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Research Article

Interaction of Palm Fatty Acid Distillate (PFAD) and High-Tannin Mangrove Fruit Extract (*Sonneratia alba*) on the In Vitro Digestibility of Fiber Fractions in Dairy Cattle Feed

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ABSTRACT

This study aimed to evaluate the interaction between Palm Fatty Acid Distillate (PFAD) and high-tannin mangrove fruit extract (*Sonneratia alba*) on the in vitro digestibility of dairy cattle feed fiber fractions. The experimental design used a completely randomized factorial 3×4 pattern with three replicates. The first factor was the PFAD level (0%, 3%, 6%) and the second factor was the mangrove fruit extract dose (0%, 0.5%, 1%, 2%). Fermentation was carried out using the Tilley and Terry (1963) rumen stage method using fresh goat rumen fluid. The parameters observed included the digestibility of neutral detergent fiber (NDF), acid detergent fiber (ADF), hemicellulose, and cellulose. The results showed that PFAD, mangrove fruit extract, and their interaction had a significant effect ($P<0.05$) on the digestibility of NDF and cellulose, but did not have a significant effect on ADF and hemicellulose. The combination of 3% PFAD and 0.5–1% extract provided the highest digestibility. Increasing PFAD and tannin reduced digestibility due to the fat coating effect and the formation of tannin complexes with structural carbohydrates. The combination of PFAD and mangrove tannin can be optimized as a functional feed additive based on local resources that improves rumen fermentation efficiency without reducing fiber digestibility.

Keywords: Dairy cattle, fiber digestibility, in vitro, PFAD, *Sonneratia alba*, tannin

INTRODUCTION

Feed quality is an important factor that determines the productivity of ruminant livestock. One innovative approach to improving rumen fermentation efficiency is to utilize natural bioactive compounds such as tannins, which can suppress methanogenesis and increase energy efficiency (Aboagye & Beauchemin, 2019; Besharati et al., 2022). However, the effects of tannins are highly dependent on their dosage and chemical structure. High concentrations can lead to reduced fiber intake and digestibility (Elihasridas et al., 2024; Chen et al., 2021).

Sonneratia alba mangrove fruit extract is known to contain high tannins (>70%) and has the potential as an alternative feed ingredient to reduce methane emissions without compromising livestock performance (Pazla et al., 2025a; Elihasridas et al., 2025). Previous studies have shown

that supplementation with 1–2% mangrove extract can reduce protozoa and methane populations by 25–30% (Elihasridas et al., 2025; Jamarun et al., 2025). However, increasing the tannin dose above the optimal threshold can suppress cellulolytic microbial activity and reduce NDF digestibility (Vasta et al., 2009; Getachew et al., 2008).

Meanwhile, Palm Fatty Acid Distillate (PFAD) is a high-energy source rich in saturated and unsaturated fatty acids, and has protective properties for feed nutrients (Pazla et al., 2025b). PFAD can form a hydrophobic layer on feed particles, reducing rapid degradation by microbes, and can be used as an encapsulation matrix for bioactive compounds such as catechins or tannins (Min & Solaiman, 2018; Pazla et al., 2025b). The interaction between PFAD and plant tannins is expected to produce a synergistic effect: PFAD stabilizes rumen conditions through energy supply, while tannins suppress protozoa and methanogen populations (Elihasridas et al., 2023; Carreño et al., 2015). However, both also have the potential to be antagonistic if the dosage is excessive, causing a decrease in fiber fraction digestibility due to a decrease in cellulolytic enzyme activity (Guerreiro et al., 2021; Ardani et al., 2024). Therefore, this study aims to evaluate the effects of the interaction between PFAD and high-tannin mangrove fruit extract on the digestibility of fiber fractions (NDF, ADF, hemicellulose, and cellulose) in vitro to find an optimal combination that is efficient and environmentally friendly.

MATERIAL AND METHODS

Research Design

This study was conducted to evaluate the interaction between Palm Fatty Acid Distillate (PFAD) and high-tannin mangrove fruit extract (*Sonneratia alba*) on the in vitro rumen digestibility of fiber fractions (NDF, ADF, hemicellulose, and cellulose). The design used was a 3×4 factorial complete randomized design (CRD) with three replicates. The first factor (A) was the PFAD level: A1 = 0% PFAD A2 = 3% PFAD A3 = 6% PFAD The second factor (B) was the dose of mangrove fruit extract: B1 = 0% B2 = 0.5% B3 = 1% B4 = 2%. There were a total of 12 treatment combinations with three replicates, resulting in 36 experimental units.

Preparation of Materials

The basic ration consists of common dairy cattle feed ingredients (Tables 1, 2, and 3). PFAD was obtained from the by-products of palm oil refining in Medan, North Sumatra. At the same time, mangrove fruit extract (*Sonneratia alba*) was collected from the coastal area of Pasaman, West Sumatra. Mangrove fruit extraction was carried out using the method of Elihasridas et al. (2025). The total tannin content of the extract obtained was 71.43%. PFAD was used in a homogeneous liquid form, without saponification, to simulate free fat in the ration.

Table 1. Nutrient Contents

Nutrient content	Feed Ingredient				
	Rice Straw	Indigofera	Corn	Bran	Tofu Dregs
Dry matter	77.62	88.10	85.55	91.05	19.85
Organic matter	88.60	92.41	93.42	98.22	95.38
Crude protein	5.65	23.98	13.58	10.05	21.91
Crude fat	1.86	3.64	3.99	2.69	9.49
Crude fiber	29.89	9.95	10.29	9.04	17.57
Ash	11.4	7.59	6.58	1.78	4.63

Nutrient content	Feed Ingredient				
	Rice Straw	Indigofera	Corn	Bran	Tofu Dregs
Nitrogen free extract	31.66	41.66	68.38	60.83	46.41
TDN	51.67	75.47	83.29	78.95	78.98
NDF	61.80	35.14	58.62	48.11	44.60
ADF	48.07	21.54	34.66	23.08	27.37
Cellulose	26.77	17.79	19.70	11.05	25.67
Hemicellulose	15.29	13.60	23.96	25.03	17.28
Lignin	10.11	2.9	7.50	3.78	5.95
Silica	8.3	0.71	0.70	2.28	-

TDN = Total Digestibility Nitrogen; NDF = Neutral Detergent Fiber; ADF = Acid Detergent Fiber

Table 2. Ration Composition

Feed Ingredient	Treatment (%)
Rice Straw	42
Indigofera	28
Corn	12
Bran	12
Tofu Dregs	5
Minerals	1
Total	100

Tabel 3. Ration Nutrient Content

Nutrient content	Value of content (%)
Dry matter	80.50
Organic matter	90.90
Crude protein	13.00
Crude Fat	18.50
Ash	3.10
Crude Fiber	8.10
Nitrogen Free Extract	42.80
TDN	66.30
NDF	50.80
ADF	34.50
Cellulose	21.20
Hemicellulose	17.00
Lignin	6.70
Silica	4.03

TDN = Total Digestibility Nitrogen; NDF = Neutral Detergent Fiber; ADF = Acid Detergent Fiber.

In Vitro Procedure

The in vitro test was conducted based on the Tilley and Terry (1963) method for the rumen stage only (without the pepsin-acid enzyme stage). Rumen fluid was obtained from adult goats that had just been slaughtered at the slaughterhouse. The rumen fluid was immediately filtered using four layers of gauze and stored in a thermos containing CO₂ to maintain anaerobic conditions and a temperature of 39 °C. A total of 2.5 g of ration samples (based on treatment

combinations) were placed in 250 mL fermentation tubes filled with 2000 mL of McDougall buffer solution and 50 mL of rumen fluid (4:1 ratio). The tubes were tightly sealed with perforated rubber and incubated for 48 hours at 39 °C in a shaker incubator. After incubation, the process was stopped by cooling with ice, followed by filtration and drying of the residue for fiber fraction analysis.

Analysis was performed on the raw material and fermentation residue to determine Neutral Detergent Fiber (NDF) Acid Detergent Fiber (ADF) Acid Detergent Lignin (ADL). The analysis method used the procedure described by Van Soest et al. (1991).

Statistical Analysis

The data were analyzed using two-way analysis of variance (ANOVA) for factorial designs according to the model:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \varepsilon_{ijk}$$

Note:

Y_{ijk}	= observation value
μ	= general average
α_i	= effect of PFAD i
β_j	= effect of extract dose j
$(\alpha\beta)_{ij}$	= interaction of PFAD times extract
ε_{ijk}	= experimental error

Further testing was conducted using Duncan's Multiple Range Test (DMRT) at a significance level of 5% using SPSS 26.0 software. Data are presented as mean $\pm$ standard error (SEM).

RESULTS AND DISCUSSION

PFAD, mangrove fruit extract, and their interaction had a significant effect ($P < 0.05$) on NDF and cellulose digestibility, but had no effect on ADF and hemicellulose (Table 4). NDF values decreased from 55.82 (control) to 54.93% in the 6% PFAD + 2% tannin combination. Cellulose also decreased from 59.0 to 57.5%. This decrease occurred due to the presence of a PFAD lipid layer that inhibited microbial adhesion to fiber particles (Pazla et al., 2025b) and the formation of tannin–protein and tannin–polysaccharide complexes that were difficult to degrade (Elihasridas et al., 2025; Pazla et al., 2025a).

In addition, tannins can also inactivate cellulase enzymes through the binding of phenolic groups to enzyme proteins (Besharati et al., 2022; Getachew et al., 2008). The combination of 3% PFAD with 0.5–1% extract provided the highest NDF and cellulose digestibility (~58%), indicating a balance between additional energy and moderate tannin effects (Min & Solaiman, 2018; Dalby et al., 2020). This combination also suppresses protozoa populations without reducing fermentation efficiency (Elihasridas et al., 2024; Vasta et al., 2009).

The absence of significant differences in ADF and hemicellulose indicates that these two fractions are relatively stable to changes in rumen microflora. The ADF fraction consists of lignocellulose that is difficult to break down even under normal fermentation conditions (Carreño et al., 2015). Hemicellulose, which is more easily digestible, can still be hydrolyzed by non-cellulolytic bacteria such as *Butyrivibrio fibrisolvens* (Guerreiro et al., 2021).

In general, these results support the research of Jamarun et al. (2025), who reported that natural tannins from mangrove fruits can be utilized within safe limits without reducing the digestibility of fiber fractions. The synergistic effect of PFAD–tannin is also in line with the results

of Pazla et al. (2025b), which showed that PFAD-CaO can be used as a delivery system for polyphenolic compounds to maintain their activity in the rumen. Thus, PFAD and mangrove tannins have the potential to be used together as functional feed additives to improve fermentation efficiency and support sustainable green livestock nutrition (Elihasridas et al., 2024; Ardani et al., 2024).

Table 4. Fiber Fraction Digestibility (ADF, NDF, Hemicellulose, Cellulose)

	Factor A	Factor B				Average
		B1	B2	B3	B4	
Hemicellulose	A1	63.04±4.05	60.42±0.37	62.12±1.09	59.34±1.35	61.23
	A2	60.44±1.26	59.59±3.83	59.51±4.07	59.27±2.64	59.60
	A3	59.15±1.82	60.76±4.53	57.88±1.52	57.71±0.63	58.87
	Average	60.74	60.26	59.84	58.77	SE = 0.77
	Factor A	Factor B				Average
		B1	B2	B3	B4	
Cellulose	A1	60.48 ^l ±0.49	59.11 ^{ijk} ±0.17	58.23 ^{efgh} ±0.50	58.20 ^{efg} ±0.62	59.01 ^b
	A2	58.14 ^{ef} ±0.45	57.11 ^{bc} ±0.68	57.04 ^b ±0.09	58.57 ^{fghij} ±0.45	57.71 ^a
	A3	58.36 ^{efghi} ±0.79	57.80 ^{cde} ±0.62	57.31 ^{bcd} ±0.31	56.34 ^a ±1.10	57.45 ^a
	Average	58.99 ^c	58.01 ^{ab}	57.53 ^a	57.70 ^{ab}	SE = 0.17
	Factor A	Factor B				Average
		B1	B2	B3	B4	
ADF	A1	54.32±1.11	54.16±0.30	53.82±0.46	53.74±0.48	54.01
	A2	54.52±1.02	54.20±0.47	53.93±0.70	55.49±0.16	54.29
	A3	54.43±0.62	54.27±0.67	53.93±0.48	53.86±0.73	54.12
	Average	54.42	54.21	53.89	54.03	SE = 0.19
	Factor A	Factor B				Average
		B1	B2	B3	B4	
NDF	A1	56.64 ^b ±0.72	55.65 ^{fgh} ±0.26	55.99 ^{fgh} ±0.67	55.00 ^{abcde} ±0.70	55.82 ^b
	A2	55.77 ^{defgh} ±0.41	55.40 ^{bcdefg} ±1.25	55.19 ^{bcdef} ±0.94	54.96 ^{abcd} ±0.47	55.33 ^{ab}
	A3	54.17 ^{abc} ±0.48	55.84 ^{defgh} ±1.16	54.65 ^{ab} ±0.58	54.54 ^a ±0.32	54.93 ^a
	Average	55.70	55.63	55.27	54.84	SE = 0.21

Note: Superscripts indicate a significant effect ($P<0.05$) A1: PFAD 0%; A2: PFAD 3%; A3: PFAD 6%; and B1: mangrove fruit extract 0%; B2: mangrove fruit extract 0.5%; B3: mangrove fruit extract 1%; B4: mangrove fruit extract 2%

CONCLUSIONS

The combination of PFAD and high-tannin mangrove fruit extract showed a significant interaction on the in vitro digestibility of fiber fractions. A PFAD level of 3% with 0.5–1% extract provided the best results for NDF and cellulose digestibility without reducing ADF and hemicellulose. The use of this combination is recommended as an efficient, environmentally friendly, and locally sourced functional feed formulation strategy.

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