

Non Starch Polysaccharidase - A Potent tool in improving fibre digestibility in Ruminants

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Abstract

Manipulation of rumen fermentation by using feed additives has been gaining popularity in ruminant nutrition in recent years. One such approach is the use of fibrolytic enzymes in the treatment of feedstuffs to enhance their digestibility. The better understanding of the production techniques, enzyme activity, mode of enzyme action and application techniques of commercial non starch polysaccharidase enzymes can help the scientific community for competent utilization of these biotechnological products for efficient utilization of the available feed resources.

Keywords: Fibre Digestion, Ruminant, Feed Enzyme, Non starch, Polysaccharide.

Non Starch Polysaccharide (NSP)

Carbohydrates in feedstuffs of plant origin are mainly grouped into monosaccharides, oligosaccharides and polysaccharides. In plants, polysaccharides are located both intra and extracellularly. The intracellular plant polysaccharides, which are non-cell wall components, are starch and fructans. Their main function is as storage components. The extra cellular or cell wall polysaccharides are arranged in definite patterns to give fibrillar polysaccharide (cellulose), matrix polysaccharide (hemi-cellulose and pectin) and encrusting substances (lignin). These cell wall components, which do not include the storage components, are commonly known as the non-starch polysaccharides (Panda and Arun, 2006). NSP can be classified into three main groups normally cellulose, non-cellulosic polymer and pectic polysaccharides (Choct, 1997).

Feed enzymes

Feed enzymes are produced by a batch fermentation process beginning with a seed culture and growth media (Cowman, 1994). Once the fermentation is complete, the enzyme protein is separated from the fermentation residues and source organisms (Beauchemin et al., 2003).

Ruminant Feed Enzymes

The rumen contains numerous microbes, which overcome the factors that limit feed utilization by non-ruminants (Bowman et al., 2002). Feeding of exogenous enzymes in ruminants was previously an unacceptable practice, because these proteins were thought to be

degraded by ruminal proteases (Kopency et al., 1987). In the last decade, researchers have reexamined the potential use of exogenous enzymes for ruminants due to higher feed costs, lower costs of enzyme production and the availability of more active and better defined enzyme preparations (Beauchemin et al., 2003). Enzyme feed additives for ruminants are concentrated extracts obtained from fungal (*Trichoderma longibrachiatum* or *Aspergillus niger* or *Aspergillus oryzae*) and bacterial (*Bacillus spp.*) fermentations. They do not contain microbial cells, because they are removed from fermentation and finally concentrated and purified. These enzymes can breakdown specific bonds in feedstuffs, not usually degraded by endogenous enzymes, thus releasing more nutrients (Sheppy, 2001).

Cellulase

The cellulase system contains mainly endoglucanase, exoglucanase or cellobiohydrolase and β -glucosidase or cellobiase. Most endoglucanases attack internal glycosidic bonds of cellooligosaccharides and release mainly cellobiose and cellobiose, while cellobiohydrolase hydrolyze the second glycosidic bond from either the reducing or non-reducing end of the cello-oligosaccharides. However, β -1, 4 glucosidase sequentially removes one glucose unit from either the non-reducing end or both ends (Bhat and Hazlewood, 2001).

Xylanase

The main enzymes involved in degrading the xylan core polymer to soluble sugars are xylanase and β -1, 4 xylosidase (Bhat and Hazlewood, 2001). The

xylanase include endoxylanase which yield xylo-oligomers, and β -1, 4 xylosidase, which in turn yield xylose.

Hemicellulase

The two main enzymes involved in hemicellulose breakdown are endoxylanase and endomannanases, which attack the backbone structure of xylans and mannans (Viikari et al., 1993). Other hemicellulase enzymes involved primarily in the digestion of side chains include β -mannosidase or α -L-arabinofuranosidase, α -D-glucuronidase, α -D-galactosidase, acetyl xylan esterases and ferulic acid esterases (Bhat and Hazlewood, 2001).

Enzyme Activity

Enzyme activities can be assayed by in vitro method by measuring production of end products (e.g. reducing sugars) per unit time, using a specified substrate, under defined conditions (Beauchemin, 1998). Various methods are available to assay the enzymatic activities like DNSA (Dinitrosalicylic acid) reducing sugar method (Bailey, 1988), Nelson Somogyi reducing sugar method (Somogyi, 1960) and also some viscosimetric method (Mc Cleary, 2001). Out of these, reducing sugar method gives a true measure of glycosidic bonds cleaved and thus predicts enzyme activity (Mc Cleary, 2001).

Most enzyme preparation having predominant self activity may also contain substantial array of minor activities. For example, an enzyme preparation with predominant xylanase activity may also have sufficient protease, cellulase, pectinase, beta glucanase or other enzymatic activities (Beauchemin, 1998).

A temperature of approximately 60 °C and pH between 4 and 5 are the optimal condition for estimating enzyme activity of most commercial enzymes (Coughlan, 1992).

Mode of Enzyme Action

Exogenous enzymes in the rumen are generally more stable than previously thought particularly when applied to feed prior to ingestion (Morgavi et al., 2000). Application of enzymes to feed enhances the binding of the enzymes with the substrate, which increase the resistance of the enzymes to proteolysis and prolongs their residence time within the rumen.

Morgavi et al. (2000) demonstrated synergism between exogenous enzymes and ruminal enzymes such that the net combined hydrolytic effect in the rumen was much greater than that estimated from the individual activities. Wang et al. (2001) reported that enzyme supplementation increased number of non-fibrolytic and fibrolytic bacteria in a batch culture system using rumen fluid. Stimulation of rumen microbial numbers through the use of enzymes could result in greater microbial biomass, which could

provide more total polysaccharidase activity to digest feedstuffs. Consistent with this hypothesis, Yang et al. (1999) reported that enzyme supplementation of dairy cow diets increased feed digestion in the rumen and the flow of microbial protein from the rumen.

Hristov et al. (1996) proved that pre-consumptive application of exogenous enzymes cause the release of soluble carbohydrates. Krause et al. (1998) reported that there was a partial solubilization of NDF and ADF when enzyme was applied prior to feeding. Nsereko et al. (2000) demonstrated compelling evidence that applying enzymes to feed cause structural changes to occur, thereby making feed more amenable to degradation.

Wang et al. (2001) proposed that the positive effect of exogenous fibrolytic enzymes on enhancing microbial colonization is likely due to enzymatic hydrolysis of the substrate, which produce reducing sugars that attract secondary colonization or to removal of barriers to microbial attachment to feed particles by cleaving the linkage between phenolic compounds and polysaccharide. Wang et al. (2003) observed that although the reducing sugars produced from the hydrolysis of straw increased microbial adhesion to feed particles, extensive enzymatic hydrolysis of barley straw prior to exposure to ruminal microorganisms actually reduced colonization compared to that of minimally hydrolyzed straw.

Adding exogenous enzymes to the diet increase the hydrolytic capacity of the rumen mainly due to increased bacterial attachment (Yang et al., 1999; Morgavi et al., 2000; Wang et al., 2001), stimulation of rumen microbial populations (Wang et al., 2001; Nsereko et al., 2002) and synergistic effect with hydrolases of ruminal microorganisms (Morgavi et al., 2000). Thus improvements in digestibility are not limited to the dietary component to which the enzymes are applied, which explains why fibrolytic enzymes can be effective when added to the concentrate portion of the diet (Beauchemin et al., 2003).

Level of Enzyme Application

Lewis et al. (1999) noted that a medium level of enzyme supplementation produced more milk than a low or high level of application, and Beauchemin et al. (2003) stated that a high level of enzyme application was less effective than a low level at increasing total tract digestibility. The reason for poor response at higher level is partly attributed to negative feedback inhibition, which is one of the classical modes of regulation of enzyme action. This feedback mechanism occurs, when enzyme action is inhibited by production of a critical concentration of a product of the enzyme-substrate interaction. For instance fermentation of sugar produced by cell wall hydrolysis may reduce the ruminal pH to levels that inhibit cell wall digestion

(Andesogan, 2005). An alternative hypothesis is that excessive enzyme application blocks binding sites for enzymes or may prevent substrate colonization (Beauchemin et al., 2003).

Methods of Providing Enzymes to Animals

Applying fibrolytic exogenous enzymes in a liquid form on feeds prior to consumption can have a positive effect on performance (Yang et al., 1999). In contrast infusion of enzymes into the rumen has not been effective (Lewis et al., 1999). The close association of enzymes with feed may enable some form of pre-ingestive attack of the enzymes upon the plant fibre or enhance binding of the enzymes to the feed thereby increasing the resistance of the enzymes to proteolysis in the rumen (Beauchemin et al., 2003).

Lewis et al. (1996) observed an increase in total tract NDF digestibility, when an enzyme solution was applied to dry hay prior to feeding, but there was no difference between applying the enzyme immediately before feeding and a twenty four hour incubation period. In vitro trials have reported similar results (Colombatto, 2000). Thus there is no requirement for a reaction phase or incubation time between treatment and feeding of forages.

Beauchemin. et al. (1999) suggested that enzymes should be applied to a large portion of the diet to increase the chance that enzymes endure in the rumen. Adding enzymes to a small portion of the diet may allow rapid passage of enzyme from the rumen, lessening the enzyme effect in the rumen (Beauchemin et al., 2003).

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