# DIETARY FACTORS INFLUENCING MILK YIELD AND MILK PROTEIN YIELD IN DAIRY COWS

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## INTRODUCTION

As a collective term, dry matter intake (**DMI**) is a major determinant of milk production in dairy cows (NRC, 2001). A survey of a large dataset including nutritional studies published in J. Dairy Sci. (vol. 1 through 82) showed a moderate linear regression fit ($R^2$ = 0.47) between DMI and milk yield (Hristov et al., 2000; Fig. 1). Dietary energy and crude protein, however, are not uniform entities. Feed evaluation and diet formulation models, using various procedures, have divided dietary crude protein and energy yielding substrates into fractions characterized by different rate and extent of ruminal degradation (Jarrige, 1989; Sniffen et al., 1992; NRC, 2001). It is likely, that dietary protein and carbohydrate fractions will have differential effects on milk and milk protein yields (**MY** and **MPY**, respectively) in dairy cows. Therefore, there is a need to investigate the relationships between specific dietary protein and carbohydrate fractions with production parameters as well as the possibility of using these relationships for prediction of MY and MPY in dairy cows.

The objective of this study was to investigate ten years of published nutritional studies, derive nutrient composition and intake data using two nutritional programs, determine correlation coefficients between specific dietary components and DMI, MY, and MPY, and determine dietary nutrients most responsible for the variation in MY and MPY in Holstein dairy cows.

## MATERIALS AND METHODS

### Input data

Diets (776) from feeding trials (229) conducted in the U.S. and Canada involving Holstein cows published in J. Dairy Sci. (volumes 73 through 82) were analyzed for nutrient composition using two diet evaluation programs, CPM Dairy (version 1.0, University of Pennsylvania, Kennett Square, PA, Cornell University, Ithaca, NY and William H. Miner Agricultural Research Institute, Chazy, NY; **CPM**) and NRC (2001; **NRC**). Diets having ingredients with

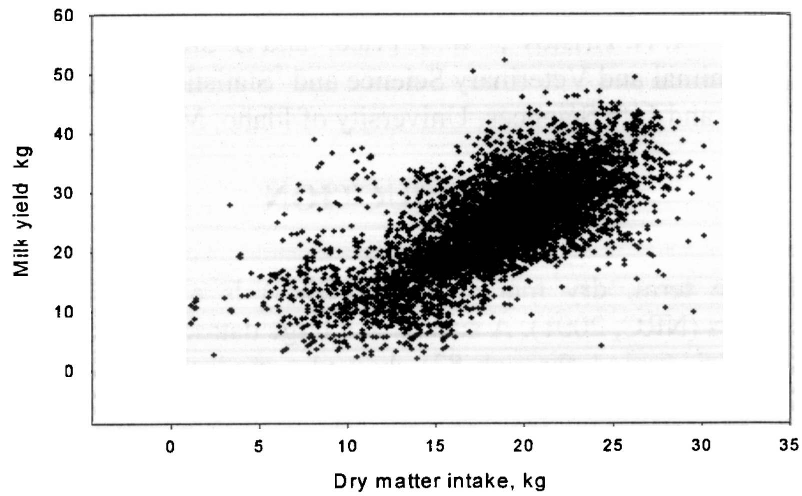


**Figure 1. Relationship between DMI and milk yield in dairy cows (n = 5814). Data from Hristov et al. (2000).**

unknown composition, effects on the composition, or utilization of the overall diet (e.g., ionophores, silage inoculants, etc.) were not used. From these trials, only the untreated, control diet was utilized in the analysis. Feeds (and level of processinglisted in the publications were matched with feeds from feeding model libraries. The chemical composition of dietary energy and proteinaceous concentrates was not edited. Where available, CP and NDF content of forages were modified to reflect actual dietary concentrations. Cows were grouped into two categories according to DIM: early-lactation cows (DIM < 100, 467 observations; **ELC**) and mid- and late-lactation cows (DIM ≥ 100, 286 observations; **MLLC**). The earliest DIM was 16, except in three observations where DIM was 3, 5, and 5. In 23 observations DIM was not available and hence, these observations were omitted from the analysis. Trials, in which DMI exceeded 30 kg/d (4 observations) or milk yield was less than 20 kg/d (19 observations) were not considered in the analysis. The average (∀SE) DMI of the cows involved in this study was 22.0∀0.09 kg/d with a minimum of 12.2 and a maximum of 29.9 kg/d. The average MY was 31.5∀0.20 kg/d and had a minimum and maximum of 20.1 and 46.4 kg/d, respectively. Body weight of experimental cows was published in 556 of the observations used in the analysis: 366 observations for ELC and 164 observations for MLLC. Mean BW was 602∀2.0 kg (minimum 476, maximum 750 kg). The dietary variables derived from the feeding programs are shown in Table 1. The response variables of interest were MY and MPY. Both, the relative concentration in dietary DM and daily nutrient intake variables were investigated. In addition, ratios between protein and energy variables and BW were considered for having potential relationships with MY and MPY.

A similar data set was obtained from nutritional studies published in vol. 83 of J. Dairy Sci. for model validation. A total of 68 observations were used in the validation process; 48

observations involved ELC and 20 – MLLC cows. Cow BW was published for 58 of the observations.

### Statistical analyses

**Variable Selection**: As an initial step in the data analysis, each data set was inspected for similarity among the nutritional variables. Since these data were derived from computational nutritional programs, some variables were essentially numeric derivatives of others, and thus, were redundant. This situation could be quickly assessed by calculating simple linear correlations among the nutritional variables. Variable pairs having correlation values larger than 0.80 to 0.90 were considered to indicate a redundancy. In such cases, only one variable of the pair was selected for further consideration. Preference for one variable over the pair was given based on biological relevance or reliability of the analytical procedure used to derive the variable. For example, in the CPM-derived dataset, preference was given to NDF over effective NDF (r = 0.85) and to NSC over CHO fraction B1 (r = 0.97). Certain variables were derived from each other (ME and NEL, for example) and would consequently give identical model estimates.

*For the remaining variables, the correlation matrix was subjected to a Principal Components Analysis (**PCA**) procedure. In PCA, the goal is to reduce the dimension of a large multivariate data set by identifying linear combinations of the variables, i.e. PCA axes, which uniquely partition the variability of the data. Typically, the first two or three PCA axes will account for a majority of the variability in the data. Within each axis, every variable is assigned a coefficient or loading according to its contribution to the axis. The relationships among the variables and their relative dominance can then be inferred by examining the magnitude of the coefficients. Variables showing large loading values (coefficients) are considered more influential than those with smaller values, therefore, would be selected as good candidates for the subsequent modeling process.*

In addition to the PCA, correlation coefficients between DMI and the response variables and the nutritional variables were assessed. Nutritional variables showing strong correlations with the responses ($r \geq 0.70$) were considered as good candidates for modeling. Through these screening processes, a final set of potential modeling variables was selected for each of the responses, MY and MPY in the ELC and MLLC groups.

**Modeling**: While the redundancy among the nutritional variables was addressed in the initial variable selection process, the remaining variables still had sizable correlations with one another. When correlated variables are used as regressors in the same model, it will result in dependency among regression variables (collinearity), which will in turn produce biased estimates and inflated standard errors. To avoid this situation, regression models included only nutritional variables with relatively small correlations. This resulted in several candidate models for each response.

*A further consideration for the modeling process concerned the nature of the data. The nutritional data presented here was collected from multiple independent studies conducted over a large range of years. When a meta-analysis of this type is carried out, the random* *effects of multiple studies should be accounted for (St-Pierre, 2001). Failure to do so can result in estimates with significant bias and poor precision. Thus, a mixed model was used to incorporate the random study effects having the general form:*

$$\mathbf{Y} = \mathbf{X}\exists + \mathbf{Z}( + „ )$$

where Y was the response being modeled, X was a matrix of the nutritional variables and ∃ was a vector of the regression coefficients. These terms represent the fixed portion of the model and are equivalent to those found in a standard multiple linear regression. The additional components, Z and (, accounted for the random effects due to the various studies. Z represented either a portion or all of the variables present in X, and ( was a vector of the parameters corresponding to Z. , was the random error term assumed to be normally distributed with a mean equal to 0 and a constant variance.

*Estimation was carried out using the method of restricted maximum likelihood (**REML**) considering the random model components as unstructured. This allowed for a covariance structure among the regressors. Model fitting and assessment was done separately for each nutritional program and DIM group. All estimated models were assessed and a final model for each response was selected based on fit, parsimony, parameter significance, and biological relevance. The adequacy of final models in each case was further evaluated through residual analysis.*

**Validation**: As a last measure of the performance for each model, a validation procedure was carried out on an independent data set collected from vol. 83 of J. Dairy Sci. Residual values between the estimated models and the validation data were computed and assessed as to their distribution, structure, and magnitude.

All statistical computations were carried out using SAS (1999).

## RESULTS

The main dietary ingredients and summary statistics are shown in Table 2. Corn silage, alfalfa silage and hay were the predominant forage components of the diets with a mean content of 32, 39, and 27% of dietary DM, respectively. The dataset included diets varying widely in forage to concentrate ratio. Forage content in dietary DM was as high as 78% (corn silage), 81% (alfalfa silage), or 100% (whole alfalfa hay diets). Energy concentrate was mostly corn (603 diets) with 97 diets having barley as main energy concentrate. Corn or barley grain represented approximately 25% of dietary DM. Maximum inclusion of grain was from 53 (high-moisture corn) to 62% (barley) of DM. There was less variation in the source of ruminally available protein, with solvent-extracted soybean meal being the main protein supplement, included in 583 diets. Corn gluten meal, fish meal, blood meal, and meat-bone meal were the sources of ruminally undegradable protein. Animal fat and urea were supplemented to 111 and 114 of the diets studied, respectively.

Chemical composition of the diets was estimated using NRC (2001). CP content averaged 17.8% of DM and ranged from 10.3 to 28.1%. The diets represented a wide range of ruminal protein degradability: from 5.8 to 22.2 % of DM. Converted to DM basis, metabolizable protein (**MP**) content of the diets varied from 6.9 to 16.3%. The energy content of the diets was comparatively more consistent: average NEL concentration was 1.61 Mcal/kg DM and varied from 1.32 to 1.96 Mcal/kg. Neutral-detergent fiber and ADF concentrations varied from 18 to 54% and from 12 to 35% of DM, respectively. Total digestible nutrients concentration, discounted for level of intake (**DTDN**), was 65.3% of dietary DM on average with the maximum TDN diet having 34% more TDN than the lowest. In NRC (2001), non-fiber carbohydrates (**NFC**) represent primarily the starch content of the diet, but also include pectins, sugars, and organic acids. The diets included in this study had 41.6% NFC content (DM basis) on average and varied from 14 to 56% NFC. Phosphorus and particularly Ca concentrations varied significantly among the diets: from 0.30 to 0.90 and from 0.20 to 1.70% of DM, respectively.

Protein and carbohydrate (**CHO**) dietary fractions were estimated using CPM Dairy. The diets encompassed a significant range of soluble protein, and fractions A, B1, and B2 protein. On average, the diets studied had 37.2% crude protein solubility, 25.7% NPN, 11.6% soluble protein (B1 protein), and 47.5% available insoluble protein content (as % of dietary CP). Concentration of CHO fractions A, B1, and B2 averaged 7.0, 52.8, and 28.3% (of the total dietary CHO), respectively and, similar to the protein fraction, diets varied significantly in carbohydrate composition. Average fermentability of dietary CHO was 41.2%.

Simple correlations between the potential regressor variables and DMI, MY, and MPY are shown on Tables 3 and 4 (CPM and NRC, respectively). Correlations between composition variables and DMI, MY, and MPY were usually low, regardless of the nutritional program used. Comparatively higher, negative correlations were found between NDF content of the diet (% of DM) and MY and MPY. Both program libraries indicated a similar NDF content and the respective correlation coefficients for MY and MPY were similar (-0.29 and –0.33 and –0.29 and –0.34, CPM and NRC, respectively). CPM also produced comparatively high (and negative) correlations between fermentable CHO fractions and DMI, MY, and MPY. Correlations between the intake variables and the response variables were higher for both nutritional programs relative to those of the composition variables. The two nutritional programs differed in their estimates of the relationship between dietary energy intake and the response variables, which most likely represents the new approach in estimating energy content/TDN of feeds in NRC (NRC, 2001). The correlations between energy content of the diet and MY and MPY were high, but similar between the two programs. Intake of CP (both programs), MP and MP from bacteria (NRC), NSC and NFC (CPM and NRC, respectively), TDN (NRC), CHO fraction B1 (CPM), and amino acid flow to the intestine (NRC) were moderately correlated to MPY and MY.

Due to the poorly defined relationships between MY and MPY and the composition variables, only the intake variables were considered for further investigation.

## CPM Dairy Program

The CPM PCA results are given in Table 5. For both ELC and MLLC, the first three PCA axes accounted for 73.0 and 73.6% of the variability, respectively. The first axis, representing the primary source of variability, was dominated by intake variables NEL, NSC, CHO fraction B1, and fermentable total CHO, as indicated by the relative magnitude of the coefficients. These are generally energy (predominantly starch)-related components of the diet. The second PCA axis indicated that intake of NDF, effective NDF, and fermentable NDF were important variables, which are related to dietary fiber intake. The third axis had high loadings for RDP, soluble protein, and protein fractions A and B1 intakes, or protein-related dietary variables.

Utilizing these variables, several candidate models were assessed with the mixed model approach. Assessment of multiple models was necessary to avoid collinearity between certain regressors such as NDF and effective NDF. The fit of each model was assessed using the Bayesian Information Criteria (**BIC**) with those models having the lowest BIC value being selected as best.

Table 6 gives parameter estimates for MY and MPY for both ELC and MLLC cows. In all models, the parameter estimates were significant and residual analysis showed no trends, patterns, or bias. The predicted values followed the observed data points well and indicated good fit.

The MY/ELC model included NEL and CP intakes and BW. Energy intake accounted for the majority of the variability. This model explained 42.4% of the variation in milk yield. Variables in the MPY model were NEL intake and RDP intake. Similar to the MY model, NEL intake accounted for the largest portion of the variability with the MPY model accounting for 44.9% of the variation of MPY. Both models included a carbohydrate and a protein component. Cow BW was significant only in the MY model for ELC.

The components of the MY model in MLLC were identical to those for ELC: NEL intake, CP intake, and BW with NEL intake being the dominant factor relative to the other regressor variables. The model accounted for 38.8% of the variation for MY of MLLC cows. Milk protein yield was best predicted by NEL intake and RUP intake, with 47.3% of the variation in MPY explained. A larger proportion of the variation in MPY was explained by NEL intake. As was seen in ELC, the models for MLLC included carbohydrate/energy component and a protein component.

Models for MY and MPY predicted well in ELC validation data, but slightly under predicted MY and particularly MPY in MLLC. Residual means (∀SE) for MY and MPY were 0.553∀0.2092 and 0.708∀0.2359 in ELC and 0.996∀0.2545 and 1.251∀0.2397 in MLLC, respectively. Residual plots in these cases indicated a random pattern, however on average, the residuals were greater than zero suggesting some potential bias for these models.

## NRC (2001) Program

The PCA results for the NRC program are given in Table 7. As was the case with CPM, three PCA axes were sufficient to describe the majority of the intake variability for both ELC and MLLC (81.5 and 78.7%, respectively). The first set of axes contained variables representing dietary protein and energy components: MP, MP from bacteria and from RUP (**MPRUP**), amino acids flow, NEL, and DTDN (both ELC and MLLC). The second and third axes had large loadings for ME, Ca, NDF, forage NDF, and NFC. Unlike CPM, NRC axes were not well defined and tended to represent a mix of energy, protein, and other variables.

These variables were again incorporated into a series of mixed effects regression models to investigate their relationships with MY and MPY in ELC and MLLC cows. Similar to CPM, the parameter estimates were significant and residual analysis showed no trends, patterns, or bias. The predicted values followed the observed data points well and indicated a good fit.

When diets were analyzed using NRC, NDF, NEL and P intakes, and BW defined the best model for MY in ELC (Table 8). Relatively, NEL intake accounted for most of the variation. The estimate for NDF intake was negative, indicating an inverse effect of this dietary parameter on MY. This was the only model, in which intake of a mineral (P) accounted for a significant portion of the variation, however, only 44.6% of the overall variability in MY was accounted for. Intake of DTDN, NDF, and MPRUP determined MPY in ELC. This model was dominated by DTDN intake and accounted for 44.5% of the variation in MPY in ELC. Energy intake was the only regressor in the MY and MPY models in MLLC cows. These models explained 40.7 and 45.6% of the variability in MY and MPY, respectively, in MLLC.

The NRC MY and MPY models predicted MY well and slightly under predicted MPY in ELC in the validation data set. Both MY and MPY under predicted in MLLC: mean residuals (∀SE) were 0.468∀0.2043 and 0.958∀0.2413 for ELC and 1.274∀0.2706 and 1.083∀0.2520 for MLLC, respectively. Similar to the CPM validation, the residuals for these models were acceptable in distribution, scatter, and pattern, but appeared to indicate some potential bias.

## DISCUSSION

The diets included in this study encompassed a large variety of feeds (a total of 88), but the majority utilized ingredients, which were typical for North American dairy diets, i.e. alfalfa silage and hay, corn silage, corn grain, and soybean meal. Therefore, the results from this meta-analysis will be most applicable to Holstein cows fed alfalfa/corn/soybean meal-based diets.

Regarding chemical composition, the diets represented a large variation in protein, energy, fiber, starch, and DTDN content. Diets with CP content between 16.2 and 19.8% of DM represented 72.3% of the total while those with NEL concentration of 1.525 to 1.675 Mcal/kg DM represented 73.6% of the total. RDP and RUP contents of 10.2 to 13.8 and 4.5 to 6.3% of DM represented 68.0 and 60.4% of the total diets. Diets with NDF and forage NDF content of 25.0 to 32.5 and 16.0 to 28.0% of DM were 64.8 and 78.6% of the total (41.9% of the diets had forage NDF content between 20 and 24% of DM) and diets with ADF content of 17.3 to 21.8% of DM were 56.5% of the total. Calculated discounted TDN content varied less among diets; 75% of the diets had DTDN concentration between 63.0 and 67.5%. A similar variation in dietary CP, NDF, and ADF content from a comparatively smaller dataset was reported by Holter et al. (1997). This composition of diets is in line with practical levels of feeding and should ensure the results from this study are applicable to most diets fed to dairy cows in North America.

*Correlations between DMI, MY, and MPY and composition variables, with the exception of some relationships between MY and MPY and NDF and fermentable NDF content of the diet, were poor. As a result, a reasonable model to predict production parameters based on diet composition was not derived. In contrast, Holter et al. (1997) found comparatively higher correlations of dietary CP with MY and MPY (r = 42 and 38%, respectively); dietary fiber variables (NDF, ADF, and forage NDF) were highly and negatively correlated to DMI, MY, and MPY (r = -44, -69, and –68% and –43, -72, and –70% for NDF and ADF, respectively). The authors reported similar results for Jersey cows (Holter et al., 1996). In an overview of 93 nutrition studies (10 years of published data), Moloi (1998) found an approximately exponential decrease in MPY with increasing NDF content of the diet up to 34% of DM. Data summarized by Allen (2000) suggested decreased DMI with increasing NDF content of the diet. In this latter study, CP content did not produce an apparent effect on DMI. Our data indicated that MY and MPY were negatively affected by NDF and effective NDF or forage NDF content of the diet (both CPM and NRC programs). Negative effects of fermentable starch (CHO fraction B1) and fermentable total CHO and NDF (fractions B2 + C) were also present when diets were analyzed using CPM.*

*In general, however, intake variables had a much stronger correlation to MY and MPY. Energy intake and TDN intake corrected for the level of DMI (NRC) were the only dietary intake variables that were highly correlated with MY and MPY. Correlations of intake of NDF, effective NDF, and forage NDF with MY and MPY were either low and negative or non-significant. This corresponds to the previously observed negative effect of fiber concentration in the diet on production parameters (Holter et al., 1997; Moloi, 1998; Allen, 2000). Amino acids flowing to the small intestine are major precursors for and determine milk protein synthesis in dairy cows (NRC, 2001). The comprehensive amino acid submodel of NRC (NRC, 2001) predicts amino acid flow to the intestine based on RUP and microbial amino acid flows, thereby reflecting the overall digestibility of the diet (correlation between DTND intake and amino acid flow was 0.82). Consequently, good correlations between NRC predicted amino acid flow and the response variables were not unexpected in this study.*

*The models derived for MY and MPY using CPM or NRC reflected the relationships existing between the dietary variables and production parameters in this analysis. Milk yield and MPY were mostly determined by energy intake or energy and DTDN intakes as a measure of digestible energy intake in the NRC-based model: the correlation coefficient between NEL intake and DTDN intake was 98.7% (over all observations). In both programs, NEL or DTDN intake were the primary regressor variables when modeling MY and MPY regardless of the stage of lactation. Moloi (1998) found that diet-related variables, such as degradable intake protein, NDF, and NFC content of the diet, impact MPY in dairy cows. Energy content of the* *diet, however, was not found to be related to MPY and no single variable was distinguished as having major influence on MPY. Crude protein, RDP or RUP intakes were factors in MY and MPY when diet composition was determined with CPM (both lactation phases). Unlike CPM, no protein fractions were found to influence MY in ELC when diet composition was determined using NRC, except for MPRUP in the case of MPY. Another difference between the two nutritional programs was the presence of NDF intake in both MY and MPY models for ELC when diets were analyzed with NRC while MY and MPY in MLLC were best explained by NEL intake alone. The lack of a protein component in the MPY/MLLC model may demonstrate the differences in the essential amino acids requirements between the early and late stages of lactation in the dairy cow (Schwab, 1995).*

*The strong presence of energy intake in the MPY models indicates the critical importance of available energy for MY and MPY in the dairy cow. The unique digestive tract of the ruminant animal results in atypical conversion of dietary energy-yielding substances into products other than glucose (Tyrrell, 1980). Thus, the majority of the digestible energy in the cow is derived from VFA: acetate provides from 25 to 35, propionate – from 15 to 30, and butyrate – from 8 to 15% of the digestible energy. Little digestible energy is derived directly from carbohydrates (5 to 10%) and the contribution of amino acids can be substantial (15-25%) (AFRC, 1998). Milk volume is lactose-related and glucose is the main precursor of milk lactose (Mepham, 1982). Consequently, MPY, as a function of milk volume, is likely to be related to glucose flow to the mammary gland. Energetically, glucose and acetate are accounting for up to 58% of the $CO_2$ production in the udder indicating the importance of the carbohydrate component of the diet as metabolic fuel within the mammary gland (AFRC, 1998).*

*Diets analyzed with NRC showed milk production in cows with DIM less than 100 days to be affected by P level in the diet (P intake). This effect is somewhat surprising in the light of recently published research showing no negative effects from reduced P feeding on milk production in Holstein cows (Wu and Satter, 2000). Phosphorus is an essential macro-mineral in ruminant nutrition and its deficiency may negatively affect microbial protein synthesis in the rumen (Durand and Kawashima, 1980; Petri et al., 1988) although the levels fed in the studied diets (average P concentration of 0.44% of DM) were significantly higher than the 0.38% recommended by Wu and Satter (2000).*

*Models for MY and MPY predicted validation data moderately well in ELC, but over estimated MY and MPY in MLLC. In all cases, the validation residuals were random with no visual trends. This indicated that the slopes (rates of response to the dietary variables) of the relationships were reasonable, but in some cases, the models were biased downwards in magnitude. The lack of validation for MLLC could be partially due to fewer observations both in model fitting and validation data sets. Best-fit models based on NRC-derived diets had only one regressor (NEL intake) and were particularly biased toward cows in the post-peak lactation stage, suggesting that some other factors may have been necessary for modeling in these cases.*

## *CONCLUSIONS*

Analysis of the data indicated that relationships between dietary nutrients and milk yield and milk protein yield were not clearly defined in dairy cows. Depending on the program used to derive nutrient composition of the diet, net energy of lactation intake or total digestible nutrients intake was the most important variable for predicting milk and milk protein yields. The derived models generally under predicted the validation data.

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**Table 1. Dietary variables investigated**

| Variable[2] | Feeding model[1] CPM Dairy | NRC 2001 |
|---|---|---|
| Crude protein (CP), % DM | X | X |
| Rumen degradable protein (RDP), % DM | X | X |
| Rumen undegradable protein (RUP), % DM | X | X |
| Soluble protein (SP), % DM | X | - |
| Metabolizable protein (MP), % DM | X | X |
| MP form feed | X | X |
| MP from bacteria | X | X |
| Protein fraction A, % of total | X | - |
| Protein fraction B1, % of total | X | - |
| Protein fraction B2, % of total | X | - |
| Metabolizable energy, Mcal/ kg DM | X | X |
| NEL, Mcal/kg DM | X | X |
| NDF, % of DM | X | X |
| Forage NDF, % of DM | - | X |
| Effective NDF, % of NDF | X | - |
| ADF, % of DM | - | X |
| Non-structural CHO[3], % of DM | X | - |
| Non-fiber CHO, % of DM | - | X |
| CHO fraction A, % of total | X | - |
| CHO fraction B1, % of total | X | - |
| CHO fraction B2, % of total | X | - |
| Fermentable CHO fraction B1, % | X | - |
| Fermentable NDF (B2 +C), % | X | - |
| Fermentable total CHO, % | X | - |
| Discounted TDN, % of DM | - | X |
| Amino acids flow to the intestine, g/d | - | X |
| Digestible amino acids flow, g/d | - | X |
| Fat , % of DM | - | X |
| Ca and P, % of DM | - | X |

[1] X = present; - = not present.
[2] Both, concentration in dietary DM and daily intake were investigated.
[3] Carbohydrate

**Table 2. Main ingredients of the investigated diets along with their associated number of observations, average, minimum, maximum, and standard deviation (SD)**

| Variable | No. of Diets | Average | Min | Max | SD |
|---|---|---|---|---|---|
| | | % of DM or as indicated | | | |
| Ingredients | | | | | |
| Alfalfa silage | 377 | 39 | 5 | 99 | 20.2 |
| Alfalfa hay | 223 | 27 | 4 | 81 | 17.5 |
| Corn silage | 432 | 32 | 4 | 78 | 14.4 |
| Corn grain, coarse ground | 477 | 26 | 1 | 58 | 11.4 |
| High-moisture corn | 126 | 27 | 0.1 | 53 | 11.0 |
| Barley grain, rolled | 117 | 26 | 2 | 62 | 16.0 |
| Dry distiller grains | 97 | 8 | 0.2 | 36 | 7.6 |
| Corn gluten meal | 124 | 3.6 | 0.6 | 21 | 3.1 |
| Soybean, hulls | 113 | 8 | 0.2 | 34 | 6.7 |
| Soybean meal, solvent | 583 | 9 | 0.1 | 27 | 5.4 |
| Whole cottonseed | 132 | 10 | 0.5 | 20 | 3.1 |
| Fish meal | 92 | 4 | 0.8 | 19 | 3.3 |
| Blood meal | 116 | 2 | 0.2 | 8.7 | 1.4 |
| Meat-bone meal | 107 | 2 | 0.2 | 8 | 1.6 |
| Molasses | 170 | 2 | 0.4 | 8 | 1.4 |
| Animal fat | 111 | 3 | 0.1 | 8 | 1.6 |
| Urea | 114 | 0.5 | 0.02 | 1.9 | 0.38 |
| Composition[1] | | | | | |
| CP | | 17.8 | 10.3 | 28.1 | 2.22 |
| RDP | | 12.2 | 5.8 | 22.2 | 2.09 |
| RUP | | 5.6 | 2.9 | 11.6 | 1.22 |
| MP | | 10.3 | 6.9 | 16.3 | 1.11 |
| ME, Mcal/kg DM | | 2.54 | 2.14 | 2.96 | 0.096 |
| NEL, Mcal/kg DM | | 1.61 | 1.32 | 1.96 | 0.075 |
| NDF | | 30.6 | 18.2 | 53.7 | 4.78 |
| ADF | | 20.4 | 11.9 | 35.0 | 3.63 |
| NFC[2] | | 41.6 | 17.4 | 56.2 | 5.60 |
| Discounted TDN | | 65.3 | 55.9 | 74.7 | 2.35 |
| Ca | | 0.61 | 0.20 | 1.70 | 0.274 |
| P | | 0.44 | 0.30 | 0.90 | 0.078 |
| Carbohydrate and protein fractions[3] | | | | | |
| Soluble protein, % of CP | | 37.2 | 17.5 | 60.0 | 8.04 |
| Protein fraction A, % of CP | | 25.7 | 9.0 | 52.0 | 7.40 |
| Protein fraction B1, % of CP | | 11.6 | 1.0 | 23.0 | 3.64 |
| Protein fraction B2, % of CP | | 47.5 | 19.0 | 69.0 | 9.05 |
| CHO[4] fraction A, % of total | | 7.0 | 2.0 | 16.0 | 1.92 |
| CHO fraction B1, % of total | | 52.8 | 21.0 | 72.0 | 7.41 |
| CHO fraction B2, % of total | | 28.3 | 11.0 | 57.0 | 7.40 |
| Total fermentable CHO, % DM | | 41.2 | 27.5 | 53.4 | 3.87 |

[1] NRC, 2001.
[2] Non-fiber carbohydrates.
[3] CPM Dairy.
[4] Carbohydrate.

**Table 3. Coefficients of correlation between dietary variables and DMI, MY, and MPY (all observations, CPM Dairy)**

| | Composition[1] | | | Intake[2] | |
|---|---|---|---|---|---|
| Variable | DMI | MY | MPY | MY | MPY |
| Crude protein (CP) | NS | 0.14 | 0.13 | 0.52 | 0.54 |
| Rumen degradable protein (RUP) | -0.09 | NS | NS | 0.41 | 0.43 |
| Rumen undegradable protein (RDP) | 0.12 | 0.16 | 0.15 | 0.43 | 0.44 |
| *Soluble protein (SP)* | *0.02* | *0.10* | *NS* | *0.34* | *0.32* |
| Metabolizable protein (MP) | NS | NS | NS | 0.43 | 0.46 |
| MP form feed | 0.13 | 0.17 | 0.20 | 0.41 | 0.45 |
| MP from bacteria | -0.20 | -0.22 | -0.23 | 0.23 | 0.26 |
| Protein fraction A | NS | NS | NS | 0.26 | 0.22 |
| Protein fraction B1 | 0.19 | 0.11 | 0.14 | 0.32 | 0.34 |
| Protein fraction B2 | NS | NS | NS | 0.35 | 0.43 |
| Metabolizable energy | NS | 0.17 | 0.19 | 0.60 | 0.64 |
| Net energy of lactation | NS | 0.18 | 0.19 | 0.60 | 0.64 |
| NDF | -0.09 | -0.29 | -0.33 | 0.11 | 0.10 |
| Effective NDF | -0.12 | -0.28 | -0.32 | -0.10 | -0.13 |
| Non-structural CHO[3] | NS | 0.11 | 0.19 | 0.45 | 0.54 |
| CHO fraction A | NS | NS | -0.08 | 0.21 | 0.15 |
| CHO fraction B1 | 0.09 | 0.20 | 0.27 | 0.42 | 0.52 |
| CHO fraction B2 | -0.14 | -0.28 | -0.30 | NS | NS |
| Fermentable CHO fraction B1 | -0.11 | -0.33 | -0.36 | 0.32 | 0.42 |
| Fermentable NDF (B2 +C) | -0.30 | -0.35 | -0.38 | -0.12 | -0.12 |
| Fermentable total CHO | -0.33 | -0.30 | -0.26 | 0.30 | 0.37 |

[1] Percentage of dietary DM or concentration (Mcal/kg DM, for example). DMI, dry matter intake; MY, milk yield; MPY, milk protein yield. Significant at $P < 0.05$; NS – non-significant.
[2] Intake of a nutrient, g/d or Mcal/d.
[3] Carbohydrate.

**Table 4. Coefficients of correlation between dietary variables and DMI, MY, and MPY (all observations; NRC, 2001)**

| Variable | Composition[1] | | | Intake[2] | |
|---|---|---|---|---|---|
| | DMI | MY | MPY | MY | MPY |
| Crude protein (CP) | NS | 0.15 | 0.14 | 0.53 | 0.54 |
| Rumen degradable protein (RUP) | NS | 0.11 | 0.09 | 0.44 | 0.45 |
| Rumen undegradable protein (RDP) | NS | 0.11 | 0.07 | 0.37 | 0.39 |
| Metabolizable protein (MP) | NS | 0.13 | 0.15 | 0.53 | 0.57 |
| MP form feed | | | - | 0.37 | 0.41 |
| MP from bacteria | - | - | - | 0.59 | 0.64 |
| Metabolizable energy | -0.37 | NS | NS | 0.60 | 0.64 |
| Net energy of lactation | -0.34 | NS | NS | 0.61 | 0.65 |
| NDF | -0.14 | -0.29 | -0.34 | 0.13 | 0.11 |
| Forage NDF | -0.23 | -0.24 | -0.32 | 0.13 | NS |
| ADF | NS | -0.09 | -0.18 | 0.26 | 0.20 |
| Non-fiber CHO[3] | 0.11 | 0.10 | 0.19 | 0.42 | 0.52 |
| Discounted TDN | -0.34 | NS | NS | 0.59 | 0.63 |
| Amino acid flow to the intestine | - | - | - | 0.54 | 0.58 |
| Digestible amino acid flow | - | - | - | 0.53 | 0.58 |
| Fat | NS | 0.24 | 0.17 | 0.41 | 0.35 |

[1] Percentage of dietary DM or concentration (Mcal/kg DM, for example). DMI, dry matter intake; MY, milk yield; MPY, milk protein yield. Significant at $P < 0.05$; NS – non-significant.
[2] Intake of a nutrient, g/d or Mcal/d.
[3] Carbohydrate.

**Table 5. Summary of Principal Component Analysis of CPM-derived model variables**

| Variable[2] | ELC[1] Axis loadings | | | MLLC Axis loadings | | |
|---|---|---|---|---|---|---|
| | PCA1 | PCA2 | PCA3 | PCA1 | PCA2 | PCA3 |
| CP | 0.2432 | 0.1553 | 0.2716 | 0.2044 | 0.2095 | 0.2049 |
| RUP | 0.2108 | 0.2020 | -0.0064 | 0.1835 | 0.1300 | -0.1364 |
| RDP | 0.1898 | 0.0800 | 0.3884 | 0.1461 | 0.1883 | 0.3607 |
| Soluble protein | 0.1222 | 0.0272 | 0.4706 | 0.0622 | 0.1431 | 0.4568 |
| Metabolizable protein (MP) | 0.2666 | 0.1858 | -0.1687 | 0.2311 | 0.2416 | -0.2121 |
| MP from bacteria | 0.2166 | 0.0762 | -0.1600 | 0.1609 | 0.2422 | -0.0457 |
| MP from RUP | 0.2025 | 0.1792 | -0.1162 | 0.2009 | 0.1374 | -0.2314 |
| Protein fraction A | 0.0839 | 0.0233 | 0.4294 | 0.0147 | 0.1484 | 0.3886 |
| Protein fraction B1 | 0.1462 | 0.0270 | 0.3452 | 0.1176 | 0.0522 | 0.3219 |
| Protein fraction B2 | 0.2018 | 0.1216 | -0.1116 | 0.2248 | 0.1214 | -0.1992 |
| NEL | 0.3065 | 0.0323 | -0.0603 | 0.3000 | 0.0611 | -0.0541 |
| NDF | 0.0919 | 0.4024 | 0.0216 | -0.0981 | 0.4090 | -0.0567 |
| Effective NDF | -0.0223 | 0.3955 | 0.0320 | -0.1946 | 0.2880 | -0.9113 |
| Non-structural carbohydrate | 0.2982 | -0.1931 | -0.0593 | 0.3072 | -0.0856 | -0.0227 |
| CHO fraction B1 | 0.2833 | -0.2186 | -0.0806 | 0.2989 | -0.1377 | -0.0503 |
| Fermentable total CHO | 0.2686 | 0.0148 | -0.2112 | 0.2493 | 0.1355 | -0.1207 |
| Fermentable NDF (B2 + C) | -0.0342 | 0.3854 | -0.2042 | -0.1146 | 0.2996 | -0.2611 |
| Fermentable CHO fraction B1 | 0.2651 | -0.2087 | -0.1217 | 0.2916 | -0.0866 | -0.0110 |
| Variability explained, % | 36.8 | 20.4 | 15.8 | 39.3 | 18.4 | 15.9 |
| Total explained | | 73.0 | | | 73.6 | |

[1] ELC – early lactation cows (DIM < 100); MLLC - mid- and late-lactation cows (DIM > 100).
[2] Intake variables.

**Table 6. Estimates, standard errors and significance for the fixed effects of the final candidate models under the CPM Dairy program. Response variables were MY and MPY in early and mid-to-late lactation cows**

| Variable[1] | Estimate | SE | DF | t value | P |
|---|---|---|---|---|---|
| | Early-lactation cows (DIM < 100) | | | | |
| MY | | | | | |
| Intercept | 0.3889 | 2.9134 | 115 | 0.13 | 0.8940 |
| NEL | 0.3942 | 0.04195 | 88 | 9.40 | <0.0001 |
| CP | 1.1438 | 0.30880 | 65 | 3.70 | 0.0004 |
| BW | 0.0233 | 0.00462 | 94 | 5.05 | <0.0001 |
| MPY | | | | | |
| Intercept | 0.2777 | 0.04660 | 141 | 5.96 | <0.0001 |
| NEL | 0.0172 | 0.00125 | 107 | 13.74 | <0.0001 |
| RDP | 0.0340 | 0.01122 | 79 | 3.03 | 0.0033 |
| | Mid- and late-lactation cows (DIM > 100) | | | | |
| MY | | | | | |
| Intercept | 22.1995 | 4.77190 | 51 | 4.65 | <0.0001 |
| NEL | 0.3916 | 0.08975 | 34 | 4.36 | <0.0001 |
| CP | 0.7714 | 0.42510 | 75 | 1.81 | 0.0735 |
| BW | -0.01714 | 0.00631 | 75 | -2.71 | 0.0082 |
| MPY | | | | | |
| Intercept | 0.3686 | 0.06672 | 85 | 5.52 | <0.0001 |
| NEL | 0.0136 | 0.00179 | 58 | 7.59 | <0.0001 |
| RUP | 0.0521 | 0.01119 | 122 | 4.66 | <0.0001 |

[1] MY and MPY, Milk yield, and Milk protein yield; NSC, NDF, CP, NEL, RDP, RUP, and BW, Non structural carbohydrate intake, NDF intake, CP intake, NEL intake, RUP intake, and RDP intake.

**Table 7. Summary of Principal Component Analysis of NRC-derived model variables**

| | ELC[1] Axis loadings | | | MLLC Axis loadings | | |
|---|---|---|---|---|---|---|
| Variable[2] | PCA1 | PCA2 | PCA3 | PCA1 | PCA2 | PCA3 |
| CP | 0.2422 | 0.1996 | 0.0596 | 0.2381 | 0.1777 | 0.3127 |
| RUP | 0.2285 | -0.1543 | 0.3938 | 0.2337 | -0.1939 | -0.0977 |
| RDP | 0.1745 | 0.3276 | -0.1331 | 0.1563 | 0.3274 | 0.4349 |
| Metabolizable protein (MP) | 0.2803 | -0.1070 | 0.1534 | 0.2855 | -0.1157 | -0.0447 |
| MP from bacteria | 0.2697 | 0.0697 | -0.2211 | 0.2715 | 0.1252 | 0.0811 |
| MP from RUP | 0.2297 | -0.2003 | 0.3567 | 0.2306 | -0.2475 | -0.1046 |
| Amino acids flow, g/d | 0.2819 | -0.0861 | 0.1272 | 0.2888 | -0.0777 | -0.0082 |
| NEL | 0.2772 | -0.0373 | -0.1870 | 0.2804 | 0.0254 | -0.0918 |
| Metabolizable energy | -0.0330 | -0.5018 | -0.0892 | -0.0101 | -0.4913 | 0.2474 |
| DTDN | 0.2731 | -0.0013 | -0.2116 | 0.2717 | 0.0561 | -0.1476 |
| NDF | 0.1348 | 0.2767 | 0.3130 | 0.0968 | 0.3434 | -0.4451 |
| Forage NDF | 0.0406 | 0.4092 | 0.2564 | -0.0078 | 0.4159 | -0.3964 |
| Non-fiber carbohydrate | 0.1728 | -0.0389 | -0.4730 | 0.1957 | -0.0614 | -0.0431 |
| Fat | 0.1354 | -0.2645 | 0.0545 | 0.1732 | -0.1155 | -0.0578 |
| Ca | 0.0799 | 0.3984 | -0.0865 | 0.0564 | 0.3598 | 0.4436 |
| P | 0.2031 | 0.0827 | 0.0209 | 0.1538 | 0.0540 | 0.1342 |
| Variability explained, % | 57.8 | 15.3 | 8.3 | 55.7 | 14.8 | 8.3 |
| Total explained | | 81.5 | | | 78.7 | |

[1] ELC – early lactation cows (DIM < 100); MLLC - mid- and late-lactation cows (DIM > 100).
[2] Intake variables.

**Table 8. Estimates, standard errors and significance for the fixed effects of the final candidate models under the NRC (2001) program. Response variables were MY and MPY in early and mid-to-late lactation cows**

| Variable[1] | Estimate | SE | DF | t value | P |
|---|---|---|---|---|---|
| Early-lactation cows (DIM < 100) | | | | | |
| MY | | | | | |
| Intercept | 5.6505 | 2.64480 | 115 | 2.14 | 0.0348 |
| NEL | 0.4032 | 0.03372 | 67 | 11.96 | <0.0001 |
| NDF | -0.4612 | 0.16790 | 86 | -2.75 | 0.0073 |
| P | 0.0244 | 0.00707 | 93 | 3.46 | 0.0008 |
| BW | 0.02214 | 0.00434 | 93 | 5.09 | <0.0001 |
| MPY | | | | | |
| Intercept | 0.2782 | 0.04743 | 141 | 5.87 | <0.0001 |
| DTDN | 0.00005 | 0.00000 | 207 | 16.04 | <0.0001 |
| NDF | -0.0178 | 0.00471 | 103 | -3.77 | 0.0003 |
| MPRUP | 0.0001 | 0.00002 | 207 | 6.45 | <0.0001 |
| Mid- and late-lactation cows (DIM > 100) | | | | | |
| MY | | | | | |
| Intercept | 13.6245 | 2.00470 | 87 | 6.80 | <0.0001 |
| NEL | 0.4110 | 0.05044 | 60 | 8.15 | <0.0001 |
| MPY | | | | | |
| Intercept | 0.4366 | 0.05697 | 85 | 7.66 | <0.0001 |
| NEL | 0.0131 | 0.00142 | 58 | 9.21 | <0.0001 |

[1] MY and MPY, Milk yield, and Milk protein yield; DTDN, NDF, NEL, RDP, MPRUP, P, Discounted TDN intake, NDF intake, NEL intake, RDP intake, Metabolizable protein from ruminally undegraded protein intake, and P intake.