



Spleen Indices of Mixed Breed Rabbits Fed Graded Levels of some Common Forages in the Coastal Savannah Ecological Zone of Ghana

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Abstract

This study sought to examine the immune response of rabbits fed different dietary levels of four common forages in Coastal Savannah Ecological Zone of Ghana. A total of 48 eight-week old mixed-breed rabbits were offered-- concentrate containing graded levels of 0, 10, 20 and 30% *Brassica oleracea* outer leaves or *Musa paradisiaca* leaves, *Megathyrsus maximus*, and *Stylosanthes guianensis* aerial parts for one week and 12-weeks adaptation and testing periods respectively. Spleen from 8 representative rabbits from each treatment group were harvested at the end of the feeding period and used to determine the spleen indices. No significant differences were found in the body weights, 1926 to 2314g, spleen weights, 1.00 to 1.50g, and spleen indices, 0.05 to 0.07g of rabbits fed concentrate containing the graded levels of the leaves. Rabbits fed diets containing 10% inclusion level of *Brassica oleracea* had the highest body weight, while the highest spleen weight and spleen index were found in rabbits fed 30% inclusion level of *Musa paradisiaca* leaves and *Stylosanthes guianensis* aerial parts. The study concludes that feeding diets containing up to 30% inclusion level of *Brassica oleracea* outer leaves, *Musa paradisiaca* leave and *Megathyrsus maximus* and *Stylosanthes guianensis* aerial parts to growing rabbits enhance the immune response of the animals thereby ensuring good performance and health.

Keywords: Body Weight; Brassica Oleracea; Dietary Level; Megathyrsus Maximus; Musa Paradisiaca; Stylosanthes Guianensis

Introduction

The spleen is the largest secondary immune organ in the body [1,6] and is responsible for initiating immune reactions to blood-borne antigens and for filtering the blood of foreign material and old or damaged red blood cells [1]. It has a wide range of immunologic functions alongside its roles in hematopoiesis and red blood cell clearance. The physical organization of the spleen allows it to filter blood of pathogens and abnormal cells and facilitate low-probability interactions between antigen-presenting cells and cognate lymphocytes [6]. It also plays a modulatory role that balances pro- and anti-inflammatory responses [9].

Brassica oleracea outer leaves, *Megathyrsus maximus* and *Stylosanthes guianensis* aerial parts and *Musa paradisiaca* leaves are among the common forages fed to rabbits in the Coastal Savannah ecological zone of Ghana [10]. *Stylosanthes guianensis* is a short-lived, erect or semi-erect perennial legume that can reach a height of 1-1.5 m. The fresh aerial parts contain 22.1% dry matter (DM) as feed, crude protein (CP) 14.0% DM, crude fibre (CF) 31.2% DM, ether extract (EE) 2.4% DM, ash 8.8% DM and MJ/kg gross energy (GE) 18.5% DM [3]. *Megathyrsus maximus* (Guinea grass) is a fast-growing leafy grass with a good nutritional value. The grass has DM 22.7% as feed, CP 11.2% DM, CF 37.3% DM, and GE 18.1 MJ/kg

DM [2]. *Musa paradisiaca* leaves, considered waste materials after harvesting the fruit [11], contain CP 19.4% DM, crude lipid 1.8% DM, CF 8.1% DM, ash 7.2% DM and carbohydrate 54.6% DM [8].

Notwithstanding the importance of the immunologic functions of the spleen in rabbits, information is scarce regarding the dietary effects on immune response of rabbits kept in the tropics. The objective of the research was to examine the immune response of rabbits fed different dietary levels of *Brassica oleracea* outer leaves, *Musa paradisiaca* leaves, *Megathyrus maximus*, and *Stylosanthes guianensis* aerial parts in the Coastal Savannah zone of Ghana.

Materials and Methods

Study location, animals and housing

The study was carried out at the Rabbitry Unit of the Council for Scientific and Industrial Research - Animal Research Institute, Frafraha Station, situated on latitude 44° 5' North and longitude 0° 9' East in the Coastal Savannah ecological zone of Ghana. A total of 96 eight-week-old clinically healthy population of mixed-breed and mixed-sex local, California, New Zealand White and Chinchilla rabbits, weighing between 745.2 to 920.8g, were used for the study.

Two rabbits of same-sex were housed in a wire mesh cage with a wooden frame, measuring 40 x 40 x 50 cm (width x length x height, and equipped with a plastic feeding trough and an automatic watering facility. Six hutches, with three rows of six cages each, used in housing the rabbits, were placed 70 cm above ground level under *Azadirachta indica* canopy in an area with natural ventilation and lighting [11].

Experimental diets

Brassica oleracea outer leaves, *Musa paradisiaca* leaves, *Megathyrus maximus* and *Stylosanthes guianensis* aerial parts, comprising tender stalks with leaves, were harvested from the surroundings of the experimental station. They were cleaned of soil and other contaminants with clean water, cut into small pieces, dried to 88.4 to 90.2% DM content, milled and incorporated into diets containing maize, soymeal, wheat bran, corn cob, oyster shell, salt, lysine, methionine and premix, at 0, 10, 20 and 30% inclusion levels. The ingredients in the diets were adjusted to achieve CP levels from 16.8 to 17.9%, metabolizable energy of 2989.0 to 3196.0 kcal/kg, and nitrogen free extract of 60.5 to 66.0% DM to meet the requirement of the rabbits. All the diets contained premix of similar composition.

Experimental design

Forty-eight (48) rabbits were randomly allotted on a weight and sex equalization basis to four dietary treatments with 12 rabbits per treatment in a completely randomized design. The treatments were composite diet without forage as control, and concentrate diets of 10, 20 and 30% inclusion levels of *Brassica oleracea* outer leaves. Similar experimental design and conditions were employed concurrently for rabbits fed diets containing *Musa paradisiac* leaves, *Megathyrus maximus* and *Stylosanthes guianensis* aerial parts at 10, 20 and 30% inclusion levels, using a common control diet and rabbits. The animals were acclimated to the experimental conditions and diets for 7 days followed by a 12 - week feeding regime. Salt-licks were supplied during the experiment, and feed and freshwater were provided *ad libitum* [11].

Harvesting of spleen

Eight (8) rabbits (4 males and 4 females) from each treatment group, were randomly selected at the end of the 12-week feeding period. The body weights of the rabbits were taken, after which they were stunned, bled by severing the carotid artery and jugular vein at the level of the atlas vertebra with a sharp knife. They were allowed to bleed for 30 minutes with their heads down, carefully eviscerated, and the internal organs removed. The spleen was separated from the other internal organs, blotted dry of blood and weighed.

Data collection and analysis

The immune organ index, represented by the spleen index, was calculated according to the formula by [7] as: Spleen index = spleen weight/body weight x 100. Data collected, comprising body weight at slaughter and spleen weight of the rabbits, were subjected to one-way analysis of variance (ANOVA) as outlined by the Generalized Linear Model of the GenStat Discovery Edition [14] using the model below: $Y_{ij} = \mu + T_i + E_{ij}$. Y_{ij} is the response variable such as body weight at slaughter, spleen weight and spleen index. μ is the overall mean; T_i is the experimental diets and E_{ij} is the residual error. Least Significant Difference (LSD) Test was used to separate means at ($p < 0.05$) level of significance and the results were expressed as means \pm Standard Error of means (SEM).

Results

The crude protein contents in the diets containing graded levels of *Brassica oleracea* outer leaves, *Musa paradisiaca* leaves, *Megathyrus maximus* and *Stylosanthes guianensis* aerial parts.

Table 1 shows the CP contents in diets containing *Brassica oleracea* outer leaves, *Musa paradisiaca* leaves, *Megathyrus maxi-*

Forage	Forage inclusion level of CP (%DM)			
	0	10	20	30
<i>Brassica oleracea</i>	17.6	17.4	16.8	17.7
<i>Megathyrsus maximus</i>	17.6	17.8	16.8	17.8
<i>Musa paradisiaca</i>	17.6	16.8	16.9	17.7
<i>Stylosanthes guianenses</i>	17.6	17.8	17.6	17.9

Table 1: Dietary crude protein content of graded levels of *Brassica oleracea* outer leaves, *Musa paradisiaca*, *Megathyrsus maximus* and *Stylosanthes guianenses* aerial parts.

mus and *Stylosanthes guianensis* aerial parts. The CP contents in the diets ranged from 16.8 to 17.9% DM. The diets containing 30% inclusion level of *Stylosanthes guianensis* aerial parts had the highest CP contents, while the lowest CP content was found in diets containing 20% inclusion levels of *Brassica oleracea* outer leaves and *Megathyrsus maximus* aerial parts.

Effects of different dietary inclusion levels of *Brassica oleracea* outer leaves on body weight, spleen weight and spleen index values of rabbits

The body weights, spleen weights and spleen indices of rabbits fed diets containing 0, 10, 20 and 30% inclusion levels of *Brassica*

oleracea outer leaves ranged from 2112 to 2314g, 1.12 to 1.25g and 0.05 to 0.06g respectively (Tables 2). No significant ($p > 0.05$) differences were found in the body weights, spleen weights and spleen indices of rabbits fed the various diets. However, the body and spleen weights tended to be lowest in rabbits fed 30% inclusion levels of the leaves, while the spleen index recorded lowest values in rabbits fed 10 and 30% graded levels of the leaves.

Effects of different dietary inclusion levels of *Megathyrsus maximus* aerial parts on body weight, spleen weight and spleen index values of rabbits

Effects of different dietary inclusion levels of *Megathyrsus maximus* aerial parts on body weight, spleen weight and spleen index

Parameter	<i>Brassica oleracea</i> inclusion level (%)				± SEM	p-value
	0	10	20	30		
Body weight at slaughter (g)	2131	2314	2165	2112	99.3	0.493
Spleen weight (g)	1.25	1.25	1.25	1.12	0.27	0.982
Spleen index (g)	0.06	0.05	0.06	0.05	0.01	0.968

Table 2: Body weight, spleen weight and spleen index values of rabbits fed graded levels of *Brassica oleracea* outer leaves.
SEM: Standard error of means.

values of rabbits are shown in table 3. Body weight, spleen weight and index values were not significantly ($p > 0.05$) different among rabbits fed 0, 10, 20 and 30% inclusion levels of *Megathyrsus maximus* aerial parts. However, the body weight tended to be higher for rabbits fed diet containing 20% inclusion level of the leaves, while the spleen weight and index values tended to be higher in rabbits fed 0 and 20% inclusion levels of the aerial parts.

Effects of different dietary inclusion levels of *Musa paradisiaca* leaves on body weight, spleen weight and spleen index values of rabbits.

Table 4 shows that the body weights, spleen weights and spleen indices were similar ($p > 0.05$) among rabbits fed diets containing 0, 10, 20 and 30% inclusion levels of *Musa paradisiaca* leaves,

However, the values tended to be higher in rabbits fed the diet containing 30% inclusion level of the leaves.

Effects of different dietary inclusion levels of *Stylosanthes Guinness* aerial parts on body weight, spleen weight and spleen index values of rabbits

Effects of different dietary inclusion levels of *Stylosanthes guianensis* aerial parts on body weight and spleen values of rabbits are presented in table 5. Body weight, spleen weight and index values were similar ($p > 0.05$) among rabbits fed 0, 10, 20 and 30% inclusion levels of *Stylosanthes guianensis* aerial parts. The body weight, spleen weight and spleen index values tended to be higher in rabbits fed the diet containing 30% inclusion level of the aerial parts of the plant.

Parameter	Megathyrus maximus inclusion level (%)				± SEM	p-value
	0	10	20	30		
Body weight at slaughter (g)	2131	1926	2200	2164	124.1	0.436
Spleen weight (g)	1.25	1.00	1.25	1.00	0.18	0.588
Spleen index (g)	0.06	0.05	0.06	0.05	0.01	0.763

Table 3: Body weight, spleen weight and spleen index values of rabbits fed graded levels of *Megathyrus maximus* aerial parts.
SEM: Standard Error of Means

Parameter	Musa paradisiaca inclusion level (%)				± SEM	p-value
	0	10	20	30		
Body weight at slaughter (g)	2131	2110	2248	2285	77.3	0.339
Spleen weight (g)	1.25	1.25	1.00	1.50	0.23	0.517
Spleen index (g)	0.06	0.06	0.05	0.07	0.011	0.602

Table 4: Body weight, spleen weight and spleen index values of rabbits fed graded levels of *Musa paradisiaca* leaves.
SEM: Standard error of means

Parameter	Stylosanthes guianensis inclusion level (%)				± SEM	p-value
	0	10	20	30		
Body weight at slaughter (g)	2131	2136	2248	2266	125.1	0.806
Spleen weight (g)	1.25	1.00	1.00	1.50	0.19	0.248
Spleen index (g)	0.06	0.05	0.05	0.07	0.015	0.407

Table 5: Body weight, spleen weight and spleen index values of rabbits fed graded levels of *Stylosanthes guianensis* leaves.
SEM: Standard error of means

Discussion

The dietary CP content in the diets containing the graded levels of the *Brassica oleracea* outer leaves, *Musa paradisiaca* leaves, *Megathyrus maximus*, and *Stylosanthes guianenses* aerial parts (16.8 to 17.9% DM) fell within the recommended values of 15 to 18% DM for various classes of rabbits [4]. The rabbits fed diets containing 10% inclusion level of *Brassica oleracea* outer leaves, *Megathyrus maximus* and *Stylosanthes guianenses* aerial parts tended to have the lowest spleen index of 0.05 g, while the highest spleen index of 0.07 g was found in rabbits fed diets containing 30% inclusion level of *Musa paradisiaca* leaves and *Stylosanthes guianenses* aerial parts, which also had the highest dietary CP content of 17.9% DM. The higher than the 16% CP in the various diets fed to the rabbits in the present study enhanced their immune responses [5].

The body weight and spleen weight of 1926 to 2341g and 1.00 to 1.50g respectively obtained for rabbits in the present study were higher than the corresponding weights of 1350g and 0.55g recorded for 20-week-old local Algerian rabbits [12]. However, the spleen indices of 0.05 to 0.07 obtained for rabbits in the pres-

ent study were lower than the calculated spleen index of 0.11 for the rabbits in the study by [12], resulting largely from the higher spleen weights of the rabbits in the current study. The good health exhibited by the rabbits throughout the experimental period resulted from the ability of their spleens in protecting them from invading pathogens [13].

Conclusion

The good health status of the rabbits during the experimental period implies that the functions of the spleen were not adversely affected by the diets fed to the rabbits and suggests that feeding diets containing up to 30% inclusion level of *Brassica oleracea* outer leaves, *Musa paradisiaca* leaves, and *Megathyrus maximus* and *Stylosanthes guianensis* aerial parts to growing rabbits enhance the immune response of the rabbits thereby ensuring good performance and health.

Authors' Contributions

- Doris Yaa Osei conceived and designed the study, conducted data gathering and wrote the article.

- Peter Asiedu designed the study, conducted data gathering, and reviewed the article.
- Felix Owusu Sarkwa designed the study, conducted data gathering, and reviewed the article.
- Eric Timpong-Jones designed the study, conducted data gathering, and reviewed the article.
- David Amedorme conducted data gathering, performed statistical analyses and reviewed the article.

Competing Interests

The authors declare there are no conflicts of interest.

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Ethical Standards

Not applicable as the rabbits were slaughtered under humane conditions.

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