Effect of Extreme Walking Conditions for Dairy Cows on Milk Yield, Chemical Composition, and Somatic Cell Count[1]

J. B. COULON, P. PRADEL,[2] T. COCHARD,[3] and B. POUTREL[3]
Institute National de la Recherche Agronomique,
Laboratoire Adaptation des Herbivores aux Milieux,
63122 Saint Genès Champanelle, France

ABSTRACT

Thirty-two cows (16 Montbeliardes and 16 Tarentaises) in midlactation were used in an experiment utilizing a 2 × 2 factorial arrangement of treatments. Throughout the trial, cows received first-cutting cocksfoot hay for ad libitum intake supplemented with a fixed amount of concentrate that was individually adapted to the milk yield of each cow. During a 23-d experimental period, one group of cows walked 9.6 km/d; the other group of cows remained in the barn. Cows that walked daily ate less hay (–1.3 and –2.1 kg/d of dry matter for Tarentaise and Montbeliarde cows, respectively) and yielded less milk (–1.7 and –2.5 kg/d for Tarentaise and Montbeliarde cows, respectively) than did cows that did not walk daily. A residual effect of walking on milk yield was observed during the 10 d following the experimental period. For both breeds, fat content and, to a lesser extent, protein content were higher (+6.4 and +1.0 g/kg, respectively) for cows that walked. Somatic cell count was also higher for cows that walked (+115,000 cells/ml). This difference was more marked in cows that were initially infected by a minor or major pathogen (+185,000 cells/ml) than in uninfected cows (+47,000 cells/ml) and on the 1st d of walking when walking was linked to increases in pH, bovine serum albumin, and immunoglobulin G_1 contents of milk (+0.08 unit, +0.16 g/L, and +0.19 g/L, respectively). Throughout the experimental period, walking induced a rise in body temperature (+1°C) and in plasma nonesterified fatty acids (+0.63 m*M*/L). On the 1st d of walking, plasma glucose, lactic acid, and cortisol contents were significantly higher for cows that walked (+0.25 g/L, +0.64 g/L, and +28.8 ng/ml, respectively).

(**Key words**: dairy cows, walking, milk composition, somatic cell count)

Abbreviation key: **UFL** = unité fourragère lait (feed unit for lactation).

INTRODUCTION

Under certain environmental conditions (scarce forage or hilly grazing), walking constitutes a major activity for cows that increases nutritional requirements (20, 24, 29) and sometimes reduces yield (23). The distance covered may vary according to the type of animal (3, 19), although few studies have been conducted with dairy cows (34), and most of those studies have examined cows that walked short distances (16, 18). Earlier trials (9, 11) have shown that the reduction in milk yield that results from long walks (9 to 12 km) was significant, varied according to animal type, and was associated with a considerable increase in milk SCC without clinical signs of mastitis. This effect may precipitate important economic consequences when the price of milk is set in consideration of SCC. These results were obtained after a single exercise. The goal of the study was to analyze the effect of repeated walking on milk yield, chemical composition, and SCC in two different breeds of cows to specify whether these cows were capable of adapting to regular exercise.

MATERIALS AND METHODS

Cows and Treatments

Thirty-two dairy cows (16 Montbeliarde cows and 16 Tarentaise cows) were included in an experiment carried out between May 13 and July 4, 1996. Cows were in midlactation (171 d) at the beginning of the experiment; 8 cows were in first lactation, and 24 cows were in second lactation or later. Cows were housed in individual stalls. Two groups of 8 cows of each breed were formed according to parity, milk yield, and milk composition as observed during the last 2 wk of May (preexperimental period). All cows were free of clinical mastitis and foot lesions at the

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[2]Experimental farm, 15190 Marcenat, France.
[3]Institut National de la Recherche Agronomique, Laboratoire de Pathologie Infectieuse et Immunologie, 37380 Nouzilly, France.

beginning of the trial. During the 23-d experiment, one group of cows walked every day, and the other group of cows remained at the barn. Cows were walked after the morning milking (between 0830 and 1100 h) on a 3.2-km course with a total elevation change of 80 m. The course was covered three times consecutively according to the procedures defined by D'Hour et al. (11). The exception to this routine was that during the first 3 d, cows covered the course four times. The surface of the course varied: one-third was tarred, and the remainder was ungraded or grassy. During walking, cows were accompanied by the same two people, one leading the way 10 m ahead of the group, and the other following. The second person walked at a constant speed of about 1 m/s, remaining at least 1 m behind the last cow. If the last cow walked faster, the person following the herd also walked faster. If the last cow slowed below 1 m/s or stopped, she was made to move ahead. Cows were not allowed to eat or drink during walking. During a 10-d postexperimental period, all of the cows were managed together at the stabling.

Throughout the trial, all cows were fed a diet composed of first-cutting cocksfoot hay [0.67 **UFL** (unité fourragère lait; feed unit for lactation) (36) and 57 g of true protein truly digestible in the small intestine/kg of DM (35)] provided for ad libitum consumption and a protein concentrate (1.03 UFL and 100 g of true protein truly digestible in the small intestine/kg of DM). The concentrate amounts were individually established for the duration of the trial according to the milk yield recorded for each cow during the 2 wk preceding the beginning of the trial (i.e., 0, 1, 2, 3 and 4 kg/d for milk yields of 12, 14, 16, 18, and 20 kg/d of FCM). In addition, all cows received 0.5 kg/d of soybean meal and 0.15 kg/d of mineral supplement (60 g/kg of P and 220 g/kg of Ca). Hay was fed in two daily meals, and concentrate was fed in one meal, except when quantities were >3 kg/d at which time the excess was fed in a second meal.

Measurements

Feed intake and feeding behavior. The quantities of feed ingested were measured individually each day. On 3 d of the experimental period (d 1, 8, and 20), orts were measured twice daily (1600 and 0700 h) to assess the quantities of feed consumed in the first hours following the return of the cows to the stable. Feed DM content was determined daily for hay and weekly for concentrate. The chemical composition of the hay was determined once during the experiment. The DM and OM digestibilities of the hay were determined using six wethers during a 1-wk measurement period following a 2-wk adaptation period. Their nutritional value was computed according to the formulas of Andrieu and Demarquilly (1). The quantities of water consumed by each group of cows were measured every day.

On 3 d of the experimental period (d 1, 8, and 20), the behavior of the cows (eating, standing, or lying down) was visually checked and recorded every 5 min between their return to the barn (1100 h) and the evening milking (1530 h) for the group of cows that walked and between the two milkings for the group of cows that remained at the barn.

Cows were weighed once weekly, on Tuesdays, in early afternoon.

Milk yield and composition. Milk yield was individually weighed at each milking. Milk composition (fat, protein, and lactose contents) was determined at each milking (infrared method; Synergy, Anadis, Alès, France). The SCC were determined every day at the evening milking (Somacount; Bentley Instruments, Chaska, MN).

On 1 d of the preexperimental period and on 3 d of experimental period (d 1, 8, and 20), pH, and contents of casein, soluble protein (31), BSA (28), IgG_1 (21), Ca (14), and P (13) were measured in individual milk samples collected at the evening milking.

Plasma metabolites. Once prior to the experiment and on 3 d (d 1, 8, and 20) during the experiment, body temperature was measured, and a blood sample was taken by caudal venipuncture from each cow upon her return to the barn. Plasma was prepared and frozen for later analysis of glucose (4), NEFA (6), urea (Merck, Darmstadt, Germany), lactic acid (BioMérieux, Marcy l'Etoile, France), and cortisol contents (5).

Bacteriological diagnosis of IMI. Foremilk samples from all quarters were collected aseptically for diagnosis of IMI within the preexperimental week and on experimental d 1 and 8. Milk samples were frozen and stored before bacterial examination. A 0.025-ml aliquot of each quarter milk sample was plated on esculin sheep blood agar. The plates were incubated aerobically for 24 to 48 h at 37°C before examination for bacterial identification. Cows were considered to be infected when at least one quarter was infected by a minor pathogen (*Corynebacterium bovis* or coagulase-negative staphylococci) or by a major pathogen (*Staphylococcus aureus* or *Streptococcus* spp.) Cows were considered to be uninfected when all four quarters were free of IMI.

Differential cell counts in milk samples. The distribution of the main cellular populations, polymorphonuclear cells and mononuclear cells, was determined by May-Grunwald-Giemsa staining from composite milk samples collected on experimental d 1 and 8. Briefly, cells were spun down onto glass slides by centrifugation ($100 \times g$ for 10 min) of 200 μl of washed skimmed milk supplemented with normal bovine serum with a cytocentrifuge (Cytospin SCA 0030; Shandon, Life Science International SA, Runcorn, UK) and then stained. The slides were examined under the microscope with a 100× oil immersion objective.

Mammary and foot characteristics. Before the experimental period, udder conformation of each cow was rated according to eight criteria (cleavage depth, distance from the knuckle to the floor, front and rear balance, lateral teat deviation, front and rear teat deviation, height of rear ligament, and teat length). A synthetic score was computed by summing the deviations from the optimal scores on each criterion. During the preexperimental and postexperimental periods, the walking performance of each cow was scored on a five-point scale (0 = normal walk to 5 = lameness). Hoof length and height were measured on the same days.

Data Analysis

The effect of walking was assessed by considering the performance of the cow over the entire experimental period (23 d), the postexperimental period (10 d), and during some days (d 1, 8, and 20) when particular measurements were taken. These data were processed by ANOVA (32). The fixed factor effects included in the model were treatment, breed, the interaction of treatment and breed, and the value of the variables observed during the preexperimental period as a covariate when available. The SCC were expressed as log-transformed values. Values presented in Figure 4 are geometric means. For behavior variables, time spent eating, ruminating, lying, or standing was computed either during the interval between the two milkings or during the interval between the return of cows to the stable after walking and the evening milking. When the udder infection status of a cow remained unchanged throughout the experimental period, the cows were considered eligible for the evaluation of the effect of walking on milk SCC. For SCC analysis, the fixed factor effects included in the model were treatment, infection status, and the interaction of treatment and infection status.

RESULTS

Gait and Hoof Condition

Walking induced marked changes in the hoof condition of the cows. Although most cows (13 of 16) had normal or slightly impaired gait before the experiment, cows of both breeds in the walking group had highly impaired gait (5 of 16) or lameness (6 of 16) at the end of the experiment (Table 1). Hoof height of the hind legs was not modified, but hoof length was significantly reduced ($P < 0.01$) for cows in the walking group relative to cows in the group that remained at the barn (–0.8 cm).

TABLE 1. Effect of walking on cow gait.

	Tarentaise		Montbeliarde	
Gait	Resting	Walking	Resting	Walking
	(no. of cows)			
Preexperimental period				
Normal	7	7	5	6
Affected	1	1	2	1
Lameness	0	0	1	1
Postexperimental period				
Normal	6	2	6	3
Affected	2	3	1	2
Lameness	0	3	1	3

Intake Amounts, Milk Composition, and Yield

Two cows had to be withdrawn from performance analysis; one cow had clinical mastitis once during the preexperimental period and twice during the experimental period, and the other cow had severe lameness because of walking. The lameness was noted on d 10 of the experimental period and prevented the cow from walking.

Milk yield during the walking period was lower on average for cows in the walking group than for cows in the group that remained at the barn ($P < 0.01$; Table 2). The difference between the groups of cows was smaller for Tarentaise cows (–1.7 kg/d) than for Montbeliarde cows (–2.5 kg/d). For the latter cows, the difference increased during the experimental period (Figure 1) from 2 kg/d during the 1st wk of walking to 3.3 kg/d during the last week of walking. However, this interaction was not significant. Milk yield began to decrease on the 1st d of walking. The decrease was greater ($P < 0.01$) when milk yield during the experimental period was higher and when udder conformation score was worse (Figure 2). Collectively, these two variables explained 76% of the yield decrease variability for cows that walked daily. The decrease in milk yield for both breeds was equally associated with a sharp increase in fat content (+6.4 g/kg between the group that walked and the group that remained at the barn; $P < 0.01$), especially during the 1st d of walking (Figure 3). Consequently, the amount of fat yielded did not differ greatly between the two groups [–31 g/d on average over the experimental period ($P < 0.05$)]. Protein content was higher on average for cows of both breeds that walked (+1.0 g/kg; $P < 0.05$), but that effect was only significant at the beginning of the walking period (Figure 3) and became nonsignificant from d 14. Therefore, the amounts of protein yielded were lower for cows that walked (–81 g/d; $P < 0.01$). The proportion of casein in total proteins was not altered by walking except on the 1st d when it was significantly lower in cows that walked (0.79 vs. 0.81; $P < 0.05$) because of a proportionally higher increase in the soluble protein content (+0.9 g/kg; +14%) than in the casein content (+1 g/kg; +4%). Lactose content decreased during the first 4 walking d (–2.2 g/kg on the 1st day; $P < 0.05$) but remained identical for both

TABLE 2. Milk yield, feed intake, and behavior data.

	Tarentaise		Montbeliarde			Significance[1]	
	Resting	Walking	Resting	Walking	RSD[2]	Trt	Trt × Breed
						P	
Cows, no.	8	7	8	7			
Week before walking							
BW, kg	568	577	629	625	41	NS[3]	NS
Milk yield, kg/d	10.6	10.6	15.0	14.8	2.8	NS	NS
Fat concentration, g/kg	38.1	37.7	38.4	37.0	2.9	NS	NS
Protein concentration, g/kg	32.9	34.0	32.2	32.1	2.2	NS	NS
Lactose concentration, g/kg	47.3	47.0	47.9	47.7	1.8	NS	NS
Concentrate intake, kg of DM/d	2.2	2.4	3.3	3.3	1.0	NS	NS
Hay intake, kg of DM/d	9.2	9.4	11.5	10.9	1.0	NS	NS
Experimental period							
BW, kg	570	539	628	580	15	**	NS
Hay intake, kg of DM/d	10.0	8.7	12.8	10.7	1.2	**	NS
Concentrate intake, kg of DM/d	1.4	1.1	2.0	1.8	0.9	NS	NS
Milk yield, kg/d	8.2	6.5	11.4	8.9	1.1	**	NS
Fat concentration, g/kg	38.5	45.0	37.2	43.4	2.1	**	NS
Protein concentration, g/kg	31.3	32.1	29.9	30.8	1.1	*	NS
Lactose concentration, g/kg	45.8	45.5	46.7	46.8	0.9	NS	NS
Fat yield, g/d	316	292	425	387	40	*	NS
Protein yield, g/d	249	206	342	275	31	**	NS
Ca,[4] g/kg	1.31	1.32	1.24	1.33	0.10	NS	NS
P,[4] g/kg	0.93	0.93	0.92	0.88	0.08	NS	NS
Casein:protein[4]	0.81	0.80	0.81	0.80	0.02	NS	NS
Energy supply, UFL[5]/d	7.7	6.6	10.3	8.4	1.5	*	NS
Energy balance, UFL/d	–0.6	–1.1	0.3	–0.5	1.5	NS	NS
N Balance, g of PDI[6]/d	153	107	245	166	164	NS	NS
Time spent lying,[7] min	127	139	107	121	28	NS	NS
Time spent eating,[7] min	157	82	162	110	20	**	NS
Time spent lying,[8] min	123	139	104	121	27	NS	NS
Time spent eating,[8] min	73	82	80	110	18	**	†
Postexperimental period							
BW, kg	578	550	637	601	19	**	NS
Hay intake, kg of DM/d	9.3	9.3	11.6	10.7	1.4	NS	NS
Milk yield, kg/d	6.4	4.9	8.6	5.9	1.3	**	NS
Fat concentration, g/kg	40.6	42.7	41.7	43.8	2.4	NS	NS
Protein concentration, g/kg	31.5	32.0	30.1	31.6	1.6	NS	NS

[1]Trt = Treatment; Trt × breed = interaction of treatment and breed.

[2]Residual standard deviation.

[3]$P > 0.1$.

[4]Mean of the three samplings taken on d 1, 8, and 20.

[5]Unité fourragère lait (feed unit for lactation).

[6]Protein digestible in the small intestine (35).

[7]Between the end of the morning milking and the beginning of the evening milking.

[8]Between the end of walking and the beginning of the evening milking.

†$P < 0.1$.

*$P < 0.05$.

**$P < 0.01$.

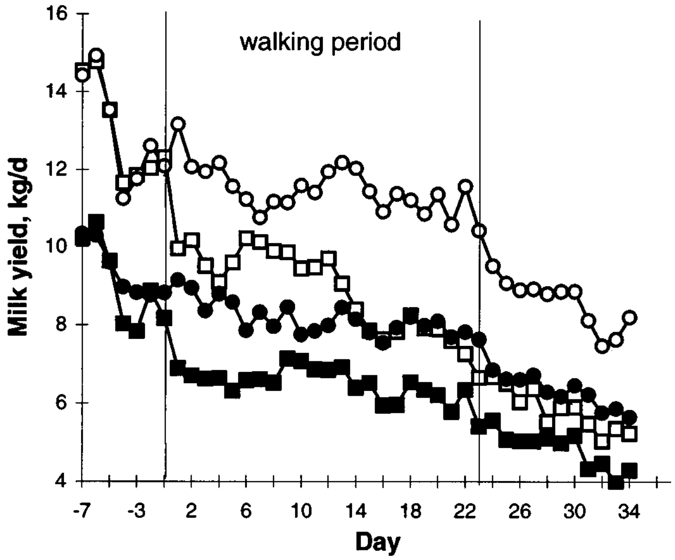

Figure 1. Milk yield pattern for Tarentaise cows that walked (■) or remained at the barn (●) and for Montbeliarde cows that walked (□) or remained at the barn (o).

treatment groups thereafter (Figure 3). Milk Ca and P contents did not differ significantly between treatments. The live weight of cows that walked was 40 kg lower than that of cows that remained at the barn ($P < 0.01$). Live weight variations, computed between the last week of the preexperimental period and the 1st wk of the postexperimental period (i.e., under identical weighing conditions) were –22 and +10 kg for cows that walked daily and those that remained at the barn, respectively ($P < 0.01$).

During the experimental period, quantities of forage ingested between the morning and evening milkings were 2.0 kg of DM lower for cows that walked. Some of this difference was recovered overnight after the 1st d of walking (+0.8 kg of DM) but not later (–0.6 kg of DM on d 8 and 20). On a daily scale, the difference between the two groups was 1.8 kg/d of DM on average ($P < 0.01$), but this difference increased during the course of the experimental period (1.5, 2.1, and 3.5 kg/d of DM on d 1, 8, and 20, respectively). Differences were twice as great in Montbeliarde cows as in Tarentaise cows (–2.3 vs. –1.2 kg of DM/d). Therefore, energy supplies were lower for cows that walked (–1.1 and –1.9 UFL for Tarentaise and Montbeliarde cows, respectively). The volumes of water consumed by each group of cows, on average, were slightly higher for cows that remained at the barn (77 vs. 68 L/d per cow for cows that walked).

On walking days, the cows that walked, once back at the barns, spent 20 min more eating (9 min for Tarentaise cows and 30 min for Montbeliarde cows) than did those that were kept at the barn ($P < 0.01$). The cows that remained at the barn ate 64 min longer between the two milkings than did the walking cows ($P < 0.01$). Based on intake amounts, the rate of forage intake was not altered by walking (26.9 and 25.4 min/kg of DM for cows that remained at the barn and for cows that walked, respectively). Lying time between the two milkings did not differ between treatments, but standing time (without eating) was

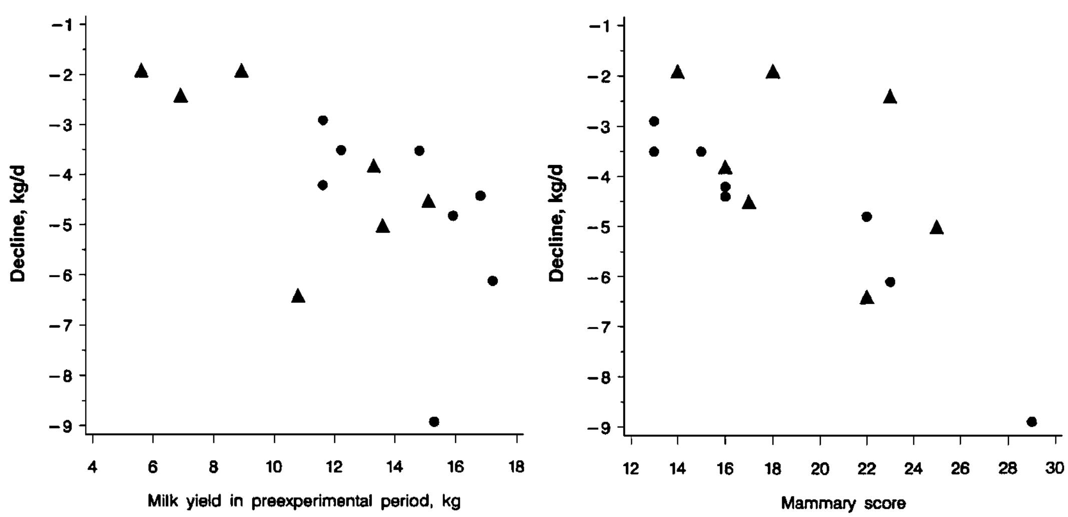

Figure 2. Relationship between the decline in milk yield during the 1st d of walking and the milk yield in the preexperimental period or the mammary score. The mammary score was computed by summing the deviations from the optimal scores for each of the following criteria: cleavage depth, distance from the knuckle to the floor, front and rear balance, lateral teat deviation, front and rear teat deviation, height of rear ligament, and teat length. Legend: Tarentaise cows (▲); Montbeliarde cows (●).

higher for cows that remained at the barn (80 min) than for cows that walked (44 min).

Postexperimental milk yield remained lower ($P < 0.01$) for cows that walked (–2.1 kg/d). Differences in milk composition and intake amounts did not differ significantly between treatments (Table 2).

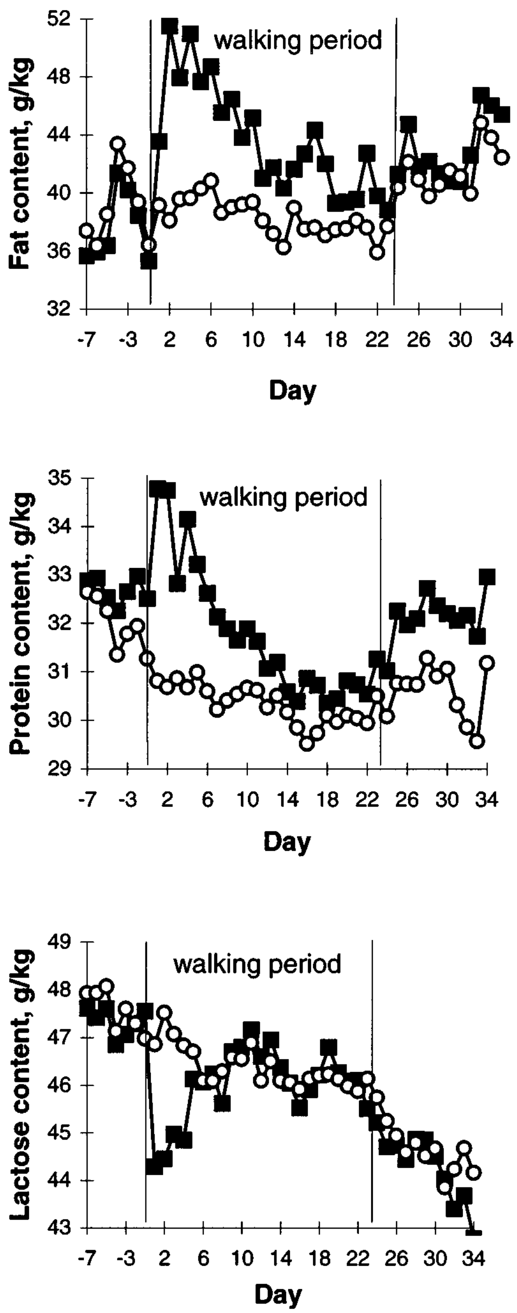

Figure 3. Milk composition pattern for cows that walked (■) and for cows that remained at the barn (o).

Blood Metabolites

During the 3 d of measurement, walking increased ($P < 0.01$) body temperature, which was greater in Tarentaise cows (+1.2°C) than in Montbeliarde cows (+0.7°C). Walking also introduced an increase ($P < 0.01$) in plasma NEFA (+0.63 m*M*/L) (Table 3). On walking d 1, plasma glucose and lactic acid contents were higher by 0.25 ($P < 0.01$) and 0.64 g/L ($P < 0.05$), respectively. No difference between the two groups remained during the following days. Cortisol concentrations were increased on walking d 1 and 8 for cows that walked (+28.8 and +6.6 ng/ml; $P < 0.01$). On walking d 1, a positive correlation was observed between cortisol and glucose concentrations ($R^2 = 0.52$). No significant difference in blood urea between groups was evident, regardless of the day of measurement.

SCC

During the trial, the infection status of 27 cows did not change (19 infected and 8 uninfected), and these cows were included in this analysis. Of the 19 infected cows, 14 were infected by a minor pathogen (6 cows that walked and 8 cows that remained at the barn), and 5 were infected by a major pathogen (2 cows that walked and 3 cows that remained at the barn). Before walking, the SCC of uninfected cows was lower ($P < 0.01$) than SCC of infected cows (Table 4). The SCC was considerably increased at the first milking following the first walk ($\overline{X}$ = +350,000 cells/ml), but no case of clinical mastitis was detected. The difference between cows that walked and cows that remained at the barn later decreased during the course of the experiment (Figure 4). For the entire walking period, mean SCC was 115,000 cells/ml higher for cows that walked. This mean difference was lower in uninfected cows (+47,000 cells/ml over the experimental period) than in infected cows (+185,000 cells/ml), but that interaction was not significant (Table 4; Figure 4). During the postexperimental period, SCC were comparable for uninfected cows of both groups (64,000 and 86,000 cells/ml for cows that remained at the barn and cows that walked, respectively). The SCC were higher, but not significantly, for infected cows that walked than for infected cows that remained at the barn (304,000 vs. 202,000 cells/ml, respectively).

Regardless of infection status, on the 1st d of walking, cows had a higher pH (+0.08; $P < 0.01$), a higher BSA content (+0.16 g/L; $P < 0.01$), and a higher IgG_1 content (+0.19 g/L: $P < 0.05$) than did cows that remained at the barn. Later, no difference was observed in pH, BSA, or IgG_1 between groups (Table 4). The increase in SCC on the 1st walking d varied

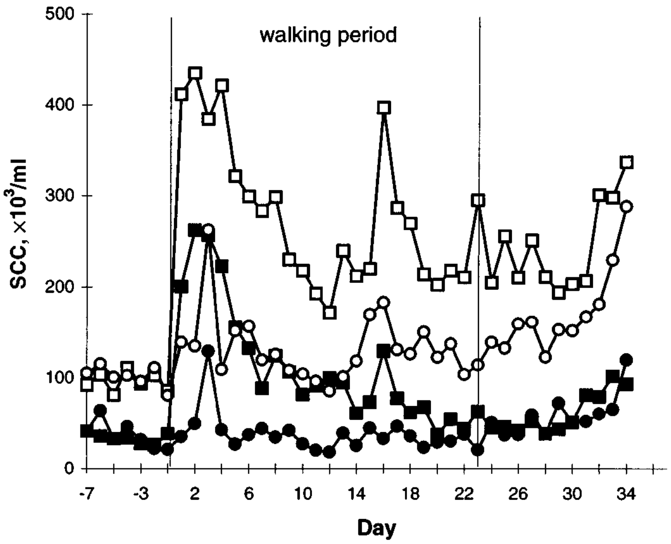

Figure 4. Geometric mean SCC for uninfected cows that walked (■), uninfected cows that remained at the barn (●), infected cows that walked (□), and infected cows that remained at the barn (o).

greatly among cows. Increased SCC was low or null in 5 cows (<100,000 cells/ml) and >500,000 cells/ml in 4 cows. This increase was not linked to the baseline milk SCC or to the magnitude of milk yield loss, but was greater as BSA concentrations increased (R^2 = 0.86; P < 0.001). Increased SCC was only weakly correlated with udder conformation (R^2 = 0.23; P < 0.1).

Regardless of udder infection status, cell type was modified by walking (Table 4). During the 1st d of walking, the proportion of polymorphonuclear cells was higher (P < 0.05) for cows that walked than for cows that remained at the stabling (66% vs. 50%).

DISCUSSION

This experiment confirmed the significant influence of prolonged walking on milk yield and composition (9), even for low yielding cows. The effect of walking would likely be more marked for higher

TABLE 3. Body temperature and blood metabolites.

	Tarentaise		Montbeliarde			Significance[1]	
	Resting	Walking	Resting	Walking	RSD[2]	Trt	Trt × Breed
						P	
Cows, no.	8	7	8	7			
d 1							
Body temperature, °C	38.0	38.8	38.3	39.0	0.3	**	NS[3]
Glucose, g/L	0.59	0.81	0.53	0.81	0.16	**	NS
Urea, g/L	0.28	0.31	0.32	0.33	0.04	NS	NS
NEFA, m*M*/L	0.30	0.72	0.14	0.71	0.19	**	NS
Lactic acid, m*M*/L	1.09	1.46	0.77	1.69	0.69	*	NS
Cortisol, ng/ml	5.3	37.1	2.9	28.6	9.8	**	NS
d 8							
Body temperature, °C	38.3	39.7	38.4	39.2	0.5	**	†
Glucose, g/L	0.60	0.59	0.52	0.54	0.04	NS	NS
Urea, g/L	0.29	0.27	0.32	0.30	0.05	NS	NS
NEFA, m*M*/L	0.33	1.38	0.21	0.86	0.28	**	†
Lactic acid, m*M*/L	0.92	1.13	1.27	1.17	0.64	NS	NS
Cortisol, ng/ml	3.4	14.0	4.7	7.4	5.1	**	†
d 20							
Body temperature, °C	37.9	39.4	38.4	39.3	0.4	**	*
Glucose, g/L	0.61	0.62	0.53	0.57	0.05	NS	NS
Urea, g/L	0.30	0.32	0.34	0.33	0.05	NS	NS
NEFA, m*M*/L	0.22	0.87	0.15	0.57	0.30	**	NS
Lactic acid, m*M*/L	0.83	0.64	0.96	1.02	0.49	NS	NS
Cortisol, ng/ml	4.6	6.7	5.6	8.1	3.8	NS	NS

[1]Trt = Treatment; Trt × Breed = interaction of treatment and breed.

[2]Residual standard deviation.

[3]P > 0.1.

†P < 0.1.

*P < 0.05.

**P < 0.01.

yielding cows (11) as was indicated by the correlation between milk yield loss and milk yield recorded before the experiment. The decline in milk yield on the 1st d of walking (about 3 kg/d) far exceeded that observed for cows kept on pasture with the same physical activity (7, 11, 22, 34). In the present study, cows were not accustomed to walking, which might partially account for this difference. Before the beginning of the trial, the only exercise was walking from stalls to the milking parlor (i.e., around 200 m/d). The marked effect of walking might also have been enhanced because walking took place just after the morning milking when the cows were fed their main meal diet (12). The physiological consequences of walking on hoof condition and the considerable changes in blood constituents [indicators of stress (cortisol)], body reserve mobilization, and reaction to exercise observed on the 1st d of walking reflected that cows were not trained for the walking course. The consequences and changes were consistent with observations made for cows (11, 27), ewes (2), and pigs (39). Most of these differences disappeared on the 8th d of walking, reflecting metabolic adaptation to walking. However, the difference in milk yield between walking and sedentary cows increased during the trial, which was contrary to the observations of Thomson and Barnes (34). As observed by Henning (17), Matthewman et al. (23), and Zerbini et al. (38), this difference was due to the lack of additional forage intake by cows to compensate for the increased energy requirements induced by exercise. In fact, the walking cows ate less forage than did the sedentary cows because of restricted access time, which was not compensated for by a higher intake rate or a suffi-

TABLE 4. Effect of walking and infection status of cows on SCC and some milk characteristics.

	Uninfected		Infected			Significance[1]		
	Resting	Walking	Resting	Walking	RSD[2]	Inf	Trt	Inf × Trt
						P		
Cows, no.	3	5	11	8				
SCC, log/ml								
Week before walking	4.65	4.63	5.07	5.00	0.27	**	NS[3]	NS
d 1	4.55	5.30	5.14	5.61	0.46	*	**	NS
d 8	4.54	5.10	5.10	5.47	0.49	*	*	NS
d 20	4.47	4.58	5.09	5.31	0.30	**	NS	NS
Experimental period	4.63	4.91	5.16	5.48	0.30	**	*	NS
pH								
Week before walking	6.71	6.73	6.76	6.71	0.06	NS	NS	NS
d 1	6.59	6.69	6.62	6.68	0.06	NS	**	NS
d 8	6.54	6.56	6.59	6.58	0.06	NS	NS	NS
d 20	6.60	6.62	6.64	6.63	0.04	NS	NS	NS
BSA, g/L								
Week before walking	0.18	0.14	0.17	0.19	0.05	NS	NS	NS
d 1	0.10	0.25	0.15	0.32	0.13	NS	**	NS
d 8	0.17	0.19	0.15	0.17	0.09	NS	NS	NS
d 20	0.13	0.13	0.13	0.18	0.06	NS	NS	NS
IgG_1, g/L								
Week before walking	0.49	0.50	0.59	0.59	0.21	NS	NS	NS
d 1	0.40	0.57	0.48	0.68	0.18	NS	*	NS
d 8	0.34	0.57	0.57	0.50	0.29	NS	NS	NS
d 20	0.44	0.46	0.49	0.66	0.20	NS	NS	NS
Polymorphonuclear cells, %								
d 1	34	67	54	65	23	NS	*	NS
d 8	43	55	56	65	15	NS	NS	NS
Polymorphonuclear cells, log/ml								
d 1	4.06	5.08	4.96	5.45	0.62	*	*	NS
d 8	4.18	5.01	4.91	5.56	0.54	*	**	NS

[1]Inf = Infection, Trt = treatment, and Inf × Trt = interaction of infection and treatment.

[2]Residual standard deviation.

[3]$P > 0.05$.

*$P < 0.05$.

**$P < 0.01$.

cient increase in nocturnal intake. Fatigue might have been responsible for that difference. Metz (25) demonstrated that dairy cows required a minimum amount of rest during the day.

The increase in energy requirements that was linked to walking can be estimated, according to the calculations of Ribeiro et al. (29) and considering the circuit characteristics, to be approximately 3380 kcal of metabolizable energy [i.e., 1.2 UFL (assuming effective utilization of metabolizable energy for lactation of 0.6)]. Overall, the effect of walking resulted in a difference of 2.7 UFL between sedentary and walking cows [1.2 UFL because of the increase in energy requirements plus 1.5 UFL because of the lower energy supply (Table 2)]. This difference corresponded with 6.2 kg of FCM if maximal marginal efficiency is 2.3 kg/UFL. However, the response of the cows was far below this theoretical value because of the ability to mobilize rapidly the body reserves, as indicated by the extremely high increase in plasma NEFA content observed immediately after walking and throughout the walking period (2, 11, 22, 27).

As has been commonly observed (9, 11, 34), the decrease in milk yield was accompanied by a considerable increase in fat content, which was linked to fat concentration, as has often been observed when feed supply fluctuated widely (33). Cows mobilize their body reserves to synthesize more concentrated milk fat. Contrary to previous observations when dietary supply was reduced (10), milk protein content was not reduced in cows that walked; instead, milk protein increased. This response was due to a greater (during the first 10 d of walking) or equivalent (the following days) decrease in milk yield than in protein yield.

As observed previously (7, 11), the milk yield response to walking differed between breeds. This difference was not due to increased intake by cows after exercise but essentially to the difference in yield between breeds, as is illustrated by Figure 2. However, this disparity could also have been due to morphological (lower live weight) or metabolic differences (higher body temperature and higher plasma NEFA and cortisol contents) that enable Tarentaise cows to spend less energy when walking and to mobilize body reserves more efficiently.

The effects of walking on milk yield were maintained during the postexperimental period, which demonstrates that the walking effect was not solely due to changes in energy balance. When such was the case, no residual effect was observed when the high energy supply was restored, as when cows that are underfed during the winter season are returned to grazing (8, 15). Therefore, walking likely alters the synthesizing capacity of the udder in an irreversible manner. This effect was perhaps emphasized because cows were in the second part of lactation (6th to 7th mo). Despite the low number of cows, this trial showed that the initial infection status of the udder partly controlled the effect of walking. In the postexperimental period, the milk yield difference between cows that remained at the barn and cows that walked was not significant for uninfected cows (1.2 kg/d; $P > 0.1$) but significant for infected cows (2.5 kg/d; $P < 0.01$).

This experiment elicited a considerable increase in milk SCC after the 1st d of walking and confirmed our previous results (9). This increase in SCC occurred essentially during the 1st d of walking and, as is commonly observed in cows with mastitis or those with infected udders (28, 30), the increased SCC was associated with higher pH, lower milk lactose, and elevated BSA and IgG_1 concentrations. These effects reflect an increase in capillary permeability (28). The indirect effect of the concentration of somatic cells, which was linked to reduced milk yield, can only explain a small part of this result because we observed no interindividual relationship between the decrease in milk yield and the increase in SCC. This experiment showed that a significant increase in milk SCC may occur in uninfected cows also, indicating a perhaps traumatic, noninfectious inflammation induced by walking, as was suggested by the relationship between udder conformation and the sharp decrease in milk yield. Although substantial increases in SCC have occasionally been associated with thermal stress or drug treatment (37) in the absence of mammary infection, those results are disputed (26). However, the effect of walking is higher in cows that were already infected, suggesting that the SCC increase is the cumulative result of infection and traumatic inflammation.

CONCLUSIONS

Extreme exercise of untrained cows lowered milk yield, altered milk composition, and raised SCC. The practical consequences of these results can be considerable, particularly in cows managed on pasture in an extensive system. These results may lead to a marked reduction of the milk price paid to farmers. This study has shown that such consequences are highly dependent on the initial characteristics of the cow (e.g., infection status and udder conformation). Therefore, in uninfected cows, the mean milk SCC was never >300,000 cells/ml during the experimental period; under these conditions, walking had no economic impact that could be linked to SCC. In practice, although walking distances may sometimes total those of this experiment, cows are generally trained and spread exercise performance over a longer period. Further studies should be undertaken to assess the effect of training and exercise distribution throughout the day on dairy cow performance.

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REFERENCES

1 Andrieu, J., and C. Demarquilly. 1987. Valeur nutritive des fourrages: tables et prévisions. Bull. Tech. Ctr. Rech. Zootech. Vet. Theix Inst. Natl. Rech. Agron. 70:61–73.
2 Animut, G., and K. D. Chandler. 1996. Effects of exercise on mammary metabolism in the lactating ewe. Small Ruminant Res. 20:205–214.
3 Arnold, G. W., and M. L. Dudzinski. 1978. Ethology of Free-Ranging Domestic Animals. Elsevier, Amsterdam, The Netherlands.
4 Bergmeyer, H. U., E. Bernt, F. Smith, and H. Stork. 1974. D-Glucose determination with hexokinase and glucose-6-phosphate-deshydrogenase. Pages 1196–1201 *in* Methods of Enzymatic Analysis. Vol 3. H. U. Bergmeyer, ed. Acad. Press, London, England.
5 Boissy, A., and M. F. Bouissou. 1994. Effects of androgen treatment on behavioral and physiological responses of heifers to fear-eliciting situations. Horm. Behav. 28:66–83.
6 Chilliard, Y., D. Bauchart, and J. Barnouin. 1984. Determination of plasma non-esterified fatty acids in herbivores and man: a comparison of values obtained by manual or automatic chromatographic, titrimetric, colorimetric and enzymatic methods. Reprod. Nutr. Dev. 24:469–482.
7 Coulon, J. B., and J. P. Garel. 1996. Aptitude à la marche de vaches laitières de types génétiques différents: influence d'un exercice répété sur la production laitière. Ann. Zootech. (Paris) 45:349–355.
8 Coulon, J. B., J. P. Garel, and A. Hoden. 1986. Evolution de la production et de la composition du lait à la mise à l'herbe. Bull. Tech. Ctr. Rech. Zootech. Vet. Theix Inst. Natl. Rech. Agron. 66: 23–29.
9 Coulon, J. B., and P. Pradel. 1997. Effect of walking on roughage intake and milk yield and composition of Montbéliardes and Tarentaises dairy cows. Ann. Zootech. (Paris) 46:139–146.
10 Coulon, J. B., and B. Rémond. 1991. Variations in milk output and milk protein content in response to the level of energy supply to the dairy cow: a review. Livest Prod. Sci. 29:31–47.
11 D'hour, P., A. Hauwuy, J. B. Coulon, and J. P. Garel. 1994. Walking and dairy cattle performance. Ann. Zootech. (Paris) 43:369–378.
12 Dulphy, J. P., and P. Faverdin. 1987. L'ingestion alimentaire chez les ruminants: modalités et phénomènes associés. Reprod. Nutr. Dev. 27:129–155.
13 International Dairy Federation. 1990. Milk: determination of total phosphorus content—spectrometric method. IDF Stand. 42B. Int. Dairy Fed., Brussels, Belgium.
14 International Dairy Federation. 1992. Milk and dried milk: determination of calcium content—flame atomic absorption method. IDF Stand. 154. Int. Dairy Fed., Brussels, Belgium.
15 Gordon, F. J. 1984. The effect of level of concentrate supplementation given with grass silage during the winter on the total performance of autumn-calving dairy cows. J. Agric. Sci. (Camb.) 102:163–179.
16 Gustafson, G. M., J. Luthman, and E. Burstedt. 1993. Effect of daily exercise on performance, feed efficiency and energy balance of tied dairy cows. Acta Agric. Scand. 43:219–227.
17 Henning, P. H. 1987. The effect of increased energy demand through walking exercise on intake and ruminal characteristics of sheep fed a roughage diet. J. Agric. Sci. (Camb.) 109:53–59.
18 Lamb, R. C., B. O. Barker, M. J. Anderson, and J. L. Walters. 1979. Effects of forced exercise on two-year-old Holstein heifers. J. Dairy Sci. 62:1791–1797.
19 Lathrop, W. J., D. D. Kress, K. M. Havstad, D. E. Doornbos, and E. L. Ayers. 1988. Grazing behaviour of rangeland beef cows differing in milk production. Appl. Anim. Behav. Sci. 21: 315–327.
20 Lawrence, P. R., and R. J. Stibbards. 1990. The energy cost of walking, carrying and pulling loads on flat surfaces by Brahman cattle and swamp buffalo. Anim. Prod. 50:29–39.
21 Levieux, D. 1991. Dosage des IgG du lait de vache par immunodiffusion radiale semi-automatisée, pour la détection du colostrum, des laits de mammites ou de fin de gestation. I. Mise au point du dosage. Lait 71:327–338.
22 Matthewman, R. W., J. Merrit, A. J. Smith, P. Phillips, and J. D. Oldham. 1989. Effects of exercise on lactational performance in cattle. Proc. Nutr. Soc. 48:92A.
23 Matthewman, R. W., J. D. Oldham, and G. W. Horgan. 1993. A note on the effect of sustained exercise on straw intake in lactating cattle. Anim. Prod. 57:491–494.
24 Mendez, D. G., O. N. Di Marco, and P. M. Corva. 1996. Energy expenditure of cattle walking on a flat terrain. Anim. Sci. 63: 39–44.
25 Metz, J.H.M. 1984. The reaction of cows to a short-term deprivation of lying. Appl. Anim. Behav. Sci. 13:301–307.
26 Paape, M. J., A. J. Kral, C. Desjardins, W. D. Schultze, and R. H. Miller. 1973. Failure or either corticosteroids, or ACTH to increase the leucocyte concentration in milk. Am. J. Vet. Res. 34:353–356.
27 Pearson, R. A., and R. F. Archibald. 1989. Biochemical and haematological changes associated with short periods of work in draught oxen. Anim. Prod. 48:375–384.
28 Poutrel, B., J. P. Caffin, and P. Rainard. 1983. Physiological and pathological factors influencing bovine serum albumin content of milk. J. Dairy Sci. 66:535–541.
29 Ribiero, J. M., J. M. Brockway, and A.J.F. Webster. 1977. A note on the energy cost of walking in cattle. Anim. Prod. 25: 107–110.
30 Rook, J.A.F., and J. V. Wheelock. 1967. Reviews of the progress of dairy science. Section C. Dairy Chemistry. The secretion of water and of water-soluble constituents in milk. J. Dairy Res. 34:273–287.
31 Rowland, S. J. 1938. The determination of the nitrogen distribution in milk. J. Dairy Res. 9:42–46.
32 SAS® User's Guide: Statistics, Version 5 Edition. 1987. SAS Inst., Inc., Cary, NC.
33 Stobbs, T. H., and D. J. Brett. 1974. Milk yield and composition of milk and blood as indicators of energy intake by Jersey cows. Aust. J. Agric. Res. 25:657–666.
34 Thomson, N. A., and M. L. Barnes. 1993. Effect of distance walked on dairy production and milk quality. Proc. N.Z. Soc. Anim. Prod. 53:69–72.
35 Vérité, R., and J. L. Peyraud. 1989. Protein: the PDI system. Pages 33–47 *in* Ruminant Nutrition. Recommended Allowances and Feed Tables. R. Jarrige, ed. Inst. Natl. Rech. Agron. and J. Libbey Eurotext, London, England.
36 Vermorel, M. 1989. Energy: the feed unit systems. Pages 23–32 *in* Ruminant Nutrition. Recommended Allowances and Feed Tables. R. Jarrige, ed. Inst. Natl. Rech. Agron. and J. Libbey Eurotext, London, England.
37 Wegner, T. N., J. D. Schuh, F. E. Nelson, and G. H. Stott. 1974. Effect of stress on blood leucocyte and milk somatic cell counts in dairy cows. J. Dairy Sci. 59:949–956.
38 Zerbini, E., A. G. Wold, and D. Demissie. 1996. Effect of draught force and diet on dry matter intake, milk production and live-weight change in non-pregnant and pregnant cows. Anim. Sci. 62:225–231.
39 Zhang, S. H., D. P. Hennessy, P. D. Cranwell, D. E. Noonan, and H. J. Francis. 1992. Physiological responses to exercise and hypoglycaemia stress in pigs of differing adrenal responsiveness. Comp. Biochem. Physiol. 103A:695–703.