This is a post-peer-review, pre-copyedit version of an article published in Organic Agriculture.
The final authenticated version is available online at: https://doi.org/10.1007/s13165-015-0137-z

Title page:

Yield and herbage quality from organic grass clover leys - a meta-analysis of Norwegian field trials

Håvard Steinshamn, Steffen A. Adler, Randi B. Frøseth, Tor Lunnan, Torfinn Torp and Anne Kjersti Bakken

Affiliation and addresses

Håvard Steinshamn, Steffen Adler and Randi B Frøseth: NIBIO - Norwegian Institute of Bioeconomy Research, Gunnars veg 6, 6630 Tingvoll, Norway

Tor Lunnan: NIBIO - Norwegian Institute of Bioeconomy Research, Nyhagevegen 35, 2940 Heggenes, Norway

Torfinn Torp: NIBIO - Norwegian Institute of Bioeconomy Research, P.O. Box 115, 1431 Ås, Norway

Anne Kjersti Bakken: NIBIO - Norwegian Institute of Bioeconomy Research, Vinnavegen 38, 7512 Stjørdal, Norway

Corresponding author, e-mail, telephone:

Håvard Steinshamn, havard.steinshamn@nibio.no, +47 404 80 314

Abstract

A meta-analysis based on experiments in organically cultivated grasslands in Norway was conducted to quantify the effects of management factors on herbage yield and feed quality. A data set was collected that included 496 treatment means from experiments in five studies carried out at eight locations with the latitude range of 58.8°N to 69.6°N between 1993 and 2010. We tested the effect of harvesting system (two *vs.* three cuts annually), plant developmental stage at the first cut, growth period (temperature sum) and the herbage clover proportion. Plant maturity at the first cut and herbage clover proportion explained to a large extent herbage yield and quality of the first cut and annual yield. The timing of the first cut influenced also the yield and herbage quality of the second cut. The analysis confirmed the importance of legumes performance for herbage yield and quality from grasslands in organic production. Estimated annual herbage DM yield harvested at standardized plant development stage and at average clover proportion was 9% higher in the two- compared to the three-cut system. The crude protein concentration and in vitro dry matter digestibility was 17% and 3% higher and the NDF concentration 7% lower in the annual herbage from the three-cut than from the two-cut system, respectively.

The empirical equations developed in this study may be applied to explore different options for grassland management as basis for ration and production planning and in scenario analysis of economic performance of individual and model farms. The equations do also reveal in numeric terms the trade-offs in management practice between high yields, yield digestibility, NDF and crude protein content in organic forage production relying on red clover N_2 fixation as the engine in the system.

Keywords;

Grass-legume mixture; herbage yield; herbage quality; empirical relationships

Introduction

Organic dairy milk production in Norway has been intensified. The annual yield per cow has increased by 18%, from 5890 kg in 2007 to 6960 kg in 2013, largely driven by increased plane of nutrition; concentrate feeding increased 28%, from 9050 to 11540 MJ Net Energy Lactation (NEL) per cow and year, in the same period (TINE Rådgivning 2014). Grass clover silage is the basic feedstuff and accounts currently for about 47% of the total NEL intake, approximately 15500 MJ NEL/cow and year (TINE Rådgivning 2014). The intensification in feeding is accompanied by increased focus by the dairy farmers on achieving high forage quality by taking the first cut at an early stage of plant development and by taking three cuts per year instead of two. Grassland yield in organic production depends on growth factors such as soil and climate, weather during both winter and the growing season, genetic potential, botanical composition, in particular content of legumes, fertilization level and harvesting system (Riesinger and Herzon 2008; Steinshamn 1997; Steinshamn 2001). Herbage feed quality is largely influenced by the same factors, but phenological stage of development and cutting frequency are probably the most important factors that influence the nutritive value of preserved forage (Buxton 1996). In organically managed ruminant production systems, grasslands legumes have an essential role due to their symbiotic N_2 fixation, high yield capacity and high feeding value. The regrowth after the first and second cut contain more clover than the spring growth (Eriksen et al. 2012; Riesinger and Herzon 2008; Steinshamn and Thuen 2008). The regrowth herbage has, therefore, usually higher crude protein (CP) concentration than the first growth, while the first cut herbage usually has higher energy value (Bakken et al 2014; Lunnan 2004a; Steinshamn 1997). Under Fennoscandian conditions, the cultivated grassland species, dominated by timothy (*Phleum pratense* L), meadow fescue (*Festuca pratensis* Huds.) and red clover (*Trifolium pratense* L.), have rapid generative growth during spring that generate high yield but also fast decline in feed quality due to increase in stem leaf ratio. In Norway, the majority of the grasslands used for silage production is harvested twice annually. In order to produce high quality silage, the seasonal first cut should be taken at early stage of development, which may necessitate an additional cut annually. Such intensification, with early first cut and increased harvesting frequency, may however compromise annual DM yield and the persistency of the sown species (Grønnerød 1988). Changes in cutting time and frequency will also change the proportion each cut contributes to the annual yield and it affects the nutritional quality in each cut. Less is known about how the yield level and quality of the total annual yield are influenced by cutting regimes, i.e. if the grassland is cut two or three times annually. This knowledge is needed for strategical planning of the grassland management in order to meet the herbage yield and quality level requirements of the planned livestock production. Trials in organically cultivated grassland have been carried out in Norway since the early 1990s. The objectives were various, but in most studies harvesting system, either the time of the first cut or number of cuts per season, or the effect of seed mixtures were tested. The results from each of these studies have given valuable information on herbage yield and quality of organically managed grassland under Norwegian conditions. However, we believe that by combining the findings from these studies using statistical methods it is possible to improve the precision of the quantitative relationships between agronomic factors and herbage yield and quality (Halling et al. 2004). Thus, the objective of the present study was to work out generalized and quantitative relationships between different management options that can serve as basis for strategic planning of organic forage production.

Material and methods

Data material

Data were collected from cultivation experiments in organically managed grasslands carried out in the period from 1994 to 2010 on experimental stations in Norway (Frøseth et al. 2014; Johansen et al. 2008; Lunnan 2004a; Lunnan 2004b; Røthe et al. 2007). Data were extracted from both published and unpublished sources as the average across replicates within each cut. The management of the experiments was carried out according to the prevailing national organic standards, which is in accordance with EU standards (Commission 1991), at the time when the experiments were laid out. Only data from experiments where clover proportion of DM yield and herbage feed quality were measured was included. Furthermore, it was required that at least two cuts and maximum three cuts per season were taken. Data from the first to the fourth production year were included. An overview of the experiments with experimental factors, location and production years, is presented in Table 1a and b. The main experimental factor in the experiments was cutting system, and in many of the experiments, it was combined with seed mixtures. Fertilization level with animal manure was confounded with study and location, and within study (Table 1a). Thus, it was not possible to test the effect of fertilization. Seed mixtures tested varied between studies and partly also between locations within study, which made it not feasible to do an overall test of seed mixture. In all studies and locations, the grass species timothy (*Phleum pratense* L.) and meadow fescue (*Festuca pratensis* Huds.) were included in mixtures with each other or in mixtures with other species (Table 1b). Red clover (*Trifolium pratense* L.) was included as the legume species in all studies, either as the sole legume species or in mixture with white clover (*Trifolium repens* L.). Results from treatments seeded with only grass at establishment were included in the analysis. The cutting systems compared were either development stage in the first cut or number of cuts per season. We were able to test the effects of cutting system (2 *vs* 3 cuts), development stage in the first cut, length of the regrowth period and proportion of clover in herbage DM yield.

Calculations

Plant development stage at the first cut was determined differently in the studies. In order to have a uniform measure, the commencement of growth in spring and the phenological stage of timothy at the first cut, expressed as mean stage by count (MSC, Moore et al. 1991), was estimated using a grassland growth model (Bonesmo 2004) and weather data for each study, location and year. In the regrowth, we used the effective temperature sum (accumulated mean daily temperatures using 0 °C as base temperature) between cuts as the indicator of plant maturity. Herbage clover proportion and feed quality parameters in annual yield (sum of all cuts per year) were calculated as the weighted mean of all cuts.

Herbage quality analysis

Herbage samples in all studies were dried at 60°C for 48 h and thereafter chopped and milled to pass through a 1-mm screen using a Cyclotec™ 1093 Sample mill (Foss Companies, Hillerød, Denmark). The herbage concentrations of crude protein (CP), in vitro dry matter digestibility (IVDMD), neutral detergent fibre (NDF), and herbage clover proportion were determined at NIBIO Løken, Vollbu, Norway, by near-infrared reflectance spectrometry (Fystro and Lunnan 2006).

Statistical analysis

The data was analysed using the mixed model procedure (proc mixed) in SAS (SAS 2004). Three different models were used for 1) first cut data, 2) second cut data, and 3) annual yield data. The models were:

1. $Y_{ijkl} = \mu + S_i + L_{j(i)} + A_{k(ij)} + \phi_l + \beta_1 d + \beta_2 d^2 + \beta_3 c + \beta_4 c^2 + \beta_5 d\cdot c + \varepsilon_{ijkl}$

2. $Y_{ijkl} = \mu + S_i + L_{j(i)} + A_{k(ij)} + \phi_l + \beta_1 t + \beta_2 t^2 + \beta_3 d + \beta_4 t\cdot d + \beta_5 c + \beta_6 c^2 + \beta_7 t\cdot c + \varepsilon_{ijkl}$

3. $Y_{ijklm} = \mu + S_i + L_{j(i)} + A_{k(ij)} + \phi_l + \eta_m + \beta_1 d + \beta_2 d^2 + \gamma_m d + \beta_3 t + \beta_4 t^2 + \xi_m t + \beta_5 d\cdot t + \beta_6 c + \beta_7 c^2 + \tau_m c + \beta_8 t\cdot c + \beta_9 c\cdot d + \varepsilon_{ijklm}$

Y_{ijkl} and Y_{ijklm} are the dependent variables (yield and feed quality), μ is the overall mean, S_i is the random effect of Study i, $L_{j(i)}$ is the random effect of Location j within Study i, $A_{k(ij)}$ is the random effect of Year k within Location j within Study i, ϕ_l is the fixed effect of Production year l, η_m is the fixed effect of cut m (model 3), d (d_{ijkl} in model 1 og 2, and d_{ijklm} in model 3) is the fixed effect of phenological stage (continuous variable), c (c_{ijkl} in model 1 and 2, and c_{ijklm} in model 3) is the fixed effect of the herbage clover proportion (continuous variable), t (t_{ijkl} in model 2, and t_{ijklm} in model 3) is the fixed effect of the temperature sum between the first and second cut (continuous variable). ε_{ijkl} in model 1 and 2, and ε_{ijklm} in model 3 are the random error terms. The β_n, γ_m, ξ_m and τ_m are the regression coefficients. γ_m, ξ_m, and τ_m account for interactions between cut *m* and *d*, cut *m* and *t*, and cut *m* and *c* respectively.

We used REML («Restricted maximum likelihood») to estimate the variance components in the model, and if the models did not converge MIVQUE0 («Minimum variance quadratic unbiased estimators») was used. The fixed effect terms were included if their P-values were less than the significance level 5%. However, the effect of development stage (*d*), in model 1 and 3, and temperature sum (*t*) between first and second cut, in model 2 and 3, were included even when their P-value was greater than 5%. Factors included in an interaction necessitated that the factors themselves are also included in the model. If an interaction was included, the terms in that interaction were not tested because the meaning of such terms may be misinterpreted. Likewise, a linear term was not tested if it is included in a quadratic term.

The final models for the response variables, e.g. herbage DM yield and CP concentration, may include many terms with interactions. Expressed as estimate of multiple regression coefficient, the results may be difficult to understand and interpret. We therefore choose graphical presentation of the agronomical interesting features of the models, according to the same principles as used by Wachendorf et al. (2001). Briefly, predictions for the crop maturity (phenological stage in the first cut and temperature sum after first cut in regrowth) by herbage clover proportion interactions are presented as curve for a range of maturity stages (continuous) and for a low and high level of clover proportion (approximately its mean ± s.d.). The range used for prediction from the independent variables was selected to exclude values close to the observed minimum and maximum of the variable, and predictions outside the range of the observed data were excluded. In addition, we used the LSMEANS/AT, CONTRAST and ESTIMATE statements in the SAS MIXED procedure to test the effect of cutting system, two *vs.* three cuts, on annual DM yield and herbage quality. The timing of the first cut and second cut were standardized to MSC=2.8 and 1 000 d°C in the two-cut system and MSC=2.4 and 600 d°C in the three-cut system, respectively, and the herbage clover proportion was set to the 0.25 in both systems.

Results

For all variables describing the dataset, there were wide ranges in score (Table 2). The statistical analysis with parameter estimates are presented in Tables 3-6, and the relationships between the independent variables plant development stage, length of regrowth period and herbage clover proportion and the response variables herbage yield and forage quality parameters are illustrated in Fig. 1-5.

Herbage yield

Herbage DM yield increased with increasing plant maturity in both spring growth and regrowth (Table 3, Fig. 1). Second cut yield was higher after an early first than late first cut, at early stage of regrowth (Fig 1.b). However, the regrowth yield increase with ageing of the plant stand was steeper after a late than early first cut (positive temperature sum by phenological stage at first cut interaction, Table 3, Fig. 1b). The effect of length of regrowth period after first cut on annual yield was small and not significant (Table 3, Fig. 1c). Herbage DM yield increased at a decreasing rate with increasing herbage clover proportion both in the spring growth, regrowth and in annual yield, as indicated by the negative quadratic term (Table 3, Fig. 2). The optimum clover proportion for herbage DM yield was 0.45-0.50 in the first cut and 0.60-0.70 for the annual yield (Table 3, Fig. 2). The positive effect of clover on herbage yield increased with plant maturity (phenological stage) in the first cut and in the regrowth (temperature sum) and, consequently, also for the annual yield as seen by the positive plant maturity by herbage clover proportion interaction (Table 3, Fig. 1 and 2). Furthermore, the positive effect of clover on annual yield was stronger in the three-cut than in the two-cut system (cutting system by clover proportion interaction, Table 3).

Herbage digestibility

The herbage IVDMD declined with plant maturity in the spring growth, in regrowth, and, consequently, in the annual yield (Table 4, Fig. 3). Herbage clover proportion had no effect in the spring growth but increasing clover proportion decreased herbage IVDMD in the regrowth and annual yield (Table 4, Fig. 3). Interactions modified the clover effect in the regrowth, as the lowering effect became stronger with increasing plant maturity (negative temperature sum by clover proportion interaction, Table 4, Fig. 3) and was weaker after an early than late first cut (negative development stage by clover interaction, Table 4). Timing of the first cut had small effect on

IVDMD of the annual yield in the three-cut system, but IDVMD declined with advanced maturity in the two-cut system (positive cutting system by temperature sum interaction, Table 3b, Fig. 2c). The negative effect of regrowth length (temperature sum) on IVDMD was stronger in the three-cut than in the two-cut system (positive cutting system by temperature sum interaction, Table 4, Fig. 3), and the effect of regrowth period increased with delayed harvest time in the first cut (negative development stage in the first cut by temperature sum interaction). The negative effect of clover on IVDMD in annual yield diminished with delayed harvest time of the first cut (positive development stage by clover interaction, Table 4).

Herbage crude protein

The CP concentration deceased with advancing plant stand maturity in spring growth, regrowth and in annual yield (Table 5, Fig. 4). A late first cut resulted in higher herbage CP concentration at the early stage of regrowth than an early first cut (negative phenological stage in the first cut by regrowth temperature sum interaction, Table 5, Fig. 4b). The CP in regrowth herbage decreased with the duration of the period, which also was reflected in the annual yield (Fig. 4b and c). The CP concentration increased strongly with increasing clover proportion in spring growth, regrowth and in annual yield (Table 5, Fig. 4). The clover effect on regrowth CP was stronger after a late than early first cut, which also was reflected in the annual herbage (positive clover proportion by phenological stage in the first cut interaction, Table 5, Fig. 4b and d). The CP in annual yield decreased with delayed first cut, both in the two- and three- cut system. However, the effect of phenological stage was stronger in the two-cut system (negative cutting system by phenological stage of first cut interaction, Table 5, Fig. 4c). The positive effect of clover on CP of annual yield diminished with the length of the regrowth period (negative clover proportion by temperature interaction).

Herbage neutral detergent fibre

The herbage fibre concentration, measured as neutral detergent fibre, increased with plant maturity and decreased with increasing herbage clover proportion (Table 6, Fig. 5). Herbage NDF concentration in regrowth was affected by the timing of the first cut by decreasing with advancing spring growth stage (Table 6, Fig. 5). Herbage NDF decreased with maturity (temperature sum) after an early first cut and increased after a late first cut (positive quadratic term of temperature sum, Table 6). Clover had strong impact on herbage NDF and decreased with increasing proportion in both spring growth and in regrowth, and consequently in annual yield. For each unit (0.01) increase in herbage clover proportion, NDF concentration decreased by approximately 2 and 1 g kg DM^{-1} in spring and regrowth herbage, respectively. The clover lowering effect on NDF was stronger in the three- than in the two-cut system and increased with plant maturity of the first cut (Cutting system by clover proportion interaction and MSC by clover proportion interaction, Table 6, Fig. 5d).

Cutting system

Cutting system, two *vs.* three cuts annually, had strong impact on herbage yield and quality of each individual cut. However, when compared at annual level, there were small differences in yield between two and three cut systems. First cut at developmental stage MSC = 2.4 and 2.8 in a standard three- and two-cut system, respectively, gave on average 9% higher DM yield in the two- than the three-cut system (7200 *vs.* 7837, SE 450 kg DM ha^{-1}, P=0.005). The weighted mean annual herbage CP concentration was on average 17% higher in the three- compared to two-cut system (124 vs. 103, SE 6.7 g kg DM^{-1}, P<0.001). The effect of cutting system was otherwise relatively small for the annual weighted herbage quality parameters IVDMD and NDF as the three-cut system resulted on average 3% higher IVDMD (737 *vs.* 717, SE 27 g kg DM^{-1}, P=0.051) and 7% lower NDF (502 *vs.* 535, SE 16 g kg DM^{-1}, P<0.001) concentrations.

Discussion

Plant maturity and legume proportion explained to a large extent the variation in herbage yield and quality in the present study. Estimates of variance components (not shown) for the selected models indicate that there were effects of year within location for most variables analyzed. Environmental factors alter herbage quality, even when analyzed at similar maturity stage and these factors may differ between years. Such factors are temperature, water deficit, solar radiation, and nutrient availability (Buxton 1996). We were not able to take into account the effect of the grass species or cultivars included, and in particular, the proportion of different grass species in the herbage has most likely influenced both yield and quality.

Herbage yield

Higher herbage yield in the early stage of regrowth of an early compared to a late first cut is likely due to higher proportion of generative shoots in the regrowth after an early than a late first cut. Generative shoots have higher growth potential than vegetative shoots that dominate in grass regrowth after a late first cut (Bonesmo 2000; Bonesmo and Skjelvåg 1999). The positive relationship between herbage clover proportion and DM yield is in accordance with other findings in grasslands with low nitrogen input (Fagerberg and Torssell 1995; Kleen et al. 2011; Mallarino and Wedin 1990; Steinshamn 2001). Annual yields were highest at a legume proportion of about 0.60-0.70, which is similar to the level 0.62-0.67 Mallarino and Wedin (1990) found in experiments with red clover-tall fescue mixtures. Many studies have demonstrated higher DM yield in mixtures of grass and legumes than the highest yielding pure stands of the same species, even at high nitrogen fertilization rates (Finn et al. 2013; Kirwan et al. 2007; Sturludóttir et al. 2014). One reason for stronger yield increase with high than low clover proportion with increasing plant maturity in spring growh is likely due to red clover's slower rate of development than the companion grasses (Fagerberg 1988; Steinshamn 1997). Another reason why clover proportion was more important for dry matter production at late than early developmental stages was probably increasing N dilution and limitation parallel to advancement in development and dry matter accumulation (Lemaire and Gastal 1997). The N limitation got more severe in canopies with a low than a high clover content.

Herbage digestibility

Previous studies have revealed that digestibility of the spring growth herbage improves with increasing herbage red clover proportion (Lunnan 1989; Steinshamn 1997), and that the declining rate with maturity is slower in red clover than in timothy, the dominating grass species in Scandinavian leys (Hetta et al. 2004; Rinne and Nykänen 2000; Steinshamn 1997). This positive effect of red clover on digestibility in spring growth is due to slower development rate of red clover and higher leaf stem ratio than in companion grasses (Buxton and Brasche 1991; Steinshamn 1997). The reason why we did not find this effect of clover in the current study is uncertain, but other grass species than timothy were also included in the mixtures in the current study. Another possibility is that in the grass dominating treatments included in the current meta-analysis, the concentration of non-structural carbohydrates with high digestibility were probably high due to low N supply. The clover lowering effect on IVDMD in regrowth herbage is in accordance with other findings (Buxton and Redfearn 1997; Fagerberg and Ekbohm 1995; Øyen and Aase 1988). This is because the regrowth of red clover is dominated by generative shoots with lower leaf stem ratio than the spring growth, and the highly lignified stems of red clover reduce the IVDMD (Buxton and Hornstein 1986).

Crude protein

Higher herbage CP concentration in early stage of regrowth after a late compared to an early first cut is likely due to higher proportion of vegetative shoots and lower yield level in the sward after a late cut. Regrowth of red clover has high proportion of stems and lower leaf stem ratio than spring growth. Stronger decrease in CP concentration in annual yield with delayed first cut in the two-cut than in the three-cut system is because of higher proportion of first cut yield in the annual yield in a two-cut system. The strong effect of herbage clover proportion on herbage CP concentration is in accordance with other studies on grass-clover leys with low nitrogen fertilization levels (Fagerberg and Ekbohm 1995; Gierus et al. 2012; Kleen et al. 2011; Steinshamn 1997).

NDF

The increased herbage NDF concentration with plant maturity is because accumulation of stem mass exceeds increment of leaf mass and thus the leaf stem ratio decreases (Buxton 1996). Stems contain more cell wall and less photosynthetic active tissues than leaves (Wilson and Kennedy 1996). The clover lowering effect on NDF is due to the general lower NDF concentration in legumes than in grass. Higher fiber concentrations at the start of the regrowth period after an early than late cut is as for CP due to higher proportion of stems after an early cut. Stronger clover lowering effect on the fiber content of the annual yield in the three- than in the two-cut system is likely due to the contribution from the third cut, as the third cut in general had low NDF concentration (figures not shown).

Conclusion and Implications

The two-cut system yielded annually more DM than the three-cut system and the difference in herbage feed quality between cutting system was modest, which implies that economically the two-cut system is superior to the three-cut system. The differences in annual DM yield, IVDMD and CP concentration in annual yield between the two- and three-cut systems diminished while the difference in NDF concentration increased with increasing clover proportion. Thus, the feeding value of the herbage from the three-cut system, relative to the two-cut system, is higher with increasing clover proportion. However, the strategic choice between two- or three-cuts annually depends on many factors such as total feed requirement, farm area and animal production goal. The equations developed may be used to quantify the effect of management factors, like cutting frequency, timing of cuts on herbage yield and quality under different climatic conditions. The accuracy of the estimates is modest, but for strategic management planning it will probably be sufficient. Data generated may be used in ration and dairy production planning and further in economic consequence analysis of management options.

Funding: The project was funded by the Norwegian Agricultural Agreement Research Fund (Project number 207755 in The Research Council of Norway), the County Governors of Sør- and Nord-Trøndelag, the Sør- and Nord-Trøndelag County Authorities, TINE SA and the Norwegian Agricultural Extension Service.

Conflict of Interest: The authors declare that they have no conflict of interest.

References

Bakken AK, Vaga M, Hetta M, Randby ÅT, Steinhsamn H (2014) Feed value of restrictedly and extensively fermented organic grass-clover silages from spring and summer growth. Grassland Science in Europe 19: 603-605.

Bonesmo H (2000) Regrowth rates of timothy and meadow fescue as related to the content of remaining water-soluble carbohydrates and non-elongated tillers. Acta Agr Scand B-S P 50:22-27 doi:10.1080/090647100750014376

Bonesmo H (2004) Phenological development in timothy and meadow fescue as related to daily mean temperature and day length. Grassland Science in Europe 9: 799-801.

Bonesmo H, Skjelvåg AO (1999) Regrowth rates of timothy and meadow fescue cut at five phenological stages. Acta Agr Scand B-S P 49:209-215 doi:10.1080/713782028

Buxton DR (1996) Quality-related characteristics of forages as influenced by plant environment and agronomic factors Anim Feed Sci Tech 59:37-49 doi: 10.1016/0377-8401(95)00885-3

Buxton DR, Brasche MR (1991) Digestibility of structural carbohydrates in cool-season grass and legume forages. Crop Sci 31:1338-1345 doi:10.2135/cropsci1991.0011183X003100050052x

Buxton DR, Hornstein JS (1986) Cell-wall concentration and components in stratified canopies of alfalfa, birdsfoot trefoil, and red clover. Crop Sci 26:180-184 doi:10.2135/cropsci1986.0011183X002600010043x

Buxton DR, Redfearn DD (1997) Plant limitations to fiber digestion and utilization. J Nutr 127:814S-818S

Commission E (1991) Council Regulation (EEC) No 2092/91 of 24 June 1991 on organic production of agricultural products and indications referring thereto on agricultural products and foodstuffs. Official Journal of the European Communities L 198:1-15

Eriksen J, Askegaard M, Søegaard K (2012) Complementary effects of red clover inclusion in ryegrass–white clover swards for grazing and cutting. Grass Forage Sci 69:241-250 doi:10.1111/gfs.12025

Fagerberg B (1988) Phenological development in timothy, red clover and lucerne. Acta Agr Scand 38:159-170 doi:10.1080/00015128809438480

Fagerberg B, Ekbohm G (1995) Variation in clover content and in nutritional value of grass-clover leys. Swedish University of Agricultural Science. Crop Production Science 23: 46

Fagerberg B, Torssell B (1995) Estimation of variation in growth potential of leys with different red clover contents, nitrogen applications and cutting regimes Wirtschaftseigene Futter 41:145-162

Finn JA, Kirwan L, Connolly J et al. (2013) Ecosystem function enhanced by combining four functional types of plant species in intensively managed grassland mixtures: a 3-year continental-scale field experiment. J Appl Ecol 50:365-375 doi:10.1111/1365-2664.12041

Frøseth RB, Bakken AK, Bleken MA, Riley H, Pommeresche R, Thorup-Kristensen K, Hansen S (2014) Effects of green manure herbage management and its digestate from biogas production on barley yield, N recovery, soil structure and earthworm populations Eur J Agron 52, Part B: 90-102 doi:10.1016/j.eja.2013.10.006

Fystro G, Lunnan T (2006) Analysar av grovfôrkvalitet på NIRS. In: Kristoffersen AØ (ed) Plantemøtet Østlandet 2006, Bioforsk FOKUS (3): 180-182

Gierus M, Kleen J, Loges R, Taube F (2012) Forage legume species determine the nutritional quality of binary mixtures with perennial ryegrass in the first production year Anim Feed Sci Tech 172:150-161 doi:10.1016/j.anifeedsci.2011.12.026

Grønnerød B The effect of cutting intensity on yield, quality and persistence of timothy. In: Halevy AH (ed) Proceedings of the 12th General Meeting of EGF, Dublin, 4-7 July 1988. CRC Handbook of Flowering. pp 520-521

Halling MA, Topp CFE, Doyle CJ (2004) Aspects of the productivity of forage legumes in Northern Europe. Grass Forage Sci 59, 331-344. doi:10.1111/j.1365-2494.2004.00435.x

Hetta M, Gustavsson AM, Cone JW, Martinsson K (2004) In vitro degradation characteristics of timothy and red clover at different harvest times Acta Agr Scand A-An 54:20-29 doi: 10.1080/09064700410024337

Johansen A, Bakken AK, Hansen S (2008) Balancing milk yield and stocking rate to the output from an organic crop rotation system Grassland Science in Europe 13:592-594

Kirwan L, Lüscher A, Sebastià MT et al. (2007) Evenness drives consistent diversity effects in intensive grassland systems across 28 European sites. J Ecol 95:530-539 doi:10.1111/j.1365-2745.2007.01225.x

Kleen J, Taube F, Gierus M (2011) Agronomic performance and nutritive value of forage legumes in binary mixtures with perennial ryegrass under different defoliation systems. J Agric Sci 149:73-84 doi:10.1017/S0021859610000456

Lemaire G, Gastal F (1997) N uptake and distribution in plant canopies. In: Lemaire G (ed) Diagnosis of the Nitrogen Status in Crops. Springer Berlin Heidelberg, pp 3-43. doi:10.1007/978-3-642-60684-7_1

Lunnan T (1989) Raudkløver, kvitkløver, luserne og kaukasisk strekbelg i blanding med timotei og i reinebestand Norsk landbruksforsking 3:25-39

Lunnan T (2004a) Avling, kvalitet og varigheit i okologisk klovereng Grønn Kunnskap 8:136-143

Lunnan T (2004b) Samanlikning av lang- og kortvarig eng – effektar på avling, fôrkvalitet og plantebestand Grønn Kunnskap 8:192-200

Mallarino AP, Wedin WF (1990) Effect of species and proportion of legume on herbage yield and nitrogen concentration of legume-grass mixtures Grass Forage Sci 45:393-402 doi: 10.1111/j.1365-2494.1990.tb01964.x

Moore KJ, Moser LE, Vogel KP, Waller SS, Johnson BE, Pedersen JF (1991) Describing and quantifying growth stages of perennial forage grasses Agron J 83:1073-1077 doi:10.2134/agronj1991.00021962008300060027x

Øyen J, Aase K (1988) Rødkløver i blanding med gras. Avling og kløverinnhold ved ulik N-gjødsling og høstingspraksis Norsk landbruksforsking 2:41-49

Riesinger P, Herzon I (2008) Variability of herbage production in mixed leys as related to ley age and environmental factors: a farm survey. Agr Food Sci 17:394-412

Rinne M, Nykänen A (2000) Timing of primary growth harvest affects the yield and nutritive value of timothy-red clover mixtures Agr Food Sci Finland 9:121-134

Røthe G, Adler S, Steinshamn H (2007) Hvitkløver og rødkløver i blanding med grasarter ved ulike høsteregimer i økologisk landbruk Bioforsk FOKUS 2 (13): 24-26

SAS (2004) SAS/STATr 9.1 User's Guide. SAS Institute Inc., Cary, NC, USA,

Steinshamn H (1997) Growth potential and herbage quality of low input grass-clover leys. Dissertation, Agricultural University of Norway.

Steinshamn H (2001) Effects of cattle slurry on the growth potential and clover proportion of organically managed grass-clover leys. Acta Agric Scand, B-S P 51:113-124 doi: 10.1080/09064710127615

Steinshamn H, Thuen E (2008) White or red clover-grass silage in organic dairy milk production: Grassland productivity and milk production responses with different levels of concentrate Livest Sci 119:202-215 doi:10.1016/j.livsci.2008.04.004

TINE Rådgivning (2014). Faglig rapport 2013. TINE Rådgivning og Medlem. https://medlem.tine.no/cms/aktuelt/nyheter/statistikk/faglig-rapport. Accessed 26 June 2015

Sturludóttir E, Brophy C, Bélanger G, Gustavsson AM, Jørgensen M, Lunnan T, Helgadóttir Á (2014) Benefits of mixing grasses and legumes for herbage yield and nutritive value in Northern Europe and Canada. Grass Forage Sci 69:229-240 doi:10.1111/gfs.12037

Wachendorf M, Collins, RP, Elgersma A et al. (2001) Overwintering and growing season dynamics of Trifolium repens L. in mixture with Lolium perenne L.: A model approach to plant-environment interactions. Ann Bot 88:683-702 doi:10.1006/anbo.2001.1496

Wilson JR, Kennedy PM (1996) Plant and animal constraints to voluntary feed intake associated with fibre characteristics and particle breakdown and passage in ruminants Aust J Agric Res 47:199-225 doi:10.1071/AR9960199

Figure captions

Fig. 1 Predictions of herbage dry matter yield (kg ha^{-1}). a) Yield in first cut according to stage of phenological development (MSC = mean stage by count) and herbage clover proportion (0.05 and 0.30). b) Yield in second cut according to regrowth period (temperature sum) after early (grey) and late (black) first cut (MSC of 2.4 and 2.8, respectively) and herbage clover proportion (0.15 and 0.50). c) Total annual yield at a clover herbage proportion of 0.25 according to MSC at first cut and temperature sum between first and second cut in two- (black, short = 700 and long = 1000 °C) and three-cut systems (grey, short = 500 and long = 700 d°C). d) Total annual yield according to MSC in the first cut and herbage clover proportion (0.10 and 0.40), where regrowth temperature sum was set to 600 and 850 d°C in three- (grey) and two-cut (black) system, respectively. Open horizontal bars indicate the range of values of MSC (a, c and d) and temperature sum (b) for which differences between curves were significant at 5% level.

Fig. 2 Predictions of annual yield (kg DM ha^{-1}). a) Annual yield according to clover herbage proportion and stage of phenological development at first cut (MSC = 2.3 and 2.5) in a three-cut system, and b) Annual yield according to clover herbage proportion temperature sum between first and second cut (700 and 900 d°C) in a two-cut system. Open horizontal bars indicate the range of values of clover herbage proportion for which differences between curves are significant at 5% level.

Fig. 3 Predictions of herbage in vitro dry matter digestibility (IVDMD, g kg^{-1} DM). IVDM in first cut according to stage of phenological development (MSC = mean stage by count) and herbage clover proportion (0.05 and 0.30). b) IVDMD in second cut according to regrowth period (temperature sum) after early (grey) and late (black) first cut (MSC of 2.4 and 2.8, respectively) and herbage clover proportion (0.15 and 0.50). c) IVDMD in annual yield at a clover herbage proportion of 0.25 according to MSC at first cut and temperature sum between first and second cut in two- (black, short = 700 and long = 1000 d°C) and three-cut systems (grey, short = 500 and long = 700 d°C). d) IVDMD in annual yield according to MSC in the first cut and herbage clover proportion (0.10 and 0.40), where regrowth temperature sum was set to 600 and 850 d°C in three- (grey) and two-cut (black) system, respectively. Open horizontal bars indicate the range of values of MSC (a, c and d) and temperature sum (b) for which differences between curves were significant at 5% level.

Fig. 4 Predictions of herbage crude protein (CP) concentration (g kg^{-1} DM). a) CP in first cut according to stage of phenological development (MSC = mean stage by count) and herbage clover proportion (0.05 and 0.30). b) CP in second cut according to regrowth period (temperature sum) after early (grey) and late (black) first cut (MSC of 2.4 and 2.8, respectively) and herbage clover proportion (0.15 and 0.50). c) CP in annual yield at a clover herbage proportion of 0.25 according to MSC at first cut and temperature sum between first and second cut in two- (black, short = 700 and long = 1000 d°C) and three-cut systems (grey, short = 500 and long = 700 d°C). d) CP in annual yield according to MSC in the first cut and herbage clover proportion (0.10 and 0.40), where regrowth temperature sum was set to 600 and 850 d°C in three- (grey) and two-cut (black) system, respectively. Open horizontal bars indicate the range of values of MSC (a, c and d) and temperature sum (b) for which differences between curves were significant at 5% level.

Fig. 5 Predictions of herbage NDF concentration (g kg^{-1} DM). a) NDF in first cut according to stage of phenological development (MSC = mean stage by count) and herbage clover proportion (0.05 and 0.30). b) NDF in second cut according to regrowth period (temperature sum) after early (grey) and late (black) first cut (MSC of 2.4 and 2.8, respectively) and herbage clover proportion (0.15 and 0.50). c) NDF in annual yield at a clover herbage proportion of 0.25

according to MSC at first cut and temperature sum between first and second cut in two- (black, short = 700 and long = 1000 d°C) and three-cut systems (grey, short = 500 and long = 700 d°C). d) NDF in annual yield according to MSC in the first cut and herbage clover proportion (0.10 and 0.40), where regrowth temperature sum was set to 600 and 850 d°C in three- (grey) and two-cut (black) system, respectively. Open horizontal bars indicate the range of values of MSC (a, c and d) and temperature sum (b) for which differences between curves were significant at 5% level.

Table 1a. Studies, experimental factors, years, production years, locations, and number of observations from each study

Study	Experimental factors	Year	Production years	Location					Number of observations
				Name	Position	Altitude (m above sea level)	Soil type	Fertilization in production years	
1	Cutting system (2 and 3 cuts) and seed mixtures (6 mixtures)	1993-2003	1 - 4	Løken	61.12°N,9.06°E	560	Silty sand	2 cuts: 20 Mg ha^{-1} cattle slurry in spring. 3 cuts: 10 Mg ha^{-1} sheep manure after 1. cut	252
2	Cutting system (early and late 1. cut) and seed mixture (4 mixtures)	2005-2006	1 - 2	Tingvoll Vågønes Holt	62.91°N,8.18°E 67.28°N,14.45°E 69.65°N,18.91°E	23 26 12	Silty sand Silty sand Sandy loam	None	36
3	Cutting system (2 and 3 cuts)	2004-2010	1 - 2	Kvithamar	63.49°N,10.88°E	28	Silty clay loam	9 Mg ha^{-1} cattle slurry in spring	56
4	Cutting system (early, normal and late 1st cut), seed mixtures (3)	2000-2003	1 - 4	Særheim Løken Kvithamar Holt	58.76°N,5.65°E 61.12°N,9.06°E 63.49°N,10.88°E 69.65°N,18.91°E	90 527 28 12	Sand Silty sand Silty clay loam Sandy loam	15 Mg ha^{-1} cattle slurry in spring	144
5	Without and with removal of green manure (using only data from the treatment with removal of plant material)	2009	1	Ås Apelsvoll Kvithamar Værnes	59.66°N,10.78°E 60.70°N,10.86°E 63.49°N,10.88°E 63.49°N,10.88°E	94 255 28 10	Clay loam Sandy loam Loamy clay Sandy loam	None	8

1. Lunnan, T. 2004. Samanlikning av lang- og kortvarig eng – effektar på avling, fôrkvalitet og plantebestand. Grønn Kunnskap 8 (2): 192-200. The three-cut system simulated a forage production system on sheep farms with spring and autumn grazing and one cut for hay production, while the two-cut system simulated a forage production system on dairy farms with two cuts for silage production. The level and type of manure were according to the production system.
2. Røthe, G., Adler, S. & Steinshamn, H. 2007. Hvitkløver og rødkløver i blanding med grasarter ved ulike høsteregimer i økologisk landbruk. Bioforsk FOKUS 2(13): 24-26.
3. Johansen, A., Bakken, A.K. and Hansen, S. 2008. Balancing milk yield and stocking rate to the output from an organic crop rotation system. Proceedings of the 22th General Meeting of the European Grassland Federation, Uppsala, Sweden. Grassland Science in Europe 13: 592-594.
4. Lunnan, T. 2004 Avling og kvalitet og varigheit i økologisk kløvereng. Grønn Kunnskap 8 (2): 136-143.
5. Frøseth R B, Bakken A K, Bleken M A, Riley H, Pommeresche R, Thorup-Kristensen K, Hansen S. 2014. Effects of green manure herbage management and its digestate from biogas production on barley yield, N recovery, soil structure and earthworm populations. European Journal of Agronomy 52: 90-102.

Table 1b. Grass and legume species, cultivars, and range in seeding rates used in the studies (Table 1a)

Study	**Grasses**		**Legumes**		**Total seeding rate, kg ha^{-1}**
	Species (cultivars)	**Seeding rate, kg ha^{-1}**	**Species (cultivars)**	**Seeding rate, kg ha^{-1}**	
1	*Phleum pratense* L. (80%'Grindstad, 20%'Bodin'[a]/'Vega' [b])	3.75 – 15.0	*Trifolium pratense* L. (50% 'Kolpo', 50% 'Bjursele')	0.63 – 5.0	25
	Festcua pratensis Huds. ('Salten')	0 – 6.25	*Trifolium repens* L. (50% 'Undrom'[a], 25% 'Milkanova'[a], 25% 'Sonja'[a]/100% 'Norstar'[b])	0.6 – 1.25	
	Poa Pratensis L. (50% 'Leikra' [a], 50%'Enaldo' [a]/100% 'Entopper' [b])	0 – 5.0	*Galega orientalis* Lam. (non-specific)	0 – 17.5	
	Festuca rubra L. ('Leik')	0 – 2.50	*Medicago sativa* L. ('Peace'[a]/'Live'[b])	0 – 8.75	
	Bromus inermis Leyss. ('Løfar'[a]/'Leif'[b])	0 – 13.75			
2	*Phleum pratense* L. ('Grindstad')	10	*Trifolium pratense* L. ('Kolpo')	0 – 5.0	30
	Festcua pratensis Huds. ('Fure')	10	*Trifolium repens* L. ('Snowy')	0 – 2.5	
	Poa Pratensis L. ('Entopper')	5			
3	*Phleum pratense* L. ('Grindstad')	14.1	*Trifolium pratense* L. ('Bjursele')	1.5	23.1
	Festcua pratensis Huds. ('Fure')	3.9	*Trifolium repens* L. ('Milkanova')	1.2	
	Lolium perenne L. ('Napoleon')	1.2	*Trifolium hybridum* L. ('Alpo')	1.2	
4	*Phleum pratense* L. ('Grindstad', 'Vega', 'Engmo')	17.5	*Trifolium pratense* L. ('Bjursele','Betty','Nordi','Kolpo')	5	25
	Festcua pratensis Huds. ('Fure', 'Salten')	7.5			
5	*Phleum pratense* L. ('Grindstad')	2	*Trifolium pratense* L. ('Nordi')	4	20
	Festcua pratensis Huds. ('Fure')	7			
	Lolium perenne L. ('Napoleon')	7			

Study 1. Cultivars used in the first [a] and second [b] rotation, respectively
Study 4. Location and timothy cultivars: Særheim 100%'Grindstad', Kvithamar and Løken 50%'Grindstad', 50%'Vega', Holt 100%'Engmo'. Location and red clover cultivars: Løken and Holt 50%'Bjursele', 50%'Betty', Særheim and Kvithamar: 50%'Nordi', 50%'Kolpo'. Location and meadow fescue cultivar: Særheim 100%'Fure', Løken, Kvithamar and Holt 100% 'Salten'.

Table 2. Mean, standard deviation, minimum and maximum values of independent (*italics*) and response variables in the three statistical models and the range of these variables in the predictions for graphical display

Variabel	**Mean**	**s.d.**	**Min**	**Max**	**Range in predictions**
Model 1: 1st cut (n=496)					
Mean stage by count (D, also in model 2 and 3)	2.59	0.44	1.73	3.60	2.0 - 3.0
Clover proportion in DM yield (C)	0.19	0.18	0.0	0.87	0.05 - 0.30
Yield (kg DM ha^{-1})	3798	2022	330	8910	1,750 - 6,250
In vitro dry matter digestibility (IVDMD, g kg DM^{-1})	747	59	619	907	700 - 816
Crude protein (CP, g kg DM^{-1})	117	40	42	238	65 - 161
Neutral detergent fiber (NDF, g kg DM^{-1})	496	93	242	676	393 - 589
Model 2: 2nd cut (n=496)					
Temperature sum between 1st and 2nd cut (T1)	758	194	225	1456	500 - 1000
C proportion7	0.29	24	0	98	0.15 - 0.50
Yield (kg DM ha^{-1})	2563	1326	90	6960	1580 - 3780
IVDMD (g kg DM^{-1})	726	48	532	863	700 - 766
CP (g kg DM^{-1})	125	23	76	201	104 - 146
NDF (g kg DM^{-1})	467	68	283	629	448 - 517
Model 3: Annual (n=292 in 2 cuts and n=204 in 3 cuts)					
T1, 2 cuts	843	195	225	1456	700 - 1000
T1, 3 cuts	636	108	449	959	500 - 700
D, 2 cuts	2.84	0.24	2.35	3.25	2.45 - 3.00
D, 3 cuts	2.23	0.41	1.73	3.60	2.00 - 2.50
C, proportion, 2 cuts	0.23	0.20	0.0	0.77	0.10 - 0.40
C, proportion, 3 cuts	0.27	0.20	0.0	0.89	0.10 - 0.40
Yield (kg DM ha^{-1}), 2 cuts	7024	2283	1800	13870	6200 - 9500
Yield (kg DM ha^{-1}), 3 cuts	6835	2215	2810	12473	5675 - 9100
IVDMD (g kg DM^{-1}), 2 cuts	716	34	605	795	698 - 761
IVDMD (g kg DM^{-1}), 3 cuts	736	27	645	806	720 - 755
CP (g kg DM^{-1}), 2 cuts	105	21	48	173	80 - 134
CP (g kg DM^{-1}), 3 cuts	129	17	70	175	99 - 136
NDF (g kg DM^{-1}), 2 cuts	516	57	360	659	472 - 568
NDF (g kg DM^{-1}), 3 cuts	475	58	324	647	476 - 544

Table 3. Parameter estimates for the models of dry matter yield (kg DM ha^{-1})

	Model 1: 1st cut				Model 2: 2nd cut				Model 3: Annual[3]			
Effect	**Estimate**	**SE[2]**	***t*[1]**	**P > |*t*|**	**Estimate**	**SE[2]**	***t*[1]**	**P > |*t*|**	**Estimate**	**SE[2]**	***t*[1]**	**P > |*t*|**
Intercept	-8344	1276	-	-	10321	739	-	-	5773	1329.8	-	-
Number of cuts (S)[4]	-	-	-	-	-	-	-	-	729.8	421.4	-	-
Production year (E)[5]	118.8	193.4	0.61	0.539	-312	120	-2.6	0.009	-375.6	265.6	-1.41	0.158
Developmental stage (D)[6]	5892	981.0	-	-	-2870.3	231.2	-	-	160.4	373.3	-	-
D×D	-496.0	190.5	-2.60	<0.001	-	-	-	-	-	-	-	-
Temperature sum (T)[7]	-	-	-	-	-9.37	1.39	-	-	-1.32	0.88	-	-
T×T	-	-	-	-	0.0036	0.00058	6.10	<0.001	-	-	-	-
T×D	-	-	-	-	1.93	0.34	5.73	<0.001	-	-	-	-
Clover proportion (C)[8]	-366	1877.1	-	-	1750.6	562.8	-	-	-7606.8	2546.5	-	-
C×C	-5211.1	1450.6	-3.59	<0.001	-2697.4	540.2	-4.99	<0.001	-7057.7	1493.8	-4.72	<0.001
S×C	-	-	-	-	-	-	-	-	-4275.3	724.5	-5.90	<0.001
T×C	-	-	-	-	3.93	0.49	8.03	<0.001	9.28	1.33	6.99	<0.001
D×C	2043.5	623.0	3.28	0.001	-	-	-	-	5123.4	771.9	6.64	<0.001
RMSE[9]	626				406				889			
Coefficient of variation, %	16.5				15.8				12.8			

[1] t-values and probabilities (P-values) of main effects were omitted when the effects was included in a significant interaction

[2]SE standard error

[3]Annual is the annual accumulated yield of all cuts

[4]The estimate is 2 cuts relative to 3 cuts. The estimated value for 3 cuts is 0, used by SAS as the reference value as it is the second of two levels.

[5]Production year was included even if the term was not significant in order to predict values and run tests of significance of independent variables. The estimated value is for the second production year relative to the fourth production year, used by SAS as the reference value as it is the fourth of four levels.

[6]Developmental stage (D) is the phenological stage of development expressed as mean stage by count of timothy in the first cut

[7]Temperature sum (T) is the cumulative sum of mean daily temperature between the 1st and 2nd cut

[8]Clover proportion of DM yield (C)

[9]RMSE = Root mean squared error

Table 4. Parameter estimates for the models of herbage in vitro dry matter digestibility (g kg DM^{-1})

	Model 1: 1st cut				Model 2: 2nd cut				Model 3: Annual[3]			
Effect	**Estimate**	**SE**	***t*** [1]	**P > |*t*|**	**Estimate**	**SE** [2]	***t***	**P > |*t*|**	**Estimate**	**SE**	***t***	**P > |*t*|**
Intercept	887.8	38.3	-	-	708.7	46.8	-	-	564.5	55.2	-	-
Number of cuts (S)[4]	-	-	-	-	-	-	-	-	-43.1	20.9	-	-
Production year (E)[5]	8.6	6.0	1.45	0.151	9.7	6.9	1.40	0.163	3.9	4.2	0.93	0.354
Developmental Stage (D)[6]	8.66	29.75	-	-	-2.53	14.85	-	-	240.2	33.3	-	-
D×D	-24.37	5.85	-4.17	<0.001	-	-	-	-	-53.12	5.56	-9.56	<0.001
Temperature sum (T)[7]	-	-	-	-	-0.063	0.087	-	-	-0.010	0.046	-	-
S×T	-	-	-	-	-	-	-	-	0.108	0.029	3.70	<0.001
T×D	-	-	-	-	0.090	0.022	4.15	<0.001	-0.054	0.018	-3.09	<0.001
Clover proportion (C)[8]	-9.70	11.08	-0.87	0.400	198.9	49.88	-	-	-250.5	40.19	-6.23	-
T×C	-	-	-	-	-0.136	0.024	-5.58	<0.001	-	-	-	-
D×C	-	-	-	-	-70.95	16.75	-4.24	<0.001	77.2	13.27	5.82	<0.001
RMSE[9]	24.8				21.9				14.2			
Coefficient of variation, %	3.3				3.0				2.0			

[1] t-values and probabilities (P-values) of main effects were omitted when the effects was included in a significant interaction

[2]SE standard error

[3]Annual is the weighted average of the annual yield

[4]The estimate is 2 cuts relative to 3 cuts. The estimated value for 3 cuts is 0, used by SAS as the reference value as it is the second of two levels.

[5]Production year was included even if the term was not significant in order to predict values and run tests of significance of independent variables. The estimated value is for the second production year relative to the fourth production year, used by SAS as the reference value as it is the fourth of four levels.

[6]Developmental stage (D) is the phenological stage of development expressed as mean stage by count of timothy in the first cut

[7]Temperature sum (T) is the cumulative sum of mean daily temperature between the 1st and 2nd cut

[8]Clover proportion of DM yield (C)

[9]RMSE = Root mean squared error

Table 5. Parameter estimates for the models of herbage crude protein concentration (CP, g kg DM^{-1})

	Model 1: 1st cut				Model 2: 2nd cut				Model 3: Annual[3]			
Effect	**Estimate**	**SE**	***t*** [1]	**P > |*t*|**	**Estimate**	**SE** [2]	***t***	**P > |*t*|**	**Estimate**	**SE**	***t***	**P > |*t*|**
Intercept	519.6	24.8	-	-	-6.2	18.8	-	-	250.2	61.2	-	-
Number of cuts (S)[4]	-	-	-	-	-	-	-	-	100.0	13.3	-	-
Production year (E)[5]	-14.9	4.4	-3.4	<0.001	-12.2	2.8	-4.29	<0.001	-18.2	2.3	-7.79	<0.001
Developmental stage (D)[6]	-257.1	19.0	-	-	48.4	6.0	-	-	-46.12	6.06	-	-
D×D	36.4	3.74	9.73	<0.001	-	-	-	-	-	-	-	-
S×D									-32.36	4.77	-6.78	<0.001
Temperature sum (T)[7]	-	-	-	-	0.165	0.035	-	-	-0.111	0.022	-	-
T×T	-	-	-	-	-0.00005	0.000014	-3.70	<0.001	-	-	-	-
T×D	-	-	-	-	-0.049	0.0088	-5.59	<0.001	0.0256	0.0076	3.36	<0.001
Clover proportion (C)[7]	84.53	5.25	16.08	<0.001	-52.9	21.4	-	-	71.27	24.24	-	-
C×C	-	-	-	-	-	-	-	-	-39.12	14.12	-2.77	0.006
T×C	-	-	-	-	-	-	-	-	-0.041	0.011	-3.64	<0.001
D×C	-	-	-	-	43.80	7.34	5.97	<0.001	24.20	6.57	3.68	<0.001
RMSE[9]	15.7				9.9				8.6			
Coefficient of variation, %	13.4				7.9				7.5			

[1] t-values and probabilities (P-values) of main effects were omitted when the effects was included in a significant interaction
[2]SE standard error
[3]Annual is the weighted average of the annual yield
[4]The estimate is 2 cuts relative to 3 cuts. The estimated value for 3 cuts is 0, used by SAS as the reference value as it is the second of two levels.
[5]Production year was included even if the term was not significant in order to predict values and run tests of significance of independent variables. The estimated value is for the second production year relative to the fourth production year, used by SAS as the reference value as it is the fourth of four levels.
[6]Developmental stage (D) is the phenological stage of development expressed as mean stage by count of timothy in the first cut
[7]Temperature sum (T) is the cumulative sum of mean daily temperature between the 1st and 2nd cut
[8]Clover proportion of DM yield (C)
[9]RMSE = Root mean squared error

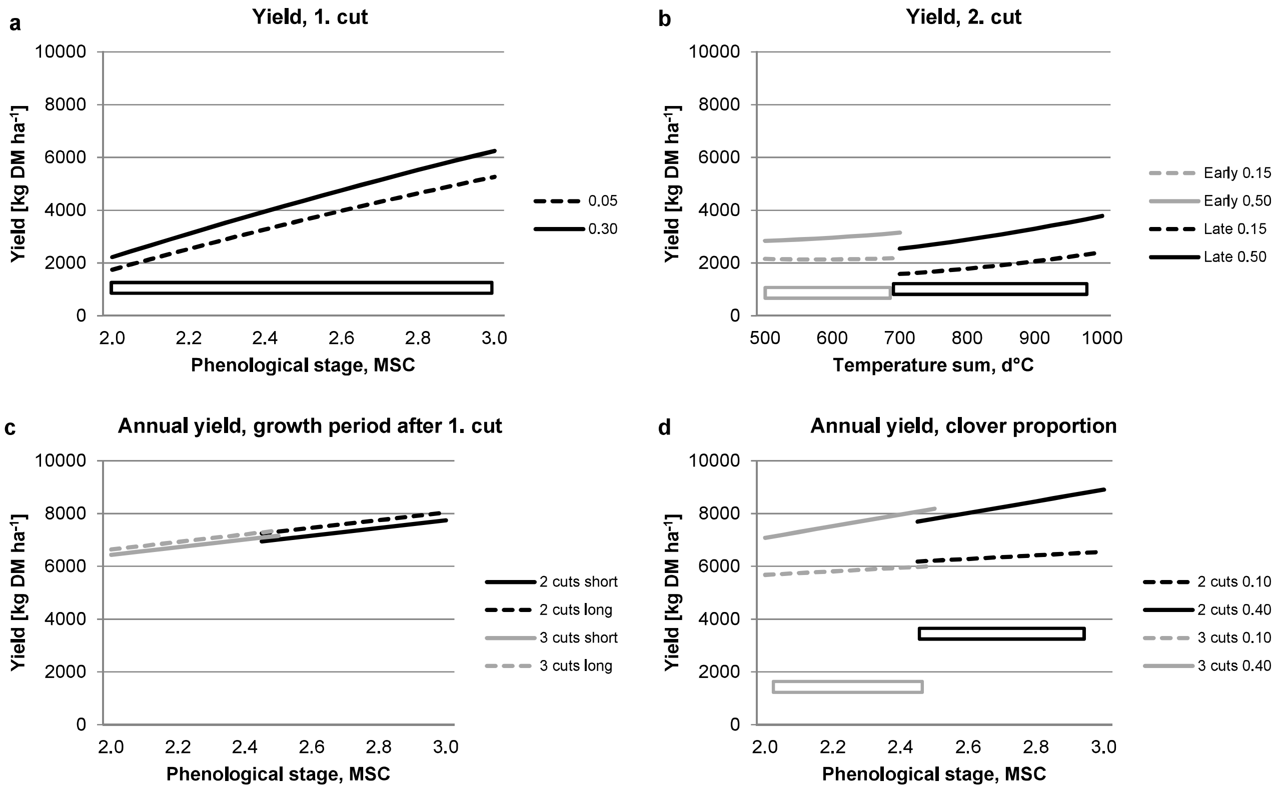
Table 6. Parameter estimates for the models of herbage NDF (g kg DM^{-1})

	Model 1: 1st cut				Model 2: 2nd cut				Model 3: Annual[3]			
Effect	**Estimate**	**SE**	***t*** [1]	**P > |*t*|**	**Estimate**	**SE**[2]	***t***	**P > |*t*|**	**Estimate**	**SE**	***t***	**P > |*t*|**
Intercept	-197.4	62.4	-	-	793.5	49.9	-	-	578.3	51.7	-	-
Number of cuts (S)[4]	-	-	-	-	-	-	-	-	-164.2	30.1	-	-
Production year (E)[5]	16.4	9.8	1.68	0.044	-5.16	8.3	-0.62	0.534	18.2	6.2	2.92	0.004
Developmental stage (D)[6]	415.9	48.7	-	-	-55.1	9.57	9.57	<0.001	-144.3	37.9	-	-
D×D	-52.1	9.57	-5.45	<0.001	-	-	-	-	42.4	7.48	5.66	<0.001
S×D	-	-	-	-	-	-	-	-	39.0	10.5	3.72	<0.001
Temperature sum (T)[7]	-	-	-	-	-0.374	0.083	-	-	0.105	0.017	6.10	<0.001
T×T	-	-	-	-	0.000248	0.000051	4.86	<0.001	-	-	-	-
Clover proportion (C)[8]	-164.5	18.5	-8.89	<0.001	-91.38	20.4	-4.47	0.001	-20.9	46.9	-	-
C×S	-	-	-	-	-	-	-	-	89.4	20.2	4.43	<0.001
D×C	-	-	-	-	-	-	-	-	-69.8	18.1	-3.86	<0.001
RMSE[9]	40.5				25.2				25.4			
Coefficient of variation, %	8.2				5.4				5.1			

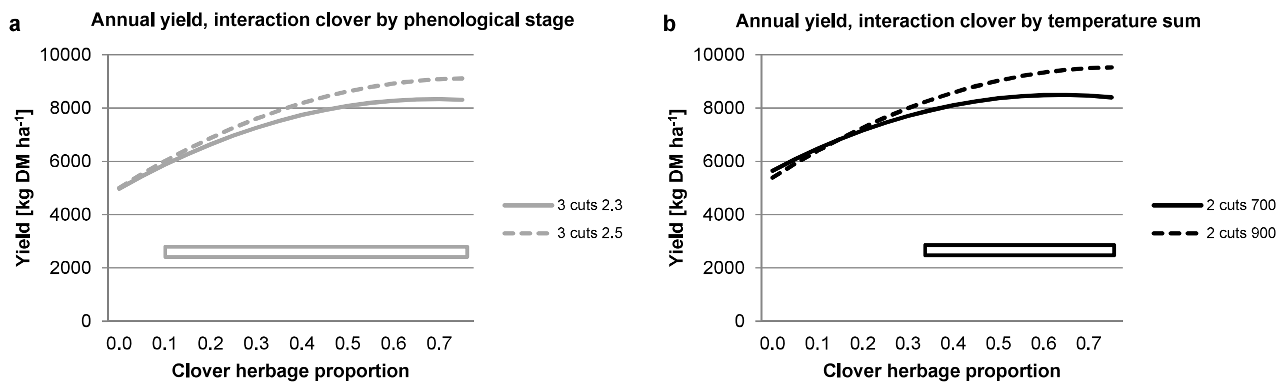
[1] t-values and probabilities (P-values) of main effects were omitted when the effects was included in a significant interaction
[2]SE standard error
[3]Annual is the weighted average of the annual yield
[4]The estimate is 2 cuts relative to 3 cuts. The estimated value for 3 cuts is 0, used by SAS as the reference value as it is the second of two levels.
[5]Production year was included even if the term was not significant in order to predict values and run tests of significance of independent variables. The estimated value is for the second production year relative to the fourth production year, used by SAS as the reference value as it is the fourth of four levels.
[6]Developmental stage (D) is the phenological stage of development expressed as mean stage by count of timothy in the first cut
[7]Temperature sum (T) is the cumulative sum of mean daily temperature between the 1st and 2nd cut
[8]Clover proportion of DM yield (C)
[9]RMSE = Root mean squared error

line figure

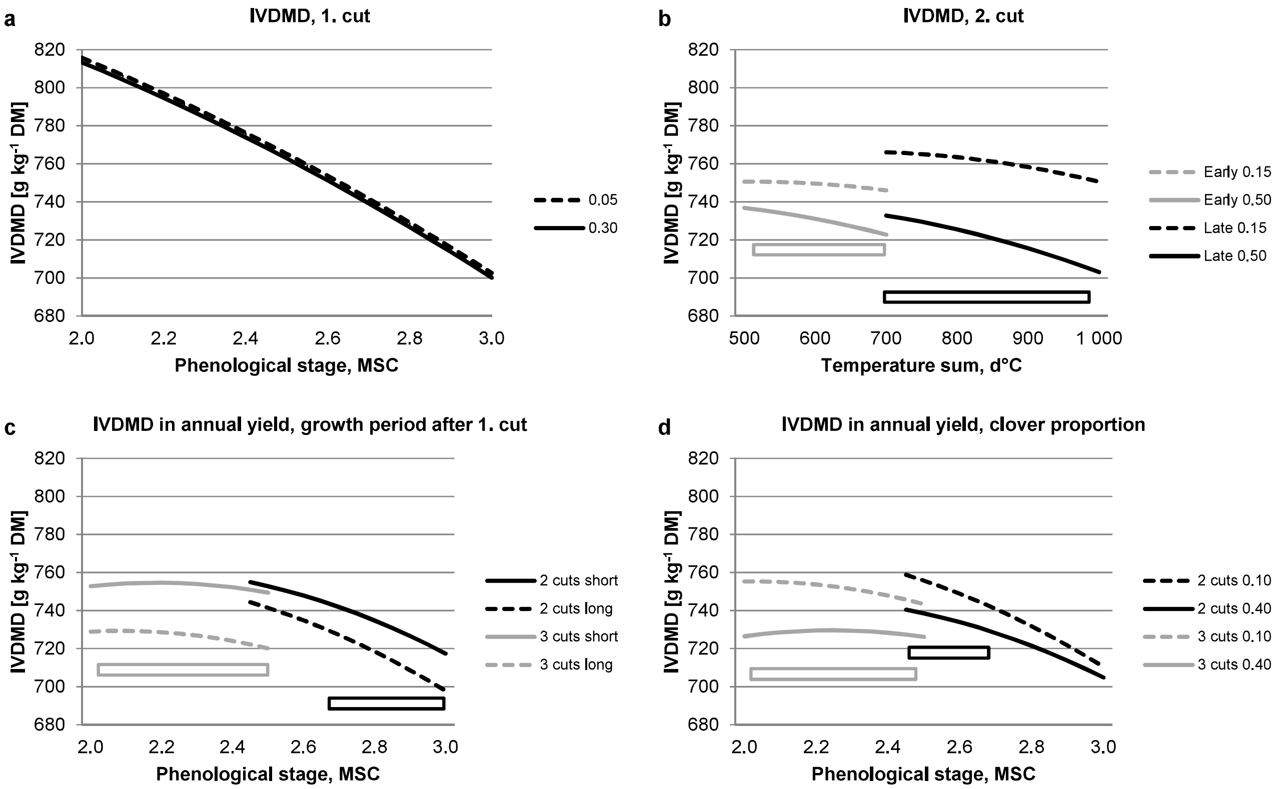
Click here to download line figure Fig 1 rev.pdf



line figure Click here to download line figure Fig 2 rev.pdf

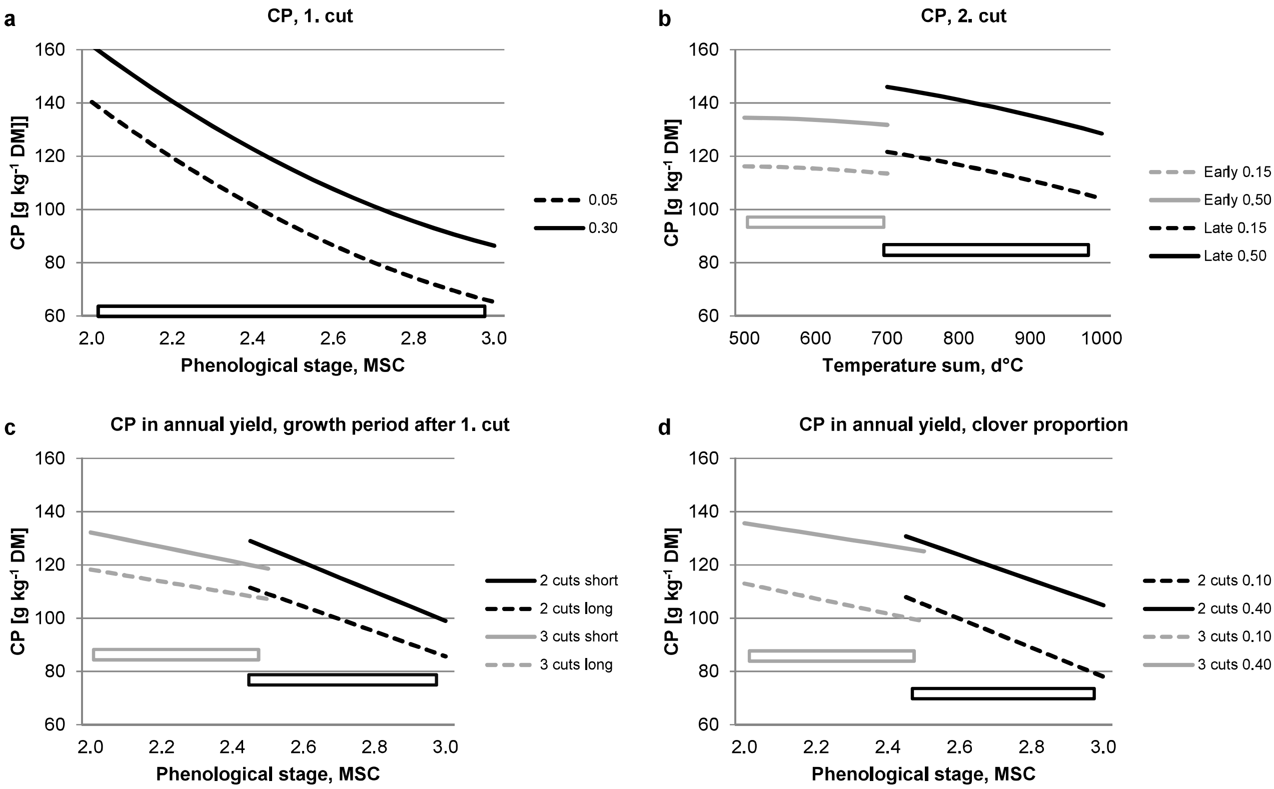


line figure Click here to download line figure Fig 3 rev.pdf



line figure

Click here to download line figure Fig 4 rev.pdf



line figure Click here to download line figure Fig 5 rev.pdf