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Feeding corn and barley concentrates to grazing dairy cows. Milk production, plasma metabolite, responses to insulin and glucose challenges to β-adrenergic stimuli *

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Summary — Pastures with high degradable protein content may induce insulin resistance, reduced insulin release and high responses to β-adrenergic signals. Twenty-seven multiparous lactating Holstein cows (85 ± 17 days postpartum) grazing oat *(Avena sativa)* sward and rye grass *(Lolium perenne)* pasture were fed pasture alone (PA) or pasture supplemented with corn (CO) or barley (BA) based concentrates. The experiment was conducted at the Agricultural Research Station of Balcarce (INTA), Argentina (37°45'S, 58°18'W). The experimental period lasted 36 days, from 2 July to 6 August. On a dry matter (DM) basis, concentrates contained CO (75%) or BA (80%), soybean meal (19.6% for CO and 5.6% for BA), sunflower meal (2.8% for CO and 11.8% for BA) and a vitamin-mineral complex (2.6%). Cows consumed concentrates at a rate of 6.33 and 5.31 kg cow^{-1} day^{-1} for CO and BA respectively. Total herbage allowance ranged from 21.07 to 29.82 kg DM cow^{-1}. Total pasture DM intake was higher ($P < 0.05$) in the PA treatment (11.93 kg cow^{-1} day^{-1}) than that of cows fed either the CO (11.02 kg cow^{-1} day^{-1}) or the BA (10.75 kg cow^{-1} day^{-1}) based concentrates. The milk production of cows was lower in the PA treatment (17.0 kg cow^{-1} day^{-1}) compared to CO (21.5 kg cow^{-1} day^{-1}) but not to BA (18.4 kg cow^{-1} day^{-1}) based concentrates ($P < 0.05$). Milk yield from cows receiving the CO or the BA based concentrates did not differ ($P < 0.05$). The fat content of milk from cows receiving the BA concentrate (28.4 g kg^{-1}) was lower than that of unsupplemented cows (PA = 34.5 g kg^{-1}) ($P < 0.05$) but not from cows receiving the CO based concentrate (30.1 g kg^{-1}). Milk fat content did not differ between cows fed the CO compared to unsupplemented PA cows ($P > 0.05$). Milk protein or lactose content did not differ significantly between treatment groups. Milk protein yield was higher in the CO ($P < 0.05$) treatment. Live weight gain was higher in CO concentrate (0.532 kg cow^{-1} day^{-1}) compared to PA (–0.035 kg cow^{-1} day^{-1}) and similar between PA and BA (0.381 kg cow^{-1} day^{-1}) or the two concentrates ($P < 0.05$). Plasma levels of glu-

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cose (G), triglyceride (TG) and non-esterified fatty acids (NEFA) were not affected by treatments ($P > 0.05$). Uraemia was lower in BA (21.2 mg dL^{-1}) compared to PA (24.2 mg dL^{-1}) but not to CO (22.3 mg dL^{-1}) treatment ($P < 0.05$). Plasma levels of insulin (INS) were higher in the BA (8.89 μU mL^{-1}) compared to PA (6.19 μU mL^{-1}) and CO (6.63 μU mL^{-1}) treatments ($P < 0.05$). Plasma INS was negatively correlated to milk yield (–0.354, $P < 0.01$) and to milk fat content (–0.374, $P < 0.01$). Plasma G responses to INS challenge were not affected by treatments ($P < 0.52$). Plasma NEFA concentrations 30 min after INS challenge were lower than the pre-injection levels ($P < 0.05$) but were not affected by treatments ($P > 0.05$). The antilipolytic effect of INS was no longer observed 60 min after the injection ($P > 0.05$) and NEFA concentrations 90 min after INS challenge resulted in higher than basal levels ($P < 0.05$). G concentrations 5, 10 and 15 min after glucose challenge were higher than pre-injection levels ($P < 0.05$) but similar between treatments. Calculated G concentration inmediately after injection (131.9, 130.7 and 141.9 mg dL^{-1}), G distribution volume (40.2, 41.6 and 40.3 L) and the fractional rate of G clearance (k) (–0.041; –0.045 and –0.059) did not differ between PA, CO and BA treatments, respectively ($P > 0.05$). Decreases in NEFA after G challenge ($P < 0.60$) and plasma INS responses to G challenge ($P > 0.05$) did not differ between treatments. The surface under the INS curve tended to be lower ($P < 0.09$) with CO (304 μU min^{-1}) compared to PA (417 μU min^{-1}) and BA (469 μU min^{-1}). The fractional rate of INS clearance tended to be lower in PA (–0.02794) compared to CO (–0.04566) and BA (–0.05848) treatments. Higher β-adrenergic responses, resistance to INS action, alterations in G metabolism or in the INS secretory response to G did not differ between unsupplemented cows exposed to higher ammonia absorption and cows receiving concentrates with different rumen starch degradability.

milk yield / corn / barley / plasma metabolites / glucose challenge / β-adrenergic responses

Résumé — Concentrés à base de maïs et d'orge pour vaches laitières. Production de lait, métabolites sanguins, réponses β-adrénergiques aux injections d'insuline et de glucose. Les fourrages contenant des matières azotées qui sont fortement et rapidement dégradées dans le rumen, peuvent induire une résistance à l'action de l'insuline, une diminution de la libération pancréatique de l'hormone ainsi que des réponses β-adrénergiques élevées. Vingt-sept vaches laitières multipares de race Holstein (85 ± 17 jours en lactation) en conditions de pâturage (prairies d'avoine *(Avena sativa)* et de ray-grass *(Lolium perenne)* ont été réparties dans trois traitements : fourrage seul à volonté (PA), fourrage à volonté plus un concentré à base de maïs (CO) et fourrage à volonté plus un concentré à base d'orge (BA). L'expérimentation, qui a été réalisée à la station de recherche agronomique de l'INTA de Balcarce, Argentine (37° 45' Sud, 58° 18' Ouest), a duré 36 jours, du 2 juillet jusqu'au 6 août. Sur la base de la matière sèche (MS), les aliments concentrés ont été constitués de grains de maïs (75 %) ou d'orge (80 %), de farine de soja (19,6 % dans CO et 5,6 % dans BA), de farine de tournesol (2,8 % dans CO et 11,8 % dans BA) et d'un mélange de vitamines et de minéraux (2,6 %). L'ingestion du concentré a été de 6,33 kg $vache^{-1}$ $jour^{-1}$ dans CO et de 5,31 kg $vache^{-1}$ $jour^{-1}$ dans BA. La disponibilité totale du fourrage était de 21,07 à 29,82 kg MS $vache^{-1}$. L'ingestion totale de MS provenant du fourrage a été plus élevée ($p < 0,05$) dans le traitement PA (11,93 kg $vache^{-1}$ $jour^{-1}$) que dans les traitements CO (11,02 kg $vache^{-1}$ $jour^{-1}$) et BA (10,75 kg $vache^{-1}$ $jour^{-1}$). La production laitière a été plus faible dans le traitement PA (17,0 kg $vache^{-1}$ $jour^{-1}$) que dans le traitement CO (21,5 kg $vache^{-1}$ $jour^{-1}$) ($p < 0,05$). Il n'y a pas eu de différences significatives entre BA (18,4 kg $vache^{-1}$ $jour^{-1}$) et les deux autres traitements ($p > 0,05$). Le taux butyreux des vaches recevant le concentré à base de BA (28,4 g kg^{-1}) a été plus faible que celui des vaches non supplémentées (PA = 34,5 g kg^{-1}) ($p < 0,05$). Il n'y a pas eu de différences de taux butyreux entre le traitement CO (30,1 g kg^{-1}) et les autres ($p > 0,05$). Le taux protéique et les teneurs en lactose n'ont pas varié. Le rendement en protéines a augmenté dans le traitement CO ($p < 0,05$). L'augmentation du poids vif a été plus élevée dans CO (0,532 kg $vache^{-1}$ $jour^{-1}$) que dans le traitement PA (–0,035 kg $vache^{-1}$ $jour^{-1}$). Il n'y a pas eu de différences entre PA et BA (0,381 kg $vache^{-1}$ $jour^{-1}$) ou entre les deux traitements avec supplémentation ($p < 0,05$). Les teneurs plasmatiques en glucose (G), en triglycérides (TG) et en acides gras non-estérifiés (NEFA) n'ont pas été modifiées ($p > 0,05$). Les teneurs plasmatiques en urée ont été plus faibles dans le traitement BA (21,2 mg dL^{-1}) que dans le traitement PA (24,2 mg dL^{-1}) sans changement par rapport au traitement CO (22,3 mg dL^{-1}) ($p < 0,05$). Les teneurs plasmatiques en insuline (INS) ont été significativement augmentées dans le

lot BA (8,89 μU mL^{-1}) par rapport aux lots PA (6,19 μU mL^{-1}) et CO (6,63 μU mL^{-1}) ($p < 0,05$). Les teneurs plasmatiques d'insuline ont été négativement associées à la production laitière (–0,354, $p < 0,01$) et au taux butyreux (–0,374, $p < 0,01$). Les variations de concentration de G après une injection intrajugulaire d'insuline n'ont pas été affectées par les traitements ($p < 0,52$). Les teneurs plasmatiques des NEFA 30 minutes après l'injection d'insuline ont été plus faibles que les valeurs avant l'injection ($p < 0,05$) mais n'ont pas été affectées par les traitements ($p > 0,05$). L'effet antilipolytique de l'insuline n'est plus observé 60 minutes après l'injection de l'hormone ($p > 0,05$) et les teneurs en NEFA 90 minutes après l'injection d'insuline ont été accrues par rapport aux concentrations avant l'injection ($p < 0,05$). Les teneurs plasmatiques en G, 5, 10, et 15 minutes après une injection intrajugulaire de G ont été significativement augmentées par rapport aux concentrations pré-injection ($p < 0,05$). L'hyperglycémie induite reste la même entre les traitements ($p > 0,05$). Les valeurs calculées de concentration plasmatique de G immédiatement après l'injection (131,9 mg dL^{-1}, 130,7 mg dL^{-1} et 141,9 mg dL^{-1}), le volume de distribution de G (40,2 L, 41,6 L et 40,3 L) et le taux de disparition de G injecté (–0,041, –0,045 et –0,059) n'ont pas été différents entre les traitements PA, CO et BA respectivement ($p > 0,05$). La diminution des teneurs plasmatiques des NEFA ainsi que l'augmentation de l'insulinémie après l'injection de G n'ont pas été différentes entre les traitements ($p > 0,05$). La surface sous la courbe d'insuline a eu tendance à être plus faible ($p < 0,09$) dans le traitement CO (304 μU $minute^{-1}$) que dans les traitements PA (417 μU $minute^{-1}$) et BA (469 μU $minute^{-1}$). Le taux de disparition de l'insuline a montré une tendance à être plus faible dans PA (–0,02794) que dans CO (–0,04566) et BA (–0,05848). Des réponses accrues aux stimuli de type β-adrénergique, des phénomènes de résistance à l'action de l'insuline, des altérations dans les paramètres associés au métabolisme du glucose ou dans l'hyperinsulinémie induite par le glucose n'ont pas été observées entre les vaches non supplémentées et exposées à une absorption majeure d'ammoniac, et les vaches supplémentées avec des sources d'amidon de différente dégradabilité dans le rumen.

production laitière / maïs / orge / métabolites sanguins / injection de glucose / réponses β-adrénergiques

INTRODUCTION

In the Argentine dairy production systems, a high proportion of nutrients comes from grazed temperate pastures, and cereal based concentrates are often used to correct pasture imbalances. One of the most frequent imbalances observed in autumn–winter forages is a high crude protein (CP) content with a rapid rate of rumen dissimilation of that CP. This is frequently coupled to a low non-structural carbohydrate (NSCHO) content in the forage. In this situation, considerable loss of the nitrogen-containing moieties may occur in the rumen, due to their availability largely exceeding that of a source of energy like NSCHO. The dietary protein is then largely destroyed by rumen microorganisms and turned into ammonia (NH_3). It has been reported that cattle fed such diets have exibited reduced growth rate and milk production with a high rumen NH_3 concentration of 160–600 mg L^{-1} (Rearte and Santini, 1989).

A lack of synchronization between availability of nitrogen and energy in the rumen means that substantial amounts of NH_3 are absorbed and conducted to the liver where it is converted into urea. Increased supply of free NH_3 does not seem to affect the activity of the urea cycle enzymes. As a result, the portal supply of NH_3 may exceed the capacity of the urea cycle (Madsen, 1983) and subclinical NH_3 toxicity may develop (Symonds et al, 1981; Beever, 1993).

Elevated blood NH_3 concentrations may induce derangements in intermediary metabolism like hyperglycaemia, increased plasma non-esterified fatty acids (NEFA) concentration, underutilization of glucose, insulin resistance, reduced insulin secretion, and enhanced responses to β-adrenergic sig-

nals (Feldman and Lebovitz, 1971; Symonds et al, 1981; Visek, 1984; Fernández et al, 1988; Gagliostro et al, 1994).

Supplementation with corn and barley based concentrates (degradable starch) can provide a more synchronized supply of readily available carbohydrates, reducing the risk of hepatic NH_3 overload and subclinical NH_3 toxicity.

Attention has focused on formulating diets where NSCHO and nitrogen should be present simultaneously in the rumen in order to improve microbial growth and milk yield. However, there is a lack of a clear zootechnical benefit when a rapidly degradable starch (eg, barley grain) is replaced by a slowly degradable starch (eg, maize grain) to enhance milk yield or to change its composition (Nocek and Tamminga, 1991; Sauvant and van Milgen, 1995).

The objectives of this experiment were: 1) to test the hypothesis that a concentrate containing a source of high rumen degradable NSCHO (barley grain) may support a greater milk production than one with a less rumen degradable NSCHO present (maize grain); and 2) to characterize the responses to insulin and β-adrenergic signals and to investigate the possible alterations in glucose metabolism or in the insulin secretory response to glucose between unsupplemented grazing dairy cows exposed to higher NH_3 absorption and grazing cows receiving concentrates with different rumen starch degradability.

MATERIALS AND METHODS

The experiment was conducted during winter, at the Agricultural Research Station of Balcarce (INTA, 37 ° 45'S, 58° 18'W) in Argentina, with 10 days of pretreatment (21 to 30 June) and 36 days of treatment (from 2 July to 6 August).

Twenty-seven multiparous Holstein cows were assigned to three treatments (each of nine cows) on the basis of their milk production for the 10 days before allocation. At the start of the experiment, the cows were 85 (± 17) days into lactation, weighing 537 (± 67) kg. The cows grazed pasture alone (PA) or were supplemented with barley (BA) or corn (CO) based concentrates. On a dry matter (DM) basis, concentrates contained corn (75%) or barley (80%) grains, soybean meal (19.6% for CO and 5.6% for BA), sunflower meal (2.8% for CO and 11.8% for BA) and a vitamin-mineral complex (2.6%). Supplemental protein sources were varied in order to obtain isoproteic concentrates. Components were pelleted and concentrates (87% DM) were offered at a rate of 7 kg cow^{-1} day^{-1} in two equal feeds at milking times (06.00 and 15.00 hours) in individual feeding stalls. The amount of concentrates fed and refusals were recorded daily. Concentrates were analysed for in vitro DM digestibility (IVDMD), CP, neutral detergent fibre (NDF) and starch.

Cows strip-grazed an oat (*Avena sativa*) sward (between the morning and the afternoon milking times) and a rye grass (*Lolium perenne*) pasture (after the afternoon milking time). The amount of each pasture on offer before grazing was estimated once a week and the grazing surface was calculated in order to obtain a total DM allowance of 25 kg cow^{-1} day^{-1}. Herbage mass was calculated using the regression equations between sward height and the herbage mass. The equations were obtained for the pre- and post-grazing strips by cutting a limited number of quadrats to ground level to correlate with the sward height.

Pasture intake was measured four times during the experiment (13, 20, 26 July and 2 August) by the difference in the herbage mass measured at the beginning and at the end of a period of 24 h of grazing (Meijs et al, 1982). In order to obtain a normal grazing behaviour, measurements were carried out using three grazing strips with three groups of three cows in each treatment. One hundred measures of sward height were taken before and after grazing in each grazing strip to estimate herbage mass as described earlier. In order to assess the quality of the grazed herbage, samples were obtained by hand plucking at the grazing height during each intake measurement. They were analysed for DM, IVDMD, CP, NDF and NSCHO.

Milk yield was recorded daily during the experiment. The fat, protein and lactose contents of milk were determined from samples obtained on 2 consecutive days during each experimental week using a Foss 605 Milko-Scan (Foss Electric, Hillerod, Demmark). Cows were weighed

immediately after the morning milking at the beginning and at the end of the experiment.

Blood samples from jugular vein were taken at 08.00 hours once a week during the 5 experimental weeks. In the second week of the experiment, preprandial (08.00 hours) blood samples were taken before (INS-0) and 30 (INS-30), 60 (INS-60) and 90 (INS-90) min after an intravenous bovine insulin (INS) challenge (0.12 U kg body weight [BW]$^{-1}$; Betasint, Beta Laboratory, Buenos Aires, Argentina). In week 3, samples were taken before (ISO-0) and 15 min after (ISO-15) an isoproterenol (ISO) challenge (4 nmol kg BW^{-1}, Proterenal, Phoenix Laboratory, Buenos Aires, Argentina) and in week 4, before (G-0) and 5 (G-5), 10 (G-10) and 15 (G-15) min after an intravenous glucose (G) challenge (100 mg kg BW^{-1}). Fractional rate of G and INS clearance *(k)* was calculated as the slope of the regression of time on lnC where C = G (or INS) concentration at × 5, 10 and 15 – G-0 G (or INS) concentration. The intercept (Co) represented the G (or the INS) concentration inmediately after the end of the G injection. The distribution volume for G (VOL-G) was calculated as the ratio between the injected G mass and Co (Ghrön et al, 1987). Surfaces under the INS or the G curves were calculated from the concentration curves of G and INS between 0 and 15 min after G injection.

Blood was collected into a heparinized flask (5 U/mL blood) and plasma was immediately obtained by centrifugation (2 000 *g* for 15 min) and stored at –24°C until analysis. Commercial enzymatic kits were used for G (Wiener Laboratory, Rosario, Argentina), NEFA (NEFA C-Test, Wako, TX, USA) and urea (Wiener Laboratory, Rosario, Argentina). Plasma INS was determined by radioimmunoassay with separation by means of immunoprecipitating reagent (standards used were 10, 22, 50, 100 and 200 μU/mL calibrated against WHO 66/304).

Milk yield, milk composition, pasture intake, plasma metabolites and INS were analysed using a split-plot design with treatment as main plot, week as secondary plot and the corresponding interaction. Live weight gain (LWG) (calculated as the difference between live weight at the end and at the beginning of the experiment), surfaces under the INS and the G curves, *k* values for INS and G, Co and VOL-G values were analysed by one-way analysis of variance. Responses to INS, ISO and G were analysed using a split-plot design with treatment as main plot, sampling time as the secondary plot and the corresponding interaction. Differences between means for treatment, week or the interaction effects were determined using the Newman-Keuls test ($P < 0.05$).

RESULTS

CO and BA based concentrates were consumed at a rate of 6.33 and 5.31 kg cow^{-1} day^{-1}, respectively, which represented about 2.9 and 2.5 kg cow^{-1} day^{-1} of starch. Values for IVDMD, CP and NDF content of the concentrates (% DM) were respectively 86.1 and 92.4; 15.8 and 18.4 and 20.1 and 13.9 for BA and CO.

Values of herbage mass in the pre-grazing strips and the quality of the forage apparently consumed by the cows are presented in table I. Herbage allowance and forage DM intake for the oat sward and the rye grass pasture are presented in table II. Cows graz-

Table I. Herbage mass and forage quality of the oat sward and the rye grass pasture [1].

	Oat sward	*Rye grass*
Herbage mass (kg DM ha^{-1})[2]	3172 (± 482)	3131 (± 229)
Dry matter (g kg^{-1})	200 (± 18)	247 (± 17)
Crude protein (g kg DM^{-1})	239 (± 48)	159 (± 37)
NDF (g kg DM^{-1})	352 (± 42)	364 (± 17)
IVDMD (%)	81.7 (± 1.9)	79.6 (± 1.6)
NSCHO (g kg DM^{-1})	169 (± 42)	195 (± 29)

[1] Mean (± standard deviation) values of the four intake measurements; [2] pre-grazing strips. NDF: neutral detergent fiber; IVDMD: in vitro dry matter digestibility; NSCHO: non-structural carbohydrate.

Table II. Forage intake in grazing dairy cows (85 ± 17 days postpartum) fed pasture supplemented with corn (CO) (6.33 kg day^{-1}) or barley (BA) (5.31 kg day^{-1}) based concentrates or pasture alone (PA).

	PA	*CO*	*BA*	*RSD*[1]	*Treatment* $P <$	*Week* $P <$	$T \times W$ $P <$
Oat sward							
Herbage allowance (kg DM cow^{-1} day^{-1})	12.79	12.58	12.81	1.14	0.904	0.004	0.745
Utilization coefficient (%)[2]	44.4[a]	39.8[b]	36.0[c]	0.94	0.005	0.043	0.456
Intake (kg DM cow^{-1} day^{-1})	5.75	4.92	4.66	0.82	0.206	0.198	0.574
Rye grass pasture							
Herbage allowance (kg DM cow^{-1} day^{-1})	12.41	12.43	12.85	1.42	0.802	0.019	0.103
Utilization coefficient (%)[2]	49.8	49.0	47.4	4.68	0.649	0.016	0.395
Intake (kg DM cow^{-1} day^{-1})	6.18	6.09	6.10	1.09	0.988	0.051	0.471
Total							
Herbage allowance (kg DM cow^{-1} day^{-1})	25.19	25.00	25.67	2.17	0.833	0.000	0.666
Utilization coefficient (%)[2]	47.3	44.1	41.6	2.07	0.063	0.044	0.325
Intake (kg DM cow^{-1} day^{-1})	11.93[a]	11.02[b]	10.75[b]	0.35	0.038	0.023	0.468

[1] Residual standard deviation for treatment factor; [2] (herbage mass at the beginning of grazing–herbage mass at the end of grazing) / herbage mass at the beginning of grazing × 100. [a b c] Within rows, means with different letters differ ($P < 0.05$). T × W : treatment × sampling time.

ing pasture alone consumed more forage DM than cows supplemented with CO and BA concentrates ($P < 0.05$). The same trend was observed when forage DM intake was expressed relatively to body weight of cows (24.56, 20.80 and 20.22 g DM kg^{-1} BW for PA, CO and BA, respectively, $P < 0.10$). The mean calculated substitution rate was 0.144 and 0.222 kg of forage DM kg^{-1} concentrate fed. There were no differences in total pasture DM intake between the CO and BA groups ($P > 0.05$).

Milk production, milk composition and LWG of cows are presented in table III. The milk production of cows fed the CO based concentrate was significantly higher ($P < 0.05$) than that of unsupplemented cows (PA) but not for cows fed the BA based concentrate. The fat content of milk from cows receiving the BA concentrate was significantly lower than that observed in cows fed PA ($P < 0.05$). There were no significant differences between PA and CO or between CO and BA treatments ($P > 0.05$). The feeding of CO or BA based concentrates did not increase the protein or the lactose content of milk above that of the PA group ($P > 0.05$). Milk fat yield did not differ between treatments but milk protein yield was higher in the CO than in the PA and BA groups ($P < 0.05$). The CO group gained more weight than the PA group ($P < 0.05$). No significant differences in LWG were detected between the supplemented groups or between the PA and the BA groups ($P > 0.05$).

Plasma metabolite and INS concentrations are presented in table IV. There were no significant differences between treatments ($P > 0.05$) in the mean values (5 weeks) for plasma G. The treatment × week interaction ($P < 0.001$) showed higher plasma G concentration for CO (90.5 mg/dL) compared to PA (79.9 mg/dL) and BA (71.9 mg/dL) treatments ($P < 0.05$) in week 1. In week 5, the respective figures were higher in PA (71.9 mg/dL) and in CO (72.5 mg/dL) compared to BA (66.6 mg/dL) ($P < 0.05$). No significant differences between treatments were detected for the mean plasma NEFA and TG concentrations. In spite of the significant treatment × week interaction for triglyceride (TG), there were no differences between treatments within weeks of sampling ($P > 0.05$).

Table III. Milk production, milk composition and live weight gain (LWG) in grazing dairy cows (85 ± 17 days postpartum) fed pasture supplemented with corn (CO) (6.33 kg day^{-1}) or barley (BA) (5.31 kg day^{-1}) based concentrates or pasture alone (PA).

	PA	*CO*	*BA*	*RSD* [1]	*Treatment* $P <$	*Week* $P <$	*T × W* $P <$
Milk (kg cow^{-1} day^{-1})	17.0[a]	21.5	18.4[ab]	6.40	0.025	0.009	0.266
Milk fat (%)	3.45[a]	3.01[ab]	2.84[b]	0.97	0.047	0.005	0.0471
Fat yield (g day^{-1})	587	651	525	289	0.210	0.026	0.035
Milk protein (%)	3.28	3.38	3.31	0.52	0.732	0.299	0.384
Protein yield (g day^{-1})	556[a]	724[b]	610[a]	223	0.017	0.116	0.364
Lactose (%)	4.78	4.84	4.80	0.36	0.748	0.005	0.349
Lactose yield (g day^{-1})	813[a]	1035[b]	883[ab]	325	0.030	0.028	0.198
LWG (g day^{-1})	−35[a]	532[b]	381[ab]	18.9	0.045	–	–

[1] Residual standard deviation for treatment factor. [a b] Within rows, means with different letters differ ($P < 0.05$). T × W : treatment × week.

Table IV. Plasma metabolite and insulin concentrations in grazing dairy cows (85 ± 17 days postpartum) fed pasture supplemented with corn (CO) (6.33 kg day^{-1}) or barley (BA) (5.31 kg day–1) based concentrates or pasture alone (PA)[1].

	PA	*CO*	*BA*	*RSD* [2]	*Treatment* $P <$	*Week* $P <$	*T × W* $P <$
Glucose (mg dL^{-1})	74.8	76.0	72.0	7.7	0.075	0.000	0.001
Urea (mg dL^{-1})	24.2^{a}	22.3^{ab}	21.2^{b}	5.0	0.034	0.000	0.006
Triglyceride (mg dL^{-1})	609	573	613	193	0.582	0.000	0.029
NEFA (μEq L^{-1})	522	462	488	288	0.624	0.000	0.384
Insulin (μU mL^{-1})	6.19^{a}	6.63^{a}	8.89^{b}	3.11	0.041	0.339	0.024

[1] Mean values of the 5 experimental weeks or of weeks 1 and 4 for insulin; [2] residual standard deviation for treatment factor. NEFA: non-esterified fatty acids; T × W: treatment × week.

Plasma urea was higher in the PA than the BA group ($P < 0.05$). Non-significant differences were detected between CO and the other treatments ($P > 0.05$). The treatment × week interaction ($P < 0.006$) showed that in week 3 (PA= 22.3 mg dL^{-1}, BA= 19.0 mg dL^{-1}) and in week 5 (PA= 29.6 mg dL^{-1}, BA= 23.0 mg dL^{-1}) the BA (but not the CO) based concentrate induced lower plasma urea levels compared to PA treatment ($P < 0.05$). Plasma urea and NEFA were correlated in PA ($r = 0.371$; $P < 0.01$) and in CO ($r = 0.317$; $P < 0.05$) but not in BA treatment. When data from the three treatments were pooled, plasma urea and milk yield were negatively correlated ($r = -0.204$; $P < 0.05$).

Plasma levels of INS were higher in BA cows compared to the other two treatments ($P < 0.05$). Significant correlations between plasma INS and metabolites were not detected. When data from the three treatments were pooled, plasma INS was negatively correlated to milk yield ($r = -0.354$; $P < 0.01$) and to milk fat content ($r = -0.374$; $P < 0.01$). Milk protein content tended to increase when plasma INS levels increased up to 10 μU mL^{-1} and then, a decrease was observed (data not shown).

Neither treatment ($P < 0.516$) nor interaction ($P < 0.928$) effects were detected in the decrease in plasma G concentration after a single intravenous INS injection. Average G values for the three treatments were 76.9 mg dL^{-1} (INS-0), 38.1 mg dL^{-1} (INS-30), 50.1 mg dL^{-1} (INS-60) and 63.3 mg dL^{-1} (INS-90). The hypoglycaemic action of INS lasted up to 90 min after the hormone injection ($P < 0.05$).

Decreases in plasma NEFA concentration were observed at 30 min after INS injection and they were not affected by concentrate feeding ($P < 0.466$). Treatment × time of sampling interaction was not detected ($P < 0.13$). Mean values from all treatments showed that the antilipolytic effect of INS was not observed longer at INS-60 samples (348 μEq L^{-1}) ($P > 0.05$) and the mean plasma NEFA concentration observed at INS-90 (414 μEq L^{-1}) was higher than the mean basal INS-0 values of 349 μEq L^{-1}. However, this lipomobilization effect at INS-90 was only observed in the PA treatment (+150; $P < 0.01$).

Plasma NEFA and G responses to isoproterenol injection are presented in table V. In both metabolites, neither the pre-injection plasma levels nor the concentration

Table V. Changes in blood plasma glucose and non-esterified fatty acid (NEFA) concentrations after isoproterenol (ISO) injection [1] in grazing dairy cows (85 ± 17 days postpartum) fed pasture supplemented with corn (CO) (6.33 kg day^{-1}) or barley (BA) (5.31 kg day^{-1}) based concentrates or pasture alone (PA).

	PA	*CO*	*BA*	*Mean*
Glucose (mg dL^{-1})				
ISO-0	74.7	71.8	70.0	72.2[a]
ISO-15	84.3	81.3	80.0	81.9[a]
NEFA (μEq L^{-1})				
ISO-0	558	412	364	445[b]
ISO-15	906	722	770	799[b]

[1] 4 nmol kg^{-1} body weight intravenously. Treatment × sampling hour interaction was not observed either for glucose ($P < 0.964$) or NEFA ($P < 0.542$). [ab] Without columns, means with different letters differ ($P < 0.05$).

increase after isoproterenol injection were affected by treatments ($P > 0.10$). Interaction between treatment and sampling hour were not detected. Similar conclusions were obtained when data were analysed as the differences between ISO-15 minus ISO-0 concentrations for G ($P < 0.965$) and NEFA ($P < 0.541$).

Neither treatment ($P < 0.74$) nor interaction ($P < 0.347$) effects were detected for the increase in plasma G concentration after a single intravenous G injection. Average G values from the three treatments showed increases ($P < 0.05$) in G concentration of 48.8, 38.7 and 31.4 mg dL^{-1} in G-5, G-10 and G-15 samples compared to basal G-0 values of 70.3 mg dL^{-1}. The surface under the G curve was not changed by treatments (PA = 93.9; CO = 88.0; BA = 89.9 mg dL^{-1} min^{-1}, $P < 0.14$). The calculated Co values for G were not affected ($P < 0.56$) by treatments (131.9, 130.7 and 141.9 mg dL^{-1} for PA, CO and BA respectively). The calculated distribution volume for G was 40.22, 41.56 and 40.33 L ($P < 0.91$) and the G k values obtained were –0.041, –0.045 and –0.059 ($P < 0.51$) in PA, CO and BA treatments, respectively.

Decreases in plasma NEFA concentrations after G challenge did not differ between treatments ($P < 0.598$) and interaction between treatment × sampling time was not detected ($P < 0.818$). Decreases in plasma NEFA concentration were observed at 5, 10 and 15 min after G injection compared to G-0 values ($P < 0.0001$ for sampling time). Average plasma NEFA from the three treatments were 418 (G-0), 334 (G-5), 327 (G-10) and 289 (G-15) μEq L^{-1}.

Uraemia was increased at 10 (17.84 mg dL^{-1}) and at 15 (17.73 mg dL^{-1}) min after G injection compared to the basal G-0 values of 16.06 mg dL^{-1} ($P < 0.0001$ for sampling time) without significant differences between treatments ($P < 0.833$). Treatment × sampling time interaction was not significant ($P < 0.359$).

There was no significant difference between treatments in the secretory INS response to G injection ($P < 0.113$). Sampling time was highly significant ($P < 0.001$) and no treatment × sampling time interaction was detected ($P < 0.212$). Average INS values from all treatments were 7.57 μU mL^{-1} (G-0), 32.27 μU mL^{-1} (G-5), 31.74 μU mL^{-1} (G-10) and 23.97 μU mL^{-1} (G-15). The calculated Co values for INS tended to be

higher in BA (50.7 μU mL^{-1}) compared to CO (33.0 μU mL^{-1}) and to PA (39.4 μU mL^{-1}) treatments ($P < 0.11$). The surface under the INS curve tended to be higher in BA (469 μU min^{-1}) compared to CO (304 μU min^{-1}) and to PA (417 μU min^{-1}) treatments ($P < 0.09$). The ratio between the INS and the G areas tended to be higher in BA (1.97) compared to the CO (1.66) and to PA (1.24) treatments ($P < 0.11$). The estimated k values for INS were –0.0279, -0.04566 and –0.05848 in PA, CO and BA treatments respectively ($P < 0.533$).

DISCUSSION

In this experiment, forage fibre content did not exceed the values considered critical for maximum intake and milk production (ie, 500–550 g NDF kg^{-1} DM; Paterson et al, 1994) and pasture CP content fell within the range of 150–250 g CP kg^{-1} DM proposed by Minson (1990) to obtain high values of forage DM digestibility. The observed coefficients of IVDMD of the forage apparently consumed by cows were indeed very high (table I). These data suggest that forage quality was adequate. Forage DM intake was relatively low (2.33% of BW in PA treatment) and the depressing effects of concentrate feeding on forage DM intake were low (table II). The mean substitution rates observed were low (0.14–0.22) compared to the values of 0.40-0.60 observed by Meijs (1981) when 2–4 kg of concentrate were fed to lactating dairy cows. These results could be linked to low values of DM allowance because total herbage allowance and forage DM intake were positively correlated ($r = 0.643$, $P < 0.001$). When pasture intake is low reduced substitution rate coefficients are obtained (Meijs, 1981; Meijs and Hoekstra, 1984). The lack of differences in pasture DM intake between the cows fed the CO and BA based concentrates could be explained in part by the absence of any effect of type of supplement on kinetics of cell wall degradation in the rumen, as was observed by Kloster et al (1994b) using the same type of concentrates.

The higher milk production observed in cows fed the CO based concentrate compared to PA could not be explained by a higher milk lactose content (table III). As the forage intake was only slightly depressed in CO fed cows compared to PA cows (–0.91 kg DM cow^{-1} day^{-1}; table II), the higher milk yield observed could be explained by a higher total energy intake of cows. When energy intake is enhanced by concentrate feeding, an increase in milk yield could be expected (Rémond, 1985; Oldham and Emmans, 1988; Nocek and Tamminga, 1991). The lack of differences in milk yield between BA and PA treatments could be explained by the lower DM intake of the BA concentrate (–15.3% compared to CO) and also by the lower net energy (NE) content of the BA concentrate (–10.1% compared to CO) that can be calculated from feed tables (Inra, 1988). The difference in NE intake between BA and CO fed cows arising from the lower DM intake of concentrate observed can be calculated to be of 23.8%. The lower intake of concentrate observed in BA could be explained by a lower palatability of the barley as reported in other studies using complete mixed rations (Casper and Schingoethe, 1989; McCarthy et al, 1989; Casper et al, 1990). The absence of significant differences in milk yield between the BA and the CO based concentrates is in agreement with the observation that there is a lack of a clear zootechnical benefit of replacing a rapidly degradable starch (eg, barley grain) by a slowly degradable one (eg, maize grain) to enhance milk yield (Nocek and Tamminga, 1991; Sauvant and van Milgen, 1995). Other experimental results obtained seem to confirm this observation (DePeters and Taylor, 1985; de Visser et al, 1990; Grings et al, 1992; Kloster et al, 1994a; Gagliostro et al, 1996). The milk fat content was sharply decreased

in the BA respect to the PA treatment (–6.1 g kg^{-1}, $P < 0.01$). BA based supplements have had either no effect on (DePeters and Taylor, 1985; Casper and Schingoethe, 1989; Weiss et al, 1989; Grings et al, 1992; Kloster et al, 1994a; Gagliostro et al, 1996) or have decreased (Herrera-Saldana and Huber, 1989; Casper et al, 1990) milk fat. The negative effects on milk fat were sometimes followed by a higher rumen propionate proportion (Casper et al, 1990) or by a lower rate of acetate to propionate in the rumen (Herrera-Saldana and Huber, 1989), although higher rumen propionate levels without a negative effect on milk fat were also observed (McCarthy et al, 1989; Weiss et al, 1989). Rumen-derived propionate increases hepatic gluconeogenesis and plasma insulin concentrations which may promote the utilization of nutrients (acetate, G, D-3-hydroxybutyrate, TG) by other tissues (muscles and adipose tissue) rather than mammary tissue (Chilliard, 1987). The rumen acetate/propionate ratio was not measured, but the plasma insulin levels were higher in BA fed cows (table IV) and plasma insulin was negatively correlated to milk fat. The sharp decrease in the milk fat content of cows receiving the BA based concentrate should be an important factor to be considered in formulating diets with a source of rapidly and highly rumen degradable starch.

As observed here (table III), as well as in previous reports (DePeters and Taylor, 1985; Herrera-Saldana and Huber, 1989; McCarthy et al, 1989; Weiss et al, 1989; Casper et al, 1990; Grings et al, 1992; Kloster et al, 1994a; Gagliostro et al, 1996), milk protein content does not seem to be affected when CO is replaced by BA in the diet of lactating dairy cows.

Ruminal NH_3 concentration is the main source of variation in plasma urea levels. In this experiment, NH_3 concentration ranged from 5 to 26 mg dL^{-1} (measured with six additional fistulated cows in a latin square design) without differences ($P < 0.15$) in the mean values for PA (15.7 mg dL^{-1}), CO (14.1 mg dL^{-1}) and BA (15.7 mg dL^{-1}) (unpublished results). The observed capacity of the BA based concentrate to decrease uraemia (table IV) may be important in grazing situations where extensive forage protein degradation and high rumen NH_3 concentrations are observed. The absence of significant differences between CO and BA in uraemia is in accordance with other studies conducted indoors (Casper and Schingoethe, 1989; Casper et al, 1990) or in grazing conditions (Gagliostro et al, 1996) and could be explained by the lack of effect of type of concentrate on ruminal NH_3 (Kloster et al, 1994b; Gagliostro et al, 1996) and on kinetic parameters of protein digestion in the rumen (Kloster et al, 1994b). The lack of differences in basal (table IV) or stimulated (table V) plasma NEFA levels suggests that the cows fed only PA were not in negative energy balance in spite of the difference observed in LWG (table III).

In our experiment, supplementation of the forage diet with the BA or the CO based concentrates was practiced in an attempt to correct a deficiency in NSCHO that is usually and spontaneously observed in fresh forages during the autunm–winter seasons and to prevent the potential risk of an excess of NH_3 absorption from the rumen.

Glucose (Soar et al, 1973; Spires and Clark, 1979; Symonds et al, 1981; Visek, 1984; Fernández et al, 1988) and NEFA (Visek, 1984; Fernández et al, 1988) are elevated when liver detoxification capacity of NH_3 is exceeded and NH_3 can reach systemic circulation. In Hereford steers, plasma glucose concentrations exhibited an increase above pre-injection levels (12%) when ammonium chloride was infused into the jugular vein (Fernández et al, 1988). In dairy cows, blood glucose concentrations increased up to 98.8 mg dL^{-1} when ammonium acetate was infused via a mesenteric vein and uraemia reached values of

42 mg dL^{-1} (Symonds et al, 1981). In our experiment, no significant differences between treatments ($P > 0.05$) were observed in the mean values of the 5 experimental weeks for plasma glucose (table IV). It has been suggested that the hyperglycaemia associated with hyperammonemia can apparently be explained by the combined effects of a reduced utilization of glucose by insulin-sensitive extrahepatic tissues, a reduced insulin secretion by the pancreatic β-cells and an enhanced hepatic glycogenolysis (Spires and Clark, 1979; Symonds et al, 1981; Visek, 1984; Fernández et al, 1988). Concerning the former, the lack of differences in the hypoglycemic effects of insulin suggested that the responsiveness of peripheral glucose utilization to INS was similar between forage-fed cows and cows fed the CO or BA based supplements. The calculated Co values of glucose and the peak in glucose plasma concentrations obtained 5 min after the injection of glucose reflected a similar extent of induced hyperglycaemia in the cows exposed to higher NH_3 absorption and in the supplemented cows. Moreover, there were no significant differences between the glucose disappearance rate, expressed as *k*. Changes in the rate of glucose disposal after exogenous glucose administration would have reflected alterations in peripheral glucose utilization (Sasaki et al, 1984). The similar calculated distribution volume between treatments may have also been reflecting an equal ability of the injected glucose to reach and penetrate into the intracellular compartments. The lack of differences in the areas under the glucose curve adds additional support to speculate an absence of any sign of insulin resistance or alterations in glucose metabolism in the PA cows and hence exposed to a higher NH_3 absorption from the rumen. Plasma glucose increased 12% during ammonium chloride infusion in steers and remained elevated 180 min post-infusion in spite of the hyperinsulinemia observed during this time, probably reflecting insulin resistance after the infusion period (Fernández et al, 1988).

It has been demonstrated that the ammonium ion alters in vitro insulin secretion at concentrations normally found in blood and that this ion may modulate insulin release in physiological states (Feldman and Lebovitz, 1971). Concerning the hyphothesis of a reduced pancreatic ability to secrete insulin in cattle exposed to high amounts of NH_3 absorption, we have not observed either a significant lower increase in the plasma insulin concentration after glucose injection or a significant lower insulin area, Co values or changes in the rate of insulin disposal after exogenous glucose administration in PA cows ($P > 0.05$). Nevertheless, it is important to note that the basal insulin concentration was higher in the BA group (table IV) and the areas under the insulin curve ($P < 0.09$), the calculated insulin concentration inmediately after the end of the glucose injection ($P < 0.11$) and the insulinogenic index (ratio between the insulin and the glucose areas) ($P < 0.11$) tended to be higher in the BA treatment where the lower values of uraemia were observed (table IV). Plasma urea increased 39% and plama insulin concentration decreased 26 to 46% during ammonium chloride infusion in steers (Fernández et al, 1988). The increase in insulin secretion in ruminants is markedly stimulated by feeding, being greater with high-concentrate than with high-roughage diets (Chilliard, 1987; Sasaki et al, 1984). Glucose injection was given here in preprandial conditions in order to minimize the insulinotropic effects of volatile fatty acids (mainly propionate) that may have complicated the interpretation of the insulin secretory response to glucose injection between diets. Inflow of glucose into the glucose pool may occur from both gluconeogenesis and glycogenolysis. The similarity extent in the plasma glucose levels induced by isoproterenol injection (table V) suggested that an enhanced hepatic glycogenolysis was probably not occurring in the unsupple-

mented cows. Concerning gluconeogenesis, it was also postulated that NH_3 overload decreases hepatic glucose production from propionate (Weekes et al, 1978; Aiello et al, 1985; Demigné et al, 1986b; Rémésy et al, 1986). Although propionate was injected (100 mg kg^{-1} BW; Demigné et al, 1986a; Ghrön et al, 1987) into the jugular vein of all cows it did not yield hyperglycaemia in any treatment at either 60 min (71.9 mg dL^{-1}) or 90 min (70.9 mg dL^{-1}) after propionate injection compared to basal (pre-injection) glucose values of 70.4 mg dL^{-1} ($P < 0.703$ for hour of sampling) (results not presented).

The liver plays a critical role for the maintenance of non-toxic concentrations of NH_3 in the plasma and such a function requires high quantities of energy to be expended (Rémésy et al, 1986). The increase in uraemia 10 and 15 min after the glucose injection suggests that the formation of urea may have been limited by energy and that the portal supply of NH_3 may have not been exceeding the capacity of the urea cycle. It has been demonstrated that propionate availability stimulates urea production in sheep hepatocytes (Rémésy et al, 1986), but in our experiment, uraemia was not increased at 60 min after intrajugular propionate injection (26.9 mg dL^{-1}) compared to the pre-injection plasma urea levels (26.8 mg dL^{-1}) ($P < 0.988$) (results not shown).

Total catecholamines tended to increase with subclinical NH_3 toxicity (Fernández et al, 1988) and this may account for the higher basal (Fernández et al, 1988) or stimulated (Gagliostro et al, 1994) plasma NEFA concentrations observed during hyperammonemia. In our experiment, plasma catecholamine levels have not been measured but higher basal or stimulated (ISO) plasma NEFA levels were not observed in the PA treatment. This result was consistent with the lack of treatment effect on the antilipolytic effects of insulin and glucose and with the absence of changes in the hypoglycaemic effects of insulin because a reduction in adipose tissue uptake of glucose may induce a lower rate of fatty acid re-esterification and enhanced lipomobilization (Chilliard, 1987; Vernon, 1988).

The fact that the plasma NEFA concentration 90 min after insulin injection was higher than the pre-injection values in PA (+152 μEq L^{-1}, $P < 0.01$) may reflect a compensatory mechanism to upset the insulin-induced hypoglycaemia. If glucose availability to muscle cells is reduced, NEFA will be oxidized at a greater rate (Madsen, 1983).

Hyperglycaemia coupled to insulin resistance, high glucose and NEFA increments after isoproterenol challenge were observed in grazing dairy cows when rumen ammonia and plasma urea concentrations averaged 21.57 and 38 mg dL^{-1}, respectively (Gagliostro et al, 1994), values that were considerably higher than those observed in this experiment (14.1–15.7 for NH_3 and 21.2–24.2 mg dL^{-1} for uraemia).

In conclusion, changes in the responses to insulin and β-adrenergic signals or alterations in glucose metabolism or in the insulin secretory response to glucose were not observed between unsupplemented grazing dairy cows (exposed to higher NH_3 absorption) and the cows receiving concentrates with different rumen starch degradability. In our experiment, we preferred not to utilize non-protein nitrogen to induce the ammonia challenge in order to explore the possible metabolic derangements of NH_3 overload in naturally occurring grazing conditions. In the pastures utilized, the NSCHO/CP ratio observed (0.71–1.23) was higher than that obtained by Gagliostro et al (1994) (0.24) where insulin resistance and enhanced β-adrenergic responses were detected. Thus, it is likely that insufficient NH_3 challenge was presented in the PA treatment to upset carbohydrate and lipid metabolism or to negatively affect the secretory insulin response to glucose challenge.

The fact that there were no differences in milk yield between the two concentrates tested seems to confirm that there is a lack of a clear zootechnical benefit when replacing a rapidly degradable starch by a slowly degradable starch. The decrease in the milk fat content of cows receiving the BA based concentrate is an important factor to be considered in formulating diets with a source of rapidly and highly rumen degradable starch. On the other hand, the decrease in uraemia observed with the BA based supplement may reflect a better equilibrium between the fermentability of dietary nitrogen and the utilization of NH_3 in the rumen. This may be of interest in autumn-winter forages with a low NSCHO/CP ratio in order to avoid the metabolic risk of an excess of NH_3 absorption from the rumen.

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REFERENCES

Aiello RJ, Fink BR, Meacham CA, Armentano LE (1985) Gluconeogenesis in isolated goat hepatocytes and their response to calcium, ammonia, and other key metabolites. *J Dairy Sci* 68 (suppl 1), 140 (abstr)

Beever DE (1993) Rumen function. *In: Quantitative Aspects of Ruminant Digestion and Metabolism* (JM Forbes, J France, eds) CAB International, UK, 187-215

Casper DP, Schingoethe DJ (1989) Lactational response of dairy cows to diets varying in ruminal solubilities of carbohydrate and crude protein. *J Dairy Sci* 72, 928-941

Casper DP, Schingoethe DJ, Eisenbeisz WA (1990) Response of early lactation dairy cows fed diets varying in source of nonstructural carbohydrate and crude protein. *J Dairy Sci* 73, 1039-1050

Chilliard Y (1987) Revue bibliographique : variations quantitatives et métabolisme des lipides dans le tissu adipeux et le foie au cours du cycle gestation-lactation. 2e partie: chez la brebis et la vache. *Reprod Nutr Dev* 27, 327-398

De Visser H, Van Der Togt PL, Tamminga S (1990) Structural and non-structural carbohydrates in concentrate supplements of silage-based dairy cow rations. 1. Feed intake and milk production. *Neth J Dairy Agric Sci* 38, 487-498

Demigné C, Yacoub C, Rémésy C (1986a) Comparaison des effets glucogéniques et anticétogéniques du propionate, du propylène glycol et du glucose, chez le mouton. *Reprod Nutr Dev* 26, 367-368

Demigné C, Yacoub C, Rémésy C, Fafournoux P (1986b) Propionate and butyrate metabolism in rat or sheep hepatocytes. *Biochim Biophys Acta* 875, 535-542

DePeters EJ, Taylor SJ (1985) Effects of feeding corn or barley on composition of milk and diet digestibility. *J Dairy Sci* 68, 2027-2032

Feldman JM, Lebovitz HE (1971) Ammonium ion, a modulator of insulin secretion. *Am J Physiol* 221, 1027-1032

Fernández JM, Croom WJ, Johnson AD, Jaquette RD, Edens FW (1988) Subclinical ammonia toxicity in steers: effects on blood metabolite and regulatory hormone concentrations. *J Anim Sci* 66, 3259-3266

Gagliostro GA, Lavandera SE, Santini FJ (1994). Responses to insulin and β-adrenergic stimuli in pastures with high protein content. *Proc Soc Nutr Phys* 3, 217

Gagliostro GA, Lavandera SE, Santini FJ, Fernández HH (1996) Supplementing barley and corn based concentrates to grazing dairy cows in spring: milk yield and composition, plasma metabolite and insulin concentration and responses to hormonal signals. *Rev Arg Prod Anim* 16, 101-117

Ghrön Y, Bruss ML, Huffman EM (1987) Non linear pharmacokinetics and conversion to glucose of intravenous sodium propionate in dairy cattle. *J Dairy Sci* 70, 2272-2279

Grings EE, Roffler RE, Deitelhoff DP (1992) Evaluation of corn and barley as energy sources for cows in early lactation fed alfalfa-based diets. *J Dairy Sci* 75, 193-200

Herrera-Saldana R, Huber JT (1989) Influence of varying protein and starch degradabilities on performance of lactating cows. *J Dairy Sci* 72, 1477-1483

Inra (1988) *Alimentation des bovins, ovins et caprins* (Jarrige R, ed), Inra Editions, Versailles, France

Kloster AM, Santini FJ, Rearte DH, Fernández, HH, Elizalde JC (1994a) Suplementación de vacas lecheras con concentrados de distinta degradabilidad ruminal. 1. Producción de leche y evolución del peso vivo. *Rev Arg Prod Anim* 14 (suppl 1),17-18

Kloster AM, Santini FJ, Rearte DH, Fernández HH, Elizalde JC (1994b) Suplementación de vacas lecheras con concentrados de distinta degradabilidad ruminal. 2. Ambiente y digestión ruminal. *Rev Arg Prod Anim* 14 (suppl 1), 18-19

Madsen A (1983) Metabolism in skeletal muscle cells. *In: The Molecular Basis of Animal Production.* (Riis PM, ed) Elsevier, New York, 9-28

McCarthy RD JR, Klusmeyer TH, Vicini JL, Clark JH (1989) Effects of source of protein and carbohydrate on rumen fermentation and passage of nutrients to small intestine of lactating cows. *J Dairy Sci* 72, 2002-2016

Meijs JAC (1981) Herbage intake by grazing dairy cows. Doctoral thesis, Wageningen, 264 p

Meijs JAC, Walters RJK, Keen A (1982) Sward methods. *In: Herbage intake handbook* (Leaver JD, ed) Grassland Resarch Institute, Hurley, 11-36

Meijs JAC, Hoekstra JA (1984) Concentrate supplementation of grazing dairy cows. 1. Effects of concentrate intake and herbage allowance on herbage intake. *Grass Forage Sci* 39, 59-66

Minson DJ (1990) *Forage in Ruminant Nutrition.* Academic Press Inc, San Diego, CA

Nocek JE, Tamminga S (1991) Site of digestion of starch in the gastrointestinal tract of dairy cows and its effects on milk yield and composition. *J Dairy Sci* 74, 3598-3629

Oldham JD, Emmans GC (1988) Prediction of responses to protein and energy yielding nutrients. *In: Nutrition and Lactation in the Dairy Cow* (Garnsworthy PC, ed) Butterworths, London, 76-96

Paterson JA, Belyea RL, Bowman JP, Kerley MS, Williams JE. (1994) The impact of forage quality and supplementation regimen on ruminant animal intake and performance. *In: Forage Quality, Evaluation, and Utilization* (Fahey GC Jr, ed) Madison, WI, 59-114

Rearte DH, Santini FJ (1989) Digestión ruminal y producción en animales en pastoreo. *Rev Arg Prod Anim* 9, 93-106

Rémésy C, Chilliard Y, Rayssiguier Y, Mazur A, Demigné C (1986) Le métabolisme hépatique des glucides et des lipides chez les ruminants: principales interactions durant la gestation et la lactation. *Reprod Nutr Dev* 26, 205-226

Rémond B (1985) Influence de l'alimentation sur la composition du lait de vache. 2. Taux protéique : facteurs généraux. *Bull Tech CRZV Theix, INRA* 62, 53-67

Sasaki Y, Takahashi H, Aso H, Hikosaka K, Hagino A, Oda S (1984) Insulin response to glucose and glucose tolerance following feeding in sheep. *Br J Nutr* 52,351-358

Sauvant D, van Milgen J (1995) Dynamic aspects of carbohydrate and protein breakdown and the associated microbial matter synthesis. *In: Ruminant physiology: Digestion, Metabolism, Growth and Reproduction* (v Engelhardt W, Leonhard-Marek S, Breves G, Giesecke D, eds) Ferdinand Enke Verlag, Stuttgart, Germany, 71-91

Soar JB, Buttery PJ, Lewis D (1973) Ammonia toxicity in sheep. *Proc Nutr Soc* 32, 77A

Spires HR, Clark JH (1979) Effect of intraruminal urea administration on glucose metabolism in dairy steers. *J Nutr* 109, 1438-1447

Symonds HW, Mather DL, Collis KA (1981) The maximum capacity of the liver of the adult dairy cow to metabolize ammonia. *Br J Nutr* 46, 481-486

Vernon RG (1988) The partition of nutrients during the lactation cycle. *In: Nutrition and Lactation in the Dairy Cow* (Garnsworthy PC, ed) Butterworths, London, 32-53

Visek WJ (1984) Ammonia: its effects on biological systems, metabolic hormones, and reproduction. *J Dairy Sci* 67, 481-498

Weekes TEC, Richardson RI, Geddes N (1978) The effect of ammonia on gluconeogenesis by isolated sheep liver cells. *Proc Nutr Soc* 38, 3A (abstr)

Weis WP, Fisher GR, Erickson GM (1989) Effect of source of neutral detergent fiber and starch on nutrient utililization by dairy cows. *J Dairy Sci* 72, 2308-2315