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HISTOLOGICAL AND HISTOCHEMICAL EVALUATION OF THE SMALL INTESTINE AND CAECAL APPENDIX OF RABBITS FED TRITICALE-BASED DIETS WITH ADDED XYLANASE

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Abstract: The inclusion of triticale in rabbit diets can negatively affect production parameters due to the presence of arabinoxylans that increase the viscosity of intestinal contents, making digestion and absorption of nutrients difficult. Therefore, the addition of the enzyme xylanase can improve intestinal morphology and the digestive process. This study aimed to conduct histological and histochemical evaluations of the small intestine and caecal appendix in growing-finishing rabbits fed diets containing 14% triticale supplemented with varying levels of xylanase. The rabbits had an initial average live weight of 822 g and reached a final weight of 2097 g. For this purpose, forty 35-d-old male rabbits crossbred New Zealand × California were used. Rabbits were housed in individual cages and randomly divided into four experimental treatments (xylanase at doses of 0, 4000, 8000 and 12 000 xylanase units (XU)/kg of dry matter). At the end of the experimental period, rabbits were slaughtered at seventy days of age and samples were taken from the duodenum, jejunum, ileum and caecal appendix. Samples were stained using the standard Haematoxylin-Eosin (HE) technique for the histological evaluation and Alcian Blue (PAS) for the histochemical evaluation. The addition of xylanase linearly increased the height of villi in the duodenum, jejunum and ileum, while villi width was linearly reduced in jejunum and increased in ileum. Crypt depth was linearly reduced by xylanase dose in duodenum, while the response was linear and quadratic in jejunum and ileum. The villi height to crypt depth ratio was linearly increased by the xylanase dose in the duodenum and jejunum, although the effect was quadratic in the ileum. The dome height of the caecal appendix increased linearly with the addition of xylanase. Epithelial mucus count was linearly reduced by xylanase dose in the base of the duodenum, jejunum and caecal appendix, although this linear reduction was only observed in the villi of the jejunum and in the caecal appendix. In conclusion, the addition of xylanase positively affected the histological and histochemical characteristics of the small intestine and caecal appendix, so its use could improve digestive and productive performance in rabbits.

Key Words: rabbit, diet, triticale, xylanase, histology, intestinal villi, caecal appendix.

INTRODUCTION

The functioning of the digestive system is influenced by the quantity and quality of the feed consumed, the type of ingredients that the feed is composed of, the digestion and absorption of nutrients and the microbiota resident in the intestine. When an adequate balance of these factors is achieved, it is possible to achieve a good productive performance (Makovicky *et al.*, 2014). In rabbits, the post-weaning period is one of the most critical phases due to the immaturity of the digestive tract, which makes them more sensitive to digestive diseases of bacterial, parasitic and viral origin (Romero *et al.*, 2010). Therefore, feeding strategies focus mainly on promoting digestive maturation

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and the immune system associated with the gastrointestinal tract, in addition to the establishment of a beneficial microbiota, through the use of balanced diets of high digestibility and including additives that improve the histology and physiology of the intestine (Gidenne and García, 2006). However, under practical conditions there are feeds that can be integrated into rabbit diets, but their use is limited by the presence of anti-nutritional factors. Triticale (x *Triticosecale* Wittm.) is an hybrid cereal available in several regions of the world and has established itself as a viable option for the livestock sector due to its nutritional properties, representing an energy source with starch content that varies from 63 to 69%, neutral detergent fibre that ranges between 8 to 13.8% and crude protein that varies from 9 to 20% (Lozano del Río *et al.*, 2004; McGoverin *et al.*, 2011; Rakha *et al.*, 2011), with the potential to integrate it into fattening rabbit diets. However, its content of arabinoxylans (6.85%) and β-glucans (0.7%) may limit its use because these components act as anti-nutritional factors by increasing viscosity and reducing nutrient absorption (Rakha *et al.*, 2011), while also causing morphological alterations in the digestive tract (Wang *et al.*, 2021). In this context, previous studies have shown that the supplementation of enzymes such as xylanase in diets containing high concentrations of non-starch polysaccharides improves the efficiency of feed utilization in poultry and pigs (Mendes *et al.*, 2013; Wang *et al.*, 2021). Therefore, the aim of the present work was to evaluate histologically and histochemically the small intestine and caecal appendix of rabbits in the growth-finishing stage fed with diets containing triticale with added xylanase.

MATERIAL AND METHODS

Animals and treatments

The experimental research protocol was approved by the ethics committee of the Faculty of Veterinary Medicine and Zootechnics, Autonomous University of the State of Mexico with registration number 03/2022 CICUAL-DISP. Forty male New Zealand × California crossbreed rabbits (*Oryctolagus cuniculus;* 35-d-old) were used. Rabbits were housed in individual cages (40 × 40 × 105 cm) of galvanized steel containing their respective feeder and drinker, in the rabbit experimental area of the Faculty of Veterinary Medicine and Zootechnics of the Autonomous University of the State of Mexico, located in Cerrillo Piedras Blancas, Toluca, State of Mexico. The duration of the experimental period was 35 days, where the rabbits started at 35 d of age and were randomly distributed and assigned to one of the four experimental diets, with *ad libitum* access to food and water during the whole experimental period; the average daily feed intake was reported by Galeano *et al.* (2024). The basal diet and calculated chemical composition are shown in Table 1. The composition of the basal diet (Table 1) was calculated according to Villamide *et al*. (2020). The diets were formulated to satisfy the dietary requirements of fattening rabbits according to de Blas and Mateos (2020). Diets were supplied in the form of 2.5 mm diameter pellets. The treatments consisted of the addition of xylanase to the basal diet at doses of 0, 4000, 8000 and 12 000 xylanase units (XU)/kg of dry matter. The xylanase enzyme (XilaBlend 6X, Enzimas y Productos Químicos, S.A. de C.V, Mexico) used in the present study is of fungal origin (*Aspergillus niger*), being of the endo-1,4-β xylanase type with a minimum guaranteed enzymatic activity of 6000 XU/g, where each XU corresponds to the amount of enzyme that releases one nmol of reducing sugar, such as xylose, from xylan per second. Enzyme was stored at room temperature and in a dry place until the food was mixed and pelletized.

Sample collection

At the end of the experimental period, when rabbits reached 70 d of age and an average weight of 2097 g, they were slaughtered according to the Mexican Official Standard procedures (NOM-033-SAG/ZOO-2014). Subsequently, tissue samples were collected from the duodenum, jejunum, ileum and caecal appendix for histological examination. Histological and histochemical staining

The collected tissue samples were fixed in 10% formalin, consecutively washed with water to remove excess fixative and dehydrated in alcohol solutions with progressive concentration gradients (70, 80, 92, 98 and 100°). Once dehydrated, the tissues were embedded in paraffin and then cut at 4 μm thickness on the microtome (Leica RM2125 RTS). The sections obtained were placed on slides, the paraffin was removed and they were rehydrated and then stained using the standard HE technique for histological evaluation and Alcian Blue (PAS) for histochemical evaluation. The slides were examined with an optical microscope (Leica DM1000) (Aguilar, 2008).

Measurement of histological and histochemical sections

The height of the villi was evaluated in the sections of the duodenum, jejunum and ileum, measuring from the top of the villi to the transition of the crypt; the width of the villi was measured at half the height; the depth of the crypt was defined as the invagination between two villi; the villi height:crypt depth ratio was calculated by dividing the height of the villi by the depth of the contiguous crypt and the area of the intestinal villi was calculated taking into account that the villus is considered as a cylindrical structure, using the following formula described by Nain *et al.* (2012):

With respect to the histological section of the caecal appendix, the height and width of the dome was measured, which corresponds to the pyramidal structure (Valdivia *et al.*, 2007). In samples stained with PAS, goblet cells were counted per villus and base of villi in each tissue in the sections of the duodenum, jejunum, ileum and caecal appendix.

Statistical analysis

The individual rabbit was the experimental unit. The data were analysed applying a completely randomized experimental design, where the collected data were processed with an analysis of variance according to the general linear model procedure with the help of the SAS statistical package (2002), under the following statistical model:

$$Yij = \beta0 + \beta1\ xi + \mu ij$$

Where Yij is the dependent variable being evaluated, β0, β1 the regression parameters, xi the xylanase level and µij the random error. Before performing the statistical analysis on the studied variables, the assumptions of normality and homogeneity of variance were verified. When statistical differences were detected, the comparison of means was carried out using the Tukey method, considering that $P<0.05$ indicates a statistically significant difference (Steel *et al.*, 1997), and linear and quadratic effects were determined by polynomial orthogonal contrasts with a significance level of $P<0.05$.

Table 1: Ingredient and nutrient composition (as feed) of the experimental diet.

Ingredients	%
Alfalfa	40.00
Triticale	14.00
Soybean meal	10.50
Wheat bran	14.00
Oat hay	16.00
Premix[1]	2.50
Nucleus[2]	0.46
Vegetable oil	2.70
Calcium carbonate	0.20
Chemical composition	
Dry matter (%)	91.11
Digestible energy (Mcal/kg)	2.500
Crude protein (%)	16.00
Neutral detergent fibre (%)	34.68
Acid detergent fibre (%)	19.50
Ether extract (%)	4.50
Calcium (%)	0.86
Phosphorus (%)	0.40
Starch (%)	12.70

[1]Premix composition per kg diet: Sodium (0.03%) and potassium (2.42%), vitamin A: 10 000 IU; vitamin D3: 2000 IU; vitamin E: 50 mg; vitamin K3: 2.5 mg; vitamin B1: 5 mg; vitamin B2: 10 mg; nicotinic acid: 20 mg, pantothenic acid: 50 mg; folic acid: 2.5 mg; vitamin B12: 1 mg; choline chloride: 400 mg; Fe: 100 mg; Zn:50 mg; Cu: 40 mg; Mn: 30 mg; I: 0.5 mg; Se: 0.05 mg; $CaHPO_4$: 15 000 mg.

[2]Nucleus composition per 100 kg of diet: *Saccharomyces cerevisiae* 50 g, Zeolite 100 g, Pellet Compactor 300 g, Coccidiostat 33 g, Calcium carbonate 1100 g, Betaine 100 g, Phytase 10 g, Calcium 14.28%, Phosphorus 0.026%, ash 46.34%.

RESULTS

Histological structure of the small intestine and caecal appendix

The sections of the intestinal villi are seen in Figure 1. The dimensions of the intestinal villi were affected ($P<0.05$) by the addition of xylanase (Table 2). In the duodenum section, an ascending linear effect ($P=0.004$) was observed in the height of the villi, with villi being longer ($P<0.05$) in the rabbits fed with 8000 and 12000 XU/kg of xylanase as compared with doses of 0 and 4000 XU/kg. No significant differences among doses of xylanase were for crypt depth ($P>0.05$). Nevertheless, a descending linear effect ($P=0.025$) was exhibited, since the greater the amount of xylanase, the lower the crypt depth.

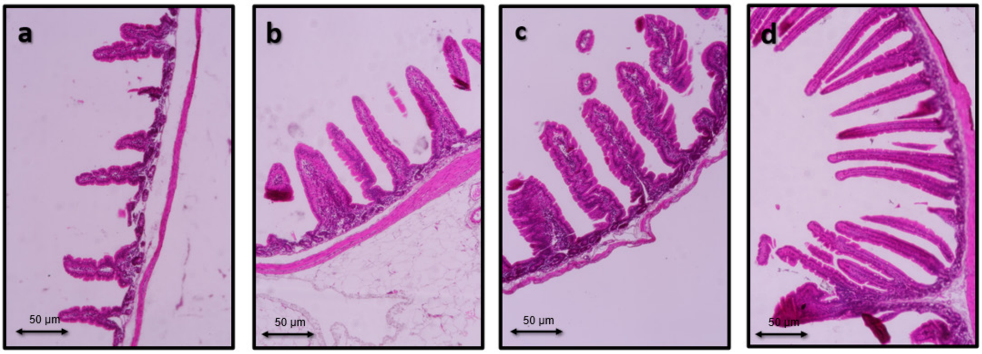

Figure 1: Histological images of the intestinal villi of the duodenum in New Zealand × California rabbits at 70 d of age. Rabbits were fed the diet containing 140 g/kg triticale with added xylanase (from 35 to 70 d of age). Xylanase dietary doses: a) 0 XU/kg, b) 4000 XU/kg, c) 8000 XU/kg, d) 12000 XU/kg. XU: xylanase units.

As concerns the villi height:crypt depth ratio, an ascending linear effect (P=0.001) of the xylanase dose was also observed, the ratio being higher (P<0.05) in rabbits fed with the doses of 8000 and 12 000 XU/kg as compared with control rabbits fed the diet devoid of xylanase.

In the jejunum section, an ascending linear effect was found in the measurements of villi height (P=0.017) and the villi height:crypt depth ratio (P=0.001), and a quadratic effect of the dose for the variables: villi width (P=0.007), crypt depth (P=0.003) and calculated villi area (P=0.025). The height of the villi of the jejunum was higher (P<0.05) at the

Table 2: Measurement of intestinal villi sections of rabbits fed the diet containing 140 g/kg triticale with added xylanase.

	Dietary dose of xylanase (XU/kg)					*P*-Value		
	0	4000	8000	12000	SEM	Tx	Linear	Quadratic
Duodenum								
Villi height (µm)	1459.80[a]	1573.78[a]	1818.14[b]	1811.93[b]	93.662	0.025	0.004	0.526
Villi width (µm)	275.49	280.02	291.54	270.76	17.035	0.844	0.972	0.464
Crypt depth (µm)	393.46	400.22	367.75	332.08	20.441	0.101	0.025	0.308
V/C ratio	3.90[a]	4.10[ab]	5.15[bc]	5.75[c]	0.272	0.001	0.001	0.475
Area (mm^2)	1275.896	1381.346	1684.174	1550.487	139.643	0.195	0.082	0.399
Jejunum								
Villi height (µm)	1571.90[ab]	1498.30[a]	1631.20[ab]	1988.80[b]	122.355	0.040	0.017	0.089
Villi width (µm)	404.95[b]	359.27[b]	258.89[a]	284.65[a]	12.322	0.001	0.001	0.007
Crypt depth (µm)	449.39[b]	314.90[a]	272.04[a]	308.59[a]	25.859	0.001	0.001	0.003
V/C ratio	3.90[a]	4.71[ab]	5.89[b]	6.28[b]	0.418	0.001	0.001	0.619
Area (mm^2)	2306.783[b]	1892.618[ab]	1443.841[a]	1755.640[ab]	152.873	0.004	0.005	0.025
Ileum								
Villi height (µm)	952.20[a]	1257.70[c]	1117.10[b]	1246.80[c]	79.315	0.037	0.045	0.277
Villi width (µm)	245.55[a]	270.79[ab]	299.35[bc]	329.31[c]	11.992	0.001	0.001	0.846
Crypt depth (µm)	202.67[a]	387.58[b]	303.84[ab]	243.94[a]	28.760	0.001	0.758	0.001
V/C ratio	4.94[ab]	3.88[a]	4.14[ab]	5.34[b]	0.336	0.0158	0.335	0.002
Area (mm^2)	737.493[a]	1291.105[b]	1150.081[ab]	1277.756[b]	129.912	0.017	0.017	0.112

SEM: standard error of the mean. n=10 per treatment. Tx: Treatments. XU: xylanase units.
[a,b] Means in the same row with different superscripts are different (P<0.05).
V/C: Villi height/crypt depth ratio.

Table 3: Measurements in caecal appendix slices of the dome area of rabbits fed the diet containing 140 g/kg triticale with added xylanase.

	Dietary dose of xylanase (XU/kg)				SEM	*P*-Value		
	0	4000	8000	12000		Tx	Linear	Quadratic
Dome width (μm)	1221.40	1325.80	1498.40	1432.10	123.081	0.424	0.159	0.496
Dome height (μm)	797.92[a]	961.85[ab]	1092.77[b]	1112.01[b]	644.568	0.009	0.001	0.275

SEM: standard error of the mean. n=10 per treatment. Tx: Treatments. XU: xylanase units.
[a,b] Means in the same row with different superscripts are different ($P<0.05$).

dose of 12 000 XU/kg, in contrast to the rabbits fed with 4000 XU/kg, which had a lower and similar height in the rabbits fed with the dose of 0 and 8000 XU/kg. The depth of the jejunal crypt was lower ($P<0.05$) in rabbits fed with any of the doses of xylanase compared to the 0 XU/kg dose. On the contrary, the ratio of villi height:crypt depth was higher ($P<0.05$) in rabbits fed with the xylanase enzyme and lower in the 0 XU/kg treatment. The calculated area was lower ($P<0.05$) in the 8000 XU/kg treatment compared to the treatment without xylanase.

With respect to the ileum section, it was observed that the height of the villi was higher ($P<0.05$) in the rabbits fed with 4000 and 12 000 XU/kg, followed by the rabbits fed with 8000 XU/kg and a lower height in the dose control, reporting a linear effect ($P=0.045$) ascending in the height of the villi; in the measurement of the width of the villi, an ascending linear effect ($P=0.001$) was also observed, with the width being higher ($P<0.05$) with the 12 000 XU/kg dose compared to dose 0. Regarding the depth of the crypt, this was higher ($P<0.05$) in the ileum of rabbits to which the dose of 4000 XU/kg was added; compared to rabbits with the doses of 0 and 12 000 XU/kg, a quadratic effect ($P=0.001$) of the xylanase dose on crypt depth was observed.

The villi height:crypt depth ratio was higher ($P<0.05$) in rabbits with the 12 000 XU/kg dose, compared to the dose of 4000 XU/kg. The calculated villi area of the ileum was higher ($P<0.05$) in rabbits with the doses of 4000 and 12 000 XU/kg compared to dose 0, but similar with the 8000 XU/kg dose, and the nature of the effect was linear ($P=0.017$) ascending.

Table 3 shows the measurement of the height of the dome of the caecal appendix that was affected in an ascending linear manner ($P=0.001$) by the xylanase dose. Rabbits fed diets with 8000 and 12000 XU/g of xylanase added showed the largest ($P<0.005$) dome area as compared with rabbits receiving the 0 dose. Figure 2 shows the histological sections of the caecal appendix in New Zealand × California rabbits at 70 d of age.

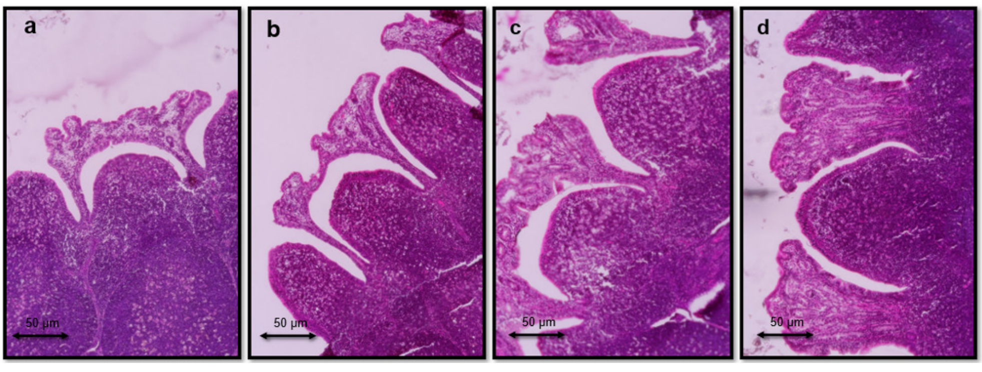

Figure 2: Histological images of the caecal appendix of New Zealand × California rabbits at 70 d of age (10x). Rabbits were fed the diet containing 140 g/kg triticale with added xylanase (from 35 to 70 d of age). Xylanase dietary doses: a) 0 XU/kg, b) 4000 XU/kg, c) 8000 XU/kg, d) 12000 XU/kg. The pyramidal structure corresponds to the dome. XU: xylanase units.

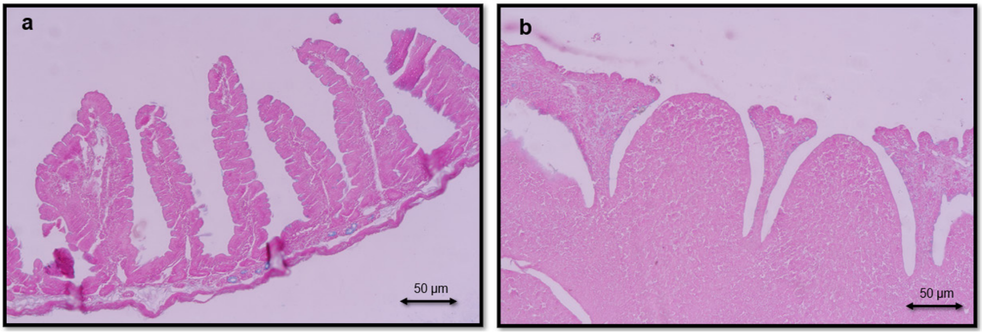

Figure 3: Histochemical images of intestinal villi (a) and caecal appendix (b) in New Zealand × California rabbits at 70 d of age. Rabbits were fed the diet containing 140 g/kg triticale with added xylanase (from 35 to 70 d of age). Counting of blue goblet cells.

Histochemical counting of the small intestine and caecal appendix

Figure 3 shows the study histochemical results for villi in the small intestine and in the caecal appendix with PAS staining for counting goblet cells from rabbits fed diets with added xylanase. Table 4 shows that the goblet cell count in each section of the small intestine was affected ($P<0.05$) by the dose of xylanase, where the 4000 XU/kg dose was the one that generated the highest count in the villi of the duodenum with respect to the 8000 XU/kg, while at the base of the hair the count was affected linearly (P=0.001) descending, and equally higher ($P<0.05$) with the addition of 4000 XU/kg, compared to the doses of 12 000 XU/kg, while in the jejunum section the goblet cell count was higher ($P<0.05$) in the 0 XU/kg treatment, in both the villi and the base, with respect to the different doses of xylanase that were used, and the dose effect was quadratic (P=0.037, P=0.016).

In the case of the ileum section, significant differences ($P<0.05$) were also observed in the count of goblet cells at the base of the villi, being higher in the rabbits fed with the dose of 4000 XU/kg compared to the rabbits with the diet without xylanase and similar in rabbits with the dose of 8000 XU/kg, also observing a quadratic effect (P=0.001)

Table 4: Counts of goblet cells present in the intestinal villi of the small intestine and caecal appendix of rabbits fed the diet containing 140 g/kg triticale with added xylanase.

	Dietary dose of xylanase (XU/kg)					*P*-Value		
	0	4000	8000	12 000	SEM	Tx	Linear	Quadratic
Duodenum								
Villi	6.64^{ab}	8.85^{b}	5.50^{a}	6.83^{ab}	0.657	0.011	0.350	0.506
Base of villi	2.29^{bc}	2.70^{c}	1.51^{ab}	1.36^{b}	0.213	0.001	0.001	0.195
Jejunum								
Villi	11.37^{b}	6.87^{a}	7.12^{a}	5.79^{a}	0.722	0.001	0.001	0.037
Base of villi	3.64^{b}	1.34^{a}	2.07^{a}	1.50^{a}	0.338	0.001	0.001	0.016
Ileum								
Villi	5.80	6.41	6.13	5.97	0.357	0.662	0.883	0.288
Base of villi	1.38^{a}	1.52^{c}	1.46^{bc}	1.44^{ab}	0.017	0.001	0.111	0.001
Cecil appendix								
Villi	0.62^{b}	0.24^{a}	0.30^{a}	0.32^{a}	0.076	0.006	0.018	0.013
Base of villi	9.88	8.41	7.82	7.44	0.826	0.194	0.041	0.515

SEM: standard error of the mean. n=10 per treatment. Tx: Treatments. XU: xylanase units.
[a,b] Means in the same row with different superscripts are different ($P<0.05$).

of the dose for this variable. In the caecal appendix, significant differences (P<0.05) were observed in the count of goblet cells in the villi, being higher in rabbits fed with the diet without xylanase compared to the three doses of xylanase. Furthermore, the dose effect was quadratic (P=0.013) in the villi count and linear (P=0.041) descending in the goblet cell count at the base.

DISCUSSION

In rabbits, there are no reports showing the effect of the inclusion of xylanase in diets of fattening rabbits containing triticale on the morphology of the small intestine and the caecal appendix.

Histological structure of the small intestine and caecal appendix

Intestinal morphology is the main indicator of intestinal health (Laudadio et al., 2012). Morphological alterations of the small intestine can affect the area of nutrient absorption and therefore their transport on this surface (Seyyedin and Nazem, 2017). The present study shows that adding the enzyme xylanase to diets for fattening rabbits that contained 14% triticale favoured the growth (height and width) of the villi of the duodenum, jejunum and ileum in a linear manner. This result coincides with that of Luo *et al.* (2009), who reported that in broiler chickens fed diets containing 40% wheat, the addition of xylanase (500 and 1000 XU/kg of feed) improved villi height and crypt depth in the small intestine, results that could be attributed to the fact that xylanase can degrade non-starch polysaccharides and thereby counteract the negative effects on intestinal morphology, reducing viscosity in the small intestine, thus improving the rate of absorption and digestion of nutrients (Šimić *et al.*, 2023). Donaldson *et al.* (2021) reported that rye can be added to broiler diets as an alternative source of energy; moreover, adding xylanase improved the morphology of the intestinal mucosa, increased the absorption surface area and the proportion of villi and crypts. Short-chain fatty acids (SCFAs), particularly butyric acid, have been shown to influence intestinal morphology (Peng *et al.*, 2009; Fang *et al.*, 2020). Carbohydrates, including xylanase, can enhance intestinal morphology by stimulating the growth of short chain fatty acids producing bacteria through two mechanisms: generating fermentable oligosaccharides and decreasing other fermentable substrates in the intestine's distal region (Patience *et al.*, 2022). The ratio of the height of the villi and the depth of the crypts of the three segments was increased by adding the enzyme xylanase, due to the activity of the enzyme in the different sections of the intestine. The enzyme added in the present study has its maximum enzymatic activity at a pH of 4.8 to 6.4, these values being found at the beginning of the duodenum. However, in the jejunum and ileum the pH changed, becoming slightly more alkaline (results reported in Galeano *et al.*, 2024), therefore, less growth of intestinal villi was observed. Likewise, in the height of the dome of the caecal appendix, there was an increase in the treatments containing xylanase; this effect could be due to the fact that there are microorganisms from the Firmicutes and Bacteroidetes phyla that are capable of producing xylanase and that act at a different pH than the enzyme added to the treatments (Biddle *et al.*, 2013; Frese *et al.*, 2015; Cotozzolo *et al.*, 2020). In the present study, the difference in villi growth could be due to the activity of the enzyme depending on its origin. That is, the added enzyme acted in the duodenum and the enzyme produced in the caecum had its greatest activity in the caecal appendix. Although the linear ratio of xylanase dose with the height of the dome of the caecal appendix could be related to the breakdown of arabinoxylan chains into shorter oligosaccharides that serve as substrates for the growth of bacteria producing SCFAs which helps to raise the concentration of these metabolites, which are known to improve the morphology and physiology of microvilli (Boontiam *et al.*, 2022).

Histochemical counting of the small intestine and caecal appendix

Maturation of the gastrointestinal tract in rabbits after weaning is determined by the early consumption of solid feed, the secretion of enzymes in the gastrointestinal tract and the proper functioning of the caecum, since the latter does not yet develop properly at the time of weaning, and it is not until 42 d of age that the caecal appendix reaches maturity (Castellini *et al.*, 2007). In rabbits at 30 d of age, the duodenal wall becomes histologically more mature with wide villi and PAS-AB positive goblet cells begin to be observed and, at 60 d of age, the duodenal villi become wider, and the presence of goblet cells is evenly distributed along the villi, being active in the secretion of mucus (Elnasharty *et al.*, 2013).

The mucin secreted by the goblet cells, especially in the openings of the crypts in the different regions of the intestine, is part of the immune system associated with the rabbit's gastrointestinal tract and is a fundamental part of the growth of microbial biofilms in the intestine that help maintain a balance of beneficial bacteria and help protect the animal against harmful pathogens, as they function as a protective barrier (Smith *et al.*, 2009; Kim and Khan, 2013; Pelaseyed *et al.*, 2014).

Feeding ingredients with high levels of xylan and arabinoxylans present in triticale causes increased viscosity in the digestive tract (Boontiam *et al.*, 2022), which is the reason why a higher number of goblet cell counts were observed in the intestinal villi of the small intestine and caecal appendix of rabbits that did not have xylanase added, as these goblet cells are responsible for the production and preservation of the protective mucous layer through the synthesis and secretion of mucins (Pelaseyed *et al.*, 2014).

CONCLUSION

The addition of xylanase enhances both histological and histochemical parameters of the small intestine and caecal appendix in growing-finishing rabbits fed triticale-based diets. These improvements in intestinal morphology suggest enhanced digestive capacity and nutrient absorption, which may lead to better productive performance in commercial rabbit production.

Authors contribution: Galeano Diaz J.P.: investigation, methodology, data curation and writing – original draft. Galeano Torres P.: resources, supervision, project administration and writing – review & editing. Garrido Fariña G.I.: conceptualization, supervision, methodology and writing – review & editing. Domínguez Vara I.A.: supervision, validation, project administration and writing – review & editing. Morales Almaraz E.: supervision, project administration and writing – review & editing.

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