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Productivity and Topsoil Quality of Young and Old Permanent Grassland: An On-Farm Comparison

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Abstract: Renewing agricultural grasslands for improved yields and forage quality generally involves eliminating standing vegetation with herbicides, ploughing and reseeded. However, grassland renewal may negatively affect soil quality and related ecosystem services. On clay soil in the north of the Netherlands, we measured grass productivity and soil chemical parameters of ‘young’ (5–15 years since last grassland renewal) and ‘old’ (>20 years since last grassland renewal) permanent grasslands, located as pairs at 10 different dairy farms. We found no significant difference with old permanent grassland in herbage dry matter yield and fertilizer nitrogen (N) response, whereas herbage N yield was lower in young permanent grassland. Moreover, the young grassland soil contained less soil organic matter (SOM), soil organic carbon (C) and soil organic N compared to the old grassland soil. Grass productivity was positively correlated with SOM and related parameters such as soil organic C, soil organic N and potentially mineralizable N. We conclude that on clay soils with 70% desirable grasses (i.e., *Lolium perenne* and *Phleum pratense*) or more, the presumed yield benefit of grassland renewal is offset by a loss of soil quality (SOM and N-total). The current practice of renewing grassland after 10 years without considering the botanical composition, is counter-productive and not sustainable.

Keywords: grassland renewal; grassland productivity; nitrogen; production system; soil functions; soil organic matter; sustainable land-use

1. Introduction

Grasslands are among the most widespread ecosystems on earth [1]. In the EU-28 in 2011, permanent grassland covered 61 million hectares i.e., about 35% of the total utilized agricultural area [2]. Grassland is an important land use for sustainable agriculture and delivers ecosystem services such as water retention, protection of water quality, sequestration of soil carbon, protection of soil from erosion, support of biodiversity and provision of forage for animals [3–5].

The grassland area in the Netherlands is approximately one million ha, which is mainly used for dairy farming [6]. In 2015, 75% of this area was classified as permanent grassland, i.e., grassland that has not been included in crop rotation for at least five years [6]. Due to the presumed value of

permanent grassland, the member states of the European Union have stipulated that the ratio of areas of permanent grassland to the total agricultural area will not decrease by more than 5% compared to a reference ratio in 2015 (EU Regulation N° 1307/2013). Recently, the Horizon 2020 SUPER-G project (Sustainable PERmanent Grassland Systems and Policies; Grant Agreement no. 774124) has started with, among others, as a goal to increase the understanding of the importance and functioning of permanent grassland in Europe [7].

On clay soils in the Netherlands, permanent grasslands are renewed (i.e., destroyed by herbicides, ploughed and reseeded) on average once every 10 years [8–11]. Grasslands can be renewed to improve levelling and soil physical quality such as drainage capacity and to counteract soil compaction [9,12]. However, the main reason for grassland renewal is to improve the productivity and feeding quality of the harvested grass and to remove unwanted species such as couch grass (*Elymus repens* L.) [13–16]. In the Netherlands, dairy farmers are advised to renew their grassland when the percentage of perennial ryegrass (*Lolium perenne* L.) drops below 50% or when the sward comprises more than 10% couch grass [17]. Another motivation for grassland renewal is to introduce the most recently developed ryegrass varieties for increasing feed production and quality [12,15,17–20]. Plant breeding programs have accomplished a theoretical yield increase of 3% per decade, plus enhanced digestibility, and many studies have shown that, with this increase, reseeding is economically attractive for farmers [18,19,21–23]. However, these studies are based on the comparison of older grass varieties with new varieties sown at the same time on the same field. In studies where newly sown varieties were compared with older grassland on one location, thus adding possible soil quality effects of reseeding to the genetic improvement, no significant difference in grass productivity was found [10,24–27]. This difference in results between studies that compared grass varieties sown at the same time and studies that compared old permanent grassland to young grassland at one location can possibly be explained by loss of soil quality, and specifically a loss of soil organic matter (SOM) and related parameters such as soil organic carbon (C), soil organic nitrogen (N) and potentially mineralizable N (PMN) caused by the decomposition of SOM after ploughing [28–30]. However, there is some evidence that possible negative effects of soil quality on the productivity of grassland with grassland renewal mainly occurs during the first years (“the years of depression”), after which productivity increases as the grassland enters the “permanent grassland phase” [31].

In the international terminology for grazing lands and grazing animals, no distinction is made between young and old permanent grassland [32]. The European Grassland Federation defines permanent grassland as grassland that has not been renewed after destruction by ploughing or spraying (herbicide) for 10 years or longer [33]. There are a number of different definitions of young and old grassland [11,13,25,34–38] ranging in age from 1–15 years for young and 10–100 years for old grassland. Moreover, Hoogerkamp [31] mentioned that the “years of depression” i.e., the decrease in productivity of grassland after renewal, usually starts 1 to 4 years after sowing and lasts for several years. Therefore in this study, we set the minimum age of young grassland at 5 years and the maximum age of young grassland at 15 years. The minimum age of old grassland was set at 20 years since grassland renewal.

The first objective of the current study was to compare grass productivity and soil chemical quality of young (5–15 years) with old (>20 years) permanent grassland on clay soil. The second objective was to investigate the relationship between grassland age, soil chemical quality and grass productivity. It is not feasible practically to test these effects in a standard (short-term) experiment, due to the long time-scale during which the expected changes in productivity and soil quality take place. Therefore, the experiment was carried out on-farm and multi-locational, where we could make use of the large range in grassland age that exists in practice.

To this end, we selected 10 dairy farms on marine clay soil in the north of the Netherlands. At each farm, two grasslands were selected, one young and one old grassland. To minimize the effect of botanical composition on grass productivity, only fields that contained at least 70% desirable grasses (i.e., *Lolium perenne* and *Phleum pratense*), were included in this study.

We hypothesized that:

- i. young grasslands (5–15 years since grassland renewal) have higher grass productivity than old grasslands (>20 years since grassland renewal) since in young grasslands the most recently developed ryegrass varieties are used and the so-called “years of depression” (a temporary period in which the productivity is lower) have come to an end.
- ii. the soil chemical quality, expressed as SOM, soil organic C and N and potentially mineralizable N (PMN), of young grassland soils, is lower than the soil chemical quality of old grassland soils.
- iii. grass productivity parameters of young and old grassland are positively correlated with SOM and related soil parameters.

2. Materials and Methods

2.1. Experimental Setup

The study was conducted in 2014, at 10 conventional dairy farms on marine clay soil in the north of the Netherlands. At each farm, a young and an old grassland were selected as a pair. Grassland ages were recorded, except for a few old grasslands where the exact age was not known and was set at 30 years. Other criteria for the selection of the grasslands were: marine clay soil, well-drained, use of the most recently developed commercially available grass varieties at the time of renewal (based on the VCU testing program for the Dutch Variety List in which the available grass varieties are ranked each year), no visual soil compaction, no clover seeded and containing at least 70% desirable grasses (i.e., *Lolium perenne* and *Phleum pratense*) based on visual estimation in March 2014. These criteria were applied to minimize the differences between the grasslands.

Botanical composition was measured in June 2014 according to the Braun-Blanquet method [39]. The number of plant species was counted, and the percentage of soil covered by desirable grasses, other grasses, legumes and herbs was assessed. Following interviews with the farmers the management and fertilization history of the grasslands was registered.

On each grassland, an experimental plot was laid out in February 2014 on a representative place in the center of the field. The layout of the experimental plot of 15 m × 9 m followed the experimental set up described by Van Eekeren et al. [40] and Deru et al. [41]. Briefly, the first 10 m of the experimental plot were split into three fertilization sub-plots of 10 m × 3 m. These sub-plots were fertilized with 0, 150 or 300 kg N ha⁻¹ yr⁻¹ with calcium ammonium nitrate (CAN, 27% N) and ample P, K and S to prevent nutrient limitations other than N, in the form of triple superphosphate and potassium sulfate granulate, at a rate of 60 kg P₂O₅ ha⁻¹, 195 kg SO₄ ha⁻¹ and 180 kg K₂O ha⁻¹ after the first grass harvest and 40 kg P₂O₅ ha⁻¹, 130 kg SO₄ ha⁻¹ and 120 kg K₂O ha⁻¹ after the second harvest. Of the annual CAN fertilization, 33% was given before the first harvest, 27% directly after the first harvest, 23% directly after the second harvest and the remaining 17% directly after the third harvest. The remaining 5 m × 9 m sub-plot was not fertilized and was used to measure soil quality parameters.

2.2. Grass Productivity

Grass was harvested four times in 2014 (8–9 May, 18–19 June, 6–7 August and 1–2 October), according to the average harvesting regime of the farms. Plots were cut at a stubble height of 6 cm, using a “Haldrup” small-plot harvester (J. Haldrup a/s, Løgstør, Denmark). Grass was weighed and sampled for dry matter (DM) and total N analysis. After drying for 48 hours at 70 °C the grass was analyzed for residual moisture content (at 105 °C) to determine DM. The Kjeldahl method [42] was used for determining total N content.

2.3. Soil Sampling and Analysis

Soil samples were taken on 29 and 30 April 2014. On each unfertilized 5 m × 9 m sub-plot, a field-moist bulk sample of 70 cores from the topsoil (0–10 cm; Ø 2.3 cm) was collected randomly,

sieved through 1 cm mesh, homogenized and stored at 4 °C until analysis. The composite sample was split into sub-samples before analysis.

For each site, soil from the 0–10 cm composite sample was oven-dried at 40 °C prior to analysis of soil acidity (pH-KCl), SOM, total carbon (C-total), total nitrogen (N-total), total phosphate (P-Al) and potassium (K) in solution. Soil dry matter content was determined after oven-drying of approximately 30 g of the composite sample (in duplicate) at 105 °C. Soil acidity of the oven-dried samples was measured in 1 M KCl (pH-KCl). SOM was determined by loss-on-ignition [43]. C-total was measured by incineration of dry material at 1150 °C, after which the CO₂ produced was determined by an infra-red detector (LECO Corporation, St. Joseph, Mich., USA). For determination of N-total, evolved gasses after incineration were reduced to N₂ and detected with a thermal conductivity detector (LECO Corporation, St. Joseph, Mich., USA). C:N-ratio was calculated by dividing C-total by N-total. The carbon percentage of the SOM was calculated by dividing C-total by SOM. P-Al was determined using standard methods [44]. K in solution was determined by flame photometry after soil extraction with HCl (0.1 M) and oxalic acid (0.5 M) in a 1:10 mass to volume M:V ratio, and filtration [44].

Hot-water extractable carbon (HWC) was analyzed according to the method of Ghani et al. [45]. Field moist soil was extracted for 16 h at 80 °C. PMN was determined by anaerobic (waterlogged) incubation of 16-gram soil in 40 ml water for 7 days at 40 °C [46,47]. These incubation conditions are optimal for quick mineralization of organic matter by anaerobic bacteria and prevent the conversion of released NH₄⁺ to NO₃⁻ (nitrification). After 1 week, NH₄⁺ contents were determined by Segmented Flow Analysis.

Soil particle analysis was done by a Beckman Coulter LS-230 laser with software version 3.01 and firmware version 2.02. Particle analysis was performed after removal of CaCO₃ with 1 M HCl, and addition of de-ionized water, and of 30% H₂O₂ to remove organic matter, at 80–95 °C. The actual soil water content per site was determined in the 5–10 cm layer below the soil surface, in three undisturbed ring samples containing 100 cm³ soil per unfertilized plot. Samples were weighed, oven-dried at 70 °C, and re-weighed to determine moisture content.

2.4. Weather Conditions

In 2014, the average temperature in the north of the Netherlands (measuring station Leeuwarden) was 11.1 °C and a mean rainfall of 742 mm [48]. This was relatively warm and dry compared to the 30-year averages for Leeuwarden of 9.5 °C and 870 mm [49].

2.5. Calculations and Statistical Analysis

For each grassland four grass productivity parameters were modelled according to Van Eekeren et al. [40] based on the dry matter yield (DMY) and nitrogen yield (NY) for each fertilization sub-plot (0, 150 and 300 kg N ha⁻¹), using the following equation:

$$Y = \alpha_i + \beta_i * N \text{ application} + \epsilon_{ij}$$

with:

- Y DM or N yield;
- α_i DM or N yield intercept, representing the DM or N yield of field *i* at 0 kg N ha⁻¹;
- β_i DM or N yield response to N fertilizer; the slope of the linear correlation between DM or N yield and N application;
- ϵ_{ij} random field effect, $\epsilon_{ij} \sim N(0, \sigma_v^2)$.

We assumed that the variance in yield was similar for the pertinent fertilization levels and those correlations between DM and N yield with N application were linear. The grassland DM yield and N yield without N fertilization (DMY_{N0} and NY_{N0}, respectively) were calculated as the intercept (α) of the described equation. The grassland DMY response and N yield response to fertilizer N application

(DMY-res and NY-res) were calculated as the slope (β) of the equation. These modelled grassland productivity parameters were used in further statistical analyses.

When data were not normally distributed, the grass productivity and soil parameters were log-transformed to assure normal distribution. Analysis of variance (ANOVA) was performed to test for significance of the differences between young and old grasslands, using Genstat software (18th edition, VSN International, UK). Each of the 10 farms comprised a young and an old grassland, the factor ‘farm’ was therefore used as a block factor in the ANOVA structure. Further data analysis was performed with Matlab (version 8.6 R2015b, The Mathworks). Pearson correlations and their significance were calculated for all possible parameter pairs. Furthermore, the linkage between grass productivity parameters and soil properties were explored using cross-validated stepwise regression with permutation tests to determine model significance, as described in Van Eekeren et al. [40] and Baars et al. [50].

3. Results

3.1. Field Properties

The average age, defined as the period in years since the last reseeding, was 9 years for the young and 25 years for the old grasslands (Table 1). There was no significant difference in soil texture between young and old grasslands. In 2013, the year before the experiment was implemented, the young and the old grasslands received on average 298 and 255 kg available N ha⁻¹ from a combination of slurry manure and artificial fertilizer, respectively, and these doses did not differ significantly. At the farm level, management in grazing and mowing frequency and fertilization was comparable between the young and the old grasslands.

Table 1. Characteristics and soil parameters in the 0–10 cm depth of the young (n = 10) and old (n = 10) grasslands on marine clay soil. Means, standard deviations, P-values (italic when <0.05). Desirable grasses are: *Lolium perenne* and *Phleum pratense*.

Parameter	Unit	Young Grassland		Old Grassland		P-Value
		Mean	s.d.	Mean	s.d.	
Grass age	years without cultivation	9	4	25	4	<0.001
Botanical composition						
<i>Desirable grasses</i>	%	87.1	9.6	83.2	11.2	0.439
<i>Other grasses</i>	%	11.6	9.6	13.8	11.3	0.634
<i>Legumes</i>	%	0.6	0.7	0.4	0.7	0.572
<i>Herbs</i>	%	0.8	0.8	2.6	3.8	0.111
Soil texture						
Clay	(% particles < 2 μ m)	31	8	29	7	0.301
Silt	(% particles 2–50 μ m)	47	4	46	5	0.074
Sand	(% particles > 50 μ m)	22	8	25	10	0.530
Soil organic matter (SOM)	%	10.7	3.3	13.3	2.2	0.031
C-total	g C. kg dry soil ⁻¹	45.2	18	61.0	12	0.002
C percentage of SOM	%	41.4	4.0	45.6	2.1	0.002
Hot-water extractable carbon (HWC)	μ g C. g dry soil ⁻¹	2412	1042	3356	593	0.002
N-total	g N. kg dry soil ⁻¹	4.82	1.7	6.28	1.2	<0.001
C:N-ratio		9.28	0.4	9.70	0.3	0.009
Potentially mineralizable N (PMN)	mg N. kg dry soil ⁻¹	175	53	232	32	0.001
Ca-total	kg Ca. ha ⁻¹	56.3	39	64.8	46	0.545
P-Al	mg P ₂ O ₅ .100 g soil ⁻¹	26.3	9	38.5	17	0.074
K in solution		36.0	15	33.5	8	0.552
pH-KCL		5.67	0.6	5.50	0.7	0.410
Soil water content	vol %	28.6	3.2	31.7	2.8	0.007

3.2. Botanical Composition

On average, the soil cover of the desirable grasses *Lolium perenne* and *Phleum pratense* was 85%, with no significant difference between young and old grasslands (Table 1). Other (not desirable) grasses present were *Agrostis stolonifera*, *Alopecurus pratensis*, *Bromus hordeaceus*, *Elymus repens*, *Holcus lanatus*, *Poa annua*, *Poa pratensis* and *Poa trivialis*. Next to these grasses, a small amount of *Trifolium repens*

(1%) and some dicotyledons (2%) were found in both the young and the old grasslands. Table S1. (Supplementary Materials) gives an overview of the grasses, legumes and dicotyledons that were found in the different fields.

3.3. Grass Productivity

The measured data on DMY and NY resulted in a highly significant fit of the model $Y = \alpha_i + \beta_i * N \text{ application} + \epsilon_{ij}$ on all locations ($P < 0.001$; average $R^2 = 0.96$).

The grassland DMY without N fertilization (DMY_{N0}), the grassland DMY response to fertilizer N application (DMY-res) and the N yield response to fertilizer N application (NY-res) were not significantly different between the young and old grasslands (Figure 1). However, the N yield without N fertilization (NY_{N0}) was significantly ($P < 0.05$) lower in the young grasslands compared to the old grasslands (Figure 1). We found a negative correlation between DMY_{N0} and DMY-res ($r = -0.76$; $P = 0.001$, Figure 2). Between NY_{N0} and NY-res, no significant correlation was found ($r = -0.01$; Supplementary Material Table S3).

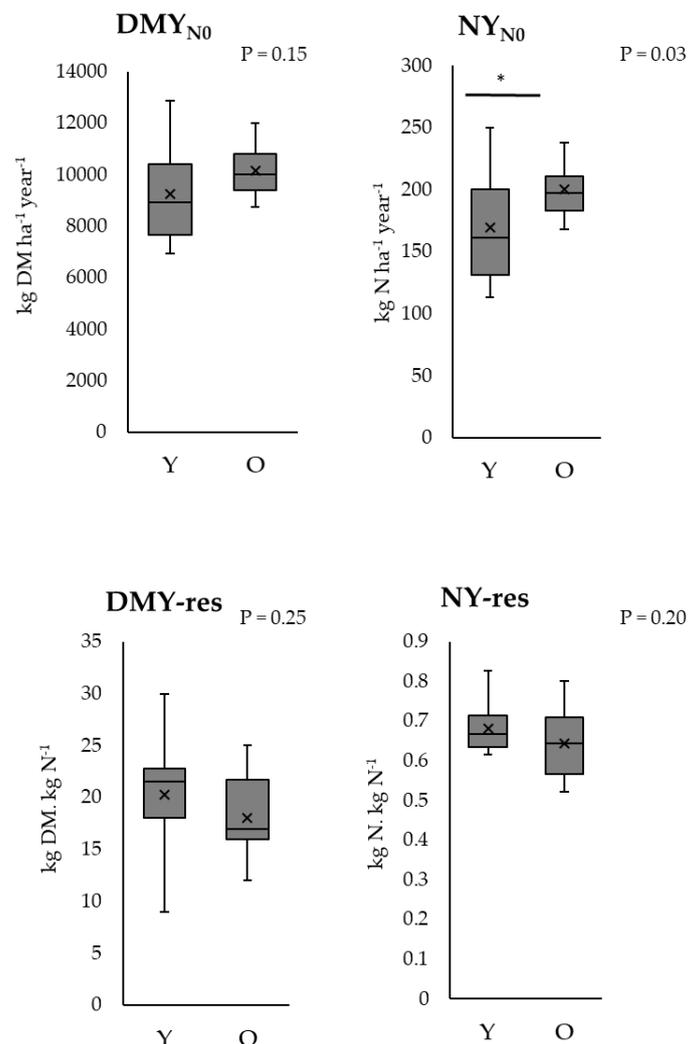


Figure 1. Boxplots with P-values of the grass productivity parameters: grassland dry matter yield without nitrogen fertilization (DMY_{N0}), grassland dry matter yield response to nitrogen fertilization (DMY-res), grassland nitrogen yield without nitrogen fertilization (NY_{N0}) and grassland nitrogen response to nitrogen fertilization (NY-res) on young (Y) and old (O) grasslands. Vertical bars represent the maximum and minimum value, crosses represent averages, asterisks represent significant ($P < 0.05$) difference between young and old grassland based on a paired ANOVA test.

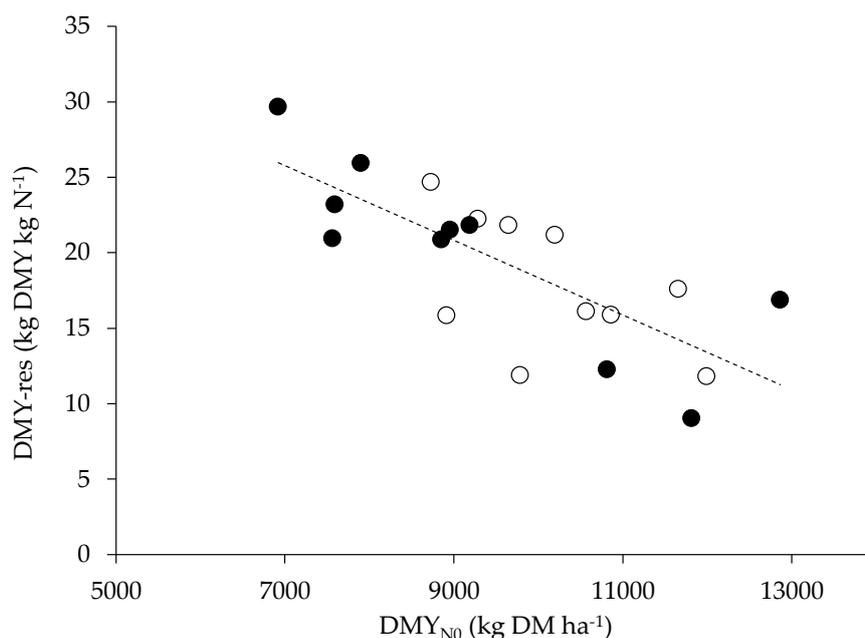


Figure 2. Dry matter yield response (DMY-res) as a function of grassland dry matter yield without nitrogen fertilization (DMY_{N0}). Closed dots represent young grassland, open dots old grassland. $r = -0.76$; $P = 0.001$.

3.4. Soil Organic Matter and Other Soil Chemical Parameters

The topsoil (0–10 cm) of the young grassland had a significantly lower SOM, C-total, C percentage of the SOM, HWC, N-total, C:N-ratio and PMN than the topsoil of old grassland (Table 1). The pH-KCl, Ca-total, P-Al and K in the solution of the topsoil did not significantly differ between young and old grassland. The young grassland soils contained significantly ($P = 0.007$) less water in the 5–10 cm soil layer at the day of sampling than the old grassland soils (Table 1). The pH-KCl and Ca-total were not correlated with any of the other measured soil parameters, except for the percentage silt, which negatively correlated with pH-KCl (Supplementary Material Table S2). SOM, C-total and N-total were negatively correlated with the percentage of sand (Table S2).

3.5. Relationships between Grass Productivity Parameters and Soil Parameters

Grass productivity parameters were significantly correlated with various soil parameters (Table S3). Of the soil parameters, N-total had the highest correlation with DMY_{N0}, DMY-res and NY_{N0} (Figure 3). In young grassland ($n = 10$) the correlations between DMY-res and SOM, C-total, C percentage of SOM, HWC and N-total were stronger than in old grassland soils. For NY-res no significant correlations with soil quality parameters were found (Table S3).

From the permuted stepwise regression procedure, DMY_{N0} was best explained by N-total (cross-validated $R^2 = 0.36$; $P = 0.04$; Table 2). With the addition of a second model parameter, the cross-validated R^2 decreased. DMY-res was best explained by N-total (negative regression weight) in combination with grassland age (positive regression weight; cross-validated $R^2 = 0.59$; $P = 0.02$). In young grassland ($n = 10$) DMY-res was best explained by N-total (negative regression weight) in combination with PMN (positive regression weight; cross-validated $R^2 = 0.69$; $P = 0.02$). NY_{N0} was best explained by N-total (positive regression weight) in combination with C:N-ratio (negative regression weight; cross-validated $R^2 = 0.61$; $P = 0.003$). In young grassland NY_{N0} was best explained by N-total (positive regression weight) in combination with C-total (negative regression weight; cross-validated $R^2 = 0.83$; $P = 0.01$). No predictors were found for grass productivity parameters in old grassland only. For NY-res no predictors were found in all fields, young nor old grassland.

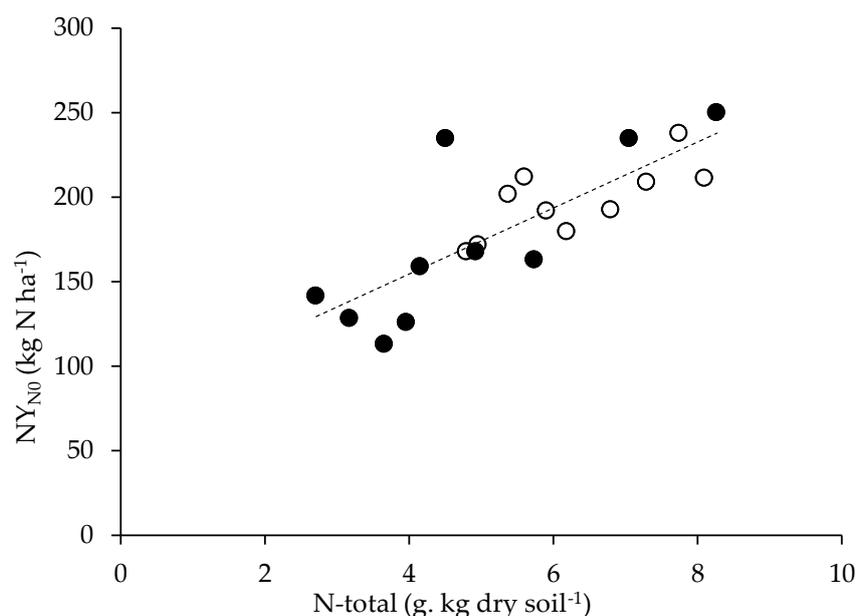


Figure 3. Grassland nitrogen yield without N fertilization (NY_{N0}) as a function of N-total of the soil. Closed dots represent young grassland, open dots old grassland; $r = 0.80$; $P = 0.001$.

Table 2. Overview of grass productivity parameters as affected by topsoil parameters by permuted stepwise regression for all ($n = 20$) and young ($n = 10$) grasslands; for old ($n = 10$) grasslands no significant predictor models were found. Significant ($P \leq 0.05$) predictor models are presented with one or two model parameters. $cv R^2 =$ cross-validated R^2 , Grass productivity parameters are described in Figure 1, topsoil parameters in Table 1. Mg_{share} : percentage magnesium in the Cation Exchange Capacity of the soil, that is further determined by calcium, potassium, sodium, hydrogen and aluminum.

Grass Productivity Parameter	All					Young				
	1st Model Parameter	2nd Model Parameter	$cv R^2$	P-Value	1st Model Parameter	2nd Model Parameter	$cv R^2$	P-Value		
DMY_{N0}	N-total	+		0.36	0.040	none				
	N-total	+	C:N-ratio	0.28	0.020					
	N-total	+	C-total	0.15	0.030					
	N-total	+	C perc SOM	0.31	0.030					
$DMY-res$	N-total	-		0.55	0.005	N-total	+		0.63	0.02
	N-total	-	C-total	0.47	0.020	N-total	-	PMN	0.69	0.02
	N-total	-	Grass age	0.59	0.020	N-total	-	C-total	0.60	0.05
NY_{N0}	N-total	+		0.56	0.004	N-total	+		0.52	0.04
	N-total	+	C-total	0.57	0.004	N-total	+	C-total	0.83	0.01
	N-total	+	C:N-ratio	0.61	0.003	N-total	+	C:N-ratio	0.63	0.02
	N-total	+	Mg_{share}	0.56	0.010					
$NY-res$	none					none				

4. Discussion

4.1. Grass Productivity

Contrary to our first hypothesis, grass productivity was not higher in young compared to old grassland (Figure 1). For $DMY-res$ and $NY-res$, there was no significant difference, whereas the NY_{N0} was actually 13% lower for young compared to old grassland. Below, we discuss these findings in relation to the three main reasons for grassland renewal, (1) botanical composition, (2) genetic improvement of ryegrass varieties and (3) grassland age.

(1) Botanical composition: In a study done in the 1970s in the Netherlands, nine experimental grassland fields with a dense sward with mainly *Lolium perenne*, *Phleum pratense*, *Poa pratensis* and *Poa trivialis* were partly reseeded [24]. In the following 10 years, DMVs and crude protein content in DM were lower in the reseeded parts in comparison with the original parts. The author mentioned that his result did not correspond with the common opinion that by definition young grasslands yield more than old grasslands. He stated that this was because good-quality old grassland was used, whereas other authors compared old grassland with an inferior botanical composition to good young grassland [24]. In our current study, young and old grasslands were not different in terms of botanical composition, most having well over 70% desirable grasses (*Lolium perenne* and *Phleum pratense*; Table 1). This minimizes the effect of botanical composition on differences in productivity between young and old grasslands in our study.

(2) Genetic improvement: An important reason for grassland renewal is the higher productivity and quality of the most recently developed ryegrass varieties [17]. A number of studies have reported increases in dry matter yield of genetically improved *Lolium perenne* varieties ranging from 0.15% [19] to 0.3% per year [18,20,21] over a period of 40 to 50 years. In our study, the average age difference between young and old grassland was 16 years. Assuming an increase of 0.3% per year in productivity by genetic improvement, the increase in productivity in these 16 years should theoretically be 4.8%. However, we found a (non-significant) decrease in DMV_{N0} of 9% of the young compared to the old grasslands. This is the productivity without N fertilization. According to Dutch legislation, farmers on a clay soil were allowed to fertilize their grasslands with 345 kg available N ha⁻¹ year⁻¹ in 2014. At the N application rate of 345 kg N ha⁻¹ year⁻¹, we also did not find a difference in the calculated DMV (based on the DMV_{N0} and DMV-res per field) of young (on average 16.2 Mg ha⁻¹) and old grasslands (on average 16.3 Mg ha⁻¹). This finding is in line with the study of Hopkins [25] who found only higher productivity of *Lolium perenne* reseeds at very high fertilizer-N rates of 450 and 900 kg N ha⁻¹ year⁻¹. Apparently, the genetic potential of the most recently developed varieties sown in the young grassland in our study was not expressed in the production environments (soil quality and fertilizer N-rates) under study.

(3) Grassland age: We chose to compare ‘young grasslands’ (5–15 years since renewal) with ‘old grasslands’ (>20 years since renewal). Other studies, however, compared grass productivity of recently (1–3 years) renovated grasslands with older grasslands [10,24–27,51]. Hoogerkamp [31] described three periods regarding the productivity of cultivated grassland when ageing: the “ley phase”, the “years of depression” and the “permanent grassland phase”. In the “ley phase”, the productivity of the young grassland is relatively high, after which it decreases (“years of depression”) and after several years it increases again to the level of old grassland, which is lower than that of the “ley phase”. According to the study of Hoogerkamp [31], the “years of depression” start 1 to 4 years after sowing and might last for several years depending on SOM content, soil compaction and a number of earthworms in the soil. Some of our ‘young grasslands’ can, therefore, be classified as still in the “years of depression” while others are already in the “permanent grassland phase”, which is in line with our findings that the variability in grass productivity was higher in young grassland and the average productivity of young grassland was not higher compared with that of old grassland (Figure 1).

4.2. Soil Parameters

The soil chemical quality, expressed as SOM, soil organic C and N and PMN, was lower in young grassland compared with old grassland (Table 1) underlining the losses in C and N in topsoil due to ploughing when grassland is renewed. This is in line with our second hypothesis. The soil water content was also significantly ($P = 0.007$) lower in young compared to old grassland soils. The water content of the soil is found to correlate positively with SOM [40,52], which may explain the lower soil water content for younger grassland soils.

SOM and labile fractions such as HWC and PMN, which are early indicators of changes in SOM [53] were positively correlated with grassland age (Table S3). This is explained by the SOM increase with grassland age [28,54].

4.3. Relationships between Grass Productivity Parameters and Soil Parameters

SOM and related soil parameters were strongly correlated with grass productivity in young as well as in old grassland, which confirmed our third hypothesis. From the measured soil parameters, N-total gave the strongest correlation with grass productivity parameters, except for NY-res (Table S3). In a New Zealand study, herbage production measured on nine old grasslands was related to N mineralization ($r = 0.87$; $P < 0.003$) [55]. In our old grasslands, there was no significant correlation between DMY_{N0} and soil N parameters (N-total and PMN), probably because the variation in DMY_{N0} and soil N parameters was smaller in the old grasslands compared to the young grasslands (Figure 1; Table 1).

In the Dutch grassland fertilization guidelines (www.bemestingsadvies.nl) the advice for N fertilization is based on the N supply capacity of the soil, defined as the non-fertilizer N supply including atmospheric deposition [34], corresponding with the NY_{N0} in our experiment. In the Dutch grassland fertilization guidelines, the N supply capacity of grassland on clay soils is estimated on the basis of N-total and corrected for grassland age. With the N supply capacity calculated with these equations, the correlation between the calculated N supply capacity and measured DMY_{N0} ($r = 0.71$; $P = 0.001$) and DMY_{res} ($r = -0.81$; $P = 0.001$), as well as the correlation with the measured NY_{N0} (N supply capacity; $r = 0.81$; $P = 0.001$) is stronger than with N-total only (Table S3). This indicates that on a marine clay soil the NY_{N0} can be predicted from N-total and grassland age.

The DMY_{res} correlated negatively with N-total ($r = -0.79$; $P < 0.01$), SOM ($r = -0.78$; $P < 0.01$), C-total ($r = -0.77$; $P < 0.01$) and soil water content ($r = -0.78$; $P < 0.01$) and also negatively with NY_{N0} ($r = -0.82$; $P < 0.01$; Table S3). This indicates that with an increasing soil N supply (due to high SOM, N-total and C-total) the response to N fertilization decreases (Figure 2). A negative correlation between NY_{N0} and NY_{res} was found by Deru et al. [41] on peat soil as well, and this negative relationship is also the basis of the official fertilization recommendations for grassland in the Netherlands (www.bemestingsadvies.nl).

4.4. Practical Implications

Dutch dairy farmers can consult the Reseeding Indicator [14] to assess when it is economically attractive to renew their grassland. According to this indicator and based on the costs in 2011, the overall costs of reseeded grassland on a clay soil (including costs for seed, ploughing, soil preparation, seeding, herbicides and soil analysis) amount to € 980 ha⁻¹ <http://webapplicaties.wur.nl/software/herinzaaiwijzerfe/>. A cost-benefit analysis by Schils et al. [12] indicated that grassland renovation is financially attractive if the new sward produces 10 to 25% more than the old sward.

Besides an economic impact, grassland renewal has an environmental impact. Many studies show an increased emission of the greenhouse gases N₂O and CO₂ and leaching of nitrogen after ploughing grassland [8,56–62]. Our study confirms that when grasslands contain at least 70% desirable grasses (i.e., *Lolium perenne* and *Phleum pratense*), long-term grass productivity does not increase and the NY_{N0} even decreases as a result of grassland renewal. Renewal does not result in higher grass productivity, most likely because of the loss of soil quality. As many studies have shown, the SOM content decreases when grassland is ploughed [38,54,55,63]. In the past, dairy farmers could compensate for this loss of SOM and related soil chemical quality through extra fertilization. However, due to current legislative prescriptions, fertilization is limited, which makes such compensation less feasible. Therefore, a strict recommendation to renew all grasslands after 10 years to improve productivity can be considered obsolete; farmers should be advised not to renew when the grassland contains 70% desirable grasses or more (i.e., *Lolium perenne* and *Phleum pratense*). When the introduction of high yielding grassland varieties is necessary, the focus should be on oversowing (i.e., non-destructively adding grass seeds to the existing sward) rather than renewing the grassland [64]. Moreover, farm management should focus

on minimizing the need for renewal by good grassland management, e.g., maintaining desirable grasses by grazing, ample fertilization, irrigation and preventing and ameliorating soil compaction [65,66].

5. Conclusions

- The grassland N yield without N fertilization (NY_{N0}) was significantly higher for old grassland compared to young grassland, as a result of higher soil organic matter, C-total and N-total contents in old grassland soils.
- There was no significant difference between old and young grassland in grassland dry matter yield without N fertilization (DMY_{N0}), grassland response ($DMY\text{-res}$), and N yield response ($NY\text{-res}$) to fertilizer N application.
- A significant part of the variation in DMY_{N0} and $DMY\text{-res}$ and NY_{N0} was explained by soil N-total content.
- On clay soils where the botanical composition of grasslands contain > 70% of desirable grasses, the presumed yield benefit of grassland renewal (sowing of the most recently developed grass varieties) is offset by a loss of soil quality (SOM and N-total). Accordingly, the current practice of grassland renewal within a relatively short time-span (every ten years) without considering the botanical composition, is counter-productive and not sustainable.

Supplementary Materials: The following are available online at <http://www.mdpi.com/2071-1050/12/7/2600/s1>, Table S1 Overview of species (grasses, legumes and herbs) plant cover (%) of the found plant species per field, mean young (m Y) and old (m O) grasslands. Table S2 Pearson's correlations (r ; $n = 20$) between topsoil quality parameters for all grasslands, parameters are described in Table 1. Table S3 Overview of Pearson's correlations (r) between grass productivity parameters and topsoil (0–10 cm) parameters for all ($n = 20$), young ($n = 10$) and old ($n = 10$) grasslands. Grass productivity parameters are described in Figure 1, topsoil parameters in Table 1. NSC: nitrogen supply capacity: calculated from N-total, corrected for grassland age according to the formulas of the Dutch grassland fertilization guideline based on Hassink [34].

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