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1 **Title:**

2 Effects of grazing adaptation on herbage intake, milk production, and body weight
3 change in lactating dairy cows after turning out to pasture in early spring

4

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1 **ABSTRACT**

2 The present study investigated the effects of adaptation to grazing in early spring on
3 the herbage intake, milk production, and body weight changes in lactating dairy cows.
4 The experiment included 12 Holstein lactating cows during early spring. Six cows
5 were allowed to gradually adapt to grazing (ADP) over 10 days. They were allowed
6 to graze on a pasture for 4 h during the first week and for 19 h during the remaining
7 3 days (adaptation period). During the 10-day period, the remaining six cows were
8 housed in a barn (CON). Cows in both groups received adequate silage, hay, and
9 concentrate during the adaptation period. Subsequently, cows in both groups were
10 allowed to graze together for 19 h for 4 weeks (experimental period). No cow
11 received supplements during the experimental period. In the first week of the
12 experimental period, the ADP cows showed a higher herbage intake than the CON
13 cows. During the subsequent weeks, there were no differences in herbage intake
14 between the two groups. At the start of the experimental period, milk production and
15 body weight of the ADP cows were higher than those of the CON cows, and this
16 difference lasted until the end of the experiment.

17

18 Key words:

19 Body weight change, Early spring, Grazing adaptation, Herbage intake, Milk
20 production

21

1 **1 INTRODUCTION**

2 Feeding management incorporating grazing for dairy cows is commonly used
3 worldwide. In temperate regions, particularly areas experiencing snowfall, grazing
4 cows are exposed to dramatic changes in feeding management between indoor
5 feeding and grazing during early spring. When cows were suddenly switched to
6 grazing from indoor feeding, dry matter intake (DMI), body weight (BW), and milk
7 production decreased temporarily (Charmley, Jannasch & Boyd, 2003; Khanal,
8 Dhiman & Boman, 2008; Mitani et al., 2020).

9 In a previous study on non-lactating dairy cows (Mitani et al., 2020), short-
10 term grazing adaptation before turning out to 1-day grazing was effective in
11 stabilizing herbage intake, ruminal fermentation parameters, and blood metabolites
12 compared with turning out to 1-day grazing without grazing adaptation, which
13 drastically decreased herbage intake and BW in the first week. Thus, to reduce
14 production loss, adaptation from indoor feeding to grazing in early spring may be
15 desirable for cows for avoiding decrease in herbage DMI.

16 However, the effectiveness of grazing adaptation is inconsistent with
17 production responses. In a series of studies in Germany, even though cows were
18 gradually adapted to grazing before turning out to 1-day grazing, their milk yield did
19 or did not recover after 3 weeks to the level similar to that of cows fed total mixed
20 ration (TMR) (Schären et al., 2016a; Hartwiger et al., 2018a). Once milk production
21 and BW decreased, several weeks were required to recover their levels (Jørgensen et
22 al., 2016). Therefore, herbage DMI immediately after turning out to 1-day grazing
23 may mediate grazing adaptability. To the best of our knowledge, no study has
24 reported the changes in herbage DMI, milk production, and BW in lactating cows in
25 response to grazing adaptation.

1 Therefore, in the present study, we investigated the effects of gradual
2 grazing adaptation prior to turning out from indoor feeding to grazing during early
3 spring on the herbage DMI, milk production, and BW of lactating dairy cows.

4 5 **2 MATERIALS AND METHODS**

6 **2.1 Animals and experimental design**

7 The methods adopted for the feeding management of cows in this study were
8 approved by the Animal Care and Welfare Committee of Hokkaido University (No.
9 15-0124).

10 The study was conducted between April 24 and May 31, 2019, at the
11 Experimental Farm of the Field Science Center for Northern Biosphere, Hokkaido
12 University (Sapporo, Japan). Twelve Holstein lactating cows with grazing experience
13 were included in the experiment. At the start of the experiment, the average (\pm
14 standard deviation) daily milk production, BW, days in milk, and parity were $29.8 \pm$
15 5.8 kg, 635 ± 46 kg, 161 ± 114 days, and 2.8 ± 1.2 , respectively. The cows were
16 divided into two groups, which did not differ in terms of average daily milk
17 production, days in milk, and parity.

18 The cows were housed in a barn prior to the experiment and received 10 kg
19 fresh matter (FM)/day of commercial formulated concentrate, 4 kg FM/day of hay,
20 and 20 kg FM/day of mixed silage (corn silage + hay, 91:9 on FM basis). The cows
21 in the period were fed enough feed to meet the energy requirement for 30 kg/day of
22 milk production according to Japanese Feeding Standard (National Agriculture and
23 Food Research Organization, 2017). Cows in one group (ADP) were subjected to two
24 phases of grazing adaptation (adaptation period): (1) from April 24 to 30, the cows
25 were grazed for 4 h (10:00 to 14:00 h) and supplied 8–10 kg FM/day of commercial

1 formulated concentrate, 3 kg FM/day of hay, and 20 kg FM/day of mixed silage and
2 (2) from May 1 to 3, the cows were grazed for 19 h (10:00 to 14:00 and 16:30 to 7:30
3 h) and supplied 5 kg FM/day of commercial formulated concentrate and 10 kg
4 FM/day of mixed silage. During this period, the remaining cows (CON) were housed
5 in the barn and supplied the same amount and composition of feeds prior to the
6 experiment.

7 Subsequently, cows in both groups were turned out to 1-day grazing together
8 on a pasture from May 4 to 31 (experimental period). No cow received supplementary
9 forage or concentrate during this period. The pasture (6.28 ha) was divided into four
10 paddocks with electric polylines for rotational grazing management with 2- or 4-day
11 intervals on each paddock. The pasture primarily constituted perennial ryegrass
12 (*Lolium perenne* L.) and white clover (*Trifolium repens* L.). All cows were allowed
13 ad libitum access to water on the pasture and mineral blocks in the barn.

14

15 **2.2 Pasture and grazing parameters**

16 Compressed herbage height was measured at 200 points in each paddock using a
17 rising plate meter immediately before each grazing rotation. Simultaneously, herbage
18 mass and compressed herbage height were measured at four points in paddock in a
19 quadrat measuring 0.5×0.5 m, and then a calibration formula was determined. Using
20 the calibration formula, herbage mass was estimated from the compressed herbage
21 height. The grass surface height was also measured at four points in paddock in a
22 quadrat. During the experimental period, herbage mass and grass sward height
23 gradually increased and were sufficient for grazing (mean \pm SD: 2.13 ± 0.14
24 t·DM·ha⁻¹ and 15.0 ± 2.7 cm, respectively).

25 From each grazed paddock, herbage samples were collected every day using

1 the hand-plucking method, mimicking the grazing behavior, at 08:00 and 16:00 h.
2 Herbage samples from each paddock were pooled and stored at -20°C . Samples of
3 commercial concentrate, hay, and mixed silage supplied during the adaptation period
4 were also collected every day, pooled for each week, and stored at -20°C . The
5 samples were lyophilized and passed through a 1-mm screen. The feed samples were
6 analyzed for DM, organic matter (OM), and crude protein (CP) content using the
7 methods recommended by the Association of Official Analytical Chemists (AOAC,
8 1990). Ash-free neutral detergent fiber (NDF) was measured according to the method
9 described by Van Soest et al. (1991). Water-soluble carbohydrate (WSC) content was
10 determined using the anthrone method (Yemm & Willis, 1954).

11 Herbage DM intake during the adaptation and experimental periods was
12 measured using the double indicator method, with lanthanum (La) as the external
13 marker by soaking sugar beet pulp in $\text{LaCl}_3\cdot 7\text{H}_2\text{O}$ solution. The cows received 50 g
14 of the La marker with 100 g of concentrate and 50 g of a vitamin–mineral mix at 08:00
15 and 15:00 h in the barn. The feces in the rectum were collected at 08:00 and 15:00 h
16 every day and pooled for days 1–3 and 4–7 of the adaptation period and days 1–3, 4–
17 7, 8–14, 15–21, and 22–28 days of the experimental period. The concentration of La
18 and C33 alkanes in the herbage, supplied feed, and fecal samples was determined as
19 described previously (Mitani et al., 2020). Herbage DM intake was estimated using
20 a previously described method (Mitani et al., 2020). Total tract apparent OM
21 digestibility was calculated based on the total OM intake and fecal OM.

22

23 **2.3 Milk production, BW, ruminal fermentation, and blood metabolites**

24 All cows were milked at 09:00 and 15:30 h daily throughout the experiment. Milk
25 production was recorded at each milking during the experiment (MMD500, Orion

1 Kikai Co. Ltd., Nagano, Japan). A milk sample was also collected at each milking
2 and pooled from consecutive milkings during the experiment. An antiseptic (2-
3 bromo-2-nitro-1,3-propanediol) was added to the pooled milk samples from each cow,
4 and the samples were sent to the Laboratory of Hokkaido Dairy Milk Recording and
5 Testing Association. Milk fat, protein, lactose, and solid-not-fat concentration was
6 measured using Fourier-transform infrared spectroscopy (MilkoScan FT+; Foss
7 Electric, Hillerød, Denmark).

8 The BW of each cow was measured at 14:00 h on days 4 and 7 during the
9 adaptation period and on days 2, 4, 7, 14, 21, and 28 during the experimental period
10 using an electronic scale (Orion kikai Co. Ltd., Nagano, Japan). Following BW
11 measurements, rumen fluid and blood samples were collected on the same days
12 during the adaptation and experimental period. The rumen fluid was collected using
13 stomach tubes, and blood samples were collected from the jugular vein using vacuum
14 blood tubes containing heparin.

15 The ruminal fluid samples were centrifuged (at 3,000 rpm and 4°C for 20
16 min), and the supernatant was stored at -80°C until further analysis. Volatile fatty
17 acid (VFA) concentration was determined using gas chromatography (GC-2010;
18 Shimadzu, Kyoto, Japan) according to the method described by Ueda et al. (2016).

19 The blood samples were centrifuged (at 3,000 rpm and 4°C for 20 min), and
20 the plasma was stored at -80°C until further analysis. Plasma urea concentration
21 (PUN) was determined using the urease-indophenol method (Wetherburn, 1967).
22 Plasma non-esterified fatty acid (NEFA) and blood glucose concentration was
23 determined using commercial kits (NEFA C-TEST Wako and Glucose CII-TEST
24 Wako, respectively; Wako Pure Chemicals, Osaka, Japan).

25

1 **2.4 Statistical analysis**

2 Statistical analysis was performed using JMP Pro 14.3 (SAS Institute Inc., Cary, NC,
3 USA). The data of the experimental period (days after turning out to 1-day grazing)
4 were analyzed with a mixed model for repeated measures using the Fit Model
5 platform in JMP. The model included treatment (CON or ADP), days of sampling,
6 and the interaction between these two parameters as the fixed effects; cows as the
7 random effect; and days of sampling as the repeated effect, adjusting the first-order
8 autoregressive structure as the correlation structure. Results of milk production and
9 BW data were also corrected by covariance data which were collected before starting
10 the experiment. If the probability value was less than 0.05, the results were
11 considered significant. When the probability value was less than 0.10, the results
12 were considered as showing a tendency. When the interaction effect was significant
13 ($P < 0.05$), the difference between treatments for each day was tested. The results
14 were presented as the least square means and standard error of the mean.

15

16 **3 RESULTS**

17 **3.1 Feed chemical compositions and feed intake**

18 The chemical composition of the feed supplied during the adaptation period and of
19 herbage during the adaptation and experimental period is shown in Table 1. The CP
20 content of herbage during the adaptation period was higher than that during the
21 experimental period, although the value remained at 17% DM throughout the
22 experiment. The NDF content of herbage increased from 24.9% during the adaptation
23 period to 34.8% DM at the end of the experiment. The WSC content of herbage was
24 the highest (26.2% DM) at the start of the experiment, but it gradually decreased to
25 16.3% DM at the end of the experiment. Corn silage and hay had a lower CP content

[Table 1]

1 and higher NDF content than herbage. The chemical compositions of the corn silage
2 and hay were within the standard ranges (National Agriculture and Food Research
3 Organization, 2009).

4 Changes in herbage DMI and total DMI are shown in Figure 1. During the
5 adaptation period, the herbage DMI of the ADP cows gradually increased, but there
6 was no difference in total DMI between the ADP and CON cows. During the
7 experimental period, the change in herbage DMI was significantly different between **[Figure 1]**
8 the ADP and CON cows ($P < 0.01$). The ADP cows consumed more than 15 kg
9 DM/day of herbage throughout the experimental period. The herbage DMI of the
10 CON cows was half that of the ADP cows at the start of the experimental period, but
11 it gradually increased until the end of the experimental period. The CON cows
12 required a week to reach the herbage DMI similar to that of the ADP cows.

13 The changes in total OM intake, total tract apparent OM digestibility, and
14 digestible OM intake are shown in Table 2. During the adaptation period, the OM
15 digestibility of the ADP cows tended to be lower than that of the CON cows ($P =$
16 0.09). At the start of the experimental period (week 1-E), OM digestibility during the
17 adaptation period was significantly decreased in the CON cows but increased in the
18 ADP cows. After week 2 of the experimental period, the OM digestibility of both
19 ADP and CON cows was similar and remained stable until the end of the experiment.
20 The digestible OM intake of the ADP cows was higher than that of the CON cows at
21 week 1 and week 3 of the experimental period. **[Table 2]**

22

23 **3.2 Ruminal fermentation and blood metabolites**

24 The changes in ruminal total VFA concentration and molar proportions of each VFA
25 are shown in Table 3. The changes in ruminal total VFA concentration tended to differ

[Table 3]

1 between the ADP and CON cows ($P = 0.08$). The total VFA concentration of the ADP
2 cows tended to be lower than that of the CON cows on days 2, 4, and 7 of the
3 experimental period, but there was no difference after week 2 of the experimental
4 period.

5 There was no difference in the proportions of acetate, propionate, and
6 butyrate between the ADP and CON cows. The changes in acetate proportion differed
7 between the two groups ($P = 0.02$), and the changes in propionate proportion tended
8 to differ ($P = 0.07$). At the start of the experimental period, the acetate proportions
9 during the adaptation period decreased but the propionate and butyrate proportions
10 during the adaptation period increased in the CON cows. However, the proportions
11 of different VFAs during the adaptation period remained unchanged in the ADP cows
12 at the start of the experimental period. In both groups, the acetate proportions
13 gradually increased but the propionate and butyrate proportions gradually decreased
14 throughout the experimental period.

15 The changes in PUN, blood glucose, and plasma NEFA concentration are
16 shown in Table 4. During the experimental period, the changes in PUN concentration
17 significantly differed between the groups ($P < 0.01$). In days 2 and 4 of the
18 experimental period, the PUN concentration of the ADP cows was lower than that of
19 the CON cows; however, there was no difference in PUN concentration between the
20 two groups after days 7 until the end of the experiment. Likewise, the changes in
21 blood glucose concentration differed between the two groups ($P < 0.02$). The blood
22 glucose concentration of the ADP cows was higher than that of the CON cows at days
23 2 and 4 of the experimental period, but there was no difference in blood glucose
24 concentration between the two groups after days 7 until the end of the experiment.
25 The mean of and changes in NEFA concentration did not differ between the two

[Table 4]

1 groups. However, the NEFA concentration of the ADP cows was numerically lower
2 than that of the CON cows at days 2 and days 4 of the experimental period.

3

4 **3.3 Milk production and BW change**

5 Changes in milk production and composition are shown in Table 5. During the
6 adaptation period, daily milk production of the ADP cows was numerically greater
7 than that of the CON cows ($P = 0.11$), although there was no difference before the
8 start of the experiment (basal data). After turning out from indoor feeding to 1-day
9 grazing, the daily milk yield of the ADP cows was maintained, while that of the CON
10 cows was decreased by 2kg. The difference in the daily milk yield between the groups
11 at the start of the experimental period was maintained until the end of experiment (P
12 = 0.08). **[Table 5]**

13 The milk fat concentration did not differ between the two groups. The milk
14 fat concentration in both groups gradually decreased until the end of the experiment.
15 The lactose concentration did not differ between the two groups. The change in
16 lactose concentration also did not differ between the two groups. During the
17 adaptation period, the milk protein and solid-not-fat concentrations of the ADP cows
18 were higher than those of the CON cows ($P = 0.03$). After turning out from indoor
19 feeding to 1-day grazing, the protein and solid-not-fat concentrations of the ADP
20 cows were maintained, although those of the CON cows decreased. The difference
21 in the protein and solid-not-fat concentrations between the groups at the start of the
22 experimental period was maintained until the end of experiment ($P = 0.02$ and 0.08 ,
23 respectively).

24 The change of 4% fat-corrected milk (FCM) yield in both groups was similar
25 as the change of daily milk yield. The difference in FCM yield between the groups

1 was greater than the difference in daily milk yield ($P = 0.04$). After turning out from
2 indoor feeding to 1-day grazing, the FCM yield in both groups gradually increased.
3 However, the difference in FCM yield between the groups at the start of the
4 experimental period was maintained until the end of the experiment ($P = 0.04$).

5 The changes in BW are shown in Figure 2. During the adaptation period, the
6 BW of the ADP cows was greater than that of the CON cows ($P < 0.01$). At the start
7 of the experimental period, BW during the adaptation period decreased by 18 and 24 **[Figure 2]**
8 kg in the ADP and CON cows, respectively. Throughout the experimental period, the
9 BW of the ADP and CON cows gradually increased. However, the difference in BW
10 between the two groups was maintained until the end of the experiment ($P = 0.05$). **[Figure 2]**

11

12 **4 DISCUSSION**

13 **4.1 Herbage intake**

14 The results of the present study clearly indicated that the total DMI significantly
15 decreased after turning out from indoor feeding to 1-day grazing when the cows were
16 not adapted to grazing. The results of continuous measurement of herbage DMI in
17 lactating cows in the present study are similar to those reported in our previous study
18 on non-lactating cows (Mitani et al., 2020). The change and difference in herbage
19 DMI during the first week of the transition period from indoor feeding to 1-day
20 grazing affected the performance of cows during the 4-week period of 1-day grazing.
21 Therefore, grazing adaptation before turning out from indoor feeding to grazing is
22 essential for maintaining herbage DMI.

23 To assess grazing adaptability, gradual changes in herbage DMI, particularly
24 in the first week of the transition period from indoor feeding to grazing, should be
25 measured. However, no study has investigated such changes in herbage DMI during

1 this period, except our present and previous studies (Mitani et al., 2020). Other
2 previous studies investigated the effects of grazing adaptation using only one- or two-
3 point measurements of herbage DMI at 3 or 4 weeks after turning out from indoor
4 feeding to 1-day grazing (Hartwiger et al., 2018b; Schären et al., 2016b). To verify
5 the feasibility of our grazing adaptation method, further research is warranted
6 through continuous herbage DMI measurements during the transition period from
7 indoor feeding to grazing.

8 It is difficult to explain why the CON cows could not ingest herbage
9 satisfactorily after turning out to 1-day grazing. The well-known theory (Forbes,
10 2007) regarding the control of voluntary intake by physical and metabolic signals can
11 not explain the results observed in the present study. According to this theory, the
12 CON cows likely ingested more herbage because their physical and metabolic signals
13 were weaker than those of the ADP cows. Forbes (2007) speculated that this
14 phenomenon occurs because the animals are not familiar with the new food and
15 environment. The sudden change in the environment from indoor to outdoor feeding
16 may have affected the herbage intake of the CON cows to a greater extent than that
17 of the ADP cows. However, to clarify this phenomenon, further research
18 investigating the changes in herbage intake in other situations, such as adaptation to
19 the outdoor environment, are imperative.

20

21 **4.2 Ruminal fermentation and blood metabolites**

22 Grazing adaptation affected changes in ruminal fermentation and blood metabolites
23 during the first week after turning out to 1-day grazing. However, the effects of
24 grazing adaptation on rumen fermentation and PUN concentration observed in the
25 present study were contrary to those observed in our previous study on non-lactating

1 cows (Mitani et al., 2020). The high total VFA concentration of the CON cows on
2 day 2 of the experimental period was not necessarily promoted by ruminal
3 fermentation, but it may be affected by the concentration effect of ruminal pool size,
4 which was smaller due to the lower herbage intake of these cows than of the ADP
5 cows.

6 The ruminal microbial flora required over a week to adapt to herbage
7 degradation during transition from indoor feeding to grazing (Hartwiger et al., 2018b;
8 Mitani et al., 2020; Schären et al., 2016b). The extremely low total tract OM
9 digestibility of the CON cows during the first week of the experimental period
10 supports this result. The high PUN concentration of the CON cows, which is
11 indicative of high NH₃-N absorption from the rumen wall, can also indirectly support
12 this result. The ruminal microbes of the CON cows could not incorporate NH₃-N into
13 microbial proteins well because they were not adapted to degrade herbage
14 carbohydrates. Synchronized degradation of carbohydrates and proteins in the rumen
15 is important for efficient microbial protein synthesis, and the degradation of
16 carbohydrates and proteins in early spring herbage natively imbalances (Van Vuuren
17 et al., 1991).

18 Furthermore, the low blood glucose concentration of the CON cows during
19 the first week of the experimental period indicates a lower energy intake due to lower
20 herbage intake and ruminal fermentation of these cows than that of the ADP cows.
21 The tendency of high NEFA concentration in the CON cows during this period also
22 supports the lower energy intake of these cows than that of the ADP cows.

23

24 **4.3. Milk production and body weight change**

25 The higher milk production of the ADP cows than that of the CON cows during the

1 adaptation period was due to the gradual adaptation of the former to early spring
2 herbage, which has a high nutritive value. Moreover, the low herbage DMI of the
3 CON cows during the first week of the experimental period was a definite indicator
4 of the difference in milk production between the ADP and CON cows at the start of
5 the experimental period. In the present study, the difference in milk production
6 between the ADP and CON cows was maintained until the end of the experiment.
7 The change and difference in BW between the ADP and CON cows showed a similar
8 trend to those in milk production.

9 Previous studies measuring the changes in milk production and BW during
10 the transition period from indoor feeding to grazing compared with TMR feeding
11 have reported similar results (Hartwiger et al., 2018a; Schären et al., 2016a). The
12 requirement of a long recovery time for milk production and BW once a cow falls to
13 the low-feeding level is known as the “carryover effect” (Jørgensen et al., 2016). In
14 lactating dairy cows, the carryover effect may be caused by mammary cell number,
15 secretory cell activity, hunger state at removal from indoor feeding, and mobilization
16 of body reserves (Jørgensen et al., 2016). In addition, the physiological status, such
17 as blood hormone levels, may affect the carryover effect. Further research is
18 warranted to elucidate the carryover effect in lactating grazing cows during and after
19 transition from indoor feeding to grazing in early spring.

20 In conclusion, the present study revealed that gradual grazing adaptation was
21 effective in preventing herbage intake reduction and maintaining milk production and
22 BW. Herbage intake drastically decreased in the first week after turning out to 1-day
23 grazing when the cows were not sufficiently adapted to grazing. Once the milk
24 production and BW drastically decreased during the transition period from indoor to
25 1-day grazing, over 4 weeks were required to recover from the reduction during the

1 transition period. This decrease in milk production and BW after turning out to
2 grazing can result in economic losses to dairy farmers. Therefore, grazing adaptation
3 of dairy cows, particularly the lactating ones, is essential.

4

5 **CONFLICT OF INTEREST**

6 Authors declare no Conflict of Interests for this article.

7

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17

1 **FIGURE LEGENDS**

2 **Figure 1.** Changes in the herbage and total dry matter intake of grazing lactating
3 cows subjected to gradual adaptation to grazing (ADP) and non-adaptation (CON)
4 during the experiment.

5 Sum of mixed silage and concentrate intake: black, herbage intake: open.

6 Week 1-E indicates days 1 to 4 and week 1-L indicates days 5 to 7 of the experimental
7 period.

8 Significance in the experimental period: treatment, $P < 0.01$; day, $P < 0.01$;
9 interaction, $P < 0.01$.

10

11 **Figure 2.** Changes in the body weight of grazing lactating cows subjected to 1-week
12 adaptation to grazing (ADP) and non-adaptation (CON) during the experiment.

13 Significance in the experimental period: treatment, $P = 0.05$; day, $P < 0.01$;
14 interaction, $P = 0.95$.

15 Plots at -10: data measured before the start of the experiment (basal data).

16 Error bar: standard error of mean

Table 1. Chemical compositions of supplied feeds and herbage during trial

	Corn silage	Hay	Formula concentrate	Herbage				
				Adaptation Period	Experimental period			
					Week 1	Week 2	Week 3	Week 4
DM, % of FM	27.6	81.1	86.8	23.0	22.3	23.5	23.2	24.2
Chemical compositions, % of DM								
OM	93.3	91.8	94.0	90.1	90.5	89.1	88.0	90.2
CP	7.5	6.2	22.4	22.1	17.2	19.4	17.2	16.3
NDFom	54.0	66.1	24.7	24.9	25.0	26.7	29.6	34.8
WSC	–	–	–	26.2	25.9	26.2	21.9	16.3

DM: dry matter, FM: fresh matter, OM: organic matter, CP: crude protein, NDFom: neutral detergent fiber, WSC: water soluble carbohydrate

Table 2. Changes in total organic matter (OM) intake and apparent OM digestibility of grazing lactating cows subjected to gradual adaptation to grazing (ADP) and non-adaptation (CON) during the experiment

	Adaptation period †		Experimental period †					SEM	Significance ($P =$) ‡		
	Early	Late	Week 1-E	Week 1-L	Week 2	Week 3	Week 4		Trt.	Day	Int.
Total OM intake, kg/day/cow											
CON	16.1	16.1	7.0 **	12.4 **	12.6	12.7 *	15.0	0.6	< .01	< .01	< .01
ADP	17.3	12.3	14.1	16.0	12.9	14.6	16.1				
Apparent OM digestibility, %											
CON	70.9	70.3	53.6 **	74.4 **	75.9	75.3	76.6	0.9	< .01	< .01	< .01
ADP	68.9	65.4	74.1	78.7	76.0	76.7	78.3				
Digestible OM intake, kg/day/cow											
CON	11.5	11.3	4.0 **	9.3 **	9.6	9.6 *	11.5	0.5	< .01	< .01	< .01
ADP	11.9	8.1	10.4	12.6	9.9	11.2	12.6				

† Early: day 1 to day 7 and Late: day 8 to day 10 in adaptation period, Week 1-E: day 1 to day 4 and Week 1-L: day 5 to day 7 in experimental period.

‡ Trt.: ADP vs. CON, Day: collection day in the experimental period, Int.: interaction between Trt. and Day.

** $P < 0.01$, * $P < 0.05$: Significantly differed between CON and ADP for each period.

Table 3. Changes in ruminal volatile fatty acids (VFA) concentration and molar proportion of each VFA of grazing lactating cows subjected to gradual adaptation to grazing (ADP) and non-adaptation (CON) during the experiment

	Adaptation period [†]		Experimental period [†]							Significance ($P =$) [‡]		
	Early	Late	Day 2	Day 4	Day 7	Week 2	Week 3	Week 4	SEM	Trt.	Day	Int.
Total VFA, mmol/dL												
CON	13.8	13.0	17.3	18.6	19.9	15.5	17.3	16.0	1.0	0.21	< .01	0.08
ADP	18.7	19.8	13.8	17.8	16.8	15.4	16.5	17.0				
Molar proportion, mol/100mol												
Acetate												
CON	65.4	66.3	56.3	56.9	55.8	57.6	61.5	64.6	0.6	0.83	< .01	0.02
ADP	58.4	55.1	54.4	56.3	57.3	57.9	62.0	64.0				
Propionate												
CON	18.6	17.9	23.4	23.1	23.8	22.9	21.6	20.8	0.4	0.90	< .01	0.07
ADP	22.2	22.2	23.0	24.3	22.8	23.1	21.2	21.1				
Butyrate												
CON	12.7	12.5	15.9	14.8	14.8	14.9	12.1	11.1	0.4	0.64	< .01	0.22
ADP	15.0	18.0	17.3	14.6	14.9	14.5	12.2	11.5				

[†] Early: day 1 to day 7 and Late: day 8 to day 10 in adaptation period, Week 1 E: day 1 to day 4 and Week 1 L: day 5 to day 7 in experimental period.

[‡] Trt.: ADP vs. CON, Day: collection day in the experimental period, Int.: interaction between Trt. and Day.

Table 4. Changes in plasma urea nitrogen, glucose, and non-esterfied fatty acids concentrations of grazing lactating cows subjected to gradual adaptation to grazing (ADP) and non-adaptation (CON) during the experiment

	Adaptation period [†]		Experimental period [†]						SEM	Significance ($P =$) [‡]		
	Early	Late	Day 2	Day 4	Day 7	Week 2	Week 3	Week 4		Trt.	Day	Int.
Plasma urea nitrogen, mg/dL												
CON	8.2	7.7	13.9 **	12.3 **	15.9	13.1	16.0	10.9	0.7	0.02	< .01	< .01
ADP	12.8	16.0	7.7	9.2	15.6	12.6	14.9	9.1				
Glucose, mg/dL												
CON	62.6	61.3	56.4 *	63.2 *	65.3	64.7	67.6	67.1	1.4	0.12	< .01	0.02
ADP	59.4	61.2	61.2	67.6	69.4	67.1	67.2	67.6				
Non esterfied fatty acids, μ mol/L												
CON	255	262	210	219	138	137	145	107	28	0.31	0.12	0.57
ADP	107	160	152	134	111	105	97	89				

[†] Early: day 1 to day 7 and Late: day 8 to day 10 in adaptation period, Week 1 E: day 1 to day 4 and Week 1 L: day 5 to day 7 in the experimental period.

[‡] Trt.: ADP vs. CON, Day: collection day in the experimental period, Int.: interaction between Trt. and Day.

** $P < 0.01$, * $P < 0.05$: Significantly differed between CON and ADP for each period.

Table 5. Changes in milk yield, 4% fat corrected milk (FCM) yield, and milk compositions of grazing lactating cows subjected to gradual adaptation to grazing (ADP) and non-adaptation (CON) during the experiment

	Adaptation period †		Experimental period †						SEM	Significance ($P =$) ‡		
	Early	Late	Day 2	Day 4	Day 7	Week 2	Week 3	Week 4		Trt.	Day	Int.
Milk yield, kg/day/cow												
CON	30.7	30.7	28.6	30.4	31.1	32.5	34.0	32.9	1.5	0.08	< .01	0.97
ADP	32.9	33.3	32.5	33.7	34.1	35.7	37.2	35.1				
4% FCM yield, kg/day/cow												
CON	29.1	29.4	29.3	30.8	31.0	32.0	31.8	31.2	1.4	0.04	0.27	0.92
ADP	33.1	33.8	34.2	34.0	33.8	35.3	35.8	34.8				
Milk composition, %												
Fat												
CON	3.88	3.92	4.36	4.25	4.08	4.01	3.66	3.73	0.17	0.73	< .01	0.12
ADP	3.98	4.07	4.39	4.14	4.10	4.05	3.86	4.05				
Protein												
CON	3.26	3.22	3.17 *	3.18 *	3.23	3.29	3.26	3.17	0.07	0.05	< .01	0.02
ADP	3.44	3.48	3.47	3.45	3.44	3.46	3.43	3.34				
Lactose												
CON	4.44	4.43	4.34	4.33	4.37	4.39	4.41	4.39	0.04	0.95	0.13	0.81
ADP	4.34	4.42	4.30	4.34	4.41	4.39	4.42	4.40				
Solid not fat												
CON	8.64	8.66	8.50	8.52	8.60	8.65	8.65	8.54	0.06	0.01	< .01	0.08
ADP	8.89	8.85	8.78	8.79	8.85	8.86	8.84	8.76				

† Early: day 1 to day 7 and Late: day 8 to day 10 in adaptation period, Week 1 E: day 1 to day 4 and Week 1 L: day 5 to day 7 in the experimental period.

‡ Trt.: ADP vs. CON, Day: collection day in the experimental period, Int.: interaction between Trt. and Day.

** $P < 0.01$, * $P < 0.05$: Significantly differed between CON and ADP for each period.



