DIGESTIBILITY AND BLOOD PARAMETERS IN THE PRERUMINANT Calf Fed A Clotting OR A NONCLOTTING MILK REPLACER 1,2

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ABSTRACT

Eight male Holstein calves 7 to 10 d of age were fed a milk replacer containing a skim milk powder subjected to low-temperature drying either with or without addition of an oxalate-NaOH buffer known to prevent curd formation. The calves were used in a completely randomized design to study the effect of milk clotting on digestibility and blood parameters. Plasma glucose and plasma insulin were similar (P > .05) for the clotting and the nonclotting milk replacers. For both treatments, concentrations of glucose and insulin reached a peak 2 h postfeeding (P < .01). Plasma triglycerides were higher (P < .01) postfeeding for the nonclotting than for the clotting milk replacer. Plasma essential amino acids and plasma urea were higher, whereas plasma calcium was lower, for the nonclotting milk (P < .01). Digestibility of dry matter, protein and fat was similar (P > .05) between clotting and nonclotting milk. The dry matter content of feces was not affected by clotting (P > .05). The data are interpreted to indicate that clotting of the milk replacer modifies blood concentrations of triglycerides, essential amino acids and urea without changing the digestibility of the diet.

(Key Words: Calves, Clotting, Coagulation, Milk, Digestibility, Digestion.)

Introduction

The effect of milk clotting on blood parameters and digestibility is usually confounded with that of treatments. The protein used for the clotting and the nonclotting milk is either different (Toullec et al., 1979) or treated to prevent curd formation in such a way that its functional properties could be modified (Jenkins and Emmons, 1982). It is, therefore, impossible to determine if the differences observed originated from the curd formation, the protein used, or the protein denaturation resulting from the pretreatment.

An oxalate-NaOH buffer developed in this laboratory prevented curd formation in the abomasum of newborn calves without denaturing milk proteins (Crugwagen, Brisson and Meissner, unpublished data). The buffer was suitable for isolating the effect of milk clotting per se, because blood parameters and digestibility were not affected by the buffer addition to a milk replacer based on a high-heat skim milk powder (Petit et al., 1987a). Therefore, the buffer was used in the present experiment to establish the importance of milk clotting on the concentration of some blood parameters and the digestibility of milk replacer constituents in young dairy calves.

Materials and Methods

Animals. Eight male Holstein calves (39 to 51 kg live weight) were obtained from the Animal Research Centre dairy cattle breeding herd at the age of 3 to 5 d. The calves were allotted to two groups of four calves, each according to a completely randomized design. Each observation obtained was the mean of four calves per treatment. They were housed individually in metabolism cages with slatted...
floors and kept in a room where temperature and relative humidity were constant. Lights were kept on all the time.

**Feed and Feeding.** The basal (clotting) milk (CM) replacer (Table 1) was formulated to contain approximately 20% lipid and 25% protein on a dry matter (DM) basis. Coagulation was prevented in the nonclotting milk (NCM) replacer by substituting 12 ml of water with an equal volume of an oxalate (.18 M)—NaOH (.25 M) buffer on each 13 g of basal milk replacer powder. The calves were fed twice daily, at 0700 and 1900. The total daily intake of fluid milk replacer was set at 10% of the initial body weight of the calves for the entire 15 d of the experiment; no refusal was registered. The clotting ability of diets was tested in vitro according to Jenkins (1982) and in vivo by visual examination when the calves were killed at the end of the trial. Extensive clotting was observed in the abomasum of calves fed CM, but no clotting was seen in those fed NCM.

**Blood Sampling.** Blood samples were obtained before the morning feeding (T₀) and 1, 2, 4, 6, 8, 12, 13, 14, 16, 18, 20 and 24 h later. The 13-h sampling corresponded to the second meal of the day at 1900. Blood sampling was done the day before the fecal collection when the calves were 10 to 12 d old. Blood samples were taken from the jugular vein into vacutainer tubes containing heparin. The blood was immediately centrifuged and the plasma isolated and frozen for subsequent analysis.

**Fecal Collection.** Collection of total feces was carried out for 5 d following a 7-d adjustment period to the diet. Daily feces were weighed and thoroughly mixed, and a 15% subsample was accumulated and stored at -15 °C for subsequent freeze-drying.

**Chemical Analysis.** Protein was measured by the Kjeldahl method. Fat was determined by the Mojonnier technique (Mojonnier and Troy, 1925) in the milk replacer and by the procedure of Lynch et al. (1963) in feces. Blood amino acids were measured with an amino acid autoanalyzer after deproteinization with salicilic acid. Plasma urea⁶, glucose⁷ and triglycerides⁸ were measured by enzymatic colorimetric methods. Plasma insulin was determined by radioimmunoassay with beef insulin as standard with polyethylene glycol separation (Desbuquois and Aurbach, 1971). Plasma calcium was determined by atomic absorption spectroscopy directly in the plasma after appropriate dilution with La₂O₃.

**Statistical Analysis.** All results were subjected to analysis of variance. Blood parameters were analyzed in a split-plot-in-time design with treatment and time postfeeding as main sources of variation. Digestibility and DM content in feces were analyzed as a randomized design with treatment as the only source of variation. The least-squares means test was used according to SAS procedure (1979) when significant differences were observed between treatments or time postfeeding.

**Results and Discussion**

The digestibilities of DM, protein and lipids were similar (P > .05) for both CM and NCM replacers (Table 2). This is in apparent agreement with other reports (Jenkins and Emmons, 1982; Van Weerden and Huisman, 1978). However, Toullec et al. (1974) observed a trend for an NCM to have lower digestibility of DM, N and organic matter compared with a CM. Their observation was for an acid-treated milk replacer with a low pH.

The DM content (Table 2) in feces was 21.5% for CM and 26.5% for NCM, but this

| Table 1. Composition of Milk Replacer |
|--------------------------------------|-----------------|
| Ingredient                           | Percentage of total |
| Fat premix⁹                          | 25.0            |
| Skim milk powderᵇ                    | 60.0            |
| Whey powder                          | 10.0            |
| Cerelose                             | 4.9             |
| Vitaminsᶜ                           | .1              |

⁶Urea-nitrogen (cat. no. 640-A), Sigma Chemical Co., St. Louis, MO.
⁷Glucose oxidase/peroxidase method (cat. no. 166 391), Boehringer Mannheim, Ville St-Laurent, Quebec, H4R 1V8.
⁸Triglycerides (cat. no. 336-20), Sigma Chemical Co., St. Louis, MO.
⁹Spray-dried product containing 80% lard; Nutribec, Quebec, Canada.
ᵇHigh-heat skim milk powder, Agropur, Coopérative Agro-alimentaire, Granby, Quebec, Canada.
ᶜVitamins (per 100 kg of dry matter): 66,000 IU of vitamin A; 117,000 IU of vitamin D₃; 660 IU of vitamin E; 440 mg of vitamin K; 275 mg of riboflavin 5-phosphate; 2,760 mg of niacinamide; 880 mg of calcium D-pantothenate; 165 mg of pyridoxine hydrochloride and 1,000 µg of vitamin B₁₂.
A difference was not significant. The absence of milk clotting in the abomasum did not produce diarrhea. This is in agreement with Toullec et al. (1974), who did not observe any difference in fecal DM content between CM and NCM obtained from the same milk powder. Diarrhea, however, was noted with NCM when the skim milk powder was heated in such a way that no clot formation occurred (Tagari and Roy, 1969). Nevertheless, heat treatment of proteins could cause digestive disorders by promoting multiplication of coliform organisms in the intestinal tract of calves (Tagari and Roy, 1969).

The glucose concentration in plasma (Figure 1) was similar from CM and NCM diets (P > .05), and the same postprandial pattern was observed. There was a sharp increase from 0 to 2 h postfeeding (P < .01). This matches the glucose peak observed for milk replacers based on low-heat skim milk powder with 5% or 25% lard (Bazin and Brisson, 1976). However, lower blood glucose peaks in calves fed NCM compared to calves fed CM were reported by Toullec et al. (1979). This could be due to the fact that they used fish protein for the NCM replacer and milk protein for the CM replacer, which resulted in different quantities of lactose in their two milk replacers.

Blood glucose concentration is a function of the hydrolysis and absorption of dietary carbohydrates (Coombe and Smith, 1974) and, therefore, could be affected by the flow rate of lactose in the duodenum. In the present experiment the same milk powder was used for both treatments and, consequently, the amounts of lactose were similar. We previously reported that the duodenal flow of lactose was the same for CM and NCM replacer based on the same milk powder (Petit et al., 1987b). The present work confirms that similar duodenal flow rates of lactose result in similar absorption rates of the hydrolysis products, as indicated by similar blood concentrations of glucose.

Plasma triglyceride concentrations were more uniform for calves fed CM than for those fed NCM (Figure 2). In calves fed CM, standard errors throughout the 24-h period remained within the range observed immediately before feeding, and there was a lack of statistical significance between times postfeeding. However, in calves fed NCM, triglyceride levels peaked 2 h postfeeding (P < .01) and gradually decreased thereafter. This was probably due to a more uniform flow rate of lipids (Petit et al., 1987a) and, consequently, a more uniform release of hydrolysis products of fat in the gut from CM than from NCM. There, the ability of a milk replacer to coagulate in the abomasum could influence plasma triglyceride levels in the preruminant calf.

<table>
<thead>
<tr>
<th>Digestibility, %</th>
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<th>Control + bufferb</th>
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<tbody>
<tr>
<td></td>
<td>Mean</td>
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<tr>
<td>Dry matter</td>
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<tr>
<td>Dry matter, %</td>
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</tr>
<tr>
<td>Feces</td>
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</tbody>
</table>

**a**Mean of four calves per treatment.

**b**Oxalate (.18 M)—NaOH (.25 M) buffer.

**c**SE = Standard error of the mean.

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**Table 2. Digestibility of Milk Replacer and Dry Matter Content of Feces (%)**

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![Figure 1](image-url)  
**Figure 1.** Concentration of glucose in plasma of calves fed a clotting (•) or a nonclotting (○) milk replacer. Vertical bars are standard errors. Arrows indicate feeding times.
Plasma essential amino acid (EAA) concentrations throughout postprandial cycles remained within the limits of the respective original standard errors at T₀ for both the calves fed NCM and calves fed CM (Figure 3). However, they were generally higher (P < .01) in calves fed NCM than in calves fed CM. Essential amino acids were expressed as the sum of threonine, valine, methionine, isoleucine, leucine, tyrosine, phenylalanine, lysine and histidine.

Cumulative duodenal flow of total N was shown to be higher in calves fed similar NCM or CM replacers (Petit et al., 1987b). The higher plasma EAA concentrations observed in NCM compared with CM calves in this experiment may have been the result of faster flow of nitrogenous material in the gut. Jenkins and Emmons (1982) also observed higher plasma EAA concentrations in calves fed NCM compared to CM replacer, but their NCM diet was pretreated with pancreatin, so the EAA blood response could have been the result of the proteolytic activity of the enzyme treatment. This response was not present when the pretreatment was applied to whole milk. It appears, therefore, that the ability of a milk replacer to curdle in the abomasum may influence the concentration of EAA in blood plasma of young calves.

Plasma urea concentrations were higher (P < .05) in NCM than in CM calves (Figure 4). This is in agreement with Jenkins and Emmons (1982), who reported that plasma urea concentration was higher in calves fed NCM replacer pretreated with pancreatin compared with the CM control. The ability of a milk replacer to coagulate in the abomasum would appear to lower plasma urea concentrations.

For both NCM and CM, there was a tendency for an increase in blood urea concentrations during the first 6 h after feeding, followed by a gradual decrease until the next feeding. In calves fed whole milk, plasma urea concentrations decreased following the morning feeding but remained unchanged after the evening meal (Williams and Smith, 1975). When synthetic milks based on nonclotting proteins were investigated, plasma urea remained constant between feedings (Nitsan et al., 1971). The patterns of plasma urea concentrations associated with time post-feeding and the factors influencing them need further investigation.

The concentration of urea in blood plasma could be associated with the rate of flow of ni-
trogenous material in the lower gut. Effectively, the duodenal flow of total N is faster in calves fed NCM compared with CM replacer (Petit et al., 1987b). It is possible, therefore, that the higher concentrations of blood urea observed in the NCM compared with CM calves, in the present trial, were associated with differences in the rate of gastric emptying of complete or partially hydrolyzed protein.

As suggested by Nitsan et al. (1971), higher blood glucose concentrations could be associated with improved use of circulating amino acids for protein synthesis, and consequently with lower plasma urea concentrations. In the present experiment, however, the glucose concentrations in the blood of both the NCM and CM calves were at the same levels throughout day and night (Figure 1). Consequently, differences in blood glucose concentrations could not explain the differences in plasma urea concentrations observed in the NCM calves compared with CM calves.

Jenkins and Emmons (1982) suggested that higher concentrations of blood urea in calves fed NCM replacer are due to an increased deamination and waste of circulating amino acids. Because protein can spare energy (Munro, 1964), higher plasma EAA concentrations in NCM compared with CM calves (Figure 3) may have increased deamination of circulating amino acids to elevate blood urea. It would appear that additional sources of readily available energy might be needed in NCM compared with CM replacers for young calves. Work is in progress to investigate this hypothesis.

Plasma insulin concentrations were similar (P > .05) for both CM and NCM diets, and they followed the same postprandial pattern (Figure 5). Plasma glucose concentrations and patterns also were similar for both CM and NCM treatments and closely resembled glucose patterns. Concentrations peaked 2 h after feeding and decreased rapidly to prefeeding levels by approximately 6 h postfeeding. Plasma glucose is probably the principal stimulator of postprandial secretion of insulin in preruminant calves fed milk protein (Grizard et al., 1982). Consequently, plasma glucose and insulin should be closely related. In the present experiment, both glucose and insulin concentrations reached a peak 2 h postfeeding and sharply declined thereafter; this is in agreement with Kalamu and Trenkle (1978). Therefore, the ability of a milk replacer to coagulate in the abomasum does not influence postprandial concentrations of plasma insulin.

Calcium concentration in plasma was lower (P < .01) in calves fed NCM than in those fed CM (Figure 6). Presumably, the oxalate buffer bound with calcium ions in the NCM replacer (Petit et al., 1987a). Because less calcium was available for absorption, plasma calcium concentration was lower for NCM replacer than for CM replacer.

In our experiment, plasma concentrations of triglycerides, EAA and urea were affected by clotting and by time postfeeding. However, the digestibility of the diet was not different for the CM and the NCM replacers. It was, concluded therefore, that previously reported decreases in digestibility of NCM replacers probably were not due to the absence of clotting.
Milk Clotting in Calves

Literature Cited


