Open Access

J. Glob. Innov. Agric. Sci., 2025, 13(2):503-508
ISSN (Online): 2788-4546; ISSN (Print):2788-4538
DOI: https://doi.org/10.22194/JGIAS/25.1562
http://www.jgiass.com

Society for Innovative Agiculture

The Effect of Using Eco-Enzymes As Starters on The Physical and Nutritional Quality of Corn Cob Fermentation

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Corn cobs are a potential source of fiber for ruminants, yet it has not been optimally utilized due to its low digestibility. The nutritional content and digestibility of corn cobs could be maximized through microbial activity during the fermentation process. By improving both, corn cobs could be used as valuable fiber sources for ruminants. The microbial activity could be improved by adding starters on the substrate. This study aims to identify the effect of eco-enzymes as starters on the physical and nutritional quality of corn cobs fermentation. This study used a completely randomized design with four treatments namely P0: corn cob without eco-enzyme; P1: corn cob + Eco-enzyme 1,5%; P2: corn cob + Eco-enzyme 3%; and P3: corn cob + Eco-enzyme 4,5% with three replications for each treatment. The results show that using eco-enzyme as a starter could prevent fungal growth in all treatments. In addition, it also improved fermentation quality on T2 and T3 treatment. Using eco-enzyme as a starter also significantly ($P<0.05$) decreases pH and crude fiber content and increases crude protein, ether extract, nitrogen-free extract, and total digestible nutrient of fermented corn cobs. It can be concluded that providing eco-enzyme starters at different levels can improve the quality of the final results of corn cob fermentation.
Keywords: Agricultural waste, corn cobs, ruminant, eco-enzyme, fermentation, physical quality, nutritional quality.

INTRODUCTION

Agricultural by-products are renewable materials, available in abundance, and have the potential to be reprocessed into more valuable products. Corn cobs are one of the agricultural by-products that have not been optimally utilized due to their low digestibility, so they are often left to pile up and pollute the environment. Berber-Villamar *et al.* (2018) stated that, currently, most of the agro-industrial waste produced globally is still not utilized and is disposed of as waste, for example, 144 million tons of corn cobs are produced annually as waste during corn processing and are discarded and burned without any benefit. To realize this potential, agricultural by-products such as corn cobs and other by-products can be utilized as raw materials for animal feed (Pandey *et al*., 2000; Qureshi *et al*., 2016; Singh-Nee Nigam and Ashok, 2009; Soccol *et al*., 2017).

Corn cobs are a potential source of fiber for feed because of their high cellulose and hemicellulose content and their abundant quantity which has not been utilized optimally. (Widiasri *et al*., 2024) stated that corn plant waste that can be used as animal feed is corn cobs. However, corn cob waste is rarely used by farmers as a source of fiber for ruminant livestock because it has a low protein content, high crude fiber, and low palatability. According to Alimon (2009) corn cobs contain 90.0% dry matter (DM), 3.0% crude protein (CP), 36.0% crude fiber (CF), 0.5% crude lipid (CL), 2.0% ash, 48.0% total digestible nutrients (TDN), 36.0% acid detergent fiber (ADF), 39.0% neutral detergent fiber (NDF), 0.12% Ca, and 0.04% P. The low nutritional quality of corn cobs can be improved through fermentation. Fermentation on agricultural waste such as corn cobs improves their physical and nutritional quality and maximizes their potential as a substitute for fiber sources for ruminants. Microbial activity during fermentation can change the composition of nutrient components in the substrate, such as protein and crude fiber. It also increases the digestibility and total digestible nutrient of corn cobs. According to Hall (2013), efficient ruminant productivity requires optimal protein and energy levels. Animal productivity can be increased by synchronizing the ruminal availability of carbohydrates and proteins.

Islamiyati, R., B. Nohong, F. Wakano, R.A. Iksan and A.M.A. Dzulqaidah. 2025. The effect of using eco-enzymes as starters on the physical and nutritional quality of corn cob fermentation. Journal of Global Innovations in Agricultural Sciences 13:503-508.
[Received 10 Jun 2024; Accepted 17 Nov 2024; Published 2 Apr 2025]

The success of a fermentation process can be increased by adding starters in the form of decomposer bacteria or water-soluble carbohydrate sources. A common study used microbials as starters to increase the success of the fermentation process. However, research on the use of eco-enzymes as starters in the fermentation of agricultural waste, especially corn cobs, has not been widespread and is almost non-existent. Based on this, it is necessary to research the effect of using eco-enzymes as starters on corn cob fermentation.

MATERIALS AND METHODS

***Place, timeline and research design*:** The research was conducted at the Ruminant Nutrition Science Laboratory and the Feed Chemistry Laboratory of the Faculty of Animal Husbandry, Hasanuddin University. The research period took place from March to July 2024. This study used a completely randomized design (CRD) (Steel and Torrie, 1993) with four treatments namely P0: corn cob without eco-enzyme; P1: corn cob + Eco-enzyme 1,5%; P2: corn cob + Eco-enzyme 3%; and P3: corn cob + Eco-enzyme 4,5%. Each treatment had three replications, so there were 12 experimental units. The data obtained in this study were analyzed using SPSS software version 26, if there was an influence then it was further tested using the Duncan method (Gaspersz, 1994).

***Eco-enzyme production process*:** Making eco-enzymes refers to the method described by Samriti *et al*. (2019). A ratio of 1:3:10 sugar (palm sugar/molasses), vegetable/fruit skin and water were mixed and allowed to expand in airtight plastic bottles. Then, these bottles were placed in a cool, dry, and well-ventilated place and protected from direct sunlight. During the fermentation process, the gas produced was released daily from the bottle. After 3 months, filtration was carried out so that the water and eco-enzyme filtrate are separated. The eco-enzyme liquid was then stored for further use. Based on the results of the eco-enzyme content test, the cellulose enzyme activity contained in the eco-enzyme was 0.5608 U/ml and the total lactic acid bacteria from the eco-enzyme was 2.2 x 106 CFU/ml with a pH of 2.97.

Eco-enzyme starter is obtained from the liquid from the previous eco-enzyme fermentation that has been harvested. The eco-enzyme is then collected into a container and measured according to the level of each treatment. After weighing is complete, the eco-enzyme was stored and grouped based on treatment.

Figure 1. Treatment process; (a) raw corn cob, (b) ground corn cobs, (c) eco-enzyme, water and molasses, (d) anaerobic-wrapped, (e) fermentation result.

***Corn cobs collection*:** The corn cobs used were Bisi-18 variety corn from corn farm in Maros Regency, South Sulawesi. The cobs were ground quite fine to a size of + 0.3 cm. Then weighed 1 kg/experimental unit each. The provision of eco-enzyme starter on corn cobs in each treatment was done by mixing the eco-enzyme that has been weighed according to the treatment with water and molasses with a ratio of 5% of the total ingredients. After mixing, the solution was sprayed on the corn cobs while stirring for even mixing. The corn cobs were then put into a silo while being compacted, then the silo is tightly closed and stored in a shady place and then left for one week for the fermentation process.

RESULTS AND DISCUSSION

***Physical conditions of corn cob fermentation*:** The results of observations of the physical condition of corn cobs fermented with eco-enzyme starters at different levels are presented in Table 1.

Table 1. Physical condition of corn cobs fermented with eco-enzyme starter at different levels.

Item	T_0	T_1	T_2	T_3
Colour	Beige	Beige	Beige	Beige
Texture	Hard	Hard	Light soft	Light soft
Mushroom growth	None	None	None	None
Odor	Typical smell of corn cobs	Typical smell of corn cobs	Fermentation smell	Fermentation smell

Source: Primary data

Based on the observation results above, it is known that the provision of eco-enzyme starter at different levels did not affect the colour of the results of corn cob fermentation. In addition, the eco-enzyme starter can also prevent fungal growth due to the antimicrobial properties of eco-enzyme which are also found in the starter used. This is in line with Gu *et al*. (2021) that eco-enzyme is a multifunctional solution that can be used as a fertilizer, plant growth hormone, pesticide, insecticide, wastewater treatment, and antimicrobial agent.

The provision of eco-enzyme starter at different levels affected the texture parameters of the final results of corn cob fermentation, this is due to the increase in the level of eco-enzyme starter given. The increase in the amount of eco-enzyme starter given to the fermentable material is positively correlated with the increase in ligninolytic enzymes that can lyse or break the bonds of cellulose, hemicellulose, and lignin in corn cobs, making the corn cobs softer. In addition, the cobs also contain a suitable substrate for the growth of molds that can synthesize ligninolytic enzymes, the ligninoolytic enzyme will work on substrates/feed materials that are high in cellulose to degrade them into simpler forms so that they can be easily utilized by livestock, Ilmi and Dwianta (2013) states that in corn stalks, cellulose is surrounded by lignin, so that lignin is the first to be broken down by fungi, Cruz *et al*. (2000), added lignin content to corn cobs by 20.3%, hemicellulose by 31.7%, and cellulose by 34.7.

Increasing the level of eco-enzyme starter in corn cob fermentation also affected the aroma of the final fermentation result. Changes in the aroma of the material also indicate the activity of microbes that digest the substrate in corn cobs, producing lactic acid and a distinctive fermentation aroma. (Sharma *et al*., 2020) stated that lignocellulosic waste is a good substrate for producing aromas, such as alcohols, aldehydes, esters, fatty acids, ketones, lactones, pyrazines, and terpenes. Furthermore, various studies have shown that several aroma compounds can exhibit biological activities such as antimicrobial, antiviral, anti-inflammatory, and antioxidant properties (Kaur *et al*., 2019; Sales *et al*., 2020).

Nutrient content of fermented corn cobs: Based on the results of the analysis of variance, it was observed that the eco-enzyme level treatment had a significant effect ($P<0.05$) on the content of crude fiber, crude fat, TDN and had no significant effect ($P>0.05$) on crude protein, NFE, pH. The higher the level of eco-enzyme starter administration on corn cobs, the lower the pH obtained, so that the fermentation process is more optimal. Shi *et al*. (2022) stated that in an efficient fermentation system, the role of pH regulation plays a role in the product spectrum, conversion efficiency, and microbial transition. The production spectrum is shifted by pH regulation, which is revealed from metabolic energy modeling.

The decrease in crude fiber content in fermented corn cobs is caused by the activity of microbes that digest lignin which in the process produces lognolytic enzymes such as lignin peroxidase so that lignin decomposition occurs more quickly. Hatakka (1994) stated that lignin peroxidase (LiP) and manganese peroxidase (MnP) are extracellular peroxidase enzymes that use H_2O_2 in degrading lignin, while laccase is an enzyme containing copper using oxygen molecules in degrading lignin.

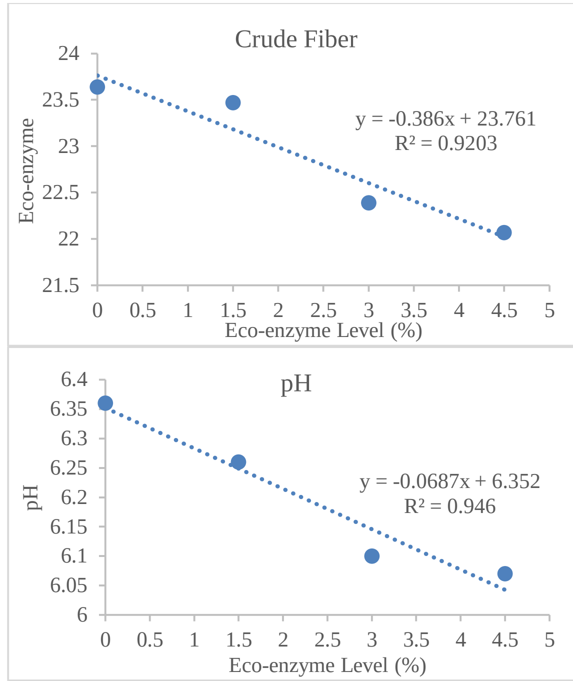

Figure 2. pH and CF content of corn cob fermentation.

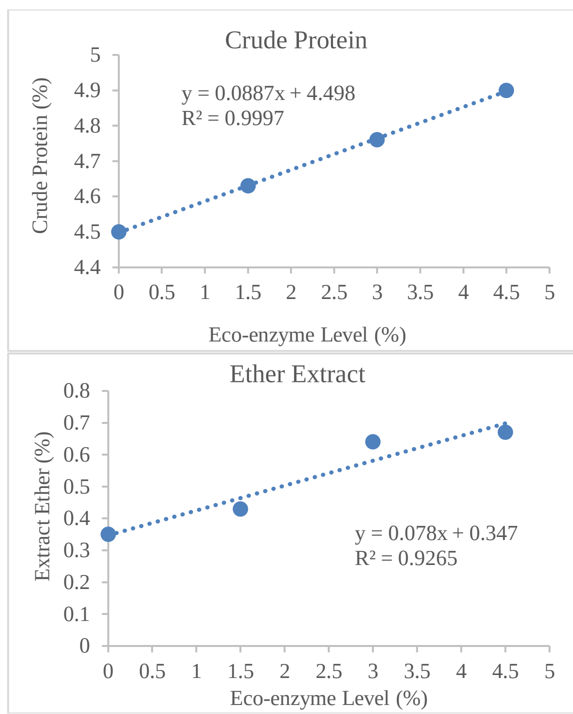

Figure 3. CP content and ether extract of corn cob fermentation.

Statistically, the crude protein content in fermented corn cobs does not differ significantly because corn cobs are basically low protein (0.7%) and high fiber (37.2%). However, if viewed based on the numbers, the higher the level of eco-enzyme starter administration, the higher the crude protein content in corn cob fermentation. The higher level of eco enzyme, the higher crude protein content. It can be looked by following the formula Y = 0.088x + 4.498. This shows that the eco-enzyme starter is able to increase the protein content in the fermented material, because the greater the number of enzymes available to break down the complex proteins in the substrate. This is in line with Gu *et al*. (2021) who states that the most common enzyme activities in eco-enzymes include protease, amylase, lipase, cellulase, and pectinase. Protease is an enzyme that breaks down proteins into smaller molecules. In addition, basically, enzymes themselves are a group of proteins that act as biocatalysts. The increase in ether extract content in the fermentation material shows that the increase in the level of the eco-enzyme starter used is positively correlated with the increase in ether extract in corn cobs. The increase in ether extract in a material shows that the feed material is of increasing quality. This is because EE is one of the components used as energy for ruminant livestock. Detmann *et al*. (2006) reported that although it has a very low concentration compared to other components in most foods produced in tropical areas, EE has a central position in obtaining energy content estimates because of its high calorie concentration.

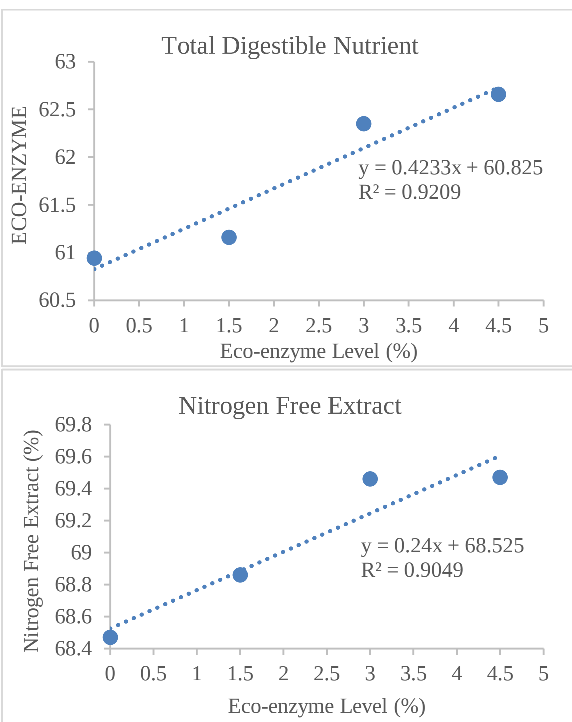

Figure 4. TDN content and pH of corn cob fermentation.

The content of nitrogen free extract (NFE) in corn cob fermentation increased along with the increase in the level of eco-enzyme starter used in each treatment, so this shows that the addition of eco-enzyme starter to the corn cob fermentation process improves the quality of the final fermentation results. This is in line with the opinion of Anggorodi (2005) who stated that NFE is a carbohydrate that is easily digested so if the NFE content is high, the quality of the feed will be better. Ikhlas *et al*. (2024) added that NFE is a soluble carbohydrate that includes monosaccharides, disaccharides, and polysaccharides that are easily soluble in acidic and alkaline solutions and have high digestibility.

The provision of eco-enzyme starter with different levels also increased the TDN of the final results of corn cob fermentation. The results obtained in this study were higher than those reported by Alimon (2009), which was 48%, this was because the corn cobs had been fermented, and the results obtained were also higher than those reported by Trihatma *et al*. (2018) that the TDN of corn cobs after fermentation was 49.67%. The much higher results in this study can be caused by the mixture of molasses (+ 80% TDN) in the material, which affects the final results of the TDN of corn cobs. Lemus (2020) stated that one of the most common ways to determine the concentration of available energy in forage is to determine the total digestible nutrients (TDN). This calculated forage parameter is easy to understand and is still a common measure of energy. Total digestible nutrients are usually the sum of digestible protein, digestible crude fiber, digestible NFE, and digestible fat.

***Conclusion*:** Providing eco-enzyme starters at different levels can improve the quality of the final results of corn cob fermentation, as indicated by a decrease in crude fiber, as well as an increase in protein and TDN levels of corn cobs. Limitations in the study related to the short fermentation period and the application of fermentation results in vivo are highly recommended for future research. The fermentation time in this study was relatively short, for further research, the time can be extended with the right environmental conditions such as temperature, humidity, and EE levels. The best results of this study can be applied in vivo to ruminant livestock with an evaluation of palatability, nutrient digestibility, and performance.

***Authors' contributions*:** Conceptualization of the manuscript and development of the methodology: R.I., B.N., and F.W.; data collection and curation: R.A. I. and A.M.A.D.; data analysis: R.I., B.N., and F.W.; data interpretation: R.I., B.N., and F.W.; funding acquisition and resources: R.I.; project administration: R.I., B.N., and F.W.; supervision: R.I. and B. N.; writing the original draft of the manuscript: R.I., B.N., and F.W.; writing, review and editing: R.I., B.N., and F.W.; All authors read and agreed to the published version of the manuscript.

***Conflict of interest*:** The authors declare no conflict of interest.

***Funding*:** This experiment was not supported by any external funding.

***Ethical statement*:** N/A

***Availability of data and material*:** Available

***Acknowledgment*:** We are thankful to every parties that are involved.

***Informed consent*:** N/A

***Consent to participate*:** All interviewees clearly understood the content and purpose of the survey and participated voluntarily.

***Consent for publication*:** All authors submitted consent to publish this research article in JGIAS.

***SDGs addressed*:** Responsible Consumption and Production.

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