






Effect of strategy for harvesting regrowth grass silage on performance in dairy cows

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ABSTRACT

The objective of this study was to evaluate the effects of feeding lactating dairy cows with regrowth silages from different 2- and 3-cut harvesting systems on milk production, efficiency of N, and energy utilization. Thirty Nordic Red cows were offered 5 experimental diets containing regrowth silages, crimped barley, and canola meal in replicated incomplete 5×4 Latin squares with four 21-d periods consisting of 14 d of feed adaptation and 7 d of sampling. Four second-cut silage diets were examined in a 2×2 factorial arrangement, enabling evaluation of effect of harvest time of the early or late first cut on second-cut silages, short or long regrowth interval within second cut, and their interaction on dairy cow performance. The third-cut silage diet harvested from early first cut and short regrowth interval of second-cut ley was compared with the second-cut silage diets to evaluate the difference in dairy cow performance between second- and third-cut silages. Postponing the first cut and extending the regrowth interval decreased dry matter intake (DMI), energy-corrected milk (ECM) yield, nutrient digestibility, and urinary energy output, but improved N efficiency (milk N/N intake). Postponing the first cut also decreased the efficiency of metabolizable energy use for lactation, but increased CH_4 yield (CH_4/DMI). Extending the regrowth interval decreased feed efficiency (ECM/DMI) and increased CH_4 intensity (CH_4/ECM). Thus, feeding regrowth silages in 2- or 3-cut systems harvested after an early first cut and short regrowth interval promoted better dairy performance and feed intake, and higher efficiency of feed and energy utilization, but with poorer N efficiency. Feeding third-cut silage improve milk yield and feed efficiency compared with second-cut silages.

Key words: dairy cow, feed efficiency, grass silage, harvesting strategy

INTRODUCTION

In northern Europe, grass silage is the main ingredient and forage source in dairy cow diets (Huhtanen et al., 2013). It is well known that early harvest of spring growth grass generally produces highly digestible silage that promotes high feed intake and milk yield (Ferris et al., 2001; Kuoppala et al., 2008; Randby et al., 2012). Furthermore, milk production is generally lower in cows fed silage harvested from regrowth compared with primary growth of grass-dominated leys (Peoples and Gordon, 1989; Khalili et al., 2005; Kuoppala et al., 2008) due to the lower digestibility of regrowth silages (Huhtanen et al., 2006), which limits intake and subsequently reduces milk production when fed to dairy cows. In addition, intake potential of silages made from regrowth compared with primary growth has been lower, even when differences in digestibility, DM concentration, and fermentation quality have been taken into account (Huhtanen et al., 2007).

Differences in digestibility of different harvests can be related to changes in temperature and day length throughout the growing period. Generally, both temperature and day length are increasing rapidly in the spring; after midsummer, temperature is still increasing, but light is starting to decline; and in late summer, both temperature and light are declining in northern Europe. Many earlier experiments have only compared silage from each cut (primary vs. regrowth), which precludes separating the effects of cut and grass maturity at harvest. Kuoppala et al. (2008) extended knowledge on silages made from regrowth grass by comparing effects of growth stage within both first- and second-cut grass silages on milk production and nutrient utilization. They reported that postponing the harvest in primary growth decreased digestible OM in silage DM (**D-value**) and subsequently decreased feed intake and milk yield of dairy cows, and this response was clearly smaller when regrowth silages were fed. Typically, in northern Europe, climatic conditions during midsummer limit the ranges of D-value of regrowth compared with the spring or primary growth but likely also makes it less sensitive to timing of harvests.

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The global climate is warming, which has increased the length of the growing season. The growing season for grass (average temperature above 5°C) in Sweden is now more than 10 d longer than it was 50 years ago (Gustavsson, 2017). Management of grasslands has gradually adapted. For example, to use the longer growing season, it is now advised to harvest leys 3 times a year for silage production (Krizsan et al., 2017). Not many dairy cow production trials feeding third-cut regrowth grass silages have been conducted, or they have given variable results regarding intake and milk production potential of the third-cut silage (Huhtanen et al., 2001; Sairanen and Juutinen, 2013; Sairanen et al., 2016). To optimize on-farm harvesting and feeding, more knowledge is needed on the production response, N utilization, and enteric CH₄ production of dairy cows to regrowth silages when different harvesting strategies are applied. We hypothesized that the effect of timing of first cut primarily is of relevance for feed value of primary growth, and that quality of silage made from second cut will decrease with increased duration of regrowth. Further, climatic conditions in northern Europe suggest that silage made of third cut is likely to have a higher feed value than silage made from second cut.

In northern Sweden, leys are typically dominated by timothy grass (*Phleum pratense*) and red clover (*Trifolium pratense*). The objective of this study was to investigate the effects of feeding lactating dairy cows with second- and third-cut silages from different 2- and 3-cut harvesting strategies for grass-dominated leys on dairy cow performance. The effects of harvest time of the early or late first cut on second-cut silages, short or long regrowth interval within second cut, and their interaction were investigated. In addition, their effects on N and energy utilization efficiency, and on CH₄ and carbon dioxide CO₂ emissions were determined.

MATERIALS AND METHODS

Grass silages were harvested in summer 2014, and the feeding experiment was conducted in spring 2015 at Röbbäcksdalens Research Farm, Swedish University of Agricultural Sciences in Umeå (63°45'N; 20°17'E). The study was carried out with the permission of the Swedish Ethics Committee on Animal Research in Umeå, in accordance with Swedish laws and regulations regarding the EU Directive 2010/63/EU on animal research.

Silage Harvest

The regrowth grass for ensiling was harvested from second- and third-year timothy leys containing some red clover (seed ratio 80:20; botanical analysis not

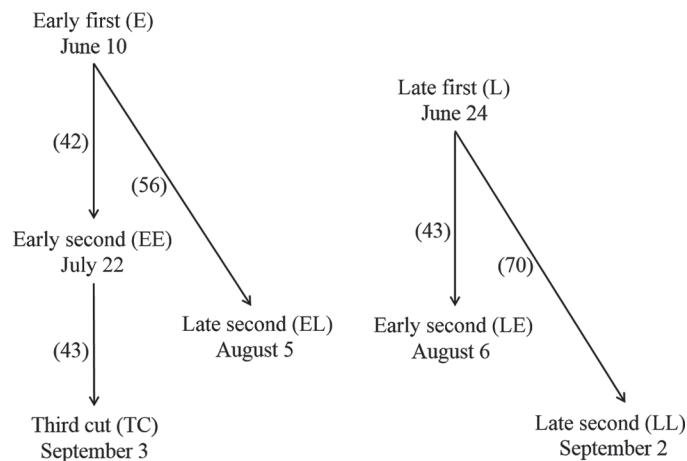


Figure 1. Schematic presentation of the strategies for harvesting regrowth materials for ensiling used in this study. Four second-cut herbage were harvested at early (EE, LE) or late (EL, LL) stage of growth after an early (E) or late (L) first cut; the third-cut (TC) herbage was harvested after the early first and second cut. Growing days from previous cut of each regrowth material are shown in parentheses.

made). The harvesting strategies used are shown in Figure 1. In summary, the leys for early (E) first cut were harvested on June 10, and the leys for late (L) first cut was harvested on June 24. Two second-cut silages were harvested on July 22 at an early (EE) stage of growth and on August 5 at a late (EL) stage from the regrowth area of E. Two further second-cut silages were harvested on August 6 at an early (LE) stage of growth and on September 2 at a late (LL) stage from the regrowth area of L. Furthermore, a third-cut (TC) silage was harvested on September 3 from the regrowth area of EE. The leys were fertilized with a commercial fertilizer (NS 27-4 Axan, Yara AB, Malmö, Sweden) providing 75, 45, and 35 kg of N/ha for the first, second, and third cut, respectively.

The grass was cut with a mower conditioner (GMT 3605 FlexP; JF-Stoll A/S, Sønderborg, Denmark), wilted overnight, and harvested with a precision chop forage wagon (ES 5000 MetaQ ProTec; JF-Stoll A/S). A formic acid-based additive (ProMyr XR 630; Perstorp Holding AB, Perstorp, Sweden) was applied at a rate of approximately 3.5 L/Mg, and the forage material was ensiled in bunker silos until used in the experiment.

Cows, Experimental Design, and Diets

Thirty lactating Nordic Red cows (21 multiparous and 9 primiparous) on average 72 (± 4.4) DIM, weighing 612 (± 14.4) kg, and yielding 31.5 (± 0.93) kg of milk/d (mean \pm SE) were used in the experiment. The feeding experiment started April 2 the year after the silage harvest. The cows were kept in an insulated

loose-house, offered a TMR ad libitum, and given free access to drinking water. The cows in the experiment were milked twice a day, at 0600 and 1500 h.

The experiment was conducted as a replicated incomplete 5×4 Latin square design with 5 treatments and 4 periods. The 4 second-cut silage treatments were fed in a 2×2 factorial arrangement to investigate the effects of harvesting time of the first cut (EE and EL vs. LE and LL), length of the regrowth interval (EE and LE vs. EL and LL), and their interaction on dairy cow performance. The fifth dietary treatment was the TC silage. Each experimental period lasted for 21 d, and recordings and samplings were conducted during the last 7 d. The cows were assigned to 6 blocks according to parity and milk yield and within block were randomly allocated to 1 of the 5 treatments.

The herbage of EE silage displayed characteristics promoting an expected highest intake of all regrowth silages in this study. The EE silage diet was formulated to meet ME and MP requirement of 35 kg of ECM yield. The other experimental diets were formulated to have the same forage to concentrate ratio based on the formulation of the EE silage diet and contained (g/kg of DM): silage (565), crimped barley (340), heat-treated rapeseed meal (ExPro-00SF; AarhusKarlshamn Ltd., Malmö, Sweden; 80) and mineral and vitamin feed (Mixa Optimal; Lantmännen Lantbruk AB, Malmö, Sweden; 15). None of the experimental diets were limiting with regard to ruminal microbial growth (the protein balance in the rumen was predicted during ration formulation). The crimped barley was rolled using a mill (Murska 1400 S2 \times 2; Murska, Ylivieska, Finland) adjusted to 0.3 mm between the rollers, treated with 4 L/Mg of propionic acid (Perstorp Holding AB, Perstorp, Sweden), and stored in air-tight bags (5.2 \times 6.0 m; Ltd. Rani Plast Oy, Terijärvi, Finland). The diets were mixed using a TMR mixer (Nolan A/S, Viborg, Denmark) and then delivered in the feed troughs 4 times (at 0300, 0900, 1500, and 2100 h) per day by an automatic feeding wagon (Roughage Intake Control feeders; Insentec B.V., Marknesse, the Netherlands).

Recordings and Sampling

The cows were offered the experimental diets in Roughage Intake Control feeders (Insentec B. V., Marknesse, the Netherlands), and their intake was recorded individually at each visit. Milk yield was recorded daily, but only the data from a 7-d series of recordings were reported, along with feed intake. The cows were weighed after morning milking for 3 d before the start of the experiment and on the last 3 d of each period. Body condition score of the cows using a 0 to 5 scale with precision increments of 0.25 (Edmonson et al., 1989)

was determined before the experiment started and on the last day of each period, by the same 2 experienced persons throughout the whole trial.

Mass fluxes of CH₄ and CO₂ were measured daily by the GreenFeed emissions monitoring (**GEM**) system (GreenFeed; C-Lock Inc., Rapid City, SD), as described by Huhtanen et al. (2015b). Gas calibrations (pure N₂ and N₂ mixed with 1,000 ppm of CH₄ and 10,000 ppm of CO₂) were performed once a week, and CO₂ recovery tests were conducted biweekly during the whole experiment. Average CO₂ recovery (mean \pm SE) was 100 \pm 0.6%. The air filters were cleaned twice a week to maintain the airflow above 26 L/s. Gas emission data during the last 7 d of each period were collected for statistical analysis. A commercial concentrate (Solid 220; Lantmännen Lantbruk, Malmö, Sweden) was given to the cows in the GEM system throughout the experiment, to encourage them to visit the units. The program was set to allow the cows to visit at a minimum of 5-h intervals and to give 8 drops of 50 g of concentrate during each visit. The interval between drops was set to 40 s.

The DM concentration was determined twice a week for the silages and once a week for the concentrate feeds. The diet composition was adjusted twice weekly to account for the changes in DM content (oven-dried at 60°C for 48 h). Dried feed samples were milled by a cutter mill (SM 2000; Retsch Ltd., Haan, Germany) to pass through a 2- or 1-mm sieve, depending on analytical purpose. In addition, silages were sampled 3 times during the recording week to provide a composite sample of fresh material for analysis of fermentation quality. The frozen silage samples were milled to pass through a 20-mm sieve by the same cutter mill as before and kept stored frozen at -20°C until analyzed.

Milk samples were collected in the last 4 consecutive milkings of each period. Feces (approximately 250 g) and urine (70 mL) spot samples from 15 multiparous cows in 3 blocks were collected at 0530 and 1530 h during 3 d in the last week of each period. After the last collection of each period, the fecal samples were oven-dried at 60°C for 48 h and milled to pass through a 1-mm sieve in a cutter mill. Fecal samples used for analysis of indigestible neutral detergent fiber (**iNDF**) were milled using mortar and pestle to pass through a 2.5-mm sieve. Urine samples were frozen at -20°C directly after collection. Fecal and urine samples were pooled by cow and within period.

Chemical Analysis

The ash, CP, and NDF concentrations of feed and fecal samples and fermentation quality of silages were analyzed as described by Gidlund et al. (2015). Urinary N was analyzed according to official method AOAC-

984.13 (AOAC, 1990) using a Foss Kjeltac 2400 Analyzer Unit (Foss Tecator AB, Höganäs, Sweden) and Cu as a digestion catalyst. Concentration of iNDF was determined by a 12-d in situ ruminal incubation in dairy cows fed a forage-based diet according to the procedure of Krizsan et al. (2015). Analysis of gross energy (GE) in feed, fecal, and urine samples was conducted according to Gordon et al. (1995) using a Parr 6400 Oxygen Bomb Calorimeter (Parr Instrument Co., Moline, IL) with benzoic acid as a standard. The milk samples were analyzed for concentrations of fat, protein, lactose, and milk urea using a near infrared reflectance analyzer (CombiFoss 6000, Foss Electric, Hillerød, Denmark).

Calculations

Potentially digestible NDF (**pdNDF**) was calculated as NDF – iNDF. Silage ME concentration was calculated assuming 16 MJ of ME/kg of digestible OM (**DOM**) according to MAFF (1975). Silage DOM was calculated from OM concentration and OM digestibility (**OMD**), which was estimated from the concentrations of iNDF and NDF (Huhtanen et al., 2013). Concentrations of ME and MP and ruminal protein balance value (**PBV**) in concentrates were calculated from analyzed composition and tabulated digestibility and degradability coefficients in Finnish feed tables (LUKE, 2017). Silage DMI (**SDMI**) index was calculated according to Huhtanen et al. (2007). The ECM yield and milk energy concentration were calculated according to Sjaunja et al. (1990). Feed efficiency was calculated as ECM yield divided by DMI. Milk N efficiency (**MNE**) was calculated as milk N output divided by feed N intake. Total-tract digestibility was calculated using iNDF as internal marker (Huhtanen et al., 1994).

The total fecal N output was calculated by using estimated total fecal DM output and determined fecal N concentration. The total fecal DM output was calculated as total iNDF intake from diet divided by iNDF concentration in feces. Daily urinary excretion was calculated as estimated urinary N excretion (**UN**) divided by urinary N concentration. The UN was estimated using the equation presented by Huhtanen et al. (2015a) including subtraction of scurf N:

$$\text{UN (g/d)} = \text{N intake (g/d)} - \text{fecal N (g/d)} \\ - \text{milk N (g/d)} - \text{scurf N (g/d)} - \text{retained N (g/d)},$$

where scurf N was estimated according to NRC (2001). Nitrogen retention was estimated from the calculated ME balance by assuming that BW gain corresponds to ME balance of 34 MJ/kg and BW loss to –28 MJ/kg

(LUKE, 2017), and BW change contains 25.2 g of N/kg (LUKE, 2017).

Energy losses in feces and urine were calculated based on their GE concentrations and daily outputs. Digestible energy (**DE**) was calculated by subtracting fecal energy from GE intake, and ME by subtracting energy loss through CH₄ and urine from DE intake. Heat production (**HP**) was calculated according to Brouwer (1965) using GEM data and urinary N output. Oxygen consumption for HP calculation was estimated using the relationship between CO₂ and O₂ derived from data in Aubry and Yan (2015; T. Yan, personal communication). Energy balance was calculated by subtracting milk energy and HP from ME intake. The efficiency of ME used for lactation (**k_l**) was calculated according to the equation given by AFRC (1990):

$$k_l = (E_l + aE_g)/(MEI - ME_m),$$

where E_l is milk energy output (MJ/d), E_g is tissue energy balance (MJ/d), MEI is ME intake (MJ/d), and ME_m is ME requirement for maintenance (0.515 × BW^{0.75} MJ/d); a = 0.84 if E_g < 0, or a = 0.95 if E_g > 0.

Statistical Analysis

Experimental data were subjected to ANOVA using the generalized linear model of SAS (SAS Inc. 2002–2003, Release 9.2; SAS Institute Inc., Cary, NC) and the following model:

$$Y_{ijkl} = \mu + B_i + P_j + C_k(B)_i + D_l + \varepsilon_{ijkl},$$

where Y_{ijkl} is the dependent variable and μ is the mean for all observations, B_i is the effect of block i, P_j is the effect of period j, C_k(B)_i is the effect of a cow k within block i, D_l is the effect of diet l, and ε_{ijkl} is the normally distributed random residual error with expected mean of zero and constant variance. Least squares means are reported and mean separation was done by orthogonal contrasts. The 4 second-cut treatments were analyzed as a 2 × 2 factorial arrangement to evaluate the effects of harvesting time of the first cut (EE and EL vs. LE and LL), length of the regrowth interval (EE and LE vs. EL and LL), and their interaction on dairy cow performance. The third-cut treatment was compared with the 4 second-cut diets to evaluate the effect of second and third cuts on dairy cow performance.

RESULTS

All the silages were well preserved, with low pH (mean 3.7), high lactic acid concentration (mean 98 g/

kg DM), and low levels of ammonia-N (mean 44 g/kg N) and butyric acid (mean < 0.3 g/kg DM; Table 1). Second-cut silages harvested after the early first cut (EE and EL) had higher concentrations of CP, GE, ME, and MP and SDMI index but lower concentrations of iNDF compared with the second-cut silages harvested after the late first cut (LE and LL). With extended regrowth interval, the trends in silage chemical composition, nutritional values, and SDMI index were similar to those with a delayed first cut. Concentrations of DM and NDF were lower in the third-cut silage, but concentrations of CP, GE, and MP were higher than in the second-cut silages. Further, EE and the third-cut displayed equivalent values of iNDF, DOM, and ME.

The composition and nutritive values of experimental diets given in Table 2 are based on observed daily intake (TMR + concentrate from GEM unit). The differences in dietary chemical composition and nutritional values reflected the differences between the silages, because silage source was the only difference between the diets.

Intake, Milk Production, and Digestibility

With extended regrowth interval, total and silage DMI decreased by 1.2 and 0.7 kg/d after early first cut, but increased by 0.3 and 0.2 kg/d after late first cut (interaction $P < 0.01$; Table 3). Postponing the first cut and increasing harvest interval decreased intake of CP, ME, MP, and PBV, and the difference was greater (interaction $P < 0.01$) after early compared with late first cut. Postponing the first cut (EE and EL vs. LE and LL) decreased ($P < 0.01$) intake of NDF. Intake of iNDF was higher ($P < 0.01$) for the regrowth silage diets after the late first cut, but the difference between regrowth interval was greater (interaction $P < 0.01$) after early compared with late first cut. Feeding the TC silage diet decreased ($P < 0.01$) intake of NDF and iNDF, but increased ($P < 0.01$) intake of CP, MP, and PBV compared with cows fed the second-cut silages. Intake of concentrate from the GEM units was on average 1.45 kg/d (results not shown), and the differences between diets were not significant ($P \geq 0.35$).

Postponing the first cut and increasing the regrowth interval (EE and LE vs. EL and LL) decreased ($P \leq 0.02$) the yield of milk, fat, and lactose and decreased concentration of milk urea. The decrease in ECM yield with increased regrowth interval was greater after early first cut compared with late first cut (interaction $P = 0.02$). Cows fed the TC silage gave higher ($P < 0.01$) yields of milk and ECM than cows fed the second-cut silages. The milk protein concentration decreased by 0.8 g/kg and increased by 0.5 g/kg with increased regrowth interval after early and late first cut, respectively (interaction $P < 0.01$). The yield of milk protein decreased

by 102 g/d and 10 g/d with increased regrowth interval after early and late first cut, respectively (interaction $P < 0.01$). Feeding the TC silage diet increased ($P < 0.01$) the concentrations of milk protein and urea and yield of milk fat, protein, and lactose compared with cows fed the second-cut silage diets.

Milk N efficiency ($P < 0.01$) increased as a consequence of postponed first cut and longer regrowth interval, and increasing the regrowth interval also led to lower ($P = 0.01$) feed efficiency. Feeding the TC silage diet improved ($P < 0.01$) feed efficiency, but decreased MNE ($P < 0.01$), compared with cows fed the second-cut silage diets. Cows fed TC silage had higher ($P < 0.01$) MNE but lower ($P = 0.02$) BW compared with cows fed the second-cut silage diets.

The total-tract digestibility of dietary components decreased ($P < 0.01$) with postponed first cut and longer regrowth interval (Table 4). The TC silage diet was more ($P \leq 0.01$) digestible than the second-cut silage diets.

Gas Emissions

Postponing the first cut decreased ($P < 0.01$) total emissions of CO₂, but increased ($P \leq 0.02$) CH₄ yield (g/kg of DMI) and CH₄/CO₂ ratio (Table 5). With longer regrowth interval, total emissions of CO₂ decreased ($P < 0.01$), but CH₄ intensity (g/kg ECM) and CH₄/CO₂ increased ($P \leq 0.02$). Total CH₄ emissions decreased by 16 g/d and increased by 16 g/d with increased regrowth interval after early and late first cut, respectively (interaction $P = 0.01$). Feeding the TC silage diet decreased ($P \leq 0.01$) CH₄ intensity and CH₄/CO₂ ratio, but increased ($P < 0.01$) total CO₂ emissions and CO₂ yield compared with cows fed the second-cut silage diets.

Efficiency of Energy Use

The GE intake and energy balance decreased by 33 and 24 MJ/d and increased by 10 and 1 MJ/d with increased regrowth interval after early and late first cut, respectively (interaction $P \leq 0.02$; Table 6). The decrease in intake of DE and ME and urinary energy output with increased regrowth interval was greater (interaction $P \leq 0.04$) after early than late first cut. Postponing the first cut decreased ($P \leq 0.04$) energy output in milk and HP. With longer regrowth interval fecal energy output increased ($P < 0.01$) and energy output in milk decreased ($P < 0.01$). Feeding the TC silage diet increased ($P \leq 0.03$) intake of DE, and energy outputs as urine, milk, and HP, but decreased ($P = 0.03$) fecal energy output compared with cows fed the second-cut silage diets.

Table 1. Chemical composition and nutritional value of silages and supplementary concentrate feeds used in experimental diets fed to dairy cows (g/kg of DM unless otherwise stated); mean and SD based on 4 determinations

Item ¹	Silage ²						Concentrate feed			
	EE	EL	LE	LL	TC		Crimped barley	Canola meal	GreenFeed concentrate	
DM, g/kg	262 (4.4)	265 (4.9)	261 (9.2)	311 (8.4)	220 (4.8)		816 (9.0)	899 (0.9)	897 (0.6)	
Chemical composition										
OM	921 (2.8)	928 (2.8)	931 (3.7)	931 (5.4)	912 (4.0)		971 (3.3)	928 (2.6)	941 (3.9)	
CP	178 (5.9)	125 (4.8)	137 (9.3)	111 (3.0)	197 (15.6)		125 (10.1)	349 (28.9)	185 (5.0)	
NDF	523 (13.0)	557 (16.0)	556 (11.0)	524 (17.9)	459 (21.3)		216 (21.1)	285 (5.9)	186 (5.2)	
Indigestible NDF	76 (8.9)	148 (7.8)	143 (13.0)	177 (9.7)	78 (10.9)		49 (0.3)	99 (0.4)	47 (0.3)	
OMD, g/kg	732 (9.8)	641 (10.1)	647 (13.0)	609 (12.1)	737 (12.8)		—	—	—	
DOM	666 (7.8)	577 (8.6)	586 (15.8)	547 (10.0)	662 (13.1)		—	—	—	
Fermentation quality										
pH	3.8 (0.03)	3.6 (0.03)	3.7 (0.02)	3.7 (0.03)	3.8 (0.01)		—	—	—	
Ammonia-N, g/kg of N	32 (4.9)	37 (3.3)	34 (1.2)	74 (5.4)	42 (8.8)		—	—	—	
Lactic acid	79 (8.1)	102 (8.3)	110 (3.8)	83 (10.7)	117 (13.0)		—	—	—	
Acetic acid	18 (2.0)	15 (1.0)	20 (1.2)	27 (2.1)	16 (1.5)		—	—	—	
Butyric acid	<0.3	<0.3	<0.3	<0.3	<0.4		—	—	—	
Gross energy, MJ/kg of DM	18.4 (0.07)	18.1 (0.13)	18.3 (0.03)	18.0 (0.08)	18.5 (0.10)		18.6 (0.10)	19.6 (0.26)	19.3 (0.11)	
Nutritional values										
ME, MJ/kg of DM	10.7 (0.13)	9.2 (0.14)	9.4 (0.25)	8.8 (0.16)	10.6 (0.21)		12.6 (0.06)	11.5 (0.04)	11.8 (0.07)	
MP	84 (1.0)	69 (0.7)	72 (0.8)	65 (1.1)	86 (2.3)		99 (1.3)	165 (7.6)	139 (1.4)	
PBV	54 (5.3)	21 (5.1)	31 (10.3)	14 (2.5)	72 (13.4)		-16 (8.7)	133 (19.9)	0.4 (3.46)	
SDMI index	98.6 (1.48)	81.3 (2.22)	80.9 (3.19)	79.8 (1.21)	90.0 (3.21)		—	—	—	
Growing degree-days	396	564	647	809	477		—	—	—	

¹OMD = organic matter digestibility; DOM = digestible organic matter; PBV = protein balance in the rumen; SDMI index = silage dry matter intake index according to Huhtanen et al. (2007); growing degree-days is calculated by taking the integral of warmth above 5°C.

²EE = early second-cut silage after early first cut; EL = late second-cut silage after early first cut; LE = early second-cut silage after late first cut; LL = late second-cut silage after late first cut; TC, third-cut silage after early first and second cut.

Table 2. Chemical composition and nutritional values of experimental diets fed to dairy cows (g/kg of DM unless otherwise stated); mean and SD for 4 determinations

Item ¹	Diet ²				
	EE	EL	LE	LL	TC
Chemical composition					
OM	926 (2.3)	929 (1.9)	931 (3.1)	931 (2.4)	921 (2.7)
CP	172 (6.1)	144 (3.7)	151 (7.7)	137 (6.6)	182 (6.3)
NDF	379 (13.7)	396 (12.2)	394 (8.9)	379 (15.8)	345 (15.1)
Indigestible NDF	72 (4.2)	114 (4.0)	110 (6.3)	130 (6.6)	75 (7.1)
Nutritional values					
ME, MJ/kg of DM	11.3 (0.07)	10.5 (0.07)	10.6 (0.13)	10.2 (0.10)	11.2 (0.12)
MP	96 (1.3)	88 (1.3)	89 (1.5)	86 (1.5)	97 (1.3)
PBV	35 (5.5)	17 (3.1)	22 (7.5)	13 (5.5)	44 (5.5)

¹PBV = protein balance in the rumen.

²Silage source differed between experimental diets: EE = diet with early second-cut silage after early first cut; EL = diet with late second-cut silage after early first cut; LE = diet with early second-cut silage after late first cut; LL = diet with late second-cut silage after late first cut; TC = diet with third-cut silage after early first and second cut.

Postponing the first cut decreased ($P < 0.01$) the ratios DE/GE, ME/GE, and energy balance/ME intake and k_i , but increased ($P < 0.01$) CH₄ energy proportion in both GE and DE, and HP and milk energy proportions in ME intake. With longer regrowth interval, the

proportions of DE and ME in GE decreased ($P < 0.01$), but CH₄ energy proportion in DE increased ($P < 0.01$). Feeding the TC silage diet increased ($P < 0.01$) the DE/GE ratio but decreased ($P = 0.02$) the ME/DE ratio compared with cows fed the second-cut silage diets.

Table 3. Intake and production data for cows fed the experimental diets (n = 24)

Item ¹	Diet ²					SEM	Probability ³			
	EE	EL	LE	LL	TC		C1	C2	C3	C4
Intake, kg/d										
Total DM	22.4	21.2	20.3	20.6	20.8	0.19	<0.01	0.01	<0.01	0.20
Silage DM	11.9	11.2	10.6	10.8	10.9	0.16	<0.01	0.03	<0.01	0.07
CP	3.9	3.0	3.1	2.8	3.8	0.03	<0.01	<0.01	<0.01	<0.01
NDF	8.5	8.4	8.0	7.8	7.2	0.09	<0.01	0.09	0.69	<0.01
Indigestible NDF	1.6	2.4	2.2	2.6	1.6	0.03	<0.01	<0.01	<0.01	<0.01
ME, MJ/d	233	201	198	193	210	2.0	<0.01	<0.01	<0.01	0.09
MP	1.97	1.69	1.67	1.61	1.81	0.016	<0.01	<0.01	<0.01	<0.01
PBV	0.72	0.32	0.43	0.23	0.84	0.017	<0.01	<0.01	<0.01	<0.01
Milk yield, kg/d	29.4	27.2	27.3	26.4	29.7	0.33	<0.01	<0.01	0.06	<0.01
ECM yield, kg/d	31.3	28.6	28.7	27.6	31.2	0.35	<0.01	<0.01	0.02	<0.01
Milk composition, g/kg										
Fat	44.4	43.2	43.7	42.7	43.0	0.50	0.26	0.03	0.86	0.32
Protein	36.8	36.0	35.6	36.1	36.7	0.18	<0.01	0.41	<0.01	<0.01
Lactose	46.6	46.3	46.4	46.3	46.4	0.17	0.82	0.22	0.53	0.89
Milk urea, mM	4.58	4.24	4.43	3.78	5.17	0.085	<0.01	<0.01	0.06	<0.01
Composition yield, g/d										
Fat	1,284	1,162	1,172	1,114	1,247	21.6	<0.01	<0.01	0.13	<0.01
Protein	1,068	966	947	937	1,068	14.2	<0.01	<0.01	<0.01	<0.01
Lactose	1,353	1,253	1,244	1,219	1,351	25.9	<0.01	0.02	0.14	<0.01
Feed efficiency, kg/kg	1.40	1.35	1.43	1.35	1.50	0.025	0.45	0.01	0.52	<0.01
N efficiency, g/kg	273	310	299	323	273	5.3	<0.01	<0.01	0.21	<0.01
BCS	3.1	3.1	3.0	3.0	3.0	0.03	0.18	0.34	0.72	0.69
BW, kg	615	617	614	615	611	1.5	0.27	0.17	0.69	0.02

¹PBV = protein balance in the rumen; feed efficiency = ECM yield/total DM intake; N efficiency = milk N output/feed N intake; BCS was assessed according to Edmonson et al. (1989).

²Silage source differed between experimental diets: EE = diet with early second-cut silage after early first cut; EL = diet with late second-cut silage after early first cut; LE = diet with early second-cut silage after late first cut; LL = diet with late second-cut silage after late first cut; TC = diet with third-cut silage after early first and second cut.

³Probability of treatments effects: C1 = effect of harvest time of first cut on second cut; C2 = effect of growth stage in second cut; C3 = interaction between the effect of harvest time of first cut and second cut; C4 = effect of second versus third cut.

Table 4. Digestibility of dietary chemical components for cows in the experiment (n = 12)

Item	Diet ¹					SEM	Probability ²			
	EE	EL	LE	LL	TC		C1	C2	C3	C4
OM	747	694	702	660	738	6.8	<0.01	<0.01	0.43	<0.01
CP	716	662	674	606	699	11.6	<0.01	<0.01	0.45	0.01
NDF	674	553	573	481	645	9.5	<0.01	<0.01	0.14	<0.01
Potentially digestible NDF	831	780	797	725	823	11.2	<0.01	<0.01	0.38	<0.01

¹Silage source differed between experimental diets: EE = diet with early second-cut silage after early first cut; EL = diet with late second-cut silage after early first cut; LE = diet with early second-cut silage after late first cut; LL = diet with late second-cut silage after late first cut; TC = diet with third-cut silage after early first and second cut.

²Probability of treatments effects: C1 = effect of harvest time of first cut on second cut; C2 = effect of growth stage in second cut; C3 = interaction between the effect of harvest time of first cut and second cut; C4 = effect of second versus third cut.

Nitrogen Metabolism

The decrease in urine and urinary N outputs with increased regrowth interval were greater (interaction $P \leq 0.01$) after early than late first cut (Table 7). The fecal N output decreased by 15 g/d and increased by 19 g/d with increased regrowth interval after early and late first cut (interaction $P = 0.03$). Postponing the first cut decreased ($P = 0.01$) the proportion of urinary N of N intake, but increased ($P < 0.01$) the proportion of fecal N of N intake. Longer regrowth interval decreased ($P < 0.01$) the proportion of urinary N of N intake, but fecal DM output and fecal N proportion of N intake increased ($P < 0.01$). Feeding the TC silage diet increased ($P < 0.01$) outputs of urine, urinary and fecal N, and urinary N proportion of N intake, but decreased ($P \leq 0.03$) fecal DM output and fecal N proportion of N intake, compared with cows fed the second-cut silage diets.

DISCUSSION

Milk production based on grass-based systems has a very important role in agriculture at northern lati-

tudes in Europe. Grass yield, and nutritive value and ensiling quality of the harvested crop are key factors determining the production cost of milk. This study represents a systematic and novel contribution of milk production responses to different harvesting strategies of regrowth silages. Diets were formulated based on feed quality characteristics of the theoretical highest intake regrowth silage (EE) to be able to display true effects of silage source on dairy cow performance.

Silage Composition

Lower concentration of CP and higher concentration of iNDF in regrowth silages after a late first cut or longer regrowth interval are in line with previous studies (Kuoppala et al., 2008; Alstrup et al., 2016). Daily changes in CP and iNDF concentration due to delayed second cut were marginally higher than those observed by Kuoppala et al. (2008). Much higher iNDF concentration in LE than in EE silage (143 vs. 76 g/kg DM) despite of the same length of growth period can be attributed to greater number of degree-days. Overall, degree-days and silage iNDF concentration were strong-

Table 5. Methane and carbon dioxide emissions for cows fed the experimental diets (n = 24)

Item	Diet ¹					SEM	Probability ²			
	EE	EL	LE	LL	TC		C1	C2	C3	C4
CH ₄										
g/d	435	419	406	422	424	6.2	0.04	0.96	0.01	0.63
g/kg of ECM	14.1	15.4	14.8	15.7	13.7	0.46	0.29	0.02	0.59	0.01
g/kg of DMI	20.2	20.7	21.1	21.5	21.3	0.33	<0.01	0.17	0.77	0.27
CO ₂										
g/d	11,544	10,883	10,644	10,495	11,450	136.2	<0.01	<0.01	0.06	<0.01
g/kg of ECM	374	406	392	394	371	14.0	0.84	0.22	0.27	0.19
g/kg of DMI	517	518	532	516	554	8.1	0.42	0.37	0.35	<0.01
CH ₄ /CO ₂ , g/kg	37.7	38.5	38.2	40.2	37.0	0.46	0.02	<0.01	0.15	<0.01

¹Silage source differed between experimental diets: EE = diet with early second-cut silage after early first cut; EL = diet with late second-cut silage after early first cut; LE = diet with early second-cut silage after late first cut; LL = diet with late second-cut silage after late first cut; TC = diet with third-cut silage after early first and second cut.

²Probability of treatments effects: C1 = effect of harvest time of first cut on second cut; C2 = effect of growth stage in second cut; C3 = interaction between the effect of harvest time of first cut and second cut; C4 = effect of second versus third cut.

Table 6. Energy intake, output, and utilization for cows fed the experimental diets (n = 12)

Item ¹	Diet ²					SEM	Probability ³			
	EE	EL	LE	LL	TC		C1	C2	C3	C4
Energy intake and output, MJ/d										
GE intake	415	382	359	369	386	5.0	<0.01	0.03	<0.01	0.41
Fecal energy	121	134	124	143	121	3.9	0.14	<0.01	0.46	0.03
DE intake	294	248	236	226	265	4.3	<0.01	<0.01	<0.01	<0.01
Urine energy	22	13	16	11	21	1.2	<0.01	<0.01	0.04	<0.01
CH ₄ energy	25	24	24	24	25	0.5	<0.01	0.66	0.20	0.19
ME intake	247	211	197	191	219	4.8	<0.01	<0.01	<0.01	0.15
Milk energy	105	98	99	94	105	1.3	<0.01	<0.01	0.70	<0.01
Heat production	120	115	114	113	121	2.0	0.04	0.10	0.39	0.03
Energy balance	22	-2	-17	-16	-7	5.7	<0.01	0.04	0.02	0.58
Energy use										
DE/GE	0.709	0.647	0.657	0.612	0.689	0.0083	<0.01	<0.01	0.30	<0.01
ME/GE	0.594	0.550	0.546	0.516	0.569	0.0096	<0.01	<0.01	0.45	0.10
ME/DE	0.840	0.850	0.832	0.841	0.825	0.0054	0.12	0.07	0.97	0.01
CH ₄ energy/GE intake	0.060	0.064	0.067	0.067	0.065	0.0014	<0.01	0.12	0.16	0.60
CH ₄ energy/DE intake	0.084	0.099	0.102	0.110	0.095	0.0028	<0.01	<0.01	0.23	0.23
Heat production/ME intake	0.493	0.558	0.603	0.612	0.560	0.0194	<0.01	0.06	0.14	0.76
Energy balance/ME intake	0.078	-0.034	-0.127	-0.122	-0.048	0.0344	<0.01	0.12	0.09	0.92
Milk energy/ME intake	0.429	0.476	0.524	0.510	0.488	0.0161	<0.01	0.30	0.06	0.87
k _l	0.692	0.662	0.634	0.625	0.643	0.0176	<0.01	0.25	0.55	0.58

¹GE = gross energy; DE = digestible energy; k_l = efficiency of ME use for lactation.

²Silage source differed between experimental diets: EE = diet with early second-cut silage after early first cut; EL = diet with late second-cut silage after early first cut; LE = diet with early second-cut silage after late first cut; LL = diet with late second-cut silage after late first cut; TC = diet with third-cut silage after early first and second cut.

³Probability of treatments effects: C1 = effect of harvest time of first cut on second cut; C2 = effect of growth stage in second cut; C3 = interaction between the effect of harvest time of first cut and second cut; C4 = effect of second versus third cut.

ly correlated ($R^2 = 0.87$). High temperatures increase lignification and will decrease digestibility (Van Soest, 1994). In earlier studies (Lindberg and Lindgren, 1988; Kuoppala et al., 2008), the NDF concentration always increased with longer regrowth interval. However, in the present study the NDF concentration of LL silage was 32 g/kg lower than that of LE silage. This suggests that the herbage grown between LE and LL was highly digestible. This could be related to rapidly shortening day length, slower degree-day accumulation and less radiation during late summer slowing down the ligni-

fication process. In addition, it might be attributable to new growth coming out during that period. In the present study, the TC silage and EE silage were rather similar in chemical compositions and nutritive values, except for lower NDF concentration in TC. With the same length of regrowth interval, the iNDF and silage DOM concentrations in TC and EE were about the same, despite the rather different growing conditions.

Conflicting effects of advancing maturity on the digestibility of regrowth silages have been reported in the literature. In this study, the average decrease in silage

Table 7. Nitrogen metabolism for cows fed the experimental diets (n = 12)

Item	Diet ¹					SEM	Probability ²			
	EE	EL	LE	LL	TC		C1	C2	C3	C4
Urine excretion										
Urine weight, kg/d	26.7	15.8	18.3	12.9	24.1	1.08	<0.01	<0.01	0.01	<0.01
N, g/d	256	161	177	130	257	7.8	<0.01	<0.01	<0.01	<0.01
N, g/kg of total N intake	395	316	357	278	404	13.4	0.01	<0.01	0.99	<0.01
Fecal output										
DM, kg/d	6.3	7.0	6.4	7.5	6.3	0.20	0.13	<0.01	0.47	0.02
N, g/d	184	169	161	180	196	7.2	0.43	0.85	0.03	<0.01
N, g/kg of total N intake	285	338	324	390	305	11.6	<0.01	<0.01	0.58	0.03

¹Silage source differed between experimental diets: EE = diet with early second-cut silage after early first cut; EL = diet with late second-cut silage after early first cut; LE = diet with early second-cut silage after late first cut; LL = diet with late second-cut silage after late first cut; TC = diet with third-cut silage after early first and second cut.

²Probability of treatments effects: C1 = effect of harvest time of first cut on second cut; C2 = effect of growth stage in second cut; C3 = interaction between the effect of harvest time of first cut and second cut; C4 = effect of second versus third cut.

OMD was 4.0 g/kg and days for second cuts, which was in line with the value of 3.50 g/kg and days in Kuoppala et al. (2008). However, Keady and O'Kiely (1998) reported that the rate of decline in OMD in regrowth of perennial ryegrass was slow, which might be explained by variations in grass species and weather conditions between years.

Short wilting time and additives were used to minimize the confounding effects of DM concentration and fermentation quality between the silages in this study. Overall, the variation in DM and fermentation quality between the second-cut silages was rather small. The lower DM concentration in TC silage was because of poorer weather conditions for wilting in late summer. In addition to the lower DM concentration, higher total acid concentration could partly explain the reduced intake of the TC silage. Total acid concentration in silage is strongly negatively correlated with SDMI in cattle (Huhtanen et al., 2007; Krizsan and Randby, 2007).

Intake, Milk Production, and Digestibility

In this study, postponing the first cut and increasing the length of the regrowth interval decreased SDMI. Silage DOM was a better predictor of SDMI than NDF concentration, reflecting the effect of potential NDF digestibility on intake (Huhtanen et al., 2007). Each 10 g increase in silage DOM resulted in a 0.180 kg increase in SDMI, which was close to the 0.175 kg increase reported in a meta-analysis ($n = 81$) by Huhtanen et al. (2007). Intake response per unit of SDMI index, which takes into account the effects of the concentrations of DM, NDF, and total acids, was 0.08 kg, which compared well with the default value of 0.10 kg (Huhtanen et al., 2007). Intake of the TC diet in this study was lower than expected on the basis of predicted intake potential suggested by Huhtanen et al. (2007), but in agreement with previous studies on third-cut grass silage (Sairanen and Juutinen, 2013; Sairanen et al., 2016). Although the DOM of the TC silage was close to that of the best second-cut silage EE (662 vs. 666 g/kg DM), the SDMI index of the TC diet was 8.6 units lower than for EE due to its lower DM concentration and higher total acid concentration. However, the lower SDMI index only accounted for about 50% of the observed difference in DMI, which suggests that other factors could be involved. For example, humid weather conditions in late summer and autumn can favor activities of epiphytic flora in herbage (Ercolani, 1991). Possible metabolites of these microbes might affect feed intake. Lower NDF intake in cows fed TC silage despite high digestibility suggests that factors other than rumen fill limited the intake of diets based on the TC silage.

In this study, every 10 g increase in silage DOM resulted in a 0.31 kg increase in ECM yield, which compared rather well with that (0.45 kg increase) estimated by Huhtanen et al. (2013) from factorial studies evaluating the effects of silage digestibility. The ECM yield was positively related to dietary MEI. In the present study the ECM yield increased by 0.09 kg/MJ increase in dietary MEI, which is identical with the 0.09 kg/MJ increase derived by Huhtanen and Nousiainen (2012) from a large data set and the 0.10 kg/MJ increase observed by Kuoppala et al. (2008) in a study comparing primary and regrowth silages harvested at different stages of maturity. The milk and ECM yields of cows fed the TC diet were similar to those of cows fed the EE diet, despite lower ME intake. The ECM yield for the TC diet was approximately 1.5 kg higher than estimated from the relationship between dietary ME intake and ECM yield. Similar results have been reported by Sairanen et al. (2016), but the reasons for the better performance of cows fed diets based on third-cut silage are still unclear. One possible reason is different partitioning of nutrients between milk and body tissues. Energy metabolism data suggested that cows fed the EE diet partitioned more energy to tissues than cows fed the TC diet.

Reported effects of extending the regrowth interval on milk fat concentration are inconsistent. In the present study the milk fat concentration decreased with extended regrowth interval. However, Kuoppala et al. (2008) and Warner et al. (2016) only observed marginal differences in milk fat concentration with extended regrowth interval. Milk fat synthesis in dairy cows can be influenced by silage factors. In addition to silage DOM and NDF concentration, fermentation characteristics can influence milk fat concentration. Increased concentrations of lactic acid in silage decrease milk fat concentration (Huhtanen et al., 2003), reflecting fermentation of lactic acid to propionate in the rumen.

Milk protein concentration was significantly higher (36.4 vs. 35.9 g/kg) in cows fed second-cut silages after early compared with late first cut, which was mainly attributable to their higher ME concentration (10.0 vs. 9.1 MJ/kg DM) and dietary MEI (217 vs. 196 MJ/d). Dietary CP concentration is the most important single factor affecting milk urea concentration (Broderick and Clayton, 1997; Nousiainen et al., 2004). Milk urea concentration increased with early first cut and shorter regrowth interval, reflecting increased dietary CP concentration.

Total-tract digestibility decreased with postponed first cut and extended regrowth interval for all nutrients, which is in line with Kuoppala et al. (2008) and Warner et al. (2016). The differences in dietary OMD were strongly positively correlated with silage

OMD ($R^2 = 0.94$), which is in line with the results of a meta-analysis ($n = 497$) by Nousiainen et al. (2009). In the present study, silage iNDF concentration was strongly negatively correlated with dietary OMD in cows ($R^2 = 0.96$), in agreement with findings in studies by Huhtanen et al. (2006) and Krizsan et al. (2014) on sheep. The difference in dietary NDF digestibility was almost completely related to pdNDF/NDF ratio ($R^2 = 1.00$). Regression coefficient was 1.26, indicating that actual NDF digestibility improved more than potential NDF digestibility. This could be attributed to the faster digestion rate of NDF in silages with higher pdNDF/NDF. Rinne et al. (2002) demonstrated a positive relationship between digestion rate of pdNDF in silages and OMD and they reported longer rumen residence time of silages with higher OMD, which could contribute to improved pdNDF digestibility of regrowth silages after an early first cut. The TC silage diet was more digestible than the second-cut silage diets, which is in line with earlier reports by Sairanen and Juutinen (2013) and Huuskonen and Pesonen (2017).

Gas Emissions

The decrease in total CH_4 emissions with increased regrowth interval after early first cut agrees with earlier findings by Warner et al. (2016). Differences between the diets in total CH_4 emissions were mainly related to DMI ($R^2 = 0.71$). In the present study, the more digestible regrowth silages after early than late first cut had lower average CH_4 yield (20.5 vs. 21.3 g/kg DMI), in agreement with Brask et al. (2013) and Warner et al. (2016). However, the effects of silage digestibility on CH_4 yield are not consistent. Beever et al. (1988) reported greater CH_4 yield with more digestible early-cut silage than late cut silage. Ramin and Huhtanen (2013) also showed a positive relationship between digestibility and CH_4 yield based on a meta-analysis with 207 observations, with the CH_4 intensity decreasing by 7.1% on average with shorter regrowth interval. The CH_4 intensity in the present study was more related to ECM yield ($R^2 = 0.91$) than total CH_4 emissions ($R^2 = 0.14$). Warner et al. (2016) also found that shorter regrowth interval decreased the CH_4 intensity, because feeding the more digestible regrowth silages from shorter regrowth interval always improved ECM yield.

The ratio of CH_4/CO_2 in breath is an ideal variable for expressing the microbial fermentation efficiency of the feed, because it directly describes the proportion of the carbon excreted that was not metabolized to CO_2 (Madsen et al., 2010). In the present study, the CH_4/CO_2 ratio was much more closely related to CO_2 production than CH_4 production ($R^2 = 0.68$ vs. 0.04). Therefore, the higher ratio of CH_4/CO_2 when postpon-

ing the first cut or extending the regrowth interval was mainly from the decreased CO_2 production from the metabolism of decreased ME intake. The lower CH_4/CO_2 ratio of the diet based on TC silage could have resulted from mobilization of body tissues, producing relatively more CO_2 but not CH_4 .

Enteric CH_4 emissions from dairy cows also represented a loss of energy. The average CH_4 -E/GE and CH_4 -E/DE was 0.065 and 0.098, respectively, which compared well with the values (0.068 and 0.089) in a meta-analysis ($n = 247$) by Yan et al. (2000) based on dairy cows fed grass silages. The CH_4 -E/DE ratio decreased with increased dietary OMD, in agreement with Ramin and Huhtanen (2013). This suggests a shift of digestion from rumen to intestine (Moss et al., 2000) and changes in rumen fermentation pattern toward increased propionate and reduced acetate (Johnson and Johnson, 1995) when cows are fed regrowth silages after an early first cut or with a shorter regrowth interval.

Energy and N Utilization

There were large differences in urinary energy output between the diets. High urinary energy output was associated with diets (EE and TC) with high OMD and CP concentrations. Similar or even higher values have been reported by Hindrichsen et al. (2006) for cows fed ryegrass silage diets and by van Dorland et al. (2006) for cows fed grass silage and fresh clover diets. Givens et al. (1989) reported large variation in urinary energy as a proportion of GE intake (mean 0.06, range 0.01–0.11, SD 0.015) in sheep fed different silages. Urinary energy is positively related to the concentrations of DOM and CP (Givens et al., 1989). In the study by Beever et al. (1988), urinary energy output was almost doubled in growing cattle fed early-cut compared with late-cut grass silage. Urinary energy output decreased by about 20% when 75% of grass silage was replaced with maize silages in a study by Cammell et al. (2000). In the present study, proportionally 0.168 of the incremental DE was lost as urinary energy with improved diet digestibility. Although there are uncertainties in estimating urinary energy output from spot sampling and calculated urinary output, possible errors in estimating urinary volume can only account for a small proportion of observed differences between the diets. For example, if fecal N output had been underestimated by 10% for the diet EE and overestimated by 10% for the diet LL, the difference between these diets in estimated urinary energy would decrease from 11.4 to 8.3 MJ/d. The close relationships between silage OMD estimated from concentrations of iNDF and NDF, and observed OMD in cows do not indicate any major relative errors in OMD and estimated fecal N output. There are also uncertain-

ties in predicting retained N from calculated energy balance. However, the N balance of dairy cows in mid-lactation is close to zero; even 1 kg/d daily gain (about 25 g N) for cows fed the EE and TC diets would reduce estimated urinary N, and consequently energy output, by about 10%. It is possible that early harvested grass silages contain low molecular weight substances such as phenols and essential oils that are absorbed, but not metabolized and excreted in urine.

Although the $\text{CH}_4\text{-E}/\text{DE}$ ratio decreased with improved diet digestibility, in agreement with Beever et al. (1988) and Ramin and Huhtanen (2013), the ME/GE ratio actually marginally decreased with diet digestibility. It appears that decreases in $\text{CH}_4\text{-E}$ and increases in UE/DE compensate for each other (Beever et al., 1988; present study), supporting the use of a constant factor for converting DOM to ME (MAFF, 1975).

The mean efficiency of ME utilization above maintenance was 0.65, which is close to values derived from respiration chamber studies (Agnew and Yan, 2000; Moraes et al., 2015). This suggests that HP estimated from CO_2 emissions measured by the GEM system and the empirical relationship between CO_2 production and O_2 consumption resulted in a reasonable mean estimate of the k_1 value. However, whether the GEM system is accurate enough to determine differences between diets needs further validation in respiration chamber studies. Because of the 3.2-fold greater effect of O_2 than CO_2 on HP, differences in respiratory quotient increase the error in k_1 estimates. Ranking of the diets according to the k_1 value was as expected according to the NRC (2001) and AFRC (1990) systems, but observed differences between the diets were greater than predicted differences. A constant maintenance requirement was used in the present study for estimating k_1 . However, increased maintenance requirement with high fiber diets has been demonstrated in several studies (Agnew and Yan, 2000). The differences in the k_1 values were more associated with dietary iNDF concentration than total NDF concentration, which can be related to greater gut fill and greater work in rumination and digestion, factors which all contribute to greater maintenance requirement (Reynolds et al., 1991). The k_1 values are also sensitive to assumed utilization of ME for body tissue gain.

In the present study, MNE increased with postponed first cut and increasing regrowth interval. This trend agrees with findings in studies investigating the effects of maturity of grass silage at harvest (e.g., Rinne et al., 1999; Kuoppala et al., 2008; Randby et al., 2012). Warner et al. (2016) also reported elevated MNE with extended regrowth interval. The MNE was closely related to N intake ($R^2 = 0.96$) or dietary CP concentration ($R^2 = 0.95$), in agreement with Castillo et al.

(2001) and Huhtanen et al. (2008). The MNE decreased by 1.16 g/kg with every 1 g/kg DM increase in dietary CP concentration, which compares well with the value of 1.20 g/kg in a meta-analysis ($n = 998$) by Huhtanen et al. (2008), especially in studies investigating the effects of silage digestibility (1.16 g/kg CP; $n = 81$). A high proportion (0.72) of incremental N intake with improved digestibility was excreted as urinary N, which is more susceptible to evaporation and leaching losses than fecal N. Milk urea N was a good predictor of urinary N output, supporting earlier results (Jonker et al., 1998; Kauffman and St-Pierre, 2001; Nousiainen et al., 2004). The recovery of incremental N intake as milk N was 0.12, which is slightly higher than reported for soybean meal (0.10) but slightly lower than reported for rapeseed meal (0.135) in a meta-analysis of protein supplementation studies (Huhtanen et al., 2011).

CONCLUSIONS

This study showed that feeding regrowth silage in 2- or 3-cut systems harvested after an early first cut and after a short regrowth interval promoted better dairy performance and feed intake, and higher efficiency of feed and energy utilization, but poorer N efficiency. Third-cut silage gave the highest milk yield and feed efficiency. A higher proportion of DE was excreted as urinary energy from silages with higher digestibility, a finding that deserves further attention. The present study generated information on dairy cow production responses to regrowth silages from different harvesting strategies. For practical applications the effects of harvesting systems on DM yield and overall farm profits should be considered, including silage nutritive value and production responses. To optimize forage harvesting and production whole farm models taking into account the effects of harvesting systems on DM yield, intake potential and nutritive value should be developed.

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