Effect of turmeric powder addition on *in vitro* rumen fermentation and digestibility

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Abstract. This study investigated the effects of adding turmeric powder on in vitro rumen fermentation characteristics and digestibility. The experimental diet comprised of 30% commercial concentrate and 70% Napier grass. The treatments were T0 (control) without turmeric powder and T1 with 2% turmeric powder inclusion. Each treatment was replicated six times. Parameters measured included rumen bacterial and protozoal populations, pH, volatile fatty acid (VFA) and ammonia (NH_3) concentrations, as well as the digestibility of dry matter (DM) and organic matter (OM). Statistical analyses were conducted using an independent sample t-test. The addition of 2% turmeric powder to the diet significantly elevated ($P<0.05$) the concentrations of total VFA and NH_3, as well as DM and OM digestibility. Conversely, the addition of 2% turmeric powder addition resulted in a significant decrease ($P<0.05$) in rumen pH but did not affect the total populations of rumen protozoa and bacteria. In conclusion, 2% turmeric powder could be an effective feed additive for enhancing rumen fermentation and digestibility.

1 Introduction

The demand for high-quality protein sources, including lamb, continues to rise in Indonesia, aligning with population growth, which has reached over 283 million by 2023. However, the sheep population in Indonesia has declined. Based on data from the Central Bureau of Statistics [1], the sheep population was recorded at 17,523,689 heads in 2020, 15,615,300 heads in 2021, and 14,063,214 heads in 2022. Low sheep productivity in Indonesia is primarily attributed to the hot and humid tropical climate, which induces heat stress, and to constraints in both feed availability and quality.

Feed quality, as indicated by its digestibility, plays a critical role in livestock productivity. High-quality feed with good digestibility supports optimal livestock performance. Digestibility is influenced by microbial activity in the rumen, where favourable conditions enhance fermentation processes and subsequently increase nutrient absorption [2]. Various additives can be incorporated to improve feed digestibility, including herbal additives such as turmeric powder.

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Turmeric (*Curcuma longa*), a rhizome plant, contains bioactive components, including essential oils (1.6%-3.9%) and curcumin (2.3%-8.4%) [3]. These compounds exhibit antioxidant, antiprotozoal, and anti-inflammatory properties, which enhance feed digestibility by inhibiting protozoal growth and promoting the proliferation of beneficial rumen bacteria. Turmeric is a potential source of natural antioxidants and can be utilized as a feed supplement [4].

Curcumin supplementation (300, 600, and 900 mg/kg) resulted in significantly increased daily weight gain but had no effect on feed intake and feed conversion ratio in sheep [5]. This study evaluated the effects of turmeric powder addition into the feed on rumen fermentation and the digestibility of feed in vitro

2 Materials and methods

2.1 Experimental diet

Two experimental diets consisting of a mixture of Napier grass and commercial concentrate with a ratio of 30:70 were prepared as follows:

- T0 (control): 30% Napier grass + 70% concentrate without turmeric.
- T1: 30% Napier grass + 70% concentrate + 2% turmeric powder.

There was no difference in the nutritional content between both dietary treatments except for the difference in turmeric addition. The nutrient composition of both feed ingredients was analyzed using a Buchi NIRFlex N-500 Solid Cell (Flawil, Switzerland). Turmeric powder was added at 2% of the concentrate weight in T1, based on previous experience using herbal additives such as betel leaves [6]. The nutritional contents of the feed used are presented in Table 1.

Table 1. Nutrient composition of Napier grass, concentrate and experimental diets.

Nutrient Composition (%)	Napier grass	Commercial concentrate	T0 (-turmeric)	T1 (+turmeric)
Dry Matter (DM)	19.9	88.6	67.99	67.99
Ash	11.7	5.8	7.57	7.57
Crude Protein (CP)	10.2	15.3	13.77	13.77
Ether extract (EE)	1.6	3.6	3.0	3.0
Crude Fiber (CF)	34.2	15.6	21.18	21.18
Nitrogen-Free Extract (NFE)*	42.3	59.6	54.41	54.41
Total Digestible Nutrients (TDN)**	51.4	70.6	64.84	64.84

*Calculated using: NFE = 100% - %Ash - %CP - %CF - %Fat
**TDN calculation based on [7].

2.2 Rumen fluid collection

Rumen fluid was obtained from three adult male sheep by using a stomach tube. The collected fluid was filtered through gauze and stored at 39°C in thermoses prefilled with water. The samples were immediately transported to the laboratory for *in vitro* fermentation and digestibility analysis. This process was repeated twice.

2.3 In vitro preparation

Napier grass was chopped to 2-3 cm in length and then oven-dried at 60°C for 24 h. The dried grass and commercial concentrate were finely ground into a mash and passed through a 1-mm sieve. McDougall's buffer solution was prepared by dissolving $NaHCO_3$ (9.8 g),

$Na_2HPO_4 \cdot 7H_2O$ (7 g), KCl (0.57 g), NaCl (0.47 g), $MgSO4 \cdot 7H_2O$ (0.12 g), and $CaCl_2$ (0.04 g) in 1 L of distilled water. Calcium chloride was added after the other components had dissolved, and the solution was saturated with CO_2 for 15 minutes. Supernatants were collected to analyze ammonia (NH_3), total volatile fatty acids (VFA), and bacterial and protozoal populations.

2.4 Measured variables

The rumen pH was measured using a calibrated pH meter. The ammonia concentrations were determined using the Conway microdiffusion method [8]. Total VFA concentrations were measured via steam distillation. The digestibility of dry matter (DM) and organic matter (OM) was evaluated following Tilley and Terry [9]. Protozoal populations were assessed following Ogimoto and Imai [10], whereas bacterial populations were determined using the spread plate method on nutrient agar.

2.5 Statistical analysis

Two treatments, i.e., T0 (without turmeric) and T1 (with 2% turmeric), and six replicates for each treatment were applied in this experiment. The data were examined using an independent sample t-test with SPSS 26.0 to compare the means of two independent groups and assess statistically significant differences.

3. Results and discussion

The effects of 2% turmeric powder addition on rumen fermentation and digestibility are presented in Table 2.

Table 2. Effects of turmeric powder addition on rumen fermentation and digestibility.

Parameter	T0	T1
Rumen pH	$7.59^a \pm 0.03$	$7.48^b \pm 0.02$
Total Bacteria (log CFU/mL)	9.52 ± 0.69	9.51 ± 0.54
Protozoa (log cells/mL)	6.13 ± 0.46	6.10 ± 0.37
NH_3 (mM)	$9.44^b \pm 0.91$	$10.68^a \pm 0.86$
Total VFA (mM)	$99.27^b \pm 3.44$	$111.34^a \pm 2.98$
DM Digestibility (%)	$65.81^b \pm 1.46$	$69.72^a \pm 1.01$
OM Digestibility (%)	$63.17^b \pm 1.46$	$67.05^a \pm 1.28$

T0: 30% Napier grass+ 70% concentrate (control), T1: Control + 2% turmeric powder, VFA: volatile fatty acid, DM: dry matter, OM: organic matter
Different superscripts within the same row differ significantly ($P<0.05$)

3.1 Rumen pH

The addition of 2% turmeric powder significantly ($P<0.05$) reduced rumen pH, which is consistent with previous findings by [5]. Despite this reduction, the pH remained within the optimal range (6.5–7.5), ensuring the maintenance of microbial activity and the fermentation processes [11].

The observed pH decrease is attributed to curcumin in turmeric, which enhances microbial activity, leading to increased VFAs production. Volatile fatty acids, including acetate, propionate, and butyrate, release hydrogen ions into the rumen fluid, lowering the pH [12]. This optimal pH range supports the growth of carbohydrate-fermenting bacteria, thereby ensuring efficient fermentation and nutrient utilization.

3.2 Total bacterial population

The addition of turmeric powder did not (P>0.05) affect the total bacterial population, which remained within the normal range (10^{10} - 10^{11} cells/mL; [13]).

The antibacterial properties of curcumin selectively promote cellulolytic and amylolytic bacteria while maintaining the overall microbial balance [5]. This finding underscores the compatibility of turmeric supplementation with the rumen's microbial ecosystem, as optimal pH conditions further support bacterial stability and activity. Similar results were reported where there was no change in the rumen bacterial population with the addition of tannin and saponin extracts [14].

3.3 Protozoal population

Protozoal populations, which account for 50% of rumen microbial biomass, remained unaffected (P>0.05) by turmeric powder supplementation, staying within the normal range (10^4 –10^6 cells/mL; [13]).

Curcumin's antiprotozoal properties likely target specific protozoal species without disrupting overall population dynamics. This balance supports bacterial activity and fermentation efficiency, as protozoa play a critical role in modulating bacterial populations and VFAs.

3.4 Ammonia (NH_3) concentration

The significant (P<0.05) increase in NH_3 concentration with turmeric powder supplementation highlights its role in enhancing proteolytic bacterial activity.

NH_3 serves as a key nitrogen source for microbial protein synthesis, supporting improved feed digestibility. The observed NH_3 levels (T0 = 9.44 and T1 = 10.68 mM/L) remained within the range (4.76 - 22.96 mM/L) reported by [15], indicating effective nitrogen utilization without adverse effects on rumen health. The influence of curcumin on protein breakdown further underscores its potential as a dietary additive to optimize rumen nitrogen metabolism. Similar results were reported in that ammonia concentration decreased with the addition of tannin and saponin extracts [14].

3.5 Total volatile fatty acids (VFAs)

Turmeric powder addition significantly (P<0.05) increased total VFA concentrations, which remained within the normal range (90.3 to 151.4 mM; [15]).

VFAs, the primary energy source for ruminants, are produced through carbohydrate fermentation by cellulolytic, hemicellulolytic, and amylolytic bacteria. The increased VFA levels observed in this study align with improved fiber digestion and microbial activity, demonstrating the role of turmeric in enhancing rumen fermentation efficiency.

3.6 Digestibility of dry matter (DMD) and organic matter (OMD)

Turmeric powder addition significantly (P<0.05) improved both DMD and OMD, surpassing the 60% threshold indicative of high-quality feed.

The role of curcumin in optimizing microbial activity and enzyme secretion likely contributed to these improvements. Enhanced digestibility aligns with increased VFA production and nitrogen utilization, further validating the efficacy of turmeric as a feed additive.

4 Conclusion

The addition of 2% turmeric powder to sheep diets did not affect rumen protozoal or bacterial populations but significantly reduced rumen pH and increased concentrations of NH_3 and total VFAs, as well as dry and organic matter digestibility. All changes remained within the normal range, indicating no negative impact on rumen function.

References

1. [BPS] Central Bureau of Statistics, Sheep Population by Province (Heads), 2020-2022. Available at: https://www.bps.go.id/en/statistics-table/2/NDczIzI=/populasi-domba-menurut%20provinsi.html (2024).
2. A.R. Castillo-González, M.E. Burrola-Barraza, J. Domínguez-Viveros, A. Chávez-Martínez, Rumen microorganisms and fermentation. Arch. Med. Vet. **46**, 3: 349-361 (2014).
3. A. Kumari, C. Prasad, R. Kumar, Essential oil and curcumin content in different varieties of turmeric (*Curcuma longa* L.). The Pharma Innovation Journal. **11**, 841 (2022).
4. E. Lukitaningsih, A. Rohman, M. Rafi, A. Fadzillah, Nurrulhidayah, A. Windarsih, In vivo antioxidant activities of Curcuma longa and Curcuma xanthorrhiza: a review. Food Research. **4**, 1: 13-19 (2019). https://doi.org/10.26656/fr.2017.4(1).172
5. G. Tian, X. Zhang, X. Hao, J. Zhang, Effects of curcumin on growth performance, ruminal fermentation rumen microbial protein synthesis and serum antioxidant capacity in housed growing lambs. Animals. **13**, 9 (2023). https://doi.org/10.3390/ani13091439
6. A.A. Yamin, A. Sudarman, D. Evvyernie, *In vitro* rumen fermentation and anti mastitis bacterial activity of diet containing betel leaf meal (*Piper betle L.*). Media Peternakan. **36**, 2: 137-142 (2013). https://doi.org/10.5398/medpet.2013.36.2.137
7. M.F. Wardeh, Model for estimating energy and protein utilization for feeds, Dissertation, Utah State University, Utah, United State of America (1981). https://doi.org/10.26076/9026-5aad
8. General Laboratory Procedure, Report of Dairy Science, (University of Wisconsin, Madison, 1966).
9. J.M.A. Tilley, R.A. Terry, A two stage technique for in the in vitro digestion of forage crops. Grass and Forage Science. **18**, 2: 104 (1963). https://doi.org/10.1111/j.1365-2494.1963.tb00335.x
10. K. Ogimoto, S. Imai, Atlas of Rumen Microbiology, (Japan Scientific Societies Press, Tokyo, 1981).
11. M. Bonnet, J.C. Lagier, D. Raoult, S. Khelaifia, Bacterial culture through selective and non-selective conditions: the evolution of culture media in clinical microbiology. New Microbes and New Infections. **34**: 100622 (2020). https://doi.org/10.1016/j.nmni.2019.100622
12. L. Chen, Y Shen, C. Wang, L. Ding, F. Zhao, M. Wang, J. Fu, H. Wang, *Megasphaera elsdenii* lactate degradation pattern shifts in rumen acidosis models. Sec. Microbial Physiology and Metabolism. **10**: 162 (2019). https://doi.org/10.3389/fmicb.2019.00162
13. D.N. Kamra, Rumen microbial ecosystem. Current Sci. **89**, 1: 124-135 (2005).
14. A. Jayanegara, Y. Yogianto, E. Wina, A. Sudarman, M. Kondo, T. Obitsu, M. Kreuzer, Combination effects of plant extracts rich in tannins and saponins as feed additives for mitigating in vitro ruminal methane and ammonia formation. Animals. **10**, 9 (2020). https://doi.org/10.3390/ani10091531
15. F. Bargo, L.D. Muller, E.S. Kolver, J.E. Delahoy, Invited Review: Production and digestion of supplemented dairy cows on pasture. J. Dairy Sci. **86**, 1: 1-42 (2003). https://doi.org/10.3168/jds.S0022-0302(03)73581-4

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