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AGRO PRODUCTIVIDAD



Avifauna

associated to
a home garden
in Valladolid,
Yucatan, Mexico

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Methods to improve the fraction of non-degradable protein in the rumen: A review

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ABSTRACT

Objective: To investigate methods to reduce dietary protein degradability at the ruminal level and to analyze their effects on ruminal fermentation based on the analysis of available literature.

Design/methodology/approach: The protection of dietary protein leads to a lower degradation at the ruminal level and an increase in the supply and utilization of amino acids.

Limitations of the study/implications: The efficacy of the processing method depends on the ingredients used in the diet.

Conclusions: The use of physical, chemical, or combination treatments is justified on raw materials with high protein value and degradability; these efficiently protect the protein from ruminal degradation and provide a better supply to the small intestine.

Keywords: Protein, Cattle, Methods.

Citation: Paredes-Díaz, D., Manríquez-Núñez, O. M., Montaña-Gómez, M. F., Ávila-Castañeda, D., & Ramírez-Briebesca, J.F. (2024). Methods to improve the fraction of nondegradable protein in the rumen: A review. *Agro Productividad*. <https://doi.org/10.32854/agrop.v17i9.3052>

Academic Editor: Jorge Cadena Iniguez

Guest Editor: Juan Franciso Aguirre Medina

Received: July 12, 2024.

Accepted: August 15, 2024.

Published on-line: October 4, 2024.

Agro Productividad, 17(9). September. 2024. pp: 101-107.

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INTRODUCTION

High-yield ruminants for meat production require diets with adequate nutrients to achieve the desired productive performance objectives (NASEM, 2016). Dietary protein is the most expensive nutrient. In the case of feedlot-finished ruminants, more rumen-undegradable protein (RUP) is needed to reach the required levels of metabolizable protein (MP) because microbial protein is insufficient to cover the needs of animals with high nitrogen (N) demand (Silva *et al.*, 2023). Protecting dietary protein to pass through the rumen intact and avoid degradation leads to increased supply and utilization of amino acids (AA) (Loregian *et al.*, 2023). It also contributes to reducing greenhouse gas emissions derived from the fermentation of certain AA (Palangi and Lackner, 2022). Therefore, it is necessary to integrate ingredients with significant levels of RUP into the diet so that ruminants can express their maximum potential and improve the profitability of production systems while reducing the negative impact on the environment (da Silva *et al.*, 2020; Valizadeh *et al.*, 2021). Nutrition researchers have suggested physical, chemical, or a combination of both methods to increase RUP in dietary protein ingredients, protecting it from degradation and fermentation in the rumen, thus optimizing its utilization (Shishir *et al.*, 2020; Roca-Fernández *et al.*, 2020; Rigon *et al.*, 2022). This review aimed to investigate methods to reduce the degradability of dietary protein at the ruminal level, increase the RUP fraction, and analyze their effects on ruminal fermentation based on the available literature.

MATERIALS Y METHODS

The literature search was performed on Google Scholar, Scopus, and Web of Science using the keywords “Rumen Undegradable Protein”, resulting in 15,900 articles published from 1978 to 2024. However, studies performed *in vitro* and with unconventional diets or feeds were omitted. It can be observed that there is little data available regarding rumen degradability, especially in recent years, probably due to the problems arising from increasingly restrictive regulations on animal welfare.

Physical Methods

Various methods have been investigated for a long time to increase the RUP; however, thermal treatments in their different modalities are more effective than chemical treatments (Iommelli *et al.*, 2022).

Extrusion

Extrusion is a widely used technique for applications in human and animal food (Lillford, 2008; Zhang *et al.*, 2011). It consists of a thermomechanical physical process where the material is subjected to kneading, compression and cooking through a sudden increase in temperature and pressure for a short time. Heat transfer and pressure cause structural changes (Maillard reaction) in the feed ingredients, such as protein denaturation, fiber solubilization, and starch gelatinization (Solanas *et al.*, 2008).

Microwave

Microwave irradiation increases the temperature of the material with dielectric properties when the energy is absorbed and converted into heat. Its use shows advantages compared to other thermal treatments due to the reduced time required, which reduces energy and operating costs (Guzik *et al.*, 2021).

Roasting

This method is commonly used in oilseeds after solvent extraction. It consists of heating the material by conduction at a variable temperature ranging from 100 to 200 °C in a time range of 0 to 4 hours with or without pressure. Pressure roasting is carried out in horizontal or vertical cylinders similar to an autoclave; however, the pressure method has certain implications, such as increasing the processing time to increase and decrease the pressure, which leads to inaccuracy in times and temperatures (Poel *et al.*, 2005). Roasting favors the increase of the RUP and alters the sensory and functional properties, improving palatability, aroma, and color and reducing the antinutritional factors of the material (Haji-Mohammadi *et al.*, 2022).

Chemical Methods

Chemical treatments that increase RUP have varied, from tannins to alcohols, acids and aldehydes (Poel *et al.*, 2005).

Tannins

Tannins have been widely used and studied in ruminant diets and are generally classified into hydrolyzable tannins (HT) and condensed tannins (CT). Of these, only CT can form complexes with dietary protein when dosed at low levels in the diet, which promotes greater resistance to microbial degradation at the ruminal level, including crude protein (CP), dry matter (DM), organic matter (OM), neutral detergent fiber (NDF) and acid detergent fiber (ADF); ammonia (NH₃) concentration is also reduced and fecal nitrogen excretion (ENF) is increased (Orzuna-Orzuna *et al.*, 2021).

Formaldehyde

The application of formaldehyde (HCHO) decreases the activity of proteolytic microorganisms through a two-step process: 1) the formation of a methyl compound and 2) a slow condensation (Brand *et al.*, 2023) promoting the formation of methyl cross-links between HCHO and dietary protein under slightly acidic to neutral ruminal pH conditions, making the protein resistant to microbial degradation without affecting its digestibility in the small intestine (Firozi *et al.*, 2024). Among the implications of using HCHO is the high variability between foods since it depends on the solubility of the dietary protein and its possible residual effect on animal tissues and secretions (Wales *et al.*, 2010).

Malic Acid

It is an intermediate organic acid in the Krebs cycle in animal tissues and the succinate-propionate pathway of ruminal microorganisms (Ke *et al.*, 2018). Direct application in feed protects dietary protein from ruminal degradation and promotes denaturation and hydrolysis under acidic conditions of the abomasum for subsequent absorption in the small intestine (Thakur *et al.*, 2023).

Physical Methods To Increase The RUP Fraction On Ruminal Fermentation

Applying methodologies to increase the RUP alters the ruminal kinetics of CP (Rigon *et al.*, 2022). Heat treatment has a significant impact by reducing the degradation of dietary protein since it modifies the molecular structure, increasing the proportion of the β -lamina secondary structure and reducing the proportion of the α -helix structure in amide I, with a minor effect on amide II (Yan *et al.* 2014), these changes occur in the proportions of the secondary structures, where the different degradabilities of proteins at the ruminal level occur (Windt *et al.* 2022). The extrusion process at moderate temperatures (110 to 160 °C) makes DM, OM, and dietary protein more degradable without affecting the total volatile fatty acids (VFA) (Risayahadi *et al.*, 2023). Still, it decreases the branched VFA (isobutyrate and isovalerate) and acetate while increasing the proportion of butyrate (Amirteymoori *et al.*, 2021). These temperature ranges used for the extrusion process are insufficient to protect dietary protein (Orias *et al.*, 2002). Processing with longer time and temperature causes overprotection due to denaturation and the formation of cross-links between reducing sugars and AA (Berenti *et al.*, 2021). However, the optimal processing temperature and time depends on multiple factors such as moisture content, carbohydrate content and type, protein content, and presence of other compounds such as sulfites, and

therefore, optimal heat treatment parameters vary from one dietary protein to another (Van Soest, 1994). On the other hand, dry or wet roasting of legume seeds decreases the proportion of α -helices concerning β -sheets, causing the dietary protein to increase its potential for post-ruminal digestion and absorption. However, roasting processing also affects the degradability of other nutrients, mainly starch (Espinosa *et al.*, 2024). In addition to structural changes, the thermal process improves nutritional quality by decreasing or altering antinutritional factors such as trypsin inhibitors, lecithin, phytic acid, tannins, etc., which generates better digestibility and availability of nutrients in the small intestine (Valizadeh *et al.*, 2021). Including heat-treated protein feeds in the ruminant diet decreases the NH_3 level at the ruminal level derived from the low degradation of dietary protein (Marques *et al.*, 2024). However, the excessive inclusion of protected protein causes a decrease in fiber degradability indirectly when it decreases microbial protein synthesis due to the reduced access to nitrogen necessary for its metabolism, mainly affecting cellulolytic bacteria (Chesini *et al.*, 2023).

Chemical Methods To Increase The Rup Fraction On Ruminal Fermentation

Chemical treatments such as the inclusion of tannins show inconsistent results on ruminal fermentation; certain authors (Krueger *et al.*, 2010) indicate that tannins, regardless of the source, do not affect ruminal variables in high-grain diets. In contrast, both Orzuna-Orzuna *et al.* (2021) and Berça *et al.* (2023) suggest that tannin supplementation in ruminant diets has a positive effect, increasing the molar proportion of propionate and butyrate while reducing the degradability of DM, OM, CP, NDF, ADF and NH_3 content. Including *Acacia mearnsii* derived tannins at increasing levels in steers-fed high forage diets caused a linear decrease in nitrogen compounds' apparent and true digestibility. In addition, it reduced the intake and degradability of OM, NDF and NH_3 content without affecting ruminal pH. However, non-fibrous carbohydrates (NFC) degradation was also affected (Orlandi *et al.*, 2015).

In contrast, another study (Ávila *et al.*, 2020) with the same tannin source (*Acacia mearnsii*) and with increasing levels of high grain diet for steers showed no effects on the intake and digestibility of DM, OM, NDF, ADF and NFC. However, the degradability of dietary protein was reduced without altering NH_3 concentration, ruminal pH decreased and VFA increased linearly with the inclusion of tannins. Therefore, the dose and source of tannins and the basal diet are important factors to assess the impact of tannins in ruminant diets. Treatment with xylose as a reducing sugar source reduces the degradability of dietary protein, increasing the RUP (Harstad and Prestløkken, 2000; Can and Yilmaz, 2002). The addition of xylose increases the total VFA compared to other thermal methods. However, there was an increase in the molar proportions of acetate butyrate (Khatibi *et al.*, 2019) and decreases in molars of valerate, isovalerate and isobutyrate without changing the NH_3 content (Ipharraguerre *et al.*, 2005); the degradability of DM, OM and NDF was not affected, but the degradability of CP was decreased (Abdollahzadeh *et al.*, 2021). Malic acid or its ionized form as malate did not affect DM and OM uptake and degradability; however, it increased the molar proportion of butyrate (Carro *et al.*, 2006), propionate (Foley *et al.*, 2009) and decreased acetate (Bharathidhasan, 2022). Carrasco *et al.* (2012) evaluated

malic acid and sodium malic acid supplements in beef cattle; the acid form decreased the NH_3 level compared to its sodium form without altering VFA levels, indicating better protection of dietary protein.

CONCLUSIONS AND IMPLICATIONS

The effectiveness of the processing method depends on the feed on which it is used, and the decrease in protein degradability is due to the effect of protection, favoring performance in ruminant animals since the use of the nutrient is improved and the ruminal fermentation processes are optimized. The use of physical, chemical, or a combination of both treatments is justified on raw materials with high protein value and degradability. These effectively protect the protein from ruminal degradation and provide a greater protein supply to the intestine. Future research should consider a joint characterization of the methods and feeds to correlate animal productive responses better.

ACKNOWLEDGEMENTS

The first author thanks CONAHCYT for its financial support for his doctoral studies (CVU: 927626) and LGAC-COLPOS: Efficient livestock farming, sustainable well-being and climate change.

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