Title: Intra and inter-annual climatic conditions have stronger effect than grazing intensity on root growth of permanent grasslands

Running head: Root production in grazed grasslands

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Abstract

- **Background and Aims**. Understanding how direct and indirect changes in climatic conditions, management, and species composition affect root production and root traits is of prime importance for grassland C sequestration service delivery.
- **Methods**. We studied during two years the dynamics of root mass production with ingrowth-cores and annual above- and below-ground biomass (ANPP, BNPP) of upland fertile grasslands subjected for 10 years to a gradient of herbage utilization by grazing.
- **Results**. We observed strong seasonal root production across treatments in both a wet and a dry year but response to grazing intensity was hardly observed within growing seasons. In abandonment, spring and autumn peaks of root growth were delayed by about one month compared to cattle treatments, possibly due to later canopy green-up and lower soil temperature. BNPP was slightly lower in abandonment compared to cattle treatments only during the dry year, whereas this effect on ANPP was observed the wet year. In response to drought, the root-to-shoot biomass ratio declined in the abandonment but not in the cattle treatment, underlining higher resistance to drought of grazed grassland communities.
- **Conclusions**. Rotational grazing pressure and climatic conditions variability had very limited effects on root growth seasonality although drought had stronger effects on BNPP than on ANPP.

Introduction

Permanent grasslands provide many services that tie in to human activities through livestock products, but also contribute to regulate greenhouse gas emission, because their soils accumulate large amounts of carbon in organic matter fractions. Intensification of management practices through changes in mowing, fertilization and grazing intensity may affect these services as well as climate variability through increased drought intensity and frequency (Conant et al. 2001; Jones and Donnelly 2004; Soussana and Duru 2007). Root activity (growth, exudation, turnover) is a main determinant of both nutrients and water uptake and a major input of C and N compounds into grassland soils. Thus, improving our understanding of grassland roots dynamics under different management and climatic conditions may help to identify management options to maintain forage production and C sequestration abilities of this ecosystem and thus its sustainability.

Different practices of management, such as mowing and grazing, modify forage production and the amount of soil C and N fluxes through direct effects of defoliation, fertilization or returns of excreta to soil on root growth and soil abiotic factors and indirect effects through species composition changes (Bardgett and Wardle 2003; Dawson et al. 2000; Soussana et al. 2004). In mown grasslands it has been shown that root mass production is generally lower when grass is frequently mown and fertilised (Leuschner et al. 2013; Picon-Cochard et al. 2009). This may be explained by changes in root-to-shoot allocation, with increase of above-ground growth in order to maximize light capture. The complexity of these phenomena in grazed grassland is greater than in mown systems owing to animals' selective defoliation of plant species, and also because returns to soil are spatially heterogeneous (Rossignol et al. 2011). In addition, level of soil fertility may buffer the degree of root response to defoliation in grazed grasslands as plants exhibit specific responses to defoliation in fertile and unfertile grasslands (Duru et al. 1998). Overall this can explain why no clear trend is found for the effects of grazing on above- and below-ground production (e.g. see syntheses of Milchunas and Lauenroth (1993) and McSherry and Ritchie (2013)), although a meta-analysis emphasizes a negative effect of grazing intensity on above- and below-ground carbon stocks compared to ungrazed systems (Zhou et al. 2017). In addition, repeated defoliations induced by grazing and mowing of grassland can simultaneously increase (i) soil temperature by increasing solar radiation reaching the soil and (ii) soil moisture due to lower leaf area index and reduction of vegetation transpiration (Moretto et al. 2001; Pineiro et al. 2010; Smith et al. 2014). Soil moisture can also be modified by high stocking rate through changes of soil bulk density due to soil compaction (Pineiro et al. 2010). These direct effects of grazing on soil abiotic factors should affect root growth of grazed grassland, although all these phenomena are not very well documented in field conditions.

Species composition change induced by management is also an important determinant of above- and below-ground response in grazed grassland; intensive practices (high grazing intensity, fertilization) generally favour the development of fast growing species (exploitative strategy) with highly digestible shoot and root tissues, low C/N and tissue density whereas at the opposite extensive practices (low grazing intensity, absence of fertilization) favour slow growing species (conservative strategy) with poorly digestible organs and high tissue density (Klumpp et al. 2009; Louault et al. 2005; Soussana and Lemaire 2014; Wardle et al. 2004). Root-to-shoot biomass allocation, but also functional traits (used as proxies of ecosystems properties like ANPP or BNPP, e.g. Laliberté and Tylianakis 2012), are thus likely to change in response to intensification of practices, e.g. from ungrazed to intensely grazed in temperate grassland (Klumpp and Soussana 2009) or in alpine meadows, steppes and desert-steppes (Zeng et al. 2015). Overall, according to Ziter and MacDougall (2013), the uncertainty surrounding nutrient-defoliation responses makes it difficult to predict whether C storage will be higher in managed compared to unmanaged grasslands. Thus soil fertility should be considered when comparing different grazing intensities in grassland, as species adapted to fertile conditions will exhibit either trait related to avoidance or to tolerance strategies toward defoliation, both having similar exploitative resource-use strategy (Louault et al. 2005).

Increased climate variability is another source of response uncertainty in managed ecosystems. As more frequent and longer period of drought associated with heat waves may threaten and shape the long-term dynamics of perennial ecosystems such as grasslands (Brookshire and Weaver 2015), it is important to understand how above- and below-ground compartments respond to climate variability. However, there are few data on above- and below-ground biomass responses to drought for grassland (Byrne et al. 2013; Wilcox et al. 2015), although some evidence shows that the ‘slow’ trait strategy (resource conservation) is associated with drought tolerance (Pérez-Ramos et al. 2012; Reich 2014). It has also been shown that the timing of drought has more influence on the below- than on the above-ground compartment especially in grazed *vs*. ungrazed grassland, as peak of shoot biomass can occur before the drought period (Frank 2007). In addition, comparing two contrasting grazed grasslands, Klumpp et al. (2011) showed that during wet years extensive managed grassland (low stocking density combined with low soil fertility) had a higher storage capacity than intensive managed grassland (moderate stocking density combined with N fertilization), whereas the reverse was observed during dry years, as a result of higher canopy senescence in extensive *vs.* intensive management. Changes in root morphology and functioning may thus be an important mechanism in plant adaptive strategies to drought, and have been less well studied than above-ground plant responses (Biswell and Weaver 1933; Dawson et al. 2000; McInenly et al. 2010). However, there are not enough data to make generalizations about combined impacts of management and climatic conditions variability such as precipitation reduction on root and shoot biomass production and plant traits defining plant strategies related to resource use and grazing intensity.

In the present experiment, we sought to assess whether grazing intensity affected root growth dynamics, root and leaf functional traits and annual below-ground biomass (BNPP) in a fertile and productive grassland and whether root response is mirrored by the annual above-ground biomass production (ANPP) and leaf traits and by changes of climatic conditions. These responses could be modulated by direct effects of grazing intensity on soil microclimate. Root and leaf traits were studied as response traits to grazing intensity and as effect traits of BNPP and ANPP, respectively. The study was carried out in a long-term field experiment for which controlled grazing intensity had been applied for 10 years. We compared abandonment of grazing and two levels of herbage utilization by grazing based on five rotations per year. In two consecutive years, the ingrowth core method was used to measure monthly root biomass production and calculate annual root production (BNPP); ANPP was measured by grazing exclusion cages and community-weighted mean leaf and root traits were assessed the first year. We tested the following hypotheses: (i) high grazing intensity increases above-ground mass at the expense of root production as a result of the direct negative effect of defoliation on root growth, whatever the climatic conditions, (ii) inter-annual climatic conditions modulate above and below-ground biomass production response to grazing intensity as a consequence of higher presence of defoliation tolerant and drought-sensitive species (*Lolium perenne* or *Trifolium repens*) in the high grazing intensity treatment; (iii) root traits respond to treatment and is a determinant of BNPP, as observed for leaf traits for ANPP.

Materials and methods

Site characteristics

The experiment took place in the long-term observatory network (ACBB-SOERE) located at St-Genès-Champanelle, France (45°43′N, 03°01′E, 880 m a.s.l.). The local climate is semi-continental with oceanic influences (mean annual temperature 8.5 °C, mean annual precipitation 784 mm, Table 1). The site supports mesotrophic multi-specific permanent grassland, dominated by species with high Ellenberg indicator values for N (Schaffers and Sykora 2000), indicating a high level of fertility for the site (Table S1; Louault et al. 2017). The soil is a cambisol with a sandy loam texture, developed on granitic bedrock. Differences in local soil composition and profile led us to consider two blocks characterized respectively by a eutric cambisol (54% sand; 26% silt; 20% clay; 7.0% organic matter; pH: 5.9) and a colluvic cambisol (50% sand; 26% silt; 24% clay; 7.4% organic matter; pH: 6.0) including some volcanic materials.

Management

Prior to the installation of this experiment in 2005, the study area had been used for intensive hay and silage production (combining grazing, mowing and fertilization), with mineral fertilization, and two years preceding the start of the experiment (2003 and 2004), the grassland site was mown three times per year without fertilization. Then, from 2005, the grassland had been managed for 10 years with a gradient of grazing intensity resulting from three treatments: abandonment (Ab), low (Cattle-) and high (Cattle+) level of herbage utilization obtained by modification of stocking density (0, 6.9 and 13.8 LSU ha^{-1}, livestock unit, respectively) with five grazing rotations each year: mid-April, late May, early July, September and November, lasting on average 9.6, 9.0, 10.7, 8.6, and 2.1 days, respectively. The two cattle treatments corresponded to two levels of herbage utilization by grazing, and had on average 15.2 ± 0.5 cm (mean ± se, Cattle-) and 7.7 ± 0.2 cm (Cattle+) residual plant height at the end of each grazing rotation, respectively. For each treatment, two replicate plots were set up per block, resulting in four replicates per treatment, and a total of 12 plots (2 blocks x 2 plots x 3 treatments). The average distance between the two blocks is about 230 m and all treatments are randomized within each block. The size of the plots differs according to treatments: 2200 m^2 for the two cattle treatments and 400 m^2 for the abandonment.

Climatic and edaphic conditions

Daily precipitation (mm) and air temperature (°C) were measured for the two years, and recorded with a meteorological station located at the site. An aridity index was calculated as precipitation minus potential evapotranspiration (P - PET, mm) with the Penman-Monteith equation. Daily soil temperature (°C) was measured with thermocouple sensors (home-made copper-constantan sensors) inserted at 20 cm depth in each plot and recorded with a HOBO data logger (U12-014, Onset Instruments, MA, USA). Daily soil volumetric water content (SWC, m^3 m^{-3}) of each plot was measured with two probes (ECHO-10, Decagon, USA), inserted horizontally at 20 cm depth, and connected to dataloggers (EM5 and EM50, Decagon, USA). From January 2014 to November 2015 (DOY 132–326), SWC was measured every 30 min and averaged at daily scale. For each plot, average values of the two probes were used. Daily relative soil water content data are shown and calculated as the ratio:

$RSWC = \frac{\text{SWC} - \text{SWCmin}}{\text{SWCmax} - \text{SWCmi}}$, where SWC is the soil moisture at a given day, SWCmin is the minimum value of soil moisture and SWCmax is the maximum value of soil moisture, both observed during the two years. For soil temperature and RSWC, values were averaged according to root growth time scale.

Root growth and root mass

Six months beforehand, shallow (0-20 cm) soil was collected on each of the two blocks of the site and sieved (5 mm mesh size) to remove stones and coarse organic matter, and then left unused outside covered under a shelter and protected from direct sunlight. Thereafter, this air-dried soil was used to fill the ingrowth-core each month.

In December 2013 and for each of the 12 plots, soil cores were collected with an auger (8 cm diameter, 0-20 cm depth) at four locations representative of the plant community in the treatment. On average mean distance between locations are 19.8 m ± 0.2, 21.7 m ± 0.1 and 17.2 m ± 0.2 for Ca+, Ca- and Ab (mean ± SD, see Fig S1), respectively. After core harvest, each hole was filled with a plastic net (8 mm mesh size) containing a fixed volume of air-dried sieved soil (ingrowth core), collected six months beforehand. Then, about each month and for two years (2 x 10 times), ingrowth cores, containing soil and the root and rhizome material that had grown therein, were extracted, and then replenished with another fixed volume of dry sieved soil. Thus monthly and annual root production (BNPP, g m^{-2} y^{-1}) were measured from February 2014 to December 2015. Root production period ranged on average 36.5 days, but with longer and shorter periods in winter and spring-summer, respectively (Table 1). In periods with absence of precipitation, a fixed volume of water was added to adjust soil humidity to field conditions. After collection, the ingrowth cores were transported to the laboratory and immediately stored at 4 °C before processing in the next five days. The roots were washed under tap water and with a 200 µm sieve, and then oven-dried (48 h, 60 °C).

In order to measure root mass stock, soil cores were collected three times (December 2013, March and June 2014) with the same auger and near the ingrowth cores locations. These samples were stored in the freezer (-18°C), and after defrosting, the roots were washed with the same procedure as that used for the ingrowth cores, and then oven-dried (48 h, 60 °C).

Root traits

Subsamples of washed roots collected with the ingrowth cores collected in June 2014, were fresh weighed, and then frozen (-18 °C) before morphology analysis. After defrosting, roots were stained with methylene blue (5 g L^{-1}) for about 5-10 minutes, rinsed in water, spread in a transparent glass box containing a thin layer of water, and covered with a transparent plastic sheet. High resolution images were recorded with a double light scanner (800 dpi, perfection V700, Epson, JA) and analyzed with WinRhizo software (PRO 2012b, Regent Instruments, CA) with the automatic procedure. Two scans per location were recorded and separately analyzed to measure root length (m), root volume (cm^3), root surface area (m^2), average root diameter (mm) and length by class diameter (13 classes: 11 with 0.1 mm interval and 2 with 0.5 mm interval). Specific root length (m g^{-1}), root tissue density (g cm^{-3}) and specific root area (m^2 g^{-1}) were calculated for fine roots as in Picon-Cochard et al. (2012).

Botanical composition

Species contribution (%) was visually observed on a circle (20 cm diameter) around each ingrowth core location in April (cattle treatments) and May (abandonment) 2014. For each zone, a score on a ten-point scale was allocated to species present according to their volume occupancy, and the percentage of each species was calculated at the plot scale by averaging values of the four zones. The list of species and their relative contributions is given in Table S2.

Above-ground biomass production

On each plot and on each sampling date, four fenced sampling areas (0.6 × 0.6 m) were used to measure accumulation of above-ground biomass after above-ground standing biomass was clipped at 5.5 cm. At each sampling date, biomass was sampled at a height of 5.5 cm, oven-dried and weighed. Measurements were made five times in the course of the year, before each grazing event in Cattle+ and Cattle- plots, and three times (spring, summer, autumn) in abandonment plots. Sampling areas were moved within the plot at each measurement date during the year. Annual above-ground net primary production (ANPP, g m^{-2} y^{-1}) was calculated as the sum of the successive biomass accumulations along the year.

Leaf traits

Community-weighted mean (CWM) trait values of leaf dry matter content (LDMC), specific leaf area (SLA) and reproductive plant height (H) were calculated for each ingrowth core zone using (i) the relative contribution of the dominant species to the community (i.e. species that account for at least 85% of the cumulated species contribution of the community) measured in 2014, and (ii) leaf trait measurements made at plot scale in 2006 and 2007. Traits were measured on ten vegetative plants using standard protocols (see methods in Louault et al. 2005). Reproductive plant height was measured on mature plants located in fenced zones to allow full plant development. CWM is expressed with the following equation: CWM = $\sum p_i \times \mathrm{trait}_i$, where p_i is the relative contribution of species *i* to the community and trait_i the trait of species *i*.

Statistical analyses

For a given date, root mass and root traits collected at each location (four ingrowth-cores in each plot), averages of data coming from the four locations were used to have a single value for each of the 12 plots and test for the effect of treatment and dates. Before ANOVA, normality of residuals was inspected with quantile-quantile plots of model residuals, and variance homogeneity was confirmed by checking the plots of model residuals *vs.* fitted values. Data were transformed if they deviated from ANOVA assumptions (square root, ln, reciprocal). Linear mixed effects models as available in the R ‘nlme’ package (Pinheiro et al. 2015) were used to perform repeated measure ANOVAs to test the effects of treatments, dates and their interactions on values of root growth, soil temperature, RSWC, and root mass stock, with plots nested in block as a random factor accounting for temporal pseudo-replication. For root growth dynamics, soil temperature and RSWC (Fig 1, Table S1), dates correspond to 20 dates and for root mass stock, dates correspond to three harvest dates (Table 2). For BNPP, ANPP and root to shoot ratio (BNPP/ANPP), data were analyzed using a nested mixed model procedure, with treatments and year used as fixed factors with plot nested in block as random factors. For leaf and root traits data, treatments were used as fixed factors with plots nested in block as a random factor. *Post hoc* tests were performed to compare significance levels across fixed factors with a Tukey test ('lsmeans' package). Principal component analyses (PCA) were performed for each year to analyze relationships between leaf and root traits, soil temperature, RSWC, root mass stock, ANPP and BNPP measured at plot level; treatments were considered as supplementary categories ('FactoMineR' package). All statistical analyses were performed in the R environment (version 3.5.2, R Core team 2012) using RStudio (Version 1.1.463).

Results

Climatic conditions during the experiment

Compared with average long-term climatic data for the site, the first and second years of the experiment had higher (+92 mm) and lower (-199 mm) precipitation, respectively (Table 1). Potential evapotranspiration (PET) in the second year was also higher than the long-term average (difference of 73 mm), leading to a negative annual climatic water balance (P - PET = -181 mm and a deficit of 271 mm compared to the long-term average). Annual temperature in the two experimental years was similar and about 0.8°C higher than the long-term average for the site (Table 1). At monthly time scale and during part of the growing season (March to September), in comparison with the first year, the second year had a cumulated water deficit difference of -266 mm and a temperature warmer by +1.9 °C than the first year. Larger differences between the two years occurred in June-July with higher temperature (+6 °C), higher water deficit (P-PET = -152.6 mm) and less precipitation (-81%) in the second year.

Dynamics of soil temperature and relative soil water content

Soil temperature was significantly affected by treatment, dates and treatment × dates (Figure 1; Table S1). For most of the dates (February to October), abandonment treatment had lower soil temperature (1.76 °C, on average) than the grazing treatments, whereas the Cattle- treatment showed significant lower soil temperature (-0.64 °C) than the Cattle+ treatment. However, this was significantly observed for a limited number of dates in early summer of both years. Relative soil water content (RSWC) fluctuated from 0.6-0.7 at the beginning of spring to 0.38 in June in the wet year and to 0.2 during the dry year, which is in accordance to variation of the atmospheric aridity index (P-PET). In the case of the dry year, from summer until autumn, RSWC remained lower than 0.4 and the aridity index was negative.

Root growth dynamics

Root growth was affected by date and treatment × date interaction (Figure 1). Each year, peak of root growth occurred twice, in spring and autumn, and growth was markedly reduced in summer and winter. Only in the second year did growth stop in summer, and it was significantly lower than the first year. Regarding treatment effect, abandonment showed significant lower root growth than the two grazing treatments for the spring period in both years, and for the autumn of the second year. While in autumn 2014, a delay of growth peaks was always observed, which led to a two-fold higher root growth for abandonment *vs.* the two cattle treatments (end of September: date 8). The two grazing treatments had similar root growth across years and seasons.

Seasonal root mass stock, BNPP, ANPP and root-to-shoot biomass ratio

Stock of root mass did not change through season and across treatment (Table 2). BNPP, ANPP and root-to-shoot biomass ratio (R/S) were significantly lower during the second year, with a stronger effect on BNPP (-44% on average) than ANPP (-24%) (Figure 2, Table 3). Only the abandonment treatment maintained their value of ANPP in the second year, which led to a 48% decline in R/S (significant treatment × year, $P < 0.01$, Table 3). Accordingly, treatment effect was only observed for BNPP the second year, with a decline of 24% for abandonment compared to cattle treatments and for ANPP the first year: Cattle+ having 22% and 68% higher values than Cattle- and abandonment, respectively, while Cattle- had 38% higher ANPP than abandonment.

Species composition, leaf and root traits

Abandonment treatment was characterized by the dominance of tall grass species: 76% in all with 27.2% of *Alopecurus pratensis*, 18.8% of *Elytrigia repens*, 11.3% of *Poa pratensis* and 10.3% of *Arrhenatherum elatius*, the presence of some forbs (19%) and the absence of legumes (Table S2 and Table 4). The two cattle treatments differed from abandonment treatment by equal presence of *Taraxacum officinale* (18% on average) and *Trifolium repens* (17% on average). Difference also concerns grass species (56% in total) with the dominance of *Dactylis glomerata* (22.2%), *A. pratensis* (7.6%) and *Schedurus arundinaceus* (5.6%) for Cattle- and *Lolium perenne* (13.6%), *D. glomerata* (9.1%) and *Poa trivialis* (7.2%) for Cattle+. Thus, the Cattle+ treatment had a higher percentage of *L. perenne* than Cattle- (Table S2).

Community-weighted mean leaf traits (CWM) were significantly modified by the treatments. Plant height and LDMC were significantly higher ($P < 0.05$ and $P < 0.0001$, respectively; Table 4) in abandonment than in the two cattle grazed treatments, whereas SLA was lower ($P < 0.05$). Unlike leaf traits, root traits were only slightly affected by the treatments. Specific root length (SRL, $P < 0.1$) and specific root area (SRA, $P < 0.05$) were lower in abandonment treatment than in Cattle-, but not Cattle+. For other root traits (diameter, RTD and root length % by class diameter) no between-treatment differences were observed (Table 4).

Co-variation of traits and production

The two main axes of the standardized PCA explained 60.1% and 56.8% of the community trait and production variation in 2014 and 2015, respectively (Figure 3). For the first year, the first PCA axis (PC1), accounting for 43.4% of the total variation, was significantly related to leaf and root traits, ANPP and soil temperature. Soil temperature, SRA and ANPP had positive loadings, and diameter, plant height and LDMC had negative loadings (Table 5). The second PCA axis (PC2), accounting for 16.7% of the total variation, was significantly and positively related to root diameter and negatively to SRA. For the second year, the first PCA axis (PC1), accounted for 37.4% of the total variation, and was significantly related to leaf and root traits, ANPP and BNPP. BNPP and SRA had negative loadings, and root diameter, plant height and ANPP had positive loadings (Table 5). The second PCA axis (PC2), accounting for 19.4% of the total variation, was significantly and positively related to RSWC and stock of root mass averaged across three dates. Finally, abandonment treatment was significantly related to PC1s with negative and positive loading s for the first and the second year, respectively.

Discussion

Ten years of contrasted management had strongly modified the functional diversity and above-ground production of this fertile upland grassland (Herfurth et al. 2015; Louault et al. 2017). Accordingly, we expected that above-ground biomass patterns would be mirrored below-ground, especially during the periods of grazing. Here we first discuss within-year differences of root growth, followed by inter-annual variation responses to grazing intensity and climatic conditions variability between the two contrasting years, and last we analyze relationships between traits and above- and below-ground production.

Seasonality of root growth was independent of grazing intensity and climatic conditions

As expected, root growth of permanent grassland is affected by seasons and peaks in spring and autumn (Garcia-Pausas et al. 2011; Pilon et al. 2013; Steinaker and Wilson 2008), but unexpectedly, grazing pressure applied by rotations and climatic conditions variability had very limited effects on this seasonality. This means that at below-ground level, plant community behavior was not affected by rotational grazing management nor by climatic conditions variability, although a severe drought occurred in summer of the second year. Only the abandonment treatment showed a delayed root growth peak in spring. This delay is probably the result of slower shoot budburst and reduced capacity to produce new green leaves in dense litter canopy, especially at the beginning of the growing season in spring (data not shown). Moreover, the tall and dense canopy of the abandonment treatment strongly modified soil temperature, with cooler soil conditions as expected in such abandoned vegetation (Picon-Cochard et al. 2006; Zhou et al. 2017; Zhu et al. 2016). As shown in some studies, light or soil water and nutrient availabilities (Edwards et al. 2004; Garcia-Pausas et al. 2011; Steinaker and Wilson 2008) are other abiotic factors determining dynamics of root growth in grasslands, as root peaks were observed before the peak of soil temperature in summer when negative climatic water balance occurred, especially in the second year. Nevertheless, plants growing in abandonment offset their slower root growth by producing similar root biomass at annual scale, especially during the wet year. The presence of tall grass species such as *A. pratensis*, *A. elatius* and *E. repens* with plant trait syndromes related to resource conservation strategy (lower SLA and SRL and higher plant height and root depth; Pagès and Picon-Cochard 2014) might explain their capacity to produce higher root biomass on a shorter-term period before canopy senescence onset. Also pre-existing soil fertility can be maintained in conditions of very low levels of herbage utilization (near-abandonment), because of the absence of biomass exportation and increased internal recycling of N within senescent plants, both contributing to an increase in total N available for plant growth (Loiseau et al. 2005).

The similar root growth dynamics of the two cattle treatments was unexpected, considering that infrequent defoliation and moderate excreta returns to the soil might increase root biomass production at the expense of shoot biomass (Klumpp et al. 2009). The absence of effect on root growth and BNPP means that grazing pressure applied on plant communities by rotations (5 rotations of 9 days each on average) was too short but enough to observe effect on ANPP, in wet conditions. Worldwide there are different ways to manage grassland by grazing (Huyghe et al. 2014), rotational or permanent grazing options with different stocking rates, durations, types of herbivores. In general, this management creates high spatial heterogeneity within the plots due to animals' selective defoliation of plant species, and also because returns to soil are spatially heterogeneous. Thus in grazed grassland, disturbance creates patches of vegetation, which should affect locally root growth and below-ground biomass of plant communities if duration of grazing is sufficient. The complexity of these phenomena in grazed grassland is greater than in mown systems owing (Rossignol et al. 2011).

Then, again, the confounding effect of soil fertility and defoliation may mask a clear response of the below-ground compartment in grazed grasslands. In view of that, we postulate that root growth in Cattle+ treatment was favored by the higher soil temperature compensating for the negative effects of frequent defoliation on root growth while the cooler soil conditions encountered in Cattle- might have slowed root growth. Soil moisture is a main determinant of plant growth and can be affected by cattle treatments. Some studies showed an increase of soil moisture in grazed compared ungrazed treatment due to lower leaf area index in the grazed conditions (Moretto et al. 2001; Pineiro et al. 2010), or an absence of effects in others (LeCain et al. 2002; Smith et al. 2014). The presence of herbivores can increase soil bulk density and consequently modify soil moisture. However, in our field conditions and after 10 years of treatments application, soil moisture was not affected by the rotational grazing, probably because the temporal scale used (monthly-based) buffer shorter-term response.

We should also consider the level of soil fertility and species composition as drivers of root growth and trait plasticity (Dawson et al. 2000). The soil fertility of our site, reflected by the nitrogen nutrition index (NNI, Lemaire and Gastal 1997), was very similar along our grazing intensity gradient (Table S1), at least in 2014. Thus in our site we had the opportunity to compare grazing intensity effect at equivalent soil fertility. Knowing that root trait plasticity generally shows larger differences with respect to soil fertility than by cutting or defoliation (Leuschner et al. 2013; Picon-Cochard et al. 2009), we can expect that under similar soil fertility grazing intensity had a less pronounced effect on root growth. Indeed, the higher presence of species tolerating defoliation, with shorter stature and root system (*L. perenne*, *P. trivialis*), but having higher shoot and root growth capacity after defoliation and also higher rhizosphere activity (Dawson et al. 2000), probably compensated for the negative effect of defoliation in the Cattle+ treatment. Also the sampling depth might have had an effect, as we expect that harvesting root systems deeper than 20 cm should give more contrasting root growth response across the two cattle treatments according to the grass species composition due to species-specific differential root depth distribution (Xu et al. 2014). Taken together, we provide evidence that higher soil temperature, high soil fertility and species composition have moderated root growth response along our grazing intensity gradient. The difficulty to assign species composition in root mixtures, however, makes it difficult to draw firm conclusions.

Climatic conditions variability shaped responses of ANPP, BNPP and root-to-shoot biomass production ratio along the grazing intensity gradient

According to meta-analyses and recent results (McSherry and Ritchie 2013; Zeng et al. 2015; Zhou et al. 2017), grazing intensity generally has negative effects on above- and below-ground biomass of grasslands whatever the climatic conditions or vegetation type, although these effects can be modulated by levels of grazing intensity. Our results do not confirm these findings, because ANPP and BNPP increased in response to grazing intensity compared to abandonment, in the wet and the dry year, respectively. Methodology issues for estimating ANPP and BNPP in grazed grasslands should thus be taken into account, as some papers report either biomass stock or fluxes measured once at peak of growth or at several periods (Scurlock et al. 2002), but also estimation of BNPP from indirect measurements (e.g. Zeng et al. 2015). Mass based on stock gives a snapshot of plant functioning, generally including mixtures of living and senescent tissues, thus depending on abiotic factors and plant growth, whereas measurements based on new shoot and root biomass reflect the growth potential of grasslands. We are aware that these methods are very different, but in response to grazing intensity, BNPP measured with ingrowth cores gave similar results as root mass stock assessed at three seasons. Nevertheless, climatic aridity index (P - PET) had stronger effects on ANPP and BNPP than grazing intensity, because severe drought had a direct negative effect on plant growth. In comparison with another experiments located alongside ours, 80% of canopy senescence was reached for a cumulated aridity index of -156 mm (Zwicke et al. 2013). As this index reached -303 mm from March to August, this confirmed that a severe drought occurred in the second year of our experiment, and explained root growth cessation in summer. At annual scale, ANPP of the two cattle treatments showed lower resistance to increased aridity (resistance defined as $ANPP_{year2}$ / $ANPP_{year1}$, being equal to 0.63) than abandonment treatment (ratio=1). For BNPP, results were inversed, leading to a lower resistance of root-to-shoot biomass ratio in abandonment than in the two cattle treatments. The absence of root growth modification by grazing at annual scale the wet year reflects well the change in root-to-shoot biomass allocation, albeit not significant. Other processes such as root turnover (mortality, rhizodeposition) are expected to change in grazed *vs.* ungrazed grassland. For our site Herfurth et al. (2015) observed similar root mass stock along a grazing disturbance gradient as in the present study, but by using a simplified C flux model, these authors showed that the Cattle+ treatment tended to accelerate

C cycling in plant communities, resulting in a higher quantity of C allocated to the soil organic matter continuum. Taken together, these results suggest that the slight BNPP increase under grazing may occur with an increase in rhizodeposition, because root turnover calculated as BNPP to root mass stock ratio (data not shown, Lauenroth and Gill 2003) was not different across treatments.

Furthermore, our results suggest that grazing treatments slow down the negative effect of aridity on root-to-shoot biomass ratio, and these treatments seem to be better adapted to buffering the negative effect of drought on grassland production than for abandoned grasslands. This is consistent with previous work showing that moderate grazing could be more beneficial than no grazing for drought resistance and recovery of ANPP and BNPP (Frank 2007; Xu et al. 2012), and that BNPP was more resistant than ANPP to change in precipitation (Yan et al. 2013). Other studies showed no prevalence effects of grazing, drought or fire observed on grassland production in North America and South Africa (Koerner and Collins 2014). Nevertheless, this points to a need for further research to determine whether grazing pressure has additive or combined effects on drought response of grasslands (Ruppert et al. 2015).

Community-weighted mean leaf and root traits as predictors of ANPP and BNPP

As shown by other studies (e.g. Diaz et al. 2007; Laliberté and Tylianakis 2012; Louault et al. 2017; Zheng et al. 2015), disturbance induced by grazing pressure has profound effects on plant community and functional traits by selecting tolerant species to defoliation such as *L. perenne*, *P. trivialis* or *T. repens*, with possible cascading effects on multiple ecosystem functions. With the capacity to regrow quickly after defoliation, these species generally exhibited high values of SLA and low values of LDMC and plant height. They contrast with species adapted to fertile soil, but with a slower regrowth capacity after defoliation such as *D. glomerata* or *F. arundinacea*, with opposite leaf trait values. In abandonment, competition for light tends to select plants with trait syndromes related to conservative strategy (tall plants, low SLA and high LDMC values). Thus, the CWM traits of the community will depend on the balance between these species groups, which are expected to affect ANPP and BNPP (Klumpp et al. 2009; Milchunas and Lauenroth 1993). Although the presence of tolerant and intolerant species to defoliation in both cattle treatments, leaf trait values were similarly and positively related to ANPP, and only differed from traits of species present in the abandonment treatment. This means that cessation of grazing strongly differentiated plant communities, whereas within the two cattle treatments differences were slighter.

For the below-ground compartment, we expected that above-ground differences were mirrored by the root growth and traits, assuming that higher root diameter values, and lower SRL and SRA values are associated with lower BNPP in abandonment compared with the two cattle treatments. Although root response to grazing (mainly through defoliation) generally reported reduction of root mass or root length (Dawson et al. 2000) our study did not confirm these assumptions. The contrasting results are possibly due to variable abundance of tolerant species to defoliation or with confounding effects of both defoliation and level of soil fertility on roots of grazed grasslands (Leuschner et al. 2013; Picon-Cochard et al. 2009; Yan et al. 2013; Ziter and McDougall 2013). Thus, root growth reductions associated with grazing may have a greater impact in locations where grazer-mediated nitrogen return is spatially decoupled from defoliation (McInenly et al. 2010). Further, higher specific root area (SRA) observed in Cattle- than in abandonment and Cattle+ treatments should reflect higher presence of species with fine roots such as *D. glomerata* or *H. lanatus* (Picon-Cochard et al. 2012), because soil fertility approximated by NNI was near comparable across treatments.

Conclusions

Similar functional diversity of the plant communities and similar soil fertility across the two cattle treatments explained the absence of changes in root mass production for these treatments. Our site disentangled confounding effects of fertility and defoliation on root production, which is not generally the case for other studies. Thus, our results suggest the prevalence of a soil fertility effect on root production response rather than a defoliation effect. However, we cannot rule out the possibility that continuous rather than rotational grazing practice would give similar results. In view of that, grazing practices information should be considered in order to give some generalizations about below-ground compartment response of fertile grassland with respect to grazing intensity. Besides, the strong effect of climatic conditions variability on ANPP and BNPP observed at short term could increase in the future as more frequent climatic extremes are expected. It is thus necessary to improve our knowledge of grazing practices that allow higher resilience of grasslands to more frequent and intense climatic events such as drought and heat waves.

Data accessibility

Data are available online: https://zenodo.org/deposit/4034903#

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Table 1. Air temperature (°C), precipitation (P, mm), potential evapotranspiration (PET) and climatic water balance: cumulated (P - PET, mm) and calculated for the 28 y period 1986-2013, mean values ± SD) and measured for the 10 dates in 2014 and 2015 corresponding to measurements of root growth and averaged (temperature) or summed (P, PET, P PET) at annual scale.

Year	Dates	Air temperature	Precipitation	PET	P - PET
	Annual long-term average	**8.5 ± 0.6**	**784 ± 1376**	**693 ± 96**	**91 ± 195**
2014	December 12 – February 23	3.7	98	37.5	60.5
	February 24 – March 23	5.3	27	46.3	-19.3
	March 24 – April 21	7.2	23.5	68.7	-45.2
	April 22 – May 25	9.2	79.5	103.1	-23.6
	May 26 – June 22	14.2	58	110.2	-52.2
	June 23 –July 20	15.1	136.5	93.9	42.6
	July 21 – August 24	14.4	90.5	100.5	-10
	August 25 – September 29	13.7	141.8	79.5	62.3
	September 30 – October 29	11.7	69	36.3	32.7
	October 30 – December 14	5.3	111	10.9	72.1
	Annual	**9.2**	**876**	**691**	**157.7**
2015	December 15 – March 1	1.3	132.5	31	101.5
	March 2 – March 29	4.5	36.5	36.8	-0.3
	March 30 – April 23	8.5	17.5	66.4	-48.9
	April 24 – May 28	11.0	66	113.6	-47.6
	May 29 – June 28	15.5	62.5	129.1	-66.6
	June 29 –July 23	21.1	26	136	-110
	July 24 – August 27	16.4	94.5	124.6	-30.1
	August 28 – September 24	12.8	77	66.3	10.7
	September 25 – October 29	7.8	55	36.1	18.9
	October 30 – December 11	7.0	54.5	25.1	29.4
	Annual	**9.4**	**585**	**766**	**-180.9**

Table 2. a) Repeated measure ANOVA is shown for treatment, date (December 2013, March 2014, June 2014) and interaction effects on root mass (g m^{-2}). Numerator (num), denominator (den) of degree of freedom (DF) and *F* values are shown. b) Root mass (g m^{-2}) of abandonment, low (Cattle-) and high (Cattle+) stocking density treatments measured in winter (December 12 2013), spring (March 20 2014), summer (June 20 2014) and averaged across the three dates. Means ± se are shown, n = 4. Superscripts ^{ns} correspond to $P > 0.05$.

a)	num/den DF	*F-value*
Treatment	2/8	1.151^{ns}
Date	2/18	2.027^{ns}
Treatment × date	4/18	1.340^{ns}

b) Date	Abandonment	Cattle-	Cattle+
December 2013	636.4 ± 133.1	403.3 ± 66.4	496.5 ± 20.6
March 2014	559.1 ± 166.2	609.2 ± 45.3	719.8 ± 47.5
June 2014	574.2 ± 84.8	482.2 ± 38.6	591.2 ± 101.7
3 dates average	589.9 ± 99.9	498.2 ± 43.6	602.5 ± 44.4

Table 3. Repeated measure ANOVA is shown for treatment, year and interaction effects on annual root production (BNPP, g m^{-2} y^{-1}), annual above-ground production (ANPP, g m^{-2} y^{-1}) and root to shoot ratio (R/S). Numerator (num), denominator (den) of degree of freedom (DF), *F* values are shown. Superscripts $^{ns, *, **, ***}$ correspond to $P > 0.05$, $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively.

		BNPP	ANPP	R/S
	num/den DF	*F-value*	*F-value*	*F-value*
Treatment	2/8	2.51^{ns}	8.10^{*}	0.46^{ns}
Year	1/9	70.72^{***}	83.77^{***}	13.09^{**}
Treatment × Year	2/9	3.83^{ns}	22.21^{**}	9.52^{**}

Table 4. Root traits measured from ingrowth core collected in June 2014 and leaf traits measured from botanical observation in abandonment (May 2014), Cattle- and Cattle+ (April 2014) treatments. Diameter: root diameter (mm); SRL: specific root length (m g^{-1}); RTD: root tissue density (g cm^{-3}); SRA: specific root area (m^2 g^{-1}); % 0-0.1 mm: percentage of length in the class diameter 0-0.1 mm; % 0.1-0.2 mm: percentage of length in the class diameter 0.1-0.2 mm; % 0.2-0.3 mm: percentage of length in the class diameter 0.2-0.3 mm; % > 0.3 mm: percentage of length in the class diameter > 0.3 mm; Community-weighted mean (CWM) Height: plant height (cm); SLA: specific leaf area (cm^2 g^{-1}); LDMC: leaf dry matter content (g g^{-1}). Means ± se are shown (n = 4). num/den DF: numerator and denominator of degree of freedom. Superscripts $^{ns, +, *, **, ***}$ correspond to $P > 0.1$, $P \leq 0.1$, $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively. For SRL and SRA, different letters correspond to significant differences between treatments.

	num/den DF	F-value	Abandonment	Cattle-	Cattle+
Root traits					
Diameter	2/8	1.61^{ns}	0.240 ± 0.015	0.210 ± 0.006	0.222 ± 0.015
SRL	2/8	3.71^{+}	237.2 ± 26.3 b	332.7 ± 30.4 a	277.8 ± 23.8 ab
RTD	2/8	0.55^{ns}	0.099 ± 0.007	0.095 ± 0.003	0.102 ± 0.007
SRA	2/8	4.96*	0.137 ± 0.011 b	0.182 ± 0.008 a	0.155 ± 0.01 ab
% 0-0.1 mm	2/8	1.28^{ns}	28.5 ± 1.1	32.9 ± 5.5	28.8 ± 2.6
% 0.1-0.2 mm	2/8	0.46^{ns}	37.7 ± 4.4	37.7 ± 2.2	39.1 ± 1.8
% 0.2-0.3 mm	2/8	0.30^{ns}	16.6 ± 1.2	16.2 ± 2.4	17.1 ± 1.9
% > 0.3 mm	2/8	1.22^{ns}	17.2 ± 5.0	13.2 ± 1.3	15.1 ± 2.1
Leaf traits					
CWM_Height	2/8	8.45^{*}	93.0 ± 3.5 a	72.8 ± 7.0 b	68.6 ± 3.8 b
CWM_SLA	2/8	5.30^{*}	205.1 ± 5.7 b	231.8 ± 7.3 a	225.5 ± 7.1 ab
CWM_LDMC	2/8	11.22^{*}	0.261 ± 0.008 a	0.227 ± 0.007 b	0.213 ± 0.010 b

Table 5. Contribution of the different variables to the first two axes of the principal component analysis (PCA) calculated for 2014 and 2015. Variables used in the PCA were annual relative soil water content (RSWC), annual soil temperature (Tsoil, °C), root diameter (Diam, mm), specific root area (SRA, m^{2} g^{-1}), root mass averaged over three dates (RootMass, g m^{-2}), annual root production (BNPP, g m^{-2} y^{-1}), plant height (Height, cm), leaf dry matter content (LDMC, g g^{-1}), annual above-ground production (ANPP, g m^{-2} y^{-1}). Treatments were added as supplementary categories.

	2014		2015	
Variable	Axis 1 (43.4 %)	Axis 2 (16.7 %)	Axis 1 (37.4 %)	Axis 2 (19.4 %)
RSWC	0.62	0.44	-0.21	**0.64**
Tsoil	**0.91**	0.09	-0.58	0.52
Diam	**-0.64**	**0.75**	**0.78**	0.53
SRA	**0.62**	**-0.58**	**-0.69**	-0.48
RootMass	-0.06	0.22	-0.07	**0.60**
BNPP	0.21	-0.23	**-0.71**	0.35
Height	**-0.82**	-0.07	**0.83**	-0.19
LDMC	**-0.83**	-0.12	**0.61**	0.03
ANPP	**0.71**	0.54	**0.57**	0.20
Suppl. Categories				
Abandonment	**-2.62**	-0.24	**2.04**	-0.27
Cattle-	1.07	-0.55	-1.21	-0.62
Cattle+	0.70	0.18	-0.83	0.90

Contribution in bold indicates significant correlation of the variables on the PCA axis ($P < 0.05$).

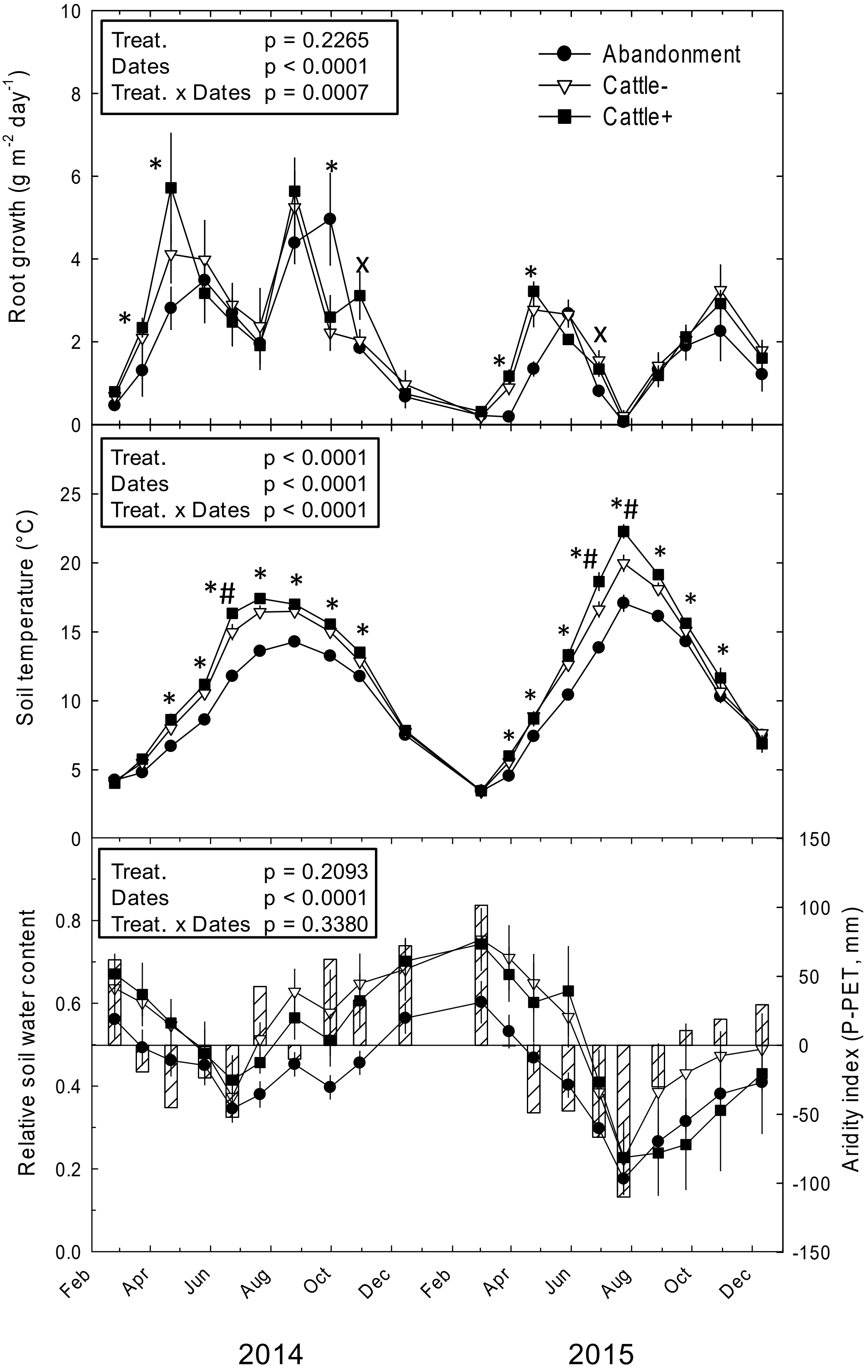
Figure captions

Figure 1. Dynamics of root growth (g m^{-2} day^{-1}), soil temperature (°C), relative soil water content and an aridity index (P-PET, mm) (hashed bars), measured over two years for abandonment, low (Cattle-) and high (Cattle+) stocking density treatments. Vertical bars correspond to 1 se (n = 4). Insets indicate P values from repeated measure two-tailed ANOVA (Treat: treatment, dates and interaction for main treatments). *: $P < 0.05$; x: $P \leq 0.1$. For soil temperature, *# corresponds to significant differences between all treatments (Abandonment < Cattle- < Cattle+).

Figure 2. Annual root biomass production (BNPP, g m^{-2} y^{-1}), annual above-ground biomass production (ANPP, g m^{-2} y^{-1}) and root-to-shoot biomass ratio measured in 2014 and 2015 for abandonment, low (Cattle-) and high (Cattle+) stocking density treatments. Vertical bars correspond to 1 se (n = 4). Within a year, different letters correspond to significant differences at $P < 0.05$.

Figure 3. Principal component analysis (PCA) combining leaf and root traits, above- and below-ground net primary production, root mass stock, relative soil water content and soil temperature measured in 2014 (a) and 2015 (b) for abandonment, low (Cattle-) and high (Cattle+) stocking density treatments. Data of each plot were used in each PCA. The first two axes are shown. Arrows show projections of the variables within the PCA. RSWC: relative soil water content; Tsoil: soil temperature (°C), Diam: root diameter (mm), SRA: specific root area (m^2 g^{-1}), RootMass: root mass averaged over 3 dates (g m^{-2}), BNPP: annual root production (g m^{-2} y^{-1}), Height: plant height (cm), LDMC: leaf dry matter content (g g^{-1}) and ANPP: annual above-ground production (g m^{-2} y^{-1}).

822 Figure 1



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825 Figure 2

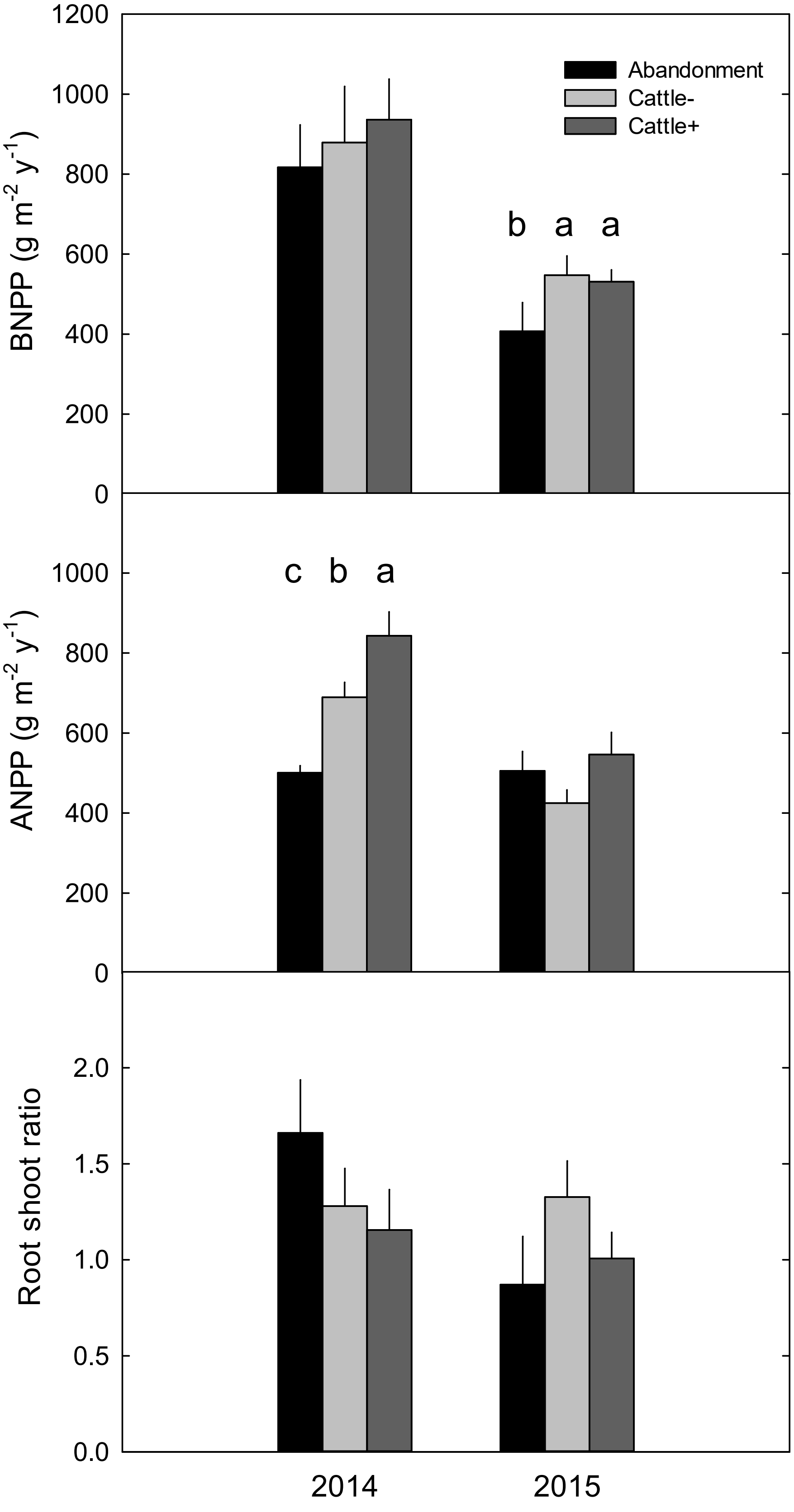

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Figure 3

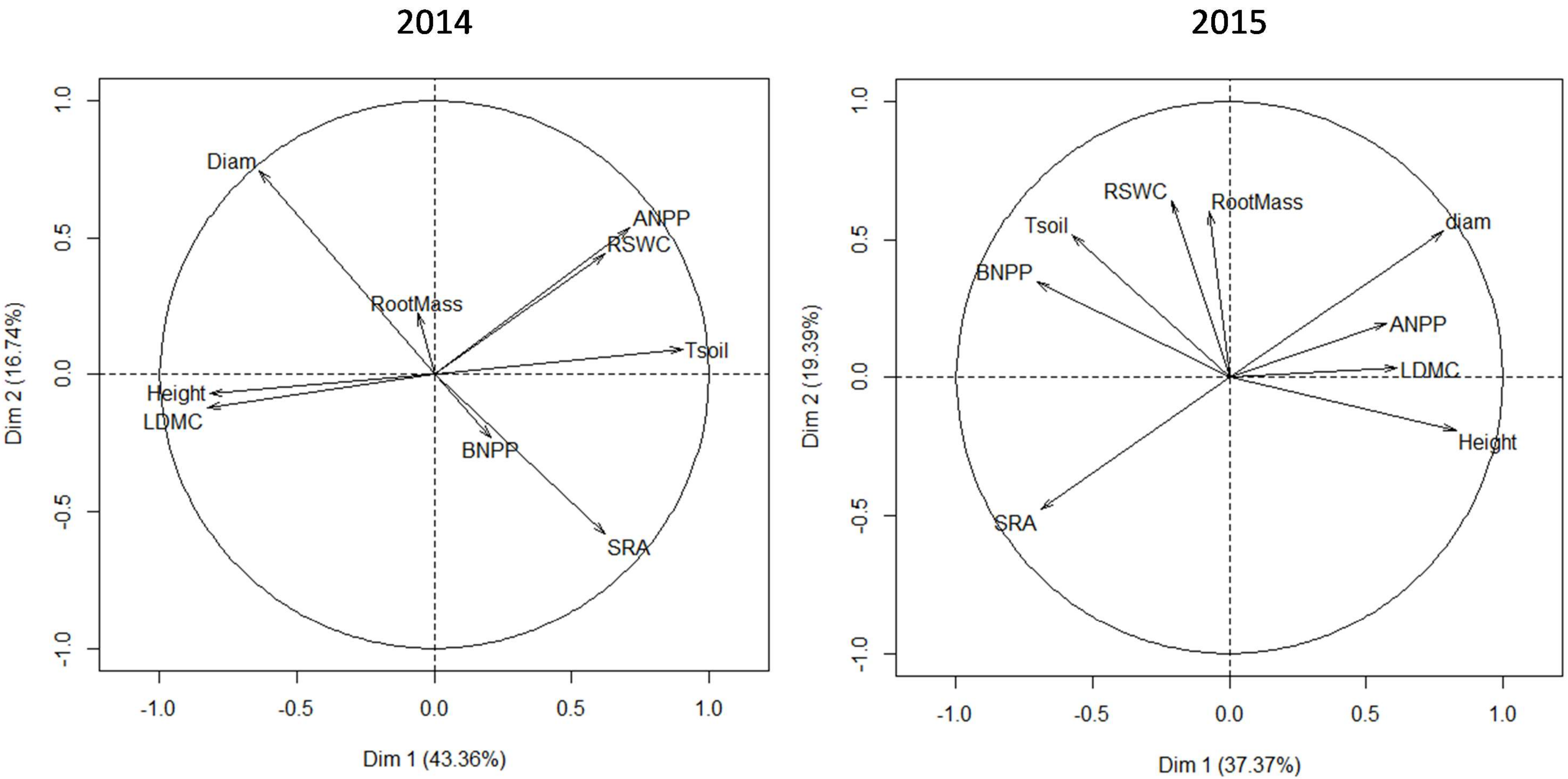
2014
Diam
ANPP
RSWC
RootMass
Tsoil
Height
LDMC
BNPP
SRA
Dim 1 (43.36%)
Dim 2 (16.74%)
-1.0
-0.5
0.0
0.5
1.0
2015
RSWC
RootMass
Tsoil
BNPP
diam
ANPP
LDMC
Height
SRA
Dim 1 (37.37%)
Dim 2 (19.39%)
-1.0
-0.5
0.0
0.5
1.0

Table S1. Repeated measure ANOVA is shown for root growth (g m^{-2} day^{-1}), soil temperature (Tsoil, °C) and relative soil water content (RSWC) responses to treatment, dates (d1 to d20) and interaction effects. Numerator (num), denominator (den) of degree of freedom (DF) and *F* values are shown. Superscripts ^{ns}, **, *** correspond to $P > 0.05$, $P < 0.001$, $P < 0.0001$, respectively.

Variables	Treatment		Dates		Treat. x Dates	
	num/den DF	F-value	num/den DF	F-value	num/den DF	F-value
Root growth	2/8	1.80^{ns}	19/171	50.40^{***}	38/171	2.096^{**}
Tsoil	2/8	33.93^{***}	19/166	944.83^{***}	38/166	9.75^{***}
RSWC	2/8	1.914^{ns}	19/163	25.287^{***}	38/163	1.097^{ns}

Table S2. Nitrogen nutrition index (NNI %, Lemaire and Gastal 1997, Cruz et al. 2006) measured on forage regrowth of May in 2014 and 2015 on the non-leguminous part to assess the effect of treatments on N availability according to grazing intensity. When legumes were below 4.5% in the herbage mass, NNI was assessed using the procedure defined by Cruz et al (2006) based on the total forage and the legume contribution. The P-values are associated with a nested mixed model: treatment used as fixed factor with plots nested in blocks as random factors. Mean ± se is shown (n = 4). For each year, different letters correspond to significant differences at P < 0.05.

Year	*P-value*	Abandonment	Cattle-	Cattle+
2014	0.146	65.64 ± 3.10 a	59.54 ± 1.78 a	63.72 ± 2.86 a
2015	0.018	69.72 ± 1.19 a	61.71 ± 1.53 b	69.25 ± 2.09 a

For each year, different letters correspond to significant differences at *: P < 0.05; **: P < 0.01; ***: P < 0.001; ns: P > 0.05.

References

Lemaire G, Gastal F (1997) N uptake and distribution on plant canopy. In: Lemaire, G (ed.) Diagnosis of the nitrogen status in crops, pp. 3-43. Springer-Verlag, Berlin, DE.

Cruz P, Jouany C, Theau J-P, Petibon P, Lecloux E, Duru M (2006) L'utilisation de l'indice de nutrition azotée en prairies naturelles avec présence de légumineuses. Fourrages 187:369-376.

Table S3. Species contribution (%) in the community present around the ingrowth core measured in April and May 2014 for Cattle-, Cattle+ and Abandonment, respectively. Mean ± se is shown (n = 4). For each species, different letters correspond to significant differences at *: P < 0.05; **: P < 0.01; ***: P < 0.001; ns: P > 0.05.

Group	Species	*P-value*	Abandonment	Cattle-	Cattle+
Grasses	*Agrostis capillaris*	ns	0.0 ± 0.0	0.6 ± 0.6	1.7 ± 1.2
	Arrhenatherum elatius	ns	10.3 ± 6.8	2.2 ± 2.2	2.5 ± 2.5
	Alopecurus pratensis	**	27.2 ± 7.9 a	7.8 ± 3.3 b	3.3 ± 1.7 b
	Dactylis glomerata	*	3.1 ± 2.7 b	22.2 ± 9.8 a	9.1 ± 3.8 ab
	Elytrigia repens	*	18.8 ± 9.9 a	2.8 ± 1.8 b	3.8 ± 2.7 b
	Schedurus arundinaceus	ns	5.0 ± 2.3	5.6 ± 2.1	6.3 ± 2.2
	Holcus lanatus	*	0.0 ± 0.0 b	4.7 ± 1.6 a	3.4 ± 1.9 a
	Lolium perenne	***	0.0 ± 0.0 b	0.9 ± 0.9 b	13.6 ± 3.8 a
	Poa pratensis	ns	11.3 ± 2.2	3.1 ± 1.5	3.4 ± 2.5
	Poa trivialis	*	0.0 ± 0.0 b	5.0 ± 2.5 a	7.2 ± 2.4 a
	Trisetum flavescens	ns	0.0 ± 0.0	2.2 ± 1.3	0.6 ± 0.4
Forbs	*Achillea millefolium*	ns	1.3 ± 0.9	3.8 ± 2.4	3.1 ± 2.3
	Anthriscus sylvestris	ns	2.5 ± 2.1	0.0 ± 0.0	0.0 ± 0.0
	Cerastium fontanum	ns	0.0 ± 0.0	1.3 ± 0.9	0.0 ± 0.0
	Cerastium glomeratum	ns	0.0 ± 0.0	0.0 ± 0.0	0.3 ± 0.3
	Cirsium arvense	ns	5.0 ± 3.5	0.0 ± 0.0	0.0 ± 0.0
	Hypocheris radicata	ns	0.0 ± 0.0	0.9 ± 0.9	0.0 ± 0.0
	Ranunculus acris	ns	0.0 ± 0.0	0.0 ± 0.0	3.8 ± 3.8
	Stellaria graminea	ns	0.6 ± 0.6	0.6 ± 0.4	0.0 ± 0.0
	Taraxacum officinale agg.	**	0.0 ± 0.0 b	17.5 ± 1.8 a	19.1 ± 6.0 a
	Urtica dioïca	*	9.7 ± 4.9 a	0.0 ± 0.0 b	0.0 ± 0.0 b
	Veronica serpyllifolia	ns	0.0 ± 0.0	0.3 ± 0.3	0.0 ± 0.0
Legumes	*Lathyrus pratensis*	ns	0.0 ± 0.0	0.3 ± 0.3	0.3 ± 0.3
	Trifolium pratense	ns	0.0 ± 0.0	0.0 ± 0.0	0.3 ± 0.3
	Trifolium repens	***	0.0 ± 0.0 b	16.3 ± 4.0 a	17.7 ± 2.5 a

Fig S1 : Scheme of the plots and blocks on the experimental site