

## Proteolysis in Different Forage Silages

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### Introduction

One of the most significant processes occurring during ensiling is proteolysis or the enzymatic breakdown of proteins to soluble nonprotein nitrogen (NPN) forms such as peptides, free amino acids and ammonia. This process occurs in the first few days of ensiling and may result in up to 85% of the crude protein in alfalfa silage being soluble NPN. This loss of true protein, particularly in silages fed to high-producing dairy cows, may significantly reduce the efficiency of nitrogen utilization by ruminants. Although considerable information is available concerning proteolysis in alfalfa and red clover, much less is known about other forage silages. The objectives of this study were to: 1) investigate the effects of maturity and dry matter (DM) content on proteolysis during ensiling in seven crops (alfalfa, red clover, orchardgrass, wheat, barley, sorghum-sudan grass and corn), and 2) determine if proteolysis can be reasonably predicted by simple means available to commercial laboratories.

### Methods

Silages of different forages were made over two years. Each forage was harvested in primary growth and at least one regrowth period per year except for small grains and corn (only primary growth). Harvesting began when a forage reached a late vegetative stage and continued through at least late flower development. This was done on a weekly basis the first year and biweekly basis the second year. One portion of mowed forage was chopped in a stationary chopper and ensiled immediately. Except for corn, four other portions were weighed onto screens and dried in the greenhouse to 35, 45, 55 and 85% DM. The 35 to 55% DM portions were chopped and ensiled upon reaching the desired DM content. The 85% DM forage was stored dry to represent hay. Ensiling was done in pint (473 ml) canning jars. Prior to ensiling, all crops were inoculated at a rate of  $10^4$  bacteria/g crop with a commercial inoculant containing *Lactobacillus plantarum* and

*Pediococcus cerevisiae* to guarantee a minimum level of lactic acid bacteria for fermentation. Silos were stored at room temperature for 30 days. Unensiled forages and silages were analyzed for DM, pH, crude protein, and soluble NPN. Silages were analyzed for fermentation products.

### Results and Discussion

In the first year, much of the growing season was cool and very wet, whereas in the second year the first half of the growing season was warm with generally below normal precipitation. This variation in weather conditions produced differences in fermentation. Most silages were well preserved, but out of 600 silages, 12 had significant levels of butyric acid in the wet year and only 2 in the dry year. Also silage pHs, particularly in the legume silages, were higher and had a greater range in the wet year compared with the dry year. Most likely, the wet year produced forages of lower sugar content and higher buffering capacity.

In spite of fermentation differences, proteolysis did not appear to be affected by year. Soluble NPN (% DM) for all crops is shown in Figure 1. What was surprising was the continuum among

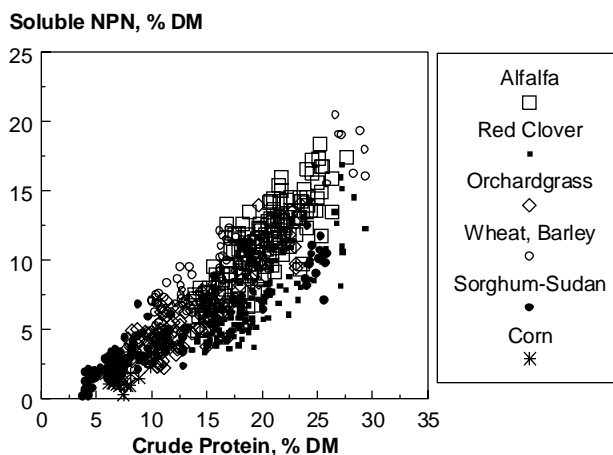


Figure 1. Soluble NPN as a function of crude protein content for all silages in both years.

crops with the exception of red clover and possibly sorghum-sudan grass. The lower NPN levels in red clover were anticipated based on earlier work at the Center, implicating the action of polyphenol oxidases. It is not known what may be affecting proteolytic activity in sorghum-sudan grass.

Stepwise regression was used to determine which factors would be useful in predicting soluble NPN (% DM basis). For all crops, crude protein and DM contents were the most important factors (Table 1). In some cases, silage pH significantly improved the prediction of soluble NPN. Excluding red clover and sorghum-sudan grass, a single regression using crude protein and DM contents as independent variables could explain 87% of the variation in

soluble NPN across the other crops. The equations are currently being used to predict soluble NPN from silages (both laboratory and field-scale silages) reported in published studies. Preliminary results indicate that the equations are reasonably predicting soluble NPN in these other studies.

### Conclusions

Overall, these results suggest that soluble NPN can be estimated from parameters that are routinely provided by forage testing laboratories. This should help forage testing laboratories to more accurately estimate the feeding value of silages for farmers. Further validation work is needed to prove that this concept in estimating soluble NPN is sound and to establish the level of error associated with the prediction equations.

Table 1. Regression equations for soluble NPN (% DM basis) in forage silages.

Crop	Number of parameters	Equation <sup>†</sup>	R <sup>2</sup> , %
Alfalfa	2	-0.840 - 0.0629 DM + 0.722 CP	77.1
	3	-0.952 - 0.0633 DM + 0.0364 PH + 0.720 CP	77.1
Red Clover	2	-3.185 - 0.0590 DM + 0.640 CP	77.3
	3	-8.242 - 0.0823 DM + 1.590 PH + 0.568 CP	80.0
Orchardgrass	2	-0.336 - 0.0326 DM + 0.553 CP	78.8
	3	2.065 - 0.0235 DM - 0.605 PH + 0.553 CP	79.2
Wheat, Barley	2	1.000 - 0.0570 DM + 0.684 CP	89.5
	3	-3.827 - 0.0895 DM + 1.596 PH + 0.609 CP	90.3
Sorghum-sudan	2	1.670 - 0.0547 DM + 0.432 CP	89.3
	3	-2.520 - 0.718 DM + 1.229 PH + 0.399 CP	90.2
Corn	2	1.250 - 0.0365 DM + 0.299 CP	59.6
	3	-0.564 - 0.0450 DM + 0.532 PH + 0.300 CP	61.2
Combined*	2	-0.947 - 0.0506 DM + 0.699 CP	86.6
	3	-1.824 - 0.0548 DM + 0.267 PH + 0.686 CP	86.7

<sup>†</sup>DM - dry matter content, %; PH - pH; CP - crude protein, % DM

\*Combined - all crop data except red clover, sorghum-sudan grass