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The Definition of Acidosis in Dairy Herds Predominantly Fed on Pasture and Concentrates[1]

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ABSTRACT

This cross-sectional survey examined the prevalence of ruminal acidosis and the effects of acidosis on the production of dairy cattle. Eight fresh cows, 3 primiparous and 5 multiparous (<100 d in milk), were selected randomly from each of 100 dairy herds in 5 regions of Australia. Rumen fluid was obtained from each cow by rumenocentesis and a stomach tube, and samples were tested for pH. Stomach tube rumen fluid samples were analyzed for volatile fatty acid, ammonia, and D-lactate concentrations. On the basis of the results of all assays, cows were categorized into 3 distinct categories (categories 1, 2, and 3) by cluster analysis. The percentages of cattle in categories 1, 2, and 3 were 10.2, 29.9, and 59.9%, respectively. Mean rumen pH for categories 1, 2, and 3 were 5.74 ± 0.47, 6.18 ± 0.44, and 6.33 ± 0.43, respectively. Biochemically, categories 1, 2, and 3 were characterized, respectively, as follows: mean total VFA concentration (m*M*), 100.74 ± 23.22, 94.79 ± 18.13, and 62.81 ± 15.65; mean ammonia concentration (m*M*), 2.46 ± 2.02, 7.79 ± 3.75, and 3.64 ± 2.03; and mean D-lactate concentration (m*M*), 0.34 ± 0.86, 0.28 ± 0.97, and 0.12 ± 0.51. Category 1 cows had higher propionate, valerate, isovalerate, and caproate concentrations and were of lower parity than cows in other categories. Cows in category 1 had higher milk production but lower milk fat content than category 2 cows. Herds were assigned to 1 of 3 groups according to the numbers of cows assigned to each category. Herds with ≥3 of the 8 cows in category 1 were classified as acidotic. Herds with ≥3 of the 8 cows in category 2 were classified as having suboptimal rumen function, and herds with ≥3 of the 8 cows in category 3 were classified as normal. Herds that had 3 or more of the 8 cows in category 1 (acidotic herds) had diets with higher energy and nonfiber carbohydrate contents and a lower neutral detergent fiber content than herds with a high prevalence of category 2 or 3 cows. The lack of significance of a herd effect in the statistical models developed suggests that the categories were robust across production systems, in which diets varied from all pasture to total mixed rations. A point prevalence of 10% (95% credible interval, 8 to 12%) of cows with an acidotic profile indicates a high risk for acidosis in the cattle sampled. The higher nonfiber carbohydrate and lower neutral detergent fiber contents of diets for herds with a high prevalence of category 1 cows (acidotic herds) indicates that there may be opportunities to reduce the risk of acidosis by dietary manipulation.
Key words: dairy cow, rumen acidosis, case definition

INTRODUCTION

Ruminal acidosis is a nutritional disorder of ruminants generally resulting from ingestion of large amounts of feeds rich in readily fermentable carbohydrates, particularly when animals are not previously conditioned to those feeds. The resulting production of large quantities of VFA and lactic acid act to lower rumen pH to nonphysiological levels. Low rumen pH can result in rumenitis, metabolic acidosis, lameness, hepatic abscessation, pneumonia, and death (Lean et al., 2000). Acidosis may be more appropriately considered a complex of conditions resulting from a similar cause: a failure to maintain effective buffering of the rumen or clearance of fermentation by-products after challenge with rapidly fermentable substrates. Brown et al. (2000) used discriminant analysis to define cows with acute and subacute acidosis in a randomized trial (n = 20) in which acidosis was induced by concentrate feeding. After examining rumen pH cut-points, they found that no single variable measured displayed a consistent response across time that could be used to identify acute or subacute ruminal acidosis. Further,

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daily fluctuations in rumen pH caused difficulty in using this as the sole measure for diagnosis of acidosis.

Australian dairy cows are predominantly pasture fed, and concentrates are typically fed twice daily at milking. Seventy-nine percent of herds in this study were fed by using this system (Bramley, 2005). The risk of acidosis may be increased when cattle are fed forages high in NFC, especially oligosaccharides, and low in physically effective fiber, such as clovers, young alfalfa, and ryegrass. Studies in Australia (Opatpatanakit et al., 1994; Clayton et al., 1999; Lean et al., 2000; Wales et al., 2001) have examined the effects of feeding starch-based concentrates to cows on pasture; however, extensive surveys of acidosis on pasture are lacking.

Some aspects of acidosis have been extensively researched and reviewed (Hungate et al., 1952; Dirksen, 1970; Huber, 1976; Slyter, 1976; Nocek, 1997; Owens et al., 1998); however, much of this research has been based on experimentally induced acidosis using large amounts of starch-based concentrate (Crichlow and Chaplin, 1985) to reduce rumen pH, often to below 5 (Nocek, 1997). Under these conditions, as more fermentable carbohydrate is fed, the growth rate of all bacteria increases, thereby increasing total VFA production. Rumen pH subsequently falls, favoring growth of *Streptococcus bovis* (Russell and Hino, 1985). The net effect of these changes is to produce a fermentation favoring lactate production and the accumulation of lactic acid, an acid 10 times stronger than either acetic, propionic, or butyric acid. Survival rates of cellulolytic bacteria decrease when pH drops to less than 6.2 (Calsamiglia et al., 1999). At rumen pH ≤4.7, the growth rate of *Strep. bovis* decreases and lactobacilli increase, producing greater amounts of lactic acid. The positive feedback effect results in fermentation stasis, absorption of D- and L-lactate, and, in severe cases, metabolic acidosis.

Although rumen pH has been used to define acidotic conditions and such methods have merit (Garrett et al., 1999), pH is a measure that merely reflects one aspect of the rumen environment. We considered that a greater understanding of rumen conditions would arise from categorizing cows by using more information on the rumen environment. This study was conducted to more accurately define rumen states present based on rumen measures in herds that were predominantly pasture based and fed varying amounts and types of concentrates.

MATERIALS AND METHODS

Herd Selection

The target population was dairy herds in New South Wales (**NSW**) and Victoria, Australia. One hundred dairy herds were selected randomly from lists supplied by local veterinary practices in 5 regions of NSW (34 herds in Finley, 17 in Camden, 9 in Taree) and Victoria (21 herds in the Western Districts and 19 in Gippsland). Herds within an area were selected randomly by using a random number chart at the time of enrollment, from a list of dairy farm clients supplied by local veterinary clinics in each area except Camden and Taree. The herds were selected by using the number arising from the random numbers table. Farmers were contacted to ensure their willingness to participate in the study. Rejection rates were less than 10% of the farmers approached in all regions except the Western Districts, where rejection rates approached 20%. Replacements for farmers who were not willing to participate were either the next herd identified by the random numbers process or, ultimately, volunteer herds from the area.

Sample Size Estimation

An initial estimate of 120 herds was made by using Pass (version 6, NCSS, Kaysville, UT) based on the number of variables anticipated in the acidotic model. However, the number of farms sampled was limited to 100 because of cost, time, and labor restraints and represents 1.2% of Victorian and NSW dairy farms (Dairy Australia, 2003).

Cow Selection

Eight cows (3 primiparous and 5 multiparous) in the first 100 d of lactation were selected randomly from each herd. The sampler was blinded from knowledge of herd tests or calving dates until after sampling so as to minimize selection biases.

Sampling and Measurements

Rumen samples were obtained by rumenocentesis and stomach tube 2 to 4 h after the morning milking when cows were fed concentrate at milking. Cows fed a TMR immediately after milking were sampled 4 to 6 h after milking. Rumenocentesis samples of 5 to 20 mL were obtained by using a 16-gauge, 3.75-cm needle attached to a 30-mL syringe. Stomach tube samples were obtained by using a custom-designed tube and equine stomach pump. The tube was removed when it contained at least 100 mL of rumen fluid. Rumen fluid was tested for the presence of saliva contamination by 1) observation of increased viscosity or obvious contamination and 2) placing a finger into the sample and withdrawing it. If a "stringing" effect was observed at the time of withdrawal, the sample was discarded and a

second sample was taken. If 3 samples were taken and saliva contamination was still present, the sample was kept and the presence of saliva contamination was noted. Although this technique helped to minimize saliva contamination, it is acknowledged that some saliva may have been present and may have influenced pH results. Rumen pH comparisons in this paper were made from rumenocentesis samples only, however, and rumen fluid taken from the stomach tubing was used to determine rumen ammonia, VFA, and D- and L-lactate concentrations.

Raw rumen fluid samples obtained by the stomach tube were centrifuged after collection, and aliquots of the supernatant were dispensed into polypropylene tubes and stored at –20°C until analysis. Rumen fluid collected by rumenocentesis was used to test rumen pH.

Analytical Procedures

Rumen pH. Rumen pH was measured immediately after collection with a calibrated pH meter (model ECpHScan-WP3, Bacto Laboratories, NSW, Australia). The meter was calibrated with buffers 4, 7, and 10 before the start of each sampling period.

Rumen VFA. Concentrations (m*M*) of propionic, acetic, isobutyric, butyric, isovaleric, valeric, and caproic acids in rumen fluid were determined (Department of Agriculture in Western Australia) by gas chromatography. The coefficients of variation (**CV**) for assays of propionic, acetic, isobutyric, butyric, isovaleric, valeric, and caproic acids were 3.8, 5.1, 3.2, 4.0, 3.4, 3.7, and 3.2%, respectively.

Rumen Ammonia. Rumen samples were analyzed for ammonia by using a Boehringer Mannheim/R-Biopharm kit (cat. no. 1 112 732, Arrow Scientific, NSW, Australia). The assay of ammonia is an enzymatic assay linked through glutamate dehydrogenase to the oxidation of NADPH. This oxidation of NADPH was monitored at 340 nm in a spectrophotometer. The CV for ammonia depended on the concentration of ammonia, with a CV of 3.7% for concentrations up to 30 mg/L and 1.1% for concentrations greater than 30 mg/L up to approximately 50 mg/L.

Rumen D-Lactate and L-Lactate. A Boehringer Manheim/R-Biopharm kit (cat. no. 1 112 821, Arrow Scientific) was used to analyze D-lactate. The oxidation of D-lactate is an enzymatic assay linked through D-lactate dehydrogenase by NAD^+. The increase in NADH was monitored at 340 nm in a spectrophotometer. The CV for assay of D-lactate also depended on the concentration of D-lactate, with a CV of 22.7% for concentrations up to 1 m*M* and 2% for concentrations greater than 1 m*M*.

Selected rumen fluid samples were also analyzed for L-lactate by using the same kit as for D-lactate. The oxidation of L-lactic acid requires the presence of the enzyme L-lactate dehydrogenase, with the increase in NADPH monitored at 340 nm in a spectrophotometer. The CV for assay of L-lactate also depended on the concentration of L-lactate, with a CV of 7.9% for concentrations up to 1 m*M* and 1.4% for concentrations greater than 1 m*M*.

Dietary Inputs. Diets from herds were obtained by questionnaire and by measurement and weighing of grain and pasture. Pasture samples, TMR samples, and some conserved forages or unknown grain mixes were submitted to the Dairy One Forage Laboratory (Ithaca, NY) for testing by near-infrared reflectance spectroscopy. Results for each feed sample included percentages of DM, NDF, CP, degradable protein, crude fat, ash, ADF, lignin, NFC, NSC, starch, and sugar. Analysis of TMR, conserved forage, and grain mix samples included DM, CP, soluble protein, ADF, NDF, fat, ash, and NFC percentages. Grains were not routinely tested for feed values; these were estimated by using values previously obtained for Australian grains. Once these inputs were determined and allowance was made for feed wastage, the amounts and composition of feeds, details of the cattle, environmental inputs, and exercise estimates were entered into the ration formulation program CPM-Dairy (Cornell*Penn*Miner 2003, version 3.0.4a) and the nutrient composition of the ration was estimated. The DMI and physically effective NDF (**peNDF**) intakes estimated from measurements in the field were compared with the predicted DMI and peNDF estimated by using CPM-Dairy to provide an upper limit of forage intake should feed intake measured in the field exceed that estimated from CPM-Dairy. For soft, short pastures, we estimated an effective NDF (**eNDF**) of 65%, and for longer pastures of greater shear strength, we estimated an eNDF of 75%. Pastures with characteristics between these were assigned an intermediate eNDF.

Statistical Analyses

Initial evaluation of data included generation of descriptive statistics, graphical evaluation of data, and bivariate analyses. A K-means cluster analysis (version 12.0 Apache Software, SPSS Inc., Chicago, IL) was used to classify cows into 1 of 3 categories based on rumen VFA, ammonia, and rumenocentesis pH at the time of sampling (referred to as categories 1, 2, and 3 in the remainder of this paper). There were 114 unclassified cases in the cluster analysis because of missing rumenocentesis data for one or more cases. These cases were

allocated to 1 of the 3 categories by using discriminant analysis (version 12.0 Apache Software, SPSS Inc.) based on the other variables measured, including stomach tube pH.

Data differences between amounts of rumen modifiers and antibiotics fed were analyzed by using one-way ANOVA. Because of the inequality of variances, between-group comparisons of the mean rumen chemistry values were assessed by using a Kruskal-Wallis one-way ANOVA and the Wilcoxon rank-sum test.

We used a generalized linear model in SPSS (version 12.0 Apache Software, SPSS Inc.) to quantify the effect of rumen pH category, DIM, and parity (as fixed effects) and herd (as a random effect) on milk volume recorded at the herd test closest to the date of sampling.

Ordinal logistic regression was used to quantify the effect of rumen chemistry values on the probability of a cow being classified into category 1, 2, or 3. A Bayesian approach was used, first to account for clustering of the data at the herd level, and second as a means for dealing with missing values in this data set. We parameterized the relationship between the logit of the outcome variable as a function of rumen propionate, butyrate, valerate, caproate, and D-lactic acid concentrations. Prior distributions were assigned to the unknown quantities (the regression coefficients for each of the parameterized fixed effects, the variability of the effect of herd, which was included as a random effect, and the missing covariates). Bayes' theorem was applied by using a Markov chain Monte Carlo algorithm implemented in WinBUGS 1.4 (Spiegelhalter et al. 1999), which allowed the data to inform the revision of the assigned prior distributions into posterior distributions that were used for inference. Flat (uninformed) prior distributions centered at zero and with small precision (inverse variance) were used for each of the fixed effects. An uninformed gamma hyperprior was used for the precision of the herd-level random effect term. For the Bayesian analyses, the Markov chain Monte Carlo sampler was run for 40,000 iterations and the first 1,000 "burn-in" samples were discarded. Convergence was quantified by using the Raftery and Lewis convergence diagnostic (Raftery and Lewis, 1992a,b). Posterior sample sizes were determined by running sufficient iterations to ensure that the Monte Carlo standard error of the mean was at least one order of magnitude smaller than the posterior standard deviation for each parameter of interest.

Herd Categories

Herds were assigned to 1 of 3 groups according to the numbers of cows assigned to each category. Herds with ≥3 out of 8 cows in category 1 were classified as acidotic (**ACID**). Herds with ≥3 out of 8 cows in category 2 were classified as having suboptimal rumen function (**SO**), and herds with ≥3 out of 8 cows in category 3 were classified as normal (**NA**). The number of herds in categories ACID, SO, and NA were 10, 35, and 69, respectively. Twenty herds met the criteria for 2 categories and were used twice in the analysis; one herd was assigned to categories ACID and SO, 2 herds were assigned to categories ACID and NA, and 17 herds were assigned to categories SO and NA.

RESULTS

Power Analysis

A sample size of 100 herds provided a power of 0.60 with $\alpha = 0.05$ to detect a difference in herd-level acidosis prevalence of 10%.

Allocation of Cows

When cows were categorized into 4 or more categories, the category size in one or more categories became too small and category definition was poor. There was no clear definition when 2 categories were tested. A total of 683 cows were used in the cluster analysis; 70 were assigned to category 1, 204 to category 2, and 409 to category 3. The 3 categories identified are proposed to represent acidotic, suboptimal rumen function, and nonacidotic cows. Cows with one or more variables missing (n = 114) were not included in the cluster analysis.

Table 1 shows the results of a K-means cluster analysis ANOVA. All covariates were significant in predicting category allocation ($P < 0.01$). Propionate ($F = 332.38$) and valerate ($F = 332.62$) were the most important factors in determining categories, followed by butyrate ($F = 291.86$), isobutyrate ($F = 243.50$), acetate ($F = 226.17$), and ammonia ($F = 203.90$). Interestingly, rumen pH ($F = 54.95$) and D-lactate ($F = 5.02$) were the least significant in predicting categories.

Category 1

Mean values for rumen measures for the 3 categories determined by using cluster analysis are given in Table 2. Data from cattle not classified by cluster analysis are presented for comparative purposes. Category 1 was characterized by a low mean rumen pH, high mean total VFA concentrations, and low to normal mean rumen ammonia concentrations of 2.46 m*M*. The mean propionate concentration was significantly higher in category 1 than in the other 2 categories ($P < 0.001$), resulting in a mean ratio for acetate:propionate of 1.53. The mean D-lactate concentration of category 1, although significantly higher than those of categories 2 and 3 ($P <$

Table 1. K-means cluster analysis ANOVA among acidotic, subclinically acidotic, and nonacidotic animals after standardizing variables to *Z*-scores (n = 683)

Item	Mean squares	Mean squares error terms	df	*F*-ratio	*P*-value
Acetate	131.811	0.583	2,680	226.172	0.0001
Propionate	168.694	0.508	2,680	332.375	0.0001
Butyrate	140.255	0.481	2,680	291.856	0.0001
Isobutyrate	130.184	0.535	2,680	243.502	0.0001
Isovalerate	95.694	0.733	2,680	130.571	0.0001
Valerate	152.686	0.459	2,680	332.620	0.0001
Caproate	70.108	0.601	2,680	116.572	0.0001
Rumen pH[1]	47.211	0.859	2,680	54.950	0.0001
D-Lactate	2.973	0.593	2,680	5.017	0.0070
Ammonia	120.556	0.591	2,680	203.895	0.0001

[1]Rumen fluid collected by rumenocentesis.

0.001), was only 0.34 ± 0.86 m*M*. The mean valerate concentration was significantly higher ($P < 0.001$) than for the other 2 categories.

Category 2

Cattle in category 2 had very different mean concentrations of almost every measure compared with category 1 (Table 2). The mean rumen pH was significantly higher for category 2 than category 1 and significantly lower than category 3 ($P < 0.001$). The mean total VFA concentration was significantly higher than category 3 ($P < 0.001$). There was no significant difference between the mean total VFA concentration in categories 1 and 2. Of the individual VFA concentrations, acetate and butyrate were significantly higher and propionate was significantly lower than in category 1 ($P < 0.001$). The lower propionate concentration in category 2 produced a higher ratio of acetate:propionate (3.04) in this category. The mean D-lactate concentration was significantly lower in category 2 compared with category 1. The mean ammonia concentration was significantly (*P*

Table 2. Means ± standard deviations of biochemical analyses for acidotic, subclinically acidotic, and nonacidotic cows determined by using cluster analysis

Item	Category 1, n = 70	Category 2, n = 204	Category 3, n = 409	Unclassified,[1] n = 114
pH (rumenocentesis)	5.74 ± 0.47[a]	6.18 ± 0.44[b]	6.33 ± 0.43[c]	6.32 ± 0.50 n = 24
Total VFA, m*M*	100.74 ± 23.22[a]	94.79 ± 18.13[a]	62.81 ± 15.65[b]	80.68 ± 26.78 n = 110
Acetate, m*M*	52.45 ± 12.37[a]	59.15 ± 11.80[b]	39.16 ± 10.74[c]	48.98 ± 16.04 n = 110
Propionate, mM	34.19 ± 10.17[a]	19.47 ± 6.18[b]	14.40 ± 4.88[c]	18.76 ± 8.43 n = 110
Butyrate, m*M*	9.13 ± 3.46[a]	12.38 ± 2.94[b]	6.89 ± 2.32[c]	9.66 ± 4.91 n = 110
Isobutyrate, m*M*	0.68 ± 0.22[a]	0.86 ± 0.22[b]	0.52 ± 0.16[c]	0.71 ± 0.31 n = 110
Isovalerate, m*M*	1.14 ± 0.37[a]	1.37 ± 0.49[b]	0.88 ± 0.26[c]	1.08 ± 0.40 n = 110
Valerate, m*M*	2.55 ± 1.20[a]	1.28 ± 0.42[b]	0.82 ± 0.36[c]	1.24 ± 0.98 n = 110
Caproate, m*M*	0.59 ± 0.56[a]	0.27 ± 0.18[b]	0.13 ± 0.17[c]	0.26 ± 0.46 n = 110
D-Lactate, m*M*	0.34 ± 0.86[a]	0.28 ± 0.97[b]	0.12 ± 0.51[c]	0.42 ± 1.88 n = 88
Ammonia, m*M*	2.46 ± 2.02[a]	7.79 ± 3.75[b]	3.64 ± 2.03[c]	4.56 ± 3.99 n = 108

[a–c]Column means within a row with different superscripts differ ($P < 0.01$) using the Wilcoxon rank-sum test.

[1]Column labeled "unclassified" was not compared statistically with other columns.

Table 3. Reasons for cows unable to be classified by using cluster analysis (n = 114)

Reason for unclassification	n
Missing rumen pH (rumenocentesis)	86
Missing D-lactate concentration	19
Missing ammonia concentration	2
Missing rumen pH and D-lactate concentration	3
Missing rumen VFA, D-lactate, and ammonia concentrations	2
All data missing	2

< 0.001) and substantially higher than in either category 1 or 3.

Category 3

Category 3 contained the largest percentage of cows (51%; Table 2). This category had a significantly higher mean rumen pH than category 1 or 2 and lower mean total VFA and D-lactate concentrations ($P < 0.001$). The mean ammonia concentration for category 3 was significantly higher than for category 1 and lower than for category 2 ($P < 0.001$). Mean acetate, propionate, and butyrate concentrations were all significantly lower than in categories 1 and 2 ($P < 0.001$) and the acetate:propionate ratio was 2.72. Mean isobutyrate, isovalerate, valerate, and caproate concentrations were also significantly lower than in categories 1 and 2 ($P < 0.001$).

Table 3 lists the reasons cows were not included in the cluster analysis. The most common reason for unclassified cows was missing rumenocentesis data. This was due to the difficulty in obtaining enough rumen fluid by rumenocentesis that was not obviously contaminated with blood to test rumen pH. Where the amount of rumen fluid collected was too small to obtain an accurate pH reading, or when blood was visually noted in the sample, the data were marked as missing. Table 4 summarizes the means and standard deviations of the variables in each category by using all cases (n = 797) as determined by the discriminant analysis. The means were similar to the initial cluster analysis results and only the unclassified cases (n = 114) were reclassified, indicating the strength of the original classification. Table 5 shows the results of the ordinal logistic regression analyses. The intraclass correlation coefficient for the effect of herd in this model was 0.65 (95% credible interval, 0.41 to 0.940).

Diet Summary for Herd Categories

The mean dietary parameters for each herd category are presented in Table 6. Significant differences were found among herd categories for estimated ME, NDF, peNDF, NFC, and starch percentages ($P < 0.05$). The mean estimated ME concentration in the diet of herd category ACID was significantly greater than that of herd category NA ($P = 0.011$). The mean NFC percentage in the diet of ACID herds was also significantly higher than those of either the SO ($P = 0.002$) or NA ($P = 0.001$) herds. Similarly, the mean starch percentage of the diet was significantly higher in ACID herds compared with SO ($P = 0.004$) or NA ($P = 0.003$) herds. The mean NDF percentage of the diet of ACID herds was significantly lower than that of SO ($P = 0.015$) or NA ($P = 0.002$) herds. The dietary peNDF was also significantly lower in ACID herds compared with SO ($P = 0.042$) or NA ($P = 0.011$) herds. All other dietary measures reported in Table 6 were not significant among categories. The percentage of herds feeding monensin, lasalocid, virginiamycin, and tylosin and the mean amounts fed from the herds feeding the product are summarized in Table 7. The prevalence of monensin or virginiamycin use and the inclusion rate of monensin or virginiamycin did not vary among the 3 herd categories.

Table 4. Means ± standard deviations of biochemical analyses for acidotic, subclinically acidotic, and nonacidotic cows (n = 797) after classification of 114 unclassified cows[1]

Item	Category 1, n = 83	Category 2, n = 243	Category 3, n = 471
pH (rumenocentesis)	5.73 ± 0.47 (73)	6.18 ± 0.43 (207)	6.33 ± 0.42 (427)
Total VFA, m*M*	100.89 ± 23.62 (83)	95.44 ± 18.41 (243)	62.93 ± 16.03 (467)
Acetate, m*M*	52.26 ± 12.52	59.54 ± 11.87	39.27 ± 11.00
Propionate, m*M*	34.04 ± 9.86	19.53 ± 6.14	14.44 ± 4.95
Butyrate, m*M*	9.49 ± 4.07	12.53 ± 3.21	6.88 ± 2.36
Isobutyrate, m*M*	0.67 ± 0.22	0.89 ± 0.23	0.52 ± 0.16
Isovalerate, m*M*	1.10 ± 0.38	1.38 ± 0.47	0.88 ± 0.26
Valerate, m*M*	2.65 ± 1.28	1.29 ± 0.42	0.82 ± 0.36
Caproate, m*M*	0.62 ± 0.67	0.28 ± 0.19	0.13 ± 0.17
D-Lactate, m*M*	0.55 ± 1.92 (81)	0.25 ± 0.90 (239)	0.14 ± 0.61 (450)
Ammonia, m*M*	2.33 ± 1.92 (82)	7.79 ± 3.91 (243)	3.56 ± 2.02 (466)

[1]Numbers in parentheses indicate the number of samples used.

Table 5. Posterior means and posterior standard deviations of the regression coefficients in the ordinal logistic regression model used to quantify the influence of rumen chemistry values on the probability of a cow being classified into category 1, 2, or 3[1]

Explanatory variable	Mean ± SD	Median (95% CI)	MC error[2]
Fixed effects			
1|2	–4.9750 ± 0.6030	–4.9030 [–6.2130–(–3.9670)]	0.08
2|3	–0.1862 ± 0.1682	–0.1830 (–0.5292–0.1358)	0.01
Propionate	1.1740 ± 0.2557	1.171 (0.7043–1.706)	0.02
Butyrate	0.8956 ± 0.1533	0.8887 (0.6166–1.236)	0.01
Valerate	2.1770 ± 0.3989	2.152 (1.4610–3.0710)	0.04
Caprionic acid	0.3793 ± 0.2188	0.3771 (–0.0413–0.8282)	0.01
D-Lactic acid	0.0794 ± 0.0947	0.0778 (–0.0970–0.2714)	<0.01
Random effect			
Herd-level variance	1.0650 ± 0.4822	1.0280 (0.0311–2.1270)	0.06
Cow-level variance	0.6852 ± 0.7588	0.3030 (0.0018–2.3060)	0.11

[1]Intraclass correlation coefficient: 0.65 (95% credible interval, 0.41 to 0.94).

[2]Monte Carlo standard error.

Table 6. Mean dietary parameters ± standard deviations of herds in each category where herds contained 37.5% or more of cows allocated to the category

Item	ACID herds,[1] n = 10	SO herds,[2] n = 35	NA herds,[3] n = 69
DMI, kg/cow per d	19.39 ± 3.01[c]	18.73 ± 3.94[c]	19.19 ± 3.74[c]
CP, %	19.93 ± 2.19[c]	19.98 ± 2.96[c]	19.62 ± 3.00[c]
Undegradable protein, % of CP	35.18 ± 3.67[c]	34.72 ± 4.71[c]	35.03 ± 4.49[c]
Soluble protein, % of CP	35.07 ± 2.40[c]	34.92 ± 4.85[c]	35.21 ± 4.21[c]
Estimated ME, MJ/kg of DM	11.02 ± 0.37[c]	10.47 ± 0.91[cd]	10.40 ± 0.79[d]
NDF, %	30.41 ± 4.27[ac]	35.65 ± 5.98[d]	36.09 ± 5.18[b]
Physically effective NDF, %	21.07 ± 3.12[c]	24.23 ± 4.68[d]	24.58 ± 4.26[d]
NFC, %	40.26 ± 4.44[a]	33.66 ± 6.45[b]	33.46 ± 5.71[b]
Starch, %	22.74 ± 5.37[a]	16.82 ± 6.23[b]	16.86 ± 5.95[b]
Total fat, %	4.01 ± 0.51[c]	4.39 ± 0.79[c]	4.44 ± 0.81[c]

[a,b]Column means within a row with different superscripts differ ($P < 0.01$).

[c,d]Column means within a row with different superscripts differ ($P < 0.05$).

[1]ACID = herds with ≥3 of the 8 cows in category 1 classified as acidotic (ACID).

[2]SO = herds with ≥3 of the 8 cows in category 2 classified as having suboptimal rumen function (SO).

[3]NA = herds with ≥3 of the 8 cows in category 3 classified as normal (NA).

Table 7. Percentage of herds feeding ionophores and antibiotics in the herd categories, and mean amounts fed

	Feeding product, % Mean ± SD fed, mg/cow per d		
Item	ACID herds[1]	SO herds[2]	NA herds[3]
Monensin	100.0 252.60 ± 100.79	65.7 258.46 ± 92.64	60.9 240.16 ± 71.75
Lasalocid	0	0	7.2 265.02 ± 40.07
Virginiamycin	50.0 156.80 ± 92.13	28.6 186.60 ± 46.32	24.6 195.29 ± 74.97
Tylosin	0	0	4.3 119.67 ± 39.50

[1]ACID = herds with ≥3 of the 8 cows in category 1 classified as acidotic (ACID).

[2]SO = herds with ≥3 of the 8 cows in category 2 classified as having suboptimal rumen function (SO).

[3]NA = herds with ≥3 of the 8 cows in category 3 classified as normal (NA).

Table 8. Production data (mean ± SD) for cows in categories for which a herd test was recorded[1]

Mean production data	Category 1, n = 39	Category 2, n = 132	Category 3, n = 276
Milk volume, L	30.00 ± 0.97[cd]	27.90 ± 0.52[c]	29.70 ± 0.36[d]
Milk fat, %	3.32 ± 0.11[ac]	3.79 ± 0.06[bc]	3.61 ± 0.04[abd]
Milk protein, %	3.15 ± 0.06[cd]	3.18 ± 0.03[c]	3.09 ± 0.02[d]
Fat:protein	1.06 ± 0.04[a]	1.20 ± 0.02[b]	1.18 ± 0.01[b]
Milk fat yield, kg	1.00 ± 0.04[c]	1.04 ± 0.02[c]	1.05 ± 0.02[c]
Milk protein yield, kg	0.94 ± 0.03[c]	0.87 ± 0.02[c]	0.91 ± 0.01[c]
SCC, cells/mL (log transformed)	10.90 ± 0.23[c]	11.00 ± 0.12[c]	11.00 ± 0.09[c]

[a,b]Column means within a row with different superscripts differ ($P < 0.01$).

[c,d]Column means within a row with different superscripts differ ($P < 0.05$).

[1]Corrected for DIM, parity, and number of days from last herd test to sampling day, controlled for the effect of herd.

Production Data Summary for Cow Categories

The mean number of days from herd test to sampling date for cows in each of the 3 categories was 11.3 ± 8.19 d, 10.7 ± 9.31 d, and 12.0 ± 9.66 d for categories 1, 2, and 3, respectively. The mean parity for cows in categories 1, 2, and 3 was 2.08 ± 1.53 calvings, 3.15 ± 2.39 calvings, and 3.05 ± 2.06 calvings, respectively. Parity was significantly lower in category 1 than in categories 2 ($P = 0.013$) and 3 ($P = 0.005$). Days in milk were not significantly different among categories of cows, and mean values for categories 1, 2, and 3 were 44.7 ± 25.47 d, 52.0 ± 26.7 d, and 47.6 ± 26.9 d, respectively.

Production data for the 3 categories after correcting for DIM, parity, and number of days from herd test to sampling date and including herd as a random effect are presented in Table 8. Cows were included in this analysis if these were in the original cluster analysis <100 DIM, and if the interval between sampling and herd testing was <31 d (1 mo). Milk production in category 1 was greater ($P = 0.057$) than in category 2, and milk production in category 2 was significantly less than that in category 3 ($P = 0.011$). The percentage of milk fat was significantly less in category 1 than in categories 2 ($P = 0.001$) and 3 ($P = 0.020$) but was greater in category 2 than in category 3 ($P = 0.022$). The percentage of milk protein was only significantly greater in category 2 than in 3 ($P = 0.024$). This resulted in a significantly lower milk fat:protein ratio in category 1 compared with category 2 ($P = 0.001$) or 3 ($P = 0.004$). The differences in milk volume among the categories resulted in a higher milk protein yield in category 1 compared with category 2 ($P = 0.055$). There was no significant difference in SCC among the 3 categories.

DISCUSSION

The 100 samples represent 1.2% of Victorian and NSW dairy farms (Dairy Australia, 2003). It was considered more valuable, in terms of understanding acidosis, to sample more cows in each herd and fewer herds. Thus, approximately 800 cows were sampled for rumen fluid. Therefore, although the statistical power for herd-level variables was modest, the statistical power for cow-level variables, such as group classification, was high.

Herds selected within each region, apart from Camden, were sampled over approximately a 1-mo period, and measures were therefore clustered within district, herd, and time of sampling. The number of herds sampled from each of the districts approximated the percentage of milk produced by the regions in 2001: Southern NSW and Northern Victoria 42%, Gippsland 25%, Western 25.2%, Central NSW 4.9%, and North Coast 3.7% of total milk, respectively (Australian Bureau of Agricultural and Resource Economics, 2004). Two of the 5 areas sampled, Camden and Taree, were difficult to obtain random samples from because of sample processing constraints, and Camden herds were overrepresented compared with milk production from the central region of NSW.

These herds may reflect farmers who were more sympathetic to research or had an awareness of acidosis, given that approximately 10% of farmers in most districts and 20% in the western districts declined to participate in the study. We observed, however, that even the most progressive farmers did not readily recognize signs of acidosis in a herd. Notwithstanding these possible sources of bias, compared with many studies that have used DHIA herds only, this study offers a data set that was sampled more widely, and one that is representative of the dairy industry in Southern Australia.

Herd Categories

Data were collected with a goal of identifying cow and herd-level risks for acidosis. Therefore, herds were grouped into categories based on individual cow classi-

fications determined by cluster analysis. The method of allocation based on ≥3 of the 8 cows being in a category resulted in some double allocation of herds. Although there was a variance inflation effect caused by counting some herds twice, this was minimal for the primary herd category of interest, ACID, because few of those herds were used twice. Had herds not been included in either category, the power of the data set would have been greatly reduced. The classification of herds by grouping ≥4 of 8 cows allocated to the same category was explored; however, this would have greatly reduced the numbers in herd groups. Although this would have alleviated the problem of herds meeting the criteria of 2 categories, it was considered more valuable to include herds in analyses, rather than to lose herds from the analysis. Consequently, *P*-values for differences among herd categories are slightly inflated.

Biochemical Analyses

Cluster analysis identified homogenous groups of cases based on rumen biochemistry. A strong correlation (R = 0.905, Pearson's correlation) between D- and L-lactate found in a subsample of 183 cows analyzed (Bramley, 2005) indicated that inclusion of either one or the other lactate or total lactate was appropriate. Given the importance of D-lactate in the pathogenesis of ruminal acidosis, the low concentrations of L-lactate, and the strong correlation of D-lactate with L-lactate, we decided to use D-lactate solely for assessing acidosis status in this study. The categories were robust, with little difference in allocation of cases assessed by cluster or by discriminant analysis. Herds were fed a wide range of diets, from TMR and zero-grazed to one herd that was fed no grain and only pasture. Despite this, the effect of herd explained only a little over half of the variation in the category assignment (Table 5).

Rumen acidosis was defined by using rumen pH. The cutoff pH at which subclinical acidosis is diagnosed is controversial, with some authors suggesting pH 5.5 for rumenocentesis samples (Garrett et al., 1999). However, in vitro fiber digestibility is reduced when pH drops below 6.2 (Grant and Mertens, 1992; Grant, 1994; Calsamiglia et al., 1999). In this study, 7.6% of cows had a rumenocentesis pH of less than 5.5, 16.1% of cows had less than 5.8, but 43% of cows had a rumen pH of less than 6.2 (Figure 1). Rumen pH fluctuates throughout the day and depends on the diet, time of feeding of concentrates, and supplementation of fiber sources. Because of the fluctuation of rumen pH during the day, diagnosis of acidosis based solely on rumen pH can be difficult. Although many of the cows in category 1 had low pH, this was not a sensitive or specific measure for group classification, suggesting that other measures of rumen function are important in determining cows at risk for acidosis.

Category 1

The rumen metabolism and milk production characteristics of cows in category 1 are consistent with an acidosis model reflecting a high rate of carbohydrate fermentation, resulting in high total VFA, propionate, and valerate concentrations, high to normal D-lactate concentrations, and low to normal rumen ammonia concentrations. Milk production was higher than for the other categories but with a low milk fat percentage, resulting in a significantly lower milk fat:protein percentage than for the other categories. Herds with a higher proportion of cows in this category, ACID herds, were fed significantly more highly fermentable carbohydrates than herds in the other categories (Table 6) in concentrations that met NRC recommendations of 36 to 44% NFC (NRC, 2001). The high NFC percentage in the diet, combined with a mean dietary NDF percentage of 30.4 ± 4.27%, the lowest of the 3 herd categories, provides conditions that favor the growth of bacteria that ferment sugars and starch. When the amount of concentrate or the carbohydrate percentage is increased in the diet and the NDF percentage is decreased, concentrations of total VFA increase, resulting in a lower rumen pH (Lana et al., 1998; Russell, 1998), results that reflect the patterns of fermentation observed in category 1 cows.

The 2 most important electron sink products in the rumen are propionate and methane, and there appears to be an inverse relationship between the 2 products (Wolin, 1960). The significantly higher valerate concentration in category 1 compared with the other categories is interesting. Valerate is normally present in very low concentrations in the rumen. The most important producer of valerate is *Megasphaera elsdenii*, found primarily in calves and cattle receiving rations high in grain (Hungate, 1966; Stewart et al., 1997). *Megasphaera elsdenii* utilizes 60 to 95% of the lactate available in the rumen (Counotte et al., 1981), and valerate is a major product formed from this substrate (Stewart et al., 1997). *Megasphaera elsdenii* takes 5 to 7 d to establish rumen populations of 5.5×10^{11} in concentration after introduction of grain to the diet (Klieve et al., 2003). Other users of lactic acid include *Selenomonas ruminantium* ssp. *lactilytica* and entodidiomorph protozoa. The significantly higher concentration of valerate in category 1 may be associated with a higher production of lactate and subsequent conversion by cows in this category. Higher concentrations of D-lactate found in this category support this contention. Valerate is

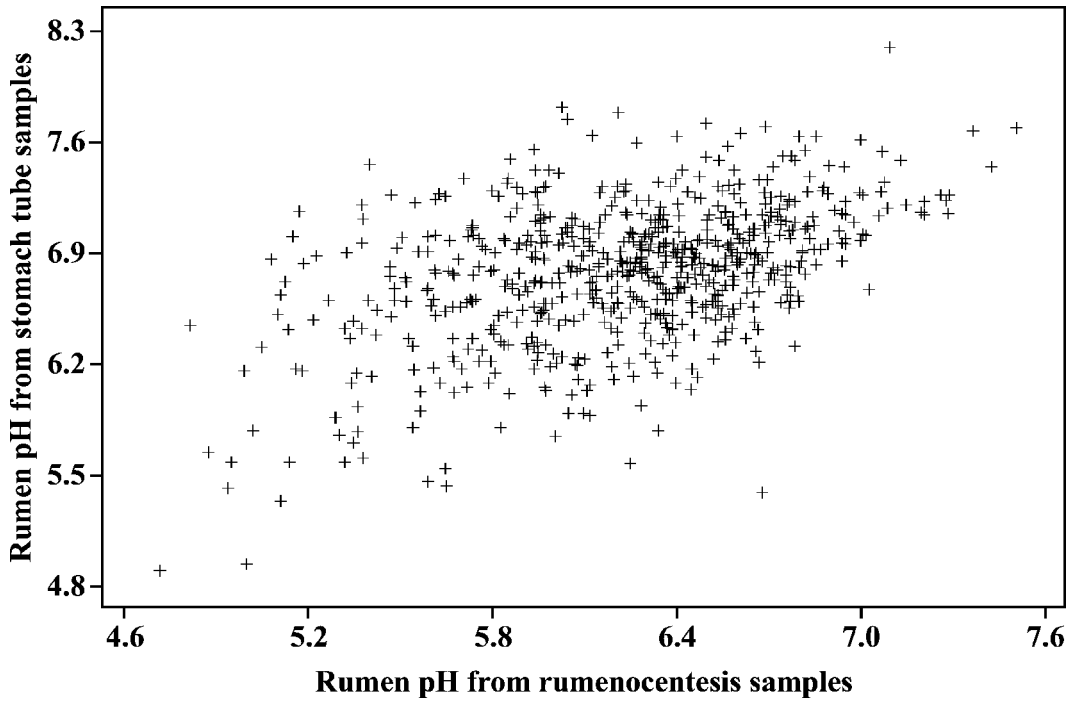

Figure 1. Scatter plot comparing rumen pH measured by rumenocentesis vs. stomach tube ($R^2 = 0.20$).

another electron sink product in the rumen and may be a useful indicator of cows at risk for acidosis.

D-Lactate concentrations in category 1 were significantly higher compared with the other categories, but were low compared with previously reported concentrations in concentrate-induced acidosis (Crichlow and Chaplin, 1985). Pastures contain significant amounts of oligofructans, and these have been used in large amounts to induce acidosis and laminitis (Theofner et al., 2004). It is therefore possible that these cows do not fit the traditional starch-induced acidosis model, where lactobacilli are known to multiply quickly, producing large amounts of lactate (Nocek, 1997), and reflect a subclinical acidosis associated with cows adapted to diets with high concentrations of rapidly fermentable starches and sugars.

Dietary CP (19.9 ± 2.19%) for ACID herds was adequate to high. The relatively low ammonia concentrations may therefore reflect either increased microbial protein synthesis and ammonia assimilation, more rapid clearance from the rumen, or failure of proteolytic bacteria to deaminate available protein. Other studies have found lower rumen ammonia concentrations to be associated with higher feeding rates of NSC (MacGregor et al., 1983; Lana et al., 1998). Lana et al. (1998) found that when the concentrate percentage was increased in the diet, deamination rates decreased. A decrease in rumen pH also reduced deamination in vitro in forage-fed cattle. However, cattle fed 90% concentrate did not have a low rumen pH, demonstrating differences in microbial populations on the 2 diets (Lana et al., 1998). Monensin fed at 350 mg/cow per d decreases ruminal ammonia production by more than 30% by inhibiting AA-fermenting bacteria (Yang and Russell, 1993). Although there was no significant difference among herd categories in the mean amount of monensin fed, all herds in this category were feeding monensin, possibly leading to some association with the lower ruminal ammonia concentration.

The activity of virginiamycin against *Strep. bovis* and *Lactobacillus* spp. has led to its use to control acidosis (Al Jassim and Rowe, 1999). Virginiamycin was fed in 50% of herds in category 1, but the mean inclusion rate in the diets was lower (156.8 ± 92.13 mg/cow per d) than the recommended rate of 200 mg/cow per d. It is possible that without feeding virginiamycin, the risk of acidosis may have been higher in these herds. However, in herds feeding virginiamycin, a lower than recommended rate of use may have reduced the effectiveness of the product.

Cows in category 1 had higher milk production than those in the other categories, but not significantly. The relationship between concentrate feeding and increased milk production on pasture-based diets is well recognized (Wales et al., 2001; Walker et al., 2001). The significantly lower milk fat percentage, after correcting

for DIM, parity, and days to herd test, was also associated with a high level of concentrate feeding, lower dietary NDF and peNDF, and lower rumen pH. This decrease was associated with the lower milk fat:protein ratio of 1.06 ± 0.036 than in the other categories, supporting previous research linking rumen pH to milk fat concentration (Kolver and de Veth, 2002) and dietary eNDF to milk fat percentage (Mertens, 1997). The mean parity of cows in category 1 was significantly lower than those in other categories, indicating that heifers, although offered diets similar to older cows, had a greater risk of acidosis, suggesting that the type of feeding system or diet preference may be a problem. If there is limited access to pasture fiber, older cows may have preferential intake of this fiber (Shepherd, 2003). Another possibility is that heifers may have fewer rumen papillae and less bacterial adaptation than mature cows.

Category 2

Dietary CP percentage, ME concentration, and NFC percentage of the diets offered were all adequate for an early-lactation dairy cow and were not significantly different from the respective values for diets offered to NA herds. These concentrations and the mean NDF percentage of 35.7 ± 5.98% should create a stable rumen environment. However, the mean rumen biochemical profile for cows in this category suggests a possible mismatch of energy and protein inputs, whereby ammonia was not assimilated into microbial protein, possibly because energetic precursors were not sufficiently available when required by bacteria. The low mean concentration of propionate probably resulted in lower glucose availability, and consequently lower milk production for cows in this category compared with either category 1 ($P = 0.057$) or 3 ($P = 0.011$).

The mean total VFA concentration observed was adequate; previous data suggest that optimal concentrations of total VFA are more than 95 m*M* (Leng and Brett, 1966). However, some studies found much lower total VFA concentrations. Russell (1998) found that nonlactating cows fed 10 kg of DM/cow per d of a diet of 100% hay had a total VFA concentration of 68.3 m*M* compared with cows fed 90% concentrate, with a total VFA concentration of 83.9 m*M*. The total VFA concentration was less than that of category 1, although not significantly, with the trend possibly explained by the lower mean dietary ME concentration and significantly lower NFC percentage of SO than ACID herds. The lower NFC percentage and significantly higher NDF and peNDF percentages in the diet of SO herds may also explain the low propionate concentration in the rumen and correspondingly higher acetate:propionate ratio (3.04), supporting previous studies (Russell, 1998).

Rumen fermentation in this category appears to have been slower and would favor more cellulolytic bacteria, such as *Fibrobacter succinogenes*, *Ruminococcus albus*, and *Ruminococcus flavefaciens*. Russell (1998) found that the substrate had a much greater impact on the fermentation end products than did pH. Cows fed 100% hay diets had an acetate:propionate ratio of 4.1 compared with a ratio of 2.2 for cows fed 90% concentrate. Cattle with higher acetate:propionate ratios had higher rumen pH values, which favored methane production in vitro (Lana et al., 1998). The significantly higher acetate and butyrate concentrations in category 2 than in category 1 may explain, in part, the significantly higher milk fat percentage in category 2 cows than in category 1 cows. Acetate and butyrate are utilized by mammary cells for energy and the production of milk fat, particularly triglycerides (Rook, 1976).

Both D-lactate and valerate concentrations in the rumen were significantly lower than in category 1. The lower D-lactate and higher pH indicates that either production of lactic acid was limited or that transfer rates of this from the rumen were sufficiently high that accumulation did not occur. More likely, production of lactic acid was less because of changes in the microbial population, such as a lower prevalence of *M. elsdenii*, resulting from lower dietary NFC and higher NDF percentages.

Mean rumen pH for this category was significantly higher than in category 1 but lower than in category 3. Although reductions in fiber digestion occur when pH falls below 6.2 (Grant and Mertens, 1992; Grant, 1994; Calsamiglia et al., 1999), a large decrease in fiber digestibility occurred only below a ruminal pH of 5.8 (de Veth and Kolver, 2001) in an in vitro study using high-quality pasture-based diets. Dry matter digestibility was optimized at pH 6.35. Therefore, it is unlikely that fiber digestion was compromised markedly for cows in this category.

Dietary CP percentage was not significantly different among the herd categories ACID, SO, and NA. All herd categories had adequate to excessive mean CP percentages based on NRC (2001) recommendations. There was also no significant difference between the undegradable protein and soluble protein fractions of the CP percentages among herds. Despite these similarities, ammonia concentrations were significantly higher in category 2 cows than in the other 2 categories ($P = 0.001$). Dietary protein is converted to ammonia by proteolytic bacteria; however, there may be a failure of N incorporation into microbial AA, and hence protein. Bacteria that primarily ferment structural carbohydrates use only ammonia as a N source, whereas NSC-fermenting bacteria use

peptides and ammonia as N sources (Russell et al., 1992). Conversion of ammonia to bacterial protein requires a number of cofactors, including VFA. The mean total concentration of VFA in this category indicates that sufficient energy was available for this process. Consequently, there may be a failure of N-energy matching in cows from this category, and this may reflect an asynchronous availability of nutrients for bacterial growth. One possibility is that the diets, although adequate on a daily basis, may be imbalanced in the supply of fermentable carbohydrates and N on an hourly basis (Shabi et al., 1998). Carbohydrate sources may also have been fed at different times in the day compared with protein sources. Feeding practices for the herds indicated that in most areas of the study, rapidly fermentable carbohydrates (wheat, triticale, barley) were fed more commonly at the time of milking than protein sources (lupins, canola meal, and urea) and in much higher quantities (Bramley, 2005). In most herds, the predominant protein source in the diet was pasture.

Cows were held off pasture routinely for 1 to 2 h twice daily and up to 6 h twice daily during milking (Bramley, 2005). Studies have reported mixed effects of asynchronous diets, with most studies examining the effects of feeding energy and protein sources with different degradabilities. Herrera-Saldana et al. (1990) found that cows fed rapidly fermentable carbohydrates (barley) and slowly fermentable protein (brewers grain) or slowly fermentable carbohydrates (grain sorghum) and rapidly fermentable protein (cottonseed meal) had higher rumen ammonia concentrations and lower microbial protein yields than those fed synchronized highly fermentable diets. Results in that study were, however, not significant and the numbers of cows used were low. Kolver and de Veth (2002) examined the effects of feeding pasture and concentrate either together or 4 h apart, with pasture fed first. Ruminal ammonia concentrations were significantly higher in cows fed the asynchronous diet compared with the synchronous diet 3 and 5 h after pasture was fed in the morning. Many milking parlor diets are predominantly or all cereal grain, a poor source of protein. The high concentration of ammonia in category 2 cows was not associated with a significantly lower milk protein percentage. Mean milk protein yield was, however, lower in this category than in category 1, suggesting that microbial protein production was lower in this group.

Category 3

Some of the mean rumen measures for category 3 indicate that cows in this category may have suboptimal rumen function. Rumen pH was 6.33 ± 0.43, but was coupled with a low total VFA concentration, a lower D-lactate concentration than in other categories, and adequate rumen ammonia concentrations. However, mean production for cows in this category was significantly higher than in category 2 ($P = 0.011$). A significantly lower milk fat percentage ($P = 0.022$) than in category 2 resulted in a milk fat:protein ratio of 1.18 ± 0.010, which was not significantly different from category 2.

None of the dietary intake variables for NA herds were significantly different from SO herds. Notwithstanding this, there were significant differences among many of the rumen parameters and production data. One possible explanation is the feeding regimens used in these herds. Although the overall dietary variables were similar, these cows may have a more synchronous diet presented throughout the 24-h period, allowing greater production. The lower total VFA concentration in this category may indicate a slower rumen fermentation relating to substrate offered compared with rumen fermentation in category 1 cows, or a rapid clearance from the rumen. The good production levels of this group suggest that VFA and microbial production from this group were good, but not optimal. It is also possible that ruminal clearance rates of VFA were higher for this group than for group 1 cows. Damage to ruminal villae, resulting in slower clearance of acids, may be a factor that may influence rumen measures and function.

CONCLUSIONS

These cows were exposed to a wide variety of diets, feeding regimens, and management strategies, which resulted in significant differences in rumen fermentation and milk production. However, despite the diversity of feeding strategies used in the study herds, the effect of herd explained only a little over half of the variation in category assignment.

Mean concentrations of almost every measure differed for each of the 3 categories identified. Category 1 cows had rumen measures consistent with a model of acidosis that reflected a rapid rate of carbohydrate fermentation, producing higher volumes of milk, but a low milk fat percentage. A point prevalence of 10% (95% credible interval, 8 to 12%) of cows with an acidotic profile indicates a high risk for acidosis in the cattle sampled. Category 2 describes cows with a possibly mismatched rumen fermentation rate, leading to lower milk production. Category 3 contains cows exposed to diets similar to those in category 2, but with lower concentrations of VFA and better milk production.

Outcomes for categories, as well as risk factors for these categories, were examined. Results from this

study are consistent with many intensive and in vitro studies conducted, but they present new insights into rumen fermentation for cows fed a wide range of diets. Cows in category 1 were more likely to be of lower parity, indicating that the risk of acidosis is higher for primiparous cows in pasture-fed systems. In particular, the study indicates the value of considering more measures of rumen function, rather than just pH, in determining cattle at risk for acidosis; it identifies the challenge between achieving high levels of milk production and maintaining cattle health; and it indicates that ruminal acidosis should be considered a complex of disorders rather than a single condition.

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