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Evaluation of corn gluten feed and soyhulls as fiber and concentrate sources in dairy diets

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The Ohio State University, 1991
EVALUATION OF CORN GLUTEN FEED AND SOYHULLS AS FIBER AND CONCENTRATE SOURCES IN DAIRY DIETS

DISSERTATION

Presented in Partial Fulfillment of the Requirements for
the Degree of Doctor of Philosophy in the Graduate School of The Ohio State University

by

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*****

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To my wife, Riffat, for her quiet understanding, voluntary support and patience.

To my son, Umar, for his gentle companionship and ingenuous love.
ACKNOWLEDGEMENTS

I wish to express my sincere gratitude and appreciation to the following people, without whose help this dissertation would be a myth:

My major advisor, Dr. J.L. Firkins, for his dedicated help, sincere concern and patient understanding even during stressful periods. I thank him for his much needed help during planning, conducting research, and especially during sampling because this help was extended in spite of personal problems. I will be forever in his debt.

Dr. D.L. Palmquist, my co-advisor, for his useful suggestions, keen interest and time conferred to me in spite of a busy schedule.

Dr. M.L. Eastridge, my committee member for his valuable guidance during research especially while conducting the second trials. His friendliness and emotional support boosted my morale.

Dr. S.C. Loerch, member of my committee, for his valuable suggestions and educated advice.

Dr. D.L. Zartman, Chairman of the Dairy Science Department, for his efforts regarding the financial aid and ever present encouragement that makes one search for the silver lining even in the gloomiest cloud.

Julia Naderer, laboratory technician, for her valuable assistance with sample collection and laboratory analyses.

Debbie Gallagher, secretary, for typing my thesis and for deciphering my otherwise illegible chirography and making sure that I took care of the little details which I may have overlooked otherwise.

My family for their love, support and encouragement.

I am indebted, too, to the United States Agency for International Development (USAID) for giving me the opportunity to prove myself and also for financing my studies and research.
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# TABLE OF CONTENTS

DEDICATION .......................................................................................................... ii  
ACKNOWLEDGEMENTS ..................................................................................... iii  
VITA ................................................................................................................. iv  
LIST OF TABLES .................................................................................................... vii  
LIST OF FIGURES .................................................................................................. viii  
A KEY TO ABBREVIATIONS ............................................................................. ix  

## CHAPTER PAGE

| I. INTRODUCTION .......................................................................................1 |
| II. LITERATURE REVIEW ...........................................................................4 |
| - Effect of NSC on digestion .................................................................4 |
| - Effect of pH on fiber digestion ............................................................ 5 |
| - Effect of NSC on fiber digestion ........................................................ 8 |
| - Effect of forage:concentrate ratio on ruminal rate of passage .......... 9 |
| - Effect of NSC on bacterial protein synthesis .................................. 10 |
| - Associative effects ............................................................................14 |
| - Effect of NSC on performance ...........................................................15 |
| - Use of SH and CGF as energy sources ........................................... 18 |
| - Comparison of SH and CGF as energy sources ............................ 21 |
| - SH and CGF as roughage sources .................................................. 22 |
| REFERENCES ............................................................................................24 |
| III. EFFECT OF REPLACING NEUTRAL DETERGENT FIBER OF FORAGE WITH SOYHULLS AND CORN GLUTEN FEED FOR DAIRY HEIFERS .......................................................................................35 |
| - INTRODUCTION .......................................................................................35 |
| - MATERIALS AND METHODS ...............................................................36 |
| - RESULTS AND DISCUSSION ...............................................................40 |
| REFERENCES ............................................................................................56 |
IV. EFFECT OF REDUCING FORAGE NDF AND NON-STRUCTURAL CARBOHYDRATES ON NUTRIENT DIGESTIBILITY AND MILK PRODUCTION BY DAIRY COWS ..........................................................61

INTRODUCTION .........................................................................................................61

MATERIALS AND METHODS ...............................................................................62

RESULTS AND DISCUSSION .............................................................................66

REFERENCES .........................................................................................................79

V. GENERAL DISCUSSION ...................................................................................86

BIBLIOGRAPHY .....................................................................................................90
LIST OF TABLES

<table>
<thead>
<tr>
<th>TABLE</th>
<th>PAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Composition of diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF</td>
<td>37</td>
</tr>
<tr>
<td>2. Ruminal characteristics in heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF</td>
<td>41</td>
</tr>
<tr>
<td>3. Organic matter (OM) digestion by heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF</td>
<td>46</td>
</tr>
<tr>
<td>4. Digestion of fiber by heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF</td>
<td>47</td>
</tr>
<tr>
<td>5. Nitrogen (N) digestion and efficiency of bacterial growth in dairy heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF</td>
<td>49</td>
</tr>
<tr>
<td>6. Ingredient composition of diets varying in concentrations of NDF and non-structural carbohydrates (NSC) from forage NDF</td>
<td>63</td>
</tr>
<tr>
<td>7. Chemical composition of experimental diets varying in concentrations of NDF and non-structural carbohydrates (NSC) from forage NDF</td>
<td>67</td>
</tr>
<tr>
<td>8. Ruminal characteristics of cows fed diets varying in proportion of NDF from forage and non-structural carbohydrates (NSC)</td>
<td>70</td>
</tr>
<tr>
<td>9. Nutrient digestibilities by cows fed diets varying in concentrations of NDF from forage and non-structural carbohydrates (NSC)</td>
<td>73</td>
</tr>
<tr>
<td>10. Dry matter intake and milk production by cows fed diets varying in proportion of NDF from forage and non-structural carbohydrates (NSC)</td>
<td>77</td>
</tr>
</tbody>
</table>
## LIST OF FIGURES

<table>
<thead>
<tr>
<th>FIGURE</th>
<th>PAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. In situ disappearance (+ SE) of alfalfa hay NDF in situ by heifers fed diets with 85, 45, 65, 45 and 65% NDF from forage for control (C85), corn gluten feed (CGF) or soyhulls (SH) diets</td>
<td>50</td>
</tr>
<tr>
<td>2. In situ disappearance (+ SE) of NDF of corn gluten feed (CGF) and soyhulls (SH) in situ by heifers fed diets containing CGF or SH in which 45 or 65% of total dietary NDF was from forage</td>
<td>51</td>
</tr>
<tr>
<td>3. Ruminal pH as affected by diets varying in proportion of NDF from forage NDF and non-structural carbohydrates (NSC)</td>
<td>69</td>
</tr>
<tr>
<td>4. Ruminal ammonia-N concentrations as affected by diets varying in proportion of NDF from forage and non-structural carbohydrates (NSC)</td>
<td>72</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Definition</td>
</tr>
<tr>
<td>--------------</td>
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</tr>
<tr>
<td>ADF</td>
<td>Acid detergent fiber</td>
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<td>ADIN</td>
<td>Acid detergent insoluble nitrogen</td>
</tr>
<tr>
<td>ADL</td>
<td>Acid detergent lignin</td>
</tr>
<tr>
<td>ANOVA</td>
<td>Analysis of variance</td>
</tr>
<tr>
<td>AOAC</td>
<td>Association of Official Analytical Chemists</td>
</tr>
<tr>
<td>BW</td>
<td>Body weight</td>
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<tr>
<td>CF</td>
<td>Crude fiber</td>
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<td>CGF</td>
<td>Corn gluten feed</td>
</tr>
<tr>
<td>CP</td>
<td>Crude protein</td>
</tr>
<tr>
<td>Co-EDTA</td>
<td>Cobalt-Ethylenediaminetetraacetate</td>
</tr>
<tr>
<td>Cr-EDTA</td>
<td>Chromium-Ethylenediaminetetraacetate</td>
</tr>
<tr>
<td>d</td>
<td>Day</td>
</tr>
<tr>
<td>DIP</td>
<td>Degradable intake protein</td>
</tr>
<tr>
<td>DM</td>
<td>Dry matter</td>
</tr>
<tr>
<td>DMD</td>
<td>Dry matter digestibility</td>
</tr>
<tr>
<td>DMI</td>
<td>Dry matter intake</td>
</tr>
<tr>
<td>FCM</td>
<td>Fat corrected milk</td>
</tr>
<tr>
<td>FTR</td>
<td>Fractional turnover rate</td>
</tr>
<tr>
<td>g</td>
<td>Gram</td>
</tr>
<tr>
<td>GLC</td>
<td>Gas-liquid chromatography</td>
</tr>
<tr>
<td>ISDMD</td>
<td>in situ dry matter digestibility</td>
</tr>
<tr>
<td>IVDMD</td>
<td>in vitro dry matter digestibility</td>
</tr>
<tr>
<td>Kcal</td>
<td>Kilocalorie</td>
</tr>
<tr>
<td>Kg</td>
<td>Kilogram</td>
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<tr>
<td>L</td>
<td>Liter</td>
</tr>
<tr>
<td>Mcal</td>
<td>Megacalorie</td>
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<tr>
<td>mg</td>
<td>Milligram</td>
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<tr>
<td>mM</td>
<td>Millimolar</td>
</tr>
<tr>
<td>NDIN</td>
<td>Neutral detergent insoluble nitrogen</td>
</tr>
<tr>
<td>NEI</td>
<td>Net energy for lactation</td>
</tr>
<tr>
<td>NSC</td>
<td>Non-structural carbohydrates</td>
</tr>
<tr>
<td>NRC</td>
<td>National Research Council</td>
</tr>
<tr>
<td>OM</td>
<td>Organic matter</td>
</tr>
<tr>
<td>SAS</td>
<td>Statistical Analysis System</td>
</tr>
<tr>
<td>SBM</td>
<td>Soybean meal</td>
</tr>
<tr>
<td>SCM</td>
<td>Solid corrected milk</td>
</tr>
<tr>
<td>SH</td>
<td>Soyhulls</td>
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<tr>
<td>SS</td>
<td>Sorghum silage</td>
</tr>
<tr>
<td>TNC</td>
<td>Total non-structural carbohydrates</td>
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<td>VFA</td>
<td>Volatile fatty acids</td>
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</tbody>
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Dairy cows require high energy diets to support high milk production. High forage diets are not only deficient in energy but also are limited by the physical capacity of the cow's digestive tract. This results in a shortage of energy, rendering high producing dairy cows unable to attain their maximum genetic potential for milk production (Clark and Davis, 1980). Therefore, formulation of high-energy diets comprising a sufficient proportion of non-structural carbohydrates (NSC) is needed to obtain maximum milk yield from dairy cows. These NSC are comprised of starch and other soluble carbohydrates from cereal grains and plant cells. However, most NSC are digested rapidly in the rumen. When NSC percentage is increased beyond a threshold level, it can result in excessive acid production by the microbial population causing reductions in ruminal fluid pH (El-Shazly et al., 1961), fiber digestion (Bines, 1976; Henning et al., 1980; Miller and Muntiferring, 1985; Wangsness and Muller, 1981), percentage milk fat (Sutton, 1989; Sutton and Morant, 1989), and perhaps dry matter intake (DMI) by cows (Joanning et al., 1981).

Adequate amounts of fiber are needed to promote rumination and salivation, which in turn help buffer ruminal pH and maintain normal rumen function. Due to the diversity of forage types and quality, balancing diets for NDF or ADF has been proposed (NRC, 1989). However, the NRC recommendations are based on limited data. The maximum and minimum amount of forage NDF (excluding concentrate NDF) in dairy diets has been suggested to be 26.7 and 17.8%, respectively (Mertens, 1988). The upper
limit of dietary forage should be used only for cows with lower milk production or when forage is abundant and cheap relative to concentrate. High producing dairy cows in early lactation can hardly maintain their milk production and body condition if fed such a high forage diet. On the other hand, cows fed minimum amounts of forage NDF may not only produce milk with low fat test, but associated metabolic disorders also can occur. These metabolic disorders include displaced abomasum, laminitis, and ruminal acidosis. For these reasons, the NRC (1989) recommends several dietary NDF percentages for cows at different stages of lactation or with varying genetic potential.

Balancing diets for NDF must be viewed with caution. For instance, the size of the fiber particles and fiber source also are critical for stimulating chewing activity. Many cereal by-products are high in NDF percentages, but their particle size is small so that rumination is not stimulated. For this reason, a minimum of 75% of total dietary NDF from forages has been suggested for lactating dairy cows (NRC, 1989). Typical diets containing forage, corn and soybean meal have 80-85% of total NDF as forage NDF. Thus, if a minimum of 75% of NDF as forage NDF is used, it must be assumed that by-product NDF is mostly ineffective as a fiber source. This NRC recommendation has been made without adequate by-product NDF information. Therefore, the effectiveness of fiber of by-product feeds and their greatest effective substitution for forage in dairy rations need to be determined.

Starches and sugars (NSC) are degraded more rapidly and completely than structural carbohydrates (NDF), thus providing more energy to ruminal bacteria to increase utilization of degraded N and increase microbial growth (Offer et al., 1978). Therefore, formulating dairy diets on the basis of NSC in conjunction with NDF may be preferable to formulating them for NSC or NDF individually. The manipulation of dietary NSC and NDF by using fibrous by-products should be practical. At a fixed forage content, flexibility to manipulate dietary NSC without using by-products is limited (Nocek and Rus-
In this dissertation, two by-product feeds, soyhulls (SH) and corn gluten feed (CGF) were chosen to replace a portion of forage fiber or concentrate in dairy rations. Corn gluten feed has 40-45% NDF that is high in hemicellulose. Soyhulls contain 61-67% NDF that is high in cellulose. Both by-products are competitive sources of energy for dairy cows in the midwest. The hypothesis was that SH or CGF can replace a portion of forage fiber and concentrate in dairy diets without affecting rumen function and lactation performance in cows. Data evaluating the effect of replacing forage or concentrates with CGF or SH on nutrient digestibility, ruminal characteristics and lactation performance are limited. The objective was to determine the effectiveness of SH or CGF to replace a portion of forage NDF and concentrate in dairy diets. Determining the most appropriate level of by-product substitution of forage or concentrate in dairy diets based on the digestibilities of nutrients and lactation performance by animals may help to increase efficiency of dairying.
CHAPTER II

LITERATURE REVIEW

Effect of NSC on digestion

It has been demonstrated that supplementation of roughages with a rapidly fermentable source of energy, such as barley, ground corn or molasses, depressed rumen cellulolysis and dry matter digestion (DMD; Burroughs et al., 1949; Chimwano et al., 1976; Kartchner, 1980). Woodford et al. (1986) conducted a trial using 4 lactating cows in a 4 x 4 Latin square design. They formulated diets containing alfalfa hay at 28, 36, 45 and 53% of total DM. Apparent digestibility of dietary constituents was not different except that NDF and ADF digestibilities increased (P < .05) as percentage forage in the diet increased (for NDF, 45.6 to 50.8%; for ADF, 46.9 to 50.7%). Observed decreases in digestibility have been associated with increasing concentrate in the diet. Mould et al. (1983) observed no difference in 24-h degradation (45%) of washed hay in situ with up to 50% supplementation with barley, but it decreased to 36% when 75% barley was supplemented. Poore et al. (1990) used six ruminally cannulated steers to determine the influence of various dietary concentrations of NSC on fiber digestion. Diets had 30, 60, or 90% of a concentrate based on flaked sorghum grain plus a 50:50 mixture of wheat straw and alfalfa hay. They reported no change in total tract digestibility of NDF, but digestibility of potentially digestible NDF in the rumen decreased (P < .05) from 92 to 70 to 48% as concentrate increased from 30 to 60 to 90% of diet DM. Digestibilities of NDF decreased (P < .05) by 72, 57 and 34% for straw, hay and grain, respectively, when concentrate was increased from 30 to 90%. This indicated that fiber digestibility was
depressed more for forage NDF than for grain NDF. Chappell and Fontenot (1968) re­
ported reduced fiber digestion when purified starch was added to forage diets fed to
sheep. It is evident that NDF digestibility decreases significantly when excessive
amounts of concentrates are added to the diet. Several factors such as concentration of
dietary concentrates, fiber source, rate of passage, DMI and ruminal pH probably are
responsible for reduced fiber digestibility associated with increased concentrates in the
diet. These will be addressed in more detail below.

Effect of pH on fiber digestion

One of the most critical factors responsible for reduced fiber digestion is pH. Ruminal microbes are very sensitive to changes in pH, and most prefer a pH between 6.2
and 6.8. This range has been demonstrated in vitro (Hiltner and Dehority, 1983; Shriver
et al., 1986; Stewart, 1977; Terry et al., 1969) and in vivo (Mould and Orskov, 1983;
Mould et al., 1983). Mould and Orskov (1983) reported a severe reduction in cellulo­
sis when pH was dropped below 6.1. Erdman (1988) reported that ADF digestion de­
creased 3.6 percentage units for every 0.1 pH unit decrease below 6.3 in the rumen.
McCullough (1969) noted that pH in rumen contents decreased with increasing amounts
of rapidly fermentable carbohydrates in the diets of cattle. When rumen contents from
these animals were incubated in vitro, the fluid with lower pH reduced cellulolysis.
Terry et al. (1969) also reported that the extent of cellulose digestion by mixed rumen
microbes was dependent on the pH of incubation media. One of the most detailed stud­
ies of the interrelationship between rumen pH and fiber digestion was done by Orskov
and Fraser (1975). They fed a common barley and dried grass diet to all sheep but varied
the methods of barley preparation. When whole barley was fed at 50 g/kg metabolic
body weight per day, ruminal pH remained above 6.0, and intake of dried grass was 42.8
g/kg metabolic body weight per day. When barley was pelleted and fed at the same rate,
the ruminal pH declined to less than 5.5, and both fiber digestibility and intake of grass
were reduced.

The effect of low ruminal pH on fiber digestion could be due partially to changing kinetics of digestion. Mertens and Loften (1980) reported a linear increase in the lag time for fiber digestion in vitro as the proportion of starch increased. Similar effects have been found in vivo (Aitchison et al., 1986). Hiltner and Dehority (1983) suggested decreased lag time for cellulose digestion by *Fibrobacter succinogenes* when soluble carbohydrates were added to the medium. They related this effect to an increase in bacterial numbers. They also reported a reduction in both pH and rate of cellulose digestion after the depletion of soluble carbohydrates in the medium.

Miller and Muntifering (1985) conducted an experiment with five rumen-fistulated Holstein steers in a Latin square design and determined the effect of dietary concentrate (0, 20, 40, 60 or 80% cracked corn) on kinetic characteristics influencing forage fiber digestion in vivo. They reported that rates of forage fiber digestion ranged from .039 \( h^{-1} \) for 20% grain to .062 \( h^{-1} \) for 80% grain but did not differ (P > .05) among treatments. They suggested that concentrate addition to forage does not reduce fiber digestibility by slowing rate of digestion. They also stated that the potential extent of forage NDF digestion was lower (P > .05) for 80% grain (28.7%) compared with all other treatments (avg., 51.2%).

Low pH could affect attachment of microbes to fiber particles, which seems to be needed by many species of bacteria before most efficient digestion occurs (Hoover, 1986). Shriver et al. (1986), using continuous cultures, reported a marked reduction in attached microbes at pH 5.8 compared to pH 6.2, which corresponded to a significant decrease in NDF digestion. It was reported that bacterial proliferation was evident during low pH conditions, but none were tightly adherent to fiber particles as viewed from electron micrographs (Cheng et al., 1983). Increased concentration of H+ may displace divalent calcium or magnesium, which may be needed by some bacteria to attach to feed
Low pH can have adverse effects on bacterial growth rates (Russell et al., 1979). Stewart (1977) reported a fall in the number of filter paper-degrading bacteria from $10^6$ to $10^3$/ml when pH was reduced from 6.9 to 6.0, resulting in a reduction in the cellulolytic activity of rumen contents. Russell and Dombrowski (1980) reported reduced yields of cellulolytic bacteria in continuous culture when pH decreased below 6.0. Reduction in the number of cellulolytic bacteria from $10^6$ to $10^4$/ml also was noted in vivo due to reduced ruminal pH (Mould and Orskov, 1983; Mould et al., 1983). This could be due to the destruction of membrane potential of cellulolytic bacteria, causing lower viability at low pH (Russel et al., 1990).

The diurnal fluctuation in ruminal pH can have detrimental effects on the rumen microbes. Goetsch and Owens (1984) conducted a trial in which four intestinally cannulated dairy cows were fed either 65, 50 or 35% concentrate diets (65C, 50C and 35C) twice daily, or a fourth, alternate (ALT) diet in which the 65C diet was fed in the morning and the 35C diet fed in the evening. They reported that ruminal digestibility of organic matter tended to decrease with increasing roughage level, but cows fed the ALT diet had the lowest organic matter digestion. Ruminal starch digestion was slightly greater, but total tract starch digestion was lower, when the ALT diet than the 50C diet was fed. This difference may be due to fluctuation in ruminal pH because cows fed the 50C diet maintained a more stable ruminal pH. It also was reported by other workers (Mould et al., 1983) that diets supplemented with molasses produced the greatest diurnal variation in the ruminal pH, but the overall pH was higher than that found with sheep fed diets supplemented with barley. Those fed the diet supplemented with molasses had lower fiber digestion. This can be due to the reduced ability of the cellulolytic microbes to adjust to changing ruminal pH.
Effect of NSC on fiber digestion

If the lower pH is the only cause for the depressed fiber digestibility, then depression can be removed by maintaining ruminal pH above 6.2. Mould and Orskov (1983) conducted a research trial in which sheep were fed diets comprising only hay or hay plus higher amounts of barley. The ruminal pH was manipulated by adding mixtures of mineral acids and NaHCO₃. They stated that microflora associated with the all hay diet were readily inhibited when ruminal fluid pH levels were maintained below 6.1. Both fiber digestion and intake were depressed. When the ruminal pH of sheep fed barley was increased above 6.2 by adding NaHCO₃, DMI, starch degradation, or cellulolysis did not increase to values for sheep fed high forage diets, indicating that high amounts of barley starch in the rumen can have detrimental effects on ruminal cellulolytic activity aside from their effects on pH. In another trial (Mould and Orskov, 1983), the ruminal pH of sheep was maintained at 6.7 ± 0.15, but the reduction in fiber degradation was not removed completely. Therefore, the residual depression in fiber digestion was caused by factors other than low pH and was termed "the carbohydrate effect". They also reported that the proportion of barley necessary to depress ruminal pH and fiber degradation was dependent on the rate of solubility of readily fermentable carbohydrates to the rumen organisms and was, therefore, influenced by the degree of processing.

The depression in cellulolysis associated with the addition of a readily degradable supplement may be due to increased competition among the rumen microbes for essential nutrients such as ammonia (Burrough et al., 1949; El-Shazly et al., 1961). Cellulolytic microbes may be unable to reproduce at a rate fast enough to compete for these nutrients and maintain themselves in the rumen when high concentrations of NSC are fed. Reduced cellulolytic numbers may decrease cellulolysis. Furthermore, mono- and disaccharides may inhibit some enzymes in the cellulase system (Mackie and White, 1990).
Effect of forage:concentrate ratio on ruminal rate of passage

The extent of degradation of a diet is related to the duration of ruminal exposure. When solid retention time was reduced by an increase in feed intake (Weston and Hogan, 1971) or by altering forage:concentrate ratio (Bines and Davey, 1970), a concurrent decrease in dry matter digestion was noted. Colucci et al. (1982) reported that ruminal passage rate of both concentrate and forage particles were reduced (28 and 44%, respectively) when concentrate levels for non-lactating dairy cows were increased from 17 to 68% of the diets fed at low intake (maintenance). However, at high intakes, cows fed diets with low (32%) and high (83%) forage had similar particle passage rates for both forage and concentrates. Colucci et al. (1990) reported that dilution rate in the rumen decreased (P < .05) linearly as the proportion of concentrates in dairy diets increased at low intakes, but no differences were detected at high intakes. Cecava et al. (1990) reported no difference in ruminal dilution rates in steers when fed high (40%) or low (24%) NDF diets at 2% body weight. The average dilution rates were 7.35 and 4.15%/h for fluid and particulate matter, respectively.

Poore et al. (1990) conducted an experiment in which six ruminally cannulated steers were used to examine the influence of various dietary forage:concentrate ratios on ruminal rate of passage. Diets had 30, 60 or 90% flaked sorghum grain plus a 50:50 mixture of wheat straw and alfalfa hay. Ruminal passage rate for straw (3.4 and 3.0%/h) and hay (4.6 and 4.7%/h) remained unaltered when concentrate increased from 30 to 60%, respectively, but passage rates for straw and hay decreased by 38 (2.2%/h) and 13% (4.1%/h) respectively, when concentrate was increased to 90%. However, passage rates for grain (5.3, 5.1 and 4.4%/h) and fluid (9.3, 10.0 and 8.2%/h) were not affected by concentrate percentage. They attributed these differential effects of dietary concentrates to differences in ruminal stratifications. As compared to sorghum grain, ruminal passage rate of alfalfa hay was relatively unaffected by dietary concentrate levels, indi-
cating that increased concentrate levels may have a greater impact on passage of low quality forage than on passage of either grain or high quality forage.

Hartnell and Satter (1979) reported no change in particulate rate of passage with variation in forage content of diets. However, liquid rate of passage was lower in cows fed diets containing 45% forage (4.7%/h) than when dietary forage concentration was raised to 67% (9.3%/h). This increased fluid dilution rate at high forage content was due to greater rumination resulting in more salivation (Luginbuhl et al., 1989).

In summary, both fluid and particle dilution rates were affected by forage:concentrate ratio and the effect was more pronounced at low (below 2% BW) rather than high intakes. Fluid dilution rate generally is depressed as concentrates are increased at the expense of forage in diets. Moreover, at low intakes particle dilution rate in the rumen often is decreased as concentrate percentage in the diet is increased. Passage of poor quality forage is decreased more than is good quality forage due to increasing concentrate in the diet.

Effect of NSC on bacterial protein synthesis

Microbial growth depends on the amount of energy obtained by the fermentation of feed organic matter. Microbes get more energy from degradation of carbohydrate than of CP or fat. The growth of microbes/unit of carbohydrate fermented increased when moderate levels of starch were added to high-fiber diets (Mathers and Miller, 1981; Chamberlain and Thomas, 1979; Offer et al., 1978; Stern et al., 1978; Hoover, 1986). Many of these researchers manipulated dietary NSC concentrations by changing the forage:concentrate ratio. They reported that the amount of concentrate in excess of 30% (DM basis) of the diet was responsible for the progressive reduction in microbial yield. The reduced microbial yield under these conditions may be due to energetic uncoupling of fermentation or reduced ruminal turnover rate (Sniffen and Robinson, 1987). Ammonia is a main source of nitrogen for bacterial growth, although peptides and amino acids
also are important. Insufficiency of these compounds at certain times after feeding may be a major factor causing energetic uncoupling, resulting in continued production of fermentation products without concomitant bacterial growth (Hespell and Bryant, 1979).

Interrelationship between protein and energy-yielding nutrients can have great effects on bacterial protein synthesis in the rumen. Stokes et al. (1991) formulated diets with three levels of NSC (54, 37 and 25% of DM), with degradable intake protein (DIP) ranging from 19 to 4% of DM. They reported that, regardless of DIP level, bacterial efficiency was lower in continuous cultures receiving diets with the lowest concentration of NSC than in those receiving 54% NSC diets. Increasing NSC from 37 to 54% resulted in a small increase (21.1 to 22.6 g of bacterial N/kg of DM digested) in bacterial efficiency, suggesting that available energy was optimized in this range. Hoover (1987) summarized 57 continuous culture fermentations of diets combining high levels (30 to 50% of DM) of NSC. These diets varied from 5.1 to 16.9% DIP. The NSC:DIP ratio for these diets ranged from 2.4 to 10.0. He reported a significant curvilinear decrease in microbial efficiency with the widening of the NSC to DIP ratio, explaining that DIP limited microbial growth in high-energy diets, particularly with a ratio above 4. It is important to note that these continuous culture studies maintained constant pH, and feeding high NSC levels (e.g. > 40%) may decrease efficiency in vivo due to low pH (discussed earlier). In another study, Stokes et al. (1991) reported that the flow of microbial N to the duodenum was the lowest in cows fed diets containing 24% NSC and 9% DIP when compared to those fed diets containing 31% NSC and 11.8% DIP or 38% NSC and 13.2% DIP. This shows that increasing NSC and DIP above 24 and 9% of DM, respectively, increases microbial protein synthesis.

Stern et al. (1986) conducted an experiment to examine the effects of total non-structural carbohydrates (TNC) and protein source on ruminal bacterial protein synthesis in continuous culture. Two levels of TNC (39.5 and 16.4%) and two levels of DIP (15.1
and 11.5%) were tested by using corn vs. beet pulp and untreated soybeans vs. alcohol-treated soybeans, respectively. They pointed out that NH\textsubscript{3}-N concentration was lower (8.5 mg/dl) for cultures receiving low TNC diets, compared to those receiving high TNC diets (17.6 mg/dl). This may be due to lower protein degradation or greater bacterial uptake of NH\textsubscript{3}-N in fermenters receiving low TNC diets. Efficiency of bacterial protein synthesis was greater for low than high TNC treatments. This unusual response in bacterial protein synthesis may be due to different energy sources (corn vs beet pulp). These findings indicate that beet pulp has lower % DIP but similar degradable energy concentration compared to corn, despite its lower TNC and higher cell wall concentration.

Stern et al. (1978) reported that dietary energy is not the only factor affecting microbial growth. They reported increased microbial yield from 15.0 to 19.5 g microbial CP/100 g DMD in continuous culture when dietary TNC concentrations were increased from 19.6 to 49.0%, even though diets were isocaloric on a gross energy basis (4.6 Kcal/g) and VFA production and DMD did not differ markedly among diets. This effect shows that a major factor affecting the utilization of degraded dietary N is the type and rate of availability of carbohydrates. McAllan and Smith (1976) formulated five diets consisting of different energy sources varying in starch concentration. They reported that ruminal microorganisms converted the highest amount of energy from the fermentation of starch into microbial protein, indicating that degradation rate of protein coincided more closely with that of starch than of cellulose and hemicellulose.

Offer et al. (1978) conducted an experiment containing soybean meal and dried grass either unsupplemented or supplemented with wheat starch, paper or equal mixtures of both. They reported that sheep fed diets containing a mixture of starch and paper had greater efficiencies of ruminal microbial protein synthesis than did those fed supplements of either starch or paper alone. When a combination of rapidly and slowly degradable carbohydrates is included in the ruminant diet, it results in the availability of a constant
source of energy for microbial growth in the rumen. Thus, maximum microbial yield is obtained due to reduced losses of NH$_3$-N in the rumen (Oldham et al., 1977; Johnson, 1976). Moreover, Offer et al. (1978) and Chamberlain and Thomas (1978) reported increased microbial N flow to the duodenum when diets containing the mixtures of readily and slowly fermentable carbohydrate vs. single sources were fed.

The source of NSC in the diet can have a prominent effect on microbial growth in the rumen. Oldham (1984) reported an increased microbial yield in cows fed diets containing barley compared to cows fed diets containing corn grain, perhaps due to greater rate and extent of degradation of barley starch. Prigge et al. (1978) used four ruminally and abomasally cannulated steers which were fed corn processed by one of four methods: 1) dry rolling, 2) steam flaking (SF), 3) propionic acid-treated, and 4) ensiled high moisture shelled corn (EHM). They reported increased microbial nitrogen flow to the duodenum of the steers fed EHM diets compared to steers fed other diets. Windschitl and Schingoethe (1984) used two rumen-fistulated Holstein cows in an experiment with a switchback design to evaluate effects of consuming dried whole whey (38% of total ration DM) on synthesis of microbial protein in the rumen. Cows were fed total mixed rations of 45% corn silage, 10% alfalfa hay, and 45% concentrate mix on a DM basis. The concentrate mix was corn and soybean meal (control) or 85% dried whole whey. Although concentrations of microbial N in duodenal digesta were constant (72-74%) for both diets, more microbial N arrived at the lower gut in cows fed whey diets. The increased dilution rates of ruminal fluid in cows fed dried whey appeared to have promoted more efficient growth of microbes due to decreasing maintenance energy as a percentage of total energy. Spicer et al. (1986), however, reported no change in microbial efficiency due to feeding diets containing 80% sorghum, corn or barley to steers.
Associative effects

Negative associative effects occur when digestibility of a feed mixture is less than that of the sum of the individual components of the diet. Byers et al. (1975) fed diets containing 100, 75, 25, or 0% corn silage plus whole shelled corn. Digestibilities of energy and DM of the mixed diets fed to steers were significantly lower (P < .01) than values predicted from all silage or all grain diets, indicating significant negative associative effects. Similarly, Bines and Davey (1970) conducted an experiment in which four diets containing 0, 20, 40 or 60% chopped straw and the remainder concentrates were fed to non-lactating dairy cows in a 4 x 4 Latin square design. All diets were isonitrogenous. The diets containing 0, 20, 40, or 60% straw had 8.05, 16.22, 25.24 or 32.47% cellulose, respectively. They reported that digestibilities of dry matter of the four diets were 81, 69, 59 and 55%, respectively. Cellulose degradation in the rumen was depressed (42.9%) when cows were fed the diet containing 20% roughage (20R) compared with those (47.1 and 49.9%) for cows fed 40R and 60R diets. Rate of breakdown of cotton threads in situ increased as the roughage content of the diet increased. Liebenberg (1979) carried out an experiment involving 36 lactating cows to determine the effect of concentrate:forage ratio on digestibility. He reported that crude fiber digestibility decreased from 54.1 to 44.6% when concentrate increased from 35 to 65%.

The level of feed intake by animals can interact with the magnitude of the associative effect. Williams et al. (1986) conducted an experiment in which steers were fed high and low starch diets at maintenance. They did not observe any major effect on the digestibility of straw in diets containing rapidly fermentable carbohydrates. Negative associative effects on digestibility were small at low level of intake (< 1.5 x maintenance) but became significant when energy intake increased to 2.5 - 3 x maintenance. Negative associative effects may be a major problem in lactating dairy cows, which normally consume above 3 x maintenance (Tyrrell and Moe, 1975).
In a study discussed earlier (Mould et al., 1983), overall average DMD was reduced by 12.2 percentage units when barley was fed to sheep at a rate of 60 g/kg. Digestibility of the hay fraction was reduced to 32.8% compared to the expected value of 52.2%. This was equivalent to a 37.2% decrease in hay digestibility. Because sheep consumed about 40% more hay, this 12.2 percentage unit decrease in digestibility increases the amount of digestible DM voided in the feces by 17 percentage units [12.2 plus (40% x 12.2%)].

Effect of NSC on performance

Ronning and Laben (1966) reported that cows fed diets with a forage:concentrate ratio of 60:40 produced greater amounts of milk than did those fed diets with forage:concentrate ratios of 90:10, 30:70 and 0:100. Cows fed the 90:10 diet could not consume enough energy to maintain milk yield, and animals receiving the 30:70 and 0:100 diets produced milk with reduced milk fat percentage when compared to other diets. Broster et al. (1977) conducted a trial in which three diets containing 60:40, 75:25 and 90:10 ratios of concentrate to hay were fed to lactating dairy cows. Milk production was 27.0, 29.0, and 34.5 kg/d but milk fat was 4.0, 3.1 and 2.7%, respectively. They recommended the 75:25 diet as an optimal diet, explaining that very high concentration of concentrates may be only temporarily acceptable during very early lactation. They also stated that the simultaneous decrease in milk fat percentage overcomes the increase in milk yield from feeding the 75:25 diet. Economic criteria can determine an upper limit to the acceptable proportion of concentrates in the diet, and it is important to re-evaluate recommendations when changes in milk pricing systems occur. When diets containing 28, 36, 45 or 53% long alfalfa hay as sole forage were fed to lactating cows, milk fat percentage increased linearly with increasing forage consumption (Woodford et al., 1986). Increasing dietary fiber increased chewing time by cows, resulting in greater acetate:propionate ratio and milk fat percentage. The point of milk fat depression oc-
curred when forage made up ≤ 45 % of DMI. Macleod et al. (1983) conducted a study in which diets with forage:concentrate ratios of 80:20, 65:35, 50:50, or 35:65 were fed to lactating cows. They reported a curvilinear increase in milk yield and a curvilinear reduction in milk fat percentage by cows fed diets with decreasing forage:concentrate ratios.

The reduced milk fat percentage by cows fed diets containing excessive amounts of NSC probably is due to the change in rumen fermentation pattern. It has been reported that, as the lipogenic (acetate and butyrate) to non-lipogenic (propionate) ratio of molar % VFA declines, milk fat percentage decreases (Sutton, 1985). The difference in forage:concentrate ratio of diets needed to maintain normal milk fat test between American work (45:55; Woodford et al., 1985) and British work (25:75; Broster et al., 1977) can be explained on the basis of forage sources. In the U.S., forages low in NDF (e.g., alfalfa and corn silage) generally are used to formulate rations for dairy cows, whereas in Britain, grass hays with higher NDF are used. Forages higher in NDF normally are lower in cell solubles and NSC, which are compensated by higher amounts of dietary concentrates.

In order to feed these higher percentages of concentrate without reducing milk fat synthesis, DM intake, and fiber digestion, considerable research has been done in Europe comparing concentrate sources with varying percentages of fiber. Fiber in concentrate feedstuffs either provides some effective fiber or dilutes NSC.

Mayne and Gordon (1984) conducted a trial using 24 British-Friesian dairy cows in early lactation in a changeover design with three periods, each of 4 wk duration. They reported that the high proportion of barley or sugarbeet pulp feeding (10 vs. 7 kg/d) significantly increased (P < .01) milk yield. All concentrates were offered in addition to the cows having ad libitum access to grass silage containing 21.3% DM and 66.8% digestible organic matter. However, energy source in the concentrates (barley vs. sugar beetpulp) did not show a significant effect on milk yield. Garnsworthy (1989) formulat-
ed diets with high starch (45.2%) and low ADF (6.9%; diet HSLF) and low starch (24.4%) with high ADF (11.75%; diet LSHF). Milk yield was not affected by treatments, but the fat percentage of milk from cows given LSHF was greater (4.26%) than that from those fed HSLF (3.71%), resulting in significant differences in fat-corrected milk (FCM) yield. Thomas et al. (1986) conducted a trial using two diets: one based on barley and second based on sugar beet pulp plus fat (SBP/FF). Both diets were formulated to be isocaloric and isonitrogenous. Increasing the amount of barley supplement in the diet caused a marked reduction in the digestibility of ADF by lactating dairy cows (68.2 vs. 54.4%). Cows fed diets containing SBP/FF produced about 1.9 kg more milk (P < .05) than did those fed diets with barley. The greater amounts of highly digestible fiber and lower NSC of the beet pulp might have helped maintain the fiber digestibility (Bhattacharya and Sleiman, 1971).

Visser et al. (1990) conducted a feeding trial in which the type of carbohydrate in concentrate mixtures (starch vs. cell wall constituents) and difference in rate of rumen degradation of carbohydrate (fast vs. slow) were studied. The basal diet, which comprised 75% of the total DM intake, consisted of wilted grass silage, corn silage and concentrates. The remaining part of the diet consisted of barley (B), corn (C), pressed ensiled beet pulp (P) or moist ensiled corn bran (CB). Diets had 20.4, 23.1, 7.8 and 14.3% starch and 35.1, 32.9, 41.2 and 43.6% NDF, for B, C, P and CB diets, respectively. They reported that cows fed these diets produced the same amount of milk, but those fed P had a greater milk fat percentage. Milk protein percentage was higher for cows fed B and C, which were relatively high in starch, than for those fed P and CB diets. The reduced milk protein percentage may be due to lower (P < .05) energy intake by these cows. Moreover, they reported higher concentrations of ruminal ammonia and isobutyrate from cows fed CB and stated that this may be due to reduced microbial protein synthesis. Phipps et al. (1987) fed fibrous (F) or starchy (S) concentrates to 99 lactating cows. They
reported that fiber digestion was significantly greater by cows fed rations containing F than S (56.5% vs. 67.3%). They also reported that concentrate type did not affect milk yield. Feeding S decreased fat concentration and fat yield (P < .05) but increased milk protein concentration (P < .001) compared with feeding F. The greater milk protein content of the high starch diet probably was due to the reduced acetate:propionate ratio in the rumen (Manson and Leaver, 1989). The increased production of propionate at the expense of acetic acid probably has a sparing effect on gluconeogenic amino acids, resulting in increased availability of amino acids for production of milk protein.

**Use of SH and CGF as energy sources**

Edionwe and Owen (1979) reported that DM, NDF, ADF and CP digestibilities were lower for lactating dairy cows fed a control diet (50% concentrate) than for those fed SH replacing 43.8% of the concentrate. Interestingly, at maintenance, the digestibility of the SH diet was 94% that of the control diet, but at 4 x maintenance, it was 104% of that of control. Nakamura and Owen (1989) conducted a trial in which lactating cows were used to evaluate SH as a replacement for corn grain in lactation diets. Soyhulls constituted 0 (corn), 50 (corn-SH), and 95% (SH) of the concentrate mixture, which was prepared as a 4.8-mm pellet. These concentrate mixtures were mixed with alfalfa silage (50:50 DM basis) and fed ad libitum as complete diets. They reported that cows fed SH diets resulted in less milk (27.3 kg/d) but with greater milk fat (3.49%) compared with cows fed corn diets (29.8 kg/d; 3.13% milk fat), resulting in similar 3.5% FCM yield and feed efficiency. Based on these two studies, it appears that SH have an energy value similar to corn in pelleted concentrate diets fed to lactating dairy cows at high intakes. Macgregor et al. (1976) also substituted SH at 0, 26.9 and 48.0% for corn into dairy diets to give 28, 38.7, and 39.5% NDF in complete feeds. Digestibilities of NDF were 47.5, 62.6, and 61.6% for 0, 26.9 and 48.0% SH diets, respectively. Likewise ADF and cellulose digestion increased as SH was added. No difference in milk production was noted.
Conrad and Hibbs (1968) reported greater digestibilities (77-79%) of N by lactating cows fed rations containing soybean flakes (SH) than those containing oats. They also reported a greater N efficiency in cows fed diets containing soybean flakes.

Anderson et al. (1988) compared SH to corn as energy supplements (0, 25 or 50% of the diet) for growing beef calves maintained on either brome grass or corn residue pastures. Addition of SH to the diet improved the whole tract digestibility of DM, ADF and cellulose when compared with calves fed the diet supplemented with corn. Percentage NDF digestion decreased by 10 units when corn was supplemented. At the 50% level, steers consuming the corn diet showed a rapid drop in ruminal pH to below 5.65, which was followed by a rapid return to its pre-feeding value. Steers fed the SH diet showed a more gradual decline in ruminal pH to 6.0 by 12 h post-feeding. Rate of solka floc digestion in situ decreased by about 50%, but it appears that the digestion of SH NDF continued. Similar results were obtained by Hubbard et al. (1986), Martin and Hubbard (1987) and Merrill and Klopfenstein (1985).

Fellner and Belyea (1991) conducted an experiment in which corn silage-based diets containing 20, 40 or 60% (DM basis) CGF were fed to lactating cows. They reported no change in milk yield, cows fed diets with 20% CGF produced milk with the highest (3.6%) milk fat test compared with those fed 40 (3.5%) and 60% (3.1%) CGF diets. They noted that milk protein percentage was greater for cows fed 40 and 60% CGF diets. Feeding increased CGF decreased DM digestibility and reduced ruminal acetate concentration but did not affect N, NDF, ADF or starch digestibilities. Feeding CGF at 16.5 and 33% of DM in diets fed to lactating cows had no effect on milk fat percentage, increased DMI and 4% FCM yield, but decreased percentage milk protein (Ohajuruka and Palmquist, 1989). Based on the lower gross efficiency of milk production, acetate:propionate ratio, rumen pH and reduced milk protein percentages, the authors suggested that CGF should be limited to 30% of concentrate mixtures or 15-20% of diet DM when used to
replace corn in diets for lactating dairy cows. Macleod et al. (1985) conducted a trial using lactating cows to examine the feeding value of CGF. They reported that feeding dried CGF resulted in greater (P < .05) DMI, solids corrected milk and protein percentages in milk when compared to cows fed the control diet.

Firkins et al. (1985) fed a control diet of cracked corn and alfalfa-orchard grass hay or CGF at 35 or 70% of DM to rams. They reported a linear decrease in ruminal pH and acetate:propionate ratio when sheep were fed dry CGF. These authors also reported decreased apparent digestibility of ADF by lambs fed diets containing dry CGF than soybean meal or wet CGF. However, Highfill et al. (1987) reported increased apparent digestibility of NDF by cows fed diets containing CGF compared with a control diet containing com-soybean meal.

Fleck et al. (1987) conducted a trial in which 32 beef cows were individually fed native grass hay and various protein supplements. Cows were fed 0, .7, 1.0, or 1.6 kg/d, respectively, of supplement for negative control (NC), soybean meal (SBM), a blend of SBM and CGF (SBM/CGF), or CGF. They reported that, compared to supplemental SBM, feeding CGF reduced (P < .01) hay intake. Calculated daily intakes of ME were 12, 17, 18 and 17 Mcal for NC, SBM, SBM/CGF and CGF, respectively. Hay intake, DM and ADF digestibilities and ME intakes tended to be higher for SBM/CGF than for the average of SBM and CGF fed alone. Compared to supplemental SBM, feeding CGF resulted in higher (P < .05) molar proportions of propionate and butyrate at 4 h after feeding. If increased ruminal concentration of propionate reflects increased ruminal production, CGF feeding may increase efficiency of use of dietary energy and may reduce loss of energy as methane from the rumen.
Comparison of SH and CGF as energy sources

Bernard et al. (1988) used 6 ruminally and abomasally cannulated steers in a 6 x 6 Latin square design to examine the effect of different supplemental energy sources on ruminal fermentation and protein metabolism. Approximately 60% of the energy was provided by sorghum silage. Supplements were formulated to provide 2.8 kg DM daily and were 15.6% soybean meal (SBM) plus 84.6% ground shelled corn, 6.1% SBM + 93.9% SH, 100% CGF, 100% wheat middlings (WM), 50% CGF plus 50% WM and 50% CGF plus 50% SH. They reported no change in ruminal and total tract digestibilities of NDF and organic matter in steers fed SH or CGF diets, but total tract digestibilities of ADF (76.5 vs. 67.8%) and N (73.3 vs. 62.34%) were higher (P < .05) when steers were fed CGF diets than when fed SH diets. Ruminal pH was significantly lower in steers fed SH diets than those fed CGF diets at 0, 1, and 2 h postfeedings, but trends reversed after 6 h postfeeding. Concentrations of acetate (43.7 vs. 32.3 mM/L) at 0 h postfeeding were higher in steers fed the SH diet than those fed the CGF diet and remained unaltered at 1, 2, 4, 6 and 8 h postfeeding between these two treatments. The concentration of propionate was higher (P < .05) at 2, 4, and 6 h postfeeding for steers fed CGF diets.

Highfill et al. (1987) conducted a study in which 4 ruminally cannulated Angus cows were used to compare CGF and SH to a concentrate containing 95% corn - 5% soybean meal (C-S) as supplements to low quality fescue hay. Cows fed 25% SH or CGF diets had higher NDF digestibilities than did those fed 25% C-S diets. In a similar study in the same report, the NDF digestibility of the SH diet was higher than that of cows fed C-S, with the CGF treatment being intermediate and not different from either SH or C-S. In situ DM digestibility (ISDMD) of hay at 48 h followed similar trends. However, these effects do not appear related to ruminal pH because mean pH was not affected (P > .05) in this trial and was always above 6.5. Therefore, CGF and SH appear
to be of similar value as energy sources in dairy diets. Limited work has been done comparing lactation potential of cows fed CGF or SH as energy sources in the same study.

**SH and CGF as roughage sources**

Bernard and McNeill (1991) conducted a trial in which CGF and SH were substituted for a portion of corn silage in the control diet to provide 22% of the total dietary DM. They reported that apparent digestibilities of ADF and NDF were greater when cows consumed SH diets; however, these diets had more than 7 percentage units more NDF. There were no differences in milk or SCM yield due to treatments, but a tendency ($P > .05$) for increased milk fat percentage was noted when cows consumed diets containing SH. Milk protein percentage and yield were greater ($P < .01$) when cows consumed diets containing CGF compared with those containing SH. Firkins et al. (1991) reported that feeding CGF at 20% of diet DM to replace corn silage to lactating dairy cows increased milk and milk protein yield.

Hsu et al. (1987) formulated diets containing com fiber (CF), which essentially is CGF without steep liquor added, or SH as roughage sources for non-lactating ruminants. The NDF concentration of the CF and SH diets were 50.5 and 59.5%, respectively. Rate of ruminal DM disappearance in situ (3 to 36 h) and extent of DM disappearance (36 h) were higher for SH than CF. Sheep fed CF and SH diets had higher digestibilities of NDF and ADF compared with sheep fed oat hulls or cotton seed hulls, probably due to the low lignin concentration of CF and SH. Total tract digestibilities of DM, OM, NDF, and ADF by sheep fed CF and SH diets were similar and were higher than 70%, indicating that these feed by-products are high in nutritive value. Steers fed SH had similar daily weight gains but greater feed intakes when compared with those fed corn diets (Hsu et al., 1987). Steers fed CF had similar daily gains, lower feed intakes and similar feed:gain ratios compared with steers fed the control diet.
Sarwar et al. (1991) conducted an experiment in which forage NDF was partially replaced by NDF of SH or CGF, and diets were fed to ruminally and duodenally cannulated heifers. Based on ruminal pH, acetate:propionate, and fiber digestibilities, they speculated that a minimum of 65% of total dietary NDF could be from forage. The National Research Council recommends 75% (1989). Froetschel and Amos (1991) conducted a trial in which ruminally fistulated steers were fed a 60:40 roughage-concentrate diet at 1.5 x maintenance. Diets had either sorghum silage (SS) or a 63:33 mixture of SS and SH as roughage. They reported that substitution of SH for SS resulted in ruminal pH and VFA patterns expected from increasing the proportion of concentrate in the diet. Ruminal pH was reduced .32 percentage units, total VFA concentration increased by 11%, and the ratio of acetate:propionate was 11% lower with SH substitution of SS. The fractional turnover rate (FTR) of ruminal ADF was increased by 22.5% with SH substitution. Whereas Klofenstein and Owens (1987) found SH NDF to be digested .4 - 2.5 times that of most forages, Anderson et al. (1987) and Sarwar et al. (1991) found no difference in rates, but did report greater extent of NDF digestion.

In conclusion, high energy supplements containing large quantities of corn or other high-starch cereal grains seriously hinder forage utilization in the rumen. The reduced forage digestibility of diets supplemented with greater amounts of cereal grains probably is due to inhibitory effects of reduced ruminal pH and high NSC concentrations. Cows fed such diets can suffer from ruminal acidosis, off feed problems, laminitis and displaced abomasum. These metabolic disorders not only cause reduction in milk yield but also increase the therapy expenditure. However, many cows need 50% or more concentrate to increase energy availability for high milk production. Therefore, identification of feed ingredients capable of eliminating or at least minimizing the adverse effects of high energy dense diets on performance of lactating cows is necessary. Corn gluten feed and SH are feed ingredients which are low in starch and high in rapidly
degradable NDF. These feed by-products can successfully replace a portion of concentrate or forage of the diet. These feed by-products may provide a suitable alternative to maintain animal performance when forage supplies or concentrates are inadequate or expensive to feed. Unfortunately, the potential of these feed by-products to provide effective fiber or to dilute NSC has not been studied completely. Their flexibility is important for dairy producers to increase efficiency of milk production under different feeding scenarios. This thesis provides evidence for the possible replacement of forage fiber and corn from dairy diets with SH or CGF.

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CHAPTER III

EFFECT OF REPLACING NEUTRAL DETERGENT FIBER OF FORAGE WITH SOYHULLS AND CORN GLUTEN FEED FOR DAIRY HEIFERS

INTRODUCTION

Feeding grains high in starch decreases fiber digestion (Joanning et al., 1981; Lonsdale et al., 1971). Mertens and Loftens (1980) suggested that the mechanism responsible for reduction of fiber digestion by cows fed mixed forage-concentrate diets was decreased cellulolytic microbial activity due to decreased ruminal pH. Formulation of dairy cattle diets for NDF concentration has been proposed as a means to accurately determine optimal proportions of feed fiber and energy to maximize FCM production (Colenbrander et al., 1986; Kilmer et al., 1979). However, the NDF procedure is only a crude chemical analysis, and physical properties of NDF from various sources also are important modulators of their ruminal degradation (Shaver et al., 1986; Woodford et al., 1986). Consequently, balancing rations based on NDF may be inappropriate if significant differences occur for physical or chemical properties of the NDF sources.

Sources of non-forage NDF generally are considered to have less "effective fiber" (needed to stimulate rumination and salivation) than do forage sources. To account for this disparity, the NRC (1989) recommends that greater than 75% of dietary NDF should be from forage, but supporting data are limited. Substituting corn gluten feed (CGF; Macleod et al., 1985) or soybean hulls (SH; Nakamura and Owen, 1989) for corn has been shown to increase percentage milk fat. Therefore, it is likely that these by-products have some "effective fiber", but part of the response could be due to decreasing non-
structural carbohydrates. The hypothesis was that SH or CGF can replace a portion of forage fiber in dairy diets without affecting performance. Therefore, research is needed to determine optimal percentages of ration NDF from forage in dairy diets containing significant amounts of fibrous concentrates. Moreover, it is unlikely that all by-products are similar in "effective fiber". Corn gluten feed (45% NDF) is high in hemicellulose, but SH (67% NDF) is high in cellulose (NRC, 1989). This study was designed to examine the effects on site of nutrient digestion by heifers when NDF of SH and dry CGF replaced forage NDF in diets with equal total NDF.

MATERIALS AND METHODS

Five Holstein heifers (2 yr old, 500 kg), fitted with ruminal cannulae and simple T cannulae in the proximal duodenum, were used in a 5 x 5 Latin square design. Five experimental diets (Table 1), each balanced for 30% NDF, were formulated but contained about 31% NDF were formulated. The contribution of forage NDF to total dietary NDF for control (C85), CGF65, CGF45, SH65 and SH45 were 85, 65, 45, 65 and 45%, respectively, with the remainder of the NDF contributed by basal ingredients and CGF or SH for appropriate diets. Moderate quality alfalfa hay was chopped without a screen. Duplicate forage samples were dry-sieved and a mean particle size (5.3 mm) determined as done by other researchers (Waldo et al., 1971). All diets were mixed daily and fed twice daily at 1% of BW for each feeding to allow for no orts.

Approximately 2 g of DM of SH, CGF and alfalfa hay (ground through 2- mm screen) were weighed into dacron bags (8 x 14 cm) with pore size averaging 51 ± 10 um (Tetko, Inc., Elmsford, NY; product number HC 7-51). Bags were inserted after feeding and incubated for 1, 2, 4, 6, 10, 16, 24, 36, 48 and 96 h. All heifers received separate bags with hay only; heifers fed diets supplemented with SH or CGF also received bags with SH or CGF, respectively. Upon removal from the rumen, bags were washed with running tap water until wash was clear (about 1 min) and were dried (55°C). The feeds
TABLE 1. Composition of diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF.

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>C85</th>
<th>CGF45</th>
<th>CGF65</th>
<th>SH45</th>
<th>SH65</th>
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<tr>
<td>Alfalfa hay, chopped</td>
<td>25.25</td>
<td>13.37</td>
<td>19.31</td>
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Chemical composition

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1 Dietary NDF from forage: 85, 45, 65, 45 and 65% for C85, CGF45, CGF65, SH45 and SH65, respectively.

2 Mean particle size was 5.3 mm by dry-sieving.

3 Vitamin premix contained 2954, 409, and 34 IU vitamins A, D, and E per gram.

4 Contained (g/kg): Na, 380; Cl, 580; Mn, 2.0; Zn, 2.0; Mg, 1.0; S, .4; Fe, 1.0; Ca, .25.

5 Contained .20 g Se/kg.
and residues were analyzed for NDF (Robertson and VanSoest, 1977). Disappearance rates of potentially digestible NDF disappearance from hay and by-products were determined by regressing the natural logarithm of percentage original NDF remaining in bags between 1 and 96 h or the end point observed visually after plotting data.

The heifers were given 10 d for adaptation to diets at the start of each experimental period, followed by a 7-d collection period. Cobalt-EDTA was prepared (Uden et al., 1980) and mixed (.5 g Co per d) with the diet daily as a duodenal fluid-phase flow marker. Chromic oxide was mixed with concentrates and used as the digestibility marker. On d 11 to 14 of each period, duodenal samples (250 ml) were collected every 6 h in a sampling scheme that allowed 1 sample for each 90 min of a 24-h schedule (16 samples). Samples were composited by animal for each period. Fecal grab samples were taken twice daily such that a sample was obtained for every 3-h interval of a 24-h time period (8 samples). Feeding of Co-EDTA and Cr₂O₃ was stopped on d 14. On d 15, about .5 kg of each meal was sprayed with an aqueous solution of ytterbium chloride (YbCl₃ 6H₂O; .75 g Yb) and fed. After consumption of the marked portion, the rest of the meal was offered. At the same time, Cr-EDTA (1 g Cr) was pulse-dosed through the rumen cannulae of each heifer. Because Cr-EDTA and Cr₂O₃ associate almost exclusively with fluid and particles, respectively, centrifugation of samples prior to Cr analyses of fluid negates any interferences by Cr₂O₃ (Firkins et al., 1986a). Ruminal samples (400 ml) were taken at 4, 8, 12, 24, 36 and 48 h post-dosing. A core sampler (Firkins et al., 1986a) was used to obtain ruminal samples, which were squeezed through 4 layers of cheesecloth. The liquid fraction was frozen, centrifuged at 25,000 x g, and analyzed subsequently for Cr concentration (Firkins et al., 1986); the solid portion was dried (55°C), ground through a 2-mm screen, and analyzed for Yb using atomic absorption spectrophotometry (Firkins et al., 1984). Fluid and particulate dilution rates from the rumen were determined by regressing the natural logarithms of Cr or Yb concentrations.
vs. time. In general, $R^2$ values of regressions were greater than .95. Ruminal volume was calculated as dose of Cr divided by y-intercept of Cr regression. Outflow was calculated by multiplying fluid dilution rate x 24 h/d x ruminal volume.

On d 15, ruminal contents (500 ml) were sampled at 3, 6, 9 and 12 h after the morning feeding. After mixing of these samples, 50 ml from each were obtained and the remaining 450 ml of sample were returned to the rumen. Ruminal pH was measured immediately, and then the sample was squeezed through 4 layers of cheese cloth, 3 ml of 6N HCl were added to terminate fermentation, and the sample was frozen. After thawing, these samples were centrifuged at 27,000 x g prior to determination of NH$_3$-N (Chaney and Marbach, 1962) and VFA concentrations by GLC (Firkins et al., 1990).

Ruminal contents (500 ml) were taken to harvest bacteria at 0800 h on d 11, 1100 h on d 12, 1400 h on d 13, and 1700 h on d 14. Following compositing by animal, bacteria were expected to have a purine:N ratio that represents an average over the feeding interval. This was necessary to account for any post-prandial changes in bacterial purine:N ratios. Each sample was blended with saline solution in a Waring blender at low speed for 1 min and then was squeezed through 8 layers of cheese cloth. Samples were frozen and later thawed and centrifuged at 500 x g to remove protozoa and feed particles and at 27,000 x g twice (including a wash with saline) to precipitate bacteria (Firkins et al., 1986b). The bacterial isolates were freeze-dried and analyzed for N (AOAC, 1984), organic matter (OM; AOAC, 1984) and purine content (Zinn and Owens, 1986).

Subsamples were taken while duodenal fluid was being stirred continuously with a homogenizer. These samples were centrifuged at 27,000 x g and analyzed for Co (Uden et al., 1980) and NH$_3$-N (Chaney and Morbach, 1962). Duodenal and fecal samples were dried at 55°C and ground in a Wiley mill (2-mm screen). Feed and dried duodenal and fecal samples were analyzed for DM, N and OM (AOAC, 1984), NDF (Robertson and Van Soest, 1977) and ADF (Goering and Van Soest, 1970). Concentrate,
duodenal and fecal samples were analyzed for Cr (Williams et al., 1962). Purine contents (Zinn and Owens, 1986) of duodenal samples were determined. Flow of bacterial N to the duodenum (g/d) was calculated by multiplying daily N flow to the duodenum times N:purine ratio of duodenal samples divided by the ratio of N:purines of the bacterial-rich isolates. Duodenal fluid and NH\textsubscript{3}-N flows to the duodenum were calculated using CoEDTA; all other flows and digestibility calculations were done using Cr\textsubscript{2}O\textsubscript{3}.

Data were analyzed using SAS (1982). The ANOVA and orthogonal comparisons were made using the CONTRAST statement in GLM. Contrasts were 1) C85 vs. average of by-product diets, 2) CGF vs. SH diets, 3) CGF45 and SH45 vs. CGF65 and SH65, and 4) interaction of by-product source vs. level of forage substitution. Least-square means were generated for some calculations because one duodenal sample was lost. Significance at (P < .05) was used unless otherwise noted.

RESULTS AND DISCUSSION

Ruminal fluid dilution rate (D) was higher in heifers fed C85 (7.45%/h) compared with SH (6.54%/h) and CGF (6.23%/h) diets (Table 2). Ruminal volumes were similar, as expected (Varga et al., 1984), so fluid outflows from the rumen followed trends that were similar to data for fluid D. The C85 diet had a higher concentration of forage, which requires more mastication and more salivation. An interaction between by-product source and level was noted for ruminal fluid D (P < .01) and outflow (P < .08). Increasing level of SH had little effect. However, fluid D and outflows by heifers fed CGF65 were similar to data for those fed C85, but were much lower when heifers were fed CGF45. Feeding CGF increased fluid flow to the duodenum in one study (Firkins et al., 1984), but the reason for this is not clear. Anderson et al. (1988) reported that there was no difference in fluid D when both toasted SH or corn were fed at 0, 12.5, 25, or 50% (DM basis) in corn stalkage-based diets to steers. Because particle size strongly influences salivation, it would be expected that most by-products, perhaps with
### TABLE 2. Ruminal characteristics in heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF.

<table>
<thead>
<tr>
<th>Item</th>
<th>C85</th>
<th>CGF45</th>
<th>CGF65</th>
<th>SH45</th>
<th>SH65</th>
<th>SE</th>
<th>Probabilities for contrasts</th>
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<td></td>
<td></td>
<td></td>
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<tr>
<td>Ruminal fluid $D^2$, %/h</td>
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<td>5.48</td>
<td>6.98</td>
<td>6.62</td>
<td>6.45</td>
<td>.291</td>
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<td>Ruminal volume, L</td>
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<td>55.3</td>
<td>57.8</td>
<td>56.3</td>
<td>55.6</td>
<td>4.64</td>
<td></td>
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<tr>
<td>Ruminal fluid outflow, L/d</td>
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<td>71.6</td>
<td>97.2</td>
<td>83.6</td>
<td>85.5</td>
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<td>Ruminal particulate $D$, %/h</td>
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<td>2.93</td>
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<td>.128</td>
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\[ \text{Acetate:propionate} \]

\[
\begin{array}{c|c|c|c|c|c|c|c}
\text{Item} & \text{C85} & \text{CGF45} & \text{CGF65} & \text{SH45} & \text{SH65} & \text{SE} & \\
\hline
\text{Ammonia-N, mg/dl} & 13.99 & 11.15 & 13.69 & 13.87 & 20.50 & 2.188 & .75 \times \text{CGF} vs SH \times \text{level} \\
\text{pH} & 6.05 & 5.75 & 5.98 & 5.55 & 5.91 & .145 & .14 \times \text{CGF} vs SH \times \text{level} \\
\text{Total VFA, mM} & 127.6 & 111.6 & 115.0 & 117.8 & 115.3 & 8.41 & .20 \times \text{CGF} vs SH \times \text{level} \\
\text{VFA, mol/100 mol} &       &       &       &       &       &       &                            \\
\text{Acetate} & 63.0 & 60.0 & 62.2 & 65.6 & 63.3 & 4.06 & .97 \times \text{CGF} vs SH \times \text{level} \\
\text{Propionate} & 19.0 & 20.5 & 18.6 & 18.2 & 19.8 & 1.88 & .90 \times \text{CGF} vs SH \times \text{level} \\
\text{Butyrate} & 13.2 & 13.9 & 14.4 & 11.8 & 12.5 & 1.99 & .99 \times \text{CGF} vs SH \times \text{level} \\
\text{BCVFA}^3 & 4.86 & 5.62 & 4.74 & 4.32 & 4.45 & .511 & .89 \times \text{CGF} vs SH \times \text{level} \\
\text{Acetate:propionate} & 3.47 & 3.30 & 3.52 & 3.73 & 3.35 & .443 & .99 \times \text{CGF} vs SH \times \text{level} \\
\end{array}
\]

\[ \text{BCVFA}^3 = \text{Butyrate} + \text{Valerate} + \text{Caproate} \]
<table>
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<tr>
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<td>.99</td>
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<tr>
<td>Acetate:propionate</td>
<td>.324</td>
<td>.62</td>
<td>.24</td>
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</table>

1 Dietary NDF from forage: 85, 45, 65, 45 and 65% for C85, CGF45, CGF65, SH45, SH45 and SH65 diets, respectively.

2 Dilution rate.

3 Branched-chain VFA were the sum of valerate, isovalerate and isobutyrate.
the exclusion of CGF, would be more similar to corn grain than forage in regard to their effects on ruminal fluid kinetics.

Ruminal particulate D was greater (P < .06) in heifers fed C85 diets than the average of those fed other diets. Anderson et al. (1988) reported values for particulate D that were similar to values of this study. Diets in this study had about 31% NDF (Table 1); consequently, rumen fill probably was not a major factor regulating particulate D. By-products should have a faster rate of degradation than would forage, so replacement of by-products for forage should decrease passage rate of undegraded particulate matter. However, data are limited to support this conclusion because few studies have evaluated replacement of forage with these by-products.

Ruminal NH$_3$-N concentrations were greater when heifers were fed SH than when fed CGF at 3 h and 6 h post-feeding. Highfill et al. (1987) reported that ruminal NH$_3$-N concentration was higher when CGF diets than SH diets were fed. The higher concentration of ruminal NH$_3$-N possibly may be due to higher amounts of urea (.58%) and SBM in SH diets compared with CGF diets (.31%) (Table 1). At 3 h, heifers fed CGF65 or SH65 tended to have greater (P < .06) NH$_3$-N concentrations than did those fed CGF45 or SH45. There may have been less available energy substrate for bacteria to use NH$_3$-N early after feeding for heifers fed the diets with more forage (CGF65 and SH65). These diets also had slightly lower CP (Table 1).

Ruminal pH was not affected by treatment at any time except 3 h, when heifers fed CGF45 or SH45 tended (P < .07) to have lower pH than did those fed CGF65 or SH65 (Table 2). Molar proportion of acetate decreased while molar proportion of propionate increased (P < .08) when 45% of NDF was from forage in comparison with values for the 65% treatments at 6 h post-feeding. Consequently, acetate:propionate was lower for 45 vs 65% of NDF from forage at 6 h post-feeding. All other VFA concentrations were unaffected among diets at the indicated times post-feeding. At 96 h post-feeding,
total VFA were higher for C85 than diets containing by-products. Hsu et al. (1987) showed that pH was lower, but acetate:propionate was higher, in sheep fed SH vs. corn fiber (much like CGF), respectively. Feeding CGF in high-forage diets to cattle decreased acetate:propionate more than did feeding SH (Bernard et al., 1988). Based on data of this study and literature values, shifts in microbial populations or end-products may have occurred when animals were fed SH vs. CGF, and probably were due to chemical differences between CGF and SH rather than to pH changes in the rumen.

Intake of organic matter was higher when heifers were fed C85 than other diets, (Table 3). This small increase in OM intake should have had little effect on kinetics of ruminal digestion. Duodenal OM flow tended to be lower (P < .08), and apparent (P < .08) and true (P < .09) ruminal digestibilities tended to be greater, when heifers were fed diets with CGF vs. SH. Because fiber digestibilities did not follow the same trend (Table 4), these results probably were due to lower apparent ruminal degradability of CP by heifers fed SH diets (Table 5). Apparent total tract OM digestion was lower (P < .06) in the C85 (69%) treatment in comparison with SH and CGF (72.3%) treatments. An interaction (P < .06) between by-product source x level for apparent total tract OM digestion also was noted. These effects were due to differences in total tract NDF digestibilities (Table 4), and will be discussed later.

Ruminal disappearance in situ of alfalfa hay or by-products is shown in Figures 1 and 2. Values for extents of digestion are shown for 36 h in Table 4 because this sampling time point most closely approximates turnover time (1/particulate D) of ruminal digesta (Table 2). When heifers were fed C85 vs. by-product diets, extent of hay NDF disappearance was higher at 36 (P < .02) and 96 h (P < .10). Feeding diets with SH depressed extent of hay disappearance at 36 h, but increased rate of hay disappearance. Also, the greater the substitution of by-product NDF for forage NDF, the greater the depression in extent of by-product at 96 h (P < .06) and alfalfa hay disappearance. The
TABLE 3. Organic matter (OM) digestion by heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF.

<table>
<thead>
<tr>
<th>Item</th>
<th>Treatments</th>
<th>Probabilities for contrasts</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C85</td>
<td>CGF45</td>
</tr>
<tr>
<td>OM intake, g/d</td>
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<td>8927</td>
</tr>
<tr>
<td>Duodenal OM flow, g/d</td>
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<td>5191</td>
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<td>Fecal OM excretion, g/d</td>
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<td>2572</td>
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<tr>
<td>Apparent ruminal OM digestion, % of intake</td>
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<td>42.0</td>
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<td>True ruminal OM digestion, % of total</td>
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<td>58.0</td>
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<tr>
<td>Apparent total tract OM digestion, % of intake</td>
<td>69.0</td>
<td>71.0</td>
</tr>
</tbody>
</table>

1 Dietary NDF from forage: 85, 45, 65, 45, and 65% for C85, CGF45, CGF65, SH45, and SH65 diets, respectively.

2 Corrected for bacterial OM in duodenal digesta.
TABLE 4. Digestion of fiber by heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF.

<table>
<thead>
<tr>
<th>Item</th>
<th>C85</th>
<th>CGF45</th>
<th>CGF65</th>
<th>SH45</th>
<th>SH65</th>
<th>SE</th>
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<tr>
<td><strong>Ruminal NDF disappearance</strong></td>
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<td></td>
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<tr>
<td>Hay, % at 36h</td>
<td>38.7</td>
<td>32.7</td>
<td>38.4</td>
<td>25.6</td>
<td>36.4</td>
<td>1.89</td>
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<tr>
<td>Hay, % at 96h</td>
<td>43.5</td>
<td>39.5</td>
<td>40.3</td>
<td>35.3</td>
<td>43.7</td>
<td>1.90</td>
</tr>
<tr>
<td>By-product, % at 36h</td>
<td>48.7</td>
<td>54.1</td>
<td>41.3</td>
<td>52.2</td>
<td>5.82</td>
<td>.45</td>
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<tr>
<td>By-product, % at 96h</td>
<td>82.7</td>
<td>86.0</td>
<td>65.9</td>
<td>88.7</td>
<td>5.93</td>
<td>.26</td>
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<tr>
<td>Hay, %/h</td>
<td>6.23</td>
<td>5.98</td>
<td>4.19</td>
<td>5.45</td>
<td>5.83</td>
<td>.427</td>
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<tr>
<td>By-product, %/h</td>
<td>2.92</td>
<td>3.59</td>
<td>3.61</td>
<td>3.34</td>
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<td><strong>Intake, g/d</strong></td>
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<td>NDF</td>
<td>3027</td>
<td>2993</td>
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<td>2979</td>
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<td>1953</td>
<td>1451</td>
<td>1670</td>
<td>1920</td>
<td>1884</td>
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<td><strong>Duodenal flow, g/d</strong></td>
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<td>1965</td>
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<td><strong>Apparent ruminal digestion, % of intake</strong></td>
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TABLE 4. continued

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<th>CGF45</th>
<th>CGF65</th>
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<td>.23</td>
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</table>

1 Dietary NDF from forage: 85, 45, 65, 45, and 65% for C85, CGF45, CGF65, SH45 and SH65 diets, respectively.

2 CGF or SH for heifers fed CGF or SH diets, respectively.

3 Rates of potentially digestible NDF disappearance.
TABLE 5. Nitrogen (N) digestion and efficiency of bacterial growth in dairy heifers fed diets with corn gluten feed (CGF) or soyhulls (SH) replacing forage NDF.

<table>
<thead>
<tr>
<th>Item</th>
<th>Treatments</th>
<th>C85</th>
<th>CGF45</th>
<th>CGF65</th>
<th>SH45</th>
<th>SH65</th>
<th>SE</th>
<th>Probabilities for contrasts</th>
</tr>
</thead>
<tbody>
<tr>
<td>N intake, g/d</td>
<td></td>
<td>195</td>
<td>198</td>
<td>191</td>
<td>207</td>
<td>200</td>
<td>2.3</td>
<td></td>
</tr>
<tr>
<td>Duodenal flow, g/d</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Ammonia N</td>
<td></td>
<td>5.1</td>
<td>5.0</td>
<td>4.8</td>
<td>5.0</td>
<td>5.1</td>
<td>.79</td>
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<td>Non-ammonia N</td>
<td></td>
<td>183.7</td>
<td>185.0</td>
<td>165.2</td>
<td>183.2</td>
<td>191.9</td>
<td>10.31</td>
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<tr>
<td>Bacterial N</td>
<td></td>
<td>105.1</td>
<td>95.4</td>
<td>89.4</td>
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<td>87.5</td>
<td>9.71</td>
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<tr>
<td>Ruminal N escape</td>
<td></td>
<td>73.2</td>
<td>84.6</td>
<td>71.0</td>
<td>91.2</td>
<td>99.3</td>
<td>4.98</td>
<td></td>
</tr>
<tr>
<td>% of intake</td>
<td></td>
<td>38.0</td>
<td>43.3</td>
<td>37.6</td>
<td>44.4</td>
<td>49.5</td>
<td>2.58</td>
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</tr>
<tr>
<td>Efficiency of bacterial growth</td>
<td></td>
<td>30.0</td>
<td>26.6</td>
<td>24.4</td>
<td>28.2</td>
<td>33.0</td>
<td>4.98</td>
<td></td>
</tr>
<tr>
<td>g N/kg OMDRA²</td>
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<td>22.0</td>
<td>20.0</td>
<td>18.5</td>
<td>20.7</td>
<td>23.0</td>
<td>2.70</td>
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</tr>
<tr>
<td>g N/kg OMDRT³</td>
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<td>79.0</td>
<td>84.0</td>
<td>76.0</td>
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<td>82.0</td>
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<td>Fecal N, g/d</td>
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</tr>
<tr>
<td>Apparent total tract</td>
<td></td>
<td>60.0</td>
<td>57.0</td>
<td>60.0</td>
<td>65.0</td>
<td>59.0</td>
<td>1.53</td>
<td></td>
</tr>
<tr>
<td>N digestion, % of N intake</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

1 Dietary NDF from forage: 85, 45, 65, 45, and 65% for C85, CGF45, CGF65, SH45 and SH65 diets, respectively.

2 Apparent ruminal organic matter digestibility.

3 True ruminal organic matter digestibility.
Figure 1. Disappearance (± SE) of alfalfa hay NDF in situ by heifers fed diets with 85, 45, 65, 45, and 65% NDF from forage for control (C85), corn gluten feed (CGF) or soyhull (SH) diets.
Figure 2. Disappearance (± SE) of NDF of corn gluten feed (CGF) and soybean hulls (SH) in situ by heifers fed diets containing CGF or SH in which 45 or 65% of total dietary NDF was from forage.
by-product source x level interaction (P < .07) at 96 h was due to lower hay NDF disappearance in situ by heifers fed SH45. Decreases in ruminal pH are known to depress fiber digestion due to inhibition of cellulolytic bacteria (Mould et al., 1983). Because pH values generally were lower when heifers were fed SH45 than when fed CGF 45 (Table 2), it appears that pH may have inhibited ruminal fiber digestion by those fed SH45 more than by those fed CGF45 (Figures 1 and 2). Rates of digestion did not follow the same trends as did extent of digestion, suggesting that reporting only rate data may be misleading. This discrepancy likely was due to correction of data for extent of digestion before calculating rate of "potentially digestible" fiber digestion. Rates and extents of NDF disappearance of CGF were similar to those reported by Kampman and Loerch (1989). Data are limited, however, for SH NDF disappearance in situ in animals fed high-concentrate diets. Extent of DM disappearance in situ at 36 h was 87.6 and 103.9% for corn fiber and SH, respectively (Hsu et al., 1987). Because that study was done with alfalfa hay-fed cattle, it is likely that digestion of by-product fiber in this study, which had much lower values, was inhibited by lower ruminal pH.

Intake of NDF was not affected by diet (Table 4), but all contrasts were significant for ADF intake. The fiber in CGF has relatively more hemicellulose (NDF-ADF) than does fiber in alfalfa hay or SH (NRC, 1989). Increased substitution of forage NDF by that in CGF, but not SH, decreased dietary ADF, explaining by-product level effects and the source x level interaction. Trends for by-product source x level interactions for duodenal NDF flow (P < .10) and ruminal NDF digestibilities (P < .08) were detected. Heifers fed CGF45 had higher ruminal digestibilities than did those fed CGF65; however, an opposite effect for by-product substitution was observed when SH were fed. As reflected by in situ data, by-product NDF should have been more digestible than forage NDF in the rumen. Thus, greater ruminal NDF digestibilities by heifers fed increasing substitution of CGF or SH for forage should have occurred. However, rapid washout of
suspended CGF fiber may have occurred when heifers were fed CGF65; particulate D was unaffected by treatment, but these animals also had high ruminal fluid outflows (Table 2). As reported by Firkins et al. (1986), ruminal fluid outflow appeared to have more impact on ruminal NDF digestibilities than did particulate D. In that study, where CGF also was fed, ruminal fluid outflow was strongly correlated to ruminal NDF digestion ($r = -0.60, P < 0.01$). The lower ruminal NDF digestibilities noted when heifers were fed SH45 vs. SH65 probably was due to ruminal pH depressing digestion of both forage and SH NDF in the SH45 diet. As discussed previously, NDF disappearance in situ was lower for hay and tended to be lower for SH, when heifers were fed SH45 than other diets. Because ruminal ADF digestibilities did not follow similar trends, it appears that hemicellulose digestion was particularly inhibited when heifers were fed SH45. As discussed before (Firkins et al., 1986), factors limiting hemicellulose digestion in the rumen may be due to limitation of xylan digestion until arabinose and other branch points are cleaved. Heifers fed CGF had lower ruminal ADF digestibilities because higher proportions of total dietary ADF were from forage in these diets, compared to SH diets.

As with ruminal data, total tract NDF digestion was lower when heifers were fed C85, which had higher proportions of NDF from forage, in comparison with diets containing by-products. Although heifers fed CGF65 or SH45 had lower ruminal NDF digestibilities, they had higher total tract NDF digestibilities than did those fed CGF45 or SH65, resulting in a significant interaction. Total tract ADF digestion was higher for SH than CGF, and a by-product source x level interaction also occurred. When ruminal digestibilities were expressed as percentages of total tract digestion, 92-93% of total tract NDF digestion occurred in the rumen when heifers were fed C85, CGF45 or SH65. These percentages are similar to others reported by Firkins et al. (1986) with similar dietary conditions. However, heifers fed CGF65 or SH45 digested only 66 and 64% of
digestible NDF in the rumen, respectively. In the study reported by Hsu et al. (1987), much more NDF was digested post-ruminally when sheep were fed SH than corn fiber. However, Bernard et al. (1988) noted that a significant proportion of CGF fiber was digested post-ruminally. Replacing forage NDF with by-product NDF should have increased total tract NDF digestibilities, as occurred with feeding SH. Low pH resulting from rapid fermentation of carbohydrates may have limited fiber digestion in the rumen but not in the total tract of heifers fed SH45. Hindgut pH was not measured in this study, but it may have been depressed in the hindgut when heifers were fed CGF45, limiting hindgut fermentation of fiber by these heifers. Siciliano-Jones and Murphy (1989a,b) reported that feeding diets similar in percentage forage to our CGF45 diets decreased cecal pH and hindgut fiber degradation. Thus, compensatory fiber digestion that occurred in the hindgut of heifers fed SH45 may not have occurred in heifers fed CGF45 due to differences in hindgut pH.

As can be seen from Table 4, ruminal ADF digestibilities sometimes were higher than were total tract ADF digestibilities. This effect could be due to the relatively larger variation in estimation of ruminal than total tract fiber digestibilities, and thus ruminal digestibilities may not be significantly different from total tract digestibilities. However, this problem has been observed before when high-concentrate diets were fed (Kerley et al., 1985). Kerley et al. (1985) explained that some phenolic-protein complexes can be formed in the gut, causing artifact fiber formation. Several feed and digesta samples were re-analysed for neutral detergent insoluble N (NDIN) and ADIN, which appeared to cause only about a 2% increase in NDF or ADF from artifact fiber in these samples. It may be due to errors resulting from overestimation of duodenal flows. It is important to note that NDF and ADF procedures were developed to analyze forage fiber. More research is needed to modify NDF and ADF procedures for concentrate and digesta samples.
Intake of N was affected by treatment (Table 5), although differences were small. Hay lost some CP during processing, so increased substitution of forage with by-product increased N intake. The SH had higher CP (15.4%) than expected (12%), explaining the effect of by-product source. However, all diets provided CP over requirements and were balanced to have adequate rumen-degradable N (NRC, 1989). Duodenal flows of NH$_3$-N, non-NH$_3$-N and bacterial N were not affected by treatment. Apparent escape of dietary N from the rumen was lower when heifers were fed C85 than the average of those fed by-products. Because basal ingredients in by-product diets were similar, it appears that the greater protein escape of SH than CGF diets was due directly to the by-products. Corn gluten feed has a very high non-protein N content (amino acids and peptides) and low protein escape from the rumen (Firkins et al., 1984). Thus, the by-product source x level interaction (P < .06) probably was due to higher than expected ruminal escape of protein by heifers fed CGF45. McDougall’s artificial saliva was acidified with 6 N H$_2$SO$_4$ to pH 5, 6, and 7, and duplicate samples of SH and CGF were incubated in buffer solutions, as done by Loerch et al. (1983). Percentages of insoluble N were 87.0, 76.9, and 87.8, respectively for SH at pH 7, 6, and 5; and were 53.8, 51.7 and 50.3, respectively, for CGF at pH 7, 6, and 5. Proteins are least soluble at their isoelectric pH (Loerch et al., 1983), and decreased pH from feeding higher concentrate diets has been shown to decrease solubility (Loerch et al., 1983) and increased ruminal escape of soybean meal (NRC, 1989). Because data of this study did not change markedly with differences in pH of the solvent, it appears that ruminal factors other than pH may have affected escape of CGF and SH protein by heifers fed the different diets.

Interactions in fecal N excretion and apparent total tract N digestibilities were significant. Heifers fed SH45 appeared to have greater N digestibilities, but those fed CGF45 had lower N digestibilities than did those fed SH65 and CGF65. Reasons for this effect are not clear.
In conclusion, if CGF or SH diets had been fed to lactating cows, it is not likely that total OM digestion would have been significantly lower than digestion of control, even if negative associative effects had been more severe than in this study. Although disappearance of by-product NDF (Table 4) may have been depressed by low ruminal pH in this study, it still should have had a greater extent of ruminal digestion than forage NDF. Nitrogen digestibilities were affected, but total non-NH$_3$-N flow to the duodenum was not changed, by these dietary manipulations (Table 5). The major responses of lactating cows to replacing forage NDF with by-product NDF probably would be milk fat percentage and DM intake. Based on rumen characteristics (Table 2), feeding SH45, and probably CGF45, likely would cause milk fat depression because negative associative affects would be exacerbated by feeding much higher intakes to lactating cows. Based on data reported by Woodford and Murphy (1988), DM intake and milk fat percentage may be reduced significantly by feeding SH45 or CGF45 but not SH65 or CGF65. Based on results in Chapter 4 and the current reasoning, 65% of dietary NDF as forage NDF should be sufficient when CGF or SH are fed. However, 45% of dietary NDF as forage NDF may be adequate when these by-products are fed; however, a lactation study is needed before application. Actual usage of these by-products as fiber sources within this context ultimately should depend on their price relative to that of forage.

REFERENCES


CHAPTER IV

EFFECT OF REDUCING FORAGE NDF AND NON-STRUCTURAL CARBOHYDRATES ON NUTRIENT DIGESTIBILITY AND MILK PRODUCTION BY DAIRY COWS

INTRODUCTION

Fiber is considered essential to maintain high feed intake, proper rumen function and normal milk fat test (NRC, 1989) in lactating cows. However, energy usually is the limiting nutrient for high producing dairy cows when considerable amounts of forage, even if of good quality, are included in the diet (Balch, 1976). As cereal grains are fed at increasing levels (e.g., 50% of the diet), acid production from fermentation of non-structural carbohydrate (NSC) overcomes the buffering capacity in the rumen, proportionately reducing ruminal fluid pH (El-Shazly et al., 1961), fiber digestion (Henning et al., 1980; Miller and Muntifering, 1985), percent milk fat (Sutton, 1989; Sutton and Morant, 1989), and perhaps DM intake (Joanning et al., 1981). These problems can be reduced by supplying a proper proportion of structural carbohydrates (NDF) in dairy diets.

Dairy diets can be balanced for NDF, but sources of NDF vary in their chemical and physical properties. Finely ground forages and cereal by-products high in NDF do not stimulate rumination and salivation relative to feeding coarsely chopped forage. According to the NRC (1989), at least 75% of dietary NDF in dairy diets should come from forages, but scientific evidence to support this recommendation for diverse situations is limited. When corn gluten feed (CGF) or soyhulls (SH) replaced forage NDF such that only 65% of total NDF (31% of diet DM) was from forage, dairy heifers maintained ruminal pH and acetate:propionate ratio, and they had greater NDF digestibility.
compared with the control treatment (85% NDF from forage; Sarwar et al., 1991). Feeding CGF at 20% of diet DM plus 1% sodium bicarbonate to replace corn silage increased yield of milk and milk protein (Firkins et al., 1991). Limited data are available on the performance and response of lactating dairy cows fed diets in which forage NDF was replaced with NDF of SH.

Cows fed diets containing 32.9% NSC diet produced more milk, and metabolizable energy was used more efficiently compared with cows fed diets containing 24.9% (Macgreger et al., 1983). In another study (Moran and Trigg, 1989), milk protein % and protein:fat ratio increased linearly with increasing dietary NSC concentration (20.5 to 35%). The increased milk protein yield mainly was due to increasing supplies of energy-yielding constituents in the diet. However, Varga and Whitsel (1991) could not establish a clear relationship between the NSC:NDF ratio by examining the rate and extent of nutrient disappearance in situ. Data are limited to evaluate effects of ranges of NSC concentrations without confounding effects of varying forage concentration. The hypothesis was that SH can replace a portion of forage fiber and concentrate in dairy diets without adversely affecting lactation performance.

The objectives of this study were to examine the potential of SH fiber to replace forage fiber with equal dietary NSC and evaluate various dietary NSC concentrations with equal amounts of dietary forage on milk yield and composition, ruminal characteristics and nutrient digestibilities by cows.

MATERIALS AND METHODS

Five ruminally cannulated, primiparous Holstein cows were used in a 5 x 5 Latin square design. Cows averaged 57 ± 40 d in lactation; much of the variation was due to one cow being 122 days in lactation. Five experimental diets (Table 6) were formulated. The control (C) diet was balanced to have 43% forage had 32% NDF, 42% NSC and 80% of the total dietary NDF from forage NDF (Table 7). Concentration of NSC was
TABLE 6. Ingredient composition of diets varying in concentrations of NDF and non-structural carbohydrates (NSC) from forage NDF.

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Diets(^1)</th>
<th>C</th>
<th>NDF70</th>
<th>NDF60</th>
<th>NSC35</th>
<th>NSC25</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaline hay</td>
<td></td>
<td>21.61</td>
<td>18.36</td>
<td>15.56</td>
<td>21.60</td>
<td>21.60</td>
</tr>
<tr>
<td>Corn silage</td>
<td></td>
<td>21.61</td>
<td>18.36</td>
<td>15.56</td>
<td>21.60</td>
<td>21.60</td>
</tr>
<tr>
<td>Corn, ground shelled</td>
<td></td>
<td>39.88</td>
<td>41.55</td>
<td>42.16</td>
<td>23.01</td>
<td>7.98</td>
</tr>
<tr>
<td>Soybean meal, 44% CP</td>
<td></td>
<td>13.40</td>
<td>14.04</td>
<td>14.69</td>
<td>6.48</td>
<td>6.86</td>
</tr>
<tr>
<td>Blood meal</td>
<td></td>
<td>1.08</td>
<td>.86</td>
<td>.76</td>
<td>1.30</td>
<td>1.73</td>
</tr>
<tr>
<td>Soybeans, roasted</td>
<td></td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>6.48</td>
<td>11.67</td>
</tr>
<tr>
<td>Soyhulls</td>
<td></td>
<td>-</td>
<td>4.32</td>
<td>8.64</td>
<td>17.29</td>
<td>31.76</td>
</tr>
<tr>
<td>T.M. Salt</td>
<td></td>
<td>.50</td>
<td>.50</td>
<td>.50</td>
<td>.50</td>
<td>.50</td>
</tr>
<tr>
<td>Selenium premix(^2)</td>
<td></td>
<td>.13</td>
<td>.13</td>
<td>.13</td>
<td>.13</td>
<td>.13</td>
</tr>
<tr>
<td>Dicalcium phosphate</td>
<td></td>
<td>.45</td>
<td>.47</td>
<td>.47</td>
<td>.51</td>
<td>.60</td>
</tr>
<tr>
<td>Limestone</td>
<td></td>
<td>1.17</td>
<td>1.20</td>
<td>1.30</td>
<td>.86</td>
<td>.43</td>
</tr>
<tr>
<td>Magnesium oxide</td>
<td></td>
<td>.17</td>
<td>.21</td>
<td>.23</td>
<td>.24</td>
<td>.28</td>
</tr>
<tr>
<td>Megalac(^3)</td>
<td></td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>.86</td>
</tr>
</tbody>
</table>

\(^1\) C = control diet, NDF70 = 70% NDF from forage, NDF60 = 60% NDF from forage, NSC35 = 35% non-structural carbohydrates, NSC25 = 25% non-structural carbohydrates.

\(^2\) Contains 198 mg of selenium per kg.

\(^3\) Calcium salts of palm oil fatty acids distillate (Church and Dwight Co., Inc., Princeton, NJ).
estimated using the difference technique (Nocek and Russell, 1988) of feedstuffs using NRC values for feedstuffs. The NDF70 (37% forage) and NDF60 (31% forage) diets were formulated to have 70 and 60%, respectively, of the total dietary NDF from forage NDF while maintaining equivalent total NDF (31%) and NSC (42%) concentrations as the C diet. To study effects of varying NSC in concentrate, NSC35 and NSC25 diets were formulated to have 35 and 25% NSC, but diets had the same percentage forage (43% of DM) as control (42% NSC). The NSC concentrations in NSC35 and NSC25 diets were attained by substituting SH, roasted soybeans and fat from calcium soups (Church and Dwight Co., Inc., Princeton, NJ) for the concentrate portion of diets. The soybeans were roasted at 149°C and steeped for 30 min, and fed whole (no reduction in particle size). All diets were formulated to be isonitrogenous and isoenergetic using NRC (1989) values for energy in all feeds and for protein in concentrates.

Moderate quality alfalfa hay was chopped without a screen. Diets were mixed daily but fed twice a day at ad libitum intakes. Cows were fed for 5 periods, each of which lasted 4 wk. The first 2 wk were allowed for dietary adaptation. Daily feed intake and milk production were averaged over the last 2 wk. Milk samples (am and pm) were taken twice weekly during the last 2 wk of each period and analyzed for milk fat and protein concentrations by infrared spectroscopy (DHI Cooperative Inc., Powell, OH). Cows were weighed once weekly during the last two weeks of each period, and weights were averaged. During wk 4, a digestibility experiment was done. Feed and orts were sampled daily from d 21-24 and 22-25, respectively, and composited by period for analysis. Chromic oxide (5 g/dose in a gelatin capsule) was dosed into the rumen at each feeding during d 15 through 25 of each period and used as the digestibility marker. Fecal grab samples were taken twice daily on d 22 to 25 of each period such that a sample was obtained for every 90-min interval of a 12-h time period (8 samples) between a.m. and p.m. feedings. Samples were composited by animal for each period. Rumen samples
were taken with a core sampler (Firkins et al., 1986) at 3, 6, 9, and 12 h after the a.m. feeding on d 22 and 23 of each period. After stirring and mixing, 40 ml from each sample were obtained, and the remaining 460 ml of sample were returned to the rumen. Ruminal pH was measured immediately. Then, the sample was squeezed through 4 layers of cheese cloth, acidified with 6 N HCl to terminate bacterial activity, and was frozen. After thawing, samples were centrifuged at 27,000 x g prior to determination of NH$_3$-N (Chaney and Marbach, 1962) and VFA concentrations by GLC (Firkins et al., 1990).

Feed offered, orts and fecal samples were analyzed for DM, N and organic matter (OM; AOAC, 1984), NDF (Van Soest, 1990) and total fatty acids (Sukhija and Palmquist, 1988). Feed samples also were analyzed for NSC using the difference technique (Nocek and Russell, 1988) or enzymatically using modifications described by Casper et al. (1990), except that reducing sugars were quantified using the method described by Miller (1960). Feed samples also were analyzed for ADF and acid detergent lignin (ADL; Goering and Van Soest, 1970) for estimation of NE$_j$ (Conrad et al., 1984). Fecal samples were analyzed for Cr (Williams et al., 1962), and digestibility calculations were made using Cr as done by Sarwar et al. (1991).

Data were analyzed using ANOVA obtained from SAS (1988). Linear and quadratic contrasts due to % NDF from forage (control, NDF70 and NDF60 treatments only) and % NSC (control, NSC35, NSC25 treatments only) were made using the CONTRAST statement in GLM. Ruminal data were analyzed as a split plot, with sampling time as the split plot. When treatment x sampling time interactions were not significant (P > .05), data were averaged among times and analyzed in the main plot. When these interactions were significant (P < .05), data were analyzed for each sampling time. Contrasts were done as described earlier. Least square means were generated for VFA due to 2 missing samples.
RESULTS AND DISCUSSION

Chemical composition of experimental diets is shown in Table 7. The dietary CP concentration was slightly higher in NDF70 and NSC35 diets, probably due to variation in hay quality among different bales or slight differences in sample homogeneity. Percentages of NSC were lower and higher using the difference and enzymatic assays, respectively, than NSC values used to balance diets; the latter were taken by difference from NRC values for feed ingredients. Because the ferricyanide (Stokes et al., 1991) procedure gave unreliable and unreproducible results, the DNS (Miller et al., 1960) method was used to quantitate reducing sugars. This method gave comparatively higher values than those obtained from the difference method. The DNS can react with eligosaccharides (Miller et al., 1960), perhaps resulting in higher concentrations of reducing sugars than the ferricyanide procedure. However, results show good agreement in ranges among treatments. Higher concentrations of ADF and NDF of NSC35 and NSC25 diets were expected and were due to added SH which contained about 70% NDF. Diets were formulated to be isocaloric based on NRC (1989) energy values for feeds, but NE\textsubscript{j} concentration, calculated from chemical analysis (Conrad et al., 1984), decreased with decreasing % NSC. This was due, in part, to the use of higher NE\textsubscript{j} (1.77 Mcal/kg; NRC, 1989) of SH than that (1.55 Mcal/kg) predicted by the equation of Conrad et al. (1984). The NRC may take into account that current practice of replacing SH for corn may decrease negative associative effects and increase NE\textsubscript{j} of SH (Nakamura and Owen, 1989). Also, the fatty acid concentrations of NSC35 and NSC25 were 2.7 and 3.9%, but expected values were 3.15 and 4.5%, respectively. Explanations for this discrepancy are not apparent. Finally, there was 0.2% ADIN (DM basis) in soybeans. If multiplied by 6.25, this artifact would increase ADL by over one percentage unit for NSC25, decreasing NE\textsubscript{j} by the Conrad et al. (1984) equation. No literature is available on the validity of correcting ADL for acid-detergent insoluble CP; thus, no correction was made. The
TABLE 7. Chemical composition of experimental diets varying in concentrations of NDF and non-structural carbohydrates (NSC) from forage NDF.

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>C</th>
<th>NDF70</th>
<th>NDF60</th>
<th>NSC35</th>
<th>NSC25</th>
</tr>
</thead>
<tbody>
<tr>
<td>CP</td>
<td>17.4</td>
<td>18.2</td>
<td>17.5</td>
<td>18.2</td>
<td>17.8</td>
</tr>
<tr>
<td>NSC&lt;sup&gt;2&lt;/sup&gt;</td>
<td>41.8</td>
<td>40.7</td>
<td>42.7</td>
<td>31.9</td>
<td>20.6</td>
</tr>
<tr>
<td>NSC&lt;sup&gt;3&lt;/sup&gt;</td>
<td>50.2</td>
<td>48.5</td>
<td>49.9</td>
<td>37.3</td>
<td>26.9</td>
</tr>
<tr>
<td>ASH</td>
<td>6.8</td>
<td>7.3</td>
<td>6.9</td>
<td>7.6</td>
<td>8.2</td>
</tr>
<tr>
<td>NDF</td>
<td>31.7</td>
<td>31.6</td>
<td>30.6</td>
<td>39.6</td>
<td>49.5</td>
</tr>
<tr>
<td>ADF</td>
<td>17.2</td>
<td>16.8</td>
<td>16.6</td>
<td>23.7</td>
<td>30.8</td>
</tr>
<tr>
<td>ADL&lt;sup&gt;4&lt;/sup&gt;</td>
<td>2.9</td>
<td>2.7</td>
<td>2.5</td>
<td>3.6</td>
<td>4.4</td>
</tr>
<tr>
<td>Fatty acids</td>
<td>2.3</td>
<td>2.2</td>
<td>2.3</td>
<td>2.7</td>
<td>3.9</td>
</tr>
<tr>
<td>NE&lt;sup&gt;5&lt;/sup&gt;</td>
<td>1.68</td>
<td>1.68</td>
<td>1.70</td>
<td>1.60</td>
<td>1.56</td>
</tr>
<tr>
<td>Ca</td>
<td>1.2</td>
<td>1.2</td>
<td>1.2</td>
<td>1.3</td>
<td>1.5</td>
</tr>
<tr>
<td>P</td>
<td>.38</td>
<td>.36</td>
<td>.35</td>
<td>.37</td>
<td>.34</td>
</tr>
<tr>
<td>Mg</td>
<td>.29</td>
<td>.35</td>
<td>.28</td>
<td>.46</td>
<td>.35</td>
</tr>
<tr>
<td>K</td>
<td>1.3</td>
<td>1.3</td>
<td>1.2</td>
<td>1.3</td>
<td>1.5</td>
</tr>
</tbody>
</table>

1. C = control diet, NDF70 = 70% NDF from forage, NDF60 = 60% NDF from forage, NSC35 = 35% non-structural carbohydrates, NSC25 = 25% non-structural carbohydrates.
2. NSC calculated by difference (Nocek and Russell, 1988).
3. Enzymatic determination of NSC (Casper et al., 1990).
4. ADL = acid detergent lignin.
5. NE<sub>1</sub> (Mcal/kg DM) calculated from (Conrad et al., 1984).
mineral contents of all diets were found to meet or exceed requirements according to NRC (1989). Levels of Ca and Mg were increased above requirements due to possible insoluble soup formation with diets containing supplemental fat, reducing cation availability.

Ruminal pH (Figure 3) followed a typical post-feeding decline and increase after 6 h, the severity of which differed among treatments (hour x treatment interaction); thus, data were analyzed at each sampling time. At 6 h, post-feeding, pH tended (P < .10) to decrease quadratically (5.78, 5.85 and 5.63 for C, NDF70 and NDF60, respectively), and at 9 h, pH tended (P < .07) to decrease linearly (5.94, 5.93 and 5.71 for C, NDF70 and NDF60, respectively) in cows fed diets containing lower amounts of forage NDF (Fig 3). Quadratic responses (P < .08, .02) for increasing ruminal pH at 6 (5.78, 5.87 and 5.92 for C, NSC35 and NSC25, respectively) and 9 h (5.94, 5.99 and 5.99 for C, NSC35 and NSC25, respectively) also were noted in cows fed diets with reducing NSC concentrations. This indicates that concentrates containing relatively high NDF and low NSC are intermediate between forage and corn in their effects on ruminal pH.

Ruminal VFA concentrations and molar percentages were averaged among all time periods because no treatment x time interaction (P > .05) were detected (Table 8). Total VFA concentration increased linearly (P < .02) with decreasing percentage NDF from forage. These results are in contrast with earlier work (Sarwar et al., 1991). Present results are an indication of more extensive fermentation in the rumen or decreased ruminal fluid volume in cows fed decreasing amounts of forage fiber. Molar percentage acetate decreased linearly (P < .07), and propionate increased linearly (P < .01), due to feeding diets decreasing in forage NDF. These effects resulted in a linear reduction (P < .01) in acetate:propionate ratio. Decreased acetate:propionate ratios commonly are observed during feeding of high grain, low forage diets (Erwin et al., 1961; Grummer et al., 1987) and are related to trends for lower pH (Figure 3) due to decreasing forage
Figure 3. Ruminal pH as affected by diets varying in proportion of NDF from forage NDF and non-structural carbohydrates (NSC). C = control, NDF70 = 70% NDF from forage, NDF60 = 60% NDF from forage, NSC35 = 35% NSC and NSC25 = 25% NSC.
TABLE 8. Ruminal characteristics of cows fed diets varying in proportion of NDF from forage NDF and non-structural carbohydrates (NSC).

<table>
<thead>
<tr>
<th>Item</th>
<th>Diets 1</th>
<th>Probabilities of contrasts 2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>NDF70</td>
</tr>
<tr>
<td>Total VFA, mM</td>
<td>113</td>
<td>113</td>
</tr>
<tr>
<td>Acetate, mol/100 mol.</td>
<td>57.0</td>
<td>57.1</td>
</tr>
<tr>
<td>Propionate, mol/100 mol.</td>
<td>23.5</td>
<td>23.8</td>
</tr>
<tr>
<td>Butyrate, mol/100 mol.</td>
<td>15.0</td>
<td>14.6</td>
</tr>
<tr>
<td>BCVFA 4, mol/100 mol.</td>
<td>4.46</td>
<td>4.35</td>
</tr>
<tr>
<td>A:P ratio 5</td>
<td>2.44</td>
<td>2.44</td>
</tr>
</tbody>
</table>

1  C = control diet, NDF70 = 70% NDF from forage, NDF60 = 60% NDF from forage, NSC35 = 35% non-structural carbohydrates, NSC25 = 25% non-structural carbohydrates.

2  L = linear; Q = quadratic.

3  Non-significant (P > .10).

4  Branched chain volatile fatty acids.

5  Acetate:propionate ratio.
NDF. A positive relationship between pH and acetate:propionate has been reported by Erdman (1988). In contrast, quadratic response occurred with molar percentage acetate (P < .01), and propionate (P < .01), in the rumen of cows fed diets decreasing in NSC concentration. This effect resulted in increased (P < .01) acetate:propionate ratio, which probably also was due to higher ruminal pH (Figure 3). The molar percentage of butyric acid decreased linearly (P < .02) due to both NDF and NSC treatments. The same effect was noted for branched chain VFA concentrations in the rumen. Because actual differences were relatively small, the biological significance of these effects is uncertain.

Ruminal NH₃-N remained unaltered among all treatments across all time periods except at 3 and 9 h post-feeding (hour x treatment interaction; Figure 4). At 3 h post-feeding, NH₃-N concentration changed quadratically (P < .10) but at 9 h, NH₃-N concentration decreased linearly (P < .05) with decreasing dietary NSC. All concentrations were above 6.9 mg/dl so it is unlikely that NH₃-N limited microbial growth or protein synthesis significantly (Hoover, 1986).

Apparent digestibility of organic matter (OM) increased quadratically (P < .01) when cows were fed diets decreasing in forage NDF (Table 9). There was no difference in OM digestibility by cows fed diets varying in NSC concentration. These data are consistent with Sarwar et al. (1991) who reported increased (P < .06) OM digestion by heifers fed diets containing SH replacing forage. Similar results were reported by Hsu et al. (1987). When corn was replaced by SH, DM digestion increased in one study (Edionwe and Owen, 1989) but was unaffected in others (Highfill et al., 1987; Nakamura and Owen, 1989).

Decreasing % NDF from forage had no effect on NDF digestibility. In an earlier report, when NDF from forage was 65 or 45% due to increased substitution of SH for forage, 96-h extent of SH NDF digestion in situ was 89 and 66%, respectively (Sarwar et al., 1991). Moreover, ruminal NDF digestibility was decreased, but total tract NDF
Figure 4. Ruminal ammonia-N concentration as affected by diets varying in proportion of NDF from forage and non-structural carbohydrates (NSC). C = control, NDF70 = 70% NDF from forage, NDF60 = 60% NDF from forage, NSC35 = 35% NSC and NSC25 = 25% NSC.
TABLE 9. Apparent total tract nutrient digestibilities (%) by cows fed diets varying in proportion of NDF from forage and non-structural carbohydrates (NSC).

<table>
<thead>
<tr>
<th>Item</th>
<th>Diets</th>
<th>Probabilities of contrasts²</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>NDF70</td>
</tr>
<tr>
<td>Organic matter</td>
<td>65.2</td>
<td>70.1</td>
</tr>
<tr>
<td>NDF</td>
<td>47.6</td>
<td>53.0</td>
</tr>
<tr>
<td>Fatty acids</td>
<td>64.5</td>
<td>63.4</td>
</tr>
<tr>
<td>N</td>
<td>64.2</td>
<td>68.4</td>
</tr>
</tbody>
</table>

1. C = control diet, NDF70 = 70% NDF from forage, NDF60 = 60% NDF from forage, NSC35 = 35% non-structural carbohydrates, NSC25 = 25% non-structural carbohydrates.

2. L = linear; Q = quadratic.

3. Non-significant (P > .10).
digestibility was increased with greater substitution of SH for forage. In the current study, however, ruminal pH values (Figure 3) were lower in cows fed NDF60 than in the previous study, probably decreasing extent of NDF digestibility in the rumen in the current study more than in the earlier one. Greater intake and hindgut passage rate may have limited compensatory digestion of NDF in the hindgut by these lactating cows compared with results from the earlier study that used heifers. It is likely that adding a ruminal buffer would have improved NDF digestion when SH replaced forage especially at the higher substitution, as occurred when corn gluten feed plus sodium bicarbonate replaced corn silage (Firkins et al., 1991).

Digestibility of NDF increased linearly (P < .04) in cows fed diets decreasing in NSC concentration (Table 9). Predicted digestibilities of NDF (Conrad et al., 1984) were 59.8, 60.5, and 59.9% for C, NSC35, and NSC25, respectively. Thus, negative associative effects caused estimated reductions in NDF digestibilities of 12.2, 4.2 and 0.2 percentage units, respectively. Higher ruminal pH (Figure 3) for these treatments may have decreased inhibitory effects of low pH on fiber digestion (Hoover, 1986). Other workers (Edionwe and Owen, 1989; Macgregor et al., 1976; Nakamura and Owen, 1989) also reported increased digestibility of fiber by cows fed fibrous compared with starchy concentrates. A reduction in digestibility of NDF or cellulose has been noted in many studies (England and Gill, 1985; Huhtanen, 1987; Kaiser et al., 1983) following the supplementation of forages with readily fermentable carbohydrates. Based on in vitro measurements, Mertens and Loften (1980) and Stewart (1977) suggested that primary mechanisms responsible for reduced fiber digestion associated with rapid starch fermentation is the reduction in cellulolytic activity. More specific explanations for the decreased fiber digestion have been attributed to increases in lag time for fiber digestion in vivo (Huhtanen, 1987; McDonald, 1981) and in vitro (Mertens and Loften, 1980). The increased lag time may be due to rumen microbes preferentially utilizing soluble car-
bohydrates before starting degradation of fiber (Mertens, 1977) or to decreased bacterial attachment to fiber due to reduced pH (Hoover, 1986). However, other workers (Miron et al., 1990; Piwonka and Firkins, 1991) have reported a reduction in rate of fiber digestion when NSC were added. El-Shazly et al. (1961) proposed that competition among ruminal microbes for essential nutrients increased when readily fermentable carbohydrates were given. Cellulolytic microorganisms may be less competitive and unable to metabolize and reproduce at a rate fast enough to maintain themselves in the rumen, and proportion of total bacteria as cellulolytics decreased. Highfill et al. (1987) reported that high-fiber energy supplements appeared to reduce negative associative effects by avoiding potential shifts in the ruminal microflora.

Fatty acid digestibilities by cows fed NSC35 and NSC25 diets were higher than that of C diets, resulting in a linear effect (P < .10). The higher digestibilities of these diets can be attributed to the added fat of soybeans and calcium soups diluting endogenous sources of fat that are more poorly digestible. Palmquist (1991) noted that fatty acids of many sources are about 77% digestible. The apparent digestibility of N remained unchanged among all treatments and is consistent with results of other workers (Bernard and McNeill, 1991; Bernard et al., 1988; Macgregor et al., 1976).

Dry matter intake by all cows remained unaltered among all treatments (Table 10), probably because differences in diet composition were not great enough to elicit responses. Varga et al. (1984) reported no difference in intake by cows fed diets with faster or slower estimated rate of NDF disappearance.

A tendency (P < .07) for a linear increase in milk yield by cows fed diets with decreasing NSC concentration was noted. Thomas et al. (1986) also reported increased milk yield by cows fed diets containing fibrous concentrate plus fat compared with cows fed starchy concentrates. In other studies in which SH replaced concentrate, no differences in milk yield occurred (Bernard and McNeill, 1991; Macgregor et al., 1976;
Nakamura and Owen, 1989).

Milk protein percentage was significantly lower (P < .01) in cows fed diets decreasing in NSC and increasing in fat (Table 10). These results supported the findings of Garnsworthy (1989), Garnsworthy (1990) and Thomas and Martin (1988). In contrast to findings of the present study, no change in milk protein percentage was noted when cows were fed diets containing various NSC concentrations (Macgregor et al., 1976) or when beet pulp (low in NSC) replaced corn (Valk et al., 1990). All diets were formulated to have similar degradable protein concentrations (60%). Thus, it is likely that the reduced milk protein content may be partially due to added fat, which often depresses milk protein concentration without altering protein yield (Palmquist and Jenkins, 1980). However, Cragle et al. (1986) decreased NSC concentration without adding fat, and cows had decreased percent milk protein. Emery (1978) reported a .015-unit increase in milk protein percentage for each additional megacalorie of net energy intake from concentrates or roughages. Thus, it can be speculated that reduced milk protein percentage by cows fed NSC25 diets may be due to reduced NE\textsubscript{L} intake and presence of added fat. Milk protein yield remained unaltered among all treatments (Table 10) and was in agreement with results of Mayne and Gordon (1984), Valk et al. (1990), Thomas et al. (1986), Hemingway et al. (1986), and Macgregor et al. (1976). Contrary to findings of this study, Nakamura and Owen (1989) and Garnsworthy (1989) reported reduced milk protein yield by cows fed fibrous concentrates replacing starchy ones. The reduced milk protein yield was due to lower milk yield.

No differences in milk fat percentages among treatments were noted (Table 10). Based on ruminal pH and acetate:propionate ratios (Table 8), cows fed NDF60 were projected to produce milk with a lower fat percentage than C-fed cows. Although traditionally it is accepted that an acetate:propionate ratio of 2.5 is needed to maintain milk fat concentration (Woodford and Murphy, 1987), others have lowered this threshold to 2.2
TABLE 10. Dry matter intake and milk production by cows fed diets varying in proportions of NDF from forage and non-structural carbohydrates (NSC).

<table>
<thead>
<tr>
<th>Item</th>
<th>C</th>
<th>NDF70</th>
<th>NDF60</th>
<th>NSC35</th>
<th>NSC25</th>
<th>SE</th>
<th>L</th>
<th>Q</th>
<th>L</th>
<th>Q</th>
</tr>
</thead>
<tbody>
<tr>
<td>DMI, kg/d</td>
<td>19.4</td>
<td>19.8</td>
<td>19.4</td>
<td>19.9</td>
<td>20.4</td>
<td>.8</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>DMI, % of BW</td>
<td>3.6</td>
<td>3.5</td>
<td>3.6</td>
<td>3.6</td>
<td>3.7</td>
<td>.1</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Milk, kg/d</td>
<td>24.7</td>
<td>24.2</td>
<td>25.2</td>
<td>25.9</td>
<td>27.7</td>
<td>.7</td>
<td>NS</td>
<td>NS</td>
<td>.07</td>
<td>NS</td>
</tr>
<tr>
<td>Milk protein, %</td>
<td>3.12</td>
<td>3.07</td>
<td>3.24</td>
<td>3.08</td>
<td>2.84</td>
<td>.04</td>
<td>NS</td>
<td>NS</td>
<td>.01</td>
<td>.01</td>
</tr>
<tr>
<td>Milk protein, kg/d</td>
<td>.77</td>
<td>.74</td>
<td>.81</td>
<td>.79</td>
<td>.79</td>
<td>.01</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Milk fat, %</td>
<td>3.46</td>
<td>3.57</td>
<td>3.61</td>
<td>3.54</td>
<td>3.51</td>
<td>.09</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Milk fat, kg/d</td>
<td>.85</td>
<td>.85</td>
<td>.96</td>
<td>.91</td>
<td>.97</td>
<td>.02</td>
<td>NS</td>
<td>NS</td>
<td>.04</td>
<td>NS</td>
</tr>
<tr>
<td>4% FCM, kg/d</td>
<td>22.7</td>
<td>22.5</td>
<td>23.7</td>
<td>24.0</td>
<td>25.7</td>
<td>.7</td>
<td>NS</td>
<td>NS</td>
<td>.04</td>
<td>NS</td>
</tr>
<tr>
<td>FCM/DMI</td>
<td>1.14</td>
<td>1.16</td>
<td>1.23</td>
<td>1.21</td>
<td>1.26</td>
<td>.06</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>FCM/NE\textsubscript{j} intake</td>
<td>.68</td>
<td>.69</td>
<td>.72</td>
<td>.75</td>
<td>.81</td>
<td>.05</td>
<td>NS</td>
<td>NS</td>
<td>.04</td>
<td>NS</td>
</tr>
</tbody>
</table>

1 C = control diet, NDF70 = 70% NDF from forage, NDF60 = 60% NDF from forage, NSC35 = 35% non-structural carbohydrates, NSC25 = 25% non-structural carbohydrates.

2 L = linear; Q = quadratic.

3 Non-significant (P > .10).

4 NE\textsubscript{j} calculated from Conrad et al. (1984).
Acetate:propionate ratios averaged above 2.26 in the present study. One cow fed NDF60 was in negative energy balance during one period, and adipose lipolysis could be one cause of high milk fat percentage by this cow (Palmquist and Conrad, 1971). Results of this trial supported the findings of Bernard and McNeil (1991), Macgregor et al. (1976), and Valk et al. (1990) who reported no change in milk fat percentage when SH or SH plus beetpulp replaced corn in diets fed to lactating dairy cows. In contrast to results of this study, Nakamura and Owen (1989), Sutton et al. (1987), Thomas et al. (1986) and Visser et al. (1991) reported increased milk fat percentage (P < .05) by cows fed diets in which corn or barley was replaced by SH or beet pulp. However, in this study, milk fat yield increased linearly (P < .04) in cows fed diets with reducing NSC concentration. Nakamura and Owen (1989), Thomas et al. (1986) and Visser et al. (1991) also reported increased milk fat yield by cows fed diets containing SH or beetpulp when compared to those fed diets containing corn or barley.

Feed efficiency (FCM/NE\textsubscript{1} intake) increased linearly (P < .04) in cows fed diets decreasing in NSC concentration (Table 10). The increased feed efficiency indicates improved utilization of NE\textsubscript{1} by cows fed diets in which a greater proportion of corn was replaced by SH while forage remained constant. These results are in contrast to results of other scientists who fed SH (Nakamura and Owen, 1989) or other fibrous concentrates (Thomas et al., 1986; Sutton et al., 1987). DM intake (and probably tissue loss) was similar among treatments. Thus, the difference is due partly to estimation of NE\textsubscript{1} of SH by technique (see earlier discussion). However, a tendency (P = .14) for a linear increase in gross efficiency (FCM/DMI) appears to substantiate our results of this study.

In conclusion, performance by cows was not affected under different feeding formulations used in this study, with the exception that decreasing NSC increased fat yield, probably due to increased pH and acetate:propionate ratio and reduced negative
associative effects. Soyhulls can replace forage such that a minimum of 60-65% of NDF as forage should have minor effects on milk production. Therefore, SH can provide a suitable alternative when forage availability is scarce or expensive to feed. It is acknowledged that more potentially digestible fiber is wasted when SH replaces forage. More information is needed before diets should be balanced for NDF, NDF from forage and NSC percentages without potentially discrediting the versatility of SH and other high-energy, fibrous by-products.

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CHAPTER V

GENERAL DISCUSSION

Most by-product feeds have a relatively high NDF percentage which is rapidly digestible, increasing their energy concentration relative to forages with similar NDF percentages. Therefore, the objectives of two independent trials (Chapter 3 and 4) were to determine the quantity of forage NDF and concentrate that could be replaced by CGF or SH in dairy diets. The hypothesis was that SH and CGF can replace a portion of forage fiber and corn in dairy diets without adversely affecting rumen function and lactation performance.

Most researchers who evaluated the use of NDF percentage in dairy diet formulation varied the amount and type of forage. They provided at least 75% of the NDF from forage to examine the influence of NDF on performance. That is why the NRC (1989) recommended that a minimum of 75% NDF should come from forage, but the scientific evidence to support this recommendation is limited. However, data from Chapters 3 and 4 are interpreted to indicate that SH or CGF can effectively replace about 15 percentage units of forage NDF in dairy diets without having any adverse effect on the lactation performance of cows.

Total tract digestibility of NDF was higher in heifers fed diets lower in forage NDF (CGF45 and SH45) in Chapter 3. Because all diets had similar concentrations of NSC (44.38, 43.25, 44.51, 42.68 and 43.67% for C85, CGF45, CGF65, SH45 and SH65, respectively), greater NDF digestibility can be mostly attributed to the source of NDF (SH or CGF vs forage) in diets. Some researchers have reported that the digestion of feed
by-product fiber is faster than that of most forages. However, data from Chapter 3 indicated that greater extent of digestion of SH fiber was the reason for higher NDF digestibility. Difference in NDF digestibility in cows (Chapter 4) fed diets lower in forage NDF relative to control was reduced and may be due to a greater intake (above 3.5% BW), which generally exacerbates negative associated effects.

Milk yield, milk composition and feed efficiency remained unchanged in cows fed diets lower in forage NDF. Replacing CGF or SH for forage tended to increase OM digestibility, as expected, because they have greater extent of NDF digestion and greater NE\textsuperscript{1} concentration than forages. However, SH or CGF supplementation probably provided some effective fiber helping to maintain a favorable ruminal environment to allow greater extent of OM digestion in by-products vs forages. In both trials, the ruminal acetate:propionate ratio decreased to 2.2 - 2.5 at about 6 h after meals when forage NDF decreased. Lack of decrease in milk fat percentage is consistent with others who showed that acetate:propionate ratio needs to drop below 2.2 for milk fat depression. Data from this research support the hypothesis that when SH (and probably CGF) is fed, forage NDF needs to be only 60% of total NDF in dairy diets (about 18% total forage NDF) to maintain lactation performance. Data from the heifer trial suggested that 45% NDF from forage may be adequate; however, this remains to be tested in lactating cows.

Previous research has indicated that optimum dietary NDF should be 28% for high producing lactating dairy cows (NRC, 1989). Diets with higher NDF percentages may be deficient in energy. However, increasing dietary NDF to about 50% resulted in an improved lactation performance (Chapter 4). The SH contributed about 49% of total dietary NDF in this diet, indicating that NRC recommendation of a minimum 75% forage NDF in dairy diets can only be applicable when dietary NDF is near the minimum recommended percentage.

When balanced for NDF, forage needed in dairy diets decreases when poor
quality forage with high NDF% is used, thus causing a rise in concentrate requirements. In such conditions, fibrous by-products should be more useful because they provide some effective fiber, increase energy density and dilute starch. Replacing good quality forage with by-products should be limited unless the forage is scarce or expensive to feed. Drought conditions usually reduce traditional feed supplies, and in such conditions, these feed by-products can act as an alternative for forages. Based on data of this research and those from literature cited in this thesis, diets should have a minimum of about 30% of DM from forage and 60% of total dietary NDF from forage. Requiring forage NDF to be 75% of total NDF (NRC, 1989) unfairly discredits the potential of many by-products to provide effective fiber.

Just as there is a minimum fiber level which is necessary for normal ruminal function, there is a maximum amount of dietary NSC which can be added to increase energy density. When this level is exceeded, ruminal dysfunction occurs, milk fat may be severely decreased and cows may go off feed. However, there also is a minimum level of NSC needed in the ration. If levels are too low, microbial protein synthesis will be limited and milk yield and milk protein percentage will decrease. Therefore, an optimum level of NSC in the diet is needed to attain maximum milk production, but in vivo data defining an optimum level of NSC are limited. Thus an experiment was conducted to evaluate the potential of SH to replace NSC in dairy diets (Chapter 4).

The ruminal acetate:propionate ratio was significantly higher in cows fed diets lower in NSC, perhaps due to a more favorable ruminal pH. Fatty acid digestibility was higher in cows fed these diets, probably due to highly digestible added fat of soybean and megalac. All cows maintained good body condition during the trial.

Higher milk yield was noted from cows fed diets lower in percent NSC, suggesting that more appropriate nutrient ratios were made available for milk synthesis when a greater proportion of corn was replaced by SH with constant dietary forage.
Reduced milk protein percentage in cows fed these diets was probably due to lower NSC intake and adding dietary fat. However, milk protein yield remained unaltered. The improved feed efficiency of cows fed diets low in NSC suggests that NE\textsubscript{I} was utilized more efficiently by cows fed diets in which a large proportion of corn was replaced by SH. Data of this study support the hypothesis that replacement of corn by SH in dairy diets does not adversely affect lactation performance. By-products should be considered as a replacement for concentrate (corn and protein source) based on their relative costs; a discount for corn should be applied because it increases negative associative effects. In contrast to the general recommendations made by many field nutritionists that dairy diets should have 35 - 40\% NSC, decreasing NSC to 25\% tended to increase milk yield and feed efficiency linearly. Thus, perhaps 25 - 35\% NSC is optimal for greatest efficiency of milk production.

Digestibility and ruminal data from short term trials are considered reliable, but lactation performance should be tested in continuous lactation trial(s) for long enough time to determine actual effects of decreasing forage NDF or total dietary NSC on yield of milk fat and protein, body weight loss, and feed efficiency. Therefore, more information is needed before diets should be balanced for NDF, NDF from forage and NSC percentages without potentially discrediting the versatility of high energy, fibrous by-products.


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