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Effects of Concentrate Level and Fermentation Time on The Protein Content of Water Hyacinth-Based Livestock Feed (*Eichhornia crassipes*)

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Abstract: This study analyzed the effects of concentrate level and fermentation time on the protein content of water hyacinth-based livestock feed. The study was initiated in February 2024 at the Chemistry Laboratory of Universitas Pendidikan Mandalika and employed a laboratory experiment with a 4 × 2 factorial Completely Randomized Design (CRD). The first factor was concentrate level: 0%, 30% (300 g), 35% (350 g), and 40% (400 g), each mixed with 1 kg of water hyacinth; the second factor was fermentation time: 7 and 14 days. Each treatment combination was replicated six times, yielding 48 experimental units. Protein content was determined using the Kjeldahl method. The data were analyzed descriptively, tested for normality and homogeneity, and subsequently analyzed using two-way ANOVA followed by the Tukey HSD post hoc test. Mean protein contents after 7 days of fermentation were 0.22%, 0.30%, 0.33%, and 0.42%, respectively, whereas the corresponding means after 14 days were 0.23%, 0.24%, 0.25%, and 0.30%. ANOVA revealed significant effects of concentrate level, fermentation time, and their interaction on protein content ($p < .001$). The best-performing combination was the 40% concentrate treatment fermented for 7 days, which produced a mean protein content of 0.42%.

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INTRODUCTION

Water hyacinth (*Eichhornia crassipes*) is an invasive aquatic plant that can disrupt the balance of aquatic ecosystems. Sindhu et al. (2017) described water hyacinth as a harmful freshwater weed in many regions because of its rapid growth and its capacity to deplete nutrients and oxygen from water bodies, thereby adversely affecting aquatic vegetation and biota. This plant commonly grows in swamps, reservoirs, and slow-flowing rivers (Nuryana, 2016). Uncontrolled water hyacinth growth can accelerate sediment accumulation, restrict oxygen diffusion into the water, threaten biodiversity, and damage aquatic ecosystems (Sudana & Mohamad, 2021).

Water hyacinth also has potential as an alternative livestock-feed resource. Its nutritional constituents, including protein, create opportunities to diversify feed ingredients and strengthen feed security. However, its utilization remains constrained by its nutrient composition, high crude-fiber content, and antinutritional compounds, which may reduce digestibility and nutrient availability for livestock (Suryani et al., 2016). Appropriate processing is therefore required to improve the quality of water hyacinth-based feed.

Fermentation is one approach that can modify the structure of crude fiber, increase nutrient availability, and reduce undesirable components. Bahrun (2010) reported that

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fermentation can improve the nutritional quality of water hyacinth, including its protein, fiber, and ash contents. Fermentation outcomes are influenced by ingredient composition, particularly the proportion of concentrate, as well as fermentation duration. Concentrate supplies additional energy and protein, whereas fermentation determines the duration of microbial activity in degrading the substrate.

Fermented feed also offers practical benefits because it can improve nutrient content, reduce odors associated with livestock and their surroundings, enhance palatability, and extend shelf life, thereby supporting feed availability during the dry season (Kusmiah et al., 2021). Nevertheless, the combination of concentrate level and fermentation time that produces the highest protein content in water hyacinth-based feed requires further investigation through controlled experiments.

This study aimed to analyze the effects of concentrate level, fermentation time, and their interaction on the protein content of water hyacinth-based livestock feed. The findings are expected to provide practical guidance for farmers and other stakeholders seeking to optimize the use of water hyacinth as an alternative feed ingredient. They may also serve as enrichment material for laboratory manuals used by school and university students.

RESEARCH METHODS

Study Design, Period, and Location

This quantitative study employed a laboratory experimental method with a 4 × 2 factorial Completely Randomized Design (CRD). The first factor was concentrate level (0%, 30%, 35%, and 40%), and the second factor was fermentation time (7 and 14 days). Each treatment combination was replicated six times, resulting in 48 experimental units. The concentrate percentages were adapted from the treatments used by Ratnaningtyas et al. (2019). The study began in February 2024 and was conducted at the Chemistry Laboratory of Universitas Pendidikan Mandalika.

Table 1. Experimental Treatment Groups Based on Concentrate Inclusion Level

Treatment Group	Concentrate Level (%)	Description
P1	0	Control group, without concentrate
P2	30	Diet containing 30% concentrate
P3	35	Diet containing 35% concentrate
P4	40	Diet containing 40% concentrate

Note: P1 served as the control treatment without concentrate supplementation.

Berikut format tabel yang lebih jelas dan konsisten untuk naskah ilmiah:

Table 2. Randomized Allocation of Experimental Units and Observation Schedule

Concentrate Level (%)	Fermentation Time (days)	Replicate 1	Replicate 2	Replicate 3	Replicate 4	Replicate 5	Replicate 6
0	7	P1U5-H7	P1U3-H7	P1U6-H7	P1U1-H7	P1U2-H7	P1U4-H7
0	14	P1U3-H14	P1U4-H14	P1U1-H14	P1U2-H14	P1U6-H14	P1U5-H14
30	7	P2U5-H7	P2U1-H7	P2U3-H7	P2U4-H7	P2U2-H7	P2U6-H7

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Concentrate Level (%)	Fermentation Time (days)	Replicate 1	Replicate 2	Replicate 3	Replicate 4	Replicate 5	Replicate 6
30	14	P2U3-H14	P2U4-H14	P2U1-H14	P2U2-H14	P2U6-H14	P2U5-H14
35	7	P3U2-H7	P3U5-H7	P3U4-H7	P3U6-H7	P3U1-H7	P3U3-H7
35	14	P3U3-H14	P3U6-H14	P3U2-H14	P3U1-H14	P3U4-H14	P3U5-H14
40	7	P4U6-H7	P4U1-H7	P4U5-H7	P4U2-H7	P4U3-H7	P4U4-H7
40	14	P4U4-H14	P4U2-H14	P4U6-H14	P4U1-H14	P4U5-H14	P4U3-H14

Note. P denotes the concentrate treatment level: P1 = 0% concentrate (control), P2 = 30% concentrate, P3 = 35% concentrate, and P4 = 40% concentrate. U denotes the experimental-unit replicate, numbered U1–U6, while H denotes the fermentation or observation time: H7 = day 7 and H14 = day 14. The order of experimental units within each treatment and fermentation time was determined through randomization.

Table 3. Description of Treatment Codes and Sample Composition

Treatment Code	Concentrate Level	Sample Composition	Total Sample Weight (g)	Fermentation Time
P1U1	0% (control)	1,000 g of water hyacinth without concentrate	1,000	7 or 14 days
P2U1	30%	1,000 g of water hyacinth + 300 g of concentrate	1,300	7 or 14 days
P3U1	35%	1,000 g of water hyacinth + 350 g of concentrate	1,350	7 or 14 days
P4U1	40%	1,000 g of water hyacinth + 400 g of concentrate	1,400	7 or 14 days

Note. P denotes the treatment level: P1 = 0% concentrate or control, P2 = 30% concentrate, P3 = 35% concentrate, and P4 = 40% concentrate. U denotes the experimental-unit replicate; therefore, U1 represents replicate 1, while the other replicates are coded U2–U6. Fermentation time is indicated by an additional code: H7 for 7 days and H14 for 14 days. For example, P2U1-H7 refers to replicate 1 of the 30% concentrate treatment fermented for 7 days.

Population and Samples

The study population consisted of water hyacinth (*Eichhornia crassipes*) collected from Bual Reservoir, Gerantung Urban Village, Central Praya District. The samples consisted of water hyacinth fermented with different concentrate levels and fermentation times.

Equipment and Materials

The research instruments comprised the equipment and materials used to prepare water hyacinth-based livestock feed and determine its protein content. Details of the equipment and materials are presented in Tables 4 and 5.

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Table 4. Equipment Used in The Study

No.	Equipment	Function
1	Bucket	Used to hold feed during fermentation after it had been mixed with concentrate.
2	Knife	Used to cut water hyacinth into smaller pieces to facilitate digestion by livestock.
3	Transparent plastic	Used to wrap the water hyacinth.
4	Analog scale	Used to weigh the water hyacinth.
5	Sack	Used to weigh the samples.
6	Volumetric flask	Used as a base when mixing water hyacinth with concentrate.
7	Glass funnel	Used to collect the filtered distillate before dilution.
8	Tarpaulin	Used to facilitate the transfer of solutions into a flask.
9	Kjeldahl flask	Used as a base for drying and chopping water hyacinth.
10	Volumetric pipette	Used for sample digestion.
11	Spatula	Used to transfer a measured volume of solution.
12	Oven	Used to transfer the ground sample.
13	Blender	Used to dry the samples.
14	Smartphone camera	Used to grind the dried samples.
15		Used to document all research activities.

Table 5. Materials Used in The Study

No.	Material	Function
1	Water hyacinth (*Eichhornia crassipes*)	Primary material used to prepare livestock feed.
2	Concentrate	Additional source of energy and protein for livestock.
3	30% H_2O_2	Used to support oxidation during the digestion process.
4	Distilled water	Used to dilute the samples.
5	Concentrated H_2SO_4	Used to digest the samples.
6	50% NaOH	Used to release ammonia during distillation.
7	0.1 N HCl	Used to capture ammonia produced during distillation.
8	Phenolphthalein indicator (PP)	Used to indicate the color change during titration.

Feed Preparation Procedure

1) Water hyacinth samples were collected from the field.
2) The water hyacinth was chopped into 3–4 cm pieces to facilitate digestion by livestock.
3) The chopped water hyacinth was dried for one day and one night to reduce its moisture content, extend its shelf life, and decrease the risk of spoilage. Drying under direct sunlight has been reported not to cause meaningful changes in the chemical composition of the material (Yanuartono et al., 2017).
4) One kilogram of water hyacinth was weighed for each experimental unit. This procedure was repeated 48 times; therefore, a total of 48 kg of water hyacinth was used.
5) SG 801 concentrate was weighed at 300 g, 350 g, and 400 g according to the assigned treatment.
6) The water hyacinth was mixed with concentrate according to the treatment group.
7) Each mixture was placed in a fermentation container.

8) The mixtures were fermented for 7 or 14 days.

Determination of Protein Content Using the Kjeldahl Method

Protein content was determined through nitrogen analysis using the Kjeldahl method. This method analyzes protein-containing materials by converting organic nitrogen into an inorganic form through three stages: digestion, distillation, and titration (Purba, 2019). Before analysis, the samples were dried in an oven at 125 °C for 1 hour, ground in a blender, sieved, and weighed to obtain 0.1-g portions.

The digestion stage decomposes organic compounds so that they can be analyzed. The addition of concentrated sulfuric acid accelerates the oxidation of carbon and hydrogen into CO, CO_2, and H_2O, while nitrogen is converted into $(NH_4)_2SO_4$ (Hulyadi, 2020; Wahyuni, 2017). During distillation, components are separated on the basis of differences in vapor pressure. The addition of NaOH converts $(NH_4)_2SO_4$ into NH_3, after which the ammonia is captured in a standard HCl solution (Ganardy, 2022; Wahyuni, 2017). The titration stage determines the concentration of the analyte from the volume of titrant required to reach the equivalence point (Indrajaya et al., 2021).

Digestion Procedure

1) A 0.1-g sample was placed in a Kjeldahl flask.
2) One milliliter of concentrated H_2SO_4 and 10 drops of 30% H_2O_2 were added.
3) The flask was gently shaken until the mixture was homogeneous.
4) The sample was digested on an electric heating bath for 10 minutes at approximately 160 °C until it turned black and no longer foamed.
5) The sample was removed and cooled, after which another 10 drops of 30% H_2O_2 were added.
6) The sample was digested again for 10 minutes until the solution became clear.
7) After cooling, the sample was filtered through Whatman No. 40 filter paper into a volumetric flask and diluted with distilled water to a final volume of 100 mL.

Distillation Procedure

1) Twenty milliliters of the digested solution was transferred to a distillation tube.
2) One milliliter of 50% NaOH was added.
3) The distillate was collected in 5 mL of 0.1 N HCl.
4) The sample was heated in a water bath for 10 minutes.
5) The distillate was diluted with 25 mL of distilled water.
6) Three drops of phenolphthalein indicator were added to the distillate.

Titration and Calculation Procedures

1) The distillate was titrated with 0.1 N NaOH until a color change occurred.
2) The initial and final titration volumes were recorded.
3) The same procedure was performed on a blank without the addition of a sample.
4) Total nitrogen content was calculated using the equation adapted from Sylvia et al. (2021).

$$Total\ N = [(V_1 - V_2) \times N \times 14.008 \times f] / (w \times 1{,}000) \times 100\%$$

After the total nitrogen content had been determined, protein content was calculated by multiplying the nitrogen content by the protein conversion factor of 6.25 (Diniz et al., 2013), using the following equation:

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$$\% \, Protein = \% \, N \times 6.25$$

Data Collection and Analysis

Data were collected through experimentation and documentation. The experiment generated protein-content data for each treatment combination, whereas documentation was used to record all stages of feed preparation and testing. The analysis began with descriptive statistics. Assumptions were evaluated using the Kolmogorov–Smirnov and Shapiro–Wilk tests for residual normality and Levene's test for homogeneity of variance. The effects of concentrate level, fermentation time, and their interaction were analyzed using two-way ANOVA. When significant differences were detected, the analysis was followed by the Tukey HSD test at a 5% significance level using SPSS.

RESULTS AND DISCUSSION

Protein Content of Fermented Water Hyacinth Feed

The observed parameter was the protein content of the feed after 7 and 14 days of fermentation. Each combination of concentrate level and fermentation time was replicated six times, yielding 48 samples. The complete observations are presented in Table 6.

Table 6. Observed Protein Content of Fermented Water Hyacinth Feed

Treatment	Fermen-tation time	Replicate U1	Replicate U2	Replicate U3	Replicate U4	Replicate U5	Replicate U6	Mean
P1 (0%)	7 days	0.21%	0.21%	0.26%	0.26%	0.17%	0.21%	0.22%
P1 (0%)	14 days	0.17%	0.21%	0.26%	0.26%	0.26%	0.21%	0.23%
P2 (30%)	7 days	0.30%	0.30%	0.30%	0.30%	0.30%	0.30%	0.30%
P2 (30%)	14 days	0.21%	0.26%	0.26%	0.26%	0.21%	0.21%	0.24%
P3 (35%)	7 days	0.35%	0.31%	0.32%	0.31%	0.35%	0.31%	0.33%
P3 (35%)	14 days	0.26%	0.26%	0.21%	0.26%	0.26%	0.26%	0.25%
P4 (40%)	7 days	0.39%	0.39%	0.39%	0.39%	0.43%	0.53%	0.42%
P4 (40%)	14 days	0.30%	0.30%	0.30%	0.30%	0.30%	0.30%	0.30%

After 7 days of fermentation, the mean protein content increased progressively with the addition of concentrate. The P1 treatment without concentrate produced a mean protein content of 0.22%, while the P2 treatment containing 30% concentrate reached 0.30%. A further increase was observed in the P3 treatment with 35% concentrate, which produced 0.33% protein, whereas the P4 treatment containing 40% concentrate yielded the highest mean protein content of 0.42%. These results indicate that increasing the concentrate level generally enhanced the protein content of the fermented material during the first seven days. The differences between consecutive treatments were 0.08 percentage points between P1 and P2, 0.03 percentage points between P2 and P3, and 0.09 percentage points between P3 and P4. The largest increase occurred between the 35% and 40% concentrate treatments.

After 14 days of fermentation, the mean protein contents were 0.23% for P1, 0.24% for P2, 0.25% for P3, and 0.30% for P4. Although the same general pattern was maintained, with higher concentrate levels producing higher protein contents, the values recorded after 14 days were lower than those obtained after 7 days for P2, P3, and P4. In contrast, the protein content of P1 increased slightly from 0.22% on day 7 to 0.23% on day 14. Overall, the P4 treatment fermented for 7 days produced the highest protein content, while the lowest value was recorded in the P1 treatment fermented for 7 days. These findings suggest that the

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combination of 40% concentrate and a 7-day fermentation period was the most effective treatment for increasing protein content under the conditions of this study.

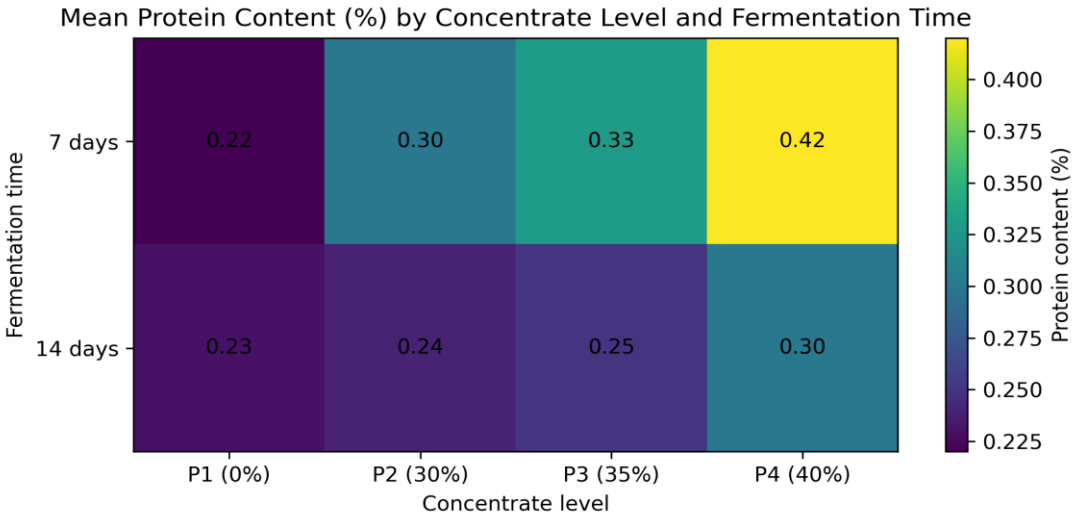

Figure 1. Mean Protein Content by Concentrate Level and Fermentation Time

Assumption Testing

The results of the residual-normality tests are presented in Table 7. The Kolmogorov–Smirnov significance value of .058 indicates that the residuals did not differ significantly from a normal distribution according to this test. However, the Shapiro–Wilk test produced a significance value of .014, indicating a departure from normality. The results of the parametric analyses should therefore be interpreted cautiously.

Table 7. Results of Residual-Normality Tests

	Kolmogorov–Smirnov[a]			Shapiro–Wilk		
	Statistic	**df**	**Sig.**	**Statistic**	**df**	**Sig.**
Standardized residual for Protein_content	.125	48	.058	.938	48	.014

The results of Levene's test are presented in Table 8. The significance value based on the mean was .011, and all other approaches also produced $p < .05$. Thus, the variances among groups were not homogeneous. Because the study used a balanced design, the two-way ANOVA results are presented in accordance with the original analysis; however, they should be interpreted in light of the violation of the homogeneity assumption. Future analyses may consider statistical approaches that are more robust to variance heterogeneity.

Table 8. Results of the Homogeneity Test

		Levene Statistic	**df1**	**df2**	**Sig.**
Protein_content	Based on Mean	6.944	1	46	.011
	Based on Median	6.221	1	46	.016
	Based on Median and with adjusted df	6.221	1	33.322	.018
	Based on trimmed mean	6.682	1	46	.013

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Effects of Concentrate Level and Fermentation Time

The two-way ANOVA showed that concentrate level had a significant effect on protein content, $F(3, 40) = 43.616$, $p < .001$. Fermentation time also had a significant effect, $F(1, 40) = 55.010$, $p < .001$. In addition, a significant interaction was found between concentrate level and fermentation time, $F(3, 40) = 9.587$, $p < .001$. This interaction indicates that the effect of concentrate level on protein content depended on fermentation duration.

Table 9. Results of The Two-Way ANOVA

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.188[a]	7	.027	30.660	.000
Intercept	3.910	1	3.910	4468.810	.000
Treatment	.114	3	.038	43.616	.000
Fermentation_time	.048	1	.048	55.010	.000
Treatment * Fermentation_time	.025	3	.008	9.587	.000
Error	.035	40	.001		
Total	4.133	48			
Corrected Total	.223	47			

Tukey HSD Post Hoc Test

The Tukey HSD test was used to evaluate differences among the eight combinations of concentrate level and fermentation time. All pairwise-comparison results are presented in Table 10 to retain the complete original dataset.

The P4 treatment fermented for 7 days showed the largest significant mean differences relative to P1 fermented for 7 days (mean difference = 0.1983, $p < .001$) and P1 fermented for 14 days (mean difference = 0.1900, $p < .001$). P3 fermented for 7 days also differed significantly from P1 fermented for 14 days (mean difference = 0.1017, $p < .001$) and P2 fermented for 14 days (mean difference = 0.0950, $p < .001$). This pattern indicates that P4 fermented for 7 days was the most effective treatment for producing a high protein content, followed by P3 fermented for 7 days.

Table 10. Results of the Tukey HSD Post Hoc Test

Group 1	Group 2	Mean difference	Adjusted p	Lower CI limit	Upper CI limit	Reject H_0
14 days_P1	14 days_P2	0.0067	0.9999	-0.0479	0.0613	No
14 days_P1	14 days_P3	0.0233	0.8668	-0.0313	0.0779	No
14 days_P1	14 days_P4	0.0717	0.0034	0.0171	0.1263	Yes
14 days_P1	7 days_P1	-0.0083	0.9997	-0.0629	0.0463	No
14 days_P1	7 days_P2	0.0717	0.0034	0.0171	0.1263	Yes
14 days_P1	7 days_P3	0.1017	0.0000	0.0471	0.1563	Yes
14 days_P1	7 days_P4	0.1900	0.0000	0.1354	0.2446	Yes
14 days_P2	14 days_P3	0.0167	0.9753	-0.0379	0.0713	No
14 days_P2	14 days_P4	0.0650	0.0102	0.0104	0.1196	Yes
14 days_P2	7 days_P1	-0.0150	0.9864	-0.0696	0.0396	No
14 days_P2	7 days_P2	0.0650	0.0102	0.0104	0.1196	Yes
14 days_P2	7 days_P3	0.0950	0.0000	0.0404	0.1496	Yes

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Group 1	Group 2	Mean difference	Adjusted p	Lower CI limit	Upper CI limit	Reject H_0
14 days_P2	7 days_P4	0.1833	0.0000	0.1287	0.2379	Yes
14 days_P3	14 days_P4	0.0483	0.1160	-0.0063	0.1029	No
14 days_P3	7 days_P1	-0.0317	0.5886	-0.0863	0.0229	No
14 days_P3	7 days_P2	0.0483	0.1160	-0.0063	0.1029	No
14 days_P3	7 days_P3	0.0783	0.0010	0.0237	0.1329	Yes
14 days_P3	7 days_P4	0.1667	0.0000	0.1121	0.2213	Yes
14 days_P4	7 days_P1	-0.0800	0.0008	-0.1346	-0.0254	Yes
14 days_P4	7 days_P2	0.0000	1.0000	-0.0546	0.0546	No
14 days_P4	7 days_P3	0.0300	0.6515	-0.0246	0.0846	No
14 days_P4	7 days_P4	0.1183	0.0000	0.0637	0.1729	Yes
7 days_P1	7 days_P2	0.0800	0.0008	0.0254	0.1346	Yes
7 days_P1	7 days_P3	0.1100	0.0000	0.0554	0.1646	Yes
7 days_P1	7 days_P4	0.1983	0.0000	0.1437	0.2529	Yes
7 days_P2	7 days_P3	0.0300	0.6515	-0.0246	0.0846	No
7 days_P2	7 days_P4	0.1183	0.0000	0.0637	0.1729	Yes
7 days_P3	7 days_P4	0.0883	0.0002	0.0337	0.1429	Yes

Discussion

Fermented water hyacinth feed was prepared through chopping, drying, mixing with concentrate, and fermentation. The results showed that increasing the concentrate level during the 7-day fermentation period was accompanied by an increase in mean protein content from 0.22% to 0.42%. During the 14-day fermentation period, protein content also increased as the concentrate level rose, but the values were lower than those obtained after 7 days for P2, P3, and P4. Thus, concentrate addition and fermentation time did not act independently; rather, their interaction determined the final protein content.

Fermentation can generally improve nutritional quality and reduce crude-fiber content through microbial activity (Nainggolan et al., 2018). Bahrun (2010) also reported improved nutritional quality in water hyacinth following fermentation. Although these studies indicate that longer fermentation can increase protein content, the present findings show that 7 days of fermentation produced higher protein contents than 14 days at concentrate levels of 30%, 35%, and 40%. This difference suggests that the optimal fermentation duration depends on ingredient composition and processing conditions.

Fermentation duration affects the physical quality and texture of feed. Kojo et al. (2015) explained that good-quality silage should not be excessively soft and should retain a texture similar to that of the original material. A relatively short fermentation period may limit the development of butyric acid-producing clostridial bacteria, which can cause silage to become soft as a result of spoilage. Conversely, excessive fermentation may reduce product yield because microorganisms enter the death phase and consequently become less capable of degrading the substrate (Rahayu & Nurwitri, 2019).

The fermentation used in this study was spontaneous because no starter culture or yeast was added. In spontaneous fermentation, microorganisms originate from the raw materials and grow according to environmental conditions, which can increase variability in the characteristics of the final product. By contrast, inoculated fermentation uses selected microorganisms as starter cultures so that the process and product can be more effectively controlled (Suprihatin, 2010).

The highest value obtained for P4 after 7 days differed from the findings of Irawati et al. (2019), who reported a high protein content after 14 days of fermentation with the addition of EM-4. This difference may be associated with the spontaneous fermentation used in the present study, whereas Irawati et al. used a starter culture. Light, pH, temperature, humidity, concentrate type, water hyacinth characteristics, and the condition of the fermentation container may also influence microbial activity and the resulting protein content.

Elferink et al. (2010) divided the silage-fermentation process into four phases. The aerobic phase lasts for several hours as oxygen between plant particles begins to decrease. The fermentation phase begins when anaerobic conditions are established and may continue for several days to several weeks, depending on ingredient composition. The stabilization phase is characterized by declining fermentation activity, during which pH, the population of lactic acid bacteria, and total acid levels become relatively stable. The feed-out phase begins when the fermentation container is opened or develops a leak, causing the anaerobic environment to become aerobic again.

In practical terms, the combination of 40% concentrate and 7 days of fermentation was the best treatment under the conditions of this study. However, because the Shapiro–Wilk and Levene tests indicated violations of some parametric assumptions, the inferential findings should be confirmed in future studies using tighter control of fermentation conditions, standardized starter cultures, and robust statistical methods.

CONCLUSION

Concentrate level, fermentation time, and their interaction significantly affected the protein content of water hyacinth-based livestock feed. The combination of 40% concentrate (400 g per 1 kg of water hyacinth) and 7 days of fermentation produced the highest mean protein content, at 0.42%. During the 7-day fermentation period, increasing the concentrate level from 0% to 40% was accompanied by an increase in protein content from 0.22% to 0.42%; during the 14-day fermentation period, it increased from 0.23% to 0.30%.

RECOMMENDATIONS

Future studies should examine other nutritional components formed during fermentation, test different feed mixtures, and add molasses or EM-4 as a starter culture. The pH, temperature, humidity, light conditions, and container seal density should be controlled and recorded. Statistical analysis should also incorporate more robust methods when the assumptions of normality or homogeneity of variance are not met.

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