



GROWTH PERFORMANCE AND BLOOD PROFILE OF FEMALE RABBITS FED DIETARY CERIUM OXIDE

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Abstract

Context: Rare earth elements, the 15 lanthanide elements with atomic numbers 57 (lanthanum) through 71 (lutetium), have been reported to be capable of producing reasonable growth-promoting effects in all classes of livestock and at the same time increase milk yield and egg production. However, concentration and type of rare earth element as well as the compositions of individual elements are important factors influencing the performance enhancing effects of REE on animals.

Objective: The effects of inclusion of different dietary concentrations of Cerium oxide (CeO), a Rare Earth Element on growth performance and blood parameters of female rabbits.

Materials and Methods: In an 8-week feeding experiment, 32 growing female rabbits were used. The rabbits were assigned to different dietary concentrations of Cerium oxide (CeO) at 0, 100, 200 and 300 ppm. The initial weight, feed intake and final weight were determined. Blood samples were collected on the last day of the experiment for hematological and serum biochemical analyses.

Results: The daily weight gain, daily dry matter intake and feed conversion ratio significantly increased ($P < 0.05$) with increase in dietary CeO concentrations. The CeO supplemented diets generally had significantly ($P < 0.05$) higher daily weight gain of 7.52 - 10.29 g compared with 6.65 g for those on the control diet. The mean corpuscular hemoglobin concentration, mean corpuscular hemoglobin, leukocyte and the mean differential leukocyte counts were significantly ($P < 0.05$) influenced while the serum biochemical parameters examined, except urea, were not significantly ($P > 0.05$) influenced by the dietary CeO.

Conclusion: This study revealed that diets supplemented with CeO, particularly at 200 ppm concentration, is capable of improving body weight gain, daily dry matter intake and feed conversion ratio in rabbits without any detrimental effect on the blood parameters indicating good health status of the animals.

Keywords: Cerium oxide; growth; hematology; rabbit; Rare earth element; serum biochemistry.

Introduction

Following the ban of all in-feed antibiotics throughout Europe in 2006 as a result of public concerns about the development and dissemination of multi-resistant bacteria, a strong demand for new, efficient, safe and inexpensive feed additive that may satisfy the needs provoked by these changes arose. Rare earth elements (REE) may therefore be a promoting approach. Despite their name, the REEs are not rare. Rare earth elements are 15 lanthanide elements with atomic numbers 57 (lanthanum) through 71 (lutetium), which are in group III A of the periodic table. In China, REEs have been in use for over 50 years as performance enhancers in both crop and animal production (Wan et al. 1998, Redling 2006). Not only the effectiveness but also the safety of REE application has been assessed in China, prior to their commercial utilization (Redling 2006). However, it is not until 1999 that growth promoting effects of rare earth elements were

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recognized beyond China (Rambeck et al. 1999). Considerable numbers of studies using REEs in China are of the opinion that these elements produce reasonable growth-promoting effects in all classes of livestock (Redling 2006) and increase in milk yield and egg production (Schuller 2001). However, these assumptions were not noticed in the tropics except for the reports of (Adu 2005, Adu et al. 2009).

This present study was therefore aimed at evaluating potentials of dietary cerium oxide, a rare earth element, on growth performance, haematology and serum biochemistry of growing female rabbits under tropical conditions, as the concentration and type of rare earth element as well as the compositions of individual rare earth element have been shown to be important factors influencing performance enhancing effects of REE on animals.

Materials and Methods

Experimental site and animals: Thirty-two female rabbits of about 12 weeks of age with an average weight of 1366.56 ± 37.54 g were used in this experiment. The animals were housed individually in a wire-mesh cage in an in-door pen. The study was carried out at the Rabbit Unit of the Teaching and Research Farm, Federal University of Technology, Akure, Nigeria and further analyses were carried out at the Research Laboratory of the Department of Animal Production and Health of the Federal University of Technology, Akure, Nigeria.

Experimental diets: Four experimental diets were formulated: control (diet 1) with non-inclusion of REE, diets 2, 3 and 4 with 100, 200 and 300ppm inclusion levels of cerium oxide, respectively as shown in Table 1. The diets were isocaloric, isonitrogenous and satisfied the nutrient requirements of the animals as recommended by National Research Council (1998).

Table 1. Gross composition (%) of growing rabbit test diets

Ingredients	Diet 1	Diet 2	Diet 3	Diet 4
	Control	100ppm CeO	150ppm CeO	200ppm CeO
Maize	32.10	32.10	32.10	32.10
Wheat offal	41.80	41.80	41.80	41.80
Ground nut cake	3.50	3.50	3.50	3.50
Palm kernel cake	20.00	20.00	20.00	20.00
Oyster shell	1.50	1.50	1.50	1.50
Bone meal	0.25	0.25	0.25	0.25
Lysine	0.15	0.15	0.15	0.15
Methionine	0.05	0.05	0.05	0.05
Vitamin Premix	0.20	0.20	0.20	0.20
Salt	0.45	0.45	0.45	0.45
Cerium oxide [ppm]	-	100	200	300
Calculated Nutrient				
Crude Fibre [%]	10.83	10.83	10.83	10.83
Crude Protein [%]	10.38	10.38	10.38	10.38
ME* [MJ/kg]	2906	2906	2906	2906
Ether Extract	4.52	4.52	4.52	4.52

ME*: Metabolisable Energy

Determination of performance characteristics: The animals were randomly assigned into one of the four groups (eight per treatment) after a 2-week physiological adjustment period. They were provided with fresh, clean water and appropriate feed for eight weeks. Feed intake for each animal was measured daily by differences between the daily feed supplied and left over and changes in live weight of the animals were taken on weekly basis during the experiment.

Hematological and serum biochemical measurement: On the last day of the experiment, blood samples were collected from the ear vein of each animal into two sets of labeled bottles, one set contained Ethyl diaminetetraacetic acid (EDTA), an anti-coagulant, for haematological analyses while the other set without EDTA for serum biochemical analyses. The packed cell volume (PCV), red blood cells (RBC), haemoglobin (Hb), leukocytes, and its differential counts (eosinophil, neutrophil, monocytes, lymphocytes), and blood constants (mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular haemoglobin (MCH)) were determined from the blood samples collected in EDTA bottles as described by Tietz (1995). The sera obtained from the blood samples in the set of bottles without anti-coagulant were decanted after centrifugation and later analysed for serum biochemistry. The serum total protein was determined by the Biuret method (Reinhold 1953) using a commercial kit (Randox Laboratories Ltd, U.K.), while the albumin value was obtained by bromocresol green method (Dumas and Biggs 1971). The globulin and albumin-globulin ratio were determined according to the method of Coles (1986). The serum creatinine and urea nitrogen were estimated by deproteinisation and Urease-Berthelot colorimetric methods, using a commercial kit (Randox Laboratories Ltd., U.K.). Also the free cholesterol was determined by nonane extraction and enzymatic colorimetric methods, respectively using commercial test kits (Quimica Clinica Aplicada, S.A.), while the serum enzymes Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST) were obtained using the Randox Laboratories Ltd, UK test kits. Glucose was determined using blood glucose meter (Accu-Chek®Active, Germany).

Statistical evaluation: The design used for this experiment was Completely Randomized Design (CRD). Data collected were subjected to statistical analysis using one way ANOVA procedure of SAS 2008. The significant treatments were compared using Duncan's multiple range tests of the same software.

Results

Performance characteristics: The performance characteristics of growing rabbits fed varied concentrations of dietary cerium oxide are shown in Table 2. Growth responses were lower in groups fed control diet compared to those fed diets supplemented with Cerium oxide. The results showed significant ($P<0.05$) influence of REE on the final live weight, total weight gain, daily weight gain, total feed intake, daily feed intake and feed conversion ratio. The daily feed intake improved by 14.22, 10.77 and 7.28% while the daily weight gain improved by 38.80, 54.75 and 13.08% for diets 2, 3, and 4 respectively, compared with the control. The feed conversion ratio improved over the control by 17.63, 28.57 and 5.13% for diets 2, 3 and 4 respectively.

Table 2. Performance characteristics of growing female rabbits fed varied concentrations of dietary cerium oxide

Parameters	Diet 1	Diet 2	Diet 3	Diet 4	±SEM
	Control	100ppm	200ppm	300ppm	
Initial live weight [g]	1366.25	1369.63	1362.88	1367.50	37.54
Final live weight [g]	1738.75 ^d	1886.38 ^b	1939.00 ^a	1788.63 ^c	44.00
Total weight gain [g]	372.50 ^d	516.75 ^b	576.13 ^a	421.13 ^c	26.23
Daily weight gain [g]	6.65 ^d	9.23 ^b	10.29 ^a	7.52 ^c	0.47
Total Feed Intake [g]	1670.13 ^d	1907.13 ^a	1849.25 ^b	1791.38 ^c	15.64
Daily dry matter intake [g]	29.82 ^d	34.06 ^a	33.03 ^b	31.99 ^c	0.28
Feed conversion ratio	4.48 ^a	3.69 ^b	3.20 ^c	4.25 ^a	0.40

^{abcd}: Means on same row with different superscripts differ significantly ($P<0.05$). SEM- Standard Error of Mean

Table 3. Haematological values of growing female rabbits fed varied concentrations of dietary cerium oxide

Parameters	Diet 1	Diet 2	Diet 3	Diet 4	±SEM
	Control	100ppm	200ppm	300ppm	
Packed cell volume [%]	33.30	32.08	33.65	33.23	2.73
Haemoglobin [g/dl]	11.70	10.85	11.38	11.60	1.22
Erythrocytes [10 ⁹ /l]	6.05	6.08	7.05	6.48	1.14
MCV [μ ³]	56.09	52.79	48.60	52.15	5.94
MCH [pg/cell]	19.68 ^a	17.86 ^{ab}	16.40 ^b	18.10 ^{ab}	1.95
MCHC [%]	34.80 ^a	33.68 ^b	33.60 ^b	33.48 ^b	0.72
Leukocytes [10 ⁹ /l]	6.70 ^a	4.78 ^{ab}	4.15 ^b	4.55 ^{ab}	1.45
Lymphocytes [%]	65.22 ^d	68.62 ^a	68.43 ^b	67.25 ^c	0.04
Neutrophils [%]	26.42 ^a	23.22 ^d	24.34 ^c	26.15 ^b	0.02
Monocytes [%]	2.39	2.51	2.23	1.98	0.23
Eosinophils [%]	6.42 ^a	5.23 ^b	5.78 ^b	5.27 ^b	0.01

^{abcd}: Means on same row with different superscripts differ significantly (P<0.05) ,

MCV- Mean corpuscular volume, MCHC- Mean corpuscular haemoglobin concentration, MCH- Mean corpuscular haemoglobin, SEM- Standard Error of Mean

Effects on haematology and serum biochemistry: Tables 3 and 4 show the hematological and serum biochemical parameters of growing female rabbits exposed to varied concentrations of dietary cerium oxide. The MCHC, MCH, leukocyte, lymphocytes, neutrophils and eosinophils counts were significantly (P<0.05) influenced by the CeO while all the serum biochemical parameters showed no significant (P>0.05) difference excepturea which was significantly (P<0.05) influenced by the CeO.

Table 4. Serum biochemistry of growing female rabbits fed varied concentrations of dietary cerium oxide

Parameters	Diet 1	Diet 2	Diet 3	Diet 4	±SEM
	Control	100ppm	200ppm	300ppm	
Total protein[g/dl]	10.60	10.38	10.50	10.32	0.34
Albumin [g/dl]	6.25	5.30	4.10	4.25	1.62
Globulin [g/dl]	4.35	5.08	6.40	6.07	1.45
Albumin/Globulin	1.44	1.04	0.64	0.70	0.31
Cholesterol [mg/dl]	35.10	34.31	37.20	34.06	3.40
Creatinine [mg/dl]	1.01	1.03	0.92	1.05	0.21
Glucose [mg/dl]	118.50	134.50	116.00	113.50	17.07
Urea [mg/dl]	2.60 ^b	2.50 ^b	2.70 ^b	3.69 ^a	0.58
ALT [IU/l]	52.40	51.45	52.70	52.48	2.28
AST [IU/l]	60.75	61.55	60.85	61.25	1.41

^{ab}: Means on same row with different superscripts differ significantly (P<0.05)

ALT: Alanine aminotransferase, AST: Aspartate aminotransferase

SEM: Standard Error of Mean

Discussion

The increased percentage daily weight gain (13.08 – 54.75%) and feed conversion ratio (5.13 – 28.57 %) observed in this study are in agreement with the findings of Adu et al.(2009), which showed an improvement of 4.4 - 13.5% and 3.32 - 16.76% for daily weight gain and feed conversion ratio respectively, for rabbits fed dietary cerium oxide. Similar values of 13.26, 5.43 and 8.50% for daily weight gain, feed intake and feed conversion ratio respectively, have been reported in pigs fed dietary REE (Xu et al.1999, Wang and Xu 2003). Also the increase in feed intake observed in this study concurred with the findings of Xu et al. (1999) and He et al. (2010) that dietary rare earth element increased the average daily feed intake in livestock.

The increase in live weight gain and feed intake could be attributed to the fact that the REE improves utilization of nutrients in the diets and also increase the secretion of digestive fluids (He et al.2010, Ou et al. 2000, Xu et al. 2004). Stabilization of intestinal micro-flora has also been reported to possibly explain growth promotion due to rare earth application (Zhang et al. 2000, Ruming et al. 2002). Influence on hormone activities as well as on cell proliferation has also been considered as possible explanation for performance-enhancing effects of rare earth elements (He et al. 2003, He et al. 2006). Some authors also suggested that the combination of antibacterial effects with anti-inflammatory properties, as observed for rare earths, may account for their performance-enhancing effects on animals (Redling 2006).

Though several studies demonstrated performance enhancing effects of rare earths on pigs, there are also some trials in which only little or no effects were observed after rare earths were added to the feed of animals (Gebert et al. 2005). The differences in the results of various researchers could be due to the fact that the impact of rare earths on animal growth differs with individual elements and their concentrations. Though rare earth elements are reported to be a chemically homogenous group sharing a great variety of biochemical characteristics, there are still differences among the individual elements. Lanthanum, for example, unlike cerium, is a cation missing unpaired f-electrons, whereas, cerium may assume two oxidation states, Ce^{3+} and Ce^{4+} .

The insignificant influence of dietary cerium oxide on most of the haematological parameters indicated that the dietary REE do not affect the status of the blood parameters as all the parameters evaluated were still within the normal ranges for rabbits (Mitruka and Rawnsley 1981, Kerr 2008, Jones 2005). The observations in the present study agreed with available reports (He et al. 2001, Adu 2005, Adu et al. 2009) that dietary REE had no negative effect on blood parameters of animals. However, the MCH gives the average haemoglobin content of a single blood cell, and it is reported that its reduction indicates iron deficiency (Schalm 1975). Therefore, its reduction by dietary REE in this study might indicate possible REE interference with iron metabolism while the significant reduction in MCHC values, though within the normal physiological ranges available in literatures (Mitruka and Rawnsley 1981, Kerr 2008, Jones 2005), indicate that dietary REE do not affect the status of red blood cells and the rabbits fed dietary REE could be considered normochromic (Coles 1986).

The significant reduction in the leukocytes, lymphocytes, neutrophils, and eosinophils counts are all indicative of possible interference of dietary REE with the soldiers of the immune system. The values of the serum biochemical parameters evaluated in this study were found to be within the normal ranges for healthy rabbits (Mitruka and Rawnsley 1981, Kerr 2008, Jones 2005) except urea which increased significantly ($P<0.05$).

The result of this study on urea is similar to the reports of He et al. (2003) and Adu et al. (2009) that dietary REE influence the values of blood urea, although an insignificant influence was reported by these authors. The significant increase in urea concentration and the insignificant increase in serum creatinine and globulin observed in this study are all possible impacts of dietary REE on protein metabolism through intermediate

metabolism. He et al. (2003) suggested that REE may form complexes with proteins which in turn may enhance protein uptake or its metabolism (Ou et al. 2000). He et al. (2001) also reported similar results with pigs fed dietary REE.

Conclusion

It can be concluded that dietary REE at 200 ppm concentration is capable of improving the body weight gain, daily dry matter intake and feed conversion ratio without any detrimental effect on the blood parameters indicating good health status of the animals.

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