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

Paredes-Díaz, David[1]; Manríquez-Núñez, O. Maritza[1]; Montaño-Gómez, Martin F.[2]; Ávila-Castañeda, Daniela[3]; Ramírez-Bribiesca; J. Efrén[3*]

[1] Instituto de Investigaciones en Ciencias Veterinarias, Universidad Autónoma de Baja California, Mexicali, Baja California, México. C.P. 21386.
[2] Facultad de Economía y Relaciones Internacionales, Universidad Autónoma de Baja California, Tijuana, Baja California, México. C.P. 22427.
[3] Colegio de Postgraduados, Campus Montecillo, Texcoco, Estado de México; México. C.P. 56264.
* Correspondence: efrenrb@colpos.mx

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.

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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 (NH_3) 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) (Risyahadi *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.

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REFERENCES

Abdollahzadeh, F., Ahmadi, F., Khani, (2021). Poultry by-product meal as a replacement to xylose-treated soybean meal in diet of early- to mid-lactation Holstein cows. *Trop Anim Health Prod* 53, 38.

Amirteymoori, E., Khezri, A., Dayani, O., Khorasani, S., Mousaie, A., and Kazemi-Bonchenari, M. (2021). Effects of linseed processing method (ground versus extruded) and dietary crude protein content on performance, digestibility, ruminal fermentation pattern, and rumen protozoa population in growing lambs. *Italian Journal of Animal Science, 20*(1), 1506-1517. doi.org/10.1080/1828051X.2021.1984324.

Ávila, A. S., Zambom, M. A., Faccenda, A., Fischer, M. L., Anschau, F. A., Venturini, T. and Faciola, A. P. (2020). Effects of black wattle (*Acacia mearnsii*) condensed tannins on intake, protozoa population, ruminal fermentation, and nutrient digestibility in Jersey steers. *Animals, 10*(6), 1011. https://doi.org/10.3390/ani10061011.

Berça, A. S., Tedeschi, L. O., da Silva Cardoso, A., and Reis, R. A. (2023). Meta-analysis of the relationship between dietary condensed tannins and methane emissions by cattle. *Animal Feed Science and Technology*, 298, 115564. doi.org/10.1016/j.anifeedsci.2022.115564.

Berenti, A. M., Yari, M., Khalaji, S., Hedayati, M., Akbarian, A., & Yu, P. (2021). Effect of extrusion of soybean meal on feed spectroscopic molecular structures and on performance, blood metabolites and nutrient digestibility of Holstein dairy calves. *Animal bioscience, 34*(5), 855. doi: 10.5713/ajas.19.0899.

Bharathidhasan, A. (2022). Effect of supplemental malic acid on methane mitigation in paddy straw based complete diet for sustainable animal production in indigenous dairy cattle. *The Indian Journal of Animal Sciences, 92*(11), 314-319. doi.org/10.56093/ijans.v92i11.100033

Can, A., and Yilmaz, A. Y. D. A. N. (2002). Usage of xylose or glucose as non-enzymatic browning agent for reducing ruminal protein degradation of soybean meal. *Small ruminant research, 46*(2-3), 173-178. https://doi.org/10.1016/S0921-4488(02)00197-9.

Carrasco, C., Medel, P., Fuentetaja, A., and Carro, M. D. (2012). Effect of malate form (acid or disodium/calcium salt) supplementation on performance, ruminal parameters and blood metabolites of feedlot cattle. *Animal Feed Science and Technology, 176*(1-4), 140-149. https://doi.org/10.1016/j.anifeedsci.2012.07.017.

Carro, M. D., Ranilla, M. J., Giráldez, F. J., and Mantecón, A. R. (2006). Effects of malate on diet digestibility, microbial protein synthesis, plasma metabolites, and performance of growing lambs fed a high-concentrate diet. *Journal of animal science, 84*(2), 405-410.

Chesini, R. G., Takiya, C. S., Dias, M. S., Silva, T. B., Nunes, A. T., Grigoletto, N. T and Rennó, F. P. (2023). Dietary replacement of soybean meal with heat-treated soybean meal or high-protein corn distillers grains on nutrient digestibility and milk composition in mid-lactation cows. *Journal of Dairy Science, 106*(1), 233-244.

Da Silva, T. R., Salcedo, Y. T. G., Vesga, D. A., and Messana, J. D. (2020). Fuentes proteicas de baja degradación ruminal y su efecto en la producción de metano en bovinos de carne. *Revista Facultad de Ciencias Agropecuarias-FAGROPEC, 12*(2), 232-240. https://doi.org/10.47847/fagropec.v12n2a5

Espinosa, M. E., Ai, Y., and Yu, P. (2024). Impact of steam pressure toasting time on the alpha helix to beta sheet ratios, nutritional value, protein subfractions, and rumen fermentation parameters of Faba bean seeds for dairy cattle. *Animal Feed Science and Technology*, 308, 115885. doi.org/10.1016/j.anifeedsci.2024.115885.

Firozi, F., Dayani, O., Tahmasbi, R., & Dadvar, P. (2024). Feeding formaldehyde-treated sesame meal to lactating Murciano-Granadina goats: implications on milk yield and composition, digestibility, rumen fermentation, and blood metabolites. *Spanish journal of agricultural research, 22*(1), 602.

Foley, P. A., Kenny, D. A., Callan, J. J., Boland, T. M., and O'Mara, F. P. (2009). Effect of DL-malic acid supplementation on feed intake, methane emission, and rumen fermentation in beef cattle. *Journal of animal science, 87*(3), 1048-1057. doi.org/10.2527/jas.2008-1026.

Guzik, P., Kulawik, P., Zając, M., and Migdał, W. (2021). Microwave applications in the food industry: an overview of recent developments. *Critical Reviews in Food Science and Nutrition, 62*(29), 7989-8008. https://doi.org/10.1080/10408398.2021.1922871.

Haji-Mohammadi, B., Hoseinkhani, A., Taghizadeh, A., & Mohammadzadeh, H. (2022). Effect of different processing methods of soybean on ruminal disappearance of crude protein and dry matter using gas production and nylon bag techniques. *Journal of Animal Science Research, 32*(1), 125-142. 10.22034/as.2022.21658.1389.

Harstad, O. M., and Prestløkken, E. (2000). Effective rumen degradability and intestinal indigestibility of individual amino acids in solvent-extracted soybean meal (SBM) and xylose-treated SBM (SoyPass®) determined *in situ*. *Animal feed science and technology, 83*(1), 31-47.

Iommelli, P., Zicarelli, F., Musco, N., Sarubbi, F., Grossi, M., Lotito, D. and Tudisco, R. (2022). Effect of cereals and legumes processing on in situ rumen protein degradability: A review. *Fermentation, 8*(8), 363. https://doi.org/10.3390/fermentation8080363

Ipharraguerre, I. R., J. H. Clark, and D. E. Freeman (2005). "Rumen fermentation and intestinal supply of nutrients in dairy cows fed rumen-protected soy products." *Journal of dairy science* 88: 2879-2892.

Ke, W. C., Ding, W. R., Ding, L. M., Xu, D. M., Zhang, P., Li, F. H., and Guo, X. S. (2018). Influences of malic acid isomers and their application levels on fermentation quality and biochemical characteristics of alfalfa silage. *Animal Feed Science and Technology*, 245, 1-9. https://doi.org/10.1016/j.anifeedsci.2018.08.012.

Khatibi S, A., Danesh Mesgaran, M., and Zahmatkesh, D. (2019). Effect of feeding of various types of soybean meal and differently processed barley grain on performance of high producing lactating Holstein dairy cows. *Iranian Journal of Applied Animal Science, 9*(4), 625-633.

Krueger, W. K., Gutierrez-Bañuelos, H., Carstens, G. E., Min, B. R., Pinchak, W. E., Gomez, R. R. and Forbes, T. D. A. (2010). Effects of dietary tannin source on performance, feed efficiency, ruminal fermentation, and carcass and non-carcass traits in steers fed a high-grain diet. *Animal Feed Science and Technology, 159*(1-2), 1-9.

Lillford, PJ (2008). Extrusión. En Food Materials Science: Principles and Practice (pp. 415-435). Nueva York, NY: Springer New York. https://doi.org/10.1007/978-0-387-71947-4_1.

Loregian, K. E., Pereira, D. A., Rigon, F., Magnani, E., Marcondes, M. I., Baumel, E. A., and Paula, E. M. (2023). Effect of tannin Inclusion on the enhancement of rumen undegradable protein of different protein sources. *Ruminants* 3-4, 413-424. doi.org/10.3390/ruminants3040034.

Marques, O. F. C., de Oliveira, E. R., Gandra, J. R., Peixoto, E. L. T., Monção, F. P., de Araújo Gabriel, A. M. and de Lima, B. M. (2024). Dietary replacement of soybean meal with heat-treated grain soybean in diets of feedlot-finished beef cattle: impacts on intake, digestibility, and ruminal parameters. *Tropical Animal Health and Production, 56*(1), 13. https://doi.org/10.1007/s11250-023-03862-3.

National Academies of Sciences, Engineering, and Medicine, Nutrient Requirements of Beef Cattle: Eighth Revised Edition; National Academy Press: Washington, DC, USA, 2016.

Orias, F., Aldrich, C. G., Elizalde, J. C., Bauer, L. L., and Merchen, N. R. (2002). The effects of dry extrusion temperature of whole soybeans on digestion of protein and amino acids by steers. *Journal of animal science, 80*(9), 2493-2501. https://doi.org/10.1093/ansci/80.9.2493.

Orlandi, T., Kozloski, G. V., Alves, T. P., Mesquita, F. R., and Ávila, S. C. (2015). Digestibility, ruminal fermentation and duodenal flux of amino acids in steers fed grass forage plus concentrate containing increasing levels of *Acacia mearnsii* tannin extract. *Animal Feed Science and Technology*, 210, 37-45. doi.org/10.1016/j.anifeedsci.2015.09.012.

Orzuna-Orzuna, J. F., Dorantes-Iturbide, G., Lara-Bueno, A., Mendoza-Martínez, G. D., Miranda-Romero, L. A., & Hernández-García, P. A. (2021). Effects of dietary tannins' supplementation on growth performance, rumen fermentation, and enteric methane emissions in beef cattle: A meta-analysis. *Sustainability, 13*(13), 7410. https://doi.org/10.3390/su13137410.

Palangi, V., and Lackner, M. (2022). Management of enteric methane emissions in ruminants using feed additives: A review. *Animals, 12*(24), 3452. https://doi.org/10.3390/ani12243452.

Poel, A. V. D., Prestløkken, E., and Goelema, J. O. (2005). Feed processing: effects on nutrient degradation and digestibility.

Rigon, F., Pereira, D. A., Loregian, K. E., Magnani, E., Marcondes, M. I., Branco, R. H., and Paula, E. M. (2022). Use of heating methods and xylose to increase rumen undegradable protein of alternative protein sources: 1) peanut meal. *Animals, 13*(1), 23. https://doi.org/10.3390/ani13010023

Risyahadi, S. T., Martin, R. S. H., Sukria, H. A., and Jayanegara, A. (2023). Effects of dietary extrusion on rumen fermentation, nutrient digestibility, performance and milk composition of dairy cattle: a meta-analysis. *Animal Bioscience, 36*(10), 1546. doi:10.5713/ab.23.0012

Roca-Fernández, A. I., Dillard, S. L., and Soder, K. J. (2020). Ruminal fermentation and enteric methane production of legumes containing condensed tannins fed in continuous culture. *Journal of Dairy Science, 103*(8), 7028-7038. https://doi.org/10.3168/jds.2019-17627.

Shishir, M. S. R., Brodie, G., Cullen, B., Kaur, R., Cho, E., and Cheng, L. (2020). Microwave heat treatment-induced changes in forage hay digestibility and cell microstructure. *Applied Sciences, 10*(22), 8017. https://doi.org/10.3390/app10228017.

Silva, F.A.S.; Benedeti, P.D.B.; Silva, L.F.C.E.; Rotta, P.P.; Menezes, A.C.B.; Marcondes, M.I.; Valadares Filho, S.C. Protein and Amino Acids Requirements for Beef Cattle. In Nutrient Requirements of Zebu and Crossbred Cattle, 4th ed.; Valadares Filho, S.C., Saraiva, D.T., Benedeti, P.D.B., Silva, F.A.S., Chizzotti, M.L., Eds.; Independent Production: Visconde de Rio Branco, MG, Brazil, 2023; pp. 201–232.

Solanas, E. M., Castrillo, C., Jover, M., and Vega, A. (2008). Effect of extrusion on *in situ* ruminal protein degradability and *in vitro* digestibility of undegraded protein from different feedstuffs. *Journal of the Science of Food and Agriculture, 88*(15), 2589-2597. doi.org/10.1002/jsfa.3345.

Thakur, S., Dey, A., and Lailer, P. C. (2023). Malic acid-heat treatment of oil cakes enhances rumen undegradable protein for effective protein utilization in buffaloes (*Bubalus bubalis*). *Indian J Anim Health, 62*(2), 212-221: doi.org/10.36062/ijah.2023.spl.02023

Valizadeh, A., Kazemi-Bonchenari, M., Khodaei-Motlagh, M., Moradi, M. H., and Salem, A. Z. M. (2021). Effects of different rumen undegradable to rumen degradable protein ratios on performance, ruminal fermentation, urinary purine derivatives, and carcass characteristics of growing lambs fed a high wheat straw-based diet. *Small Ruminant Research*, 197, 106330. doi.org/10.1016/j.smallrumres.2021.106330

Valizadeh, A., Kazemi-Bonchenari, M., Khodaei-Motlagh, M., Moradi, M. H. and Salem, A. Z. M. (2021). Effects of different rumen undegradable to rumen degradable protein ratios on performance, ruminal fermentation, urinary purine derivatives, and carcass characteristics of growing lambs fed a high wheat straw-based diet. *Small Ruminant Research*, 197, 106330. doi.org/10.1016/j.smallrumres.2021.106330.

Van Soest, P. J. (1994). Nutritional ecology of the ruminant. Cornell university press.

Wales, A. D., Allen, V. M., and Davies, R. H. (2010). Chemical treatment of animal feed and water for the control of Salmonella. *Foodborne Pathogens and Disease, 7*(1), 3-15. doi.org/10.1089/fpd.2009.03.

Windt, X., Scott, E. L., and Bitter, J. H. (2022). Fourier transform infrared spectroscopy for assessing structural and enzymatic reactivity changes induced during feather hydrolysis. *ACS omega, 7*(44), 924-930. https://doi.org/10.1021/acsomega.2c04216

Yan, X., Khan, N. A., Zhang, F., Yang, L., and Yu, P. (2014). Microwave irradiation induced changes in protein molecular structures of barley grains: relationship to changes in protein chemical profile, protein subfractions, and digestion in dairy cows. *Journal of agricultural and food chemistry, 62*(28), 6546-6555. https://doi.org/10.1021/jf501024j.

Zhang, M., Bai, X., and Zhang, Z. (2011). Extrusion process improves the functionality of soluble dietary fiber in oat bran. *Journal of Cereal Science, 54*(1), 98-103. https://doi.org/10.1016/j.jcs.2011.04.001.