

EFFECT OF AVOCADO (*Persea americana* MILL VAR. HASS) RESIDUES SUPPLEMENTATION ON RABBIT HEALTH PROFILE, AND MEAT QUALITY

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ABSTRACT

The aim of this study was to evaluate changes in body weight and serum metabolic parameters in meat-producing New Zealand rabbits fed a diet supplemented with 2% Hass avocado peels (P) and seeds (S). The evaluated variables included meat color, pH, moisture content, water-holding capacity, fatty acid profile, malondialdehyde levels as an oxidation parameter, measured by the TBARS (thiobarbituric acid reactive substances) test. It was found that serum concentrations of glucose, total cholesterol, triglycerides and transaminases remained unchanged throughout the experiment across treatments P and S, as well as in comparison to the control treatment (C) ($p < 0.05$). Supplementation with P and S did not affect the normal growth curve of the rabbits, as it was similar to that of C. There were no differences in final weight among the three treatments, nor in carcass quality. At the end of the experimental period, the meat characteristics of rabbits supplemented with P and S were similar ($p > 0.05$) to those of the C treatment for all the evaluated response variables. Thus, the addition of 2% avocado peels and seeds to the regular diet of rabbits, had not adverse effects on their normal growth and metabolism, nor did it significantly influence meat quality characteristics or oxidative stability.

Keywords: *avocado waste, New Zealand rabbits, oxidation in meat*

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INTRODUCTION

Food security is of paramount importance to ensuring a sufficient supply of nutritious food that meets the population's needs (FAO 2009; FAOSTAT 2010). Cereals, fruits, vegetables and meat are essential for fulfilling nutritional requirements. Among the meats consumed, rabbit meat (*Oryctolagus cuniculus* L.) contributes to a healthy diet due to its excellent nutritional properties, which benefit human health (Dalle Zotte *et al.*, 2011; Dalle Zotte *et al.*, 2014). It is highly digestible, rich in high-quality protein, and contains significant amounts of minerals and vitamins. Additionally, it has a low caloric content and is an important source of polyunsaturated fatty acids, which makes it susceptible to rapid oxidation. This process generates short-chain aldehydes, such as

malondialdehyde, which, along with the acids formed during oxidation, contribute to the rapid deterioration of the meat. As a result, its nutritional and sensory qualities decline, ultimately reducing its shelf life and economic value (Kakuda *et al.*, 1981; Cassens 2004; Kumar *et al.*, 2015).

To prevent the deterioration of rabbit meat, incorporating antioxidants into the rabbit's diet is an excellent strategy to stabilize oxidation reactions, extend shelf life, and improve its quality (Dalle Zotte *et al.*, 2016; Hernández-López *et al.*, 2016; Mireles-Arriaga *et al.*, 2018).

This can be achieved by using synthetic antioxidants, such as butylated hydroxytoluene (BHT), which is commonly used in animal feed (Brewer, 2011; Kumar *et al.*, 2015). Alternatively, natural antioxidants that do not pose health risks to animals or consumers can

be used. For meat preservation, essential oils from natural sources such as onion, blueberries, strawberries, rue, oregano, rosemary, and alfalfa are commonly utilized (Vekiari 1993; Hernández-Hernández *et al.*, 2009; Dal Bosco *et al.*, 2014; Cardinali *et al.*, 2015; Kone *et al.*, 2016; Ayala *et al.*, 2020). Another alternative is the use of agroindustrial byproducts that contain phenolic compounds and tocopherols, both which have antioxidants effects. Some examples include tomato peels (Dal Bosco *et al.*, 2012) and avocado sedes and peels, which have also demonstrated physiological benefits related to metabolic health, such as reducing glucose and total cholesterol levels (Espinosa-Garza *et al.*, 2019).

A large quantity of Hass avocado (*Persea Americana* Mill.) is consumed and its processed worldwide, generating significant amounts of organic waste, such as peels and seeds, which account 30% of the fruit's fresh weight. Every year, 1.6 million tons of these by-products are discarded as inedible fractions, leading to the loss of potential nutrients and bioactive compounds (Federici *et al.*, 2009; Rosero *et al.*, 2019; Jimenez *et al.*, 2021; Salazar-López *et al.*, 2020). Considering this, the present study aimed to evaluate serum metabolic parameters, oxidative stability, and the fatty acid profile of rabbit meat supplemented with avocado peels and seeds.

MATERIALS AND METHODS

Vegetal material: Hass avocados at physiological maturity were obtained in Uruapan, Michoacan. Peels (P) and seeds (S) were separated from the avocado and underwent a disinfection process via immersion in hipoclorite solution (10%) for 10 min. P and S were weighed and dried at 50°C for 36 h in a forced circulation oven (Shel-Lab FX-14, USA). Subsequently, P and S dried and disinfected were subjected to ground a 2 mm sieve (Thomas-Wiley Mill, Model 4, Thomas Scientific™, USA). Finally, P and S were stored in light-protected and hermetically sealed containers until further use.

Preparation of extracts for quantification of antioxidant activity: 100 mg of peels and pulverized seeds were individually weighed, and 3 mL of 80:20 v/v ethanol: distilled water solution was added. The extract was shaken by sonication for 1 h (Branson 2510, USA) and subsequently centrifuged at 4500 rpm at 10°C for 30 min (Hermle Z323K, Wehingen, Germany). The supernatant was stored at -20°C and light-protected for subsequent analyzes of total phenols (Singleton and Rossi 1965), FRAP (Ferric Reducing Antioxidant Power) (Benzie and Strain 1996), DPPH (2,2-diphenyl-1-picrylhydrazyl-hydrate) (Brand-Williams *et al.*, 1995) and ABTS (2,2'-azino-bis 3-ethylbenzothiazoline-6-sulfonic

acid) (Re *et al.*, 1999). All analyses were adapted to microplates.

Animal model: Fifteen New Zealand white rabbits, 35 days old and weighing 904±105 g were used. Animals were individually housed in stainless steel cages with automatic feeders (Extrona® Barcelona, Spain) in the experimental unit for small species at the Desert Areas Research Institute (IIZD). All experimental procedures were conducted following the animal care guidelines (NOM-051-ZOO-1995, NOM 062-ZOO-1999) and approved by the Technical Committee of the IIZD of the Universidad Autónoma de San Luis Potosí (N° 00126).

The animals were subjected to a five-day adaptation period with standard rabbit feed (Ganador® Malta Cleyton, CDMX, Mexico) and water *ad libitum*. Subsequently, rabbits were randomly assigned to three treatments (n=5): control treatment (C) 100% standard diet, 98% standard diet added with 2% avocado seeds (S) and 98% standard diet added with 2% avocado peels (P). Diets had similar nutritional content: 16% protein, 3% fat, 12% moisture, 17% fiber, 10% ash and 42% nitrogen-free extract. Daily net feed intake was recorded, and changes in body weight were monitored weekly throughout the study.

At the beginning and middle of the experimental period, blood samples (3 mL) were obtained by puncturing the marginal vein of each rabbit, centrifuged (Solbat Model J-40, Puebla, Mexico) at 3500 rpm for 10 min to separate the serum, and then stored at -70 °C for subsequent biochemical analysis of glucose, total cholesterol, triglycerides and transaminases ALT and AST using commercial enzymatic kits (Spinreact®, Girona, Spain).

At the end of the 5th week, rabbits were fasted for 12 hours and weighed. Slaughter was carried out according to established ethical protocols (NOM-194-SSA1-2004, NOM-033-SAG/ZOO-2014). Blood samples were obtained for serum collection, and the skin, viscera, liver, adipose tissue, and the entire carcass were dissected and weighed. Carcass was rinsed with cold running water and stored at 4°C. After maturing for 24 h, cold carcass was weighed, vacuum packed (Oster® Food Saver® Model FM 3941, USA) and frozen at -20°C, for subsequent analyses.

Quantification of color, pH and moisture: Color, pH and moisture analyses were performed 24 h *post mortem*. Color was measured on the longitudinal and flat surface of the *Longissimus dorsi* muscle at three equidistant points of the tissue (FRU®, Model WR-10QC, China). Measured color coordinates were L^* , a^* , b^* for lightness, redness, and yellowness, respectively (CIE, 1976).

For pH, 3 g of meat was liquefied with 20 mL of deionized water and its pH was measured (HANNA Edge® HI 2202, USA) (Peña-Avelino *et al.*, 2017).

For moisture analysis, 5 g of homogeneous, ground meat was placed in a previously weighed and dried capsule. Subsequently, sample were dried at 60 °C for 12 h in a forced circulation oven (Shel-Lab FX-14, USA) and weighed again (NOM-116-SSA1-1994).

Water retention capacity: Water retention capacity (WRC) was carried out according to Grau and Hamm (1953). 300 mg of meat were weighed and placed between filter paper (Whatman™ Grade 541, 125 mm, 22 µm, China). Next, the sample was placed between two glass plates (30 x 30 cm), and 10 kg were applied for 15 min. Subsequently, the weight was removed, the sample was discarded and the filter paper was weighed before and after drying at 60°C for 24 h (Shel-Lab FX-14, USA). Percentage of water released was calculated as the percentage ratio of the weight of the wet filter paper-weight of the dry filter paper/initial weight of the sample per 100.

Extraction of lipids from meat: Cold extraction of meat lipids was carried in accordance with Folch *et al.* (1957) using some adaptations. 10 g of meat was weighed and 80 mL of Folch's reagent (chloroform: methanol 2:1, v/v) was added, homogenized at 9500 rpm for 2 min (OMNI TH-01, LR60902, Georgia, USA) and centrifuged at 3000 rpm for 10 min. Subsequently, the supernatant was filtered under vacuum. Residue in the tube was washed twice consecutively with 60 mL of Folch's reagent, centrifuged and filtered again. The three supernatants were mixed and 40 mL of a 0.73% NaCl aqueous solution was added, shaken vigorously for 5 min, and centrifuged at 3000 rpm for 10 min. Organic phase was subsequently recovered by rotary evaporation and dried with nitrogen. Extracted lipid were resuspended in hexane and stored at -70°C for chromatographic analysis.

Fatty acid profile: A total of 5 µL of the lipid extract was filtered through anhydrous sodium sulfate for injection (1 µL) in the gas chromatography system (Thermo Scientific™ Trace 1300, USA) coupled to a mass spectrometry detector, equipped with an HP-INNOWax 19091N column (30m x 0.25 mm x 0.50 µm) Injector and detector temperatures were settled at 260 °C and 280 °C, respectively. Initial oven temperature was 150 °C and kept for 15 min, then increased 5 °C/min up to 150 °C, 10 min at 150 °C, and finally ramped at 5 °C/min up to 250 °C which was maintained for 17 min. Helium was used as carrier gas at a flow rate of 1.0 mL/min. Free fatty acids were identified by comparison with the retention times of individual standards and by comparison of their mass spectra with the NIST library. Quantitative data were obtained by calibration curves carried out with commercial standards (Sigma Aldrich, USA).

Oxidative stability of meat: Meat oxidation was carried out according to Botsoglou *et al.* (1994) with

modifications: 1 g ground meat, 1 mL of BHT at 1% in ethanol and 4 mL of 10% aqueous trichloroacetic acid (Meyer®, CDMX, Mexico) were added. This mixture was homogenized vigorously for 5 min and centrifuged at 3500 rpm for 15 min. Afterwards, 2 mL of the supernatant was used and 500 µL of 2-thiobarbituric acid (Sigma Aldrich, USA) [0.055 M] in 10% aqueous trichloroacetic acid was added. Samples were placed in a water bath at 90°C for 45 min (Oakton® Stable Temp, Model 12500-30, USA). Finally, samples were read at 532 nm (Agilent 8453, USA), water was used as a blank. The calibration curve was performed with 1,1,3,3-tetraethoxypropane (Sigma Aldrich, USA) [0.002 M] as a standard solution.

Statistical analysis: The data obtained from the evaluated variables were tested for normality using the Shapiro-Wilk test ($p > 0.43$), confirming that the data followed a normal distribution. This allowed for an analysis of variance to be performed under a completely randomized design with three treatments and five replications each. The analysis was conducted using the PROC GLM procedure in SAS software version 9.2 (SAS Institute, Inc., Cary, North Carolina, USA). When ANOVA detected differences between treatments, a Tukey multiple comparison test was applied, with a significance level set at $p < 0.05$.

RESULTS AND DISCUSSION

By-product yield: From the 135 processed whole fruits, the average fresh weight was 210.32 ± 31.16 , with seeds weighing 33.01 ± 9.60 and peels 24.67 ± 5.25 g. After drying, 859.13 g of peels and 2072.07 g of seeds were obtained on a by dry weight (DW) basis. The relative proportion of seeds and peels, expressed as a percentage of the whole fruit on a dry basis, was $47.07 \pm 5.16\%$ and $28.32 \pm 2.92\%$, respectively. These values align with findings reported by Rodríguez-Carpena *et al.*, (2011a) and Calderón-Oliver *et al.*, (2016).

Antioxidant capacity of byproducts: Table 1 shows the results of the analysis of total phenolics and antioxidant activity of the peel and seed extracts.

A higher content of total phenols and antioxidant activity was found in the peel compared to the seed of Hass avocados, which is expected since the peel is the physiological part of the fruit that covers the easily oxidizable pulp. The content was higher than that of other commonly consumed fruits, such as tomatoes, onions, beets and carrots, which contain an average of 4.1 mg EAG/g DW (Kähkönen *et al.*, 1999). The phenolic content in avocado residues found in this study was higher than 5.7 mg EAG/g DW in seeds and 19.7 mg EAG/g DW in peels reported by Calderón-Oliver *et al.*, (2016), but lower than the 35.11 and 78.41 mg EAG/g DW reported by Rodríguez- Carpena *et al.*, (2011) for the

seed and peel respectively, and the exceptionally high amounts of 328.8 and 527.8 mg EAG/g DW in seed and

peel reported by Rosero *et al.*, (2019).

Table 1. Quantification of total phenols and antioxidant activity analysis of peel and seed extracts.

By-product	Total phenols	FRAP	DPPH	ABTS
	(mg GAE/g DW)		(mg Trolox Eq /g DW)	
Peel	28.5 ±1.89	89.5±6.19	504.2±10.23	96.5±4.24
Seed	18.9 ±1.99	49.1±3.28	367.1± 18.59	67.3±6.64

FRAP: Ferric Reducing Antioxidant Power, **DPPH:** 2, 2-diphenyl-1-picryl-hydrazyl-hydrate, **ABTS:** 2,2'-azino-bis 3-ethylbenzothiazoline-6-sulfonic acid, **GAE:** Gallic Acid Equivalents. **DW:** dry weight. Means ± SD (n=5).

Although phenolic compounds are found in various digestible parts of fruits and vegetables (Soong and Barlow 2004), some parts that are indigestible for human metabolism and are discarded as waste. However, after processing, these by-products could have promising applications in the livestock, cosmetic, and food industries due to their high polyphenol content. The high concentration of phenolic compounds in avocado residues, specifically phenolic acids and flavonol derivatives, has been associated with a reduction in lipid and protein oxidation in processed pork meat products (Rodríguez Carpena *et al.*, 2011a, b).

Body weight change: No significant differences ($p > 0.05$) were observed in body weight change or total weight gain of the rabbits throughout the experimental period between treatments (Table 2). At the end of the

experiment, the rabbits' weight was higher than that reported by Al-Sagheer *et al.* (2017) and Alagawany *et al.* (2018) in New Zealand rabbits supplemented with olive and coconut oil, which showed weight gains of 1990 g and 2011 g, respectively, after 13 weeks of experimentation.

Net feed consumption: Feed consumption was similar across the three treatments ($p > 0.05$) (Table 2). The inclusion of peels and seeds in the respective treatments did not restrict intake throughout the experiment, which is consistent with the findings De Blas *et al.*, (1981) regarding the typical feed consumption of rabbits.

No significant differences were found between treatments regarding average body components weight after slaughter (Table 3). The cold carcass yields relative to pre-slaughter weight were 53.85%, 54.91%, and 55.19% for C, P, and S, respectively, which is consistent with the findings of Tufarelli *et al.* (2010), who reported yields of 54.5–56.8%.

Table 2. Net daily feed intake and body weight change (g).

Treatment	Basal feed intake	Final feed intake	Basal weight	Final weight	Weight gain
C	73.1	97.4	935.0	2017.0	1186.0
P	77.0	104.0	910.0	2096.0	1082.0
S	74.9	99.0	868.0	2079.0	1211.0
SEM	4.2	7.2	69.5	112.2	64.9
<i>p value</i>	NS	NS	NS	NS	NS

C: Control, P: Avocado peel (2%), S: Avocado seeds (2%), SEM: Standard error of the mean. Means (n=5).

Quantification of moisture, pH, color and WRC: The pH of the meat was similar across all treatments ($p > 0.05$) (Table 4), and was higher than the values reported by Bianospino *et al.* (2006) (5.58-5.61) and Piles *et al.* (2000) (5.7), but similar to those evidenced by Lambertini *et al.* (1996) (6.05-6.2), Dal Bosco *et al.* (1997) (6.2-6.6) and Metzger *et al.* (2003) (6.41-6.45). It has been reported that in meats with higher pH levels, the speed of the reduction reaction from oxymyoglobin to myoglobin, which has a dark red color, is promoted. Therefore, at lower pH, the meat color (L^*) becomes lighter, as muscle contraction increases light scattering (Ouhayoun and Dalle Zotte 1993; Dal Bosco *et al.*, 2002; Bízková and Tůmová 2010).

The pH in meat is influenced by inadequate handling before and after slaughter. In rabbit meat, the pH is slightly higher than in other meats, such as beef and pork (5.5-5.7). Consequently, susceptibility to microbial growth is greater, which increases the risk of foodborne infections (Blasco and Piles 1990; Faucitano *et al.*, 2010).

Meat color was not influenced by the level of avocado peels and seed supplementation ($p > 0.05$). However, brighter meats (L^*) were obtained for higher pH values with treatments S and P compared to C, which is consistent with the findings of Kone *et al.* (2016), who reported a similar trend. Regarding red (a^*) and yellow (b^*) tones, the evidence is inconsistent: Dal Bosco *et al.* (2002) and Dalle Zotte *et al.* (2009) found significant

decreases, whereas Combes *et al.* (2010) reported an increase in a^* . Additionally, Dal Bosco *et al.* (2002) detected a significant increase in b^* , while Dalle Zotte *et al.* (2009) and Combes *et al.* (2010) observed a decrease. These variations may be attributed to diet, as well as the type of housing, materials, and measurements methods, which can influence growth, movement, and ultimately, meat quality and color.

The analysis of water retention capacity (WRC) showed that 20.6% (S), 20.9% (C), and 22.3% (P) of the total water in the meat was retained, with statistical similarity between treatments. In this study, a lower WRC was observed compared to the values reported by Ramírez *et al.* (2004) at 33.29% and Ariño *et al.* (2006) at 30.8%, but higher than that reported by María *et al.* (2006), who found values ranging from 12.12% to 14.93%.

Table 3. Average body components weight of New Zealand rabbits at slaughter.

Variable	Treatment		
	C	P	S
Live weight (g)	2033.3±7.6	2148.3±83.7	2070.0±181.5
Hot carcass (g)	1161.7±28.4	1243.3±82.8	1203.3±103.9
Cold carcass (g)	1095.0±34.6	1180.0±72.6	1143.3±115.9
Skin (%)	31.0±1.9	29.7±2.2	29.0±2.2
Liver (%)	6.85±0.7	7.71±2.9	6.90±1.6
Heart and lungs (%)	1.98±0.2	1.69±0.4	1.60±0.1
Gastrointestinal viscera (%)	37.4±5.8	34.86±4.1	33.09±2.6
Fat and kidneys (%)	4.7±0.8	5.1±0.5	4.4±0.7

C: Control, P: Avocado peel (2%), S: Avocado seeds (2%). Means ± SD (n=5).

Table 4. Quality parameters of New Zealand rabbit meat.

Treatments	Moisture (%)	WRC (%)	pH	Color		
				L^*	a^*	b^*
C	75.4±0.73	20.9 ± 5.06	6.33±0.1	41.6±2.7	2.5±0.6	1.7±0.5
S	75.5±1.40	20.6 ± 3.71	6.40±0.1	44.66±6.4	2.5±0.5	1.8±0.2
P	76.5±1.35	22.3 ± 3.79	6.37±0.1	46.0±0.5	2.7±0.5	1.3±0.1

WRC: Water retention capacity, C: Control, P: Avocado peel (2%), S: Avocado seeds (2%), L^* : lightness, a^* : redness, b^* : yellowness. Means ± SD (n=5).

Serum biochemical parameters: The serum biochemical parameters evaluated at the beginning, middle and end of the experimental period are presented in **Table 5**.

At the beginning of the experiment, the measured serum parameters were statistically similar across treatments ($p>0.05$), indicating the homogeneity of the experimental units. However, during the course of the experiment, the basal glucose concentration in the treatment (S) increased statistically ($p<0.05$) compared to the intermediate and final concentrations, which remained similar to each other. Regarding total cholesterol concentration, a decreasing trend was observed over time across all treatments. However, a statistically significant difference was only found between the baseline sampling and the intermediate and final sampling in treatment (P). In general, the trends in concentration changes were similar across the evaluated treatments, possibly due to the normal variation in physiological biochemical values throughout the individual's growth curve.

For some parameters and across treatments during the course of the experiment, certain differences were observed. Specifically, the basal glucose concentration in treatment (S) increased statistically ($p<0.05$) compared to the intermediate and final concentrations, which remained similar to each other. In general, trends in concentration changes remained similar. Reported serum glucose concentrations in rabbits range from 81-183 mg/dL (Hewitt *et al.*, 1989), 75-155 mg/dL (Melillo 2007), 99-148 mg/dL (Jenkins 2008), 75-150 mg/dL (Østergaard *et al.*, 2010), and 69-194 mg/dL (Özkan *et al.*, 2012).

Reported triglycerides levels range from 45 to 204 mg/dL (Østergaard *et al.*, 2010), while total cholesterol levels have been reported as 78 mg/dL (Caisey and King 1980), 10-80 mg/dL (Melillo 2007), and 4-100 mg/dL (Østergaard *et al.*, 2010). Regarding AST and ALT concentrations, reported values include 47 and 79 U/L (Caisey and King 1980), 35-130 and 45-80 U/L (Melillo 2007), 10-78 and 27-72 U/L (Jenkins 2008) and 10-120 and 12-80 U/L (Østergaard *et al.*, 2010). Therefore, in the present study, the concentration ranges

obtained from the biochemical parameters evaluated in the marginal vein sampling (B and I), and in the slaughter (F) are consistent with and fall within the optimal ranges established for healthy adult New Zealand rabbits. The

observed variations among treatments are possibly due to the normal fluctuation of physiological biochemical values during the individual's growth curve.

Table 5. Serum parameter concentrations at the beginning (B), intermediate (I) and end (E) of the experimental period.

Parameter	Treatment	Sample		
		B	I	E
Glucose (mg/dL)	C	100.9 ± 25.7	134.6 ± 13.5	113.6 ± 19.9
	P	120.4 ± 14.3	135.0 ± 12.7	116.8 ± 22.2
	S	97.4 ± 9.1 ^b	143.2 ± 11.8 ^a	129.7 ± 33.2 ^{a,b}
Total cholesterol (mg/dL)	C	100.6 ± 16.1	83.8 ± 21.5	81.9 ± 23.8
	P	94.6 ± 21.8 ^a	62.1 ± 10.9 ^b	60.7 ± 15.1 ^b
	S	93.1 ± 19.9	69.1 ± 13.4	80.9 ± 39.9
Triglycerides (mg/dL)	C	297.8 ± 93.4 ^a	126.0 ± 36.8 ^b	131.1 ± 64.0 ^b
	P	325.8 ± 95.1 ^a	110.9 ± 46.3 ^b	126.74 ± 50.6 ^b
	S	320.7 ± 64.4 ^a	100.2 ± 22.0 ^b	109.01 ± 18.0 ^b
ALT (U/L)	C	30.6 ± 8.1	37.2 ± 10.2	46.1 ± 19.3
	P	27.0 ± 7.6 ^c	34.0 ± 6.5 ^b	48.9 ± 9.1 ^a
	S	28.2 ± 1.7	28.2 ± 4.7	50.0 ± 18.6
AST (U/L)	C	32.7 ± 4.7	26.6 ± 6.4	41.3 ± 20.6
	P	36.0 ± 8.3	26.0 ± 3.1	47.2 ± 10.7
	S	35.5 ± 4.1	22.0 ± 2.5	52.2 ± 16.7

C: Control, P: Avocado peel (2%), S: Avocado seeds (2%), ALT: alanine transaminase, AST: aspartate transaminase. Means ± SD (n=5). ^a Means with different letter per row per parameter are statistically different (p<0.05).

Oxidative stability of meat: In the present study, the supplementation level used (2%) maintained a similar oxidation level ($p > 0.05$) across all treatments. We found MDA concentrations of 1.38 mg/kg (C), 1.14 mg/kg (P) and 1.05 mg/kg (S) in raw rabbit meat. Although no statistical differences were observed, there was a tendency for lower oxidation in treatments that included avocado peel or seed (Figure 1). These findings are similar to those reported by Dal Bosco *et al.* (2002), who found 1.99 mg MDA/kg meat, and are consistent with Kone *et al.* (2016), Botsoglou *et al.* (2004), and Rotolo *et al.* (2013), who observed no significant changes in meat oxidation status with oregano supplementation at concentrations of 10, 100, and 10,000 ppm, reporting similar MDA levels to their control treatment.

In meat such as pork the oxidation state decreases when 1000 ppm of essential oils are included in the diet (Alarcon-Rojo *et al.*, 2013), possibly due to difference in fat content compared to the lean tissue of rabbits.

Physical preservation procedures, such as irradiation and freezing are also influenced by oxidative processes, not only in red meats but also in white meats like rabbit and fish. Piccini *et al.* (1986) reported that the malondialdehyde (MDA) concentration ($\mu\text{g}/\text{per kg}$) was higher in irradiated tuna (412.32) than the frozen control treatment (215.19).

Thus, in the search for natural resource alternatives to enhance preservation and prevent oxidation in perishable products, direct supplementation is often chosen in the animal models, as in the present study, or by the direct addition of antioxidants on fresh or processed meat products. This helps prevent rapid deterioration and extends shelf life. However, in excessive amounts, antioxidants can alter the organoleptic qualities, which would be undesirable for consumers. Additionally, these compounds should not be used to conceal spoiled products or masks poor and/or inadequate processing conditions.

Extraction of lipids and quantification of their fatty acids: The quantification of lipid extract in meat yielded 10.9 ± 2.0 (P), 10.8 ± 0.4 (C), and 8.1 ± 0.3 (S) % in fresh meat and crushed by the extraction technique used. There were similarities with Cambero *et al.* (1991) between 5.2 to 10.5 % and Kone *et al.* (2016) from 11.2 to 14.3 % of total lipids, and representing a lower content than other types of meat such as pork and beef (Wood *et al.*, 2008).

The composition of fatty acids in meat can be modified through dietary reformulation and is influenced by the digestive characteristics of animals (Ahmad *et al.*, 2018). Rabbits, as monogastric animals, have nutritional needs that allow them to tolerate higher fiber content than other species with similar physiology, such as birds and pigs. This characteristic encourages research on the

evaluation of various fiber-enriched alternatives for supplementation, including the use of organic waste.

Chromatographic analysis revealed statistical similarities between treatments for the quantified free fatty acids. The content of palmitic acid was higher than the other quantified fatty acids, in accordance with Cambero *et al.* (1991), who reported that palmitic acid is the major fatty acid in the total and apolar lipids of New

Zealand rabbit meat. There were no statistical differences ($p>0.05$) between treatments for quantified free fatty acids (**Table 6**). The content of palmitic acid was higher than the other quantified fatty acids, in coordination with that described by Cambero *et al.*, (1991) who says that palmitic acid is the major fatty acid in the total and apolar lipids of rabbit meat New Zealand.

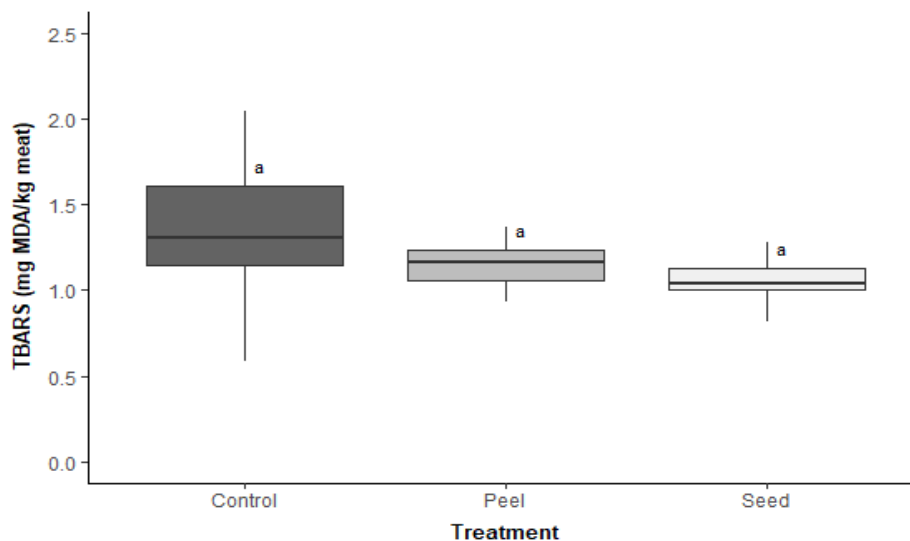


Figure 1. Quantification of malondialdehyde (MDA) in New Zealand rabbit meat. Means \pm SD (n=5).

Table 6. Concentration of free fatty acids (mg/100 g of fresh rabbit meat).

Treatment	Myristic C14	Palmitic C16	Stearic C18	Oleic C18:1	Linoleic C18:2
C	23.7 \pm 3.6	70.5 \pm 15.5	43.41 \pm 12.93	44.67 \pm 1.58	40.79 \pm 4.36
P	26.8 \pm 5.5	66.8 \pm 16.1	40.15 \pm 5.72	41.18 \pm 7.07	39.80 \pm 4.16
S	21.2 \pm 5.1	59.8 \pm 1.8	38.50 \pm 6.81	43.43 \pm 14.77	35.81 \pm 1.59

C: Control, P: Avocado peel (2%), S: Avocado seeds (2%). Means \pm SD (n=5).

Although a low dose of supplementation (2%) with avocado peels and seeds was used in the present study, higher doses could be incorporated into the rabbit diet to take advantage of the byproducts of this valuable natural resource, as they provide additional nutritional value as fiber and/or antioxidants. However, nutritional growth requirements should be considered when evaluating potential physiological and/or toxicological adverse effects, as well as the regulatory limits on supplementation, to prevent impairment of the organoleptic qualities of the meat.

Conclusion: The addition of 2% of avocado peels and seeds to the regular diet of rabbits does not alter their normal growth and metabolism. Furthermore, it does not affect the quality characteristics or oxidative stability of their meat. Therefore, the use of avocado agro-industrial by-products could be applied as supplementation in the rabbit diet without any adverse effects.

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