Sheep Excrement Increases Mass of Greenhouse Gases Emissions from Soil Growing Two Forage Crop and Multi-Cutting Reduces Intensity

Xinzhou Zhao, Lina Shi, Shanning Lou, Jiao Ning, Yarong Guo, Qianmin Jia and Fujiang Hou *

State Key Laboratory of Grassland Agro-Ecosystems, Key Laboratory of Grassland Livestock Industry Innovation Ministry of Agriculture and Rural Affairs, College of Pastoral Agriculture Science and Technology, Lanzhou University, Lanzhou 730020, China; lzhou_xzc@lzu.edu.cn (X.Z.); shln17@lzu.edu.cn (L.S.);
loushn09@lzu.edu.cn (S.L.); ningj10@lzu.edu.cn (J.N.); guyry15@lzu.edu.cn (Y.G.); guqm@lzu.edu.cn (Q.J.)

* Correspondence: cyhouf@lzu.edu.cn

Citation: Zhao, X.; Shi, L.; Lou, S.; Ning, J.; Guo, Y.; Jia, Q.; Hou, F. Sheep Excrement Increases Mass of Greenhouse Gases Emissions from Soil Growing Two Forage Crop and Multi-Cutting Reduces Intensity. Agriculture 2021, 11, 238. https://doi.org/10.3390/agriculture11030238

Academic Editors: Dominika Lewicka-Szczechak

Received: 17 February 2021 Accepted: 8 March 2021 Published: 11 March 2021

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Abstract: To explore the effects of multi-cutting and sheep excrement on greenhouse gas (GHG) emissions from grassland ecosystems which simulate grazing livestock to a certain extent, spring wheat (Triticum aestivum L., var. Yongliang 15) and common vetch (Vicia sativa L., var. Lanjian 3) were planted in pot experiments in an inland arid region in 2019. Four treatments were conducted with eight replicates: plants without sheep excrement and cutting (CK), plants with multi-cutting (MC), plants with sheep excrement (SE), and plants with multi-cutting and sheep excrement (CE). The results showed that the carbon dioxide (CO2) emission of common vetch with CE significantly was higher than that with MC at the earlier and later branching stages (p < 0.05). That of spring wheat with CE was significantly higher than that with MC at the later tillering stage (p < 0.05). Nitrogen oxide (NOx) emissions of the two forage crops with SE rose significantly more than those with MC at both stages (p < 0.05). Methane (CH4) of both forage crops with SE changed from absorption to emission (p < 0.05). Soil NO3-N content of both forages significantly increased with SE compared with MC (p < 0.05), while soil NH4-N content did not change significantly. Sheep excrement changed the CH4 sink into a CH4 source of the soil growing the two forage crops and increased the emissions of CO2 and NOx, whereas multi-cutting significantly reduced the GHG intensity of forage crops mostly by promoting the growth of the two forage crops. Future studies are suggested to identify the spatiotemporal effects of cutting and sheep excrement on GHG emissions to improve the prediction of future climate impacts from grazing activities.

Keywords: soil moisture; soil temperature; root biomass; root/shoot ratio; nitrate nitrogen (NO3-N); ammonium nitrogen (NH4-N)

1. Introduction

The rapid increase in greenhouse gas (GHG) emissions, mainly including carbon dioxide (CO2), nitrous oxide (N2O), and methane (CH4) [1], has aggravated global warming, which has become a concern for society. Globally, it is estimated that 5–20% of CO2, 15–30% of CH4, and 80–90% of N2O come from soil each year [2,3]. Grassland represents the largest area of terrestrial ecosystems, accounting for about half of surface land, and is one of the main sources and sinks of GHGs [4]. Ruminant livestock, which mostly graze on grassland in the world, contribute to 18% of global agricultural GHG emissions [5]. Therefore, it is of great significance for agricultural production, food security, and ecological protection to study multi-cutting and excrement in the grazing process and their relationship with greenhouse gas emissions, as well as to explore how GHG emissions can be reduced through the grazing management of different forage crop grasslands.
Multi-cutting, which is often considered as a simulated grazing method by cutting the stem of forage crops frequently with harvesters [6,7], can not only significantly reduce aboveground of plant but also affect soil nutrient cycling, thus altering soil GHG emissions [8]. The excrement of grazing herbivores produces CH₄, N₂O, and other GHGs through the action of soil microorganisms, in turn, further exacerbating GHG emissions [9]. Grazing livestock can inhibit CO₂ emissions and reduce them by 15.91% [10,11]. On the other hand, CO₂ emissions from pastures with grazing livestock were determined to be higher than areas with grazing bans [12]. The influence of cutting and excrement on N₂O emissions mainly comes from the feces and urine of herbivores. Sheep excrement increases the emissions of N₂O by increasing the concentration of NH₄⁺-N and NO₃⁻-N in soil [13]. Nevertheless, some studies found that cutting may reduce soil water content and organic matter, thus reducing N₂O emissions [14]. Although grazing livestock is beneficial for restraining soil CO₂ and N₂O emissions, it comes at the cost of declining plant productivity and soil fertility [11]. Grazing livestock inhibits CH₄ emissions [10]. On the other hand, there is no significant correlation between grazing livestock and CH₄ emissions in grasslands (p > 0.05) [15]. Furthermore, the cutting frequency of forage crop was often overlooked, despite being an important factor in forage harvesting or simulated grazing. There have been some important reports on GHG emission inventories from sources of ruminant agriculture based on emission factor methods [16,17] or remote sensing [18], and more field trials are worth implementing in localized emission factor studies. Moreover, studies on the emissions of the three dominant components of GHGs, global warming potential, and the mechanisms of different forage crops should be conducted under a combination of ingestion, excrement, and trampling of grazing livestock, which could be simulated by multi-cutting and animal excrements to a certain extent [19–21]. Therefore, we hypothesize that the interaction between multi-cutting and livestock excrement may significantly alter the global warming potential of different forage crops, which may exceed the influence of single factors.

To test the hypothesis, common vetch and spring wheat were selected for pot experiments in an inland arid region. The objectives of the study included assessing (1) the effects of multi-cutting and sheep excrement on the emissions of major GHG components from the soil of the two forage crops, (2) the global warming potential and GHG emission intensity of the two forage crops, and (3) the relationships among forage crops, soil, global warming trend, and GHG emission intensity. The results could be used as a reference for other related studies such as the effects resulting from the interaction of multiple factors in simulated grazing on greenhouse gas emissions.

2. Materials and Methods

2.1. Site Description

This experiment was conducted in the Linze Grassland Agriculture Station (LGAS) of Lanzhou University (100°02’ east (E), 39°15’ north (N), 1390 m above sea level), which is located in the core area of Heihe Oasis in Gansu province, an inland arid region in China. It is classified as a secondary salinization meadow with a temperate continental climate. The annual mean temperature is about 7.7 °C, with 3042 h of annual sunshine, and the annual accumulated temperature ≥0 °C is 3548 °C, while the annual accumulated temperature ≥10 °C is 3026 °C, with a frost-free period of approximately 170 days. The annual mean precipitation is 118.4 mm, more than 70% of which takes place from May to September, and the annual potential evaporation is 1830.4 mm. The soil was categorized as Aquisalix on the basis of its 0.7–0.9% salt content [1,22]; soil pH was 8.06 ± 0.09, the soil electrical conductivity was 2.87 ± 0.03 mS/cm, soil organic carbon content was 8.06 ± 0.31 g/kg, soil total nitrogen content was 0.87 ± 0.02 g/kg, and soil total phosphorus content was 0.53 ± 0.01 g/kg (unpublished data).
2.2. Experimental Design

The pot size was 30 cm in diameter and 20 cm in height. The soil was acquired from local farms, with approximately 10.8 kg per pot. Spring wheat (Triticum aestivum L., var. Yongliang 15) and common vetch (Vicia sativa L., var. Lanjian 3) were selected as the experimental forage crops. Prior to the start of the experiment, we soaked the seeds in water to promote germination. Seeds with inconsistent germination were removed following two rounds of selection. Subsequently, germinated seedlings of spring wheat plants and common vetch plants were transplanted into pots according to different varieties. Each pot of spring wheat contained eight plants, whereas each pot of common vetch contained five plants. The basal fertilizer applied in the experiment was mainly nitrogen and phosphorus, with a dosage of 22.5 g N/m² and 7.5 g P/m², respectively.

Four treatments were conducted with eight replicates: plants without sheep excrement and cutting (CK), plants with multi-cutting (MC), plants with sheep excrement (SE), and plants with multi-cutting and sheep excrement (CE). When the forage crops grew to approximately 25 cm, the treatment was carried out, in which the stubble was 5 cm in each case under MC and CE. The addition of sheep excrement included a mixture of feces and urine. The fecal input was 4 g/plot, which was determined according to the yield and digestibility of forage crop in the field experiment [23], and the urine application amount was 25 mL/plot, which was determined according to the nitrogen content of forage crop, the nitrogen deposition of grazing sheep, and the feces nitrogen content [24]. For the spring wheat, it was applied at the tillering stage on 3 and 23 June, and for the common vetch, it was applied at the branching stage on 10 June and 9 July. The spring wheat received the first cutting and the second cutting on 3 and 23 June, respectively. The common vetch received the first cutting and the second cutting on 10 June and 9 July, respectively.

2.3. Sampling and Analysis

CH₄, CO₂, and N₂O were collected and analyzed using a static chamber and gas chromatograph [25]. The chamber body featured an opaque darkroom with a size of 30 × 30 × 30 cm. Except for the opening at the bottom, all other sides were sealed, and the outer layer was sealed with a foam plate approximately 2.5 cm thick. The top of the chamber was connected with a thin tube approximately 30 cm long with an inside diameter of 1 cm. Before collecting the gas, we prepared a tray approximately 50 cm long and wide, and we moved the pot to the tray and covered the gas chamber before filling the tray with enough water to seal the chamber. When collecting greenhouse gases, three-way valves and 50-mL syringes were used to collect gases from the static chamber and transfer them to an air bag with a capacity of approximately 500 mL, which was then taken back to the room to analyze the concentrations of the gas components. The gases were collected at 0, 10, 20, and 30 min after each treatment between 9:00 a.m. and 11:00 a.m. The gas emission fluxes collected during this period are considered to represent the mean gas emissions per day. The static chamber collected four air bags at a time, and each set of collections was repeated three times. The concentrations of CH₄, CO₂, and N₂O were analyzed and measured using a gas chromatograph (GC-2014CC1188530256, produced by Shimadzu Company of Japan). The emission fluxes of the three gases were calculated as follows [26,27]:

\[
F(GHG) = \rho \times \frac{V}{R^2 \times \pi} \times \frac{P}{P_0} \times \frac{T_0}{T} \times \frac{dCt}{dt} \times 24
\]

where \(F(GHG)\) represents the emission value of each greenhouse gas (mg·m⁻²·day⁻¹ or μg·m⁻²·day⁻¹), \(\rho\) represents the density of the gas to be measured under standard conditions (CH₄ = 0.717 kg·m⁻³, CO₂ = 1.977 kg·m⁻³, N₂O = 1.978 kg·m⁻³), \(V\) represents volume of the static chamber (0.027 m³), \(R\) represents radius of the pot (0.15 m), \(P\) represents the atmospheric pressure at the sampling site (kPa), and \(P_0\) represents the standard atmospheric pressure (\(P_0 = 101.325\) kPa). \(T_0\) represents the absolute temperature under standard conditions (\(T_0 = 273.15\) K), \(T\) represents the temperature (K), and \(\frac{dCc}{dt}\) represents the change rate of gas concentration over time.
To facilitate the comparison of greenhouse gas emissions under different resources, CH₄ and N₂O were converted into CO₂ equivalents, i.e., the global warming potential (GWP). On a 100-year scale, the GWP for CH₄ and N₂O represents 25× and 298× CO₂ equivalents, respectively [28]. The greenhouse gas emission intensity (GHGI) was used to measure the emissions per unit of forage yield. The formulas for GWP and GHGI are as follows:

\[
GWP = E(CO₂) + 298 \times E(N₂O) + 25 \times E(CH₄)
\]

\[
E(GHG) = \sum_{i=1}^{n} \left( \frac{F_2 + F_1}{2} \right) \times (d_2 - d_1)
\]

\[
GHGI = \frac{GWP}{Y_h}
\]

where GWP represents the seasonal global warming potential (kg·ha⁻¹), E(CO₂), E(N₂O), E(CH₄), and E(GHG) represent the seasonal cumulative emissions, F₂ and F₁ represent the emission of each greenhouse gas after two treatments, respectively, d₂−d₁ represent the days between two treatments, GHGI represents the greenhouse gas emission intensity (kg·t⁻¹), and Y_h represents the dry matter of each forage crop (t·ha⁻¹) [29].

At the end of each treatment, all of the forage crops in the pot were harvested, and the total dry matter (tDM) and shoot biomass (SB) were measured in the laboratory. The soil around the root system in the pot was collected and screened with an 80-mesh sieve (diameter of the sieve hole, 0.18 mm) to remove impurities such as plant roots, sand, and gravel. Air-drying was carried out under natural conditions for determination of the physical and chemical properties of the soil and other conventional indicators. Soil nitrate nitrogen (NO₃⁻N) was determined using the indophenol blue colorimetric method. Soil ammonium nitrogen (NH₄⁺-N) was determined by ultraviolet (UV) spectrophotometry using a Cary 60 UV–Vis spectrometer. The root system of forage crops was washed using a root washing net to remove surface soil and was baked at 65 °C until constant weight to obtain the root biomass (RB). In addition, the soil temperature (ST) and soil moisture (SM) were monitored.

2.4. Statistical Analyses

IBM SPSS Statistics 22 was used for data processing, correlation analysis, and significance analysis. Microsoft Excel 2019 and GraphPad Prism 8 were used for plotting. The structural equation model (SEM) was developed using IBM SPSS AMOS 24. One-way analysis of variance (ANOVA) was used to study the effects of different treatments on greenhouse gas emissions, soil properties, and biomass. Duncan’s test was used for multiple comparison analysis, and the significance level was set to \( p = 0.05 \). Pearson’s correlation coefficient was used to analyze the correlation between two variables. Multivariate analysis of variance with repeated measurements for the effects of variety (V), cutting (C), excrement (E), and their interactions (V × C, V × E, C × E, and V × C × E) on each of indicators were determined using the F-test (Table 1).

| V  | C  | E  | V × C | V × E | C × E | V × C × E |
|----|----|----|-------|-------|-------|----------|
| RB | 172.986 | <0.001 | 0.270 | 0.611 | 0.326 | 0.576 | 2.215 | 0.156 | 2.215 | 0.156 | 0.108 | 0.747 | 0.051 | 0.825 |
| SB | 37.989 | <0.001 | 118.373 | <0.001 | 3.208 | 0.092 | 0.567 | 0.462 | 0.567 | 0.462 | 0.057 | 0.814 | 0.272 | 0.609 |
| RB/SB | 135.819 | <0.001 | 73.642 | <0.001 | 2.567 | 0.129 | 10.694 | <0.01 | 10.694 | <0.01 | 1.064 | 0.318 | 0.000 | 0.983 |
| tDM | 3.447 | 0.082 | 121.917 | <0.001 | 3.923 | 0.065 | 0.075 | 0.788 | 0.075 | 0.788 | 0.116 | 0.738 | 0.360 | 0.557 |
| SM | 0.176 | 0.680 | 1.038 | 0.323 | 1.189 | 0.292 | 0.211 | 0.652 | 0.211 | 0.652 | 0.011 | 0.918 | 0.055 | 0.818 |
| ST | 2.260 | 0.152 | 0.202 | 0.659 | 3.711 | 0.072 | 0.230 | 0.638 | 0.230 | 0.638 | 0.647 | 0.433 | 0.123 | 0.730 |
| NO₃⁻N | 1.300 | 0.271 | 0.085 | 0.775 | 38.503 | <0.001 | 0.705 | 0.413 | 0.705 | 0.413 | 0.102 | 0.754 | 0.006 | 0.942 |
3. Results

The multivariate analysis of variance showed that the three factors and their interaction have significant effects on the biomass of forage crops, soil physicochemical properties, and greenhouse gas emissions ($p < 0.05$) (Table 1).

3.1. Biomass of Forage Crops and Soil Physicochemical Properties

3.1.1. Aboveground Biomass and Root Biomass of Forage Crops

There were significant SB differences among the four kinds of treatments and two forage crops ($p < 0.05$) (Figure 1). The SB of common vetch under MC and CE at the first time point was significantly higher than that under other treatments, where the SB of common vetch under CE was highest, reaching 4.10 g-plot$^{-1}$, which was 75% higher than that of common vetch under CK. There was no significant difference in the SB of common vetch between CE and CK ($p > 0.05$). At the first time point, the RB difference of common vetch under each treatment was not significant ($p > 0.05$). At the second time point, the SB of common vetch under SE was significantly higher than that under other treatments ($p < 0.05$), reaching 4.64 g-plot$^{-1}$, while that under CK was significantly higher than that under MC and CE ($p < 0.05$). The RB under SE was significantly higher than that under other treatments ($p < 0.05$), reaching 0.74 g-plot$^{-1}$.

The SB of spring wheat under CE at the first time point was significantly higher than that under other treatments ($p < 0.05$), reaching 2.76 g-plot$^{-1}$, an increase of 24% with respect to MC and 49% with respect to SE. The RB of spring wheat under SE and CK at the first time point was significantly higher than that under MC and CE ($p < 0.05$). The SB of spring wheat under SE and CK at the second time point was significantly higher than that under MC and CE ($p < 0.05$). At this time, the RB of spring wheat was in the order SE > CK > MC > CE, where that under SE was significantly greater than that under other treatments ($p < 0.05$).

The RB/SE under MC and CE was lower than that under CK and SE for different crops in different periods ($p < 0.05$), which was due to cutting (Figure 1). At the second time point, the RB/SE of spring wheat under SE was significantly higher than that under CK ($p < 0.05$), while there was no significant difference between the RB/SE under SE and CK at other stages ($p > 0.05$). The RB/SE under MC and CE increased for both forage crops. At the second time point, the RB/SE for common vetch under MC increased by 18% and that under CE increased by 14% with respect to the first time point. In the same period, the RB/SE of common vetch under SE decreased by 5% compared with the first time point. At the second time point, the RB/SE of spring wheat under MC increased by 12% with respect to the first time point, and the RB/SE under CE increased by 21%. At this stage, the RB/SE of common vetch under CK was 7% lower compared to the first time point.
3.1.2. Soil Physicochemical Properties

Multi-cutting and sheep excrement had significant effects on soil available nitrogen ($p < 0.05$) (Figure 2). The addition of sheep excrement significantly increased the content of NO$_3^-$-N ($p < 0.05$). In both soils for common vetch and spring wheat, the content of NO$_3^-$-N under SE and CE was significantly higher than that under CK and MC ($p < 0.05$). Specifically, the content of soil NO$_3^-$-N of common vetch under CE was the highest ($p < 0.05$). The soil NO$_3^-$-N of spring wheat under SE was the highest ($p < 0.05$). However, there was no significant difference in NO$_3^-$-N between CK and MC and between SE and CE in both soils ($p > 0.05$), respectively. The content of soil NH$_4^+$-N of common vetch was significantly different between CK and SE or CE ($p < 0.05$), and the content of soil NH$_4^+$-N under CE was the highest ($p < 0.05$). There was no significant difference in soil NH$_4^+$-N of spring wheat among the four treatments ($p > 0.05$).

At the first time point, the SM of common vetch and spring wheat did not differ significantly among treatments ($p > 0.05$). The ST of common vetch under CE and SE was significantly higher than that under MC and CK ($p < 0.05$). The ST of spring wheat under SE was significantly higher than that under MC and CE ($p < 0.05$), while that under MC was significantly higher than that under CE ($p < 0.05$). At the second time point, there was a significant difference in ST between SE and MC or CE of common vetch and spring wheat ($p < 0.05$). At this stage, the SM of common vetch under CE was significantly higher than that under MC and SE ($p < 0.05$). The SM of spring wheat under CE was significantly higher than that under MC and SE ($p < 0.05$) (Figure 3).
Figure 2. Soil nitrate nitrogen (NO$_3^-$-N) and soil ammonium nitrogen (NH$_4^+$-N) content of the two forage crops. The significant differences were labeled with letters using Duncan’s notation, from largest to smallest: a, b, c. If there is no significant difference between two values, the same or overlapping letters are used for both values.
Figure 3. Soil moisture (SM) and soil temperature (ST) of the two forage crops. The significant differences were labeled with letters using Duncan's notation, from largest to smallest: a, b, c. If there is no significant difference between two values, the same or overlapping letters are used for both values.

3.2. Greenhouse Gas (GHG) Emissions

3.2.1. CO₂ Emissions

CO₂ Emission from Common Vetch

At the first time point, the mean CO₂ emissions of common vetch under SE were the highest and were 180% and 159% higher than those under CK and MC, respectively (p < 0.05) (Figure 4). The mean CO₂ emissions of common vetch under CE increased by 170% with respect to CK, which was 7.082 g·m⁻²·day⁻¹, with no significant difference compared to SE (p > 0.05). The mean CO₂ emission of common vetch under MC was 2.839 g·m⁻²·day⁻¹, which was not significantly different from that under CK (p > 0.05). There was no significant difference in mean CO₂ emissions among CE, SE, and MC (p > 0.05). At the second time point, the mean CO₂ emissions of common vetch under SE and CE were the largest (p < 0.05), increasing by 29% and 27% with respect to CK, respectively. There was a significant difference between MC and other treatments (p < 0.05), with emissions under MC decreasing by 8% with respect to CK to 4.894 g·m⁻²·day⁻¹.
CO₂ Emission from Spring Wheat

At the first time point, there was no significant effect of the four treatments on the mean CO₂ emissions of spring wheat ($p > 0.05$). The mean CO₂ emission of spring wheat under MC was 5% lower than that under CK ($p < 0.05$), which was 3.297 g·m⁻²·day⁻¹. At the second time point, the mean CO₂ emissions of spring wheat under CE and SE were 36% and 35% higher than under CK, respectively. There was no significant difference between MC and CK ($p > 0.05$) (Figure 4).

3.2.2. N₂O Emissions

N₂O Emission from Common Vetch

CE and SE had a significant effect on mean N₂O emissions for the two forage crops ($p < 0.05$) (Figure 4). At the first time point, the mean N₂O emissions of common vetch under SE and CE were the highest ($p < 0.05$), which were 672% and 668% higher compared with CK, respectively. There was no significant difference in N₂O emissions between CE and SE ($p > 0.05$). The mean N₂O emission of common vetch under MC was 0.435 mg·m⁻²·day⁻¹, which was not significantly different with respect to CK ($p > 0.05$). At the
second time point, the mean N₂O emissions of common vetch under SE were the highest, which were 690% higher compared with CK. Emissions under CE were 633% higher than under CK. There was no significant difference between MC and CK (p > 0.05).

N₂O Emission from Spring Wheat

At the first time point, the mean N₂O emissions of spring wheat under CE were the highest (p < 0.05), which were 645% higher than under CK. There was no significant difference between CE and SE (p > 0.05). The mean N₂O emissions of spring wheat under MC were 0.284 mg·m⁻²·day⁻¹, which were not significantly different with respect to CK (p > 0.05). At the second time point, the mean N₂O emissions of spring wheat under CE were highest at 2.351 mg·m⁻²·day⁻¹, 633% higher than with CK (p < 0.05). The mean N₂O emissions of spring wheat under SE were not significantly different from those under CE (p > 0.05). There was no significant difference in N₂O emissions between MC and CK (p > 0.05) (Figure 4).

3.2.3. CH₄ Emissions

CH₄ Emission from Common Vetch

The mean emissions of common vetch and the CH₄ emissions of spring wheat under CK and MC were negative, showing an absorption state. The mean emissions of common vetch and the CH₄ emissions of spring wheat under CE and SE were positive, showing an emission state (Figure 4). At the first time point, the mean CH₄ emissions of common vetch under CE were the highest (p < 0.05). There was no significant difference between CE and SE (p > 0.05). The mean CH₄ emissions of common vetch under MC were −0.252 mg·m⁻²·day⁻¹, which were not significantly different with respect to CK (p > 0.05). At the second time point, the mean CH₄ emissions of common vetch under SE were the highest (p < 0.05). There was no significant difference between SE and CE and between MC and CK (p > 0.05).

CH₄ Emission from Spring Wheat

At the first time point, the mean CH₄ emissions of spring wheat under SE were the highest (p < 0.05). There was no significant difference between CE and SE (p > 0.05). The mean CH₄ emissions of spring wheat under MC were −0.396 mg·m⁻²·day⁻¹, which were not significantly different with respect to CK (p > 0.05). At the second time point, the mean CH₄ emissions of spring wheat under CE were the largest (p < 0.05), while they were 0.142 mg·m⁻²·day⁻¹ for spring wheat under SE, with no significant difference with respect to CE (p > 0.05). There was no significant difference between MC and CE (p > 0.05) (Figure 4).

3.3. Total Seasonal Global Warming Potential (GWP)

The seasonal GWP was individually determined for all three greenhouse gas emissions, and the total seasonal GWP (GWPₙ) was then calculated (Figure 5). The results showed that the GWP of soil under SE and CE was significantly higher than that under CK and MC (p < 0.05).

The GWP of common vetch under CE was 2310.22 kg·ha⁻¹. The GWP of common vetch under SE was 2379.80 kg·ha⁻¹, while the GWP of common vetch under MC was 1197.96 kg·ha⁻¹. The GWP of spring wheat under SE was the highest (p < 0.05), and the GWP under CE was the second highest (p < 0.05). Furthermore, there was no difference between MC and CK with values of 1167.17 and 1195.56 kg·ha⁻¹, respectively (p > 0.05), 9% and 2% less than that under CE.
3.4. GHG Emission Intensity (GHGI).

The GHGI values for both forage crops under SE were significantly higher than those under other treatments \((p < 0.05)\) (Figure 5), which were 80.7% and 30.5% higher than when under CK. The GHGI value of common vetch under CE was significantly higher than when under MC \((p < 0.05)\), increasing by 19.4% compared with CK. The GHGI values of common vetch and spring wheat under MC were significantly lower than those under other treatments \((p < 0.05)\).

3.5. Structural Equation Model (SEM)

There were significantly positive correlations between the SB and tDM \((r \geq 0.8^{**}, p < 0.01)\) and between the CO\(_2\) and CH\(_4\) emissions \((r \geq 0.8^{**}, p < 0.01)\) of common vetch (Figure 6). There was a strong positive correlation between the NO\(_3^-\)-N content of common vetch and emissions of NO\(_2\) or CH\(_4\) \((r \geq 0.8^{**}, p < 0.01)\). There was a strong positive correlation between the CO\(_2\) emissions of common vetch and the two other greenhouse gases \((r \geq 0.8^{**}, p < 0.01)\).

There was also a positive correlation between the NO\(_3^-\)-N of spring wheat and N\(_2\)O or CH\(_4\) emissions \((r \geq 0.7^*, p < 0.05)\). In addition, there was a very strong positive correlation among the three GHGs of spring wheat \((r \geq 0.8^{**}, p < 0.01)\).

The SEM showed that forage crop variety was the main factor determining RB increase. Increases in SM, ST, and NO\(_3^-\)-N also led to increases in RB (Figure 7). The determining factors that added to SB were multi-cutting and forage crop variety. In addition, an increase in SB could also be related to sheep excrement. A decrease in SM could also lead to an increase in SB. The increases in CO\(_2\), N\(_2\)O, and CH\(_4\) emissions could mainly be achieved through increases in NO\(_3^-\)-N and NH\(_4^+\)-N, while NO\(_3^-\)-N and NH\(_4^+\)-N could be controlled as a function of sheep excrement. The increase in NH\(_4^+\)-N had a more significant impact on the increase in CO\(_2\) emissions, whereas the increase in NO\(_3^-\)-N had a more significant impact on the increase in N\(_2\)O and CH\(_4\) emissions.
Figure 6. Matrix of Pearson correlation coefficients for common vetch and spring wheat. Red rectangles represent positive correlation coefficients and blue rectangles represent negative correlation coefficients. $|r| \geq 0.7^*$ and $p < 0.05$ indicate significant correlation; $|r| \geq 0.8^{**}$ and $p < 0.01$ indicate a very significant correlation.
Figure 7. Structural equation model based on data in this study. The structural equation model shows the direct and indirect effects of soil and biomass factors on CO₂, CH₄, and N₂O emissions. The number at each arrow is the normalized path coefficient. The R² value above the variables represents the proportion of variance in the interpretation of endogenous variables (χ² = 1.967; degrees of freedom (df) = 12; p = 0.161).

4. Discussion

4.1. Effects of Multi-Cutting and Sheep Excrement on Forage Biomass and Dry Matter

Compared with the control, the dry matter of common vetch and spring wheat under multi-cutting treatment was increased by 56.39% and 70.40%, respectively. This is due to cutting increasing the absorption of inorganic nitrogen in the soil by forage crop roots. In the early and middle stages after the cutting of forage crops, nitrogen absorbed by roots from the soil is transported to the aboveground parts for their growth; however, in the later stages, this nitrogen is mainly stored in the root system [30]. As an exogenous nitrogen fertilizer, the added sheep excrement has a promoting effect on the growth of graminaceous crops, while the addition of sheep excrement has no effect on the root protein of common vetch due to its inherent biological nitrogen fixation [31,32]. We found that the RB/SB under MC and SE showed some differences. Multi-cutting increased the RB/SB and the cumulative effect of root biomass, which promoted the growth of forage, and it also improved the adversity performance of forage crops.

4.2. Effects of Multi-Cutting and Sheep Excrement on Physicochemical Properties of Soil

Grassland soil nutrients undergo N cycling in grassland ecosystems, while grassland resource management and ecological benefit adjustment have non-negligible roles [33]. Furthermore, the available nitrogen content of soil affects the soil microorganism quantity, litter decomposition rate, and soil organic carbon and nutrient long-term accumulation [34], which affect greenhouse gas emissions. As an important process of the nitrogen cycle, mineralization can transform organic nitrogen in the soil to become rapidly available for absorption and use by plants [35]. In this experiment, sheep excrement increased soil NO₃⁻-N content. The soil NO₃⁻-N content of common vetch was significantly and strongly positively correlated with GWPᵢ, and the cumulative temperature of common vetch was
significantly and strongly correlated with GWP. Nitrogen mainly exists in various forms such as organic nitrogen, NO₃⁻N, and NH₄⁺-N in livestock manure, which mainly includes undigested forage, water, livestock metabolites, and microorganisms [36]. In the process of feces degradation, organic nitrogen in sheep feces is decomposed by microorganisms and NH₄⁺ is produced. NH₄⁺ is oxidized into NO₃⁻-N under the action of related microorganisms and enzymes and it enters the soil, becoming an important part of the soil’s available nitrogen [37]. The addition of sheep excrement increases the available carbon in the soil, promotes the decomposition of carbon in the soil, increases the soil nitrogen supply level, and then increases the net nitrogen mineralization rate. The nitrogen in livestock urine is mainly in the form of urea, which mainly includes nitrogen-containing organic compounds, urea, and inorganic salts [38]. Studies have shown that the pH of urine of grazing livestock is around 8.5. The addition of urine changes the soil pH in the short term, accelerates the decomposition of soil organic matter to a certain extent, and increases the content of available nitrogen in the soil. Urea contained in urine breaks down to produce ammonia oxide, which is then nitrated to nitrate and exists in the soil in the form of NO₃⁻-N [39]. The addition of urine and feces in the treatment did not have a significant impact on soil NH₄⁺-N, and it is speculated that the aerobic conditions during the test stimulated the nitrification of NH₄⁺-N to a certain extent. In addition, different responses to SM and ST of the two herbage species were obtained in this study. This may have been because the nitrogen fixation of legumes increases the SM, while cutting reduces the density of aboveground forage crops. Therefore, the SM under MC at the first time point was significantly lower than that under other treatments. The ST of spring wheat under CK and SE was significantly higher than that under MC and CE, possibly because at the first time point, spring wheat was at the nutrient stage of absorbing sheep excreta, which was related to the endothermic effect during this process.

4.3. Effects of Multi-Cutting and Sheep Excrement on GHG Emissions

The addition of sheep excrement increased the CH₄, CO₂, and N₂O emissions in the soil, and cutting had little effect on GHG emissions. The conversion of a CH₄ sink into a CH₄ source in the soil may have been due to the fact that at the early stage of excrement addition, the soil is covered with fresh livestock manure with a high water content. At this point, the addition of feces provides available carbon sources for microbial activities and accelerates the consumption of oxygen in the soil, thus forming an anaerobic microenvironment. The activity of methane-oxidizing bacteria is then enhanced, and biodegradable organic compounds in feces are decomposed to produce a large amount of CH₄. At the same time, water and nitrogen contained in the urine interact to promote the production of CH₄ and inhibit the absorption of CH₄ by the soil [25,40]. Therefore, reducing the intensity of grassland use and afforestation can increase the absorption capacity of soil with respect to atmospheric methane [41]. After sheep excrement is added, urea hydrolysis produces CO₂ under the action of urease, and a large number of hydroxide ions are produced in this process, such that the soil pH increases in the short term. The alkaline environment is conducive to the decomposition of soil organic matter, promoting the respiration of soil microorganisms and increasing soil respiration [42]. Carbohydrates in sheep excrement can provide carbon sources for microbial activity, leading to enhanced soil respiration [43]. Studies in alpine meadow grazing areas showed that in the 0.3% and 2.9% of grassland corresponding to fecal and urine spots, respectively, there were 0.1% and 0.6% higher CO₂ emissions, respectively, compared with the total alpine meadow [40]. The increase in N₂O emissions was mainly due to the hydrolysis of inorganic substances in sheep excrement and the mineralization of organic compounds into ammonia nitrogen, which produced N₂O under nitrification and denitrification [44]. The anaerobic environment in the soil provided favorable conditions for denitrification, and the nitrogen in sheep excrement provided sufficient substrate for denitrification [27,45,46]. Studies have shown that fecal and urine spots emit N₂O into the atmosphere, with nitrogen accounting for 3.2% and 3.8% of the total nitrogen in excrement, respectively [47–49]. The nitrogen
morphism and water content in the pot experiments were fixed and controllable, while terrestrial ecosystems are highly sensitive to temperature changes [50]. However, our SEM found that soil temperature and moisture did not respond significantly to cutting and did not have a decisive effect on greenhouse gas emissions, which may be due to the fact that our sampling time was in summer.

5. Conclusions

This study showed that sheep excrement changed the CH₄ sink into a CH₄ source and increased the CO₂ and N₂O emissions in the soil of common vetch and spring wheat, and it contributed 94.65% and 44.58% to GWP in both soils at the same time, whereas multicutting significantly reduced the GHGI of the two forage crops to 37.51% and 35.09%, respectively, and promoted the growth of forage crops. Future studies should consider the spatiotemporal effects of cutting and sheep excrement on GHG emissions in order to improve the prediction of future climate impacts resulting from grazing activities.

Author Contributions: Conceptualization, F.H.; methodology, L.S. and F.H.; investigation, L.S., J.N. and Y.G.; formal analysis, X.Z. and L.S.; data curation, X.Z., L.S. and S.L.; writing—original draft preparation, X.Z. and L.S.; writing—review and editing, X.Z., Q.J. and F.H.; visualization, X.Z.; supervision, F.H.; project administration, F.H.; funding acquisition, F.H. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the Strategic Priority Research Program of the Chinese Academy of Sciences (XDA20100102); the National Key Basic Research Program of China (2014CB138706), and the Program for Innovative Research Team of Chinese Ministry of Education (IRT_17R50).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: We are very grateful to Shenghua Chang for assistance with laboratory analysis. We thank William Irvine and Angela H. from English Editing of MDPI for their improvement of the language of the manuscript. We deeply appreciate the directions of the two anonymous reviewers and editors. We sincerely thank the editors Aileen Song and Aniko Erdes for their help.

Conflicts of Interest: The authors declare no conflict of interest.

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