

# Addition of Exogenous Fibrolytic Enzymes to Lactating Dairy Cow Diets<sup>1</sup>

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## Introduction

Feed costs account for an important expense on farms. Therefore, methods to improve feed utilization are highly desired. Enzymes that are fed to cattle, rather than produced by the cow, are referred to as exogenous enzymes. Exogenous enzymes have been identified as a way to improve the digestibility of feedstuffs due to their role in targeting specific nutrients within the diet. However, inconsistent results from research studies have limited their adoption by producers.

Generally, enzymes are fed to target the fiber fraction of the diet, which is the least digestible component. However, exogenous enzymes may also be used to target the dietary protein or starch fractions as well, depending on the producer's goal. The target nutrient of an enzyme can be determined from its name. For example, fibrolytic enzymes target fiber. Fiber is a complex entity due to the makeup of the structural components in the cell wall, so enzymes considered fibrolytic would be cellulolytic (cellulose-degrading), hemicellulase (hemicellulose-degrading), and xylanase (xylan-degrading) (Beauchemin et al. 2003). Enzymes that target starch are considered amylolytic; those that are protein-specific are proteolytic. Thus, when feeding enzymes to dairy cows, it could be beneficial to incorporate multiple types to ensure that more than one portion of the diet is targeted (Meale et al. 2014). Additionally, the exogenous enzyme composition used in one scenario may not

yield results in another due to differences in diet composition. This makes it important to identify the enzymatic needs of producers on a case-by-case basis to improve feed utilization, rather than considering exogenous enzymes as a blanket solution.

Many of the exogenous enzymes used in dairies focus on targeting the fiber fraction of the diet. The focus on fibrolytic enzymes stems from a few factors. Firstly, fiber is an abundant portion of the diet for lactating dairy cattle. Therefore, finding ways to improve its use opens opportunities for producers to reduce feed costs. Secondly, this is the dietary fraction that offers the most potential benefits because fiber is the least digestible macronutrient, so it is difficult for animals to capture all of the energy available from it. Starch and protein are both highly degradable in the rumen, but any portion that is not will be subject to extensive digestion in the small intestine. However, cows are unable to degrade fiber in the small intestine, instead relying on fermentation in the rumen to obtain the vast majority of its nutrients (Beauchemin et al. 2003). Improved fiber digestion increases the overall digestibility of the diet, making dairy cows more effective at producing milk and reducing the amount of feed required. This report summarizes information on the usage of exogenous enzymes, such as their modes of action and effectiveness in lactating dairy cows. This publication is for Extension agents, nutritionists, and dairy producers.

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## Mode of Action

The overall mode of action for any exogenous enzyme is to act upon a specific nutrient within the diet. This is done through an enzyme's ability to target particular chemical bonds in macronutrients. However, the method by which an enzyme acts depends on where and how the enzyme is applied to the diet.

## Application to Feed

Applying enzymes prior to feeding is the most effective way to maximize their potential. Mixing the enzyme into the diet is simple. The primary reason for this approach, rather than top-dressing it, feeding it separately, or directly adding it to the rumen, is time. Enzymes need time to attach to the target nutrient (especially those related to fiber); adding them to the diet prior to feeding allows the enzymes to begin attachment before consumption. This reduces the lag time between when feed is consumed and when it begins to undergo ruminal degradation. There does not seem to be an ideal amount of time to apply an enzymatic treatment to the diet before it is fed, as long as the enzyme is added prior to consumption (Beauchemin et al. 2003). However, there seems to be a limit to how it can be applied to feed depending on environmental and processing factors. For instance, application to pelleted feeds can reduce efficacy due to the heat treatment that occurs during this process and can prevent increases in digestibility (Bowman et al. 2002).

Finally, adding an enzyme solution rather than a dry product seems to be beneficial. This is particularly true for diets that have a high dry matter content. Water is biochemically critical for effective enzyme function due to the hydrolysis of soluble sugars (i.e., glucose) from a complex polymer (such as cellulose). This process of hydrolysis involves the addition of water to specific bonds within a complex carbohydrate, and can be limited if there is a shortage of water in the environment (Meale et al. 2014). One feed with a high-moisture environment is silage, which can make up a large proportion of the diet and contribute significantly to the fiber fraction (NDF) consumed by the animal. While high-moisture environments can enhance enzymatic activity, they can pose quality issues, especially because the sugars are used by microorganisms that can cause spoilage. This is particularly important for silages; therefore, maintaining aerobic stability is critical to ensure good silage quality (Muck et al. 2018). Due to these issues with silage treatment, it seems better to mix the enzyme into either the concentrate or dry forage portions of the diet, rather than the entire total mixed ration. Research supports addition to the concentrate as the most effective option (Beauchemin et

al. 2003). A study comparing three methods of application saw the greatest increase in digestibility when the enzyme was added to the concentrate portion of the diet rather than as a pellet or a supplement [72.6% vs. 70.1% vs. 65.9% dry matter (DM) digestibility, respectively]. This difference in digestibility is attributed to the enzyme attachment to the concentrate feeds. The differences observed for digestibility were similar with milk production as well, despite no differences in dry matter intake (DMI), with a 4 lb/d production difference between the application to the concentrate and as a supplement (Bowman et al. 2003).

## Rumen Effects

Within the rumen, the effects of exogenous enzymes are specifically related to the improvement and facilitation of feed utilization. While a pre-treated diet has many responses to exogenous enzymes, there are major changes occurring within the rumen, too. The hydrolysis of complex carbohydrates yields simple sugars. Bacteria use these sugars for growth, stimulating more microbial growth. This can lead to increases and changes in the overall rumen microbial population (Chung et al. 2012). For instance, when fibrolytic enzymes are fed, it is reported that there are also increases in the digestibility of protein. While changes in other nutrient digestibility can be attributed to changes in the rumen microbial community, this could also be due to an overall increase in enzymatic activity in the rumen. With the addition of enzymes to the feed, some degradation does begin, creating more surface area and attachment points for bacteria once the feed arrives in the rumen. This allows more hydrolysis to occur throughout the rumen, carrying on a cycle of more substrate increasing the growth of bacteria, which in turn increases endogenous bacterial enzyme secretions (McAllister et al. 2001). Recently, we tested a hemicellulose, xylanase, and amylase mixture, and observed only minor changes in ruminal fermentation, with no increase in ruminal digestibility (Bennett et al. 2021). While there is not generally an increase in the extent of nutrient digestibility with the inclusion of enzymes in the diet, there is an increase in the rate of nutrient digestibility. This is an important distinction to make. While the bacterial community will reach a plateau on its own, enzymes help reduce the lag time required for microbial attachment (Meale et al. 2014).

Because enzymes are proteins, they are susceptible to protein degradation by ruminal microorganisms. During the attachment process, a feed-enzyme complex is formed, which has another benefit of protecting the enzyme from protein degradation in the rumen. Since they are attached to feed particles, there are fewer free enzymes flowing out of

the rumen mixed with ruminal contents (Beauchemin et al. 2003). While some exogenous enzymes are able to function in the small intestine, the extent is much more limited than in the rumen. Although this would further enhance the ability of enzymes to degrade feed and increase nutrient availability, there is limited research investigating the ability of exogenous enzymes to pass through the rumen and be effective in the small intestine (Meale et al. 2014).

## Production Responses

When consulting individual research studies, it can be hard to reach a consensus regarding the effectiveness of exogenous enzymes for lactating dairy cow production. However, numerous reviews and meta-analyses have been completed, which support the idea that exogenous enzymes are able to increase animal productivity. A comprehensive meta-analysis was done at UF/IFAS (Arriola et al. 2017) analyzing 13 studies using fibrolytic enzymes. The authors reported that there were no changes in DMI; however, there was an increase in both DM and NDF digestibility (1.36% and 2.3%, respectively), providing more energy to the animal. This was supported by an increased milk production of 1.8 lb/d, and an increase in 3.5% fat-corrected milk by 1.2 lb/d. This analysis provides evidence of the potential for cellulase and xylanase to improve milk production in lactating dairy cows; however, the increases were rather small due to the variability among trials.

The results with exogenous enzymes on production are inconsistent, likely due to variability in the doses used. Optimum dose levels are difficult to determine and vary with different dietary conditions. While any low amount will be beneficial, there is a threshold. Too high of a dose can have adverse effects. This is potentially due to the competition between the enzymes and microorganisms for attachment sites on the substrate (Meale et al. 2014). As previously mentioned, there is some degradation of the enzymatic protein by endogenous enzymes which can limit the efficacy of the enzyme. If the enzymes are being degraded before they have time to attach, then they are not able to have any positive effects on nutrient digestibility.

Part of the variability may be due to the challenge in comparing results among studies. Key details, such as enzyme activity, are not always reported, which can lead to nonequivalent comparisons. Enzyme activity determines the amount of enzyme that is functional by measuring the rate of sugars released from the substrate of interest, and it can be affected by many factors. In particular, pH can be a major factor in how effective an enzyme will be. Many have strict pH ranges within which they operate optimally.

Thus, studies may test similar feeding levels of an enzyme but have different results due to differences in the activity level of the enzyme. Additionally, enzyme activity is often reported for ideal conditions that may not be representative of the rumen (Beauchemin et al. 2003). When enzymes are fed in a production setting, it is critical to understand the enzyme activity level and base the recommended dose on that rather than using a “one size fits all” inclusion level.

## Conclusions

Table 1 summarizes the results of experiments supplementing various exogenous enzymes to lactating dairy cows. This table and the previously discussed studies show that enzymes are variable in their effectiveness. Additionally, their efficacy at improving performance is inconsistent. Nevertheless, there are generally advantages associated with increasing nutrient availability through the usage of enzymes in the diet. The rumen microbial community can benefit from the inclusion of enzymes, potentially leading to important changes that allow animal productivity to increase. Dairy cows who have been fed diets containing exogenous enzymes have shown positive responses that indicate that application of enzymes is worth considering for utilization in farms. Note that there are many enzyme-based feed products available, so we recommend that producers and nutritionists pay close attention to the target compound of the enzyme, the dose, the physiological state of the animal, and the basal diet, to ensure it will benefit their animals.

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Table 1. Summary of the results observed in experiments supplementing various exogenous enzymes to lactating dairy cows.

<b>Enzymes</b>	<b>Feeding Rate</b>	<b>Effects</b>	<b>Citation</b>
Cellulase and xylanase	1 g/d	Increased milk fat and protein, increased acetate:propionate	Bowman et al. (2002)
Endoglucanase, xylanase	0.5–1 mL/kg of diet	Altered rumen microbial community, increased methane emissions	Chung et al. (2012)
Amylase, cellulase, hemicellulase, glucanase, and pectinase	10 g/d	Increased total volatile fatty acids concentration	Oh et al. (2019)
Cellulase and xylanase	1.3 g/kg of diet	Increased dry matter and fiber digestibility	Rode et al. (1999)
Amylase, xylanase	12 g/d and 8 g/d, respectively	Decreased milk protein, decreased feed efficiency, increased butyrate production	Zilio et al. (2019)