

1 **Effect of replacing organic grass-clover silage from primary growth**
2 **with regrowth on feed intake and milk yield of dairy cows**

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14 ABSTRACT

15 Under Norwegian conditions diets based on primary growth silage (PG) typically increase milk yield compared
16 to silage prepared from the regrowth (RG). Organic PG, dominated by immature grasses, are often high in
17 energy and low in CP, whereas the opposite is the case for organic RG harvests, dominated by clover. Here, we
18 tested the hypotheses that increasing proportions of RG will reduce the total supply of metabolizable energy, but
19 increase the CP-intake, and that there is a dietary optimal mix of PG and RG to meet requirements for optimal
20 milk production. Sixteen Norwegian Red cows were used in an experiment designed with four balanced 4×4
21 Latin squares with 21-days periods to evaluate the effect of incremental replacement of PG with RG on feed
22 intake, nutrient digestion and milk production. Silages were prepared from PG and RG of an organically
23 managed grassland. Treatments comprised silages fed *ad libitum* with RG replacing PG in ratios of 0, 0.33, 0.67
24 and 1 on DM basis. Additionally concentrate was offered with 8 kg for pluriparous and 7 kg for primiparous
25 cows. The PG had higher contents metabolizable energy (ME), potentially degradable neutral detergent fiber
26 (NDF) and water soluble carbohydrates, while RG contained more crude protein (CP) and indigestible NDF.
27 The already mentioned characteristics led to higher intakes of DM, organic matter, NDF and ME, and lower
28 intakes of CP and indigestible NDF with increasing proportions of PG in the diet. Milk yield tended to be higher
29 when PG and RG were offered as a mixture than when fed alone. The milk fat concentration decreased linearly
30 with increasing proportions of RG proportion, while protein concentration was unaffected by diet. This led to a
31 similar production of energy corrected milk among cows fed diets containing PG while cows fed pure RG diet
32 produced 0.9 kg less daily. Silage energy concentration and energy intake influenced milk production more than
33 CP supply.

34

35 **Key words:** dairy cow, organic milk production, regrowth, silage, grass-clover

36 INTRODUCTION

37 Organic dairy production in Norway is becoming more intensive, where increased milk yield has been
38 achieved from increasing amounts of concentrate feed (TINE Rådgiving 2012). High-grade protein plant
39 sources have limited (e.g. rapeseed and pulses) or no (e.g., soybeans) cultivation potential in Norway. Import of
40 particularly soybeans is controversial (Leiber 2014). Homegrown forages may be cheaper to produce but only a
41 high quality forage will realize a profitable production. Grassland yield and productivity of organic dairy
42 production depend largely on biological N-fixation by legumes (Steinshamn 2010). To obtain forage with high
43 fiber digestibility and thereby high energy content, the spring growth has to be harvested when plants are at an
44 early stage of phenological development. Although the growing season in most of Norway is short, such an
45 early primary growth (PG) may constitute less than 50% of the total annual yield and be low in crude protein
46 (CP) (Steinshamn and Thuen 2008). The following regrowth (RG) harvests are often low in NDF, but its
47 digestibility and energy value are found to be inferior to that in PG (Huhtanen et al. 2007). This might have
48 nutritional and production consequences.

49 Dairy cows, fed grass silage from RG, were found to yield less milk than those fed grass silage from PG,
50 which was ascribed to lower dry matter (DM) intake of RG silage (Kuoppala et al. 2008; 2010). Bertilsson and
51 Murphy (2003) found that cows fed pure red clover silage from RG had higher feed intake than those fed pure
52 red clover silage from PG, but milk yield and composition were similar. Additionally they found higher NDF
53 content, lower proportion of potentially degradable NDF (pdNDF) and a lower rate of degradation of NDF in
54 red clover silage prepared from RG than from PG. Vanhatalo et al. (2009) found that DM intake of grass silage
55 decreased, while intake of red clover silage increased with advancing maturity, when comparing different
56 growth stages in the PG of grass and red clover silages. In organic grass-clover harvests, clover contributes to a
57 significant amount of CP in the total crop. Due to slower spring growth rate compared to grass species, the
58 clover proportion is often low in PG of organic grass-clover swards harvested at early grass maturity stages,
59 while in RG harvests the clover proportion is often high (Steinshamn and Thuen 2008). Furthermore, a slow
60 release of N from manure at low spring temperatures limits the CP content in grasses in organic fields
61 (Steinshamn 2001). Therefore, organic PG at high latitudes in Scandinavia, dominated by immature grasses, are
62 often high in energy and low in CP. The CP in conventional agriculture normally starts with high concentrations
63 and sinks rapidly with increasing maturity (Vanhatalo et al. 2009). Because of the mentioned slow N-release in
64 organic clover-dominated RG harvests, the CP concentration is higher than the PG. However, this difference in
65 estimated energy and nutrient contents of PG and RG silages is recognized, but few feeding trials with dairy

66 cows have been carried out in which PG and RG are compared (Khalili et al. 2005; Kuoppala et al. 2008;
67 Halmemies-Beauchet-Filleau et al. 2014).

68 The object of this study was to see if there is a synergetic effect on DMI and milk production when fiber and
69 CP in organic grass-clover silages from PG and RG are mixes, or if the milk production is better of when
70 feeding one of the silages alone. Based on available knowledge, the hypotheses were that increasing dietary
71 proportions of RG will reduce the total supply of metabolizable energy, but increase the N-intake, and that there
72 is a dietary optimal mix of PG and RG to meet the requirements for the highest milk energy production.

73 MATERIALS AND METHODS

74 Experimental Design and Animals

75 An experiment consisting of four balanced 4×4 Latin squares was conducted, each with 4 Norwegian Red
76 cows, 4 organic grass-clover silage diets, and four 21-day periods. Each 21-day period was split in a 9-day
77 adaption period and a 12-day sampling period. The 16 cows entered the experiment in early stage of lactation
78 with (mean \pm standard deviation) 36 ± 17 days in milk and BW of 623 ± 78 kg. At the end of the study, the BW
79 had increased to 643 ± 68 kg. One cow was excluded from the experiment in two periods due to indigestion.
80 Two squares were performed in fall 2012 (one with pluriparous cows and one with primiparous cows) and two
81 squares were performed in spring 2013 (both with pluriparous cows).

82 Grass-clover Silages

83 Both experimental silages were harvested from the same organically managed ley (Counc. of the Eur. Union
84 2007) in its second and third production year in Ås, Norway ($59^{\circ}40'19''N$, $10^{\circ}46'33''E$) in 2012. This ley has
85 been organically cultivated since 1993. Cattle slurry was applied at rates of 30 ton/ha on May 2 and 15 ton/ha on
86 June 15 in accordance with the regulations of organic production in the European Union (Counc. of the Eur.
87 Union 2007). The PG harvest was conducted at early booting stage of timothy (*Phleum pratense* L. cv.
88 'Grindstad') on June 7, and it consisted of timothy (41.9%), meadow fescue (*Festuca pratensis* Huds. cv. 'Fure',
89 25.3%), white clover (*Trifolium repens* L. cv. 'Hebe', 11.3%), smooth meadow grass (*Poa pratensis* L. cv.
90 'Knut', 8.1%), red clover (*Trifolium pratense* L. cv. 'Bjursele', 6.5%) and other legumes, grasses, herbs, and
91 weeds (6.9%). The RG was harvested on July 26 when crop NDF was estimated to be similar to that of the PG.
92 The RG consisted of white clover (39.3%), timothy (29.2%), meadow fescue (14.2%), couch grass (*Elytrigia*
93 *repens*, 6.0%), smooth meadow grass (5.2%), red clover (1.4%), and other legumes, grasses and weeds (4.7%).
94 The botanical composition was determined after walking the whole field in a diagonal grid directly before each

95 harvest. Every tenth step a handful of sward was cut with a pair of scissors. The total sample was then manually
96 sorted into botanical components. Accordingly, all species were separately dried at 105 °C to constant weight
97 (minimum 24 h) and botanical composition was expressed on DM basis.

98 **Silage Production**

99 The crops were cut with Kuhn FC 302 G (Kuhn S.A., Saverne, France) mower and wilted for 9-10 h during
100 daytime or 20-21 h over-night in PG, and 9-12 h during daytime or 27-30 h over-night in RG. Thereafter crops
101 were baled using Orkel GP 1260 (Orkel AS, Fannrem, Norway) fixed chamber roundbaler with 20 fixed knives
102 and a theoretical 54 mm chop length. Ensil®Pluss silage additive (540 g/kg formic acid, 180 g/kg propionic
103 acid; Felleskjøpet Agri SA, Lillestrøm, Norway) was applied to the crop at 5.1 and 5.2 L/ton for PG and RG,
104 respectively. Immediately after baling, each bale was weighed, and a core sample was taken to produce
105 composite samples for approximately eight bales. The bales were wrapped in 8 layers of 0.75 m wide and 0.025
106 mm thick white Trioplast 2000 (Trioplast, Smålandsstenar, Sweden) stretch plastic film using Tanco Autowrap
107 280 ARC (Tanco Autowrap Ltd., Co. Carlow, Ireland) wrapping machine. Yields of harvested herbage were
108 2.84 ton DM/ha in PG and 2.05 ton DM/ha in RG.

109 The weather was sunny with no precipitation during wilting of the PG, but humid and a few raindrops (0.1
110 mm) when mowing began in the RG. Thereafter weather cleared up. The mean daily temperature was 11.1°C
111 and total precipitation 77 mm in May and until PG harvest began on June 5. From PG until the RG harvest, the
112 mean temperature was 14.5°C, and total precipitation 157 mm.

113 **Experimental Diets**

114 Experimental treatments comprised diets with RG replacing PG in the DM ratio 0, 0.33, 0.67 and 1
115 (treatments D1, D2, D3, and D4, respectively). Cows were fed silage *ad libitum* allowing 10% refusals. Before
116 feeding, the silage was chopped using a roundbale chopper (Serigstad RBK 1202, Serigstad Agri, Bryne,
117 Norway) and further with an Epple Blasius 940 (Epple Maschinen GmbH, Wiesensteig, Germany) to a chop
118 length of 3-4 cm. For each cow, the correct proportions of the grass-clover silages were weighed in based on
119 daily DM-determinations. The cows were individually fed three times a day and feeds were divided into the
120 correct weight for each meal. Finally, the mixed-diets were thoroughly mixed by hand to minimize feed
121 selection.

122 A compound concentrate (Felleskjøpet Agri SA, Lillestrøm, Norway) was fed 8 kg/d (as fed basis) to
123 pluriparous- and 7 kg/d to primiparous cows containing peas (26.8%), oats (16.8%), wheat (16.5%), barley

124 (15%), rapeseed cake (10%), molasses (5.5%), rapeseeds (5%), CaCO₃ (1.3%), Ca(H₂PO₄)₂ (0.96%), MgO
125 (0.66%), NaCl (0.6%), Na₂SO₄ (0.63%) and a complementary micro-mineral and vitamin mix (0.25%). The
126 amount of added micro minerals and vitamins from the mix in the concentrate (per kg) was Ca (2,95 g), Cu (15
127 mg), Zn (65 mg), Mn (20 mg), I (3.5 mg), Co (0.25 mg), Se (0.3 mg), Vitamin A (5000 IU), Vitamin D (2000
128 IU) and Vitamin E (40 mg).

129 Cows were housed in a tie-stall with continuous access to water and were fed at 0630, 1415 and 2200 h.
130 Daily feed residues remained in the feed through until directly before the morning feeding. Collection and
131 recording of refusals were manually weighed once daily. Milking was conducted *in situ* daily at 0700 and 1700
132 h. All experimental procedures were carried out in accordance to the laws and regulations controlling
133 experiments with live animals in Norway, made by the Norwegian University of Life Sciences Animal Care and
134 Use Committee and the Norwegian Animal Research Authority.

135 **Sampling, Recordings and Chemical Analyses**

136 Feed intake and milk yield data from day 10 to 21 in each period were used. Daily samples of 500 g from
137 each of the two silages were collected and freeze-dried on Monday to Friday to provide a pooled sample from each
138 week. After thawing a portion of each pooled sample was oven-dried at 59°C to constant weight (minimum 24
139 h) and weighed warm. These portions formed the basis for daily DM intakes (DMI) of silage after correction for
140 volatiles (Norfor 2007b). Analyses of pH was conducted using a Thermo Orion 420A+ pH-meter with Orion
141 9107BN electrode (Thermo Scientific, Beverly, MA, USA). Other portions of the pooled samples were freeze-
142 dried. After drying, samples were equilibrated to room humidity overnight and milled through a 1-mm screen
143 (Retsch GmbH cutting mill, Haan, Germany). Dried samples were analyzed for DM, ash, fat, water soluble
144 carbohydrates (WSC) and NDF as described by Randby et al. (2010). Lignin was analyzed with H₂SO₄ corrected
145 for ash as described by Van Soest et al. (1991) modified according to AOAC (1984). Contents of acid detergent
146 fiber (ADF), NDF-N and ADF-N were equally corrected for ash, Kjeldahl-N was analyzed at Kjeltac 2460 (Foss
147 Electric, Hillerød, Denmark). Indigestible NDF (iNDF) was determined according to Norfor *in sacco* standard
148 070910 (NorFor 2007a) using Sefar Petex 07-11/5-cloth (Sefar AG, Heiden, Switzerland) and 288 h
149 intraruminal incubation. Potentially degradable NDF was calculated as NDF – iNDF. Metabolizable protein
150 content, expressed as amino acids absorbed in the small intestine (AAT) and protein balance in the rumen
151 (PBV), was calculated on background of the evaluation system according to Madsen et al. (1995), based on a
152 fixed value for ruminal efficient protein degradability in experimental silages of 0.80 (Spörndly 2003). The

153 AAT/PBV-system of Madsen et al. (1995) was chosen and considered adequate for this experiment. Minerals
154 were analyzed with inductively coupled plasma atomic emission spectroscopy on IRIS Intrepid II XSP (Thermo
155 Fisher Scientific, Waltham, MA, USA). Chemical composition of the concentrates were analyzed with the same
156 methods as used for the grass-clover silages. Additionally DM was determined at 105°C and starch was
157 determined by an enzymatic method (α -amylase and amyloglucosidase) (Megazyme, Wicklow, Ireland) in
158 samples milled through a 0.5 mm screen.

159 Aliquot milk samples were collected with fractional sampling milk meters (Tru-Test Industries Ltd,
160 Auckland, New Zealand) weekly in six subsequent milkings on day 11 to 14 and 18 to 21. Milk samples were
161 analyzed for fat, protein, lactose, urea and free fatty acids using an infrared spectrophotometer (MilkoScan
162 6000, Foss Electric, Hillerød, Denmark). Calculations of energy corrected milk yield (ECM) included the
163 concentration of lactose and was done according to Volden (2011).

164 **Digestibility Study and Feed Value Calculations**

165 *In vivo* digestibility of silages and concentrate was determined at maintenance level using three wethers per
166 feed. Both PG- and RG-silages were single fed at 880 g/d DM, while the concentrate was evaluated feeding 450
167 g concentrate and 450 g DM of RG-silage a day. The contents of metabolizable energy (ME) and net energy
168 (NE_L) were calculated on the basis of Van Es (1978).

169 **Statistical Analysis**

170 Mean values of measurements from day 10-15 and 16-21 in each period were used for feed intake and milk
171 production results. All data were analyzed using the MIXED procedures of SAS software (SAS Institute Inc
172 2011) with the following model:

$$173 Y_{ijkl} = \mu + c_i + D_j + P(S)_{kl} + S_l + e_{ijkl},$$

174 where μ is the overall mean, c is the random effect of cow ($i = 1$ through 16) and D ($j = 1$ through 4), $P(S)$ (k
175 and $l = 1$ through 4) and S ($l = 1$ through 4) are the fixed effects of diet, period within square and square,
176 respectively. Sum of squares were divided into orthogonal contrasts to assess linear and quadratic effects of the
177 diets. No cubic effects were observed and they are therefore not included.

178 Results were considered significant at $P < 0.05$, and P -values between 0.05 and 0.1 were considered trends,
179 while $P > 0.1$ were considered non-significant.

180 RESULTS

181 Chemical Contents of Experimental Feeds

182 The DM content of the wilted grass-clover crops before ensiling were 36.6% and 31.3% in PG and RG,
183 respectively. On DM basis, 1 kg PG consisted 934 g organic matter (OM), 470 g NDF and 107 g CP. These
184 proportions changed in the RG to respectively 915, 447 and 138 g/kg DM.

185 Table 1 shows the chemical composition of the experimental silages and concentrate. Silages were
186 restrictively fermented and of good quality with low NH₃-N values and no butyric acid. Contents of OM and
187 NDF were slightly higher in the PG than in RG. However, iNDF in NDF was 50% higher and WSC
188 concentration 50% lower in RG than PG, while the CP content was 19% higher in RG than in PG. The CP in
189 RG had higher concentrations of neutral detergent indigestible N (NDIN) and marginally higher concentrations
190 of acid detergent indigestible N (ADIN).

191 The PG had higher in vivo digestibility of DM, OM and NDF, and higher energy values and AAT value than
192 RG (Table 2). The in vivo digestibility of CP was higher in RG than in PG.

193 Nutrient Intake

194 Daily dietary nutrient intake of dairy cows offered the four silage diets are presented in Table 3. There was a
195 significant linear effect of treatment on intake of all dietary parameters except ADF. As such, intake of DM,
196 OM, NDF, pdNDF, WSC, starch and AAT decreased, while CP, PBV, iNDF and fat increased when replacing
197 PG with RG. Total DMI decreased with 5.5% (1.09 kg DM), when PG was totally replaced with RG. The PG
198 had at the same time higher concentrations of OM in the DM, which made the differences in OM intake between
199 diets bigger and resulted in a 6.7% reduction when replacing PG with RG. Although the total intake of N-
200 fractions in RG was higher, there were at the same time higher concentrations of NDIN and ADIN.

201 Milk Production

202 There was a tendency ($P=0.06$) to quadratic response in daily milk yield to increasing proportion of RG in the
203 diet, with the lowest yield in D4 and the highest in D3 (Table 4). However, daily ECM yield decreased linearly
204 with increasing proportion of second cut silage in the diet. Cows fed the D4 diet yielded on average 0.9 kg ECM
205 less than the average of the diets containing PG. A similar effect was seen in daily milk fat yield, while milk fat
206 concentration showed a clear linear decline to increasing proportion of RG. Protein yield showed a quadratic
207 response to dietary treatments, with similar yields in D1, D2 and D3 and a 20 g smaller daily yield in D4. The

208 increasing quadratic effect with increasing proportions of RG in the diet was more pronounced on milk urea
209 than on the protein yield.

210 **Energy and Nitrogen Use Efficiency**

211 The decreasing intake of ME with increasing proportions of RG and still with a high milk production increased
212 the conversion rate of ME into ECM (Table 4). To assess the N efficiency of turning feed N into milk N, the
213 milk N produced was divided with the feed N intake. The N efficiency decreased with increasing RG
214 proportions (Table 4).

215 **DISCUSSION**

216 **Silage Composition**

217 The energy content in grass species like timothy decreases rapidly after shooting due to lignification of plant cell
218 walls. In organically managed **Scandinavian** grasslands, the herbage CP content is generally lower than in
219 conventional because of exclusive use of manure as fertilizer (Byström et al. 2002; Steinshamn and Thuen
220 2008). Early harvested organic PG are therefore usually high in energy but low in CP. The CP content increases
221 in the RG, due to increasing proportions of clover (Steinshamn and Thuen 2008). This experiment compared the
222 effects of replacing silage from early PG with its RG on feed intake and milk production. We hypothesized that
223 dairy cows offered a mixture of PG and RG silage would perform better than feeding each silage alone.

224 We aimed for equal NDF contents in both experimental crops, but ended up with lower concentrations in the
225 RG. The result was a consequence of two factors; there was twice as much clover in RG than in PG and clover
226 species have lower NDF concentrations than grasses (Huhtanen et al. 2006). Especially in relatively mature
227 stands, clover species contribute to raising CP concentrations in grass-clover crops (Vanhatalo et al. 2008).
228 Therefore, in line with our expectations, RG had more CP but less energy due to significantly higher proportions
229 of iNDF and thereby lower digestibility. This is normally the case between PG and RG, regardless of
230 conventionally or organically harvested crops (Khalili et al. 2005; Kuoppala et al. 2008). **Higher clover**
231 **proportion with higher CP content in RG compared to PG were expected as other studies have shown an**
232 **increased clover proportion from 18% to 35% in respectively PG and RG (Govasmark et al. 2005; Steinshamn**
233 **and Thuen 2008).**

234 **Nutrient Intake and Digestibility**

235 In experiments with pure grass silages, Khalili et al. (2005) and Kuoppala et al. (2008) showed declining DMI
236 with decreasing amounts of PG, which supported the finding of the linear decline in DMI when replacing PG
237 with RG. The higher clover content in RG probably prevented a bigger difference in DMI. Increasing maturity
238 of clover species have shown smaller decreases in DMI than in grasses within PG (Vanhatalo et al. 2009),
239 however it is not certain that a pure clover diet would increase DMI, but there is an obvious positive effect of
240 mixing clover and grass on DMI (Bertilsson and Murphy 2003; Dewhurst et al. 2003; Steinshamn 2010).
241 Huhtanen et al. (2007) reported that the best way to predict silage DMI, independent of growth stage and
242 harvest, was to use digestible OM in silage (*D*-value) combined with NDF quality. The model cannot accurately
243 predict the intake response of replacement of grass with clover. Clover is known to have higher passage rate and
244 total digestibility than grasses (Kuoppala et al. 2009). Nonetheless, the linearly decreasing silage DMI from PG
245 to RG in this experiment can still be supported, as the PG was higher in both *D*-value and pdNDF content.
246 Regardless of diet, the intake of starch and free glucose ranged from 10.9 to 11.5% of the total DMI. At these
247 levels, glucose and starch do not reduce NDF digestion significantly, compared to levels exceeding 20% of
248 total DMI (Khalili and Huhtanen 1991; Stensig et al. 1998).

249 Higher energy concentration in PG increased the energy intake differences between diets compared to the
250 differences in DMI, up to 10%. The hypothesis on decreasing energy intake and increasing N-intake with
251 increasing proportion RG was met. Diets containing PG also offered the rumen microbes more energy from
252 carbohydrates, which probably gave a more effective protein synthesis (Madsen et al. 1995). Under the above
253 conditions, increased microbial activity and protein synthesis could partly compensate for lower CP in PG
254 compared to RG silage. Furthermore, the CP concentration in the concentrate was higher than in the forages and
255 increased the total CP concentration in D1 to 131 g/kg DM, which likely provided a sufficient N supply to the
256 rumen microbes for an effective protein synthesis (Schwab et al. 2005). On the other hand, the milk urea of 2.25
257 mMol/L in D1 is below 2.6 mMol/L, which is considered the lower limit for sufficient supply of rumen
258 degradable protein (RDP) (Nousiainen 2004). When comparing these findings it is clear that D1 is on the lower
259 border or below of what can be tolerated of CP in the diet for the microbial protein synthesis. On the contrary,
260 the CP of the RG had higher concentrations of both NDIN and ADIN. Whilst these N fractions are
261 predominantly associated with rumen undegradable protein (RUP), Acid detergent indigestible N is considered
262 indigestible in a diet with grass-clover silage (Van Soest 1994; Licitra et al. 1996) whereas NDIN is loosely
263 bound to fiber, and as such a valuable source of N for the animal (Van Soest 1994). The latter will most likely

264 be a part of RUP, depending on passage rates through the rumen. This meant, as the NDIN had a bigger share of
265 CP in the RG it might have probably increased the level of RUP. The diet with pure RG was lower in AAT but
266 still within recommended levels for the measured yield level, as the PBV was far above recommended levels for
267 the respective diet. Thereby, forage RDP that possibly could have supported a higher rumen protein synthesis
268 was lost due to a shortage of rumen digestible carbohydrates. Requirements of AAT were met in all diets while
269 the PBV was calculated to be above minimum requirements in all diets. Still diets with a major proportion of
270 RG had a high PBV value, which resulted in a lower microbial protein synthesis than if the diet was denser in
271 energy. That means that there could have been produced more protein of microbial origin if the energy
272 concentration had been higher (Madsen et al. 1995).

273 Intake of macro- and micro minerals, except for Se, was all within recommended levels for lactating cows.
274 Selenium was provided at around 50% of the recommended 0.35 mg/kg DM (National Research Council 2001).
275 The concentrate offered about 90% of the Se, reflecting the common challenge in Norwegian soils with low
276 concentrations of Se. The K:Mg-ratio was slightly above recommendations. A too high ratio will make an
277 impact on the Mg absorption. In this case, it will not have a practical importance (Newton et al. 1972).

278 **Milk Production and Energy Utilization**

279 **Primiparous cows constituted 25% of the participating cows in the present study. Isolated, they had about 10%**
280 **lower milk yield than the pluriparous cows. However, primiparous and pluriparous cows showed throughout**
281 **similar diet effects on milk contents and conversion rates.** Calculations based on the Nordic feed evaluation
282 system, Norfor (Volden 2011), suggest that cows in the current experiment required 128.6 MJ NE_L daily to be
283 able to produce 29.1 kg ECM/day with a body weight of 640 kg. This was barely the case in D4. There were a
284 0.3 MJ NE_L deficit in D4, while D3 offered an energy balance and the other diets had a surplus of energy. A
285 positive energy balance on most diets is in line with the BW increase measured in average for all cows during
286 the course of the experiment.

287 Milk fat concentration from cows fed diets containing PG was higher than from cows fed RG diets, possibly due
288 to a significant higher dietary proportion of pdNDF (Beauchemin 1991) and WSC (Huhtanen et al. 2003). In
289 addition, the RG contained more clover. Clover, and especially red clover, is known to reduce milk fat
290 concentrations compared to grass species (Vanhatalo et al. 2009; Steinshamn 2010). The total milk fat yield
291 remained similar in all diets except D4, due to higher milk yields in D2 and D3.

292 Milk protein yield (MPY) showed a quadratic response favoring the mixed diets, however, with small
293 differences between diets. That makes this finding less significant and shows that higher dietary CP contents
294 will not necessarily increase MPY (Huhtanen and Hristov 2009). Because of the higher CP content in RG, the N
295 utilization efficiency decreased with increasing levels of RG. This shows that clover had a poorer conversion
296 rate than grasses due to higher CP levels (Dewhurst et al. 2003). Still, compared to other studies with conversion
297 rates ranging from 0.25 to 0.33 (Kuoppala et al. 2008; Eriksson et al. 2012; Randby et al. 2012), all the
298 conversion rates in the present study were good (0.30-0.32), due to the overall moderate CP intake.

299 The slightly lower milk yield observed when cows were offered D4 supports findings of other experiments with
300 lower yields in diets with RG (Peoples and Gordon 1989; Khalili et al. 2005; Kuoppala et al. 2010). Reports by
301 Randby (1992); Vanhatalo et al. (2009) and Steinshamn (2010) suggest that feeding dairy cows legume silages
302 results in a higher silage intake and thereby a higher milk production than pure grass silage. Dewhurst et al.
303 (2003) found that 50% clover did not differ from a pure clover diet on DMI and milk production. However, it
304 was also reported that pure clover diets may decrease milk yield compared to mixed grass-clover silages
305 (Halmemies-Beauchet-Filleau et al. 2014). Based on the mentioned findings the highest feed intake and milk
306 production should have occurred in D4. However, the effect of growth, PG versus RG, with its effect on fiber
307 content and quality and thereby energy value are obviously more important; thus decreasing feed intake and
308 milk production in D4 compared to the diets containing PG. Kuoppala et al. (2008) showed a clear correlation of
309 ME intake and ECM yield. It is obvious that higher intakes ME will increase the milk yield. On the other hand,
310 the RG contributed to a better utilization of pdNDF into milk.

311 Organic milk production has limitation on the use of concentrate (Counc. of the Eur. Union 2007). Similar diets
312 to those used in this experiment, could comprise less concentrate, which would imply even lower dietary CP
313 concentrations in the total diet. Calculations suggest that there would be expectations of too low PBV in D1, if
314 the concentrate was fed at less than 4 kg a daily (Madsen et al. 1995). This means that there would be too little
315 RDP for the microbes to synthesize into microbial protein. On the other hand, the NE_L would be higher than the
316 cow could utilize for milk synthesis and, as such, the excess of energy could be partitioned into body reserves.
317 All three diets comprising RG came out with a positive PBV even in a diet theoretically without concentrate.
318 This suggests that a diet based on a mix of PG and RG would be more optimal than the pure PG or RG diets, if
319 concentrate levels were below 4 kg daily, or if the actual concentrate contained less CP than in the current study.
320 With 4 kg concentrates, or less, the hypothesis on a dietary optimal mix of PG and RG would probably have

321 been met, but with 8 kg concentrates, as in the current study, the hypothesis was rejected because pure PG was
322 equally good as the mixed diets.

323 **CONCLUSION**

324 The protein supply was not a key factor for the milk yield response. Sufficient energy supply with the PG was
325 more important. The overall observed nitrogen use efficiency was good but significantly better with PG in the
326 diet. As long as there were at least 33% PG in the diet, the ECM did not decrease. Milk yield tended to be higher
327 in mixed diets, but lower fat content compared to the diet with pure PG took the overall advantage away from
328 the mixed diets. Still in the practical use for farmers, who have to use all available resources at the farm, the
329 mixed diets will provide a higher milk production and better economy than feeding each harvest separate.

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337 **REFERENCES**

- 338 AOAC (1984) AOAC Official Method 7.074 Fiber(Acid Detergent) and Lignin in Animal Feed In: Williams, S.
339 (ed.) Official Methods of Analysis of AOAC, 14th edn, pp. 162-163. Arlington, VA, USA: Assoc. of Off. Anal.
340 Chemists. In., 14 edn.,
- 341 Beauchemin K (1991) Effects of dietary neutral detergent fiber concentration and alfalfa hay quality on
342 chewing, rumen function, and milk production of dairy cows J of Dairy Sci 74:3140-3151
- 343 Bertilsson J, Murphy M (2003) Effects of feeding clover silages on feed intake, milk production and digestion in
344 dairy cows Grass and Forage Sci 58:309-322 doi:10.1046/j.1365-2494.2003.00383.x

345 Byström S, Jonsson S, Martinsson K (2002) Organic versus conventional dairy farming – studies from the
346 Öjebyn Project. In: The UK Organic Research Conference, Aberystwyth, UK, 26-28 March 2002. .p 179-184

347 Counc. of the Eur. Union (2007) Council Regulation (EC) No. 834/2007 of 28 June 2007 on organic production
348 and labelling of organic products and repealing Regulation (EEC) No 2092/91. Off. J. Eur. Communities 189,
349 20.7.2007, 1-23.

350 Dewhurst R, Fisher W, Tweed J, Wilkins R (2003) Comparison of grass and legume silages for milk production.
351 1. Production responses with different levels of concentrate J of Dairy Sci 86:2598-2611

352 Eriksson T, Norell L, Nilsson-Linde N (2012) Nitrogen metabolism and milk production in dairy cows fed
353 semi-restricted amounts of ryegrass–legume silage with birdsfoot trefoil (*Lotus corniculatus* L.) or white clover
354 (*Trifolium repens* L.) Grass and Forage Sci 67:546-558 doi:10.1111/j.1365-2494.2012.00882.x

355 Govasmark E, Steen A, Bakken A, Steinshamn H, Strøm T, Hansen S (2005) Factors affecting the concentration
356 of Zn, Fe and Mn in herbage from organic farms and in relation to dietary requirements of ruminants Acta Agri
357 Scand, Section B — Soil & Plant Sci 55:131-142 doi:10.1080/09064710510008586

358 Halmemies-Beauchet-Filleau A, Vanhatalo A, Toivonen V, Heikkilä T, Lee MRF, Shingfield KJ (2014) Effect
359 of replacing grass silage with red clover silage on nutrient digestion, nitrogen metabolism, and milk fat
360 composition in lactating cows fed diets containing a 60:40 forage-to-concentrate ratio J of Dairy Sci 97:3761-
361 3776 doi:http://dx.doi.org/10.3168/jds.2013-7358

362 Huhtanen P, Hristov AN (2009) A meta-analysis of the effects of dietary protein concentration and degradability
363 on milk protein yield and milk N efficiency in dairy cows J Dairy Sci 92:3222-3232 doi:10.3168/jds.2008-1352

364 Huhtanen P, Nousiainen J, Rinne M (2006) Recent developments in forage evaluation with special reference to
365 practical applications Agri and Food Sci 15:31

366 Huhtanen P, Nousiainen JI, Khalili H, Jaakkola S, Heikkilä T (2003) Relationships between silage fermentation
367 characteristics and milk production parameters: analyses of literature data Livest Prod Sci 81:57-73

368 Huhtanen P, Rinne M, Nousiainen J (2007) Evaluation of the factors affecting silage intake of dairy cows: a
369 revision of the relative silage dry-matter intake index Animal 1:13

370 Khalili H, Huhtanen P (1991) Sucrose supplements in cattle given grass silage-based diet. 2. Digestion of cell
371 wall carbohydrates *Animal Feed Sci and Tech* 33:263-273 doi:[http://dx.doi.org/10.1016/0377-8401\(91\)90065-Z](http://dx.doi.org/10.1016/0377-8401(91)90065-Z)

372 Khalili H, Sairanen A, Nousiainen J, Huhtanen P (2005) Effects of silage made from primary or regrowth grass
373 and protein supplementation on dairy cow performance *Livest Prod Sci* 96:269-278

374 Kuoppala K, Ahvenjarvi S, Rinne M, Vanhatalo A (2009) Effects of feeding grass or red clover silage cut at two
375 maturity stages in dairy cows. 2. Dry matter intake and cell wall digestion kinetics *J Dairy Sci* 92:5634-5644
376 doi:10.3168/jds.2009-2250

377 Kuoppala K, Rinne M, Ahvenjarvi S, Nousiainen J, Huhtanen P (2010) The effect of harvesting strategy of grass
378 silage on digestion and nutrient supply in dairy cows *J Dairy Sci* 93:3253-3263 doi:10.3168/jds.2009-3013

379 Kuoppala K, Rinne M, Nousiainen J, Huhtanen P (2008) The effect of cutting time of grass silage in primary
380 growth and regrowth and the interactions between silage quality and concentrate level on milk production of
381 dairy cows *Livest Sci* 116:171-182 doi:<http://dx.doi.org/10.1016/j.livsci.2007.10.002>

382 Leiber F (2014) Resigning protein concentrates in dairy cattle nutrition: a problem or a chance? *Org Agri* 4:269-
383 273

384 Licitra G, Hernandez TM, VanSoest PJ (1996) Standardization of procedures for nitrogen fractionation of
385 ruminant feeds *Anim Feed Sci and Tech* 57:347-358 doi:Doi 10.1016/0377-8401(95)00837-3

386 Madsen J, Hvelplund T, Weisbjerg MR, Bertilsson J, Olsson I, Spörndly R, Harstad OM, Volden H, Tuori M,
387 Varvikko T, Huhtanen P, Olafsson BL (1995) The AAT/PBV protein evaluation system for ruminants - a
388 revision *Nor J of Agri Sci Suppl*:35

389 National Research Council (2001) *Nutrient Requirements of Dairy Cattle: Seventh Revised Edition, 2001*. The
390 Natl. Acad. Press, Washington, DC

391 Newton G, Fontenot J, Tucker R, Polan C (1972) Effects of high dietary potassium intake on the metabolism of
392 magnesium by sheep *J of Anim Sci* 35:440-445

393 NorFor (2007a) Norfor in sacco standard, Nordic Feed Evaluation System. Accessed 21.11 2014

394 Norfor (2007b) Norfor Method for Determination of Dry Matter vol 2015, 21 September 2007 edn.,

395 Nousiainen J (2004) Development of tools for the nutritional management of dairy cows on silage-based diets.
396 PhD Thesis. Univ. of Helsinki

397 Peoples A, Gordon F (1989) The influence of wilting and season of silage harvest and the fat and protein
398 concentration of the supplement on milk production and food utilization by lactating cattle *Anim Prod* 48:305-
399 317

400 Randby Å, Nørgaard P, Weisbjerg M (2010) Effect of increasing plant maturity in timothy-dominated grass
401 silage on the performance of growing/finishing Norwegian Red bulls *Grass and Forage Sci* 65:273-286

402 Randby Å, Weisbjerg M, Nørgaard P, Heringstad B (2012) Early lactation feed intake and milk yield responses
403 of dairy cows offered grass silages harvested at early maturity stages *J of Dairy Sci* 95:14

404 Randby ÅT (1992) Grass-clover silage for dairy cows. In: 14th Gen. Meet. of Eur. Grassl. Fed., Lahti, Finland,
405 8-11 June 1992 1992. pp 272-275

406 SAS Institute Inc (2011) SAS/STAT Software, Version 9.3. doi:citeulike-article-id:11105873

407 Schwab C, Huhtanen P, Hunt C, Hvelplund T (2005) Nitrogen Requirements of Cattle. In: Pfeffer E, Hristov
408 AN (eds) Nitrogen and Phosphorus Nutrition of Cattle: Reducing the Environmental Impact of Cattle
409 Operations, vol 1. CABI Publ., Cambridge, MA, USA, pp 13-70

410 Spörndly R (2003) Fodertabeller för idisslare. Rep. 257. Dep. of Anim. Nutr. Manag. Swedish Univ. Agric. Sci,
411 Uppsala, Sweden

412 Steinshamn H (2001) Effects of Cattle Slurry on the Growth Potential and Clover Proportion of Organically
413 Managed Grass-Clover Leys *Acta Agri Scand, Sect B — Soil & Plant Sci* 51:12 doi:10.1080/09064710127615

414 Steinshamn H (2010) Effect of forage legumes on feed intake, milk production and milk quality—a review *Anim*
415 *Sci Pap and Rep* 28:12

416 Steinshamn H, Thuen E (2008) White or red clover-grass silage in organic dairy milk production: Grassland
417 productivity and milk production responses with different levels of concentrate *Livest Sci* 119:14
418 doi:<http://dx.doi.org/10.1016/j.livsci.2008.04.004>

419 Stensig T, Weisbjerg MR, Hvelplund T (1998) Digestion and passage kinetics of fibre in dairy cows as affected
420 by the proportion of wheat starch or sucrose in the diet *Acta Agri Scand, Sect A — Anim Sci* 48:12
421 doi:10.1080/09064709809362413

422 TINE Rådgiving (2012) TINE SA (The Nor. Dairy Ass.), Ås, Norway.

423 Van Es AJH (1978) Feed evaluation for ruminants. I. The systems in use from May 1977-onwards in The
424 Netherlands *Livest Prod Sci* 5:15 doi:http://dx.doi.org/10.1016/0301-6226(78)90029-5

425 Van Soest PJ (1994) *Nutritional ecology of the ruminant*. 2. edn. Comstock Publ. Assoc., Ithaca, N.Y.

426 Van Soest PJ, Robertson JB, Lewis BA (1991) Methods for Dietary Fiber, Neutral Detergent Fiber, and
427 Nonstarch Polysaccharides in Relation to Animal Nutrition *J of Dairy Sci* 74:15
428 doi:http://dx.doi.org/10.3168/jds.S0022-0302(91)78551-2

429 Vanhatalo A, Kuoppala K, Ahvenjärvi S, Rinne M (2009) Effects of feeding grass or red clover silage cut at two
430 maturity stages in dairy cows. 1. Nitrogen metabolism and supply of amino acids *J of Dairy Sci* 92:14
431 doi:10.3168/jds.2009-2249

432 Vanhatalo A et al. Effects of harvest time of red clover silage on milk production and composition. In:
433 *Biodivers. and Anim. Feed: Future Chall. for Grassl. Prod. Proceedings of the 22nd Gen. Meet. of the Eur.*
434 *Grassl. Fed., Uppsala, Sweden, 9-12 June 2008., 2008. Swedish Univ. of Agri. Sci., pp 391-393*

435 Volden H (2011) *NorFor Nordic feed evaluation system Norfor - the Nordic Feed Evaluation System*, vol 130.
436 Wageningen Acad. Publ., Wageningen, The Netherlands. doi:Book_Doi 10.3920/978-90-8686-718-9

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Table 1.
The chemical composition of organic grass-clover silages and concentrate offered dairy cows.

Item	Primary growth		Regrowth		Concentrate	
	Mean	SE ^a	Mean	SE ^a	Mean	SE ^a
Dry matter, g/kg	369	0.49	336	0.38	876	3.9
pH	4.4	0.01	4.3	0.01		
<i>g/kg DM</i>						
Organic matter	932	0.47	915	0.48	922	0.69
Crude protein	116	1.0	138	0.90	165	0.25
Water soluble carbohydrates	39	2.0	26	0.64	64	0.86
Starch ^b					372	2.18
NDF ^c	501	3.4	473	2.0	154	2.8
Indigestible NDF ^c	63	1.2	97	2.6	56	1.4
Potentially digestible NDF ^c	439	3.2	377	3.7	98	3.4
Non fiber carbohydrates	246	4.0	218	2.3	566	3.0
ADF ^d	294	2.1	324	1.1	87	1.2
Acid detergent lignin	39	2.6	38	0.52	33	3.4
Fat	28	0.55	33	0.66	54	0.63
NDIN ^e	1.98	0.04	3.38	0.07	1.99	0.04
ADIN ^f	0.92	0.04	1.12	0.06	0.64	0.04
Lactic acid	13.0	0.50	17.5	0.62		
Formic acid	3.71	0.18	3.83	0.20		
Acetic acid	2.55	0.09	3.34	0.08		
Propionic acid	0.84	0.04	0.92	0.05		
Burtyric acid	0.00	0.00	0.00	0.00		
Ethanol	4.58	0.34	1.16	0.08		
Ammonia N (g/kg N)	11.5	0.94	14.0	0.81		
<i>Vitamins and Minerals</i>						
Vitamin A, IU/kg					5000	
Vitamin D, IU/kg					2000	
Vitamin E, mg/kg					40	
Ca, g/kg DM	5	0.13	7	0.20	11	0.28
P, g/kg DM	3	0.05	3	0.05	7	0.10
K, g/kg DM	20	0.39	21	0.22	10	0.07
Mg, g/kg DM	2	0.04	2	0.05	7	0.22
Na, g/kg DM	1	0.07	2	0.10	5	0.12
Cu, mg/kg DM	12	0.94	12	1.00	24	0.44
Zn, mg/kg DM	28	1.78	30	1.91	130	1.70
Mn mg/kg DM	41	2.37	52	2.56	81	15.2
Se, mg/kg DM	0.02	0.007	0.01	0.002	0.45	0.054

440 ^a Standard error

441 ^b Starch inclusive glucose.

442 ^c Neutral detergent fiber.

443 ^d Acid detergent fiber exclusive of ash.

444 ^e Neutral detergent insoluble nitrogen.

445 ^f Acid detergent insoluble nitrogen.

446 Table 2.

447 Apparent digestibility of nutrients on wethers and calculated feed values of primary growth (PG), regrowth
448 (RG) and concentrate.

Item	PG	RG	SEM ^a	P-value	Concentrate
<i>Sheep digestibility</i>					
Dry matter	0.72	0.68	0.01	0.01	0.79
Organic matter	0.75	0.71	0.00	0.01	0.83
Crude protein	0.61	0.65	0.01	0.03	0.76
NDF ^b	0.73	0.68	0.01	0.03	0.46
Fat	0.86	0.86	0.01	0.34	0.85
<i>Feed values</i>					
ME ^c , MJ/kg DM	10.3	9.7	0.67	<0.01	11.6
NE _L ^d , MJ/kg DM	6.02	5.77	0.31	<0.01	6.61
AAT ^e , g/kg DM	72.0	67.8	0.20	<0.01	71.2
PBV ^f , g/kg DM	-7.80	19.9	0.81	<0.01	36.3
D-value ^g	698	649	0.36	<0.01	728

449 ^a Standard error of the mean

450 ^b Neutral detergent fiber.

451 ^c Metabolizable Energy.

452 ^d NEL, calculated according to Van Es (1978).

453 ^e Amino acids absorbed in the intestine (Madsen et al., 1995).

454 ^f Protein balance in the rumen (Madsen et al., 1995).

455 ^g Digestible organic matter in dry matter.

456

457 Table 3.

458 Effect of diets with regrowth replacing primary growth in the DM ratio 0, 0.33, 0.67 and 1 (treatments D1, D2,
459 D3, and D4, respectively) on daily basis feed intake with orthogonal contrasts

Item	Diet				SEM	Orthogonal contrasts	
	D1	D2	D3	D4		Linear	Quadratic
<i>DM intake, kg/d</i>							
Grass-clover silage	15.1	15.0	14.4	14.0	0.49	<0.01	0.16
Concentrate	6.8	6.8	6.8	6.8	0.01		
Total	21.9	21.8	21.2	20.7	0.49	<0.01	0.14
<i>Intake, kg/d</i>							
Organic matter	20.3	19.9	19.6	19.0	0.49	<0.01	0.55
NDF ^a	8.60	8.44	7.92	7.67	0.248	<0.01	0.58
Indigestible NDF ^a	1.32	1.46	1.56	1.71	0.044	<0.01	0.74
pdNDF ^b	7.28	6.99	6.36	5.96	0.209	<0.01	0.44
ADF ^c	5.02	5.13	5.03	5.12	0.154	0.45	0.84
Acid detergent lignin	0.80	0.81	0.76	0.76	0.022	0.02	0.81
Water soluble carbohydrates	1.04	1.03	0.98	0.96	0.020	<0.01	0.85
Non fiber carbohydrates	7.29	7.31	7.13	7.05	0.125	<0.01	0.29
Starch ^d	2.39	2.39	2.39	2.38	0.004	0.04	0.18
Fat	0.85	0.87	0.87	0.89	0.020	<0.01	0.97
Crude protein	2.86	2.95	2.96	3.04	0.064	<0.01	0.84
<i>Intake, g/d</i>							
NDIN ^e	57.0	62.1	65.5	71.8	1.68	<0.01	0.48
ADIN ^f	21.7	23.4	24.4	26.4	0.63	<0.01	0.74
Ammonia-N	3.14	3.67	3.89	4.37	0.158	<0.01	0.83
AAT ^g	1565	1543	1469	1428	36.1	<0.01	0.41
PBV ^h	129	252	384	528	7.3	<0.01	0.13
MJ ME/d ⁱ	239	235	224	217	5.1	<0.01	0.38
MJ NE _L /d ^j	141	139	132	128	3.0	<0.01	0.36
DMI/BW ^k	0.0346	0.0347	0.0337	0.0331	0.0010	<0.01	0.21

460 ^a Neutral detergent fiber.

461 ^b Potentially degradable NDF.

462 ^c Acid detergent fiber exclusive of ash.

463 ^d Starch inclusive of glucose.

464 ^e Neutral detergent insoluble nitrogen.

465 ^f Acid detergent insoluble nitrogen.

466 ^g Amino acids absorbed in the intestine (Madsen et al., 1995).

467 ^h Protein balance in the rumen (Madsen et al., 1995).

468 ⁱ NE_L, calculated according to Van Es (1978).

469 ^j ME, calculated according to Van Es (1978).

470 ^k DM intake, kg/kg BW.

471 Table 4.

472 Effect of diets with regrowth replacing primary growth in the DM ratio 0, 0.33, 0.67 and 1 (treatments D1, D2,

473 D3, and D4, respectively) on milk production and efficiency measures with orthogonal contrasts.

Item	Diet				SEM ^a	Orthogonal contrasts	
	D1	D2	D3	D4		Linear	Quadratic
Milk <i>kg/d</i>	30.2	30.2	30.6	29.9	1.16	0.50	0.06
ECM ^b <i>kg/d</i>	30.0	29.9	29.9	29.1	1.20	<0.01	0.11
<i>Milk composition</i>							
Fat, %	4.07	4.02	3.93	3.88	0.143	<0.01	0.99
Protein, %	3.12	3.13	3.10	3.11	0.046	0.27	0.87
Lactose, %	4.79	4.74	4.79	4.79	0.049	0.76	0.36
Urea, <i>mmol/L</i>	2.25	2.67	3.01	3.70	0.091	<0.01	0.01
Free fatty acids, <i>mEq/L</i>	0.44	0.34	0.34	0.37	0.059	0.20	0.08
<i>Yield of milk components, kg/d</i>							
Fat	1.22	1.21	1.20	1.16	0.064	<0.01	0.19
Protein	0.936	0.941	0.942	0.922	0.0293	0.14	0.04
Lactose	1.44	1.43	1.46	1.43	0.047	0.71	0.39
<i>Efficiency measures</i>							
ECM/MJ ME	0.126	0.127	0.134	0.134	0.0034	<0.01	0.55
ECM/Dry matter intake	1.37	1.37	1.41	1.40	0.036	0.07	0.78
Milk N/Feed N ^c	0.320	0.312	0.313	0.298	0.0067	<0.01	0.30

474 ^a SEM = standard error of the means.

475 ^b ECM = Energy corrected milk, ME = Metabolizable energy.

476 ^c Proportion of nitrogen in total ingested feed converted into nitrogen in milk.

477