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RESEARCH ARTICLE

Effects of exogenous enzymes, *Lactobacillus acidophilus* or their combination on feed performance response and carcass characteristics of rabbits fed sugarcane bagasse



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Abstract

The aim of this study was to evaluate the effect of *Lactobacillus acidophilus* (LAC), exogenous enzymes of ZAD[®] (exogenous enzyme preparation) or their combination on feed conversion, and dressing of rabbits fed different treatments of sugarcane bagasse (SCB). Five rations were allotted randomly to five groups of New-Zealand White (NZW) rabbits ($n=10$) with initial live body weight of (838 ± 42.4) g and 5 weeks of age. Rabbits were fed on diets with different sources of fiber as follows, (i) a control diet composed of 100% berseem hay and 0% SCB, (ii) 50% berseem hay and 50% untreated SCB (USCB), (iii) 50% berseem hay and 50% SCB treated with *L. acidophilus* (LAC), (iv) 50% berseem hay and 50% SCB treated with ZAD[®] (ZAD), and (v) 50% berseem hay and 50% SCB treated with a combination of LAC+ZAD[®] (LZ). Treatments of SCB with *L. acidophilus*, ZAD[®] and LAC+ZAD[®] had the highest feed conversion ratio than both USCB and control. The dressing percentage of rabbits that fed the LAC and LZ diets was higher ($P<0.05$) compared with that in the other groups. Performance index (PI) for LAC group was improved ($P<0.05$) compared to that for the other groups; however, PI for USCB group was the lowest ($P<0.05$). It could be concluded that treating SCB with *L. acidophilus*, exogenous enzymes of ZAD[®] or their combination improved feed conversion and performance with more positive effects with *L. acidophilus* than the other treatments.

Keywords: exogenous enzymes, feed conversion, *Lactobacillus acidophilus*, rabbit, sugarcane bagasse

1. Introduction

Rabbits have been recognized to have a very important

role supplying humans with animal protein (Parker 2012). Moreover, rabbit occupies a vital midway between non-ruminants and ruminant animals which can effectively utilize cellulose rich feeds or with ration containing less than 20% grain (Hasanat *et al.* 2006). Rabbits are herbivores that can be successfully raised on diets with high roughage percent such as sugarcane bagasse (Roy *et al.* 2002). Simple biological characteristics, short breeding cycle, high prolificacy and better feed conversion efficiency logically place rabbit just below poultry (Hasanat *et al.* 2006). However, improving feed formulation and strategies for enhancing

Received 30 September, 2013 Accepted 19 May, 2014
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doi: 10.1016/S2095-3119(14)60827-3

the production potential of rabbit especially in tropical and subtropical regions of the world have not been fully exploited (Falcão-e-Cunha et al. 2007; AbuHafsa et al. 2014; Elghandour et al. 2015).

Several studies have been applied to incorporate exogenous enzymes, *Lactobacillus acidophilus* bacteria and their combination in rabbit diets to improve nutrients availability (Falcão-e-Cunha et al. 2007; El-Adawy et al. 2013). Some did not detect any significant effect of enzymes on rabbit performances (Falcão-e-Cunha et al. 2007); others showed lower mortality rate of rabbits besides improved feed conversion ratio (Eiben et al. 2004). Additionally, Rivero et al. (2012) showed that addition of enzymes did not affect the blood contents from red blood cells, haematocrit, haemoglobin, white blood cell, segmented neutrophil, lymphocytes, and plasma protein of lambs. These have been considered as an advantage, as they improve the impact of the supplemented enzymes on cecal fermentation pattern in the rabbit metabolism (AbdI-Rahman et al. 2010). The objective of this study was to evaluate the effect of treating sugarcane bagasse with *L. acidophilus*, the exogenous enzymes of ZAD® or their combination on feed conversion ratio, dressing percentage, performance index and economic evaluation of New-Zealand White rabbits.

2. Results

2.1. Chemical composition

The crude protein in sugarcane bagasse (SCB) treated with *Lactobacillus acidophilus* (LAC), ZAD® (ZAD) and the combination of LAC and ZAD® (LZ) treatments was higher by 35.6, 30.9 and 34.09%, respectively, in comparison with the untreated SCB (USCB) (Table 1).

Table 1 Chemical analysis (% on dry matter basis) of different experimental ingredients

Chemical analysis (%)	Berseem hay	USCB ¹⁾	Experimental ingredients ²⁾		
			LAC	ZAD	LZ
Dry matter (DM)	88.7	89.7			
DM basis (%)					
Organic matter	86.1	95.9	93.8	93.6	93.6
Crude protein	13.7	2.9	4.5	4.2	4.4
Ether extract	2.4	1.2	2.1	1.9	2.1
Crude fibre	28.4	43.8	28.3	33.2	29.9
Neutral detergent fibre	52.3	77.4	65.3	72.2	68.1
Acid detergent fibre	38.8	56.2	59.6	64.5	62.1
Nitrogen free extract	41.7	48.0	58.9	54.3	57.1

¹⁾USCB, untreated sugarcane bagasse diet.

²⁾SCB treated with *Lactobacillus acidophilus* (LAC), ZAD® (ZAD) and the combination of LAC and ZAD® (LZ).

The biological treatment of SCB with the combination of LAC and ZAD (i.e., LZ) has intermediate values of crude fibre (29.9%) and neutral detergent fibre (62.1%) compared to the other two types of treatments (i.e., LAC and ZAD) (Table 1).

2.2. Feed conversion and performance index

Feeding rabbits on diets containing biologically treated SCB lead to an improvement ($P<0.05$) in feed conversion (FC) ratio (Table 2); the highest ($P<0.05$) FC ratio was for the group fed LAC diet, while the lowest FC ratio was for the group fed USCB diet. Results also showed that there were insignificant ($P>0.05$) differences among the FC ratio of groups fed control, ZAD and LZ (Table 2).

Replacing 50% of berseem hay in the rabbit diets with biologically treated SCB led to improve the performance index (PI) for the rabbits. The highest PI was noted in rabbits fed the LAC diet which improved by about 20.11,

Table 2 Effect of feeding control and different experimental diets on the average feed conversion of growing New-Zealand White (NZW) rabbits during the period of 5-12 weeks of age¹⁾

Items	Control groups		Groups of treatments			SE	Significance
	Control	USCB	LAC	ZAD	LZ		
No. of rabbits	10	10	10	10	10		
Feed conversion (g feed g ⁻¹ gain) at							
Weeks 5–6	4.4 b	6.5 a	4.1 b	3.9 bc	3.5 c	0.35	*
Weeks 6–7	3.6	3.8	3.8	4.1	3.7	0.18	NS
Weeks 7–8	3.6 b	4.4 a	3.6 b	3.5 bc	3.3 c	0.13	*
Weeks 8–9	3.5 ab	3.8 a	2.9 c	2.9 c	3.4 b	0.11	*
Weeks 9–10	3.5 b	3.7 a	2.9 c	3.7 b	3.5 ab	0.11	*
Weeks 10–11	4.4 a	4.7 a	3.3 b	4.5 a	4.8 a	0.27	**
Weeks 11–12	3.3 bc	3.9 ab	3.5 b	4.2 a	2.8 c	0.24	*
Weeks 5–8	3.8 ab	4.6 a	3.8 ab	3.7 bc	3.5 c	0.31	*
Weeks 8–12	3.6 ab	4.0 a	3.1 b	3.7 a	3.5 ab	0.24	*
Weeks 5–12	3.8 b	4.3 a	3.4 c	3.8 b	3.5 bc	0.16	*

¹⁾Diets are 100% berseem hay (control), 50% berseem hay and 50% untreated SCB (USCB), 50% berseem hay and 50% SCB treated with *Lactobacillus acidophilus* (LAC), ZAD® (ZAD) and the combination of LAC and ZAD® (LZ). The same as below. Different letters in the same row are significantly differ. **, $P<0.01$; *, $P<0.05$; NS, not significant. The same as below.

27.50 and 23.14% in the 8th, 10th and 12th week of age, respectively, compared with that of the group fed USCB diet. The PI improved ($P<0.05$) by about 16.08% compared with those fed control diet in the 12th week followed by the group fed LZ diet then the group fed ZAD diet, while it was the lowest in the group fed USCB diet (Table 3).

2.3. Carcass characteristics

Studying the impact of using 50% untreated or biologically treated SCB in addition to 50% berseem hay in rabbits diets on carcass characteristics, results of Table 4 showed that the rabbits fed LAC diet recorded the highest dressing percent followed by the group fed LZ diet, while the lowest value was obtained by the group fed USCB diet. There was not any significant difference ($P>0.05$) in the hot and cold carcass percentage between the rabbits fed control, USCB, ZAD and LZ diets. Rabbits fed LAC diet showed significant ($P<0.05$) reduction in its alimentary tract weight (full) as a percentage of the total body weight in comparison with those fed the control and USCB diets with insignificantly decreased alimentary tract weight (empty) as a percentage of the total body weight (Table 4).

3. Discussion

About 50–60% of the feed consumed by the rabbit is used

in the fattening while 40–50% is used in the reproduction. This depends on the weaning date and slaughter weight. Efficient stock, feed quality, mortality rate and the farm management including reproduction efficiency, and slaughter age are other factors affecting feed conversion (Maertens 2009). El-Marakby (2003), Allam *et al.* (2006) and El-Banna *et al.* (2010a) showed an improvement in FC ratio for the animals fed diets contained biologically treated poor quality roughages which may be due to the high live body weight gain with low feed intake.

The highest PI value at the 8th week of age was significantly recorded with the group fed LZ diet, where at the 10th and 12th week of age by those fed LAC diet with an increment of 29.08, 27.5 and 23.14%, respectively, than those fed the USCB diet.

El-Banna *et al.* (2010b) reported that the dressing percentage of rabbits fed potato vines treated with *L. acidophilus* was increased compared with the untreated ones.

A significant ($P<0.05$) reduction in the alimentary tract weight (full) as a percentage of the total body weight was obtained for rabbits fed LAC compared with those fed the control and USCB diets. This result may be due to the lower feed intake obtained with this group. On the other hand, feeding LAC diet insignificantly decreased the alimentary tract weight (empty) as a percentage of the total body weight than feeding the control and USCB diets. Zhang *et al.* (1992), El-Hindawy *et al.* (1994) and Ayyat

Table 3 Effect of feeding control and different experimental diets on performance index (PI %) of growing NZW rabbits during the period of 5–12 weeks of age

Items	Control groups		Groups of treatments			SE	Significance
	Control	USCB	LAC	ZAD	LZ		
No. of rabbits	10	10	10	10	10		
Performance index (PI %) at							
Weeks 5–8	34.6 a	27.8 b	34.8 a	36.1 a	39.2 a	3.8	***
Weeks 8–10	45.2 b	40.9 c	56.4 a	43.9 bc	46.3 b	2.6	.
Weeks 10–12	49.9 b	41.5 c	57.9 a	50.2 b	54.6 ab	4.2	***

***, $P<0.001$.

Table 4 Carcass characteristics of growing NZW rabbits (weight and as % of pre-slaughter weight) as affected by feeding on control and different experimental diets

Items	No. of rabbits	Pre-slaughter weight (g)	Hot carcass		Cold carcass		Alimentary tract		
			g	%	g	%	Full (g)	As % of body weight Full (%)	Empty (%)
Control groups									
Control (berseem hay)	4	1696.0	946.9 b	55.8 b	926.5	54.6 ab	330.9	19.6 bd	6.1
USCB	4	1646.3	885.7 c	53.6 c	865.7	52.4 c	373.7	22.8 a	5.4
Groups of treatments									
LAC	4	1808.7	1038.1 a	57.4 a	1021.7	56.5 a	325.8	18.1 c	5.3
ZAD	4	1864.3	1036.1 a	55.3 b	1016.1	54.3 b	356.9	19.3 bc	5.5
LZ	4	1641.0	920.8 b	56.11 a	912.7	55.6 ab	330.9	18.4 c	5.3
SE		56.1	40.6	0.85	82.6	0.9	6.5	0.9	0.4
Significance		NS	.	.	NS	.	NS	.	NS

et al. (1996) found that carcass traits as a percentage of pre-slaughter weight and chemical analysis of meat were insignificantly affected by the dietary supplementation with probiotics (Lacto-Sacc or Lact-A-Bac) except the pre-slaughter weight, hot and cold carcass which increased significantly in groups which were supplemented with probiotic. Ali (1999) reported that the carcass weight and dressing weight 100 g^{-1} body weight were significantly ($P < 0.05$) higher for broiler chicks fed diet supplemented with Lacto-Sacc. Also, Florou-Paneri et al. (1993) reported that quail chicks fed diets supplemented with Lacto-Sacc had no significant effect on carcass traits.

The total feed cost kg^{-1} gain of rabbits was lower in rabbits fed LAC diet than those fed control and USCB diets by about 17.6 and 19.34%, respectively. This lower value of feed cost kg^{-1} gain may be due to the lower value of feed conversion ratio and the higher average body weight gain.

The higher selling price (L.E.) of the weight gain was obtained in the group fed LAC diet and was increased by 1.75 and 6.11% compared with the control diets (USCB and control), respectively.

4. Conclusion

It could be concluded that the replacement of 50% of berseem hay with *L. acidophilus* bacteria-treated SCB or a combination of *L. acidophilus* and ZAD (LZ) in growing rabbit diets was recommended due to its positive effects on feed intake, which leading to high growth performance and successfully improving chemical composition, carcass characteristics, productivity and economical evaluation.

5. Materials and methods

The study was carried out at the Rabbit Research Laboratory of the Animal and Fish Production Department, Faculty of Agriculture (El-Shatby), Alexandria University, Alexandria Governorate, Egypt.

5.1. Animals, management and growth trial

A total of 50 male weanling New Zealand White rabbits of about 5 weeks old and an average initial body weight of about (838 ± 42.4) g live body weight were allotted randomly into five groups, ten rabbits of each. The rabbits were housed individually in galvanized metal cages under the same managerial conditions in a well-ventilated building. The experiment was conducted to 12-week old rabbits (marketing weight). Live body weight and feed consumption (g d^{-1}) were individually recorded weekly. Each rabbit was

weighed individually before morning feeding.

Fresh water was available all the time using stainless steel nipples available in each cage. The experimental diets were offered to rabbits *ad libitum* in pelleted form. All rabbits were kept under the same hygienic, environmental and managerial conditions.

5.2. Experimental design and treatments

The control diet was formulated to meet the nutrient requirements for growing rabbits according to NRC (1984) and de Blas (1986) in which berseem clover (*Trifolium alexandrinum*) hay was the main source of fibre. The experimental design was a complete randomized design, with five treatments and ten replications each. The treatments comprised with different sources of fiber as follows, (i) a control diet composed of 100% berseem hay and 0% SCB, (ii) 50% berseem hay and 50% untreated SCB (USCB), (iii) 50% berseem hay and 50% SCB treated with *Lactobacillus acidophilus* (LAC), (iv) 50% berseem hay and 50% SCB treated with ZAD® (ZAD), and (v) 50% berseem hay and 50% SCB treated with 25% LAC+25% ZAD® (LZ). All diets were iso-nitrogenous ($(14 \pm 0.6)\%$ CP) and iso-caloric ((2093 ± 50.0) kcal kg^{-1}) and contained similar levels of micro-elements (Table 5).

The treatment of SCB with *L. acidophilus* was carried out according to (El-Adawy et al. 2013). Briefly, skimmed milk was inoculated with *L. acidophilus* at 30°C for 8 d. The SCB was sprayed with the media of 6 L skimmed 200 kg^{-1} SCB plus 50 L water 200 kg^{-1} SCB and supplemented with 5 kg soya bean plus 10 kg molasses 200 kg^{-1} of SCB and mixed in a big bottle. The treated SCB was then compressed in the form of bales which were covered tightly using polyethylene sheets and kept under room temperature (about 26°C) for 47 d.

ZAD® (patent No. 22155) is the biotechnical product made from natural sources to elevated level of cellulase enzyme from anaerobic bacteria which convert the polysaccharide into monosaccharide by specific enzymes according to the procedure of Gado (1997). It contains the following enzymes activity: cellulase, 8.2 U g^{-1} ; hemi-cellulase, 6.2 U g^{-1} ; amylase, 64.4 U g^{-1} and protease, 12.3 U g^{-1} . The treatment of SCB with ZAD® was carried out as follows: The SCB was sprayed with 2 packets of ZAD® dissolved in 50 L water 200 kg^{-1} of SCB and supplemented with 5 kg soya beans plus 10 kg molasses 200 kg^{-1} of SCB and mixed in a big bottle. The treated SCB was compressed into bales and incubated as above. For the treatment with combined *L. acidophilus* and ZAD®, the SCB was sprayed with the media by 6 L skimmed 200 kg^{-1} SCB contained *L. acidophilus* plus 50 L water 200 kg^{-1} SCB plus 2 packets

Table 5 Ingredients and chemical composition (%) of the control and experimental diets

	Control	USCB	LAC	ZAD	LZ
Ingredients					
Berseem hay	30.0	15.0	15.0	15.0	15.0
Sugarcane bagasse	0	15.0	15.0	15.0	15.0
Yellow corn	18.0	18.0	16.0	16.0	15.5
Wheat bran	16.0	16.0	16.0	16.0	16.0
Barley grain	17.0	17.0	17.0	17.0	17.0
Soya bean meal	15.0	15.0	15.0	15.0	15.0
Wheat straw	0.0	0.0	2.0	2.0	2.5
Molasses	2.0	2.0	2.0	2.0	2.0
Limestone	1.1	1.1	1.1	1.1	1.1
Table salt	0.5	0.5	0.5	0.5	0.5
Vitamin and mineral premix ¹⁾	0.2	0.2	0.2	0.2	0.2
DL-Methionine	0.1	0.1	0.1	0.1	0.1
Lysine	0.1	0.1	0.1	0.1	0.1
Chemical composition	87.6	88.3	88.5	88.4	88.6
Crude protein	15.4	13.9	14.4	14.2	14.2
Ether extract	3.5	3.2	3.4	3.3	3.4
Crude fibre	13.7	14.9	14.3	14.5	14.4
Nitrogen free extract	55.1	56.2	56.5	56.4	56.5
Digestible energy (Kcal kg ⁻¹ feed DM) ²⁾	2016.4	2074.8	2137.8	2102.4	2134.5

¹⁾ Vitamins and mineral premix per kilogram contained: V_A 2 000 000 IU, V_{D3} 150 000 IU, V_K 0.33 mg, V_{B1} 0.33 mg, V_{B2} 1.0 g, V_{B6} 0.33 g, V_{B12} 1.7 mg, pantothenic acid 3.33 g, biotin 33 mg, folic acid 0.83 g, choline chloride 200 mg, Zn 11.7 g, Mn 5.0 g, Fe 12.5 g, Mg 66.7 mg, Se 16.6 mg, Co 1.33 mg, Cu 0.5 g, I 16.6 mg, and antioxidant 10.0 g.

²⁾ Digestible energy (Kcal kg⁻¹ feed DM) was calculated.

of ZAD[®] products and supplemented with 5 kg soya bean plus 10 kg molasses 200 kg⁻¹ of SCB and mixed in a big bottle. The treated SCB was compressed into bales and incubated as above. The SCB was then dried overnight in a forced air oven at 75°C until a constant weight, and then stored in polyethylene bags until assayed.

5.3. Analytical procedures

Samples were ground to pass a 1-mm screen on a model 4 Wiley Mill. Samples were analyzed for dry matter (method #930.15), N (method #954.01), and ether extract (method #920.39), according to AOAC (1997). Crude fibre and acid detergent fibre were determined using Ankom Technology (AOAC 1997; method #973.18). Neutral detergent fibre analyses included a heat stable amylase (Van Soest *et al.* 1991) and were expressed both inclusive and exclusive of residual ash. Ash was determined by incineration at 550°C for 3 h.

5.4. Carcass characteristics trail

Twenty rabbits (taken randomly) were used for all experimental groups (four rabbits×five groups). The rabbits were starved before slaughter for 24 h. Each rabbit was weighed exactly before slaughter, and after hemodialysis, the rabbits were reweighed once again. After that, the fur and the internal organs were removed and the rabbits were reweighed again (hot carcass). Then the carcass was saved in the

fridge till the next day (24 h) to reweigh again (cold carcass).

5.5. Statistical analysis

Data were analysed by general linear model (GLM) procedure (SAS 2000). Data were analysed by adapting the following model:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where, Y_{ij} means the observation on the i th treatment; μ means overall mean; T_i means effect of the i th treatment; e_{ij} means random experimental error; means separation was performed using Duncan multiple range test.

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(Managing editor ZHANG Juan)