Chilean J. Agric. Anim. Sci., (2023) 39(3): 351-358.
https://doi.org/10.29393/CHJAA39-31EFDS60031

ISSN 0719-3890 online

ENZYMES FROM *Pleurotus ostreatus* SPENT SUBSTRATE USED FOR FATTENING RABBITS: EFFECTS ON PRODUCTIVE PERFORMANCE, CARCASS TRAITS AND MEAT CHARACTERISTICS

Darinka Miroslava Saavedra-Castillo[1a], Maricela Ayala-Martínez*[1b], Javier Piloni-Martini[1c], Aurora Quintero-Lira[1d], and Sergio Soto-Simental[1e]

[1a] Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Ave. Universidad s/n km 1. Ex Hacienda de Aquetzalpa. CP. 43600. Tulancingo, Hidalgo, México
[1b] Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Ave. Universidad s/n km 1. Ex Hacienda de Aquetzalpa. CP. 43600. Tulancingo, Hidalgo, México
https://orcid.org/0000-0001-5554-218X
[1c] Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Ave. Universidad s/n km 1. Ex Hacienda de Aquetzalpa. CP. 43600. Tulancingo, Hidalgo, México
https://orcid.org/0000-0002-1367-5010
[1d] Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Ave. Universidad s/n km 1. Ex Hacienda de Aquetzalpa. CP. 43600. Tulancingo, Hidalgo, México
https://orcid.org/0000-0003-4638-6028
[1e] Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Ave. Universidad s/n km 1. Ex Hacienda de Aquetzalpa. CP. 43600. Tulancingo, Hidalgo, México
https://orcid.org/0000-0002-6923-0926
* Corresponding author: ayalam@uaeh.edu.mx

ABSTRACT

Rabbit meat has a high nutritional value. The cost of rabbit feed is high, and thus it is important to find alternative feed sources with reduced costs. The objective was to evaluate the effects of enzymes obtained from the spent substrate of *Pleurotus ostreatus* on carcass traits and meat characteristics of fattening rabbits in order to recommend an alternative additive for rabbit feed. Spent substrate of *P. ostreatus* with different levels of enzymatic activity was used to feed growing rabbits. Ninety-sixty rabbits were selected and randomly distributed to four treatments: 0 (control without enzyme extract, C); 20000 (T20); 40000 (T40); and 60000 (T60) IU kg^{-1} of laccase enzymes from the spent substrate of *P. ostreatus*. The experiment included six repetitions (four animals each) per treatment. Rabbits were fattened over 28 d and productive performance parameters were determined. Then, the animals were slaughtered, and carcass traits and meat characteristics were measured. The results indicated that total weight gain, total feed consumption, total daily weight gain, hot carcass weight, and dressing percentage decreased ($p<0.05$) as the amount of enzyme increased. However, the T40 treatment was similar to the control ($p>0.05$) for carcass traits, texture and pH, although L*, cooking loss and hardness recorded higher values ($p<0.05$). In conclusion, the spent substrate of *P. ostreatus* can be used to feed rabbits as some parameters of productive performance and carcass traits were similar to the control treatment.

Keywords: Edible fungi, laccases, meat characteristics, carcass trait.

Received: August 9, 2023 Accepted: December 4, 2023

INTRODUCTION

Rabbit production is an important animal activity, which in some countries is associated with small producers. However, feed cost represents the largest proportion of the production process and accounts for 0.6 to 0.7 of the total costs (Maertens, 2010). One strategy for reducing feed costs is the use of agro-industrial byproducts. For example, Oseni and Lukefahr (2014) studied rabbit production in a specific low input system, indicating that there are many feedstuffs or agro-industrial byproducts that can be used due their nutritional quality. One of these byproducts is the spent mushroom substrate of *P. ostreatus*.

According to Grimm and Wösten (2018), P. *ostreatus* production generates five kg spent substrate per kilogram of mushroom and is considered as an agro-industrial waste. The authors also indicate that spent mushroom substrates can be used to feed animals, to make biofuel, to produce materials, and to obtain enzymes. *P. ostreatus* produces cellulases, xylanases and laccases (Iandolo et al., 2011). These enzymes, which are produced while growing edible fungi, can improve the digestibility of lignocellulosic materials used for mushroom cultivation (mainly wheat straw, corncob, and barley straw) and also increase some components such as crude protein, with benefits for animal feeding (Leong et al., 2022).

There are some strategies for improving feed quality. One of them is using commercial exogenous enzymes as they increase the digestibility of fiber fractions, thereby improving the availability of nutrients to complement endogenous enzyme activity (Oijha et al., 2019). However, the high cost and accessibility of enzymes limits their use. An alternative for improving feed digestibility is the use of enzymes obtained from spent substrates of some fungi as indicated by Leong et al. (2022). Recently, some commercial enzymes have been used for feeding rabbits to increase productive performance and meat quality. Olorontula et al. (2018) fed rabbits with a commercial enzyme containing cellulase, phytase, protease, α-amylase, β-glucanase, and lipase. Abdullahi et al. (2020) fed rabbits with an exogenous enzyme and indicated that it degraded fiber. The objective of this work was to evaluate the effects of enzymes obtained from the spent substrate of *Pleurotus ostreatus* on carcass traits and meat characteristics of fattening rabbits for potential use as an alternative additive in rabbit feed.

MATERIALS AND METHODS

Enzymes from the spent substrate of *Pleurotus ostreatus*

To obtain the enzyme extract, 250 g of spent *P. ostreatus* substrate were blended with 500 mL of distilled water in an Osterizer blender model 4655-13 (Oster, Mexico City, Mexico). Eight layers of gauze were used to filter the mix, and then the supernatant was centrifuged at 10 000 rpm for 20 min at 21 °C. The supernatant obtained was used to measure soluble protein content and enzymatic activity. Levels of total soluble protein were determined using Bradford reagent (Brandford, 1976), while bovine serum albumin (BioChemika, USA) was used as standard in the citrate buffer (50 mM, pH 5.3). Absorbance was read at 595 nm. The enzymatic activity of xylanases, cellulases and laccases was measured according to Ayala et al. (2011). Briefly, the extract was mixed with xylose (100 µL + 900 µL, respectively) and incubated at 50 °C for 5 min. Subsequently, 1.5 mL of 3,5-dinitrosalicylic acid (DNS) was added and the mixture was boiled for 5 min. The samples were chilled on ice and absorbance was read at 540 nm. To determine cellulase activity, carboxymethylcellulose (CMC) plus enzyme extract (100 + 900 µL, respectively) were incubated for 60 min at 50 °C. After that, DNS (1.5 mL) was added, and tubes were heated for 5 min to measure the absorbance at 540 nm. Laccases were determined mixing 200 µL of enzymatic extract with 600 µL of citrate buffer (50 mM, pH 5.0). The tubes were incubated in a water bath (Cole-Parmer, USA) at 40 °C for one min, and then 200 µL of ABTS (2,2′ Azino-bis (3-ethylbenzthiazoline-6-sulfonic acid) in citrate buffer (50mM, pH 5.0) as substrate was added to measure absorbance at 420 nm. All the absorbance measurements were performed using a Jenway 6305 spectrophotometer (Dunmow, Essex, UK).

Experimental design, animals, and feed

The protocol number CICUA/ICAP 001/2020 indicates that this experiment was conducted according to guidelines on animal care of Universidad Autonoma del Estado de Hidalgo. A one-way design was used to analyze the four treatments: Control (C) without enzyme extract; and 20000 (T20), 40000 (T40), and 60000 (T60) UI kg^{-1} of laccase enzymes from the spent substrate of *P. ostreatus*. Ninety-six California x English pot rabbits (35 d of age) were selected, and randomly distributed to the treatments. The experiment included 6 repetitions (with 4 animals each) per treatment. Rabbits were fattened at an experimental rabbitry for 28 d. Environmental conditions in the hutch were 20

°C of temperature and 70% of relative humidity. Automatic drinkers and manual feeders were adapted to cages (45 x 60 x 40 cm in size) to allocate the rabbits. To feed the rabbits, isoproteic (15% CP), isoenergetic (2.6 MCal.kg^{-1}) and isofibrous (29% NDF and 17% ADF) diets (Table 1) were used to feed daily. Diets were designed to meet nutritional requirements recommended by the NRC (1977). A pellet machine model SKJ120 (Yuezhen Machinery Co., Shandong, China) was used to manufacture the feed, which was then stored at room temperature.

Productive performance

To determine productive performance, the animals were weighed on a weekly basis and the feed was measured on daily basis using a Mettria MTNUV-40 digital scale (Mettria, Mexico City, Mexico). The obtained data were used to calculate average daily weight gain (ADWG), total weight gain (TWG), feed consumption, total feed consumption and feed conversion ratio (FCR). After the fattening period, rabbits were slaughtered by concussion, without fasting, according to the national legislation (NOM-033-SAG/ZOO-2014). Before slaughter, length and lumbar circumference of the rabbits were measured (Garcia-Valencia et al., 2022).

Carcass and meat traits

Carcass and lumbar circumference were measured once rabbits were slaughtered (n=96). Afterwards, the animals were dismembered to obtain weights of different parts of the animal. Then carcasses were maintained in refrigeration at 6 °C for 24 h. Subsequently, the carcasses were sectioned as indicated by Blasco et al. (1993).

To measure meat color on loin surface (*Longissimus lumborum*) a Minolta CM-508d spectrophotocolorimeter (Minolta, Tokyo, Japan) was used. The instrument was setup to determine CIEL*a*b* color space, D65 illuminant and 10° standard observer (AMSA, 2012). To determine pH, a Hanna model HI99163 meat pH meter (Hanna Instruments, Cluj-Napoca, Romania) was used. Right loins (*Longissimus lumborum*) of the carcasses were used to determine cooking loss. The samples were put into a plastic bag and submerged in a water bath (StableTemp, Cole Parmer, USA), and then cooked until an internal temperature of 68 °C was reached. The samples were weighed once they were cold, and cooking loss values were calculated. After the loins were weighed, a texture profile analysis (TPA) was conducted according to Echegaray et al. (2022) with some modifications. Briefly, the cooked loins were divided into six cubes of 1 cm side, and then

Table 1. Diet formulation and chemical composition.

	Treatments[1]			
Ingredient	**C**	**T20**	**T40**	**T60**
Ground corn, g kg^{-1}	176	177	178	179
Ground sorghum, g kg^{-1}	175	176	177	178
Dry Distilled Grains, g kg^{-1}	37	37	37	37
Molasses (25 % MS), g kg^{-1}	70	70	71	71
Soybean oil, g kg^{-1}	25	25	26	26
Canola meal, g kg^{-1}	38	38	39	39
Soybean meal, g kg^{-1}	192	193	194	195
Barley Straw, g kg^{-1}	277	186	93	0
Vitamins and minerals premix, g kg^{-1}	9	9	9	9
P. ostreatus extract, g kg^{-1}	0	88	176	266
Nutritional composition calculated				
Protein, %	16.00	16.00	16.00	16.00
Digestible energy, Mcal MS kg^{-1}	2.5	2.5	2.5	2.5
[2] ADF, %	14.00	15.00	16.00	18.00
[3] NDF, %	29.50	29.90	30.20	30.60
Calcium, %	17.80	17.76	17.71	17.60
Phosphorus, %	0.80	0.80	0.80	0.80
Sodium, %	0.40	0.40	0.40	0.40

[1]C= control treatment without enzyme extract, T20 = 20000, T40 = 40000, and T60 = 60000 IU kg^{-1} of laccase enzymes from the spent substrate of *P. ostreatus*. [2] ADF: Acid detergent fiber [3] NDF=Neutral detergent fiber

placed perpendicular to the fibers on the base and compressed at 50% using P/25 aluminum probe with 1 mm s^{-1} of crosshead speed test. After the samples were compressed, a TA XTPlus texture analyzer (StableMicrosystems, Surrey, UK) was used. The texture parameters (hardness, resilience, cohesiveness and springiness) were calculated using Exponent software ver. 6.1.20.0 (StableMicrosystems, Surrey, UK).

Statistical analysis

A repeated time one-way design was used to analyze productive performance, while a completely random experimental design was used to evaluate carcass characteristics and meat traits. Then, all analyses were achieved using SPSS software (IBM, Chicago, IL, USA). The statistical model used to determine influence of treatment through time was:

$$Y_{ijk} = \mu + \beta_i + \tau_j + \beta_i(\tau_j) + \varepsilon_{ijk},$$

where, Y_{ij} = dependent variable (weight, feed consumption), μ = mean, β_i = fixed effect of *i*-th rabbit within the group, τ_j = time, $\beta_i(\tau_j)$ = the behavior of treatment trough time and ε_{ijk} = experimental error associated with each dependent variable Y_{ijk}.

$$Y_{ij} = \mu + \beta_i + \varepsilon_{ij},$$

in which Y_{ij} = dependent variable, μ = mean of the population, β_i = the fixed effect of *i*-th rabbit of the group, and ε_{ij} = experimental error associated with each dependent variable Y_{ij}.

RESULTS

An enzyme extract obtained from the spent susbtrate of *P. ostreatus* was used and its effects on productive performance, carcass and meat characteristics were determined. Variables measured to improve the productive performance are shown in Table 2. There were differences ($p<0.05$) among groups fed with the spent substrate of *P. ostreatus* with different enzymatic activity. Feed consumption, final weight, and total weight gain were lower ($p<0.05$) in the group T60 for all weeks during the fattening period. Daily weight gain was higher ($p<0.05$) in the control group in weeks 2 and 3, while the control also recorded the highest value in terms of total daily weight gain. Moreover, the feed conversion ratio was lower ($p<0.05$) in the control group.

The carcass trait of rabbits that consumed enzymes from the spent substrate of *P. ostreatus* is shown in Table 3. Skin, gastrointestinal tract, carcass length, lumbar circumference, live weight, hot carcass weight, cold carcass weight, dressing carcass, head, and bone showed differences among treatments ($p<0.05$). In general, these

Table 2. Feed consumption and daily weight gain in rabbit feed with an enzyme extract from the spent substrate of *Pleurotus ostreatus*.

	Treatments[1]				
Variable	**C**	**T20**	**T40**	**T60**	**SEM**
Feed consumption 1, g	624.25Aa	615.00Ba	569.50Ba	523.25Ba	22.38
Feed consumption 2, g	668.00Aa	442.75bCa	495.08Ba	372.75Bb	65.61
Feed consumption 3, g	656.75Aa	753.50ABa	549.91bBa	499.10Ba	127.03
Feed consumption 4, g	942.62Ba	883.67Aa	937.58Aa	840.49Aa	72.87
Initial weight, g	968.18a	1011.00a	909.68a	972.18a	37.89
Final weight, g	1960.90a	1750.00ab	1717.50ab	1602.18b	94.31
Total weight gain, g	992.72a	739.00b	807.81ab	630.00b	76.51
Total feed consumption, g	2997.15a	2677.74ab	2681.92ab	2328.52b	140.81
Daily weight gain 1, g d^{-1}	38.83Aa	36.47Aa	40.31Aa	32.14Aa	3.10
Daily weight gain 2, g d^{-1}	39.48Aa	12.90Cb	17.09Bb	8.30Bb	7.44
Daily weight gain 3, g d^{-1}	32.33Aa	22.38BCab	19.37Bb	18.25Bb	4.79
Daily weight gain 4, g d^{-1}	27.27Aa	29.24ABa	33.7Aa	27.38Aa	3.96
Total daily weight gain, g d^{-1}	35.45a	26.39b	28.84ab	22.50b	2.73
Feed conversion ratio	3.10b	4.27a	3.50ab	4.07ab	0.39

[ab] Different superscript letters indicate significant differences among columns. [AB] Different superscript letters indicate significant differences among files only in feed consumption or daily weight gain. Tukey test ($p<0.05$). [1]C= control treatment without enzyme extract, T20 = 20000, T40 = 40000, and T60 = 60000 IU kg^{-1} of laccase enzymes from the spent substrate of *P. ostreatus*.

Table 3. Carcass traits in rabbits fed with an enzyme extract of the spent substrate of *Pleurotus ostreatus*.

Variable	Treatments[1]				SEM
	C	T20	T40	T60	
Skin, g kg^{-1} LW[2]	24.9a	23.9a	24.1a	22.7b	0.3
Feet, g kg^{-1} LW	24.7a	24.0a	25.1a	25.0a	0.7
Gastrointestinal tract, g kg^{-1} LW	225.6b	261.6b	246.2b	297.8a	10.9
Animal length, cm	32.2a	29.2a	30.0a	29.5a	1.1
Carcass length, cm	21.7a	20.5ab	20.3ab	19.1b	0.8
Lumbar circumference, cm	30.7a	29.3ab	29.2ab	28.6b	0.8
Lumbar carcass circumference, cm	15.8a	14.4ab	14.9ab	13.7b	0.6
Live weight, g	2017.5a	1800.7ab	1716.9ab	1600.0b	93.8
Hot carcass weight, g	1095.6a	940.7ab	911.4ab	784.8b	59.9
Cold carcass weight, g	1072.1a	917.0ab	889.1ab	782.6b	58.7
Dressing carcass, %	54.4a	51.8ab	52.7ab	48.5b	1.1
Kidney fat, g kg^{-1} HCW[2]	11.8a	11.1a	10.0a	9.0a	1.3
Scapular fat, g kg^{-1} HCW	4.1a	2.6a	4.1a	2.7a	0.5
Head, g kg^{-1} HCW	94.6b	112.2ab	111.2ab	119.3a	5.4
Forepart weight, g kg^{-1} HCW	243.6a	233.2a	235.3a	244.3a	5.2
Intermedia part weight, g kg^{-1} HCW	99.6a	105.8a	105.8a	106.6a	4.3
Hind part weight, g kg^{-1} HCW	188.1a	172.3a	177.9a	175.5a	5.7
Legs, g kg^{-1} HCW	336.7a	334.1a	331.6a	343.5a	7.4
Meat, g kg^{-1} Legs	698.9a	674.3a	689.1a	672.5a	12.9
Bone, g kg^{-1} Legs	276.3b	301.3a	287.2a	304.2a	13.0
Dissectible fat, g kg^{-1} Legs	10.3a	11.8a	8.5a	7.9a	2.1

[ab] Different superscript letters indicate significant differences among columns. Tukey test ($p<0.05$). [1]C= control treatment without enzyme extract, T20 = 20000, T40 = 40000, and T60 = 60000 IU.kg^{-1} of laccase enzymes from the spent substrate of *P. ostreatus*. [2]Hot carcass weight.

variables were higher in the control group, except for the gastrointestinal tract, head, and bone since they were similar among treatments. Carcass traits and meat characteristics were also similar ($p>0.05$) between the control and T20 groups. However, bone content was higher ($p<0.05$) in the T20 group compared to the control.

Meat characteristics are shown in Table 4. L* value increased ($p<0.05$) in response to the different levels of enzymes used. However, yellowness (b* value) and chroma decreased ($p<0.05$) when enzyme levels were high, and the control group recorded the highest values. Moreover, cooking loss increased ($p<0.05$) as the level of enzymes increased. All TPA parameters were lower ($p<0.05$) in the control group, but the parameters were similar among the groups fed with different levels of enzymes.

DISCUSSION

Daily feed consumption and daily weight gain were different among the treatments. Higher daily feed consumption was observed in the control group, thereby contributing to better daily weight gain, which can be confirmed with a high total weight gain and total feed consumption. It was only during the last fattening week that all groups showed similar behavior in some productive performance parameters. These relationships indicate a lower feed conversion rate in the control group. Piles and Sanchez (2019) reported that rabbits with restricted feeding had limited access to food, meaning that their feed efficiency was lower than ad libitum fed rabbits. Consequently, there is low growth due to the amount of feed intake, which can be used as an indicator of feed efficiency. In the present study, the productive performance parameters were low in animals fed with enzymes, except in the T20 treatment. This effect is associated with a low feed conversion rate in groups fed with high quantities of the spent substrate of *P. ostreatus*. According to Vitolo (2021), an enzyme only needs a minimum amount to catalyze any amount of substrate. The author also indicated that initial activity has a relationship with the amount of substrate. These low productive performance

Table 4. Characteristics of rabbit meat obtained from rabbits fed with an enzyme extract of the spent substrate of *Pleurotus ostreatus*.

Variable	Treatments[1]				SEM
	C	T20	T40	T60	
L*	42.51c	44.97b	46.50ab	47.79a	0.59
a*	-0.38a	-0.48a	-0.42a	-0.23a	0.16
b*	7.76a	7.62b	6.45c	6.27d	0.23
c	7.86a	7.82a	6.65b	6.38b	0.22
h	1.54a	1.49a	1.49a	1.52a	0.02
pH	5.89a	5.88a	5.92a	5.96a	0.06
Cooking loss, %	17.03b	25.98a	24.78a	25.54a	1.81
Hardness, N	1.17b	2.22a	2.51a	2.62a	0.24
Resilience	0.18b	0.23a	0.21a	0.19b	0.01
Cohesiveness	0.42b	0.51a	0.52a	0.55a	0.02
Springiness	0.47c	0.58b	0.64a	0.55b	0.09

[abc] Different superscript letters indicate significant differences among columns. Tukey test ($p<0.05$). [1]C= control treatment without enzyme extract, T20 = 20000, T40 = 40000, and T60 = 60000 IU.kg^{-1} of laccase enzymes from the spent substrate of *P. ostreatus*.

parameters in rabbit groups fed with *P. ostreatus* spent substrate are related to the fact that enzymes do not catalyze fiber components properly. In addition, during the pelletizing process, temperatures were high and enzymes were inactive, indicating that temperature is important for enzyme stability. However, the use of multi-enzyme supplementation in rabbits increased the feed conversion rate due to the increase in the digestibility of protein and fiber (Olorontula et al., 2018).

In general, carcass traits and composition were lower in groups fed with enzymes obtained from *P. ostreatus* spent substrate, except for the gastrointestinal tract, head and bone weight. In a study by Salama et al. (2019), no differences were found in carcass traits among rabbits fed with fennel along with a commercial enzyme mixture. According to Mohammed *et al.* (2018), no differences were found using broiler feed with exogenous enzymes when enzyme levels were increased. This was attributed to the effect caused by chemical differences among the ingredients used to feed animals as well as enzymatic activity. As previously mentioned, if the enzymes did not catalyze fiber components nor enhance the digestibility of nutrients, there was no improvement in carcass traits. In our study, enzymatic activity probably decreased after the pelletization process due to the temperature during the processing of the feed. It could also be the case that enzymes did not have much contact with feed fiber to catalyze this component and increase carbohydrate availability.

In terms of meat characteristics, the control group presented the highest color values, including L*, a* b* and c parameters, compared to the enzyme treatments. It should be noted that the color is the most important meat trait because consumers usually use this attribute when purchasing meat (Tomasevic et al., 2021). In contrast, Hernández-Martinez *et al.* (2018) used sorghum, which was hydrolyzed by *Trametes maxima* CU1 and *Pycnoporus sanguineus* CS2, and did not find any differences in color parameters among treatments. Similar results were found by Kong et al. (2022) with Rex rabbits fed an enzymolytic soybean meal. However, there are several pre- and post-harvest factors influencing meat color, e.g., the use of natural additives (Cardinali et al., 2015). Meat pH is an important parameter for determining shelf life and growth of microorganisms in meat (Dalle-Zotte, 2002). In this study, the pH of rabbit meat from animals fed with several levels of enzymes showed no significant differences. However, cooking loss and texture profile analysis parameters were lower in the control treatment. In another study, Hernández-Martinez et al. (2018) found low hardness values in meat from rabbits fed with hydrolyzed sorghum.

CONCLUSION

According to the results, meat obtained from rabbits fed with different levels of laccase enzymes deteriorate rabbit meat characteristics. However, the spent substrate of *P. ostreatus* can be used to

feed rabbits as some productive performance parameters and carcass traits were similar to the control treatment. It is important to continue evaluating other alternative for producing rabbit feed, including maintaining some contact time between fiber and enzymes before pelletizing the feed to prevent enzyme deterioration and improve carbohydrate availability.

Ethical statement

The authors confirm that this research protocol was summited to the ethical committee of Universidad Autónoma del Estado de Hidalgo. Number CICUA/ICAP 001/2020 indicates this experiment was conducted according to guidelines on animal care.

ACKNOWLEDGEMENTS

Saavedra-Castillo, D.M. received a grant from Consejo Nacional de Ciencia y Tecnología from the Mexican government. The authors would also like to thank Christopher Shackley for reviewing the English language.

LITERATURE CITED

Abdullahi, H.R., S.E. Alu, and A. Yakubu. 2020. Growth parameters and carcass characteristics of grower rabbits fed tiger nut offal meal-based diet supplemented with kingzyme® enzyme. Journal of Applied Agricultural Research 8(1):136-145. https://jaarbox.com/storage/app/public/files/1/Animal%20Production_1607413561.pdf

AMSA, 2012. Meat color measurement guidelines. American Meat Science Association Champaign. Il. USA. https://meatscience.org/publications-resources/printed-publications/amsa-meat-color-measurement-guidelines

Ayala, M., M.S.S. González, R.J.M. Pinos, C. Vázquez, M. Meneses, O. Loera, and G.D. Mendoza. 2011. Fibrolytic potential of spent compost of the mushroom *Agaricus bisporus* to degrade forages for ruminants. African Journal of Microbiology Research 5(3):241-249. https://doi.org/AJMR10.528.

Blasco, A., J. Ouhayoun, and G. Masoero. 1993. Harmonization of criteria and terminology in rabbit meat research. World Rabbit Science 1:3-10. https://doi.org/10.4995/wrs.1996.278

Bourne, M.C. 1978. Texture profile analysis. Food Technology 35: 62-66.

Cardinali, R., M. Cullere, A. Dal Bosco, C. Mugnai, S. Ruggeri, S. Mattioli, C. Castellini, M.M. Trabalza, and A. Dalle Zotte. 2015. Oregano, rosemary and vitamin E dietary supplementation in growing rabbits: Effect on growth performance, carcass traits, bone development and meat chemical composition. Livestock Science 175:83–89. https://doi.org/10.1016/j.livsci.2015.02.010

Dalle Zotte A. 2002. Perception of rabbit meat quality and major factors influencing the rabbit carcass and meat quality. Livest Production Science 75(1):11-32. https://doi.org/10.1016/S0301-6226(01)00308-6

Echegaray, N., M. Rosmini, M. Pateiro, R. Dominguez, P.E.S. Munekata, J.M. Lorenzo, E.M. Santos, and R. Bermudez. 2022. In: Lorenzo, J. M., Domínguez, R., Pateiro, M., Munekata, P. E. (Editors). Methods to Assess the Quality of Meat Products. Springer. New York, NY, USA.

FEDNA, 2020. Tablas FEDNA de composición y valor nutritivo de los alimentos para la fabricación de piensos compuestos. Fundación Española para el Desarrollo de la Nutrición Animal. Accesed July 2020. http://www.fundacionfedna.org/

García-Valencia, S., S. Soto-Simental, J. Ocampo-López, and M. Ayala-Martínez. 2022. Diet supplementation with *Dalbergia palo-escrito* hexane extract in fattening rabbits: its effect on productive performance, carcass traits, meat characteristics and meatballs Shelf-Life. Italian Journal of Animal Science 21(1): 1533-1542.

Grimm, D., and Wösten H.A. 2018. Mushroom cultivation in the circular economy. Applied Microbiology and Biotechnology 102(18):7795-7803. https://doi.org/10.1007/s00253-018-9226-8

Hernández-Martínez, C.A., G.F. Treviño-Cabrera, C.E. Hernández-Luna, R.H. Silva-Vázquez, E. Michael, G. Gutiérrez-Soto, and G. Méndez-Zamora. 2018. The effects of hydrolysed sorghum on growth performance and meat quality of rabbits. World Rabbit Science 26 (2):155-163. http://doi.org/10.4995/wrs.2018.7822

Hernández-Martínez, C.A., J.A. Maldonado Herrera, G. Méndez-Zamora, C.E. Hernández-Luna, and G. Gutierréz-Soto. 2017. Enzymatic extract of *Trametes maxima* CU1 on productive parameters and carcass yield of rabbits. Canadian Journal of Animal Science 97(4): 683-688. https://doi.org/10.1016/j.cofs.2021.02.012

Iandolo, D., A. Piscitelli, G. Sannia, and V. Faraco. 2011. Enzyme production by solid substrate fermentation of *Pleurotus ostreatus* and *Trametes versicolor* on tomato pomace. Applied Biochemistry and Biotechnology 163:40–51. https://doi.org/10.1007/s12010-010-9014-0

Kong, F., T. Liu, Y. Liu, Z. Yu, W. Zhang, D. Fan, J. Fan, L. Kong, B. Li, S. Chen, and B. Chen. 2022. Dietary effects of enzymolytic soybean meal inclusion on antioxidant capacity, intestinal morphology and caecal microbiota of Rex rabbits. Italian Journal of Animal Science 21(1):1220-1231. https://doi.org/10.1080/1828051X.2022.2104177

Leong, Y.K., T.W. Ma, J.S. Chang, and F.C. Yang FC. 2022. Recent advances and future directions on the valorization of spent mushroom substrate (SMS): A review. Bioresource Technology 344:126157. https://doi.org/10.1016/j.biortech.2021.126157

Maertens, L. 2010. Feeding systems for intensive production. In: Nutrition of the rabbits. 2nd edition. Eds. C. de Blas, J. Wiseman. CAB international. Oxfordshire, UK.

Mohammed, A.A., A.B. Habib, A.M. Eltrefi, E.S.A. Shulukh, and A.A. Abubaker. 2018. Effect of different levels of multi-enzymes (Natuzyme Plus®) on growth performance, carcass traits and meat quality of broiler chicken. Asian Journal of Animal Veterinary Advances 13 (1): 61 – 66. http://doi.org/10.3923/ajava.2018.

NOM-033-SAG/ZOO-2014. Mexican Official Standard. Methods to give death to domestic and wild animals. (in Spanish). [On line] Available: http://www.economianoms.gob.mx/normas/noms/2010/033sagzoo2015.pdf.

NRC. 1977. Nutrient Requirements of Rabbits. National Academies Press. https://doi.org/10.17226/35

Ojha, B.K., P.K. Singh, and N. Shrivastava. 2019. Enzymes in the animal feed industry. In: Enzymes in food biotechnology p. 93-109. Academic Press. San Diego, CA, USA. https://doi.org/10.1016/B978-0-12-813280-7.00007-4

Oloruntola, O.D., S.O. Ayodele, S.A. Adeyeye, M.H. Ogunsipe, O.T. Daramola, and E.S. Ayedun. 2018. Effect of pawpaw leaf meal and multi-enzyme supplementation in the diet on performance, digestibility, and oxidative enzyme status of rabbits. JoBAZ 79: 26. https://doi.org/10.1186/s41936-018-0039-1

Oseni, S.O., and S.D. Lukefahr. 2014. Rabbit production in low-input systems in Africa: situation, knowledge and perspectives – A review. World Rabbit Science 22:147–160. https://doi.org/10.4995/wrs.2014.1348

Piles, M., and J.P. Sánchez. 2019. Use of group records of feed intake to select for feed efficiency in rabbit. Journal of Animal Breed and Genetics 136(6):474-483. https://doi.org/10.1111/jbg.12395

Salama, W.A., A. Refaie, and M. El-Shora. 2019. Performance of growing rabbits fed diets containing fennel seed meal without or with enzyme mixture. Egyptian Journal of Rabbit Science 29(1): 45-60. https://ejrs.journals.ekb.eg/article_45673.html

Vitolo, M. 2021. Enzymes, the catalytic proteins. In: Pharmaceutical biotechnology, A. Pessoa Jr, M. Vitolo, P.F. Long (eds). CRC Press. Boca Ratón, FL, USA.