Forages for Cattle
New Methods for Determining Energy Content
and Evaluating Heat Damage

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Improved System of Forage Analysis

The crude fiber method of feed analysis has been used for more than 100 years. Although this method was an important first attempt at determining the energy content of feeds, it has a number of shortcomings.

- The crude fiber method assumes that crude fiber is the same for all forages. This is not true. The crude fibers of alfalfa, orchard grass and cottonseed hulls have different digestibilities and therefore cannot be considered the same for calculating feed energy.
- The crude fiber value for the same feed may be quite different from laboratory to laboratory because of the varying conditions under which chemists measure crude fiber. For example, the strength of acid and base used and the length of time feed is boiled in acid and base can affect crude fiber value.
- Crude fiber increases as forages mature, but this increase often does not accurately reflect the simultaneous decrease in energy content. Using the crude fiber method, the energy content of good quality forages is often underestimated and overestimated in poor quality forages.
- The crude fiber method often does not differentiate the highly digestible parts of the plant from the less digestible parts.

A new analytical approach for estimating energy content of forages was developed by Van Soest in the 1960s at the U.S.D.A. Beltsville Nutritional Research Facility. These detergent fiber analyses give more accurate estimates of forage energy values and now are used for forage analysis.

Detergent Fiber Analysis

The detergent fiber analytical method separates a forage into two parts: (1) the cell solubles, which include starches, proteins, sugars and other compounds which are highly digestible, and (2) detergent fiber, which provides structural support for the plant and is lower in digestibility.

Different types of detergent fiber are also determined. Figure 1 is a schematic diagram of the plant cell and illustrates the cell solubles, cell wall and types of fiber found in the cell wall.

Neutral detergent fiber (NDF), also called cell wall, is measured by boiling a sample of forage in a special detergent (soap) under a neutral \( \text{pH} = 7 \) condition and filtering the boiled sample through filter paper. The liquid that passes through the filter paper contains starch, sugar, protein and other compounds that were dissolved.

The part of the feed sample that does not dissolve remains on the filter paper; this residue is called NDF or cell wall.
After drying, the NDF is calculated as a percentage of the original forage sample. Figure 2 shows how NDF is determined.

NDF contains all the fiber found in the forage and consists of the following fiber components—hemicellulose, cellulose and lignin. NDF is partially digestible, ranging from 20-80 percent, depending upon forage species and stage of maturity.

NDF also maintains the original bulkiness of the feed before it was boiled in detergent. NDF is responsible for rumen fill, and we are developing equations to predict forage intake, based on forage NDF%.

**Acid detergent fiber (ADF)** is determined in much the same way, except a different detergent is used under acid (pH = 2) conditions. The sample is boiled and filtered like in the NDF procedure. Because of a different detergent and acid conditions, hemicellulose and cell solubles dissolve and are filtered away. The residue left is ADF and consists mainly of cellulose and lignin. ADF is related to dry matter digestibility and is used to predict net energy content.

**Acid detergent lignin (ADL)** is measured by further treating ADF with strong acid, which dissolves cellulose, or with permanganate (salt of permanganic acid), which oxidizes (removes) the lignin. Either approach allows calculation of amount of lignin.

**Digestibility.** ADF is partially digestible, ranging from 20-80 percent, while ADL is low in digestibility, from 0-30 percent. The ADF fraction is closely related to digestibility of the forage sample because it contains cellulose and lignin.

Cellulose is the major fiber fraction to be digested; there is less of lignin. Lignin, however, ties up cellulose—the higher the concentration of lignin, the greater the amount of cellulose tied up and made indigestible.

Two forages may have similar ADF content, 25 percent for example. Forage 1 may be 20 percent cellulose and 5 percent lignin; forage 2 may be 15 percent cellulose and 10 percent lignin. Forage 1 would be much more digestible.

Therefore, information about the amount of lignin and cellulose as well as ADF content of a forage is important in predicting energy content.

**Dry matter digestibility.** The last step in forage analysis is measuring the digestibility using laboratory techniques. The test for dry matter digestibility (DMD) simulates digestibility in the cow, but the laboratory test is much less expensive and less time consuming.

A sample of forage, some rumen fluid and certain chemicals are put into a flask and allowed to digest for a standard period, usually 48 hours. Then NDF determination is made on the contents of the flask. The residue left on the filter paper is undigested fiber, mostly lignin and cellulose.

This measures how much forage was not digested in the 48-hour period. Little additional fiber digestion would occur past that period. DMD, or the amount of digestible material, is 100 minus the NDF residue (undigested fiber). This is an improved method of estimating total digestible nutrients (TDN). TDN is estimated as DMD minus 10 percent. Table 1 summarizes the parts of the forage plant, how the detergent fiber analysis segregates these parts and the digestibility of these parts.

**Fiber content.** The amount of fiber in a forage depends upon: (1) the species and (2) the stage of maturity. Table 2

### Table 1. Summary of Detergent Fiber Fractions.

<table>
<thead>
<tr>
<th>Plant part</th>
<th>Contains</th>
<th>Detergent Fiber Fraction</th>
<th>How Determined</th>
<th>Digestibility</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Cell solubles</td>
<td>protein, sugar,</td>
<td>Cell solubles = 100 - NDF</td>
<td>Released by neutral</td>
<td>90 - 100%</td>
</tr>
<tr>
<td></td>
<td>starches, fats,</td>
<td></td>
<td>detergent extraction</td>
<td></td>
</tr>
<tr>
<td></td>
<td>pectins</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. Cell wall</td>
<td>NDF, ADF</td>
<td>Filtration, leaves residue (NDF or ADF)</td>
<td>20 - 80%</td>
<td></td>
</tr>
<tr>
<td>a. Primary wall</td>
<td>Cellulose</td>
<td>ADF - ADL</td>
<td>Filtration</td>
<td>50 - 90%</td>
</tr>
<tr>
<td></td>
<td>Some lignin</td>
<td>ADL</td>
<td>Dissolve or oxidize</td>
<td>0 - 30%</td>
</tr>
<tr>
<td></td>
<td>Heat-damaged protein</td>
<td>Kjeldahl determination</td>
<td></td>
<td></td>
</tr>
<tr>
<td>b. Secondary wall</td>
<td>Hemicellulose</td>
<td>NDF - ADL</td>
<td>Difference</td>
<td>20 - 80%</td>
</tr>
<tr>
<td></td>
<td>Most lignin</td>
<td>ADL</td>
<td>Dissolve or oxidize</td>
<td>0 - 30%</td>
</tr>
</tbody>
</table>
lists usual fiber values for some Missouri forages. From the values in Table 2, several important facts are evident:

- Grasses have higher fiber and lower energy than alfalfa cut at similar stages.
- Grasses and alfalfa increase in fiber content from early to late stages; the increase is greater for grasses than alfalfa.
- Corn for silage does not show an increase in fiber nor a decrease in energy from early to late stages. Because the corn plant is producing a large amount of starch, the concurrent increase in fiber is not evident.

### Net Energy Terminology

The term net energy (NE) is sometimes misunderstood and needs to be clearly defined. In this Guide, NE is used in the same context as in National Research Council (NRC) publications and in UMC Guide 3104 "Calculating Rations for Dairy Cattle."

For example, a lactating cow weighing 1,430 pounds and producing 65 pounds of 3.5 percent butterfat milk needs 10.9 Mcal or therms of NE for maintenance and 20.4 Mcal or therms of NE for production, which totals 31.3 Mcal or therms of NE. Because a certain weight of forage allows a cow to produce a given quantity of milk, forages are estimated to contain a certain amount of net energy. Separation of net energy into that used for maintenance and that used for production is not necessary because they are used with the same efficiency. Therefore, how much forage energy was used for maintenance and how much was used for production is of little concern in the lactating cow.

NE values used here are not calculated in the same way as the estimated net energy (ENE) values of Morrison’s *Feeds and Feeding*, although some values may be similar. Morrison’s tables of ENE underestimate energy content of high quality forages and overestimate the energy of low quality forages.

The terms Mcal, therm, therm/pound and therm/100 pounds need explanation. One Mcal and one therm are equal to 1000 kcal, the amount of heat needed to raise 400 pounds (50 gallons) of water 10°F. Therm/pound and therm/100 pounds (or cwt) refer to energy concentration in a feed.

For example, if a pound of forage were found to contain .5 therm, energy content would be expressed as .5 therm/pound or 50 therms per 100 pounds (50 therms/cwt). It would also be equivalent to .5 Mcal/pound or 50 Mcal/100 pounds.

### Determining Net Energy

**Net energy** can be measured directly only by expensive, laborious animal trials. It can be predicted using either NDF or ADF. Forages cut at different stages of maturity have different levels of fiber and energy. Older, more mature forages have higher fiber and less energy than younger succulent forages. NDF and ADF both increase as forages mature, while the DMD (or TDN) decreases. Research indicates the following relationship for net energy, NDF and DMD:

\[
\text{NE} = (\text{Mcal/lb or therms/lb}) = (0.01) \times (\text{TDN}) \times \left(2.86 - \frac{35.5}{\text{cell solubles}}\right) \\
\times 2.2 \text{ lbs/kg}
\]

- TDN = DMD - 10
- Cell solubles = 100 - % NDF

Both NDF and DMD (as TDN) are needed in the equation because as a plant matures, the increase in NDF is large, while the decrease in DMD is not so great. Using both NDF and DMD increases accuracy of the net energy value.

Some forages change in NDF and DMD more than others. If legumes, corn silage and sorghum silage increase 1 percent in NDF, DMD and TDN simultaneously decrease by 1 percent. Thus, as a legume or silage increases from 50 to 60 percent in NDF, DMD will decrease from 70 to 60 percent and TDN from 60 to 50 percent. Grasses decrease 2 percent in DMD (or TDN) for each 1 percent increase in NDF. As NDF goes up from 55 to 65 percent, DMD will decrease from 65 to 45 percent and TDN from 55 to 35 percent.

Two advantages of knowing about these relationships between NDF and DMD are: (1) They are more accurate than using crude fiber because both fiber and digestibility are measured, and (2) Stage of maturity is not necessary for estimation of energy. If only the NDF value is known, DMD...
Figure 3. The Kjeldahl procedure is used to determine protein content of forages.

can be estimated by comparing it to the same type forage which has known NDF and DMD values.

A lot of data exist for estimation of net energy from NDF. But NDF is not recognized as an official chemical method, and many commercial labs hesitate to use it until the method becomes official.

Less information is available relating ADF to forage net energy but ADF is used by most commercial feed analysis labs for estimation of net energy. Different equations are used depending upon type of forage.

1. Grasses: Net energy (Mcal/lb) = 1.50 - .0267 (% ADF).

2. Legumes: Net energy (Mcal/lb) = 1.044 - .0123 (% ADF).

3. Mixed legume and grasses: Net energy (Mcal/lb) = 1.044 - .0131 (% ADF).

4. Corn, small grain or sorghum silage: Net energy (Mcal/lb) =

\[
0.3133 \times \left( 2.86 - \frac{35.5}{100 - (1.67 \times \% ADF)} \right)
\]

5. Grains: See UMC Guidesheet 3105.

As with the NDF technique, knowing the cutting stage or date is not necessary for estimation of energy. For a particular type of feed, a given ADF content is related to a certain amount of energy. As ADF goes up or down, energy content changes in the opposite direction.

The problem with using cutting dates to estimate energy content is that this method does not take weather variations into account. The weather affects plant growth too much for cutting dates to be accurate.

Using cutting stage is more accurate than cutting date because generally a given cutting stage, such as 1/10th bloom or boot, is more closely related to chemical composition. However, heat or drought stressed forages can have elevated fiber levels compared to the same forage cut at the same stage and neither heat or nor drought stressed.

Dent stage corn silage grown in New York and Michigan usually has NDF of about 40-50 percent. Corn silage grown in Missouri usually has NDF of 50-55 percent; some silages have been found to contain 65-70 percent NDF. The higher NDF concentration decreases the net energy content of Missouri-grown corn silage and gives lower net energy values.

As a result, New York and Michigan corn silage has a TDN value of about 65-70 percent, whereas Missouri corn silage has a TDN value of about 55-60 percent. Corn silage, as well as nearly all other forages grown in hotter climates, generally has a higher fiber and lower energy content.

A lab test is the only accurate way to determine fiber and net energy content of any forage.

Measuring Protein Availability

Protein is an essential nutrient for the dairy cow. Protein availability (quality) can vary depending upon storage and harvesting methods; quality can affect milk production.

Protein content of forages usually is determined by the Kjeldahl (pronounced Kell-doll) method. In this procedure a feed sample is boiled in strong acid (H\textsubscript{2}SO\textsubscript{4}) which destroys organic matter and converts the nitrogen (N) of natural protein and N of non-protein nitrogen (NPN) compounds into ammonia, which is trapped as ammonium sulfate. Then, the ammonium sulfate is boiled in a strong base (NaOH) to release ammonia, which is trapped in a specific chemical that allows measurement of N.

Plant protein usually contains 16 percent N. Because the Kjeldahl method measures N, we calculate protein by multiplying nitrogen times 6.25 (= 100/16). We refer to this as crude protein.

Two problems of the Kjeldahl method are: (1) It cannot distinguish between the natural protein such as that of soybean meal and the NPN of compounds such as urea and ammonia, and (2) Protein that is unavailable in the cow is still measured as N by the Kjeldahl procedure.

The Kjeldahl procedure gives no indication whether protein is available or unavailable. Therefore, a procedure must be used to measure availability of protein.

Crude protein often is assumed to be completely digested by the dairy cow, but we know a certain amount of crude protein is completely unavailable. The unavailable part probably is 20-25 percent of the crude protein and is similar for most forages except those cut extremely early or extremely late. For most forages, we consider 3 percentage units of crude protein to be normally unavailable protein.

This should not be mistaken to mean that 3 percent of the crude protein is unavailable. For example, alfalfa and grass hay usually contain 15 percent and 10 percent crude protein; both have about 3 percentage units of unavailable protein which leaves 12 and 7 percentage units available protein, respectively. Unavailable protein apparently is bound to fiber and actually may not be true protein. However, for the sake of simplicity, it is calculated as N x 6.25 and is called unavailable protein.

Usually the unavailable protein content of a forage is of little concern, but in some conditions it can be a problem.
Table 3. Examples of Adjusting Crude Protein for Heat-Damaged Forages.*

<table>
<thead>
<tr>
<th>1. Normal clover hay with no heat damage</th>
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</tr>
</thead>
<tbody>
<tr>
<td>Crude protein</td>
<td>18.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unavailable protein</td>
<td>3.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heat-damaged protein</td>
<td>0 (3.0-3.0)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adjusted crude protein</td>
<td>18.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>2. Moderately heat-damaged haylage</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein</td>
<td>14.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unavailable protein</td>
<td>12.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heat-damaged protein</td>
<td>9.0% (12.0-3.0)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adjusted crude protein</td>
<td>5.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>3. Excessively heat-damaged clover hay</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein</td>
<td>19.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unavailable protein</td>
<td>15.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heat-damaged protein</td>
<td>12.0% (15.0-3.0)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adjusted crude protein</td>
<td>7.0%</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Although the latter two examples are extreme cases of heat-damaged protein, the examples are actual forages from three different farmers.

Unavailable protein can become significant in hays and haylages that become too hot during storage. This is usually more common in legumes than grasses.

Generally, the large amounts of unavailable protein are caused by excess moisture in hays and too little moisture and too much oxygen in haylages. The resulting forage turns brown to black depending on severity of overheating, and it has an odor that ranges from sweet to caramel-like to tobacco-like. Cows often relish overheated forage because the sugars become condensed and turn into syrup. We often refer to this condition as heat damage.

Farmers often assume that because overheated forages are eaten readily by cows, nutrient composition is unaffected by heat damage. Some actually think quality is improved. That is definitely not the case. Overheated forages, especially legumes, smell sweet and are dark brown to black in appearance and may contain much unavailable protein.

Apparently, when forages become overheated during the curing process, some true protein becomes tied up with carbohydrates, and less protein is available for use by the animal. Fortunately, the amount of protein made unavailable by overheating can be measured.

The ADF procedure removes available protein and leaves unavailable protein behind in the fiber residue. Determining the Kjeldahl protein content of the ADF residue (ADF-N) estimates unavailable protein. Another procedure is to digest the feed with weak acid and pepsin, an enzyme found in the small intestine of animals. Unavailable protein cannot be digested with acid-pepsin.

These two methods of determining unavailable protein, ADF-unavailable-protein (ADF-N) and pepsin-unavailable-protein, are similar and either can be used to measure heat-damaged protein. The extent of heat damage is indicated by the elevation of either measure of unavailable protein above the average baseline value of 3 percentage units of unavailable protein.

For example, if a clover hay had 12 percentage units of unavailable protein, the amount of heat-damaged protein would be 12 percentage units total unavailable protein minus 3 percentage units normal unavailable protein = 9 percentage units heat damage. This means 9 percentage units of protein are heat damaged and unavailable above the normal amount of unavailable protein (3 percentage units).

If the clover hay originally contained 19 percent crude protein and 9 of these 19 percentage units are heat-damaged, then the clover hay really contains 10 percent (19-9) adjusted crude protein (not heat-damaged).

In essence, instead of feeding a 19 percent crude protein hay, a farmer would be feeding a 10 percent crude protein hay. The most immediate effect of heat-damaged forage is reduced milk yield, that is, cows do not produce as much as they should. The only practical way to overcome this is to increase the protein content of the concentrate to make up for the amount of heat damage present.

Usually, if heat-damaged forage has been fed and the net crude protein was below requirements, milk yield will increase 2-10 pounds per day within a few days after correction.

Usually, if heat-damaged forage has been fed and the adjusted crude protein was below requirements, milk yield will increase 2-10 pounds per day within a few days after correction.

Table 4. Recommended Forage Analyses.

<table>
<thead>
<tr>
<th>Dry Matter</th>
<th>Energy</th>
<th>Protein</th>
<th>Calcium</th>
<th>Heat Damaged ADFN or Pepsin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grass Hay &amp; Haylage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Legume Hay &amp; Haylage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Legume Grass Mixtures Hay &amp; Haylage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Corn Silage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Corn Silage with NPN Added</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Sorghum Silage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Sudan Hay or Haylage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Any high dry matter haylage with brown color or known to have heated during storage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Any hay, baled wet, brown in color, or suspected to have heated during storage</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
</tbody>
</table>
A milk yield response may not occur in some cases, even though protein content of the concentrate is increased this can happen if the farmer is overfeeding protein — feeding clover hay (high in protein) and a concentrate high in protein such as 16 percent crude protein. Moderate heat damage may have reduced the protein content of the hay, but because the hay was high in protein, the hay and concentrate still provided sufficient protein to meet production needs and milk yield was not depressed. Table 3 gives three examples of adjusting crude protein for heat damage.

A crude protein determination cannot distinguish if any heat damage exists. Either the pepsin-protein or ADF-N tests must be used. Haylages, especially those that are dark-colored and/or have a sweet or tobacco smell, should be tested for heat-damaged protein.

In analyzing forages, spend money wisely and get chemical determinations that can be used effectively. Sample forages properly as well. We recommend the analyses in Table 4; additional analyses such as phosphorus, magnesium and sulfur can be used but are not necessary.