calcium ammonium nitrate $(M+)$.

3. Wildekamp. This site is a poorly drained grassland on sandy soil with a total N input via fertilisation and atmospheric deposition of about 400–500 kg N ha⁻¹ year⁻¹. The vegetation is dominated by grasses. Three treatments were used: fertilisation with $Ca(NO₃)₂$ at a rate of 80 kg N ha⁻¹ cut⁻¹ (CaNi); fertilisation with $(NH_4)_2SO_4$ at a rate of 80 kg N ha⁻¹ cut⁻¹ (AmSu); fertilisation with cattle slurry, injected with a sod-injector to a depth of 5 cm, at a rate of $15 \text{ m}^3 \text{ ha}^{-1} \text{ cut}^{-1}$, which was equal to a mineral N application rate of about 45 kg N ha⁻¹ cut⁻¹ (Slur). In the Netherlands, animal manure must be injected into the soil in order to reduce NH₃ volatilisation.

4. Zegveld. This site is a grassland on an intensively managed and moderately drained peat soil with both high and low N inputs. The vegetation is dominated by grasses. There were two sites: one with a relatively low mean ground-water level of 42 cm below the soil surface in the experimental year 1994 (Z_{low}); and a second one with a relatively high mean ground-water level of 22 cm below the surface in 1994 ($\rm Z_{high}$). On both these sites, there were three treatments: mowing, and withholding of N fertilisation since 1992 (before 1992 about 400 kg fertiliser \bar{N} ha⁻¹ year⁻¹ was applied as calcium ammonium nitrate; M–); mowing, with N application $(M+)$; grazing, with N application $(G+)$. In 1994, cumulative N application rates for both M + and G + were 378 kg N ha⁻¹ year⁻¹ for site Z_{high} . Fertiliser N was applied as calcium ammonium nitrate. The N input via atmospheric deposition was estimated to be 35 kg N ha–1 $year⁻¹$ (Erisman and Draaijers 1995).

Monitoring net CH4 emissions in the field

Net $CH₄$ emissions were measured with vented, closed flux chambers (Hutchinson and Mosier 1981; Mosier 1989). At Wolfheze, net CH₄ emissions were measured weekly to monthly from March 1996 to March 1997 with 12–24 flux chambers. At Bovenbuurtse Weilanden, net CH_4 emissions were measured 3–4 times in the period 6 May 1994 to 5 July 1994, with four to six flux chambers

Table 1 Characteristics of the Wolfheze, Bovenbuurtse Weilanden, Wildekamp and Zegveld sites. *LOI* loss-on-ignition, *n.d.* Not determined

per treatment, and 3–5 times in the period 20 January 1995 to 28 February 1995 with six to 12 flux chambers per treatment. At Wildekamp, net CH₄ emissions were measured during 29 days following fertiliser application on 30 May 1994, with four flux chambers per treatment. At Zegveld, net CH₄ emissions were measured weekly to biweekly from December 1993 to January 1995 with six to 12 flux chambers per site (two to four flux chambers per treatment). At the start of the measurements, circular, stainless steel flux chambers (internal diameter 20 cm, height 16 cm) were carefully inserted into the soil to a depth of 2–4 cm at all sites, except for treatment Slur at Wildekamp. For this treatment, rectangular flux chambers were used (length 80 cm, width 20 cm, height 16 cm) to cover a representative fraction of grassland with and without injected slurry per flux chamber. Flux chambers were closed by a stainless steel lid and covered with insulating sheets to prevent temperature changes within the chambers. At regular time intervals (20–40 min), four gas samples were taken from the headspace of the chambers with glass syringes. At each measurement date, soil temperatures were recorded at 0, 10, 20, 30, 40, and 50 cm depth. The soil moisture content of the upper 10 cm was determined gravimetrically after drying at 105° C for 24 h.

Incubation experiments

The effect of N fertilisation and grazing versus mowing on CH4 consumption capacity was studied in an incubation experiment with field-moist soil samples from different soil depths of Bovenbuurtse Weilanden. About 25 g (dry weight) homogenised soil of the layers 0–5, 5–10, 10–20, and 20–30 cm depth was incubated aerobically in bottles of 580 ml at 20 °C with an initial CH₄ concentration of about 80 μ l l⁻¹. There were three replicates per soil layer. Control bottles containing no soil were included. Gas samples from the headspace of the bottles were taken through rubber septa. The incubation lasted 1 week during which four gas samples were taken.

Analytical procedures and data acquisition

Gas samples from the field experiments were analysed for $CH₄$ within 24 h by gas chromatography (PU 4400, Unicam) using a flame ionisation detector (coefficient of variation: 0.08%). A standard CH₄ concentration of 2.0 μ l l⁻¹ (\pm 5%) was used for calibration. Gas samples of the incubation experiments were analysed for CH₄ immediately after sampling.

In the incubation experiments, the decrease in $CH₄$ concentration in the headspace of the incubation bottles showed typical first-order kinetics (1):

$$
\ln Y_t = \ln Y_0 - kt,\tag{1}
$$

in which $Y = CH_4$ concentration in the headspace (μ l l⁻¹), *t* = time (days), $k =$ the rate constant (day⁻¹). The rate constant k was normalised to dry weight (day⁻¹ g^{-1} dry soil) and used to characterise the $CH₄$ consumption capacity of the soil.

Net CH₄ emissions were calculated for the field experiments from linear regression of the time course of $CH₄$ concentrations in the headspace of the flux chambers. Annual mean net CH_4 emissions were estimated by trapezoidal integration of mean net CH4 emissions over time. Statistical differences between treatments were tested by ANOVA with a factor treatment $(P=0.05)$.

Results

Net CH₄ emissions

Net CH₄ emissions from grasslands at Wolfheze and Zegveld throughout the year are shown in Table 2. On average, Wolfheze was a net sink for $CH₄$, with an annual mean uptake of 1.1 kg CH_4 ha⁻¹ year⁻¹. At Zegveld, differences in $CH₄$ emissions between sites were significant $(P=0.05)$. The site with a relatively low ground-water level (Z_{low}) consumed, on average, 0.3 kg $CH₄$ ha⁻¹ year⁻¹, while the site with a relatively high ground-water level (Z_{high}) consumed, on average, $0.1 \text{ kg } CH_4 \text{ ha}^{-1} \text{ year}^{-1}$. There were no significant differences in net $CH₄$ emissions between the treatments. The temporal and spatial variability of net $CH₄$ emissions at Zegveld and Wolfheze are discussed by Van den Pol-van Dasselaar et al. (1997) and Van den Polvan Dasselaar et al. (1998), respectively.

Net CH₄ emissions at Bovenbuurtse Weilanden are shown in Table 2. On average, treatment $M +$ was a small sink, while the other treatments were small sources of $CH₄$. The soil moisture content was, on aver-

Table 2 Mean net CH₄ emissions from grasslands at Wolfheze, Zegveld and Bovenbuurtse Weilanden throughout the year. *Negative numbers* indicate net consumption of atmospheric CH4, *positive numbers* indicate net CH4 production. *Zlow* Zegveld, rela-

tively low ground-water level; *Zhigh* Zegveld, relatively high ground-water level; *B* Bovenbuurtse Weilanden; *M* mowing; *G* grazing; G_{low} low stocking density, G_{high} high stocking density; $$ no mineral N application; $+$ mineral N application

age, 37% (w/w) in the mowing treatments and 43% in the grazing treatments, both in summer and winter. The soil temperature of the upper 5-cm layer was, on average, $18\textdegree C$ in summer and $7.5\textdegree C$ in winter. The spatial variability of net CH_4 emissions was high, especially in the grazing treatments. Often, one or two high net CH4 production values and several low net $CH₄$ consumption values were found. There were no significant differences in net CH_4 emissions between different treatments, or between the two measurement periods.

Effect of type of fertiliser application

Type of fertilisation, i.e. mineral fertiliser or cattle slurry, markedly influenced net $CH₄$ emissions from the soil to the atmosphere at Wildekamp (Fig. 1). Mean net CH₄ emissions (\pm SE) were 0.02 \pm 0.06 mg CH₄ m⁻² day⁻¹ for CaNi, -0.09 ± 0.05 for AmSu and 7.76 \pm 2.29 for Slur. There were no significant differences between the two mineral fertiliser treatments. However, net $CH₄$ emissions were significantly different for the Slur treatment compared to the mineral fertiliser treatments, CaNi and AmSu $(P=0.05)$. Treatment Slur showed net

Fig. 1 Mean net CH₄ emissions (mg CH₄ m⁻² day⁻¹; \pm SE) after N application via $(NH_4)_2SO_4$ (*AmSu*), $Ca(NO_3)_2$ (*CaNi*) and cattle slurry injected into the soil (*Slur*), and ground-water level (*GWL*; cm below the soil surface) at Wildekamp. Each data point is the average of four measurements. *d* Day

CH4 production during the experimental period. The time course of net CH_4 production followed the time course of changes in the ground-water level, with a delay of 3–4 days (Fig. 1). During the first 2 weeks of the experimental period, the soil temperature at 5 cm depth was about 15° C. In the last 2 weeks, it gradually increased to 23 °C. The net uptake of CH_4 for all treatments at the end of the experimental period coincided with a low ground-water level and high soil temperatures.

Effect of withholding N fertilisation, and grazing versus mowing

Within sites, the effect of withholding N fertilisation for 3 years (comparison of treatments M- and $M+$ at Zegveld) or 9 years (comparison of treatments M – and M + at Bovenbuurtse Weilanden) on net CH_4 emissions was not significant ($P=0.05$; Table 2). Furthermore, the incubation experiments with soil samples from Bovenbuurtse Weilanden showed no differences in $CH₄$ consumption capacities of soil samples from treatments M– and $M + (P=0.05)$. The average profile of CH₄ consumption capacity of all treatments at Bovenbuurtse Weilanden showed a subsurface maximum at 10–20 cm depth (Fig. 2).

At Bovenbuurtse Weilanden, the effect of grazing versus mowing (comparison of M and G) and stocking density (comparison of G_{low} and G_{high}) on net CH₄ emissions was not significant $(P=0.05;$ Table 2). Furthermore, at Zegveld the effect of grazing versus mowing (comparison of $M+$ and $G+$) on net CH₄ emissions was not significant $(P=0.05;$ Table 2).

Discussion

Net CH₄ emissions

Temperate grassland soils, especially when they are well-drained and thus mainly aerobic, are generally a sink for atmospheric CH4, with an average uptake of 0–1 mg CH₄ m⁻² day⁻¹ (Mosier et al. 1991; Jarvis et al. 1994; Kruse and Iversen 1995; Dobbie et al. 1996). Our results are within this range (Table 2). However, several sites sometimes acted as a source instead of a sink. This may have been due to relatively high ground-water levels and soil moisture contents, which would have created anaerobic microsites near the soil surface. These anaerobic microsites may have produced $CH₄$, thus decreasing the sink function of the soil.

The incubation experiments with soil samples from Bovenbuurtse Weilanden showed a subsurface maximum CH4 consumption capacity at 10–20 cm depth (Fig. 2). This phenomenon has been found before (e.g. Koschorreck and Conrad 1993; Dunfield et al. 1995; Schnell and King 1996; Van den Pol-van Dasselaar et al. 1997). It might be caused by outcompetition of me-

Fig. 2 Mean normalised rate constants of $CH₄$ consumption capacity (day⁻¹ g⁻¹ dry soil; \pm SE) of soil samples from different layers (cm below the surface) at Bovenbuurtse Weilanden, determined from aerobic incubations at 20 °C

thanotrophic bacteria by other, better adapted organisms in the top-soil (Koschorreck and Conrad 1993). Alternatively, methanotrophs may be sensitive to moisture changes, and so may be unable to survive in the top-soil (Schnell and King 1996). The latter hypothesis is supported by results from Nesbit and Breitenbeck (1992), who showed that the methanotrophic activity of completely air-dried soils did not recover after rewetting.

Effect of N fertiliser type

At Wildekamp, we tested the effect of N fertiliser type on net CH_4 emissions. Net CH_4 emissions after N application via $(NH_4)_2SO_4$ were not significantly different from net CH_4 emissions after N application via $Ca(NO₃)₂$ (P=0.05), even though NH₄⁺ is often associated with a decrease in $CH₄$ uptake (Hütsch et al. 1994; King and Schnell 1994; Dunfield and Knowles 1995; Willison et al. 1995). This may have been due to conditions being favourable for $CH₄$ production during the first weeks of the experiment. Net $CH₄$ emissions from the Slur treatment were significantly higher than from the mineral fertiliser treatments (Fig. 1). The net CH4 production from the Slur treatment was probably caused by a combination of wet soil, application of easily decomposable organic material and anaerobic conditions in the slurry itself. When the ground-water level dropped at the end of the measurement period, the Slur treatment became a small net sink for $CH₄$. It has been shown that farmyard manure may stimulate the CH4 consumption capacity of a soil by increasing the microbial biomass (Willison et al. 1996). Hütsch et al. (1993) showed that application of farmyard manure did not have a significant long-term effect on the $CH₄$ consumption capacity of soils. It is probable that the application of organic manure does not affect the long-term CH4 consumption capacity of soil.

Effect of N input

N input, especially in the form of NH_4^+ , and high N turnover rates may decrease $CH₄$ uptake (e.g. Mosier et al. 1991; Hütsch et al. 1994; Willison et al. 1995), either by an immediate inhibition of methanotrophy (short-term effect) or by a change in the composition and size of the microbial community due to repeated fertiliser N applications (long-term effect).

We did not find any significant short-term effects of N fertilisation on net CH_4 emissions. At Zegveld, there was no significant effect of withholding N fertilisation for 3 years. At Bovenbuurtse Weilanden, we also found no significant effect of withholding N fertilisation, even though the period without N fertilisation of the M– treatment was 9 years and before that period only 50 kg N ha⁻¹ year⁻¹ was applied. However, combined data from Wolfheze and Zegveld, the two sites with yearround measurements, suggested that there might be a significant long-term effect of N inputs causing differences in annual mean net CH_4 emissions (Fig. 3). This could, however, also have been partly due to differences in moisture content, since Wolfheze, the site with the highest CH_4 uptake, did not only have the lowest N input, but also the lowest ground-water level. Groundwater level and soil moisture content were found to be important determining factors of temporal variability of CH4 emissions from Zegveld and Wolfheze (Van den Pol-van Dasselaar et al. 1997; Van den Pol-van Dasselaar et al. 1998). Whatever the precise cause, our data indicate that the overall effect of N fertilisation on net $CH₄$ emissions from grasslands is small or negligible at the current rates of N input in the Netherlands.

Fig. 3 Relationship between estimated mean annual N input (kg N ha⁻¹ year⁻¹) and mean annual net CH₄ emissions (kg CH₄ ha⁻¹ year⁻¹; \pm SE). *Zlow* Zegveld site with a relatively low mean ground-water level; *Zhigh* Zegveld site with a relatively high ground-water level; *M* mowing; *G* grazing; *–* no mineral N application; $+$ mineral N application; *yr* year

Effect of grazing versus that of mowing

Grazing versus mowing may affect $CH₄$ uptake through differences in the annual supply of C and N to the soil (Van den Pol-van Dasselaar and Lantinga 1995). At Zegveld, the effect of grazing versus mowing on net CH₄ emissions was not significant ($P=0.05$). This may have been due to the relatively short period of 3 years in which different treatments were applied. However, also at Bovenbuurtse Weilanden, the effect of grazing versus mowing and stocking density on net $CH₄$ emissions was not significant $(P=0.05)$, despite the fact that these treatments had been established more than 20 years before. Spatial variability was often higher in the grazing treatments than in the mowing treatments (Table 2), probably as a result of cattle excreta creating a heterogeneous pattern of methanogenesis and methanotrophy in the soil. On spots with fresh dung, conditions, i.e. high organic matter content and low O_2 content, are favourable for methanogenesis. Methanogenesis causes relatively high $CH₄$ concentrations in the soil, which may stimulate methanotrophs. Increased methanotrophy may still continue when methanogenesis has decreased. Clearly, these factors contribute to a large spatial variability of $CH₄$ emissions from grazed grasslands. However, our data indicate that the overall effect of grazing on net CH_4 emissions from grasslands is negligible. It has to be emphasised that $CH₄$ produced by cattle was not taken into account in these estimates.

Net $CH₄$ flux of grasslands in the Netherlands

About 30% of the total surface area in the Netherlands is occupied by intensively managed grasslands, amounting to 1 050 000 ha. Recent estimates of national $CH₄$ emissions (Van den Born et al. 1991; Van Amstel et al. 1993) did not consider grasslands as a sink for $CH₄$. In contrast, drained organic soils, which are mainly used as grasslands, have been estimated to be a net $CH₄$ source, representing $32-89$ Gg CH₄ year⁻¹ (35-700 kg $CH₄$ ha⁻¹ year⁻¹; Van Amstel et al. 1993). Van Amstel et al. (1993) based their estimates on research on organic soils in other countries (Aselmann and Crutzen 1989; Moore and Knowles 1989). However, our yearround measurements at Zegveld showed that drained organic soils in the Netherlands are a net sink for $CH₄$ with an annual mean net CH_4 uptake of 0.1–0.3 kg CH_4 ha–1 year–1 (see also Van den Pol-van Dasselaar et al. 1997). We consider 0.1 kg CH₄ ha⁻¹ year⁻¹, which was the mean annual CH₄ uptake at site Z_{high} , the site with the relatively high ground-water level, as the lower limit of CH4 uptake by intensively managed grasslands in the Netherlands. We based this assumption on the fact that the conditions of this site are least favourable for $CH₄$ uptake for the following reasons: (1) it has a peat soil, which has the potential of emitting $CH₄$ as it has a high organic matter content and is largely anaerobic

due to the high ground-water level; (2) it is intensively managed with high N fertilisation and it has a high N turnover rate. The net uptake of 1.1 kg CH_4 ha⁻¹ year⁻¹ at Wolfheze may be the upper limit of $CH₄$ uptake by grasslands in the Netherlands. We based this assumption on the fact that the conditions at this site are the most favourable for $CH₄$ uptake of all the sites assessed because: (1) the prevailing soil moisture contents were optimal for CH4 uptake (Van den Pol-van Dasselaar et al. 1998), (2) it is extensively managemed with no N fertilisation during past centuries and it has a low N turnover rate. We conclude that grasslands in the Netherlands (excluding wetlands/undrained peatlands that occupy only 0.5% of the total surface area) are a net sink for CH_4 with an estimated CH_4 uptake of 0.5 Gg CH_4 $year⁻¹$. Estimates of CH₄ emissions in the Netherlands need to take into account the role of grasslands in the national $CH₄$ budget.

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