Available online freely at www.isisn.org

Bioscience Research

Print ISSN: 1811-9506 Online ISSN: 2218-3973

Journal by Innovative Scientific Information & Services Network

RESEARCH ARTICLE BIOSCIENCE RESEARCH, 2020 17(2): 988-993. OPEN ACCESS

Improving the digestibility value of the feed of banana stems by fermentation using Effective Microorganism 4 (EM4) and Organic Liquid Supplement (OLS)

I Dewa Gede Mayun Permana[1], Ida Bagus Wayan Gunam[2*] and I Dewa Gede Agus Tri Putra[3]

[1]Department of Food Science and Technology, Faculty of Agricultural Technology, Udayana University, Badung, 80361, Bali **Indonesia**

[2]Department of Agro industrial Technology, Faculty of Agricultural Technology, Udayana University, Badung 80361, Bali **Indonesia**

[3]Department of Mechanical Engineering, Bali State Polytechnic, Jimbaran, Badung 80361 Bali, **Indonesia**

***Correspondence:** ibwgunam@unud.ac.id **Received 10-04-2020, Revised: 06-06-2020, Accepted: 08-06-2020 e-Published: 09-06-2020**

The use of agricultural waste as animal feed is quite beneficial in obtaining the nutritional needs of livestock. Fermentation of animal feed increases the digestibility of dry matter and organic matter. This study aims to improve the digestibility value of the feed of banana stems by fermentation. The research design used in this study was a complete randomized design with three replications. The treatment is by fermenting the banana stems using two types of starter which include effective microorganism 4 (EM4), organic liquid supplement (OLS) and without a starter. The results showed that the fermentation treatment on a banana stem with starter can increase the digestibility value of the feed by 39% and the fermentation result increases the protein, fat content, and the extract without nitrogen. The EM4 starter provides better results than the OLS.

Keywords: Banana stem, Starter, Feed, Fermentation, Digestibility value

INTRODUCTION

In Indonesia, forage is difficult to occur in the dry season, while agricultural waste is very abundant and has the potential to pollute the environment. The uses of agricultural waste as animal feed is quite advantageous in obtaining the nutritional needs of livestock and reduce pollution (Al-Arif et al., 2017; Isnawati et al., 2018). Banana plants (*Musa paradisiaca*) are able to grow well in the tropical regions, including Indonesia. All this time, what is widely used from banana plants are the fruits and a small portion of leaves used as packaging, while the stem has never been widely used. This causes a large amount of waste and ends up disrupting the environment.

Banana stems contain a high amount of water at 92.50%, while protein is 0.35% and crude fiber is 4.60%. Besides that, it contains various useful minerals such as calcium, potassium, magnesium, iron and the highly important Zn which levels are 37−163 ppm (Santoso et al., 2004). Pseudostems contain 5−8% dry matter (DM) and 3−5% crude protein (CP). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) vary between 50−70% and 30−40%, respectively (Wadhwa and Bakshi, 2013). Bagheripour et al., (2008) reported that silage water-soluble carbohydrates (WSC), acid detergent fiber (ADF), and NDF contents of pistachio products declined with increasing fermentation time. The effective degradability (ED) of banana pseudo-stems (BPS), neutral detergent fiber (NDF) is higher than banana pseudo-stems without ensiled/control (BPS0). The banana pseudo-stem used in China as a source for in the form of silage. However, information on the nutritive value of BPS on different incubation periods still lacks (Wang et al., 2016). If the water level is decreased, higher protein and mineral content can be found.

The demand for animal protein still increases, resulting in an increase in the development of the livestock sector such as cattle. The problem often faced in cattle farms is the availability of regular and quality feed. Forage feed sources are often constrained by their availability due to limited land and even narrower due to land conversion. This is a problem in the development of cattle, especially the availability of their feed, which is why an alternative source is needed.

Based on its chemical composition, banana stems can be used to feed animals such as cattle. Improving the quality of banana stems can be done by fermentation (Wang et al., 2016). In the fermentation process, a macro overhaul of molecules occurs with the microbial activity turning into a simpler molecule. As carbohydrates broke down into glucose, the protein will be hydrolyzed into peptides to easily digest by animal intestines. Fermentation increases the ileum digestibility of dry matter, organic matter and barley energy by 6% and wheat by 3% (Jørgensen et al., 2010). According to Ginting (2018) fat, crude protein, pure protein, and ash content increased significantly from 1.12%, 6.60%, 4.38%, and 4.45% to 2.09%, 15.54%, 13.37%, and 8.02% respectively when fermentation reaches 96 hours in the presence of ammonium sulfate. In addition, acids and flavor-forming compounds are formed which increases the appetite of cows, and reduce odors (Santoso et al., 2004). In a recent study, the efficiency of utilizing rice straw for ruminants increased by supplementing probiotic bacteria (Thalib, 2008).

Good quality feed is not necessarily optimally utilized by livestock, partly because of microbial fermentation in the rumen. In low-quality feed, protozoa feed on bacteria as a nitrogen source. This condition is not optimal for rumen fermentation, mainly due to the reduced population of fiber decomposing bacteria (Prayitno and Hidayat, 2013). Hungate (1988) reported that feed fermentation will produce energy and CO_2. EM4 is a mixture of microorganism cultures consisting of lactic acid producing bacteria (*Lactobacillus*), yeast and fungi that have the potential to increase biological reactions for industrial wastewater management (Higa, 1994). EM4 can improve the nutritional quality of concentrated rations, such as increasing the content of dry ingredients, organic matter, crude fat and nitrogen free extract (NFE) (Zega et al., 2017). Giving 1.2 mL of EM4 fermentation feces at the level of 10% produces the best quality. Fermented feces produce better carcass quality compared to unfermented feces without the effect of accumulation of abdominal fat (Santoso et al., 2004). For this reason, it is necessary to do research on the manufacture of animal feed from fermented banana stems.

MATERIALS AND METHODS

The materials used are *kepok* bananas (*Musa paradisiaca* L.), effective microorganisms 4 (EM4), organic liquid supplement (OLS) and the chemicals needed are NaOH, HCl, H_2SO_4 (Sigma Co., USA), petroleum-ether, ether (Merck, Germany) and distillate water. All chemicals used for this study were analytical grade.

Experimental design

This study uses a complete randomized design with three replications. The treatment given was used of 2 (two) types of starters, that was effective microorganisms 4 (EM4) and organic liquid supplements (OLS) and controls.

The banana stem was chopped to small sizes. Chopped banana stems were dried, leaving only 20% of water level. Five liters of clean water was prepared in a bucket container. Then was added into molasses as much as 200 mL and urea as much as 5 g. Then EM4 and OLS have added as much as 10 mL each. The mixture was stirred so that the ingredients dissolve evenly. After that, the ingredients in the form of chopped banana stem sprayed with a starter (EM4 and OLS) with a ratio of 1 L starter for 100 kg of ingredients (chopped banana stems) using hand sprayer. After the material was sprayed evenly, it was then wrapped and tightly closed before being fermented for 2 weeks. After the fermentation was completed, a chemical and sensory analysis was processed.

Analytical methods

The fermented banana stems were analyzed for protein, fat, ash, water and carbohydrate content with the proximate method (AOAC, 1990), dry matter digestibility (DMD), organic matter digestibility (OMD), nitrogen-free extract (NFE) based on the N.A.S method (1971) in AOAC (1990) and total digestible nutrient (TDN) based on the Fowler method (1975) and sensory testing of aroma and color (Soekarto, 1985).

RESULTS AND DISCUSSION

Content of dried and organic compounds

The dried compounds and organic material obtained from fermentation can be seen in Table 1. The results show that the content within the dried compounds in the unfermented dried material weighs heavier than the material fermented with both EM4 and OLS. This shows that very little fermented material has a macromolecular overhaul. The treatment of EM4 and OLS containing microbes will break down crude fiber into simpler molecules such as glucose and ethanol by yeast and become lactic acid by the lactic acid bacteria. As known that EM4 contains microbes such as *Lactobacillus casei*, *Saccharomyces cerevisiae*, *Rhodopseudomonas palustris* (Santosa et al., 2017), *Actinomycetes*, *Bacillus mucilaginous* (phosphate solvent bacteria), and photosynthetic bacteria. OLS contains microbes such as *Lactobacillus* sp., *Acetobacter* sp., *Saccharomyces* sp., *Aspergillus* sp., and *Streptomyces* sp. The OLS contains *Trichoderma* spp., widely used as a starter to the feed source and fermented plant on the growth of organic chemicals (Romero et al., 2016). According to Hu et al. (2008), Lactic acid bacteria increase rapidly during the first three days of fermentation and then slowly decrease until day 10 and, thereafter, the counts were maintained at around 6.7 log CFU/g during the duration of the fermentation period.

Table 1: Levels of dried and organic components of fermented banana stems (%)

Components	Treatment		
	EM4	OLS	Control
Dried Components	95.31 ± 0.35	94.34 ± 1.25	96.73 ± 0.76
Organic Components	79.84 ± 1.04	80.93 ± 0.23	81.02 ± 0.48

Effective microorganisms 4 (EM4); Organic liquid supplements (OLS). Results are presented as mean ± SD of triplicate.

Organic components in the treatment with the addition of EM4 and OLS starters drops lower than the control. This shows that during fermentation with the addition of EM4 and OLS, a breakdown of organic components in the banana stem occurs. EM4 and OLS contain microbes such as yeast and bacteria. The dominant microbes in EM4 are *Lactobacillus* spp., *Saccharomyces* spp., *Rhodo pseudomonas palustris*, *Actinomycetes* sp., *Aspergillus* sp. and *Penicillium* sp. (Higa and Parr, 2014). While the dominant microbes in OLS are *Lactobacillus* sp., *Acetobacter* sp., *Saccharomyces* sp., *Aspergillus* sp., and *Streptomyces* sp. These microbes break down macromolecules such as fiber in banana stems into simple compounds such as alcohol and volatile organic acids, which causes the reduction of organic components. Fermentation causes a decrease in carbohydrate concentration and an increase in the concentration of Klason lignin; the latter shows that some dried ingredients are lost during fermentation (Jorgensen et al., 2010). Controlled fermentation usually involves lactic acid bacteria, molds, yeasts and *Bacillus* as inoculums as they can achieve more predictable results under optimal conditions (Giraffa, 2004).

The composition of fermented banana stems

The results analysis of the chemical composition of the proximate banana stem can be seen in Table 2. There are differences in chemical composition between banana stems treated with EM4 starter, OLS, and control.

Table 2: Proximate analysis results of fermented banana stems (%)

Components	Treatments		
	EM4	OLS	Control
Protein	3.86 ± 0.30	3.68 ± 0.14	3.56 ± 0.50
Crude fiber	21.89 ± 1.61	25.34 ± 3.18	25.63 ± 0.91
Fat	3.26 ± 0.55	2.58 ± 58	2.29 ± 0.31
Ash	20.16 ± 1.04	19.07 ± 0.23	18.98 ± 0.72

EM4: Effective microorganisms 4; OLS: Organic liquid supplements. Results are presented as mean ± SD of triplicate.

Based on the obtained results shown, protein content in fermented banana stems with EM4 and OLS are higher than those without starter (control). This is because EM4 and OLS contain microbes that use free nitrogen in nature as a nitrogen source or stimulate the growth of microbes which utilizes free nitrogen. The crude protein, true protein, fat and ash contents increased significantly when the fermentation reached 96 hours (Aruna et al., 2017). Darwish et al., (2012) also reported that crude protein increased from 3.60% to 11.80% when *S. cerevisiae* was used as a biological additive after 7 days incubation at 28^{o}C.

Addition of EM4 and OLS can improve the fermentation process in banana stems (Table 3).

Table 3: Digestive value of fermented banana stems (%)

Components	Treatments		
	EM4	**OLS**	**Control**
DMD	53.93 ± 1.13	53.41 ± 4.87	24.94 ± 0.91
OMD	50.58 ± 1.03	49.14 ± 3.65	31.73 ± 0.78
NFE	46.15 ± 0.48	45.68 ± 1.24	30.97 ± 2.08
TDN	32.30 ± 4.15	28.36 ± 2.18	22.99 ± 0.64

DMD: Dry matter digestibility; OMD: Organic matter digestibility; NFE: Nitrogen free extract; TDN: Total digestible nutrient. Results are presented as mean ± SD of triplicate

Table 4: Sensory value of fermented banana stems

Components	Pretreatments		
	EM4	**OLS**	**Control**
Aroma	strong acids	strong acids	weak acids
Color	bright brown	bright brown	browns

EM4: Effective microorganisms 4; OLS: Organic liquid supplements

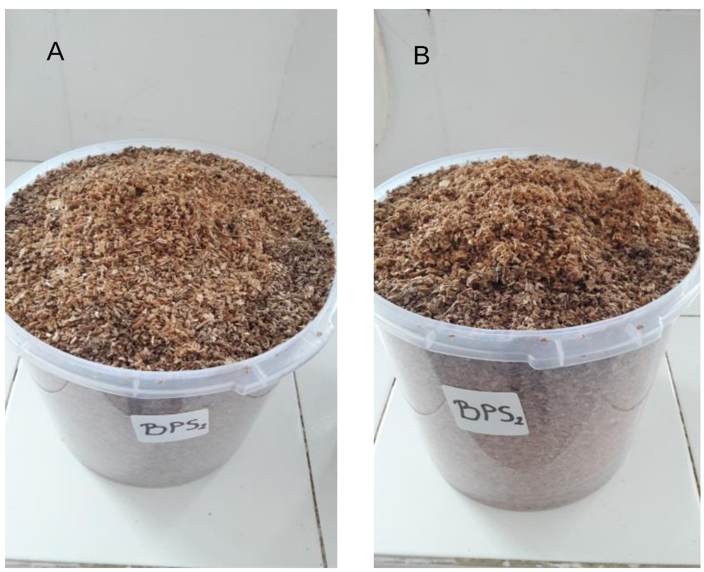

Figure 1: Cattle feed made from banana stems: (A) before fermentation, (B) after fermentation

The fermentation process causes the crude fiber component to decrease, this is caused by microbial activity that utilizes fiber as a carbon source. Microbes are able to break down fibers into simpler molecules. The formation of simpler molecules will facilitate absorption by livestock intestines to increase the digestibility of livestock. Fibers in plants are usually tightly tied to lignin, which causes the fiber to be unbreakable in the digestive system, thereby reducing digestibility. Fermentation increases the ileum digestibility of dried matter, organic matter and biomass energy from barley and wheat (Jørgensen et al., 2010). Besides that, Thalib (2008) reported that supplementing probiotic bacteria can reduce the number of pathogenic microbes in the digestive tract and help balance microbial consortium by optimizing the fermentation process.

Digestive value of banana stems

Dry matter digestibility (DMD) in fermented materials is higher than the control and between treatments, EM4 and OLS are no different. The digestion of dried components is a mixture of ingredients that can be digested by digestion, generally consisting of carbohydrate. The digestion of dried components as in vitro digestibility test. The results of the analysis of dried components digestibility of agricultural wastes are presented in Table 3.The digestibility rate of fermented and non-fermented dried wastes depends on the level of the components.

Sensory value of fermented banana stems

Observation of the aroma and color of fermented banana stems is obtained as shown in Table 4. Both starter treatments gave almost the same results but were different when compared with controls.

Fermented banana stems use effective microorganisms 4 (EM4) and organic liquid supplements (OLS) to produce a strong sour aroma. Without this aroma, the starter would be very weak. The dominant aroma is lactic acid. This shows that giving the starter EM4 and OLS improves the fermentation process. As it is known that EM4 and OLS contain lactic acid bacteria to make sure that lactic acid is the aroma that arises. Besides that, feed after fermentation would generally appear after a slightly darker than before fermentation, this is due to the process of breaking down large molecules into small molecules so that the appearance of the feed turns darker (Figure 1).

CONCLUSION

The results of this study conclude that the treatment of fermentation in banana stems with EM4 and OLS starters can improve feed digestibility and is better than control. Fermented banana stems also increase protein, fat and extract ingredients without nitrogen. EM4 starters provide better results compared to OLS. The results show that fermented banana pseudo-stems could be considered as an alternative of feeds or substitute of forage sources..

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

ACKNOWLEGEMENT

The authors are the grateful to Ministry of Research, Technology and Higher Education of the Republic of Indonesia that has funded this research through the Institutional National Strategic Research fund grant scheme which are channeled through Udayana University with the contract number: 415.70/UN14.4.A/PL/2017.

AUTHOR CONTRIBUTIONS

IDGMP conceived idea, conducted research, data collection, data interpretation, and write up. IBWG conducted literature search, writing the manuscript, and corresponding author. IDGATP designed research methodology, conducted the research, statistical analysis. All authors read and approved the final version.

REFERENCES

Al-Arif MA, Suwanti LT, Estoepangestie ATS, and Lamid M, 2017. The nutrients contents, dry matter digestibility, organic matter digestibility, total digestible nutrient, and NH_3 rumen production of three kinds of cattle feeding models, pp. 338–343. In Proceedings, The Veterinary Medicine International Conference 2017, KnE Life Sciences.

AOAC, 1990. Official methods of analysis. 16th ed. Association of official analytical chemists, Washington DC, USA.

Aruna TE, Aworh O, Raji A, Olagunju A, 2017. Protein enrichment of yam peels by fermentation with *Saccharomyces cerevisiae* (BY4743). Ann. Agric. Sci. J. 62: 33–37.

Bagheripour E, Rouzbehan Y, and Alipour D, 2008. Effects of ensiling, air-drying and addition of polyethylene glycol on *in vitro* gas production of pistachio by-products. Anim. Feed Sci. Tech. 146: 327–336.

Darwish GAMA, Bakr AA, Abdallah MMF, 2012. Nutritional value upgrading of maize stalk by using *Pleurotus ostreatus* and *Saccharomyces cerevisiae* in solid state fermentation, Ann. Agric. Sci. J. 57(1): 47–51

Fowler VR. 1975. The evaluation of feeds diets and additives by the pig producer, Proc. Of the British Society of Animal Production, 4: 11-18.

Ginting N, 2018. Comparison of isolate dadih with yeast dadih in improving nutrition quality of

Cassava Waste (CW), Proceedings, ICBBogor 2017, IOP Conf. Series: Earth Environ. Sci. 141: 01200.

Giraffa G, 2004. Studying of the dynamics of microbial populations during food fermentation. FEMS Microbiol. Rev. 28: 251–260.

Higa T, 1994. Personal communication. EM Technology Serving the Wold. 7 p.

Higa T, and Parr JF, 1994. Beneficial and effective microorganisms for a sustainable agriculture and environment. Atami, Japan: International Nature Farming Research Center.

Hu J, Lu W, Wang C, Zhu R, and Qiao J, 2008. Characteristics of solid-state fermented feed and its effects on performance and nutrient digestibility in growing-finishing pigs, Asian-Aust. J. Anim. Sci. 21(11): 1635–1641.

Hungate RE, 1988. The rumen and its microbes. Applied Science. Academic Press. New York, USA.

Isnawati, Ni'matuzahroh, and Surtiningsih T. Diversity of indigenous fungi d uring ruminant feed fermentation made of water hyacinth (*Eichhornia crassipes*) and corn (*Zea mays*) cob. Biosci. Res., 15(3): 2797-2801.

Jørgensen H, Sholly D. Pedersen AO, Canibe N, and Knudsen KEB, 2010. Fermentation of cereals - influence on digestibility of nutrients in growing pigs. Livest. Sci. 134 (1–3): 56–58.

Prayitno CH and Hidayat N, 2013, The efficacy of methanol extract of garlic (*Allium sativum*) to improve rumen fermentation products. Anim. Prod. 15(1):69–75.

Romero ES, Lopez LLMA, Porciuncula FL, Juico PP, and Galindez JL, 2016. Formulation and testing of combined organic liquid supplement from *Trichoderma* spp. and fermented plant and seed extracts on the growth of organic pechay. Ann. Trop. Res. 38(1): 53–64.

Santoso U, Nengsih F, Rozal A, Setianto J, and Kadarsih S, 2004. The effect of fermented feces on growth, fat deposition and carcass quality in broiler chickens. J. Indon. Trop. Anim. Agric. 29(1): 27–32.

Santosa B, Fitasari E, and Suliana G, 2017. Functional feed production contains three bioactive compounds from tofu dregs using effective microorganism-4 and *Lactobacillus plantarum,* Buana Sains. 17(1): 25–32.

Soekarto, ST, 1985. Organoleptic assessment, Bhratara Karya Aksara, Jakarta.

Thalib A, 2008. Utilization of probiotic-fermented rice straw as ruminant feed. Wartazoa 18(4): 198–206.

Wadhwa M, and Bakshi MPS, 2013. Utilization of fruit and vegetable wastes as livestock feed and as substrates for generation of other value-added products. RAP Publication (FAO), Ludhiana, India.

Wang CF, Muhammad AUR, Liu ZY, Huang BZ, and Cao BH, 2016. Effects of ensiling time on banana pseudo-stem silage chemical composition, fermentation and in sacco rumen degradation. J. Anim. Plant. Sci. 26 (2): 339–346.

Zega AD, Badarina I, and Hidayat, 2017. The nutritional quality of fermented beef cattle concentrate rations based on palm sludge and some local feed ingredients with bionac and EM 4. Indon. Anim. Sci. J., 12 (1): 38–46