

Protein requirement of the juvenile male red-rumped agouti (*Dasyprocta leporina*) fed diets formulated with tropical forages

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The rearing of neotropical animals for meat has the potential to improve food security in the Caribbean, South and Central America. Neo-tropical animal species are well adapted to these environments and can be maintained on unconventional feedstuffs such as forages, fruits and vegetable waste. The red-rumped agouti (*Dasyprocta leporina*) is a neotropical animal with potential for captive rearing for meat production. However, its dietary protein requirements are largely unknown. Therefore this study was done to investigate protein requirements in captive reared juvenile male agoutis. Nine juvenile male agoutis were allocated to one of three treatment groups with varying levels of crude protein (CP). The treatment groups were low crude protein (LP – 100 g/kg DM), medium crude protein (MP – 125 g/kg DM) and high crude protein (HP – 150 g/kg DM). After an initial 6-weeks adaptation period, measurements of dry matter and nutrient intake and weight gain were recorded weekly for 5 weeks. Daily CP intake was unaffected by dietary CP levels ($P > 0.05$). However, apparent CP digestibility (61.2%) and intake of digestible CP (4.5 g/day) were highest with MP diet. Dry matter intake of LP diet was 17% and 39% higher than MP and HP diets, respectively. Intake of gross energy was highest (1.2 MJ/day) in agoutis fed the LP diet. It was therefore concluded that dietary CP of 100 – 125 g/kg DM can satisfy minimum daily intake of 4.0 g digestible CP which was adequate to achieve average daily gain of approximately 5.0 g in the captive reared juvenile male agouti.

Keywords: Digestible crude protein, CP intake, dry matter intake, daily weight gain

Food and nutrition security continues to be a challenge in the Caribbean. While most Caribbean countries have enough food available to meet their daily energy requirements, imports accounted for 40 – 95% of the total food consumed within the Caribbean (Beckford and Campbell 2013). The reported total cost for food imports in CARICOM was more than US\$ 4 billion in 2011 with livestock products, along with processed foods and grains accounting for more than 25% of total import costs (FAO 2013). Therefore it is important that Caribbean islands develop their local livestock sectors as a key strategy for reducing reliance on imported animal protein and feed ingredients. Livestock production in the region has been dominated by traditional farming of domesticated species such as poultry, pigs, cattle, sheep, goats and, to some extent, rabbits. Management of these species,

especially poultry and pigs is heavily dependent on imported inputs such as animal genetics and ingredients for feed. They also require sophisticated housing and disease management and surveillance systems. Recent increases in the cost of imported grains for animal feeds and declining animal productivity in Europe and North America due to climate change further threatens food availability in the region. Therefore, alternative livestock species that are less dependent on imported inputs and are well adapted to the tropical environment of the Caribbean could significantly reduce reliance on imported feed ingredients, animal genetics and livestock products.

Neotropical wildlife species offer suitable livestock options that can be developed for food and conservation purposes. Unlike conventional livestock production systems, neotropical animals can remain productive on

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forages, fruits and vegetable waste as their sole source of feed. They are also well adapted to the tropical climate and are less susceptible to heat stress and diseases than most domesticated livestock species (Jones and Garcia 2018; Lall et al. 2018a). NRC (1991) confirmed that neotropical wildlife farming can serve as a source of economic development for rural populations; this is because of their small size, relatively higher reproductive ability and efficiency in converting waste and food by-products into meat makes them less expensive to maintain with faster returns on investment. The agouti (*Dasyprocta leporina*) is one such neotropical wildlife species with potential for domestication and commercial production. The agouti is a neotropical rodent that primarily inhabits South and Central America and some Caribbean islands where they are hunted for meat (Jones et al. 2019).

While the feeding behaviour (Lall et al. 2018b) and gross digestive anatomy (Garcia et al. 2000) of the agouti are well documented, its nutritional requirement specific to protein is not well understood. Protein nutrition is central to the development of any livestock species because of the vital role of protein in promoting health and production (Wang et al. 2020). This study investigated the protein requirements in captive reared juvenile male agouti fed isocaloric diets formulated with conventional feeds and different types and amounts of tropical forages to provide varying levels of dietary crude protein.

Materials and methods

Site description and ethical clearance

Ethical clearance (Ref: CREC-SA.0871/042021) for this research was

provided by the Research Ethics Committee of The University of the West Indies (UWI), St Augustine Campus, Trinidad. Management of these animals complied with the guidelines for use of laboratory animals for research (National Research Institute 2011). The experiment was conducted at The University of the West Indies, St Augustine Field Station (10°38' N, 61°25' W). Average daily temperature and humidity throughout the experimental period were 30 °C and 80%, respectively.

Formulation of the diets

Three isocaloric diets low crude protein (LP – 100.2 g/kg DM), medium crude protein (MP – 125.3 g/kg DM) and high crude protein (HP – 150.4 g/kg DM) contents were formulated for this study (Table 1). These diets composed of varying levels of *Leucaena leucocephala* pods, rolled oats, *Trichantera gigantea* leaves, commercial rabbit feed and tanner grass (*Brachiaria arrecta*). Diets were formulated with a stochastic model in the WINFEED 2.8 feed formulation software (WinFeed Limited, Cambridge, UK). All forages were wilted over a 2-day period before being dried in a force-draft oven at 60°C for 24 hours. The dried forages and all other feedstuffs were milled using a Thomas Wiley laboratory Mill Model 4 to pass through a 2 mm screen.

Approximately 300 ml of a 30% sugarcane molasses solution was added per kg of mixed feed ingredients and pelleted in a modified OEM 150kg/H Automatic Electric Meat Grinder (Model No. SXC-12). The feed pellets were kept at a constant size of 3 × 1 cm. The pellets were oven dried for 12 hours at 60 °C degrees in a force-draft oven.

Table 1: Diet composition and chemical constituents

	Diet/diet composition (g/kg DM)		
	LP	MP	HP
Ingredients/diet composition			
<i>Leucaena leucocephala</i> pods	0.0	33	154
Processed oats	459	0	11
<i>Trichantera gigantea</i> leaves	62	14	162
Commercial rabbit ration	103	720	673
<i>Brachiaria arrecta</i>	376	233	0
Chemical constituents			
Crude protein	100	125	150
Ether extract	15	11	9
Ash	82	89	101
Neutral detergent fibre	290	311	297
Acid detergent fibre	254	253	228
Acid detergent lignin	46	45	69
Hemicellulose	35	58	69
Cellulose	209	208	159
Gross Energy (MJ/kg DM)	17.9	17.9	17.8

LP, MP, HP represent the low, medium and high protein diets respectively

Animal management

Nine captive reared juvenile male agoutis (10 – 12 weeks old) with mean live weight of 1.4 ± 0.2 kg were allocated to one of three diets with varying crude protein (CP) levels. The animals were obtained from the agouti unit of The University of the West Indies Field Station, where they were housed in communal cages and fed a diet of commercial rabbit feed (Master Mix Feeds) supplemented with “in-season” fruits. The selected animals were placed in individual metabolism cages designed for this species. These cages allowed for the collection of refused feed and faeces while avoiding contamination of both.

The cages consisted of two parts: an upper housing area for the animals and lower area for the collection of faeces and spillage feed. The dimensions of the cages were $25 \times 16 \times 20$ cm, constructed using 2.5×5 cm wire mesh for the sides and 1.25×2.5 cm for the floor. At the back of the cages was an opening to insert metal feeders. The cages were placed on top of an iron shelf that contained slots for the insertion of the waste bins.

Feeding management

The animals were fed with a commercial rabbit ration (100 g/day/animal) and kept for an initial 4 weeks in the metabolism cages to ensure they were well adapted to the new environment. After this period, they were allowed a second adaptation phase where they were fed with the treatment diets for another 2 weeks before measurements were taken. Daily intake was monitored and adjusted when necessary to ensure at least 10% refusal. The end of this phase was determined when dry matter intake was approximately 4% of the animal live weights. Afterwards, the animals were offered the diets at rate of 200 g per day. Feeding was done once daily at 7:00 a.m. for 5 weeks. Unlimited water was provided to the animals using rabbit water feeders installed on the side of the cage and metal cups tightly secured in the cages. Faeces were collected from the sieve tray and weighed twice weekly. Intake was recorded daily by collecting the refuse and spilled feed. The animals were weighed at the end of each week in a polypropylene feed bag of known weight using a digital hanging hook scale.

Chemical analysis

Feed and faecal samples collected during the experimental period were stored prior to chemical analysis at 4 °C in a refrigerator at the food production laboratory of the Faculty of Agriculture. In preparation for chemical analysis, both feed and faecal samples were dried to constant weight at 60 °C in a forced-draft oven, then ground in a hammer mill to pass through a 2 mm screen. Approximately 1 g of ground sample was oven dried at 105 °C for 24 hours to determine dry matter (DM) content (AOAC 2005). CP was determined by the copper catalyst Kjeldahl method (AOAC, 2005; method number 976.05) for total nitrogen and CP was calculated as N × 6.25. Neutral detergent fibre (NDF) and acid detergent fibre (ADF) were sequentially analysed using an ANKOM 2000 fibre analyser.

Sodium sulphite and amylase were included in the NDF analysis. NDF and ADF were expressed inclusive of residual ash. Acid detergent lignin (ADL) was determined by solubilisation of cellulose with 72% sulphuric acid as described by Van Soest et al. (1991). The difference between NDF and ADF was calculated as hemicellulose and the difference between ADF and ADL content represented cellulose. Gross energy (GE) of the feed and faecal samples were determined using a Parr Instrument Company 6200 Series isoperibol calorimeter.

Statistical analysis and calculations

Analysis of the effects of varying CP levels (fixed factor) was done by one-way ANOVA. Treatment means were separated by Tukey's multiple comparison test using Minitab 19 statistical software. Apparent digestibility and intake of proximate fractions were calculated as follows:

Apparent DM digestibility %

$$= 100 \left(\frac{\text{daily DM intake} - \text{daily DM faecal output}}{\text{daily DM intake}} \right)$$

Daily nutrient intake

$$= \left(\frac{\text{nutrient content of feed} \times 100}{\text{DM \% of feed}} \right) \text{daily DM intake}$$

Apparent nutrient digestibility %

$$= 100 \left(\frac{\text{daily nutrient intake} - \text{daily nutrient output}}{\text{daily nutrient intake on DM basis}} \right)$$

Results

Nutrient intake and apparent digestibility

Dietary CP did not significantly affect daily CP intake. However, apparent CP digestibility (61.2%) and subsequent daily intake of digestible CP (4.5 g/day) were highest in MP diet. Apparent DM, ADF and ether extract digestibility and GE intake (1.2 MJ/day) were highest in animals fed the LP diet (Table 2).

Table 2: Nutrient intake and apparent digestibility in agouti fed diets with varying crude protein (CP) levels

Parameters	Diet/ crude protein levels			SEM	P
	LP	MP	HP		
Nutrient intake (g/day)					
Crude protein	6.9	7.3	6.3	0.33	0.088
Digestible crude protein	3.9 ^{ab}	4.5 ^a	3.1 ^b	0.27	0.006
Ether extract	1.0 ^a	0.6 ^b	0.4 ^c	0.03	0.000
Neutral detergent fibre	20.1 ^a	17.9 ^b	12.5 ^c	0.80	0.000
Acid detergent fibre	17.6 ^a	14.6 ^b	9.6 ^c	0.67	0.000
Acid detergent lignin	3.2 ^a	2.6 ^b	2.9 ^{ab}	0.13	0.016
Cellulose	14.4 ^a	11.9 ^b	6.7 ^c	0.50	0.000
Hemicellulose	2.4 ^b	3.3 ^a	2.9 ^a	0.13	0.000
Gross energy (MJ/day)	1.2 ^a	1.0 ^b	0.75 ^c	0.05	0.000
Apparent DM and nutrient digestibility (%)					
Dry matter	70.8 ^a	67.2 ^{ab}	61.6 ^b	1.6	0.001
Crude protein	54.8 ^a	61.2 ^{ab}	48.2 ^b	2.1	0.001
Ether extract	64.3 ^a	53.9 ^{ab}	49.5 ^b	3.8	0.026
Neutral detergent fibre	42.4 ^a	39.4 ^{ab}	28.9 ^b	3.3	0.016
Acid detergent fibre	37.7 ^a	32.9 ^a	18.7 ^b	3.5	0.001
Cellulose	40.8	36.9	30.2	3.2	0.074
Hemicellulose	76.3	67.7	62.6	3.9	0.067

^{a, b, c}, Treatment means within row that do not share a letter differ significantly ($P \leq 0.05$). LP, MP, HP represent the low, medium and high protein diets respectively.

Animal performance

Total and average daily weight gain varied between treatments but these variations were not significantly different, though some animals on the HP diet lost weight. Intake of DM was highest in animals offered the LP diet

(69.3 g/day) (Table 3). Dry matter intake of LP diet was 17% and 39% higher than MP and HP diets, respectively. Intake of DM and faecal output were 2.8% of the final body weight and 38.4% of feed intake, respectively in animals fed HP diet.

Table 3: Performance (mean \pm SE) of agouti fed diets containing different levels of CP

Performance parameters	Diet/ Crude Protein levels			SEM	P
	LP	MP	HP		
Initial weight (kg)	1.5	1.4	1.4	0.27	0.957
Final weight (kg)	1.7	1.6	1.3	0.20	0.382
Total weight gain (g)	198	164	-40	95.6	0.251
Average daily gain (g)	5.7	4.7	-1.1	2.77	0.251
ADG (g)/ g CP intake	0.82	0.64	- 0.17	0.40	0.319
DM intake (g/day)	69.3 ^a	57.5 ^b	42.1 ^c	2.7	0.000
DM intake (% final BW)	4.4 ^a	4.5 ^a	2.8 ^b	0.13	0.000
Faecal output (g DM/day)	19.7 ^a	18.4 ^a	15.7 ^b	0.53	0.000
Faecal output (% DM intake)	29.2 ^b	32.8 ^{ab}	38.4 ^a	1.6	0.001

^{a, b, c}, Treatment means within row that do not share a letter differ significantly ($P \leq 0.05$). LP, MP, HP represent the low, medium and high protein diets respectively.

Discussion

Crude protein intake and digestibility

Despite increasing concentrations of dietary CP, intake of CP was similar between treatments because DM intake decreased as dietary CP increased. Intake of CP and digestible CP by agoutis in the present study was less than half the requirements for growing/fattening rabbit reared on commercial concentrate feeds (Iyeghe-Erakpotobor et al. 2007; Marín-García et al. 2020). The low DM intake observed with the HP diet was associated with low apparent DM and CP digestibility. Higher lignin concentration of the HP diet due to higher proportion of *L. leucocephala* pods and *T. gigantea* leaves could account, in part, for the low DM digestibility and intake from the HP diet. Indeed, lignin is indigestible (Moore and Jung 2001), and could therefore limit feed digestibility and intake. The lignin content of the LP diet was similar to the upper limit (65.0 g/kg as fed) of lignin in commercial rabbit feeds (Gidenne 2015) which could be an indication that other factors may have had a greater influence on the digestibility of the HP diet or the lignin tolerance level of the non-domesticated agouti that is accustomed to selecting mainly fruits and seeds is lower than that of the rabbit. Lignin negatively affects digestibility by shielding plant polysaccharides from enzymatic hydrolysis (Jung and Allen 1995). The activity of lignin can also be regulated by composition (Jung and Allen 1995) and spatial distribution of lignin within the plant cell (Dryden 2008) in addition to its concentration. The lower apparent DM and CP digestibility of the HP diet could be a result of high levels of phenolic compounds or other anti-nutritive factors. The HP diet contained the highest portion of *L. leucocephala* pods which are rich in condensed tannin. Condensed tannin can reduce CP digestibility by bonding to plant protein forming tannin-protein complex making some protein resistant to microbial digestion (Adewale et al. 2018).

However, low tannin content around 0.45 – 0.5% DM, was reported to increase weight gain and DM intake (Maertens and Štruklec 2006), while concentrations around 4% of DM had no significant effect on DM digestibility in rabbits (Mashamaite et al. 2009). This could confirm that tannin had no influence on DM or CP digestibility in MP diet because *L. leucocephala* pods only contributed 3.3% of the diet DM. Garcia et al. (2000) confirmed that the agouti, like the rabbit, can obtain significant amounts of its nutrient via hind-gut fermentation because it has a functional caecum that is proportionally larger than the forestomach. In fact, volatile fatty acids produced from caecal fermentation in the rabbit can satisfy as much as 50% of their maintenance energy requirements (Gidenne 2015). This justifies the inclusion of forages in the diet of the rabbit and the agouti. The potential effect of anti-nutritional compounds may be an indication that protein digestion in the agouti reared in captivity is affected, to a large extent, on caecal fermentation. Therefore, the nature and source of the protein offered to this animal is critical. For example, higher CP digestibility (68.2 – 71.1%) from diets with tortula yeast or soybean meal fed to mature-adult agouti was previously reported (Hosken et al. 2015). Variations in the amino acid profile of the protein source can account for differences in protein digestibility. Torula yeast, in particular, has a lysine content of 4.5%, which is much higher than that of soybean meal (Rodriguez et al. 2011). While the concentrations of essential amino acid of the forage protein sources used in the present study were unknown, the profile of essential amino acids may be lower than required to support rapid growth because non-protein nitrogen constitutes a large fraction of forage CP (Hughes et al. 2021). It is therefore clear that significant research is needed to understand amino acids requirements, the most suitable forage types and optimum inclusion rates for different classes of agoutis reared in captivity.

Animal performance

Average daily weight gain of agoutis in this study was superior to those reported by John and Jones (2020) in adult agoutis. On the other hand, the ADG observed in this study was lower than those reported in a recent study with young agoutis that were weaned at different stages and fed with commercial rabbit feed (Singh and Jones 2021). Clearly, stage of development has a significant bearing on growth response (Lui and Barron 2011). This highlights the need to develop feeds for the specific physiological stages of the agouti as is done for domesticated livestock species like pigs and poultry. Similar ADG from the LP and MP diets is an indication that dietary CP of 100 – 125 g/kg DM exceeded maintenance requirements and is capable of supporting growth of approximately 5.0 g per day in the juvenile agouti providing at least 4.0 g daily intake of digestible CP is achieved. Because of variation in protein quality due to protein source, digestible CP is a much better description for protein requirement of the agouti, especially when forage is included in the feed. Dry matter intake in growing rabbits at a similar stage of growth to the agoutis is around 30 – 60% higher than observed for the agoutis in this study (Iyeghe-Erakpotobor et al. 2007; Marín-García et al. 2020). Considering that rabbit production has benefitted from significant genetic improvement and development of feeds to suit their specific needs unlike the non-domesticated agouti, DM intake relative to body weight is a more suitable standardised approach to determine intake adequacy. Indeed, daily DM intake of LP and MP diets as a percentage of body weight was around 4.5% which is above the intake/live weight ratio of agouti in other studies (Hosken et al. 2015) and also higher than the target for most domesticated livestock species including the rabbit and different classes of ruminants. This can be taken as confirmation that NDF (290 – 311 g/kg DM), ADF (253 g/kg DM) and ADL (46.0 g/kg DM) contents of the LP and MP diets pose no

serious limitation to intake. Indeed, these fibre and lignin contents are within the recommended range for commercial feeds for growing rabbits (Gidenne 2015). Low intake of energy, digestible CP and DM were responsible for the negative growth of the agoutis fed the HP diet. This was possibly influenced by anti-nutritive compounds such as condensed tannin in *L. leucocephala* pods. The highest intake of DM and lowest faecal output relative to DM consumed for LP diet could be an indication of the animal's desire to satisfy its protein and energy requirement when fed a low protein diet. Therefore, DM intake was high and the feed and nutrients consumed were more efficiently used. It is worthy to note that intake of energy was also highest from the LP diet.

This observation offers support to the chemostatic intake regulation theory which suggests that intake of lower quality diets tend to increase because the animal's appetite is regulated by the concentration of glycogen reserves and blood metabolites such as glucose (Cheeke 2005).

Conclusion and recommendation

Dietary CP of 100 – 125 g/kg DM that can satisfy minimum daily intake of 4.0 g digestible CP is adequate to achieve ADG of approximately 5.0 g in the captive reared juvenile male agouti. Due to the variations in protein type, due to the source of the protein, digestible CP is a more accurate description of the protein requirement of the agouti, especially where forage-based protein is included in the diet. Further work on the protein requirements of the juvenile male agouti using protein sources free of anti-nutritive compounds must be done to expand knowledge and understanding of protein requirements, utilisation and nitrogen metabolism in this neo-tropical species under captive rearing. Also, techniques to collect urine samples free from faecal and feed contamination must be developed to facilitate nitrogen balance studies.

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Conflict of interest declaration

The authors declare there are no actual or potential conflict of interest associated with this work.

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