



The nitrogen, carbon and greenhouse gas budget of a grazed, cut and fertilised temperate grassland

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30 Abstract

Intensively managed grazed grasslands in temperate climates are globally important environments for the exchange of the greenhouse gases (GHGs) carbon dioxide (CO₂), nitrous oxide (N₂O) and methane (CH₄). We assessed the N and C budget of a mostly grazed, occasionally cut, and fertilized grassland in SE Scotland by measuring or modelling all relevant imports and exports to the field as well as changes in soil C and N pools over time. The N budget was dominated by import from inorganic and organic fertilisers (21.9 g N m² yr⁻¹) and losses from leaching (5.3 g N m² yr⁻¹), N₂ emissions and NO_x and NH₃ volatilisation (6.4 g N m² yr⁻¹). The efficiency of N use by animal products (meat and wool) averaged 11%. On average over nine years (2002-2010) the balance of N fluxes suggested that 7.2 ± 4.6 g N m⁻² y⁻¹ (mean ± confidence interval at p > 0.95) were stored in the soil. The largest component of the C budget was the net ecosystem exchange of CO₂ (NEE), at an average uptake rate of 218 ± 155 g C m⁻² y⁻¹ over the nine years. This sink strength was offset by carbon export from the field mainly as harvest (48.9 g C m² yr⁻¹) and leaching (16.4 g C m² yr⁻¹). The other export terms, CH₄ emissions from the soil, manure applications and enteric fermentation were negligible and only contributed to 0.02-4.2 % of the total C losses. Only a small fraction of C was incorporated into the body of the grazing animals. Inclusion of these C losses in the budget resulted in a C sink strength of 163 ± 140 g C m⁻² y⁻¹. On the contrary, soil stock measurements taken in May 2004 and May 2011 indicated that the grassland sequestered N in the 0-60 cm soil layer at 4.51 ± 2.64 g N m⁻² y⁻¹ and lost C at a rate of 29.08 ± 38.19 g C m⁻² y⁻¹, respectively. Potential reasons for the discrepancy between these estimates are probably an underestimation of C and N losses, especially from leaching fluxes as well as from animal respiration. The average greenhouse gas (GHG) balance of the grassland was -366 ± 601 g CO₂ eq m⁻² y⁻¹ and strongly affected by CH₄ and N₂O emissions. The GHG sink strength of the NEE was reduced by 54% by CH₄ and N₂O emissions. Enteric fermentation from the ruminating sheep proved to be an important CH₄ source, exceeding the contribution of N₂O to the GHG budget in some years.

55 *Keywords:* grassland, carbon stocks, carbon sequestration, nitrogen cycling, budget, greenhouse gases



Introduction

60 Nitrogen (N) is an essential component of proteins and genetic material and therefore required by all
living organisms. Before N can be used by most organisms, inert atmospheric molecular nitrogen (N_2)
has to be transformed to reactive nitrogen (Nr). In an agricultural system Nr is added from inorganic
fertiliser and cultivation-induced biological N fixation and as organic compounds from organic manure
applications and dung from grazing animals. Generally N inputs into agricultural systems exceed
65 outputs in the form of crops or animal off-takes (meat, milk and wool). In a steady state system the
exceeding Nr is converted back to N_2 through complete denitrification (Galloway et al., 2003).
However, in agricultural systems the surplus of Nr is generally only partly converted to N_2 , while the
rest is lost to the atmosphere or aquatic ecosystems as Nr, causing various environmental problems.

Carbon (C) and N cycles in grasslands are intricately linked and tightly coupled in extensively
managed low N grasslands, with sinks and sources in equilibrium. Converting such systems to
70 intensively managed N fertilised grasslands in the short term may increase the soil organic carbon
(SOC) pool from decomposed plant litter and root material as well as through rhizodeposition (Rees et
al., 2005) until a new equilibrium is reached (Soussana and Lemaire, 2014). In the case of the
Broadbalk experiment, Rothamsted, this equilibrium was achieved after 50 years (Powlson et al,
2011). After the conversion to intensive N management, the tight coupling of the N and C cycles
75 becomes disrupted, leading to emissions of N_2O and CH_4 at rates which may outweigh the benefits of
C sequestration. Several studies indicate that managed grasslands can sequester C (Kim et al., 1992;
Jones et al., 2006; Soussana et al., 2004; Ammann et al., 2007) however, uncertainties are high
(Janssens et al., 2003). On the contrary, Smith (2014) concluded from long-term experiments and
chronosequence studies, that changes in agronomic management may lead to short-term C
80 sequestration, but in the long-term, under constant management and environmental conditions, C
stocks are relatively stable. In a grassland ecosystem the C balance is determined by the net biome
exchange (the difference between total C input and losses). In managed grassland ecosystems exports
through biomass harvesting, the addition of organic manures (from organic fertiliser additions and
animal excretion) as well as CO_2 and CH_4 losses from animal respiration and enteric fermentation can
85 make significant contributions to the C budget.



Worldwide an estimated 26 % of land consists of managed grassland (FAOstat, 2008). The impact of Nr losses, C sequestration and GHG emissions (CO₂, CH₄ and N₂O) from managed grasslands on the environment is therefore of global importance and will become even more relevant in the future as an increased standard of living in developed countries is expected to result in a rapid growth of livestock farming (Caro et al, 2014). Nutrient budgets are a valuable tool to summarise and understand nutrient cycling in agroecosystems and to assess their impact on the environment. As imbalances are not sustainable in the long term, N and C budgets can be used as indicators and regulatory policy instruments for nutrient management in order to reduce losses and increase efficiency. So far, different Nr species have been looked at in separate studies according to their form and impact. Few studies have attempted to calculate N budgets from managed grasslands (e.g. Ammann et al., 2009; Chen et al., 2004; Nunez et al., 2010, Kramberger et al., 2015), whereas C budgets have been assessed more often and are available for various ecosystems (e.g. Aubinet et al., 2000; Soussana et al., 2007; Ammann et al., 2007, Rytter et al. 2015). To calculate the total C and N budget of an ecosystem all import and export processes have to be assessed by measuring or estimating all imports and exports to an ecosystem. The other method is to measure differences in N and C stocks in the soil over time. This approach has the advantage that it requires the measurement of only a single component of the system. However, a large number of samples are needed at an interval of more than 5 years before detectable changes may be statistically significant (Smith, 2004). Moreover this approach does not provide any information about the different processes leading to the final budget.

In this study we assessed the C and N budget from an intensively managed grassland in Southern Scotland using both approaches. Here we report one of the most detailed analyses of C and N fluxes from a grassland ecosystem over 9 years (2002-2010). This study allowed an analysis of the importance of common grassland management practices such as cutting for silage, grazing of cattle and sheep at different stocking densities, N input by inorganic and organic fertiliser applications, as well as different weather conditions on the N, C and GHG balance. The data were collected in the frame of the three European projects GREENGRASS (EC EVK²-CT2001-00105), CarboEurope (GOCE-CT2003-505572) and NitroEurope (contract 017841).



115 2. Methods

2.1 Site description

The experimental site, Easter Bush, is located in South East Scotland, 10 km South of Edinburgh (03°02'W, 55°52' N, 190 m a.s.l). Mean annual rainfall (2002-2010) was $947 \pm$ something mm and the mean annual temperature was $9.0 \pm$ something °C. The field has been under permanent grassland management for more than 20 years with a species composition of >99% rye grass (*Lolium Perenne*) and < 0.5% clover (*Trifolium repens*). The soil type is an imperfectly drained Macmerry soil series, Rowanhill soil association (Eutric Cambisol) with a pH of 5.1 (in H₂O) and a clay fraction of 20-26%. The ground water table was assumed to be at 0.85 m depth on average and the main rooting zone extends down to 0.31 m below soil surface.

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2.2 Grassland management

The grassland was grazed continuously throughout the experimental period by heifers in calf, ewes and lambs at different stocking density (Table 1 and Figure 1a). Animals were counted several times per week and it was assumed that the animal number stayed constant between observations. Livestock units used for heifers, ewes and lambs were 0.75, 0.10 and 0.04, respectively (1 livestock unit has a standard live weight of 600 kg head⁻¹ (Scottish Agricultural College, 1995). Lambs were present on the field from April to September only. The grass was cut for silage on the 1st of June and 8th of August 2002 and on the 29th of May 2003. Ammonium nitrate fertiliser was applied to the field 3-4 times per year, usually between March and July ($56 \text{ kg N ha}^{-1} \text{ application}^{-1}$ on average). In 2008 an additional fifth mineral N application was applied, using urea instead of ammonium nitrate fertiliser. Organic manure was applied on the 28th of September 2004 and 27th of March 2005 as cattle slurry, using a vacuum slurry spreader. Rates of N and C input from fertiliser and manure and export from harvest are shown in Table 4 and 5 and in Fig. 1 a) and b).

140 2.3. Annual budget calculations



We assessed the N and C budget by measuring or estimating the import and export of all relevant fluxes to and from the grassland field on an annual basis. Throughout the manuscript all fluxes are presented following the sign convention used in micrometeorology; fluxes from the ecosystem to the atmosphere are positive (exported from the field), while negative values indicate fluxes from the atmosphere to the ecosystem (imported to the field). We set the system boundary for inputs and exports of N and C by the field perimeters (covering an area of 5.4 ha). The balance of all imports and exports results in the observed changes of N and C on this field over time.

The change in the N balance (ΔN) over time (Δt) of our grassland ecosystem can be written as:

$$\begin{aligned} \Delta N/\Delta t = & FN_{\text{org fert.}} + FN_{\text{synt fert.}} + FN_{N_2 \text{ (biol. fixation)}} + FN_{\text{dep.}} + \\ & FN_{\text{harvest}} + FN_{\text{animal}} + FN_{\text{leaching}} + FN_{\text{NH}_3/\text{NO}_x(\text{fert., manure, animal})} + \\ & FN_{\text{NO}_x(\text{soil})} + FN_{\text{N}_2\text{O}} + FN_{\text{N}_2(\text{denitr.})} \end{aligned} \quad (1)$$

N imports include the addition of N from organic and inorganic fertiliser ($FN_{\text{org fert.}} + FN_{\text{synt fert.}}$), the fixation of N_2 through biological fixation ($FN_{N_2 \text{ (biol. fixation)}}$) and the deposition of NH_3 , HNO_3 , NH_4^+ , NO_3^- from dry, and NH_4^+ and NO_3^- from wet deposition (summarised as $FN_{\text{dep.}}$). Exports include the N lost from plant biomass at cuts for silage (FN_{harvest}), the off-take of N in meat and wool from animals (FN_{animal}), the loss of organic and inorganic dissolved N through leaching (FN_{leaching}), the NH_3 and NO_x emissions from volatilisation of inorganic and organic fertiliser spreading as well as from animal excretion ($FN_{\text{NH}_3/\text{NO}_x(\text{fert., manure, animal})}$), the emission of NO_x from the soil ($FN_{\text{NO}_x(\text{soil})}$), the emission of N_2O from the soil ($FN_{\text{N}_2\text{O}}$) and the loss of N_2 from total denitrification ($FN_{\text{N}_2(\text{denitr.})}$).

The change in the C balance (ΔC) over time equals the net biome production (NBP) and can be written for our site as:

$$\Delta C/\Delta t = \text{NBP} = FC_{\text{CO}_2} + FC_{\text{org fert}} + FC_{\text{animal}} + FC_{\text{CH}_4} + FC_{\text{leaching}} + FC_{\text{harvest}} \quad (2)$$

FC_{CO_2} represents the net ecosystem exchange (NEE) of CO_2 and $FC_{\text{org fert}}$ is the C input through manure application. Carbon input from animal excretion was not included in the budget as it was assumed to be



recycled C from plant and soil uptake. FC_{animal} includes the C off-take through animal weight increase and wool production. As grazing cows were heifers in calf, there was no C off-take through milk to be considered. Methane emissions from enteric fermentation by ruminants, animal excretion and manure application as well as CH_4 fluxes from the soil are included in FC_{CH_4} . FC_{leaching} is the C lost through dissolved organic and inorganic C and dissolved CH_4 leaching and FC_{harvest} represents the C lost from the system through plant biomass export from harvests (cut for silage). Carbon emissions from farm operations (i.e. tractor emissions) or off farm emissions (i.e. fertiliser manufacture) are not included in the C budget.

Details of methods to quantify each N and C budget component, as listed in Eq. (1) and (2), are described under Sect. 2.4 to 2.11. Some budget components were measured throughout the 9 years presented, while others were only measured in some years or not at all. Missing data were derived from the literature, models or averages from available data from other years.

The annual net GHG exchange (NGHGE) was calculated from annual NEE (FC_{CO_2}), CH_4 (FC_{CH_4}) and N_2O (FN_{N_2O}) fluxes using global warming potentials (GWPs) at the 100-year time horizon (IPCC, 2013):

$$NGHGE = (FC_{CO_2}) + FC_{CH_4} * k_{CH_4} + FN_{N_2O} * k_{N_2O} \quad (3)$$

Where;

$$k_{CH_4} = 9.09, \text{ since } 1 \text{ kg } CH_4\text{-C} = 9.09 \text{ kg } CO_2\text{-C}$$

$$k_{N_2O} = 127, \text{ since } 1 \text{ kg } N_2O\text{-N} = 127 \text{ kg } CO_2\text{-C}$$

In addition the net annual greenhouse gas balance (NGHGB) was calculated by including the loss of C through animal meat and wool production, harvest off take, C leaching and input by organic fertiliser application:

$$NGHGB = NGHGE + FC_{\text{org fert}} + FC_{\text{animal}} + FC_{\text{leach}} + FC_{\text{harvest}} \quad (4)$$



2.4 Nitrogen and carbon import by fertiliser and manure ($FN_{\text{synt fert}} + FN_{\text{org fert.}} + FC_{\text{org fert.}}$)

Mineral fertiliser was applied by a spreader as either ammonium nitrate or urea. Data of application rates and N content were obtained from the farmer. Six month old cattle slurry was spread by a vacuum slurry tanker. Three samples from the slurry tank were taken at each application and analysed for ammoniacal nitrogen (NH_3 and NH_4^+), dry matter content, total N, total C, pH and nitrate. The total N and C import to the field by the slurry was calculated by the volume of the slurry applied and the N and C analyses of the slurry.

2.5 Nitrogen and carbon export by harvest ($FN_{\text{harvest}} + FC_{\text{harvest}}$)

The farmer estimated a harvest of 15 t fresh weight (FW) $\text{ha}^{-1} \text{y}^{-1}$ at the first cut and 10 t FW $\text{ha}^{-1} \text{y}^{-1}$ at the second cut of a year. As there were two cuts in 2002 and one cut in 2003 the estimated harvest was 25 t FW $\text{ha}^{-1} \text{y}^{-1}$ for 2002 and 10 t FW $\text{ha}^{-1} \text{y}^{-1}$ for 2003. A subsample of harvested vegetation was collected and dried at 80°C for plant N and C analysis using a Carbo-Erba/400 automated N and C analyser.

2.6 Nitrogen and carbon export by meat and wool ($FN_{\text{animal}} + FC_{\text{animal}}$)

It was estimated by the farmer that heifers increased in weight by 0.8kg per day (starting weight of 450 kg). The ewe weight was assumed to be constant (60 kg), whereas lambs were brought to the field at a weight of 5 kg and removed when they reached a weight of 45 kg. The total meat export was calculated from the daily weight increase of heifers and lambs multiplied by the animal number per day. To calculate the N and C export from meat we assumed an N content of 3.5 % and a C content of 21 % (Flindt, 2002). Ewes were sheared annually in June, yielding an estimated 2.5 kg of wool per sheep. Wool N and C export was calculated from wool production multiplied by the average sheep number in June, assuming a N and C content of wool of 16.5 and 50 %, respectively (Roche J., 1995)

2.7 Nitrogen and carbon leaching ($FN_{\text{leaching}} + FC_{\text{leaching}}$)



Two sets of ten glass suction cups (pore size $<1 \mu\text{m}$, ecoTech, Bonn, Germany) for soil water and four
230 Teflon suction cups (ecoTech, Bonn, Germany) for soil gas collection were installed in August 2006.
One set was located on a slope, another on a hollow. For the budget calculations we only used results
from the slope location as the hollow location was frequently water logged. Suction cups were installed
horizontally from a soil pit beneath the A horizon (30 cm depth) and at 90cm depth and connected to 2-l
glass bottles in an insulated aluminium box placed into the soil pit. Samples were collected every two to
235 three weeks. For further details and description of dissolved organic and inorganic C (DIC, DOC) and
dissolved CH_4 analysis see Kindler et al. (2011). Dissolved inorganic and organic N (DIN, DON) and
total N (TN) concentrations in leachate water were analysed by colorimetric analysis (San⁺⁺, Automated
Wet Chemistry Analyzer - Continuous Flow Analyzer (CFA), Skalar, The Netherlands). Leachate C and
N concentrations were measured from October 1st 2006 - March 30th 2008. Dissolved C and N were
240 calculated by multiplying concentrations of DIC, DOC and dissolved CH_4 or DIN and DON
respectively, with leachate volume. The latter was derived from a soil water model based on daily
precipitation and evaporation data (Kindler et al., 2011). For the remaining years N was simulated using
the LandscapeDNDC model (Haas et al., 2013, with the model tested and validated with comprehensive
measured data. LandscapeDNDC is a process based biogeochemical model with unifying functionalities
245 of the agricultural-DNDC (e.g. Li et al., 1992; Li 2000) and the ForestDNDC model (e.g. Kesik et al.,
2005; Stange et al., 2000), particularly suitable for ecosystem N turnover and associated losses of N
trace gases and nitrate leaching (Wolf et al., 2012; Chirinda et al., 2011; Kiese et al., 2011). For C
leaching linear regression models describing the relationship between calculated C leaching fluxes and
leachate volume for the measurement period (DOC; $y = 0.0186x - 0.0695$, $R^2 = 0.8663$, DIC; $y = 0.021x$
250 $- 0.0008$, $R^2 = 0.8056$ and dissolved CH_4 : $y = 0.0019x - 0.0135$, $R^2 = 0.7623$) were used to extrapolate
to the remaining years.

2.8 Gaseous N fluxes

255 2.8.1 N deposition (FN_{dep})

Wet N deposition



Wet N deposition was determined from daily samples collected by an automatic precipitation sampler (Eigenbrodt® precipitation collector 181/KS, Königsmoor, D) at Auchencorth Moss (3°14'35W, 55°47'34 N), 17 km south west of Easter Bush (Skiba et al., 2013, McKenzie et al., 2015). The precipitation collector was only open during rainfall and closed automatically when precipitation ceased. Precipitation samples were analysed for NO_3^- and NH_4^+ by ion chromatography (Methrom AG, Switzerland). Typical detection limits were 0.5 μM for NH_4^+ and 0.4 μM for NO_3^- . Annual inorganic N deposition at this site was then adjusted to annual rainfall amounts at Easter Bush. For years where no data were available (2002, 2003), an average mineral N concentration per mm rainfall for 2004-2009 was taken and adjusted to the annual rainfall amount at Easter Bush in 2002 and 2003.

Dry N deposition

Cumulative monthly concentrations of gaseous and aerosol N species (NH_3 , HNO_3 , particulate NH_4^+ and NO_3^-) were collected from another field, about 300m distance from our study field, using a DELTA system (DENuder for Long Term Atmospheric) (Sutton et al., 2001). The DELTA system comprised of a denuder filter sampling train, an air pump (providing a sampling flow rate of 0.2-0.4 L min^{-1}) and a high sensitivity dry gas meter to record sampled volumes (Tang et al., 2009) set at 1.5 m height above ground. N dry deposition fluxes were calculated using the average flux from four different inferential models; the UK CBED scheme (Concentration Based Estimated Deposition technique (Smith et al., 2000), the Dutch IDEM model (Bleeker, 2000), (Erisman et al., 1994), the dry deposition module of the Environment Canada model CDRY (Zhang et al., 2001; Zhang et al., 2003) and the surface exchange scheme EMEP (Simpson et al., 2003; Tuovinen et al., 2009), as described in detail by Flechard et al. (2011).

2.8.2 N_2O fluxes ($\text{FN}_{\text{N}_2\text{O}}$)

From June 2002 to July 2003 N_2O fluxes were measured continuously by eddy covariance (EC) using an ultra-sonic anemometer coupled with a Tunable Diode Laser absorption spectrometer (TDL) at a frequency of 10 Hz. For details see Di Marco et al. (2004). The detection limit for the TDL was estimated to be 1 ppbV and the detection limit for a 30 min averaging period of the N_2O flux



285 measurement was estimated at $11 \text{ ng N}_2\text{O-N m}^{-2} \text{ s}^{-1}$. From August 2006 to December 2009 N_2O fluxes were measured using manual closed static chambers (Clayton et al., 1994, Skiba et al., 2013). Four chambers (0.4 m diameter, 0.2 m height) were inserted into the soil to 0.03 – 0.07 m depth and were accessible for animals to graze. Chambers were closed usually between 10:00 and 12:00 for 60 minutes with an aluminium lid fitted with a draft excluder. Samples of 200 ml were collected by syringe and
290 injected into Tedlar bags at the beginning and the end of the closure time through a three way tap fitted into the lid. In the laboratory samples were transferred to glass vials and analyzed for N_2O using a Hewlett Packard 5890 series II gas chromatograph (Agilent Technologies, Stockport, UK), fitted with an electron capture detector (detection limit: $\text{N}_2\text{O} < 33 \text{ ppbV}$). Fluxes were calculated from the change of gas concentration with time of closure, multiplied by the volume of enclosed space and divided by its
295 surface. Linearity tests were performed in between measurements showing a linearity of up to 120 minutes with an average $R^2 = 0.96$. The minimal detectable flux was $12 \text{ ng N}_2\text{O-N m}^{-2} \text{ s}^{-1}$. Fluxes were measured weekly and more frequently during fertilisation. Cumulative fluxes were calculated by gapfilling data for missing days using linear interpolation and summing up all gapfilled data over each calendar year. For the periods where no N_2O fluxes were measured (January -May 2002, July 2003-
300 March 2004, May 2004-July 2006) fluxes were simulated by LandscapeDNDC (Haas et al., 2013).

2.8.3 NO_x fluxes ($\text{FN}_{\text{NO}_x(\text{soil})}$)

NO_x fluxes from the soil were only measured for a short period (June 2009-August 2010). The NO_x fluxes were measured using an autochamber system described in detail by Butterbach-Bahl et al.
305 (1997). Four Perspex chambers (0.5 m x 0.5 m x 0.15 m; total volume 0.0375 m^3) were fastened onto shallow frames and moved fortnightly to a second position to allow free grazing of the first chamber set. One control chamber was placed onto a Perspex surface to account for ozone/ NO_x reactions inside tubing and chamber. Measurements were made 4 times per day, every 6 hours for an 8 min period per chamber. An in-house Labview program controlled chamber closure and activated a solenoid valve
310 system to sample from the 4 chambers in sequence, interlaced with sampling from the control chamber. PTFE tubing (25 m in length, ID x OD; 4.35 x 6.35 mm) connected chambers to the NO_x (42i-TL Trace Level NO_x Analyzer, Thermo Scientific US) and ozone (Model 49i Ozone Analyzer, Thermo



Scientific, US) analysers located inside the mains-powered field cabin. Fluxes were calculated from the difference between control (on Perspex) and sample chambers (on grass), the flowrate into the analysers (11 lpm) and the surface area of the frames (0.25 m²). We used simulated data from Landscape DNDC for years where no NO_x fluxes were measured.

2.8.4 NH₄ + NO_x volatilisation (FN_{NH₃/NO_x} (fert., manure, animal))

The fraction of nitrogen that volatilises as NH₄ and NO_x from applied synthetic fertiliser or cattle slurry application and animal excretion was estimated to be 10% and 20% of total N applied, respectively (IPCC, 2006b). The animal excretion amount was estimated in accordance with the IPCC Guidelines (IPCC, 2006a). The amount of N excretion (N_{ex}) from animals depends on the total N intake (N_{intake}) and total N retention (N_{retention}) of the animal. N_{intake} (amount of N consumed by the animal) depends on the gross energy (GE) intake (see section 2.10) and the crude protein content (CP%) of the feed, assumed to be 15.6% (MAFF, 1990). N_{retention} represents the fraction of N intake retained by the animal as meat, milk or wool. For lactating ewes the milk production was estimated at 0.618 l animal⁻¹ d⁻¹ and the milk protein content (Milk PR%) at 5.3% (Atti et al., 2006). Daily N excretions were thus calculated as 0.0263 kg N animal⁻¹ d⁻¹ for ewes and varied between 0.0019-0.0106 kg N animal⁻¹ d⁻¹ for lambs and 0.096-0.1013 kg N animal⁻¹ d⁻¹ for heifers.

2.8.5 N₂ emission by total denitrification (FN_{N₂(denitr.)})

Di-nitrogen (N₂) emissions resulting from total denitrification in the soil was not measured in our experiment. We therefore used the N₂ emission rates from LandscapeDNDC simulations.

2.8.6 Biological N₂ fixation (FN_{N₂} (biol. fixation))

The species composition was measured by the visual estimation method (Braun-Blanquet, 1964). As the legume fraction (*Trifolium repens*) was smaller than 0.5% at each measuring point we assumed the nitrogen fixation through plants to be zero.

2.9 Exchange of CO₂ (FC_{CO₂})



NEE was measured by an eddy covariance system consisting of a fast response 3D ultrasonic anemometer (Metek USA-1, Metek GmbH, Elmshorn, Germany) and a fast closed path CO₂-H₂O analyser (LI-COR 7000 infra-red gas analyzer (IRGA), LI-COR, Lincoln, NE, USA). Wind velocity components were measured at 2.5m above ground and data were logged at 20 Hz by a PC running a custom LabView data acquisition program. Air was sampled 0.2 m below the sensor head of the anemometer using 6.3 mm (1/4 in. OD) Dekabon tubing. The IRGA was located in a field laboratory ca. 10 m from the mast. Lag times between wind data and trace gas concentrations were synchronised and taken into account in the offline data-processing (Helfter et al., 2014). Quality control of the eddy covariance data followed the procedure proposed by Foken and Wichura (1996). Data were filtered out if the friction velocity (u_*) was smaller than 0.2 m s⁻¹ (insufficient turbulence), CO₂ concentrations fell outside a plausible interval (330–450 ppm), CO₂ fluxes fell outside the range -50 to 50 μmol m⁻² s⁻¹ and latent (LE) and sensible (H) heat fluxes fell outside the range -250 to 800 W m⁻². Missing NEE data were gap-filled using the online tool developed at the Max Planck Institute for Biogeochemistry, Jena, Germany¹ (Reichstein et al., 2005) NEE is the arithmetic sum of the gross primary production (GPP) and total ecosystem respiration (TER). Flux partitioning of measured NEE into GPP and TER was calculated by the same online tool used for gapfilling. In this flux partitioning approach, daytime TER is obtained by extrapolation of a night time parameterisation of NEE on air temperature and GPP is the difference between ecosystem respiration and NEE. Contrarily to unmanaged ecosystem, TER at our site also includes the respiratory loss of CO₂ by grazing animals. Net primary production (NPP), which represents the annual plant growth (difference between GPP and autotrophic respiration) was calculated as 50% of GPP (Waring et al., 1998).

2.10 Methane fluxes (FC_{CH₄})

Methane fluxes from the soil were measured with closed static chambers simultaneously with the N₂O measurements (see Sect. 2.8.2). The same GC was fitted with a flame injection detector (detection limit: CH₄ < 70 ppbV). The minimal detectable flux was 17 ng CH₄-C m⁻² s⁻¹. Fluxes were measured weekly and more frequently at fertiliser events. As measured soil CH₄ fluxes were close to zero and did not

¹ <http://www.bgc-jena.mpg.de/~MDIwork/eddyproc/upload.php>



370 vary significantly between months, we calculated CH₄ for months where no CH₄ fluxes were measured (January-May 2002, July 2003-March 2004, May 2004-July 2006), as an average monthly cumulative flux from other years.

Methane emissions from grazing animals, i.e. animal excretion and enteric fermentation, were estimated following the IPCC Tier 2 methodology (IPCC, 2006a: Stewart et al., 2009). For animal excretion only solid volatile production was considered, as urine has no effect on CH₄ emissions (Jarvis et al., 1995). The calculation of CH₄ emissions from excretion was based on the amount of volatile solids (VS) excreted, the maximum CH₄ producing capacity (B₀) of the manure and the CH₄ conversion factor (MCF), which is specific to the storage type (pasture, in our study). The amount of VS excreted depended largely on the GE intake of the animal. The GE intake (based on digestible energy of feed intake, milk production, pregnancy, current weight, mature weight, rate of weight gain and IPCC constants) in our study was estimated at 19.5 MJ animal⁻¹ d⁻¹ for ewes, while it ranged from 7.9 to 14.9 MJ animal⁻¹ d⁻¹ for lambs and from 160.9 to 169.7 MJ animal⁻¹ d⁻¹ for heifers. Emission factors for excretion were calculated as 0.198 kg CH₄ head⁻¹ y⁻¹ for ewes and varied between 1.64-1.73 kg CH₄ head⁻¹ y⁻¹ for heifers and 0.081-0.152 kg CH₄ head⁻¹ y⁻¹ for lambs. Methane emission factors for enteric fermentation were calculated from GE intake and CH₄ conversion factors (Y_m). Depending on animal type and live weight, emission factors were 7.6 kg CH₄ head⁻¹ y⁻¹ for ewes and varied between 60.1-63.8 kg CH₄ head⁻¹ y⁻¹ for heifers and 2.0-4.0 kg CH₄ head⁻¹ y⁻¹ for lambs. Annual emissions from excretion and enteric fermentation were calculated from daily CH₄ emissions per animal multiplied by the animal number.

Methane emissions from slurry applications were assumed to be small. As no chamber measurements were conducted at the time of slurry spreading, the emissions were estimated as 0.07 % of the applied assuming that emissions were comparable to those in a related study (Jones et al., 2006), where CH₄ was measured from chambers after slurry application on a nearby field in 2002 and 2003.

2.11 VOC

Fluxes of non-methane volatile organic compounds (VOC) were not measured. We assumed similar VOC emissions to those reported by Davison et al. (2008) for an intensively managed grassland in



Switzerland, where the daily average flux of methanol, acetaldehyde and acetone over 3 days after cutting were 21.1, 5.1. and 2.6 nmol m⁻² s⁻¹, respectively. Based on those values, annual VOC emissions from our field were estimated to be in the order of 0.03% of the annual C offtake in harvest and 0.08 % of annual C off-take by grazing animals. We therefore assumed VOC emissions to be negligible.

2.12 Soil N and C measurements

Total N and C content of the soil were measured in May 2004 and May 2011. One hundred soil cores with an inner diameter of 8.7 (2004) and 8.3 cm (2009, both corers from Eijkelkamp Agrisearch Equipment BV, Giesbeek, The Netherlands) were collected along a regular grid with a distance of 10 m between sampling points on both occasions. Cores were separated into layers of 0-5, 5-10, 10-20, 20-30, 30-40, 40-50 and 50-60 cm. Coarse stones of a diameter > 4 mm and roots of a diameter >1mm were removed from the samples prior to drying at 40 °C. Stone and root samples were air-dried separately. Then, soil samples were sieved to < 2 mm. Particles > 2 mm were combined with the coarse stones. Dry weights of roots and combined stone fractions were determined. Total N and C concentrations in < 2 mm soil separates were determined using dry combustion (VarioMax, Elementar Analysensysteme GmbH, Hanau, Germany). As the site contains no inorganic C, total C equals organic C. As bulk density varies spatially and over time (e.g. through compaction by livestock), the soil N and C content per unit ground area to a fixed depth will also change, without any change in the mass fraction of N and C in dry soil. Therefore, total N and C stocks were calculated on an equivalent soil mass (ESM) basis, so that comparisons between years were valid (see Gifford and Roderick, 2003, Wendt and Hauser, 2013). A cubic polynomial was fitted to the data, to predict cumulative N and C with cumulative soil mass in the profile. A soil mass of 800 kg m⁻² was used (Table 7), which corresponds to approximately 60-cm depth, which was the depth of the corer. Uncertainty in the estimates of stock change was based on the prediction intervals in the cubic polynomial at a soil mass of 800 kg m⁻².

2.13 Ancillary measurements



425 Soil temperature and volumetric soil moisture were continuously recorded at four depths (3.5, 7.5, 15 and 30 cm) by temperature probes (temperature probe 107, Campbell Scientific, Loughborough, UK) and TDR probes (TDR 100, Campbell Scientific, Loughborough, UK), respectively, the latter installed in June 2002. Rain was measured by a tipping bucket rain gauge, while air temperature and relative humidity were measured by an integrated humidity and temperature transmitter (HUMITTER[®], Vaisala Ltd, Suffolk, UK).

430 **2.14 Statistical and uncertainty analysis**

435 Random error was determined as 2σ -standard error (95% confidence) of the overall mean according to Gaussian statistics. Analyses of variance (ANOVA) were used to test if values were significantly different from zero ($p < 0.05$). For systematic errors the uncertainty range of measurements as well as of parameterisations and literature based estimates was estimated according to expert judgment. To calculate the combined effect of systematic uncertainties of each flux component on the C and N budget simple Gaussian error propagation rules were used. Confidence intervals are given at the 95% confidence level.

3. Results

440 **3.1 Climate and management**

445 The meteorological conditions exhibited substantial inter-annual variability in the study period 2002-2010 (Table 2 and Fig. 2). Annual rainfall ranged from 575 mm to 1238 mm with highest monthly rainfalls at 280 mm month⁻¹ in September 2002. Lowest annual reported rainfall was in 2010; this low value was caused by a gap in data from January-March, due to snowfall. Average annual air temperature ranged from 8.3 to 9.6 °C with highest daily air temperatures of 30.4 °C in August 2005 and lowest in December 2010 at -10.3 °C. Highest average monthly air temperatures were measured in July 2006 at 17°C and lowest monthly average air temperatures at 2°C in November 2009. In 2003 the highest average annual temperature (9.6° C) and lowest annual rainfall (680 mm) were measured with a correspondingly low annual soil water content of 31 %. The duration of the growing season was defined 450 per calendar year as the period bounded by the first and last 5 consecutive days with mean daily air



temperature ≥ 5 °C. The length of the growing season (LGS) varied between 151 days (2006) and 242 days (2009) (Table 2).

Livestock stocking density exhibited both intra- and inter-annual variability. The average annual stocking density was lowest in 2002 and 2003 at $0.27 \text{ LSU ha}^{-1} \text{ y}^{-1}$ and $0.54 \text{ LSU ha}^{-1} \text{ y}^{-1}$, respectively (Table 1), which were the years where the grass was cut for silage and no lambs were present in the field. In 2007, 2008, 2009 and 2010 no heifers were present in the field. Highest annual average stocking density occurred in 2004 and 2007 at $0.99 \text{ LSU ha}^{-1} \text{ y}^{-1}$ and $0.91 \text{ LSU ha}^{-1} \text{ y}^{-1}$, respectively. Maximum monthly stocking density occurred in September 2006 at 13.8 LSU ha^{-1} , while interim periods with no grazing at all were observed in all years (Fig. 1a). Mineral N fertiliser was applied split into 3 to 5 applications per year, ranging from 2.5 to $9.6 \text{ g N m}^{-2} \text{ application}^{-1}$ (Fig. 1b). Organic manure was applied in 2004 and 2005 as cattle slurry, spread at a rate of 6.9 and $15.8 \text{ g N m}^{-2} \text{ application}^{-1}$, respectively, which resulted in a C input of 55.4 and $171.8 \text{ g C m}^{-2} \text{ application}^{-1}$, respectively (Fig. 1b and c). The grass was only cut in 2002 and 2003. Harvested biomass in 2002 and 2003 ranged from 2.60 to $3.75 \text{ t DW ha}^{-1} \text{ cut}^{-1}$ which resulted in an N off-take ranging from 1.7 to $4.7 \text{ g N m}^{-2} \text{ cut}^{-1}$ and a C removal from the field ranging from 113.1 to $169.5 \text{ g C m}^{-2} \text{ cut}^{-1}$ (Fig. 1c).

3.2 Uncertainty analysis

Systematic uncertainties for each component of the C and N budget are shown in Table 3. Uncertainty values were estimated according to expert judgment. The systematic uncertainty of the N input from mineral fertiliser was assumed to be minimal (1 %), while the systematic uncertainty of the N and C spread by the manure was assumed to be 17 % on average for the C and N analysis. Together with an uncertainty of 10 % of the volume spread, this resulted in a total uncertainty of 20 %. The uncertainty of the C and N analysis for harvest were 4 and 12 %, respectively. We assumed an error of 10% in the farmer's estimate of the harvest amount, which resulted in a total uncertainty of 16 % for N and 11 % for C off take. We attributed a systematic uncertainty of 30 % to the modelled data for C and N leaching. The systematic uncertainty of the meat and wool consists of the estimated uncertainty in the animal weight, animal numbers and literature values for wool and meat C and N contents. We assign an uncertainty for animal weight of 10 %, for animal numbers of 5 % and for literature values of wool and



meat C and N content of 3 %, resulting in a total uncertainty of 12 %. The uncertainty of wet N
480 deposition was 30 % resulting from the error of sample analysis and a potential bias from dry deposition
on the funnel. The uncertainty of dry N deposition consisted of an error of 7 % for the analysis of
DELTA samples and an 80% uncertainty of the variation of the output from the four models, which
resulted in a total uncertainty of 80%. The systematic uncertainty attributed to the annual cumulative
N₂O fluxes was 30 %, due to the uncertainty of gapfilling. The uncertainty attributed to the modelled
485 NO_x fluxes is 30 %. The uncertainty attributed to the NH₄ and NO_x volatilisation was 30 % from
applied synthetic fertiliser and 50 % from cattle slurry application and animal excretion. The uncertainty
attributed to the N₂ fluxes was 30 %. The total uncertainty for NEE values was estimated to be 80 g C
m⁻² y⁻¹ (Levy et al., submitted). The systematic uncertainty of annual cumulative soil CH₄ fluxes was
very high at 160 %, due to the uncertainty of gap filling and as values were close to zero. The
490 uncertainty of CH₄ from enteric fermentation and animal excretion estimates were each assumed to be
20%, according to IPCC (2006a). The uncertainty of CH₄ fluxes from organic manure application was
estimated at 120 %.

3.3. N budget

495 In our grassland system the N balance is the difference between the N input through fertiliser and
atmospheric deposition and the N output through harvest, animal export, leaching and gaseous
emissions. The total resulting balance over the nine years, derived from flux calculations and
estimations, showed that N was stored at an average rate of -7.21 ± 4.6 g N m⁻² y⁻¹ (p<0.05). From 2003
to 2010, N was stored at a rate of -3.1 to -17.9 g N m⁻² y⁻¹, whilst in 2002 N was lost at a rate of 6.3 g N
500 m⁻² y⁻¹ (Table 4). The major N input consisted of inorganic fertiliser, ranging from -11 to -25.9 g N m⁻²
y⁻¹, averaging at -19.2 g N m⁻² y⁻¹, while N deposition represented only between 1.9 and 5.9% of the
total N input. During the years where N was stored, a significant positive correlation between total N
input from fertiliser and N storage was observed ($R^2 = 0.55$). Largest losses resulted from leaching at an
average rate of 5.34 ± 3.4 g N m⁻² y⁻¹ and were estimated to be highest in 2002 at 14.9 g N m⁻² y⁻¹ and
505 lowest in 2003 at 0.09 g N m² y⁻¹. We found a strong correlation between N leaching and rainfall ($R^2 =$
0.82), if values from 2004 were excluded, a weak correlation between livestock density and N leaching



if the years 2002 and 2004 were excluded ($R^2=0.47$), while no correlation with total N input could be found. The total N off take through meat and wool ranged from $0.15\text{--}3.12\text{ g N m}^{-2}\text{ y}^{-1}$, while the total annual N offtake from harvest was $5.0\text{ g N m}^{-2}\text{ y}^{-1}$ in 2002 and $4.68\text{ g N m}^{-2}\text{ y}^{-1}$ in 2003. Amongst gaseous exchanges, highest losses were estimated from N_2 emissions, averaging at $2.76\text{ g N m}^{-2}\text{ y}^{-1}$ with maximum losses of $4.12\text{ g N m}^{-2}\text{ y}^{-1}$ in 2009, although in 2004 and 2005 losses from NO_x/NH_3 volatilisation from excretion and organic fertilisation exceeded losses from N_2 emissions. Losses through NO_x from the soil were always less than 1% of the total N exchange ($0.2\text{ g N m}^{-2}\text{ y}^{-1}$). Nitrous oxide emissions ranged from 0.11 to $1.27\text{ g N m}^{-2}\text{ y}^{-1}$, representing 1.3–8.4 % of the total N export. Annual N_2O emissions showed no correlation with precipitation, livestock density or total N input. However, there was a positive correlation with rainfall if 2004 and 2007 data were excluded ($R^2=0.78$); with livestock density if the years 2002 and 2004 were excluded ($r^2=0.70$); and with total N input if the years 2002, 2003 and 2010 were excluded ($R^2=0.76$). N_2O emission factors (percentage of N lost from total N inputs by mineral and organic fertiliser), ranged between 0.6 and 7.5 % (Table 6).

To investigate the influence of different managements on the N and C budget, we separated experimental years into harvested and grazed (2002 and 2003) and grazed only years (2004–2010 Fig. 3 and 4). During the harvested years, the main loss of N from the system occurred through leaching (39.2% of total N inputs), followed by the export through harvest (25.2%), while the export from animals (meat and wool) accounted for less than 2 % of total losses (Fig. 3a). The main loss to the atmosphere resulted from total denitrification (N_2 ; 15.4%), followed by NO_x/NH_3 volatilisation from inorganic N fertiliser applications (9.5%), while N_2O emissions accounted for 3.3%, NO_x/NH_3 volatilisation from excretion for 2.7% and NO_x from soil for less than 1%. The residual 2% represents the N storage in the soil and the uncertainty of the budget. When grazed-only years were considered (Fig. 3b), the residual part was the highest at 38.6%. Losses through leaching (19.9%) and N_2 (11.4%) were lower in grazed years compared to harvested years, while the export through grazing animals were considerably higher at 15.8% (sum of N loss through meat, wool and NO_x/NH_3 volatilisation from excretion). An additional loss occurred in grazed years through the volatilisation of NO_x/NH_3 from organic fertiliser applications in 2004 and 2005 (3%). Losses through N_2O and NO_x/NH_3 from inorganic fertiliser were comparable to harvested years at 2.5% and 8.3%, respectively.



535 Cumulative soil N stocks were derived from soil core measurements taken in May 2004 and
May 2011. Nitrogen storage over the 7 years was calculated from the cumulative equivalent soil mass
(ESM) for the soil mass increment of 800 kg m^{-2} , which corresponds to approximate 60 cm depth. The
estimated N storage over the 7 years was $-4.51 \pm 2.64 \text{ g N m}^{-2} \text{ y}^{-1}$ (Table 7) and was a significant N
accumulation to the soil ($p < 0.01$). The estimated N storage derived from flux calculations between
540 2004 and 2010, however was $-9.20 \pm 4.10 \text{ g N m}^{-2} \text{ y}^{-1}$, which is 2 times more than that calculated by
sequential soil analysis.

3.4. C budget

Annual C inputs through photosynthesis (GPP) varied between -982.1 and $-2162.9 \text{ g C m}^{-2}$, and losses
545 through autotrophic and heterotrophic respiration (TER) varied between 972.1 and $2183.2 \text{ g C m}^{-2}$, both
considerably larger than any other C fluxes (Table 5). If only the NEE was considered (difference
between GPP and TER), the grassland acted as a sink for CO_2 at an average of $218 \pm 155 \text{ g C g C m}^{-2} \text{ y}^{-1}$,
and the CO_2 uptake was significantly different from zero ($p < 0.05$). The sink strength ranged from $-$
 $10 \text{ g C m}^{-2} \text{ y}^{-1}$ (2006) to $-606 \text{ g C m}^{-2} \text{ y}^{-1}$ (2009), only in 2004, the grassland was a small source of CO_2
550 ($72 \text{ g C m}^{-2} \text{ y}^{-1}$). Taking into account all C inputs and outputs (NBP), C was sequestered on average at
 $164 \pm 140 \text{ g C m}^{-2} \text{ y}^{-1}$ over the nine years, although the storage was not significantly different from zero
($p < 0.05$). In 2004 and 2006 C was lost from the ecosystem. The major C import resulted from NEE in
all years apart from 2005, when the C input from manure application was larger. Highest C export
occurred from harvest in 2002 and 2003 (270.6 and $169.5 \text{ g C m}^{-2} \text{ y}^{-1}$ respectively), while second largest
555 export in 2002 and 2003 and largest exports in other years was leaching (6.8 to $25.1 \text{ g C m}^{-2} \text{ y}^{-1}$). The
measured C leaching value for 2007 ($15.4 \text{ g C m}^{-2} \text{ y}^{-1}$, table 5) differs from the leaching value published
for Easter Bush by Kindler et al. (2011), as we only used values of one of the two measured sites in this
manuscript (slope, not hollow, as the hollow location was frequently water logged). The third largest C
loss consisted of C export from meat in 2004-2010, ranging from 6.4 - $15.8 \text{ g C m}^{-2} \text{ y}^{-1}$. In 2002 and
560 2003, when no lambs were present in the field, C export from meat was exceeded by CH_4 losses from
enteric fermentation. Carbon export from wool ranged from 0.5 to $2.1 \text{ g C m}^{-2} \text{ y}^{-1}$. CH_4 emissions from
organic fertilisation, soil processes and animal excretion were always less than 1 % of the total C losses.



CH₄ losses from enteric fermentation ranged from 1.5 to 5.7 g C m⁻² y⁻¹, corresponding to 0.5–22.5 % of all C losses from the ecosystem. The annual carbon balance (NBP) was dominated by the NEE. A high livestock density tended to reduce the net sink strength. A significant negative correlation of NEE as well as NBP with stocking density could be seen ($R^2=0.77$ and $R^2=0.83$, respectively), if the years with cuts (2002 and 2003) were excluded. The NBP correlated positively with rainfall ($R^2=0.48$) whereas the correlation improved if the dry year 2003 was excluded ($R^2=0.78$). There was only a weak correlation between NEE and rainfall ($R^2=0.38$ for all years, $R^2=0.47$ without the year 2003).

Net primary production (NPP) in years when grass was harvested and grazed (2002 and 2003) and grazed only (2004–2010) are presented in Figure 4. In both management types most C was lost through ecosystem respiration, (67% and 71% of NPP, respectively). Harvest export represented 21% of NPP. Leaching accounted for 1.5% of NPP during harvested years and 2.2% in grazed only years. Animal export (meat and wool) consisted of 1.5% of NPP in grazed only years and was less than 0.2% of NPP in grazed and harvested years. The sum of all CH₄ emissions (from organic fertilisation, excretion, enteric fermentation and soil) was less than 1% of the NPP. The residual part, which includes the C storage in the soil as well as the uncertainty of the budget, was estimated at 10% and 24% of NPP in harvested and grazed or grazed years, respectively.

The C content for the cumulative soil mass increment 0–800 kg m⁻² (~ 0–60 cm) was lower in 2011 compared to 2004, resulting in a C loss of 29.08 ± 38.19 g C m⁻² (Table 7). In comparison, based on flux calculations C was stored at 180 ± 180 g C m⁻² y⁻¹ over the 7 years. However, neither C loss calculated by sequential soil analysis, nor C storage estimated from flux calculations were significantly different from zero.

3.5. Greenhouse gas budget

In order to calculate the global warming potential for the Easter Bush grassland fluxes of the greenhouse gases CO₂, N₂O and CH₄ were expressed in CO₂ equivalents considering the different global warming potentials for each gas at the 100 year time horizon (1 for CO₂, 298 for N₂O and 25 for CH₄, IPCC, 2013). Average greenhouse gas fluxes, net GHG exchange (NGHGE) and attributed net GHG balance (NGHGB) for 2002–2010 are shown in Figure 5. The CO₂ storage from the NEE provided



the largest term in the annual GHG budget. Carbon dioxide (NEE) was sequestered over the 9 years at a rate of $-799 \pm 567 \text{ g CO}_2 \text{ m}^{-2} \text{ y}^{-1}$. This storage was significantly different from zero ($p < 0.05$). On average, the net GHG exchange (NGHGE) was highly correlated with NEE ($R^2=0.96$). On average the grassland was a source of the GHGs CH_4 and N_2O at a rate of 148 ± 30 and $285 \pm 131 \text{ g CO}_2 \text{ m}^{-2} \text{ y}^{-1}$, respectively, both being significantly different from zero ($p < 0.001$ and $p < 0.01$, respectively). Nitrous oxide losses ranged from $52 \text{ g CO}_2 \text{ eq. m}^{-2} \text{ y}^{-1}$ (2004) to $588 \text{ g CO}_2 \text{ eq. m}^{-2} \text{ y}^{-1}$ (2007) (data for each year not shown). Methane from soil processes, manure input as well as animal excretion, accounted for less than 5% of total CH_4 emissions. Methane emissions from enteric fermentation ranged from $53 \text{ g CO}_2 \text{ eq. m}^{-2} \text{ y}^{-1}$ (2002) to $199 \text{ g CO}_2 \text{ eq. m}^{-2} \text{ y}^{-1}$ (2004). The CH_4 emissions, which were predominately (> 97%) of ruminant origin weakened the sink strength of NEE by 18 %. If both CH_4 and N_2O were considered the total trade-off of NEE was a substantial 54% and increased to a total of 67 %, if only grazed years were considered. On average the grassland represented a GHG sink of $-366 \pm 601 \text{ g CO}_2 \text{ m}^{-2} \text{ y}^{-1}$, if only NEE, CH_4 and N_2O were included (NGHGE). If all C components ($\text{FC}_{\text{org.fert}}$, $\text{FC}_{\text{animal}}$, $\text{FC}_{\text{leaching}}$, $\text{FC}_{\text{harvest}}$) are included, the sink strength of the grassland decreased to $-182 \pm 560 \text{ g CO}_2 \text{ m}^{-2} \text{ y}^{-1}$ (NGHGB). This represents a weakening of the sink strength of the NGHGE by 50 %, mainly due to the export of harvest. However, it has to be noted that in harvested years the return of the manure, resulting from the grass fed to livestock off -site, would reduce the GHG balance. If only grazed years were considered the sink strength increased slightly by 5.4 %, due to the C input from manure in 2004 and 2005. Both, NGHGE and NGHGB were not significantly different from zero.

4. Discussion

4.1. N balance

Our experimental field has been under grazing/cutting management for more than 20 years with regular N inputs from mineral fertilizers, manure and animal excretion. As biological N_2 fixation by legumes is inhibited by soil mineral N (Streeter, 1988), the legume fraction was less than 1% and therefore a negligible source of N in our system. Atmospheric N deposition (wet and dry) accounted only for a small fraction of the total N input on our managed grassland. This is in contrast to semi natural systems, where atmospheric N deposition represents the main N input (Pheonix et al., 2006, Bleeker et al., 2011).



620 The main N inputs in our study were from inorganic and organic fertilizer additions. The amount of N added through fertilizer was governed by recommended maximum levels (SRUC, 2013) and lies within the range of N applied in other European studies with similar management (e.g. Laws et al., 2000; Allard et al., 2007; Ammann et al., 2009). Nitrogen added through the excretion from grazing animals was not considered an N input as this represents an internal redistribution of N within the system.

625 4.1.1 N use efficiency

The ratio between N input and percentage of N uptake into the crop or animal products (meat, wool and milk) is defined as the N use efficiency (NUE). In our study a substantial amount of N was removed by harvest, with an NUE of herbage in cut years (2002 and 2003) of 25% (Figure 3a). This seems low compared to reported N efficiencies of 55-80% in harvested herbage from managed temperate grasslands (Ball and Ryden 1984; Ammann et al., 2009). The inclusion of grazing ruminants alters the NUE of herbage as the nitrogen in the grazed grass is consumed and converted to meat, milk, wool, or is excreted. The lower NUE in the grass production in our study is therefore partly due to grazing. Furthermore, it has been shown that the proportion partitioned to plant uptake decreases as the total amount of soil inorganic N increases (Scholefield et al., 1991), which is a further explanation for a low NUE in herbage in our high N input system. There are different mitigation options to reduce N losses and thus increase NUE. The introduction of clover into grassland has been shown to reduce the requirement of N input from fertilisation, thereby resulting in the same yield (Herrmann et al., 2001; Ledgard, 2001). Adherences with fertiliser recommendation systems and avoidance of over fertilisation is also likely to increase the efficiency of N use without compromising productivity (Rees et al., 2013).
640 The use of nitrification inhibitors applied onto grassland has been shown to result in a reduction of N₂O emissions (McTaggart et al., 1997). Furthermore, a novel approach to reduce N losses from sheep urine, by infusing N process inhibitors into the gastrointestinal tract of the animals, has been demonstrated by Ledgard et al. (2008), however, the evidence for this as a mitigation option is still limited, and could face legal and ethical challenges.

645 The NUE in crops is significantly higher compared to the NUE in animal production. The NUE of animal products on our grassland system ranged from 5 to 18% in grazed years (2004-2010), with an



average of 10.6 %. This is in agreement with the NUE reported for sheep of 6.2 % by Van der Hoek (1998) and studies for beef production systems, which reported N efficiencies range from 6 to 12% (Whitehead et al., 1986; Tyson et al., 1992) and 5-20% (Ball and Ryden, 1984). Approximately 85% of crops produced are used for animal feed, which is significantly less efficient than if the crops were used to feed humans directly. A measure to reduce N pollution could therefore be the reduction of meat consumption (Smith et al. 2013).

4.1.2 N loss to the environment:

Nitrogen leaches from grassland soils in the form of nitrate (NO_3^-), ammonium (NH_4^+) and dissolved organic N (DON). Whereas NO_3^- is highly mobile in water and can be easily leached into groundwater, NH_4^+ is less prone to leaching as it is mostly bound to soil particles (Brady and Weil, 2002). Leaching depends on the water-holding capacity of the soil, amount of rainfall, water use by plants and soil nutrient content, which are in turn influenced by management. Leaching occurs predominantly from late autumn to early spring when precipitation often exceeds evapotranspiration (Askegaard et al., 2005). In our field leaching losses varied widely over the years. This variation can mainly be explained by differences in precipitation. Overall, leaching from our field ($5.3 \pm 3.4 \text{ g N m}^{-2} \text{ y}^{-1}$) was comparable to values measured at intensively grazed pastures in Ireland ($1.8\text{-}6.4 \text{ g N m}^{-2} \text{ y}^{-1}$, Watson et al., 2007) and England ($3.8\text{-}13.3 \text{ g N m}^{-2} \text{ y}^{-1}$, Scholefield et al., 1993) or croplands (e.g. Bechmann et al., 1998), max. leaching losses of $10.4 \text{ g N m}^{-2} \text{ y}^{-1}$). However, leaching from our study was high compared to the Swiss NitroEurope site, where a maximum loss of $3.5 \text{ kg N ha}^{-1} \text{ y}^{-1}$ was estimated from an ungrazed grass/clover sward, despite annual rainfall and N inputs comparable to our site (Ammann et al., 2009). This difference can be explained by the different plant cover and management. It has been shown that clover introduction can reduce leaching (Owens et al., 1994), whereas grazing tends to increase leaching (Cuttle and Scholefield, 1995). Grazed grasslands tend to have higher N leaching rates than cut grasslands as the N added as fertiliser is not removed by harvest, but returned to the soil in urine and dung from consumed herbage, prone to leaching. The uneven distribution of excreted organic N further enhances leaching due to the formation of N hotspots, which has been observed at outdoor pig farms (e.g. (Eriksen, 2001). Ryden et al., (1984a) showed a 5.6 times higher leaching loss from grazed



675 compared to cut grassland with 36% of total N inputs lost from grazed compared to 6% lost from cut
grassland. On our site leaching equaled about 20 % of total inputs in grazed years, compared to 39% in
the cut years. However, the higher value in cut years was due to the high rainfall in 2002.

Due to high atmospheric N₂ background, N₂ fluxes cannot be measured directly in the field.
680 However, there are different methods to measure N₂ fluxes indirectly, which have been summarized by
Groffman et al. (2006). In our study, we estimated N₂ losses using the process based biogeochemical
model LandscapeDNDC (Haas et al, 2013, Molina-Herrera et al. 2016). They represented the highest
gaseous losses from our grassland in most years, with an average of 12.6 % of total N inputs and 14 %
of inorganic fertilizer N inputs. This is comparable with the average N₂ loss of 12.5 % from inorganic N
685 applications measured by the acetylene inhibition method from a fertilized and cut, but ungrazed
grassland in Switzerland (Rudaz et al., 1999). Using the same method, van der Salm et al. (2007)
reported a higher loss of 22% of total N input from a cattle grazed pasture on a heavy clay soil in the
Netherlands. Apart from the impact of the heavy clay soil, which could have enhanced denitrification
due to reduced oxygen concentrations, grazing is likely to have enhanced denitrification rates in van der
690 Salm's study. Grazing not only enhances denitrification through soil compaction caused by trampling
animals but also due to the formation of N hot spots resulting from unevenly distributed soil N from
excretion. In our study N₂ losses simulated by LandscapeDNDC are based on average (per ha⁻¹) changes
of the soil N pool instead of the more uneven distribution of soil N in hot spots like urine patches.
Therefore it is likely that N₂ losses in our study have been underestimated.

695

Nitrous oxide emissions are influenced by both management and environmental conditions
(Flechard et al., 2007, Bell et al., 2015; Cowan et al., 2015). In our study N₂O fluxes showed typical
temporal variations with high N₂O peaks after N application decreasing to background levels after < 1
to 20 days, increased losses during wetter periods, and reduced losses during the colder winter months
700 (Skiba et al., 2013). Spatial variability was high due to the uneven distribution of excreta and urine and
uneven soil compaction from grazing animals (Jones et al., 2011). Values measured in our study (0.1 to
1.3 g N m⁻² y⁻¹) are within the range of literature values from reported grazed as well as un-grazed



705 European grasslands (Velthof and Oenema, 1997; Leahy et al., 2004; Flechard et al., 2007). Generally
N₂O losses are higher from grazed grassland compared to cut, ungrazed pasture (Velthof and Oenema,
1995; Luo et al., 1999) due to a more anaerobic environment as a consequence of soil compaction
caused by animal treading and the influence of N and C from the deposition of animal excreta to the soil
(Oenema et al., 1997). We could only find correlations between annual N₂O emissions and stocking
density, rainfall or total N input if certain years were excluded. This shows that N₂O emissions are not a
uniform fraction of N applied, as suggested by the Tier 1 IPCC methodology, but are also influenced by
710 the type of N applied, by stocking density, and by the rainfall at the time of fertilization (Jones et al.,
2007; Flechard et al., 2007). We found a relationship between the cumulative precipitation 1 week
before plus 3 weeks after fertilization and N₂O emissions ($R^2=0.53$) (Skiba et al., 2013). This
relationship, together with the influence of stocking density and type of N applied needs to be
considered when developing Tier 2 emission factors. Emission factors, calculated as a simple fraction of
715 total N input (mineral and organic fertilizer) showed a variation of 0.6 and 7.4% on our field. In five out
of eight years this value was above the uncertainty range (0.3 - 3 %) given by IPCC Tier 1 guidelines
(IPCC, 2006b). However, it has been shown that the N₂O emission factor from managed grassland can
be higher, especially under wet conditions and with a high soil C content as this is the case for Scottish
soils (Jones et al., 2007; Dobbie et al., 1999; Bell et al., 2015).

720 In grazed pastures NH₃ volatilizes from urine patches, decomposing dung as well as from
fertilizers containing urea and NH₄⁺ (Twigg et al 2011). Increased rates of NH₃ losses have been
associated with high stocking densities under a rotational grazing system by Ryden and Mc Neill
(1984). In our study, N volatilized as NH₃ and NO_x from inorganic and organic fertiliser and animal
725 excretion, before it was incorporated into the soil, and accounted for a considerable amount of total N,
with losses of 13 % in cut and grazed years (2002, 2003) and 17 % in grazed only years. Apart from
2004, where stocking rates were highest, NO_x and NH₃ volatilizations from inorganic fertilizer
applications exceeded those from animal excretion, while those from organic manure applications
exceeded those from inorganic fertilizers (2004, 2005). However there is a high uncertainty attributed to
730 those estimates.



735 Soil NO_x emissions result predominately from microbial nitrification of either added N fertilizers or following the mineralization of soil organic matter, animal excretions or added manure. Emissions tend to be linked with aerobic soil conditions (Davidson, 1991). In relation to the total N loss from our grassland system, soil NO_x emissions were estimated to be negligible, accounting for less than 1% of the total budget.

4.1.3 N storage in the soil

740 Results from soil analysis taken in May 2004 and May 2011 indicate that our field has stored N. The N budget assessed from the net N flux balance also showed that N was stored in the soil of our grassland, although at a higher rate (average N storage of $-7.2 \pm 4.6 \text{ g N m}^{-2} \text{ y}^{-1}$ over all 9 years and average N storage of $-9.16 \pm 4.09 \text{ g N m}^{-2} \text{ y}^{-1}$ in grazed years, 2004-2011). The slight shifts in measurement periods (May 2004 – May 2011) for the soil stock calculations and the period (Jan 2004 – Dec 2010), is presumed to be insignificant in this comparison.

745 Results from both methods are within the range of literature values. Neeteson and Hassink (1997) found a N accumulation in SOM of $0\text{-}25 \text{ g N m}^{-2} \text{ y}^{-1}$ from two cattle grazed farms in the Netherlands, while Watson et al. (2007) reported a N storage in grazed Irish grasslands ranging from $10\text{-}15.2 \text{ g N m}^{-2} \text{ y}^{-1}$, depending on N inputs. Soil N storage assessed from soil measurements from a cut grassland close to our field, where plots were treated with cattle slurry, stored N over 6 years at a rate of $-2.17 \text{ g N m}^{-2} \text{ y}^{-1}$ in the top 10 cm, while, in the same experiment, a N loss was observed from mineral N and urea treatments (4.5 and $8.3 \text{ g N m}^{-1} \text{ y}^{-1}$, respectively) (Jones et al., 2007). In contrast, Schipper et al. 750 (2007) reported an average loss of $9.1 \text{ g N m}^{-2} \text{ y}^{-1}$ in the top 100cm from managed grasslands over 20 years in New Zealand.

755 The reason for the difference between methods (flux measurements vs sequential soil sampling) in our study might lie in a possible underestimation of losses from flux measurements. Uncertainties of our estimates are high, especially those from N losses. The largest absolute systematic uncertainty for the N balance was attributed to N leaching. Leaching was modelled for most years, whereas the model was validated using measured data from October 1st 2006 - March 30th 2008. The spatial variability of leaching was not considered in the measured data, as only one location has been used. The uncertainty



of the leaching estimate would therefore be reduced if the model could be validated with data measured
760 from several locations. The second highest systematic uncertainty was attributed to losses through N₂,
NO_x/NH₃ emission from excretion, NO_x/NH₃ emission from inorganic fertilization and inputs from
organic fertilization. Combined uncertainties from all components lead to a total systematic uncertainty
in the N balance of 2.1 g N m⁻² y⁻¹ (2004-2010).

765 4.2. Carbon balance

4.2.1. Net ecosystem exchange

We observed large variations of NEE between years, caused by varying management and environmental
conditions. The maximum uptake of CO₂ measured in our study is close to the upper range of NEE
reported for temperate grasslands (100 to 600 g C m⁻² y⁻¹, (IPCC, 1996). On an annual basis our
770 grassland site was a sink for atmospheric CO₂ in most years. NEE was only positive in 2004, which was
likely to be due to a combination of slurry spreading and a high livestock density. Generally, grazing
causes a very gradual impact on the CO₂ uptake as a part of the field is defoliated each day. The reduced
leaf area index (LAI) then leads to a reduced CO₂ uptake by plants. In addition to the reduced LAI,
grazing presents a source of CO₂ from animal respiration, thereby reducing the CO₂ sink of the
775 grassland (Levy et al., submitted). Indeed, annual NEE of all years correlated negatively with livestock
density if years with cuts were excluded. On average over the 9 years the magnitude of the NEE on our
grassland (-218.0 ± 154.5 g C m⁻² y⁻¹) was close to the average NEE measured in a comparison of nine
European grasslands over two years (240 ± 70 g C m⁻² y⁻¹) by Soussana et al. (2007) and comparable to
the CO₂ sink capacity of managed Irish grasslands measured by Byrne et al. (2007) (290 ± 50 g C m⁻² y⁻¹)
780 or Leahy et al. (2004) (257 g C m⁻² y⁻¹). Despite high variability over the 9 years, the average NEE
value was significantly different from zero (p < 0.05). The NEE represents the difference between the
gross primary production (GPP) and the total ecosystem respiration (TER), both influenced by
temperature, precipitation and management, though GPP is mainly controlled by PAR above a certain
temperature threshold. The range of the calculated annual GPP (-982 to -2163 g C m⁻² y⁻¹) and TER
785 (972 to 2183 g C m⁻² y⁻¹) from our field were within reported values for other managed grasslands.
Gilmanov et al. (2007) reported the GPP of 18 intensively managed European grasslands ranging from



467 to 1874 g C m⁻² y⁻¹ and TER ranging from 493 to 1541 g C m⁻² y⁻¹, while Mudge et al. (2011) reported values of 2000 g C m⁻² y⁻¹ for GPP and TER from a intensively grazed dairy pasture in New Zealand.

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4.2.2. Net biome production

The total C budget (=NBP), which includes all components of C import and export in addition to the CO₂ exchange, was negative on average, meaning that C was stored in the grassland over the 9 years. However, due to the high variability between years, NBP was not significantly different from zero (p = 0.05), suggesting that our site is carbon neutral. The average C storage value on our site (164 ± 140 g C m⁻² y⁻¹) is higher than most estimates reported in literature, but due to the high annual variation, still within the range of reported values; Soussana et al. (2007) reported C storage estimates from European grazed and cut grasslands of 104 ± 73 g C m⁻² y⁻¹, and Mudge et al. (2011) reported for a grazed and cut grassland in New Zealand fluxes of 59 ± 56 g C m⁻² y⁻¹ and 90 ± 56 g C m⁻² y⁻¹ in two consecutive years. NBP estimates from a Swiss grassland cut for silage was shown to sequester C at a rate of 147 ± 130 g C m⁻² y⁻¹ (Ammann et al., 2007), while estimates from a cut grassland in Germany was shown to vary from being a sink (-28 g C m⁻² y⁻¹) to being a source of C (+25 g C m⁻² y⁻¹), depending on years (Prescher et al., 2010). The inclusion of all C imports and exports lead to a weakening of the C sink strength assessed from NEE measurements in 5 years and even changed the grassland from being a sink to being a source in 2006. Due to the C export from harvest, C sequestration tends to be lower in cut systems. This is represented in the lower residual value of NPP in cut years (Figure 4a) compared to the residual value from grazed only years (Figure 4 b), whereas the residual value represents the C storage in the soil as well as the uncertainty of the budget. The grassland off-take from harvest weakened the annual C sink capacity assessed from the NEE by 51 % (2002) and 43 % (2003). However, it has to be kept in mind that the herbage yielded from cuts will end up as animal feed; C will be digested and respired off-site, releasing CO₂ to the atmosphere as well as being returned to the grassland as manure. It is likely that much of the organic C in the manure is decomposed and evolved to the atmosphere as CO₂, with very little being retained in soil because of the lack of contact between manure and soil: there is some evidence of this from two long-term grassland experiments in the UK (Hopkins et al., 2009).

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815 When the only management was grazing (2004-2010) the NEE showed to be a good proxy of the NBP. In those years the plant biomass was digested on-site by the grazing animals and thereby contributed to total ecosystem respiration

Only a small fraction of the digested C was incorporated into the body of the grazing animal as meat and wool, while the largest part was respired as CO₂ shortly after intake.

820 We estimated that CH₄ emissions from grazing animals were only 0.7 % of NPP. Methane emissions were also measured by eddy covariance technique over several months in 2010 on the same field (Dengel et al., 2011). By dividing CH₄ fluxes by the number of sheep in the field each day, Dengel et al. calculated CH₄ emissions per head of livestock as 7.4 kg CH₄ head⁻¹ y⁻¹ for sheep, which is close to the emission factor used in our budget of 7.6 kg CH₄ head⁻¹ y⁻¹ for ewes, showing that our estimates were
825 realistic. Methane emissions from slurry spreading were relatively high on specific days (up to 0.28 g C m⁻² d⁻¹, measured with chamber method), however, they were negligible on an annual basis as peaks only lasted for 2–3 days.

Carbon leaching from managed grasslands has not been reported in many studies. Kindler et al.
830 (2011) reported C leaching from various European ecosystems, whereas the measured data (2007) from our experimental field was part of the study. Our data (30.0 g C m⁻¹ y⁻¹, average of two locations as published in Kindler et al. (2011) were close to the average value (29.4 g C m⁻¹ y⁻¹) of the reported European grasslands, which showed a range of C losses of 6.5-42.5 g C m⁻¹ y⁻¹. Higher losses have been observed by McTiernan et al. (2001), who measured DOC export from grassland lysimeter plots treated
835 with N fertilizer and slurry over two months. Up-scaled to one year, they measured DOC loss between 25.2 and 70.8 g C m⁻² y⁻¹, all above what we measured in our study. Important factors controlling the magnitude of C leaching have shown to be drainage, the topsoil C/N ratio and the saturation of the subsoil's sorption capacity for organic C (Kindler et al., 2011; McTiernan et al., 2001). In waterlogged
840 soils the soil organic matter (SOM) decomposition and groundwater recharge tend to be reduced and thus the amount of C prone to leaching compared to that under more aerobic conditions associated with drainage. Although our field was drained more than 50 years ago, the drainage system does not operate very well, resulting in large puddles of standing water during prolonged periods of rain. The measured



845 data used for the budget were taken at one sampling point, which was not in a waterlogged area. Therefore our leaching estimates are highly uncertain and could be significantly lower and C exports overestimated. The spatial heterogeneity within the grassland field caused by uneven water management as well as faeces and urine patches requires to sample at more points in order to obtain a representative leaching value.

850 The systematic uncertainty of the C balance is mainly determined by the error of the CO₂ exchange, followed by the systematic uncertainty of the harvest export, organic fertilizer input and leaching losses. Combined uncertainties from all components lead to a total systematic uncertainty of the C balance of 18.3 g C m⁻² y⁻¹.

4.2.3 C sequestration

855 Unlike forests, most of the C stored by grasslands is contained within soil organic matter. Carbon sequestration in grasslands can therefore be either determined directly from measuring soil organic carbon changes or indirectly by measuring the net C balance flux. If measuring soil C changes, the internationally recommended practice in carbon accounting is to express soil C stocks to a depth of 30 cm (IPCC, 1997). However, as the bulk density often changes over time with land use, the soil C content per unit ground area to a fixed depth will also change even without any change in the mass fraction of C in dry soil. By using the ESM method this problem is avoided, by considering the whole soil mass present in the 0-60 cm soil layer. A comparison of the C storage calculated from the net C flux balance from 2004-2010 with C stock changes measured from soil sample analysis (Table 7) show that, although the flux balance estimated a C sequestration, while based on soil measurements C was lost, 865 neither value was significantly different from zero. A literature search by Soussana et al. (2010) showed that generally C sequestration calculations on grassland were lower if derived from SOC stock changes (average -5±30 g C m⁻² y⁻¹) compared to C flux balances (average -22±56 g C m⁻² y⁻¹), although these estimates were not significantly different from each other. However, in none of those reviewed studies were C flux and C stock change measured in the same field experiment. A reason for the discrepancy 870 between calculation methods in our study might lie in a possible underestimation of C exports in the



875 flux balance calculation, leading to an overestimation of C storage in the soil. One underestimated flux could be the export of DIC and DOC. Leaching was only measured in one year (2008), while values for remaining years were estimated using a simple regression model with an attributed high uncertainty of 30 % ($4.9 \text{ g C m}^{-2} \text{ y}^{-1}$ of average fluxes). Further uncertainty could be due to the use of only one
880 sampling location, which might not be representative of the whole field due to high spatial heterogeneity (see Sect. 4.1.2.). Indeed, Siemens (2003) hypothesized that the underestimation of C leaching from soils can explain a large part of the difference between atmosphere- and land-based estimates of the C uptake of European terrestrial ecosystems. Gapfilling can introduce uncertainties in the NEE data especially for years with low data capture. Furthermore, CO_2 losses from animal respiration could be
885 underestimated at times due to the animals moving out of the footprint of the EC mast. Using animal respiration values from chamber experiments of $12.1 \text{ g CO}_2 \text{ kg}^{-1} \text{ live weight d}^{-1}$ for cows and $11.7 \text{ g CO}_2 \text{ kg}^{-1} \text{ live weight d}^{-1}$ for sheep and lambs (Shane Troy, SRUC, personal communication), we estimated a maximum CO_2 loss from animal respiration of $53 \text{ g C m}^{-2} \text{ y}^{-1}$ (2002-2010) or $59 \text{ g C m}^{-2} \text{ y}^{-1}$ (2004-2010). So if we assume that all animal respiration has been missed by eddy covariance
890 measurements then the C sink estimated from NEE measurements would be reduced by 24 % (2002-2010) or 33 % (2004-2010). This theoretical maximum 33% reduction would reduce the net carbon balance to $\sim 122 \text{ g C m}^{-2} \text{ y}^{-1}$ (2004-2010).

In the literature, losses as well as storage of C at various rates have been reported from managed grasslands assessed from soil stock measurements. Soil stock measurements from our field are
895 comparable with the C sequestration of $10\text{-}30 \text{ g C m}^{-2} \text{ y}^{-1}$, measured from US rangelands (0-60 cm, Schuman, et al., 2002), while Watson et al. (2007) measured a C storage at $112\text{-}145 \text{ g C m}^{-2} \text{ y}^{-1}$ in the top 15 cm soil layer from a grazed Irish grassland. Bellamy et al. (2005) showed no evidence of increased C in the topsoil of grasslands in England and Wales and Hopkins et al. (2009) found no significant change of SOC over time in two UK long term experiments. Depending on the study,
900 managed grasslands in Belgium were shown to either lose ($90 \text{ g C m}^{-2} \text{ y}^{-1}$, Lettens et al., 2005a) or sequester carbon ($4.4 \text{ g C m}^{-2} \text{ y}^{-1}$ in 0-30 cm, Goidts and Van Wesemael, 2007; $22.5 \text{ g C m}^{-2} \text{ y}^{-1}$ in 0-30 cm, Lettens et al. 2005b). Schipper et al. (2007) reported losses of C from pastures in New Zealand over 20 years at an average rate of $106 \text{ g C m}^{-2} \text{ y}^{-1}$ (top 100 cm), whereas these losses were a result of an



earlier land use change from forestry. The above mentioned results are contrasting and inconclusive,
900 because observed C sinks in grasslands are the effect of land management or land use change prior to
the beginning of the C stock change measurement. Soussana et al (2014) concluded in a theoretical
study that grassland SOC sequestration has a strong potential to partly mitigate the GHG balance of
ruminant production systems at low grazing intensities, but not with intensive systems. Smith (2014)
examined evidence from repeated soil surveys, long term grassland experiments and simple mass
905 balance calculations and concluded that, although grasslands can act as C sinks, they cannot act as a
perpetual C sink and thus could not be used as an offset for GHG emissions.

4.3 Greenhouse gas budget

In the overall N and C budget N_2O and CH_4 emissions were negligible in terms of N and C losses from
910 the system (1 – 8 % of total N losses and 0.6 - 4.5 % of total C losses, respectively). However, in terms
of CO_2 equivalents, N_2O emissions as well as CH_4 emissions strongly affected the GHG budget. Since
the radiative forcing effect of N_2O is 298 times greater than that of CO_2 a relatively small emission of
 N_2O can exert a strong influence on the total radiative forcing budget of an ecosystem. Indeed, the sink
strength of the NEE was weakened by N_2O emissions by 29 % over all years. Methane emissions from
915 soil processes, manure input and animal excretion were negligible in terms of the C budget as well as in
the GHG budget. In contrast, enteric fermentation proved to be an important GHG source. The positive
correlation of CH_4 emissions with the stock density indicates that any changes in animal production will
have a major impact on the global CH_4 budget. The weakening of the GHG sink strength of the NEE by
 N_2O and CH_4 emissions, show the importance of those two gases in terms of global warming. Thus,
920 adapting the management of grasslands by adding fertilizer or manure to increase plant growth and thus
improve C sequestration in soils may increase N_2O emissions, while changing land use from cropland to
pasture in the attempt to reduce C losses from soils might lead to increased CH_4 losses from grazing
animals.

5. Conclusion

In our study only a small proportion of the N inputs from inorganic fertilizer and organic manure were



converted to animal outputs or stored in the soil, while the main part was lost through leaching and gaseous emissions. An improvement of the NUE would mean both an economic profit for the farmer as well as an environmental benefit. Estimates from flux budget calculations indicated that our grassland was sequestering C. However, although grasslands can act as C sinks, they can not act as a perpetual C sink and thus could not be used as an offset for GHG emissions. Instead, as it is easier and faster for soils to lose than to gain carbon, care must be taken to preserve C loss by management options, rather than trying to increase carbon stocks in grasslands. There was a discrepancy between soil stock measurements and flux budget calculations for the C as well as the N budget. The reason for the discrepancy between C storage estimates might lie in a possible underestimation of C exports such as leaching and animal respiration as well as the uncertainty due to gapfilling in the NEE data. The N budget storage might have been overestimated by the flux calculations through a possible overestimation of N losses, mainly through leaching as well as through N₂ and NO_xNH₄ emissions. Our data have shown that the information about the potential of managed grasslands to act as sinks or sources for GHG is important for mitigation and adaption purposes. High plant productivity, stimulated by fertilisation, resulted in high plant CO₂ fixation. However, increased N losses through N₂O emissions counteracted the benefits of C sequestration in terms of GHG emissions. Furthermore, CH₄ emissions from enteric fermentation largely reduced the positive effect of CO₂ uptake, especially in years where NEE rates were small. We therefore conclude that CO₂ exchange alone is not sufficient for the estimation of the GWP of a managed grassland ecosystem.

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**Table 1.** Average annual livestock densities [LSU ha⁻¹ y⁻¹].

	2002	2003	2004	2005	2006	2007	2008	2009	2010
Heifers	0.12	0.38	0.05	0.15	0.27	0	0	0	0
Ewes	0.14	0.16	0.82	0.56	0.51	0.68	0.68	0.61	0.53
Lambs	0	0	0.12	0.12	0.12	0.23	0.14	0.11	0.12
all animals	0.27	0.54	0.99	0.83	0.90	0.91	0.83	0.72	0.65

1450

Table 2. Weather characteristics of each measurement year.

	2002	2003	2004	2005	2006	2007	2008	2009	2010
Annual mean temperature [°C]	9.2	9.6	8.9	8.8	9.3	9.1	8.6	8.9	8.3
Maximum temperature [°C]	23.6	29.5	27.4	30.4	26.6	21.4	23.5	28.0	24.0
Minimum temperature [°C]	-5.1	-8.4	-4.9	-6.6	-5.5	-7.5	-5.8	-7.8	-10.3
Annual rainfall [mm]	1238	680	1169	1028	1120	904	1065	744	575
Soil water content [%]	36.9	31.0	40.3	45.2	36.6	37.7	41.5	39.4	-
Water filled pore space [%]	68.0	57.2	74.3	83.3	67.5	69.5	76.5	72.6	-
Length of growing season	180	196	156	177	151	186	193	242	226

1455

Table 3. Systematic uncertainties attributed to each budget component. Combined uncertainties were calculated according to simple Gaussian error propagation rules.

Nitrogen budget component	N [%]	Carbon budget component	C [%]
Mineral fertiliser	1		
Organic manure ^a	20	Organic manure ^a	20
Harvest ^b	16	Harvest ^b	11
Leaching	30	Leaching	30
Animal (wool and meat) ^c	12	Animal (wool and meat) ^a	12
Wet deposition	30	CH ₄ soil	160
Dry deposition ^d	80	CH ₄ enteric	20
N ₂ O	30	CH ₄ excretion	20
NO _x soil	30	CH ₄ organic	120
NH ₄ volatilisation	30		
NO _x volatilisation	50		
N ₂	30		

^acombined uncertainties of C and N analysis (17%) and volume spread (10%)^bcombined uncertainty of total C (4%) and N (12%) analysis and farmer's estimate in harvest amount (10%)^ccombined uncertainties from animal numbers (5%), animal weight (10%) and literature values for C and N content for meat and wool (3%)^dcombined uncertainty of DELTA sample analysis (7%) and variation of outputs from the four models (80%)

1460



Table 4. Nitrogen budget and balance for each measurement year and average values, confidence intervals at $p > 0.95$ (CI) and systematic uncertainties (uncert.) for 2002-2010 [$\text{g N m}^{-2} \text{y}^{-1}$]. Negative numbers represent uptake while positive numbers represent loss of N from this grassland ecosystem.

	2002	2003	2004	2005	2006	2007	2008	2009	2010	2002-2010		
										average	CI	uncert.
Organic fertilisation	0	0	-6.9	-15.8	0	0	0	0	0	-2.5	3.6	0.2
Inorganic fertilisation	-20.6	-16.0	-11.0	-17.3	-22.4	-17.3	-25.9	-25.0	-19.0	-19.4	3.1	0.2
Wet deposition	-0.4	-0.6	-0.6	-0.7	-0.6	-0.6	-0.5	-0.4	-0.5*	-0.5	0.1	0.2
Dry deposition	-0.5	-0.4	-0.3	-0.3	-0.2	-0.3	-0.2	-0.2	-0.3*	-0.3	0.1	0.2
Harvest	5.0	4.7	0	0	0	0	0	0	0	1.1	1.4	0.2
Meat	0.2	0.5	1.9	2.6	2.2	2.4	1.5	1.1	1.2	1.5	0.5	0.2
Wool	0	0	0.6	0.5	0.4	0.7	0.7	0.2	0.2	0.4	0.2	0.0
Leaching	15.0	0.1	0.1	4.6	10.6	4.2	5.6	2.6	5.3*	5.3	3.4	1.6
N ₂	3.7	2.2	1.3	1.7	2.8	3.0	3.3	4.1	2.8*	2.8	0.6	0.8
N ₂ O	1.1	0.1	0.1	0.4	0.9	1.3	0.8	0.4	0.4	0.6	0.3	0.2
NO _x (soil)	0.3	0.1	0	0.1	0.2	0.2	0.3	0.1	0.1	0.2	0.1	0.1
NO _x ,NH ₃ (inorg.fert.)	2.1	1.6	1.1	1.7	2.2	1.7	2.6	2.5	1.9	1.9	0.3	0.6
NO _x ,NH ₃ (org.fert.)	0	0	1.4	3.2	0	0	0	0	0	0.5	0.7	0.3
NO _x ,NH ₃ (excretion)	0.4	0.7	1.7	1.3	1.3	1.6	1.5	1.3	1.2	1.2	0.3	0.6
N balance	6.3	-7.0	-10.6	-17.9	-2.5	-3.1	-10.3	-13.2	-6.6	-7.2	4.6	2.1

*average value of 2002-2009



Table 5. Carbon budget and balance for each measurement year and average values, confidence intervals at $p > 0.95$ (CI) and systematic uncertainties (uncert.) for 2002-2010 [$\text{g C m}^{-2} \text{y}^{-1}$]. Negative numbers represent uptake, while positive numbers represent loss of C from the grassland ecosystem.

	2002	2003	2004	2005	2006	2007	2008	2009	2010	2002-2010		
											average	CI
GPP	-2162.9	-1982.0	-2111.4	-1662.4	-982.1	-1722.7	-1441.2	-1722.4	-2015.4	-1755.8	244.4	105.3
TER	1726.9	1725.9	2183.2	1638.5	972.1	1606.7	1324.0	1116.7	1547.0	1537.9	236.2	92.3
NPP	-1081.5	-991.0	-1055.7	-831.2	-491.1	-861.3	-720.6	-861.2	-1007.7	-877.9	122.2	-52.8
CO ₂ (NEE)	-436.0	-256.1	71.8	-24.0	-10.0	-115.9	-117.1	-605.7	-468.4	-217.9	154.5	80.0
Organic fert.	0.0	0.0	-55.4	-171.8	0.0	0.0	0.0	0.0	0.0	-25.2	37.8	5.0
Harvest	270.6	169.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	48.9	65.5	5.4
Meat	0.9	3.0	11.5	15.8	13.1	14.5	9.1	6.4	7.3	9.1	3.4	1.1
Wool	0.0	0.0	1.7	1.5	1.3	2.1	2.0	0.7	0.5	1.1	0.5	0.1
Leaching*	25.1	7.0	22.1	18.7	19.4	15.4	17.0	6.8	16.4*	16.4	4.3	4.9
CH ₄ (organic fert.)	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
CH ₄ (soil)	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0	0.0	0.1
CH ₄ (excretion)	0.0	0.1	0.2	0.1	0.2	0.2	0.1	0.1	0.1	0.1	0.0	0.0
CH ₄ (enteric ferm.)	1.5	3.2	5.7	4.8	5.2	5.2	4.8	4.1	3.8	4.3	0.8	0.9
C balance (NBP)	-137.8	-73.3	57.7	-154.7	29.3	-78.6	-84.0	-587.6	-440.3	-163.2	139.5	15.9

*average value of 2002-2009



Table 6. Annual N₂O exchange, total N input by fertiliser (mineral and organic) and N₂O emission factors, expressed as percentage of total N inputs in 2002-2010.

	N ₂ O flux [g N m ⁻² y ⁻¹]	Total N input [g N m ⁻² y ⁻¹]	EF [%]
2002	1.14	20.60	5.5
2003	0.14	15.98	0.9
2004	0.11	11.00	0.6
2005	0.36	17.25	1.1
2006	0.88	22.43	3.9
2007	1.25	17.25	7.2
2008	0.84	25.93	3.2
2009	0.41	24.95	1.6
2010	0.35	18.98	1.9

Table 7. N and C budget (g N or C m⁻² y⁻¹) over 7 years based on repeated soil N and C stock inventories (May 2004 and May 2011) and flux budget calculations (January 2004 - December 2010). Soil stock changes are based on a soil mass of 800 kg m⁻², which corresponds to approximately 60 cm depth. The flux budgets are averages for the years 2004 – 2010 (Table 4 & 5). Numbers in brackets represent confidence intervals. Negative numbers are sinks.

	N balance	C balance
soil stock change	-4.51 (2.64)	29.08 (38.19)
flux budget	-9.20 (4.10)	-180.7 (180)

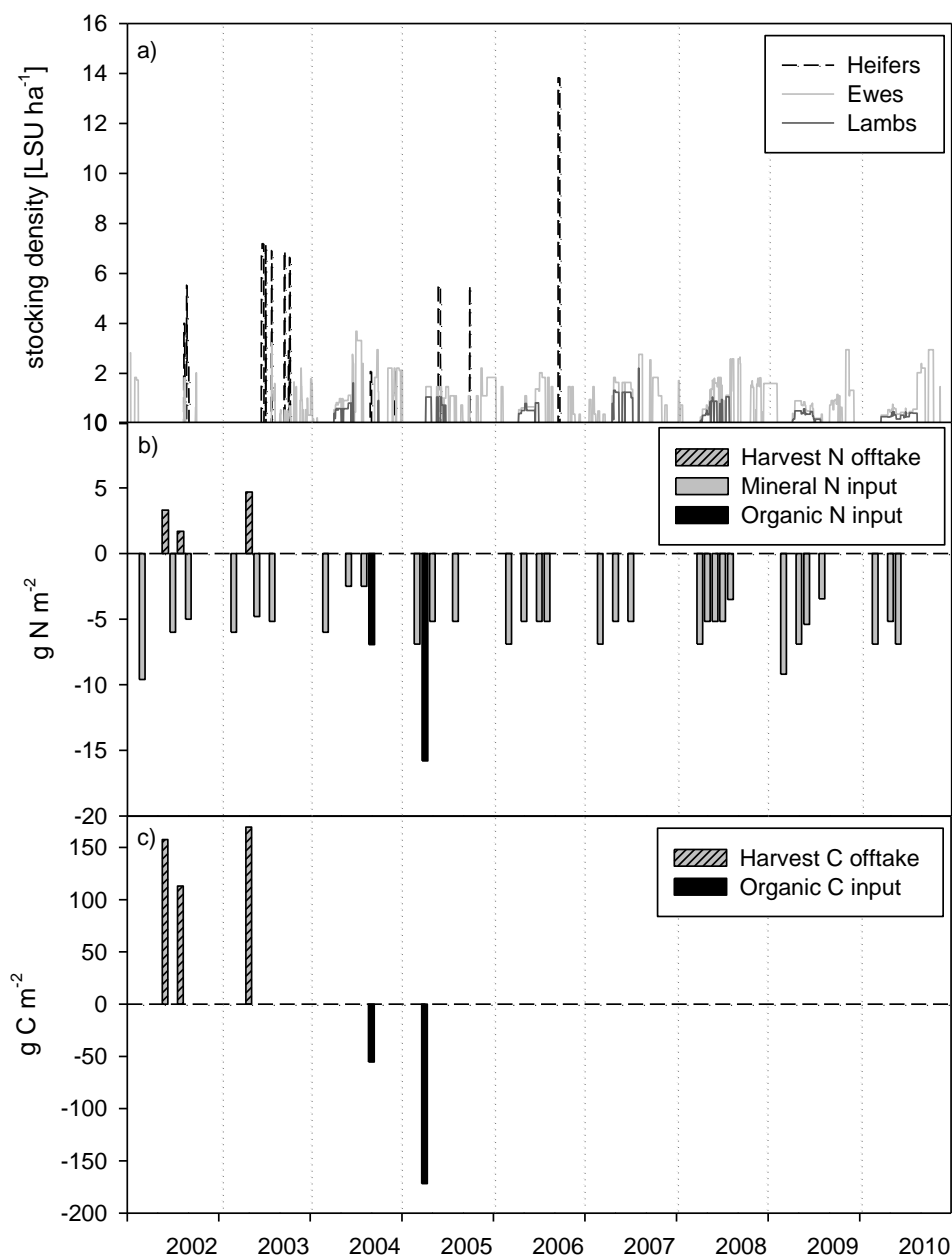


Figure 1. Livestock density (c), nitrogen (c) and carbon (b) input and export from inorganic and organic fertiliser and harvest from 2002-2010.

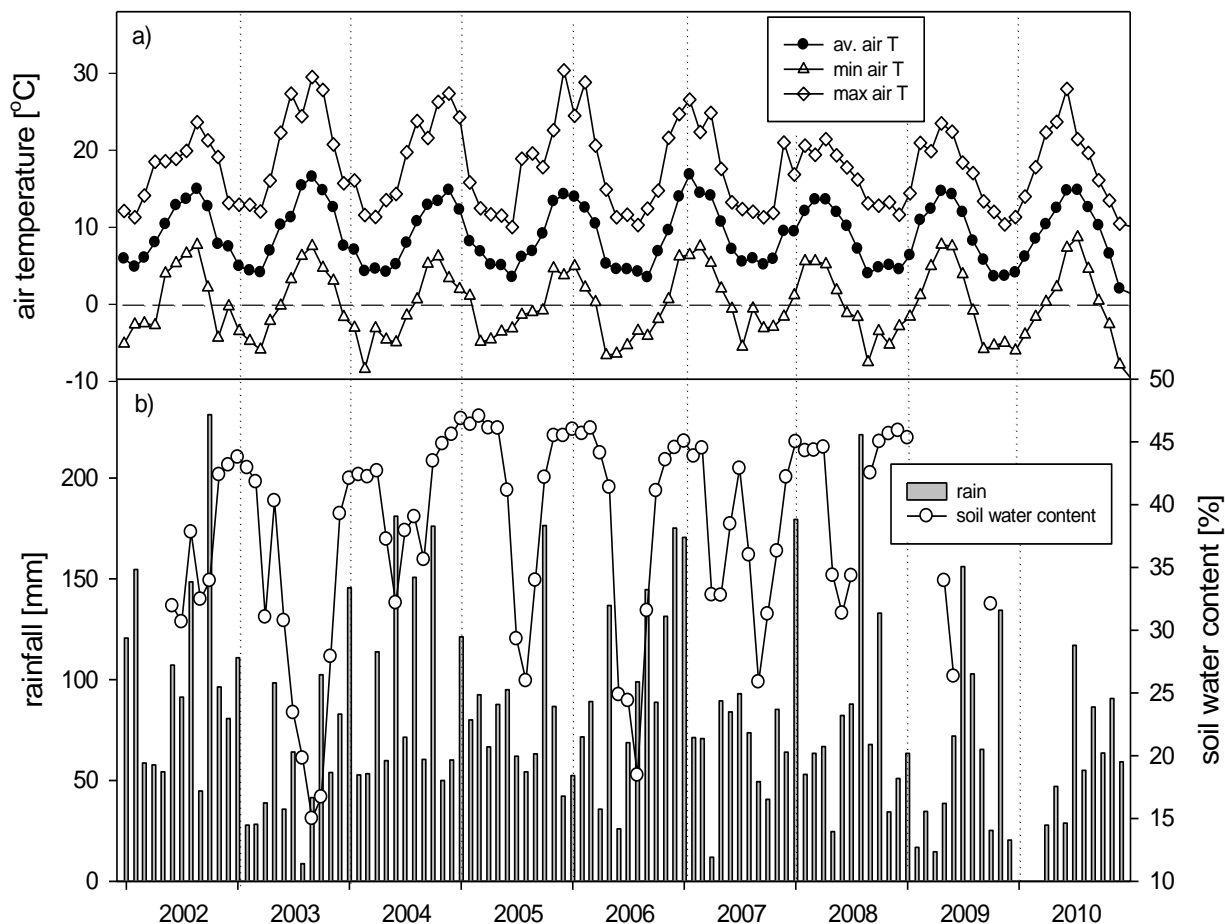


Figure 2. Maximum, minimum and average monthly air temperature, derived from daily averages (a) and monthly cumulative rainfall and soil water content (b) from 2002-2010.

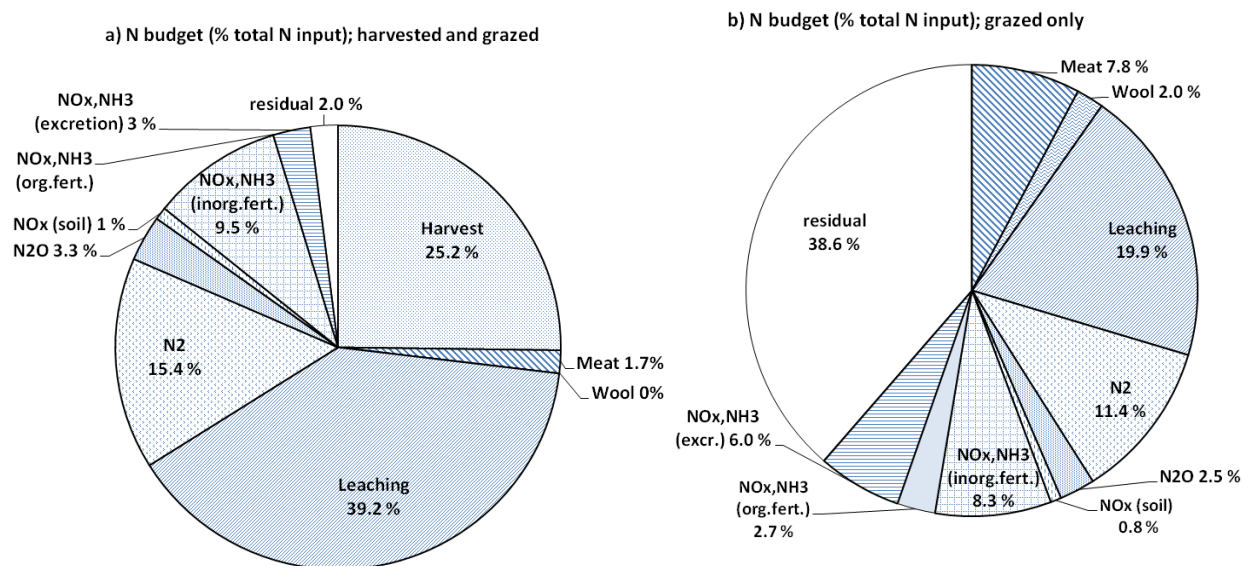


Figure 3. Mean annual nitrogen budget for Easter Bush, showing the fate of total N input (fertiliser and deposition) in (a) years when harvested for silage (2002 and 2003) and (b) in years when only grazing took place (2004–2010). The residual term includes all the error in the budget calculation, as well as any net accumulation of soil organic nitrogen.

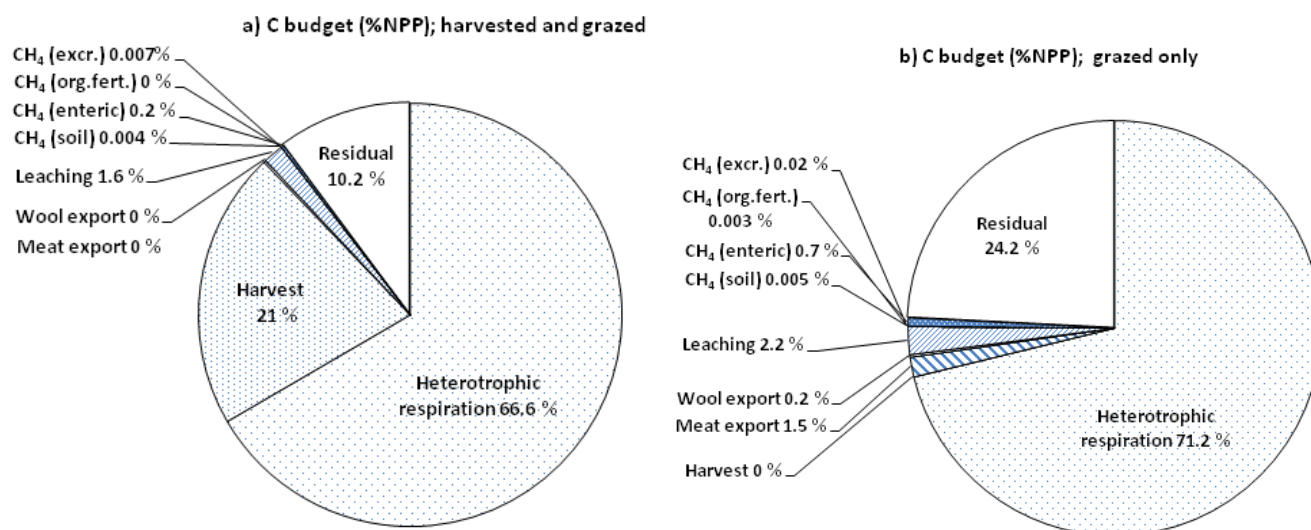


Figure 4. Mean annual carbon budget for Easter Bush, showing the fate of net primary productivity (NPP) in (a) years when harvested for silage (2002 and 2003) and (b) in years when only grazing took place (2004–2010). Heterotrophic respiration includes the respiration of soil microbes, cows and sheep. The residual term includes all the error in the budget calculation, as well as any net accumulation of soil organic carbon.

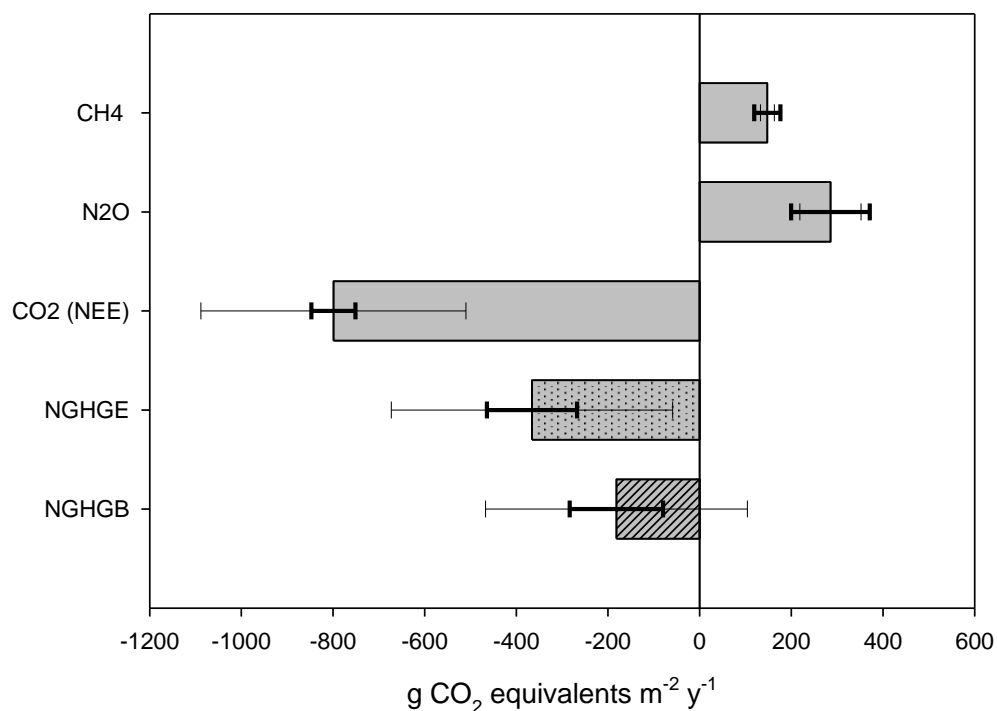


Figure 5. Average greenhouse gas fluxes, net GHG exchange (NGHGE) and attributed net GHG balance (NGHGB, includes $FC_{\text{org fert.}}$, FC_{animal} , FC_{leaching} , FC_{harvest}) for 2002-2010. Positive values correspond to losses and negative values to storage of greenhouse gases to and from the grassland system, respectively. The CH₄ component comprises CH₄ fluxes from enteric fermentation, animal excretion, slurry application and soil exchange, while the N₂O component is the N₂O flux from the soil. Global warming potentials of 298 and 25 were used for N₂O and CH₄ respectively, using a time horizon of 100 yrs (IPCC, 2013). Thin error bars represent variations (confidence intervals at $p > 0.95$) between years, while thick error bars represent the systematic uncertainty of each value.