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Scholars Journal of Agriculture and Veterinary Sciences Abbreviated Key Title: Sch J Agric Vet Sci ISSN 2348–8883 (Print) | ISSN 2348–1854 (Online) Journal homepage: https://saspublishers.com/sjavs/ Improving Rumen Fermentability and Fiber Fraction Digestion of Fermented Rice Straw with A Cattle Feed Supplement and A Concentrate Ramaiyulis*, Eva Yulia, Nelzi Fati, Salvia, Nilawati Animal husbandry study program, Agriculture Polytechnic Payakumbuh, 26271, Indonesia DOI: 10.36347/sjavs.2020.v07i02.003 | Received: 08.02.2020 | Accepted: 15.02.2020 | Published: 18.02.2020 *Corresponding author: Ramaiyulis Abstract Original Research Article This study aimed to obtain the best ration with the use of fermented rice straw (RF) as a basal feed for cattle farms. The RF was made by fermenting rice straw using Rhizopus oligosporus. The concentrate consisted of many locally available feed ingredients. The supplement was composed of several feeds as a multinutrient to supplement any deficiencies in the ration. There were 4 treatment rations: RF = 100% RF (control), RFS = RF + 10% supplement, RFSC = RFS + 10% concentrate, and RFSC2 = RFS + 20% concentrate. The rations were tested by in vitro digestion using bovine rumen fluid and 48 hours of incubation at 39 °C under anaerobic conditions. The results showed that the addition of the supplement significantly increased the digestibility of the dry matter, organic matter, crude protein, and hemicellulose, while the addition of the concentrate significantly increased the concentration of VFAs and the digestibility of NDF and cellulose. The best composition was an 80:10:10 (% DM) mixture of RF, supplement and concentrate. Keywords: fermented rice straw, supplement, concentrate, cattle ration, digestibility. INTRODUCTION The Indonesian government targets beef selfsufficiency in 2020 by stimulating domestic beef production which currently only reaches 68% [1]. Development of beef production through small-scale community farms is limited by the availability of grasslands and pasture fields for supplying forage

continuously. Extensively raising of cattle in the districts of Pesisir Selatan and Sijunjung, fulfilling forages for cattle by releasing cattle roaming the highways and house yards wich disturb farmers' horticultural gardens and leading to the pollution of the environment [2]. Therefore, it is necessary to develop alternative forage feed sources from local resources. Optimization of the use of rice straw agricultural waste has several factors limiting its use as forage. Rice straw mostly (> 60%) consists of cell walls composed of cellulose, hemicellulose, lignin, and silica [3]. The limiting factor of rice straw use as forage is its low nutritional value and digestibility; thus, it is unable to provide adequate nutrition for high-producing ruminants [4]. Rice straw contains high silica (12-16%) and lignin (6-7%) contents, which inhibit rumen microbial degradation during the digestive process [5]. Rice straw must undergo pretreatment before it is turned into animal feed. Pretreatment of rice straw with fungi is a practical, low-cost and environmentally friendly way to increase the nutritional value and straw digestibility [6]. Processing straw by fermentation 6can increase the nutritional value and digestibility of rice straw as a forage feed [7]. Rice 9straw can be used as a substitute forage feed, although it cannot be used as a complete ration, and its use must be supplemented with a concentrate [8]. Feed originating from local sources such as sago pith, bran, cassava, and coconut pulp, as a concentrate of inexpensive, valuable energy sources. The addition of concentrate to fermented rice straw is expected to provide a readily available carbohydrate source 4that can be used for microbial growth and digestive activity in the rumen [9]. The addition of concentrate with the cattle feed supplement reported by [10] can interact positively to increase the digestibility of low-quality forage. The cattle feed supplement is reported to be able to optimize the rate of microbial biomass production in the rumen [11] so that more microbes will participate in the digestion process. The effect of adding a concentrate and a supplement to a ration of fermented rice straw was hypothesized to improve the digestibility of fermented rice straw, but this needs to be investigated because there is currently little information available about this interaction. 10This study aimed to determine the ability of the cattle feed supplement and concentrate addition to improve the digestibility of fermented rice straw

in the rumen in vitro. The results of this study can be used as a basis for in vivo research to optimize the use of fermented rice straw as forage for beef cattle. MATERIALS AND METHODS Rations preparation Rice straw is a byproduct of rice (Oryza sativa) harvesting and, in this study, was of the Cisokan variety cultivated in Nagari Batu Balang, Harau subdistrict, Lima Puluh Kota district. Mature rice plants were cut ± 30 cm in depth from the clumps and then threshed to remove their grains. The rice straw was chopped with a chopper machine to a size of 1-2 cm. Then, it was mixed with 10% bran (dry basis) and inoculated with the fungus Rhizopus oligosporus using tempeh juice and incubated for 21 days under anaerobic conditions. The nutrient content of fermented rice straw is shown in Table 1. Table-1: Composition and nutrient content of the treatment ration Items Treatment Rations RF Supplement Concentrate RF RFS RFSC RFSC2 Ingredients (% DM) Fermented rice straw 100 90 80 70 - - - *Supplements - 10 10 10 - - - **Concentrates - - 10 20 - - - Chemical composition (% DM) Organic matters 87.06 87.18 87.89 88.59 87.06 88.26 94.12 Crude protein 9.82 11.17 11.19 11.21 9.82 23.31 11.64 Crude fiber 30.69 28.65 26.67 24.70 30.69 10.31 10.19 NDF 70.35 66.03 62.65 59.27 70.35 27.16 36.56 ADF 45.33 42.15 39.83 37.51 45.33 13.56 22.10 Cellulose 28.43 26.53 25.65 24.77 28.43 9.44 19.64 Hemicellulose 25.02 23.88 22.82 21.77 25.02 13.60 14.46 Lignins 8.99 8.09 7.19 6.29 8.99 0.82 0.96 Tannins - 0.05 0.05 0.05 - - - * Supplements: cattle feed supplement with 5% addition of gambier leaf residue (Ramaiyulis [11]), consist of brown sugar (15%), bran (27%), coconut meal (12%), soybean meal (15%), tapioca (15%), urea (5%), salt (3%), mineral (3%), and gambier leaf residue (5%). ** Concentrates: sago pith (30%), bran (30%), cassava (20%), and coconut pulp (20%). DM: dry matter; NDF: neutral detergent fiber; ADF: acid detergent fiber. RF: fermented rice straw; RFS: RF + 10% supplements; RFSC: RFS + 10% concentrate: RFSC2: RFS + 20% concentrate. The supplement was provided according to cattle feed supplement with a 5% addition of gambier leaf residue [11], consist of brown sugar (15%), bran (27%), coconut meal (12%), soybean meal (15%), tapioca (15%), urea (5%), salt (3%), mineral (3%), and gambier leaf residue (5%). The supplements were produced by heating brown sugar at a temperature of 90 °C until the sugar melted; then, it

was poured into a mixture of ingredients according to the composition in the formula. Dough was molded into pellets that were 5 mm in diameter with a pellet machine and dried in an oven at 60 °C. The concentrate consist of local feed ingredients, namely, sago pith (30%), bran (30%), cassava (20%), and coconut pulp (20%), is shown in Table 1. The fermented rice straw, cattle feed supplement, and concentrate were utilized in the combinations shown in Table 1 and were evaluated in 4 replications. In Vitro experiment An in vitro experiment was carried out following the method of [12] using the rumen fluid of Bali beef cattle (Bos sondaicus) obtained after animals were slaughtered in abattoirs. Rumen liquid was mixed with McDougall buffer solution consisting of 19.6 g NaHCO3, 7.42 g Na2HPO4·7H2O, 1.14 g KCl, 0.94 g NaCl, 0.24 g MgSO4·7H2O and 0.08 g CaCl2·2H2O per liter of distilled water at a ratio of 1:4 and a pH of 6.8. The allocation of the in vitro treatments followed a randomized complete block design with individual rumen fluid donors as blocks. A feed sample of 2.5 grams was put into an Erlenmeyer flask, 250 ml of a mixture of rumen fluid and McDougall buffer was added, and 2 Erlenmeyer flasks were made for each treatment unit. Then, the mixture was purged for 30 seconds with CO2 gas to create an anaerobic environment in the Erlenmeyers, which were then covered with a ventilated rubber cap. The Erlenmeyers were placed in a water-bath shaker (Precision, USA) and were incubated at 39 °C for 48 hours. Fermentation was stopped by immersing the Erlenmeyers in cold water. Parameter measurements After completing incubation, the pH of the rumen fluid was measured using a pH meter (Hi9807phep). Next, the samples were centrifuged (Hitachi CR21, Japan) at 3,000 rpm for 15 minutes at 4 °C. The supernatant was used for NH3 analysis by the Conway microdiffusion method and volatile fatty acids (VFA) analysis by the steam distillation method [13]. The 12residue was washed twice with distilled water, centrifuged again and then filtered using Whatman 41 filter papers before being dried in a 60 °C oven for 24 hours. Then, proximate analysis [13] was performed to determine the content 13of dry matter, organic matter, crude protein and crude fiber in the residue. Fiber fraction analysis was performed following the method of [14] to determine the contents of the neutral detergent fiber (NDF), acid 2detergent fiber (ADF), acid

detergent lignin (ADL), hemicellulose (NDF-ADF) and cellulose (ADF-ADL). NDF was determined by boiling 0.5 g of sample with 100 ml of a neutral detergent solution (NDS) for 1 hour. The NDS consisted of disodium ethylene diaminetetra acetate, sodium lauryl sulfate, sodium tetraborate, monoglycolether, sodium dihydrogenphosphate, and distilled water. Then, the sample was filtered in a glass crucible (coarse porosity 1), dried in a 105 °C oven and weighed. The ADF was determined in the same way but using an acid detergent solution (ADS), which consisted of cetyl trimethylammonium bromide, sulfuric acid, and distilled water. An addition of 72% H2SO4 was used to separate ADL. Statistical analysis Measured parameters were analyzed using Statistical Package for Social Science (SPSS, version 13.0, SPSS Inc., and Chicago, IL). Single-factor analysis of variance (ANOVA) was used to analyze data following a statistical model: $Yij = \mu + \tau i + \beta j + \varepsilon ij$ where Yij is the observed value, μ is the overall mean, τi is the effect of the different treatments, βj is the effect of the different blocks, and εij is the residual random error. If a significant effect was expressed at the probability level of $P < 0.05$, it was followed by Duncan's new multiple range test to determine the average value that was significantly different at $P < 0.05$. RESULTS AND DISCUSSION Rumen fermentability The in vitro rumen fermentability of fermented rice straw (RF) with the addition of a supplement and concentrate is shown in Table 2. The addition of the supplement to the RF did not significantly influence the VFA concentrations, but the addition of 10% the concentrate increased the VFA concentrations ($P < 0.05$). The VFA concentration of rumen fluid is closely related to the rate of rumen fermentation, especially the degradation of carbohydrates that produce carbon chains and protein degradation, which liberates carbon chains that are reflected in the VFA concentrations in rumen fluid [15]. The addition of concentrate consisting of raw materials of sago pith, bran, and cassava supplied soluble carbohydrates, thereby increasing the degradation of carbohydrates and increasing the concentration of VFAs in the rumen. VFAs are the most important source of metabolic energy in ruminants and supply 55-60% of the energy needed [16]. In addition, VFAs are needed by rumen microbes as a source of carbon chains for microbial protein synthesis [17]. The RFS ration produced the highest

NH3 concentration (P <0.01) compared to that of all other rations. NH3 is the end product of crude protein degradation and NH3 deamination in the rumen. The condensed tannin content in supplements acts as protein protection from rumen microbial degradation [18]. Tannin is a polyphenol compound that is capable of binding and precipitating proteins, thus protecting them from rumen microbial degradation, which causes a decrease in NH3 in the rumen [19]. Therefore, the production of NH3 in this study came from the breakdown of urea contained in supplements to NH3. Table-2: Concentrations of VFA, NH3, and pH in the rumen in vitro of fermented rice straw with the addition of supplements and concentrates Rumen Parameters Treatment Ration SE P-value RF RFS RFSC RFSC2 VFA, mM 130.00b 130.50b 142.50a 148.75a 8.09 0.028 NH3, mM 3.50b 5.17a 4.08b 4.33b 0.23 0.008 pH 6.99 6.99 6.98 6.92 0.02 0.051 a,b,c differences in superscripts indicate significantly different mean values VFA: volatille fatty acids; SE: standard error of mean: P: probability RF: 100% fermented rice straw; RFS: RF + 10% supplements; RFSC: RFS + 10% concentrate: RFSC2: RFS + 20% concentrate NH3 is the main nitrogen source used by rumen microbes to live and breed and to produce microbial proteins [20]. The effectiveness of NH3 use must be accompanied by the synchronization of NH3 concentrations with the availability of carbon chains [21]. The high NH3 content in the RFS treatment was due to the hydrolysis of urea to NH3 in the rumen occurring faster than its use by rumen microbes. The lack of availability of carbon framework sources causes slow use of NH3 by microbes. The low VFA concentration in the RFS treatment indicated the lack of availability of carbon frameworks that can be used by microbes in forming microbial proteins with NH3 as the amide group. The addition of a supplement and a concentrate did not affect the pH of the rumen fluid (P> 0.05). Rumen acidity indicates the condition of the rumen environment and ensures an optimal environment for the rumen microbes. Cellulolytic bacteria that ferment straw live in the rumen under neutral pH conditions and are sensitive to a low rumen pH [22]. A decrease in pH usually occurs because of the fast rate of fermentation of concentrate feed that is easily degraded, but the addition of concentrates up to 20% did not cause a decrease in the rumen pH. Nutrient digestion Table

3 shows the digestibility of nutrients in the in vitro rumen of the fermented rice straw with the addition of supplements and concentrates. The lowest digestibility of dry matter and organic matter was found in the RF rations (control) and increased with the addition of the supplement (RFS), and the highest digestibility was found with the addition of 20% concentrate (RFSC2) (P <0.01). The digestion of dry matter and organic matter in the rumen showed the percentage of nutrients available to livestock as a result of fermentation by microbes in the rumen. Rumen digestion constitutes 85% of the total digestion of nutrients in the digestive tract of ruminants [15]. Table-3. Digestion of rumen in vitro of fermented rice straw with the addition of supplements and concentrates Digestibility Parameters Treatment Ration SE P-value RF RFS RFSC RFSC2 DMD 26.28c 30.69b 34.18b 41.43a 0.01 0.003 OMD 28.52c 32.71b 34.77b 43.38a 0.02 0.008 CPD 44.98b 57.53a 61.42a 59.49a 0.04 0.048 CFD 24.19 24.08 22.86 24.82 0.02 0.520 a,b,c differences in superscripts indicate significantly different mean values DMD: dry matter digestibility; OMD: organic matter digestibility; CPD: crude protein digestibility; CFD: crude fiber digestibility; SE: standard error of means; P: probability RF: 100% fermented rice straw; RFS: RF + 10% supplements; RFSC: RFS + 10% concentrate: RFSC2: RFS + 20% concentrate Crude protein digestibility was found to be lowest in the control rations, and supplementation increased (P <0.05) the digestibility of crude protein, while the addition of the concentrate had no significant effect. The supplement contained easily degradable urea and carbohydrates as an available energy source, which allows for optimization of rumen microbial growth [11]. The protein content of the rations was relatively low, ranging between 9.82 and 11.21%, while the standard of SNI for fattening beef cattle ration contains at least 13% crude protein [23]. The addition of the supplement and concentrate on the fermented rice straw did not significantly affect the digestibility of crude fiber in the rumen (P> 0.05). The fermented rice straw exhibited high lignin content (8.99%) (Table 1), and lignin bonding with crude fiber inhibits the action of cellulase enzymes to digest crude fiber. The addition of the supplement and concentrate was not able to remove the limitation of crude fiber digestibility in the fermented rice straw. Fiber fraction digestion In Table 4, the in vitro

digestibility of the fiber fraction of the rations showed that the NDF and cellulose digestibility increased due to the addition of the concentrate (P <0.05), while the addition of the supplement had no significant effect. NDF showed the cell wall fraction content in the rations. Rice straw contains high amounts of silica (12-16%) compared to those in other agricultural wastes (only 3-5%) [5], and a high silica content inhibits the digestion of NDF. The addition of the supplement did not significantly influence the digestibility of NDF, while the addition of the concentrate was able to increase the digestibility of NDF. The concentrate contained a fast available energy source that could be used for microbial growth and activity in the rumen, increasing NDF digestion [24]. The physical, chemical and biological treatment of straw can break lignocellulosic bonds and increase the digestibility of NDF and cellulose in fermented rice straw [6]. Table-4: Digestion of fiber fractions of fermented rice straw with the addition of supplements and concentrates Fiber fraction Digestibility Treatment Ration SE P-value RF RFS RFSC RFSC2 NDF 13.31b 16.95b 20.95a 20.56a 0.02 0.013 ADF 21.89b 23.43b 24.86b 29.53a 0.02 0.040 Cellulose 34.26b 39.11b 45.47a 52.16a 0.02 0.004 Hemicellulose 23.91b 34.53a 36.45a 36.87a 0.04 0.014 a,b,c differences in superscripts indicate significantly different mean values 2NDF: neutral detergent fiber; ADF: acid detergent fiber; SE: standard error of means; P: probability RF: 100% fermented rice straw; RFS: RF + 10% supplements; RFSC: RFS + 10% concentrate: RFSC2: RFS + 20% concentrate The digestion of hemicellulose increased with the addition of the supplement (P <0.05) but was not affected by the addition of the concentrate. Hemicellulose is known to be more easily digested than other cell wall components due to its amorphous structure and lower polymerization levels [25]. Increased digestibility of hemicellulose with the addition of the supplement and increased digestibility of cellulose with the addition of the concentrate showed that both the supplement and concentrate played a synergistic role in increasing the 17digestibility of cell wall components as a source of energy for livestock. ADF digestibility only increased with the addition of 20% concentrate (P <0.05). The constituent components of ADF bind strongly to lignin, which makes ADF components difficult to penetrate by rumen microbial enzymes [26]. At the 20%

concentrate level, the starch content was increased, and the starch content in rations is known to be positively correlated with the ability of microbes to digest plant cell walls [27]. The addition of the supplement and concentrate decreased the fiber fraction content of the ration, thus increasing the content of food substances that were easily degraded in the ration. The availability of easily degraded food substances increases the ability of microbes to digest fiber fractions, thereby increasing the digestibility of fiber fractions [28].

CONCLUSIONS In vitro rumen digestibility of fermented rice straw can be improved by the addition of supplements and a concentrate. Supplements can increase the digestibility of dry matter, organic matter, crude protein, and hemicellulose, while the addition of concentrate can increase the digestibility of NDF and cellulose. The best ration composition with the use of fermented rice straw was an 80:10:10 ratios (% DM basis) of fermented rice straw, supplement, and concentrate.

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Sources

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