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THE EFFECT OF DIFFERENT FIBRE AND STARCH DIETARY LEVELS ON HAEMATOLOGY OF POST-WEANING RABBITS

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Abstract

The objective of this study was to evaluate the effect of different fibre and starch dietary levels on the blood parameters of post-weaning rabbits. A control group was fed a post-weaning, growing rabbit diet containing 138 g/kg dry matter (DM) crude fibre and 168 g/kg DM starch. Three rabbit groups were fed diets with 147, 156, or 164 g/kg of DM crude fibre, and three groups were fed diets having 190, 223, or 247 g/kg DM starch. The experiment started with six week-old rabbits and lasted 28 days. Blood glucose levels were, on average, 7% lower in the groups fed fibre-enriched diets ($P = 0.048$) and 10% higher in the rabbit groups fed starch-enriched diets than in the control. The higher the dietary level for fibre and starch diets, the greater the deviation from normal in glucose levels. Total protein levels were not significantly modified, excepting a 13% increase in the globulin fraction of fibre and starch-diet fed groups and a subsequent decrease (8.3% and, respectively 13.8%) in the albumin/globulin ratio, but no differences according to the level of fibre or starch. Compared to the control, blood urea was 17% lower and 10% higher in rabbits fed the high crude fibre high starch diets respectively. Alanine aminotransferase, aspartate aminotransferase, and alkaline phosphatase showed relatively steady activity regardless of diet. The fibre diet levels were positively correlated with protein ($r = 0.87$) and HDL cholesterol ($r = 0.78$), but negatively correlated with glucose ($r = -0.72$), urea ($r = -0.91$) and triglycerides ($r = -0.78$). Contrastingly, dietary starch levels correlated positively with triglycerides ($r = 0.92$) and LDL cholesterol ($r = 0.86$). A negligible relationship was found between cholesterol and fibre ($r = 0.15$) or starch ($r = 0.01$) enriched diets.

Key words*: blood biochemistry, diet, fiber, growing rabbit, starch.*

INTRODUCTION

In rabbit metabolism and nutrition research, diets with differing structures or chemical compositions are often tested. Most dietary studies aim to determine the effects of different levels of protein, starch or fibre on metabolism or nutrition physiology (Eiben et al., 2008; El-Tahan et al., 2012; Wang et al., 2012).
The structure and composition of these diets, which sometimes deviate in high degree from the standard diet, could influence blood parameters and subsequently, the physiological status and welfare of individuals (Atansuyi et al., 2012; Ewuola et al., 2012).
The aim of this study was to reveal the extent to which different starch or fibre dietary levels could influence the haematology of growing rabbits.

MATERIALS AND METHODS

Animals and experimental design

Research was performed on 6-week old New Zealand x Californian (males and females) weaned (at 28 days old) rabbits with an average initial weight of 1.038 ± 54 g.
Two weeks later, the young rabbits were equally divided into seven groups: a control and six experimental groups (10 rabbits in each one).
Rabbits were housed in individual cages (250 × 400 × 300 mm), and maintained at temperatures ranging from 22°C to 26°C. Water and pelleted feeds were provided *ad libitum*.
A light cycle of 12 h from 07:00 h to 19:00.h was maintained throughout the trial.
Diets were calculated according to weaned rabbit nutrient requirements described by De

Blas and Mateos (2010): a control diet and three diets each with increasing crude fibre (FL1, FL2, FL3) and starch (SL1, SL2, SL3) levels respectively (Table 1).

Table 1. Calculated chemical composition of control and experimental post-weaning rabbit group diets

Chemical composition	Control	Fibre enriched diets			Starch enriched diets		
		FL1	FL2	FL3	SL1	SL2	SL3
Dry matter (DM, g/kg)	886	883	882	878	885	892	889
Crude protein (g/kg DM)	127	138	136	135	128	127	119
Crude fibre (g/kg DM)	138	147	156	164	121	123	121
Starch (g/kg DM)	167	149	147	146	190	223	247
Digestible energy (MJ/kg DM)*	10.04	9.93	9.51	9.28	99.34	10.72	10.73

*Calculated according to energetic values of ingredients (tables INRA, 1999)

All diets consisted of maize, barley, oats, wheat bran, full-fat soya, sunflower seed meal, dehydrated alfalfa meal, and a vitamin mineral premix. The diet per kg contained 10,000 IU vitamin A, 2,500 IU vitamin D_3, 80 IU vitamin E acetate, 2.0 mg thiamine, 4.5 mg riboflavin, 3.4 mg pyridoxine, 10 mg pantothenic acid, 20 mg niacin, 0.030 mg vitamin B_{12}, 100 mg Fe, 100 mg Zn, 52.2 mg Mn, 0.4 mg I, and 0.4 mg Se. We enriched the fibre diets by adding wheat straw and bran in partial substitutions of maize and barley (FL1 = 6.0 % fibre enriched diet, FL2 = 12.7% and FL3 = 18.8%). The starch diets were enriched with the addition of maize starch in partial substitutions of maize and dehydrated alfalfa meal (SL1 = 13.7% starch enriched diet, SL2 = 33.4%, SL3 = 47.4%). The trial lasted 28 days with the rabbits being 42 and 70 days old at the start and end of it respectively. Feed intake, body weight, and clinical parameters of the animals were monitored.

Blood sampling and chemical analyses

At the end of the experimental feeding period, blood samples were taken at 10:00 h, from the marginal vein of the ears of each rabbit. Fresh blood samples, from 7-8 randomly selected rabbits from the control and each experimental group, were used to analyse the blood morphological parameters: red blood cells (RBC), haemoglobin, packed cell volume (PCV), mean corpuscular haemoglobin concentration (MCHC), mean corpuscular volume (MCV) and mean corpuscular haemoglobin (MCH) using an ABACUS Junior Vet automatic haematology analyser. The blood serum was obtained after blood coagulation, 10 min of centrifugation at 14,000 G, and decantation. The sera were stored at 4°C, until biochemical processing, for no longer than 24 h. The glucose was measured using an enzymatic method (Ashwell, 1957). Total serum protein and serum albumin were determined according to Gornall et al., (1949). Alkaline phosphatase (ALP, EC 3.1.3.1), aspartate aminotransferase (AST, EC 2.6.1.1), and alanine aminotransferase (ALT, EC 2.6.1.2) activities were assayed by spectrophotometric methods as described by Richterich (1967). Triglycerides, total cholesterol, high-density lipoprotein (HDL)-cholesterol, low-density lipoprotein (LDL)-cholesterol, and urea were measured according to Manta et al. (1976).

Statistical analysis

A two-way ANOVA was performed for the descriptive statistics and statistical comparisons of the blood parameters of each rabbit group using a General Linear Model procedure of SAS (2002). When any null hypothesis was rejected, a Tukey's *post hoc* test was performed to compare the groups. The relationship between starch or fibre enrichment levels and the blood biochemical parameters were statistically analysed using Pearson correlation (r). The significance level of null hypothesis rejection was declared at $P < 0.05$.

RESULTS AND DISCUSSIONS

Results of intake and growth suggested that animals maintained their physiological status regardless of the fibre or starch dietary levels (Table 2). A 13.7% starch supplementation (in SL1) resulted in a 25% and 63% increase in intake and growth respectively. Rabbit groups

fed by starch-enriched forages showed productive performance (weight gain) superior to control. Similar results were reported by Wang et al. (2012) in growing meat rabbits.

Table 2. Daily gain and feed intake of rabbit fed on increasing fibre or starch dietary level

Item	Control (n=10)	FL1 (n=10)	FL2 (n=10)	FL3 (n=10)	SL1 (n=10)	SL2 (n=10)	SL3 (n=10)	P values
Daily gain (g/d)£	21.1	28.4	22.4	21.4	34.3*	30.4	20.6	0.005
Feed intake (g/d)	81.5	87.0	79.0	75.5	101.6*	98.8*	94.6	0.019

n= number of animals; *: significant difference from the control group at the level P < 0.05. £: period = 42 to 70-d old.

Similarly, different levels of fibre and starch enriched diets did not modify the analysed blood morphological parameters of the rabbits (P>0.05, Table 3). Surprisingly, there seemed to be a correlation between RBC count and dietary levels of starch and fibre. These unchanged physiological features suggest that the hematopoietic process remained in the normal range, at least during the experimental period and for the tested levels of fibre or starch. This is also in agreement with Ewuola et al. (2012), who reported no significant modifications of blood parameters in rabbit fed on different dietary levels of water spinach leaf meal, but a good intake, growth and health performances were observed.

Table 3. Morpho-haematological parameters in post-weaning rabbits fed for 28 days on increasing levels of a fibre or starch diet

Item	Con-trol	Fibre enriched diets				*P* values	r	Starch enriched diets				*P* values	r
		FL1	FL2	FL3	SD			SL1	SL2	SL3	SD		
n=	8	8	7	8				10	9	9			
RBC (10^6/µL)	6.58	6.65	6.50	6.36	0.43	0.059	0.81	6.50	6.44	5.89	0.41	0.054	0.86
Haemoglobin (g/dL)	10.6	11.0	10.7	9.8	1.5	0.059	0.66	10.0	10.5	10.2	1.0	0.932	0.22
PCV (%)	36.7	37.7	36.4	35.0	2.3	0.069	0.71	36.4	36.8	36.0	3.0	0.060	0.56
MCHC (g/dL of RBC)	28.8	29.1	29.3	28.0	1.3	0.069	-	27.4	28.6	28.3	3.3	0.491	-
MCV (fL)	55.7	56.6	56.0	55.8	3.0	0.088	-	56.0	57.1	61.1	2.9	0.059	-
MCH (pg)	16.1	16.5	16.4	15.4	0.5	0.831	-	15.3	16.3	17.3	2.5	0.048	-

SD: standard deviation; n = sample size; r = correlation coefficient; RBC = red blood cells; PCV = packet cell volume; MCHC = mean corpuscular haemoglobin concentration; MCV = mean corpuscular volume; MCH = mean corpuscular haemoglobin

Fibre diets had a different effect on blood biochemical parameters than the starch diets. Compared to the control, blood glucose levels reduced by 3% in FL1, 12% in FL2, and 7% in FL3, but increased by 12% in SL1, 7% in SL2 and 11% in SL3 (Table 4).

Maintaining the normal limits of glucose concentrations regardless of diet showed that rabbits were able to control the intermediary metabolism of carbohydrates for the tested fibre or starch dietary levels. Instead, different levels (and higher versus control) of blood glucose were reported by Ewuola et al. (2012) in rabbits fed on different dietary levels of water spinach leaf meal.

Total protein level were only slightly affected by diet with a little increase in the globulin fraction (+13%, in FL1-FL3 and 14% in SL1-SL3 groups), and a subsequent decrease (8% in FL and 14% in SL groups) in the albumin/globulin ratio. A positive correlation between proteins and globulins with fibre level independent variables were found (r = 0.87 for total protein and r = 0.88 for globulins).

This is in contrast with findings by Oboh et al. (2007), where the total protein, albumin and globulin were lowered (*P*<0.05) in rabbits after eight weeks on a high carbohydrate/low fat diet.

Blood urea was lower in all the experimental groups than the control group, regardless of fibre or starch levels. Urea levels decreased in FL and SL groups (−3% in FL1, −22% in FL2 and FL3; −32% in SL1 and SL2, −19% in SL3 [P<0.05]).

There was a negative correlation between urea levels and fibre levels (r = −0.91), and, less so, in starch levels (r = −0.51). The decrease in urea levels could suggest a decrease in protein mobilization and the use of protein as a metabolic fuel (Atansuyi et al., 2012; Wang et al., 2012).

Table 4. Blood biochemical parameters in post-weaning rabbits fed for 28 days on increasing fibre or starch dietary levels

Item	Control	Fibre enriched diets			SD	*P* values	r	Starch enriched diets			SD	*P* value	r
		FL1	FL2	FL3				SL1	SL2	SL3			
n	*10*	*10*	*10*	*10*				*10*	*10*	*10*			
Glucose*	87.0$^{a;b}$	84.6	77.0^{a}	81.4^{b}	2.2	0.048	0.72	99.0	94.0	97.5	6.60	0.032	0.59
Protein**	7.3	7.3	7.5	7.5	4.4	0.509	0.87	7.7	7.5	7.6	0.40	0.090	0.47
Albumin (A)**	3.7	3.7	3.5	3.6	0.6	0.454	0.65	3.7	3.3	3.6	0.33	0.193	0.52
Globulins (G)**	3.4	3.6	4.0	3.9	0.8	1.054	0.88	3.9	3.9	3.8	0.55	0.232	0.62
A/G ratio	1.08	1.02	0.87	0.92	-	-	0.85	0.94	0.85	0.94	-	-	0.69
Urea (mmol/L)	6.9$^{a;b;c}$	6.7	5.5^{a}	5.5^{b}	0.4	0.031	0.91	5.2^{a}	5.2^{b}	5.8^{c}	0.23	0.002	0.51
AST***	50.3	55.4	52.3	49.4	3.0	0.065	0.24	50.0	52.9	46.4	5.0	0.094	0.38
ALT***	38.8	36.9	40.1	42.0	1.7	0.298	0.63	35.9	38.4	33.0	3.7	0.059	0.67
ALP***	104.0	120.4	110.0	96.0	6.4	0.243	0.39	120.1	110.0	114.1	8.6	0.165	0.34
Cholesterol*	89.9	87.7	92.3	89.3	8.8	0.404	0.15	85.0	79.0	92.1	4.4	0.332	0.01
Triglycerides*	60.0$^{a;b}$	48.1^{a}	46.4	48.3	2.7	0.020	0.78	55.4^{b}	54.5	44.0	1.7	0.044	0.92
HDL chol.*	35.0$^{a;b}$	29.9	32.0^{a}	30.0	2.4	0.039	0.61	29.0	33.3^{b}	31.3	2.0	0.047	0.33
LDL chol.*	40.2$^{a;b}$	54.5	50.0^{a}	54.9	6.2	0.033	0.78	46.0^{b}	46.2	48.0	2.3	0.022	0.86

Legend: as in Table 2. Means with the same letter in the same row differ (P<0.05); * mg/dL; ** g/dL; *** IU.

The almost steady-state activities of ALT, AST, and ALP may represent the integrity of the liver health parameters and the ability to maintain a constant protein synthesis activity in the liver regardless of the starch or fibre dietary levels.

Increasing the fibre or starch levels did not change total cholesterol ($P > 0.05$) but affected the lipid metabolism profile. Triglycerides and HDL decreased in FL1 (−20% and −17%, respectively) and SL1 (−8% and −17%, respectively). A strong negative relationship between triglycerides and starch diets was found (r = −0.92). On the other hand, LDL increased in FL1 and in SL1 and correlated positively with dietary starch level independent variables (r = 0.86). Variations in triglycerides, HDL or LDL levels were not necessarily proportional to the changes in fibre or starch dietary levels, which could reveal a high physiological capacity of the rabbit to correct the levels of these blood parameters. No relationship was found between cholesterol and fibre (r = 0.15) or starch (r = 0.01) levels. Instead, direct correlations between triglycerides and pancreatic lipase activity have been reported by Dojana et al. (2012).

CONCLUSIONS

Increasing fibre or starch dietary levels had an effect on the blood biochemical parameters of weaned rabbits. Blood glucose and protein levels were slightly modified. Urea was directly correlated with fibre and starch dietary levels. Triglycerides and HDL decreased in both, fibre and starch enriched diets. Variations in triglycerides, HDL, and LDL levels were not proportional to fibre or starch dietary levels.

REFERENCES

Atansuyi A.J., Akinyemi M.B., Omo-Akeju M.O., Chineke C.A., Aletor V.A., 2012. Haematological indices of rabbits fed graded levels of two fibre sources. In: Proc. 10th World Rabbit Congress, 2012, Sharm El-Sheikh, Egypt, p. 713-717.

Ashwell G., 1957. Colorimetric analysis of sugars. In: Methods in enzymology. Kaplan N.O., Colowick S.P. (ed) Acad Press New York, 3, 85.

De Blas J.C., Mateos G.G., 2010. Feed Formulation. In: De Blas C., Wiseman J. (ed) The Nutrition of the rabbit. CABI Publishing, CAB International, Wallingford, Oxfordshire, UK, p. 222-232.

Dojană N., Codreanu I., Preda C., 2012. Enzyme output capacity of the rabbit exocrine pancreas to adapt differently to food substrate concentration changes. Scientific works, C series, Veterinary Medicine, LVIII(3), p. 134-141.

Eiben C., Gippert T., Gódor-Surmann K., Podmaniczky B., Kustos K., 2008. Influence of dietary protein reduction and enzyme and/or amino acid supplementation on fattening performance of rabbits. In: Proc. 9th World Rabbit Congress, June 2008, Verona, Italy, p. 637-641.

El-Tahan H.M., Amber K., Morsy W.A., 2012. Effect of dietary starch levels on performance and digestibility of growing rabbits. In: Proc. 10th World Rabbit Congress, September, 2012, Sharm El-Sheikh, Egypt, p. 501-505.

Ewuola E.O., Jimoh O.A., Atuma O.V., Soipe O.D., 2012. Haematological and serum biochemical response of growing rabbits fed graded levels of Moringa oleifera leaf meal. In: Proc. 10th World Rabbit Congress, 2012, Sharm El-Sheikh, Egypt, p. 679-683.

Gornall A. G., Bardawil C. J., David M.M., 1949. Determination of serum proteins by means of the biuret reagent J. Biol. Chem., 177: p. 751 -766.

Manta I., Cucuianu M., Benga G., Hodârnău A., 1976. Biochemical methods in clinic laboratory. "Dacia" (ed.), Cluj, Romania.

Oboh H.A., Omofoma C.O., Olumese F.E., Eiya B., 2007. Effects of High Carbohydrate Low Fat Nigerian-Like Diet on Biochemical Indices in Rabbits. Pakistan Journal of Nutrition, 6, p. 399-403.

Richterich R., 1967. Chimie clinique, théoretique et pratique. Doin, Paris.

Wang X., Ma M., Sun L., Wang C., Zhu Y., Li F., 2012. Effects of different protein, fibre and energy levels on growth performance and the development of digestive organs in growing meat rabbit. In: Proc. 10th World Rabbit Congress, Sharm El-Sheikh, Egypt, p. 641-645.

***SAS, 2002. SAS/STAT User's Guide (Version 2.1.3 for Windows). SAS Inst. Inc., Cary, NC, USA.