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Influence of Graded Levels of Dietary Protein Supplementation on Growth, Nutrient Digestibility and Nitrogen Balance of Yankasa Rams in Semi-Arid Nigeria.

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Abstract-- **This study evaluated the effects of graded levels of dietary protein on growth performance, nutrient digestibility, and nitrogen balance of growing Yankasa rams. The objective was to determine growth performance, nutrient digestibility and nitrogen balance of Yankasa rams. Twelve (12) rams with an average weight of 16 kg and 12 months of age were randomly assigned to four dietary treatments in a completely randomized design (CRD), with three animals per treatment. The control diet contained 13.4% crude protein (CP), while the other three treatments were formulated to provide 20%, 40%, and 60% increases in CP, respectively. The basal diet consisted of fresh grass and soybean hay. The experiment lasted 90 days. Feed intake and weight changes were recorded daily and fortnightly, respectively. Digestibility was determined through a 7-day total faecal collection period. Data were analyzed using ANOVA in SAS, and treatment means were separated using Duncan's Multiple Range Test. Results showed that the diet containing 60% protein supplementation had the highest crude protein content and significantly (P<0.05) improved growth performance. Rams fed the highest protein level (60%; 16.15% CP) recorded higher feed intake, final weight, daily weight gain (67.95 g/day), and improved feed conversion ratio, representing a 278% increase in weight gain over the control. Nutrient digestibility increased significantly (P<0.05) with increasing protein levels, with dry matter (57.18–67.32%) and crude protein (63.36–77.95%) digestibility highest in the 60% treatment. Nitrogen balance was positive and highest (7.83 g/day; 47.87%) in the 60% protein treatment. In conclusion, increasing dietary protein improved growth performance, nutrient utilization, and nitrogen retention of growing Yankasa rams. A diet containing 60% protein supplementation (16.15% CP) is recommended for optimal growth performance. It is recommended that farmers in semi-arid region of Nigeria should consider supplementing their Yankasa rams with concentrate diets to improve growth performance and nutrients utilization and further research is required to investigate the long-term effect of supplementation on reproductive performance and health of Yankasa rams.**

I. Introduction

Nutrition remains the foundation of sustainable livestock production, particularly in efforts to meet the growing demand for animal protein.

In the seasonally dry tropics, ruminant productivity is largely constrained by fluctuations in feed availability and quality between the wet and dry seasons (Muhammad, 2008). During the dry season, forages are typically characterized by low crude protein and energy contents, resulting in inadequate nutrient intake to meet maintenance and production requirements. Consequently, ruminants experience weight loss, reduced growth rate, poor digestibility, and overall low productivity (Nigele *et al*, 2010; Adewumi & Ajayi, 2010; Alemayehu, 2006). The low nutritive value of dry-season forages imposes significant nutritional stress on animals grazing without supplementation, leading to depressed voluntary intake and inefficient nutrient utilization (Jiwuba *et al*, 2021). To address this challenge, supplementation of forage-based diets with energy- and protein-rich concentrate feeds has been widely recommended to bridge the nutrient deficit and improve pasture utilization.

Concentrate supplementation enhances rumen fermentation by improving rumen microbial activity, fibre digestibility, and forage intake (Leng, 2011; Peyraud, 2001). Adequate protein and energy supplementation also optimize rumen fermentation by maintaining appropriate pH and ammonia-nitrogen (NH_3-N) concentrations, thereby promoting microbial protein synthesis and volatile fatty acid production, with potential reductions in methane emission (Khampa & Wanapat, 2007). Such improvements in rumen function are critical for increasing ruminant productivity under tropical production systems.

The Yankasa sheep, an important indigenous breed in northern Nigeria, contributes significantly to meat production but is highly affected by seasonal feed scarcity. Therefore, this study evaluated the effects of varying dietary protein levels in concentrate supplements on growth performance, nutrient digestibility, nitrogen balance, and blood profile of Yankasa rams.

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II. Materials And Methods

Description of the Study Area

The experiment was conducted at the Bayero University Kano Livestock Teaching and Research Farm, Department of Animal Science, Faculty of Agriculture, located in Kano, Nigeria (Longitude 9°30′–12°30′ E and Latitude 8°30′–9°30′ N). The area is characterized by a semi-arid climate with mean annual temperature ranging from 38 to 43°C and relative humidity between 40 and 51% (Olofin, 2007). Annual rainfall ranges from 800 to 1000 mm (KNARDA, 2012). Major crops cultivated include sorghum, millet, maize, groundnut, cowpea, sesame, and rice (Muhammad *et al*, 2007).

Experimental Animals and Management

Twelve (12) Yankasa growing rams, approximately 12 months old with an average initial body weight of 16 kg, were used for the study.

Prior to the commencement of the experiment, pens were thoroughly cleaned, disinfected, and allowed to dry for two weeks. The animals were dewormed orally with albendazole to control internal parasites, while an acaricide (amitraz solution) was applied externally against ectoparasites. Long-acting oxytetracycline (LA) was administered intramuscularly at a dosage of 1 mL per 10 kg body weight as a prophylactic measure against bacterial infections. The rams were individually housed in well-ventilated pens and managed under standard husbandry practices throughout the experimental period. Clean drinking water was provided ad libitum.

Experimental Diets and Treatments

Feed supplements were collected from private ranches and analyzed for crude protein (CP) content. The mean CP value obtained was used to formulate the control diet. Subsequently, concentrate rations were formulated to increase crude protein levels in increments of 20, 40 and 60%. The ingredient composition and calculated nutrient profile of the experimental diets are presented in Table 1.

Table 1:
Composition (%) of Ingredients in the supplementary concentrate

Ingredients	Treatments			
	1	2	3	4
Wheat bran	50	48	38	30
Maize bran	35	26	24	24
Soy Bean Meal	4	12	20	28
Cotton Seed Cake	5	8	12	12
Salt	2	2	2	2
Bone meal	3	3	3	3
Toxin binder	1	1	1	1
Total	100	100	100	100
Calculated analysis				
Crude protein	13.4	16.2	18.8	21.3
ME (kcal/kg)	2620	2626	2717	2802

(contr.) = crude protein of supplements from farmers' farms, (20%), (40%) & (60%) = contr. improved by 20, 40 & 60%.

Experimental Procedure and Design

A 90-day feeding trial was conducted using a Completely Randomized Design (CRD). The experimental rams were randomly allotted to four dietary treatments with three animals per treatment. Experimental diets were offered at 5% of body weight on a dry matter basis. The dietary regimen consisted of 40% concentrate supplement and 60% basal diet (fresh grass and hay).

The daily feed allowance was weighed and divided into two equal portions. One portion was offered at 09:00 h and the second at 16:00 h. Weekly feed allowance was adjusted based on the previous week's intake. Sheep were weighed at the commencement of the experiment and subsequently at fortnightly intervals before morning feeding to monitor body weight changes.

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Digestibility Trial

At the end of the growth trial, a digestibility study was conducted. Rams from each treatment were fitted with faecal collection bags and allowed a 14-day adaptation period, followed by a 7-day total collection period. Feed refusals were collected and weighed every morning before fresh feed was offered. Daily faecal output was collected, weighed, and oven-dried at 65°C for 24 hours. Urine was collected in labeled containers containing 5 mL of 0.1 M tetraoxosulphate (VI) acid (H_2SO_4) to prevent nitrogen losses and stored under refrigeration pending analysis. At the end of the collection period, faecal samples for each animal were bulked, thoroughly mixed, and a 25% subsample taken for laboratory analysis. Apparent nutrient digestibility was calculated as:

Nutrient digestibility (%) = Nutrient intake – Nutrient in faeces x 100

Nutrient intake

Data Collection and Calculations

Daily feed intake and body weight changes were recorded. Feed offered and refused was recorded individually daily. All data collected were calculated using the following formula:

Feed intake (kg) = feed offered (kg) – feed refused (kg)

Total weight gain (kg) = final weight gain (kg) – initial weight gain (kg)

Average daily weight gain (kg) = final weight gain (kg) – initial weight gain (kg)

number of feeding days

Feed Conversion Ratio (%) = Feed intake / Weight gain

Lower FCR = better efficiency.

Laboratory Analyses

Feed, orts, and faecal samples were oven-dried at 65°C for 72 hours according to Association of Official Analytical Chemists (2023). Dried samples were ground using a hammer mill to pass through a 1-mm sieve and stored in airtight containers for analysis. Proximate composition (crude protein, crude fibre, ether extract, ash, and nitrogen-free extract) was determined following standard procedures of AOAC (2023). Crude protein was calculated as Nitrogen × 6.25. Neutral Detergent Fibre (NDF) and Acid Detergent Fibre (ADF) were determined according to the method of Van Soest (1991).

Statistical Analysis

All data generated were subjected to Analysis of Variance (ANOVA) using the Completely Randomized Design model in SAS (2003). Treatment means were separated using Duncan's Multiple Range test at 5% probability level ($P < 0.05$). The statistical model used was: $Y_{ij} = \mu + T_i + e_{ij}$ Where: Y_{ij} = observation, μ = overall mean, T_i = treatment effect, e_{ij} = experimental error

III. RESULTS

Chemical Composition of Experimental Diets

The chemical composition of the experimental diets is presented in Table 2. Significant differences ($P < 0.05$) were observed for all parameters except dry matter (DM). The 60% protein level diet recorded the highest values for crude protein (CP), ether extract (EE), hemicellulose (HEM), and Metabolizable energy (ME). In contrast, the control diet had the highest values for crude fibre (CF), ash, nitrogen-free extract (NFE), neutral detergent fibre (NDF), and acid detergent fibre (ADF). These results indicate progressive improvement in nutrient density with increasing concentrate inclusion.

Table 2:
Chemical composition of experimental diets (%)

Parameters	Treatments			
	(Contr)	(20%)	(40%)	(60%)
Dry matter	75.96	76.08	75.38	75.59
Crude protein	14.26[d]	14.94[c]	15.45[b]	16.15[a]
Crude fibre	29.10[c]	27.84[b]	27.15[a]	26.79[a]
Ether extract	6.27[b]	7.65[a]	7.55[a]	7.75[a]
Ash	6.95[a]	6.49[b]	6.13[c]	6.41[d]
Nitrogen-Free-Extract	44.11[a]	42.15[bc]	41.41[c]	39.97[d]
Neutral-detergent-fibre	52.64[a]	51.40[b]	47.95[c]	45.64[d]
Acid-detergent-fibre	35.39[a]	34.72[b]	32.73[c]	27.93[d]
Hemicellulose	17.25[a]	16.68[b]	15.22[c]	17.71[a]
Metabolizable energy (kcal/kg)	1736.6[d]	1876.3[bc]	1886.9[b]	1929.2[a]

abc Values with the same superscript within a row are not significantly Different (P≤0.05). (contr.) = crude protein of supplements from farmers' farms, (20%), (40%) & (60%) = contr. improved by 20, 40 & 60%.

Growth Performance of Yankasa Rams

The growth performance of Yankasa rams fed different protein levels is presented in Table 3. Significant differences ($P < 0.05$) were observed for all performance parameters except feed conversion ratio (FCR), which showed no significant difference ($P > 0.05$). Rams fed the 60% protein level diet had significantly higher ($P < 0.05$) final weight, total weight, daily weight, and feed intake compared to other treatments.

Table 3: Growth performance of Yankasa rams fed different levels of protein

Parameters	Treatments				SEM
	(contr.)	(20%)	(40%)	(60%)	
Initial weight (kg)	16.26	16.36	16.09	16.04	4.833
Final weight (kg)	17.97[a]	18.88[ab]	18.19[b]	22.49[a]	4.217
Weight gain (kg)	1.71[b]	2.52[b]	2.10[b]	6.46[a]	3.180
Daily weight gain (g/d)	17.96[b]	26.49[b]	22.11[b]	67.95[a]	0.0003
Total feed intake (kg)	0.80[d]	0.81[c]	0.83[b]	0.87[a]	0.0000
Daily feed intake (g/d)	0.49[d]	0.50[c]	0.51[b]	0.54[a]	0.0000
Feed conversion ratio	0.47	0.32	0.39	0.13	224.32

Nutrient Intake and Digestibility

Nutrient intake and digestibility values are presented in Table 4. Significant differences ($P < 0.05$) were observed for all intake parameters except Neutral-Detergent-Fibre intake (NDFI) and Acid-Detergent-Fibre intake (ADFI). The 60% protein level recorded the highest intake values for dry matter, crude protein, ether extract, and ash intakes. However, NDFI and ADFI were higher in the control treatment and 20% protein level.

Digestibility coefficients differed significantly ($P < 0.05$) among treatments. The 60% protein level consistently showed superior digestibility of dry matter, crude protein, crude fibre, nitrogen-free extract, NDF, and ADF. These findings indicate improved nutrient utilization with increasing dietary protein levels.

Table 4. Nutrient Intake and Digestibility of Yankasa rams fed different levels of protein

Parameters	Treatments				SEM
	(Contr.)	(20%)	(40%)	(60%)	
Nutrient intake (g/day)					
Dry matter intake	372c	380b	384b	408a	0.0004
Crude protein intake	69d	75c	79b	87a	0.0004
Ether extract intake	31c	38b	39b	42a	0.0001
Ash intake	34a	32b	31b	35a	0.0004
Neutral-detergent-fibre intake	258a	257a	245a	247	0.0007
Acid-detergent-fibre intake	173a	174a	167a	151a	0.0010
Digestibility (%)					
Dry matter	57.18^{c}	62.21^{b}	66.25^{a}	67.32^{a}	4.875
Crude protein	63.36^{d}	68.02^{c}	74.90^{b}	77.95^{a}	0.073
Crude fibre	39.26^{c}	52.95^{b}	58.08^{a}	58.74^{a}	3.938
Nitrogen-free-extract	18.25^{d}	32.99^{c}	39.69^{b}	43.97^{a}	9.188
Neutral-detergent-fibre	34.43^{b}	35.25^{b}	36.25^{b}	53.38^{a}	0.897
Acid-detergent-fibre	21.01^{d}	24.58^{c}	31.23^{b}	49.02^{a}	0.891

abc Values with the same superscript within a row are not significantly Different (P≤0.05). (contr.) = crude protein of supplements from farmers' farms, (20%), (40%) & (60%) = contr. improved by 20, 40 & 60%.

Nitrogen Balance

Nitrogen balance parameters are presented in Table 5. Significant differences ($P < 0.05$) were observed across treatments for all nitrogen indices measured. Sixty percent (60%) protein level diet recorded the highest nitrogen intake, nitrogen absorbed, nitrogen balance, and nitrogen retention percentage.

Urinary nitrogen was highest in 40% protein treatment. The progressive increase in nitrogen retention with higher protein levels indicates improved protein utilization and metabolic efficiency in rams fed higher concentrate diets.

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Table 5. Nitrogen balance of Yankasa rams fed different levels of protein

Parameters (g/day)	Treatments				SEM
	(contr.)	(20%)	(40%)	(60%)	
N intake	11.65[d]	13.35[c]	14.56[b]	16.36[a]	0.159
Faecal N	6.06	6.25	6.15	7.34	0.420
Urinary N	0.84[b]	1.31[a]	1.58[a]	1.19[a]	0.028
N loss	6.89[c]	7.56[b]	7.73[b]	8.53[a]	0.452
N absorbed	5.59[c]	7.09[b]	8.40[a]	9.02[a]	0.817
N balance	4.76[d]	5.79[c]	6.83[b]	7.83[a]	0.863
N balance (%)	40.68[b]	43.39[b]	46.83[a]	47.87[a]	34.696

abc Values with the same superscript within a row are not significantly Different ($P \leq 0.05$). (contr.) = crude protein of supplements from farmers' farms, (20%), (40%) & (60%) = contr. improved by 20, 40 & 60%.

IV. Discussion

Chemical Composition of Experimental Diets

The chemical composition of the experimental diets indicates their suitability as supplements for growing ruminants. The adequate dry matter (DM) content observed supports optimal voluntary intake and proper rumen function, as diets with appropriate DM enhance microbial fermentation efficiency (McDonald *et al*, 2011). Crude protein (CP) increased progressively from 14.26% in the control diet to 16.15% at 60% inclusion, meeting the protein requirements for growing sheep. Adequate CP enhances nitrogen availability for rumen microorganisms, promoting fibre degradation and nutrient utilization (Abu *et al*, 2023; NRC, 2007). The progressive improvement in CP content likely contributed to the enhanced growth and nutrient digestibility observed.

Fibre fractions (CF, NDF, and ADF) declined with increasing protein levels, suggesting improved digestibility and reduced rumen fill. High NDF and ADF are known to limit voluntary intake and energy availability in sheep (McDonald *et al*, 2011). Therefore, their reduction implies improved feed efficiency and greater energy utilization. Ether extract (EE) increased to 7–8%, enhancing dietary energy density without exceeding recommended levels. Moderate fat inclusion improves energy supply and growth performance when maintained within safe limits (NRC, 2007). Although ash content slightly declined, values remained within acceptable ranges, indicating adequate mineral supply necessary for metabolic and skeletal functions (McDowell, 2003).

Nitrogen-free extract (NFE) decreased with higher protein inclusion, reflecting lower rapidly fermentable carbohydrates. Excess fermentable carbohydrates may predispose animals to rumen acidosis (Plaizier *et al*, 2017); thus, the observed reduction may support rumen stability while maintaining energy supply through increased EE and improved digestibility (Beauchemin *et al*, 2020). Metabolizable energy (ME) increased significantly from 1,736.6 to 1,929.2 kcal/kg, demonstrating improved dietary energy density. Energy is often the primary limiting nutrient in sheep production systems (Solaiman *et al*, 2018). Higher ME supports anabolic processes, growth, and improved feed efficiency (Mekonnen *et al*, 2021; González-Rivas *et al*, 2022).

Growth Performance of Yankasa Rams Fed Different Protein Levels

Weight gain reflects the efficiency of feed utilization in ruminants. In this study, rams fed the 60% protein diet showed the highest weight gain, likely due to the higher crude protein (CP) content enhancing rumen microbial activity, improving roughage utilization, and increasing feed intake (Hassan, 2014; Munza, 2021). This aligns with Ferdous *et al* (2011), who reported that higher levels of concentrate supplementation improve live weight gain. Daily weight gain (DWG) was also highest in the 60% protein treatment, which may be attributed to the diet's higher dry matter (DM) content and lower neutral and acid detergent fiber (NDF and ADF), facilitating nutrient absorption and utilization (Osita *et al*, 2019).

DWG in this study ranged from 13.05 to 67.94 g/day, lower than values reported by Abu *et al* (2023) for the same breed (40.87–75.73 g/day) and Teklehaymanot (2019) (106.67 g/day), possibly reflecting differences in diet composition and experimental conditions.

Feed conversion ratio (FCR) was lowest in the 60% protein treatment (0.13), indicating more efficient feed utilization, likely due to higher feed palatability and nutrient availability (Audu *et al*, 2018). Feed intake increased with protein level, reflecting improved palatability and enhanced supply of nitrogen and readily available carbohydrates for rumen microbes, which stimulate microbial growth and increase fractional outflow of digesta (Aregheore & Perera, 2004; Shem *et al*, 2003). Total and daily feed intake in this study were higher than those reported by Audu *et al* (2023) and Dan Abba (2021), but lower than values reported by Garba *et al* (2021) and Mbahi *et al* (2023), likely due to differences in breed, feed composition, and environmental conditions.

Overall, the improved weight gain and FCR observed can be attributed to the quality of nutrients in the supplemented diets, which enhanced nutrient availability and utilization (Tripathi *et al*, 2006). Increasing the concentrate-to-roughage ratio improved feed consumption, conversion efficiency, final weight, and daily weight gain (Atsbha *et al*, 2021).

Nutrient Intake and Digestibility of Yankasa Rams Fed Graded Levels of Protein

Nutrient intake increased progressively with increasing dietary protein level, consistent with the positive association between digestibility and voluntary feed intake (Detmann *et al*, 2014). Dry matter intake (DMI) increased from 2.15% at 20% protein level to 9.68% at 60% protein level. The values obtained were higher than those reported by Sani *et al* (2022) for Yankasa rams fed millet stover–based diets, suggesting improved diet quality in the present study. The relatively low neutral detergent fibre (NDF) content may have reduced rumen fill limitation, thereby enhancing intake (McDonald *et al*, 2011). Similar responses of DMI to increased crude protein (CP) concentration have been reported by Ahamefule *et al.,* (2006).

Crude protein intake (69–87 g/day) increased proportionally with dietary CP level and fell within the range reported by Yusuf *et al.,* (2022), although exceeding values documented by Sani *et al* (2022) and Abubakar *et al.,* (2005). Ether extract intake was comparable to Garba *et al.,* (2021), while variations observed in NDF and ADF intakes relative to previous studies likely reflect differences in ingredient composition and supplementation strategy.

Digestibility coefficients improved significantly with increasing protein level. Dry matter digestibility (DMD) recorded the highest value at 60% protein inclusion. These values align with reports by Atsbha *et al.,* (2021) and Teklehymanot (2019) and exceed those of Udo *et al.,* (2021). Enhanced DMD with protein supplementation has been attributed to increased nitrogen availability for rumen microbial proliferation and activity (Hagos, 2014; Gebrelassie, 2012). Crude protein digestibility (63.36–77.95%) similarly increased with dietary protein level, supporting the positive relationship between CP concentration and nutrient utilization (McDonald *et al*, 2011). The improved CP digestibility suggests efficient ruminal degradation and microbial protein synthesis. Differences compared with earlier studies may be due to breed, age, feeding level, and diet composition.

Fibre digestibility parameters (CFD, NDFD, and ADFD) showed improvement with increasing CP level, indicating enhanced degradation of structural carbohydrates. Adequate nitrogen supply stimulates rumen microbial fermentation of fibre fractions (Banerjee, 2009; McDonald *et al*, 2011). Although some values differed from previous reports, such variations are commonly associated with differences in dietary formulation and experimental conditions. Nitrogen-free extract digestibility was highest at 60% protein level, indicating improved utilization of non-structural carbohydrates. Overall, diets containing 60% protein level consistently produced superior intake and digestibility values, followed by the 30% level. The digestibility coefficients obtained (>50%) exceed the minimum recommended threshold (40–50%) for ruminant feeds, confirming adequate ruminal degradation.

Nitrogen Balance of Yankasa Rams Fed Different Levels of Protein

Nitrogen balance is a reliable indicator of the biological value of dietary protein (Hassan *et al.*, 2016; Bastos *et al.*, 2014; Abdu *et al.*, 2012; Babayemi & Bamikole, 2006). Positive nitrogen balance signifies that nitrogen intake exceeds nitrogen losses through faeces and urine, indicating adequate protein supply for growth and tissue deposition. In the present study, nitrogen retention values (40.68–47.87%) showed that all rams were in positive nitrogen balance, suggesting sufficient dietary protein across treatments.

The significant differences observed among treatments may be attributed to variations in crude protein (CP) content and nitrogen intake. Nitrogen retention generally increases with higher nitrogen consumption and digestibility (Okeniyi *et al*, 2010). Thus, the superior nitrogen retention recorded in rams fed the higher-protein diet could be linked to improved protein availability and efficient utilization (Foster *et al*, 2009).

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High urinary nitrogen losses are often linked to increased rumen protein degradability (McDonald *et al*, 2011). Therefore, improved nitrogen retention may result from reduced urinary nitrogen excretion and enhanced metabolic utilization. The overall positive nitrogen retention suggests that dietary protein levels were adequate to support rumen microbial activity and muscle growth in the rams.

Nitrogen retention percentages (40.68–47.87%) in this study were slightly lower than the 43.7–89.75% reported by Abu *et al* (2023), Ahmed *et al* (2022), Yakubu *et al* (2021), Yusuf *et al* (2022), and Jokthan *et al* (2013), such variations may be due to differences in crude protein levels, ingredient composition, protein bioavailability, and overall nitrogen intake (Lamidi, 2009).

V. Conclusion And Recommendation

Conclusion

Inclusion, Increasing dietary protein levels improved feed intake, growth performance, nutrient digestibility, nitrogen balances and physiological status of Yankasa rams. Rams fed the diet containing 60% protein supplementation (16.15% crude protein) showed superior performance in terms of weight gain, feed utilization and nutrient digestibility. Therefore, supplementation with a diet containing approximately 16% crude protein is recommended for optimal growth performance of Yankasa rams under semi-arid conditions.

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