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**EFFECT OF TOTAL REPLACEMENT OF SOYBEAN MEAL BY LUPINE SEEDS  
(*L. ALBUS* AND *L. LUTEUS*) ON CARCASS CHARACTERISTICS  
AND MEAT FATTY ACIDS COMPOSITION OF GROWING RABBITS**

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## EFFECT OF TOTAL REPLACEMENT OF SOYBEAN MEAL BY LUPINE SEEDS (*L. ALBUS* AND *L. LUTEUS*) ON CARCASS CHARACTERISTICS AND MEAT FATTY ACIDS COMPOSITION OF GROWING RABBITS

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### ABSTRACT

The present study aimed to evaluate the effect of total replacement of soybean meal (SBM) by two species of lupine seeds (*L. albus*, cv. Nacional and *L. luteus*, cv. Mister) on carcass characteristics and fatty acids composition of growing hybrid of rabbits both sexes (New Zealand x Californian) at slaughter. Three diets were formulated to be nearly isonitrogenous and isoenergetic; the control diet (CTD), containing 15% of SBM as the main protein source, and the other two diets with complete replacement of SBM by seeds of LA (LAD) and LL (LLD). At weaning (35 days), rabbits were housed in groups of 2 and randomly assigned into 3 groups according to diet (ten replicates per treatment) and controlled for 5 weeks. At the end of the trial (69 days age), thirty rabbits (ten of each treatment) were slaughtered, and the head, liver, hind leg and dissectible fat were weighed to estimate each percentage in relation to the whole carcass. Colour and pH were measured after 24h chilled carcass. Fatty acid composition and cholesterol content were also determined. Total replacement of soybean meal by lupine seeds had no significant effect ( $p>0.05$ ) on the contribution of the dressing out percentage, carcass part yield, dissectible fat, pH and colour parameters. Rabbits fed with LLD presented a similar ( $p<0.05$ ) final weigh to those fed with CTD; however, LAD had a negative effect on weight at slaughter. Diets containing lupine seeds seem to have had a negative effect on the amount of SFA content compared to the control diet ( $p<0.01$ ). Both, LAD and LLD diets presented higher PUFA values (27.44 and 28.98 vs 24.12 for control,  $p<0.01$ ) The results of PUFA n-6/ PUFA n-3 ratio were higher in rabbits fed with LLD and the lowest in samples from the LAD group. Incorporation of lupine seeds in the diets did not affect carcass characteristics but improved the fatty acid profile.

**Key words:** rabbit, feed, lupines, meat, fatty acids.

### INTRODUCTION

In the context of a strong EU dependence on the import of soybean meal (SBM) to meet protein requirements in the formulation of compound feeds, it is important to study alternative proteins sources. Lupine is one of the grain legumes grown in Europe that can be used as a source of protein for livestock feeding (Gresta *et al.*, 2010). It has been proved that it can supplement and replace other protein sources without negative impact on performances of growing rabbits (Gugotek *et al.*, 2018; Uhlířová *et al.*, 2018) and can have a positive impact of the fatty acid profile of rabbit meat (Volek and Marounek, 2011; Volek *et al.*, 2018).

Thus, the present study aimed to evaluate the effect of total replacement of SBM by two species of lupine seeds (*L. albus*, cv. Nacional and *L. luteus*, cv. Mister) on carcass characteristics and fatty acids composition at the slaughter of growing hybrids rabbits New Zealand x Californian.

## MATERIALS AND METHODS

### Animals and experimental design

Sixty weaned hybrid rabbits (New Zealand x Californian) from both sexes were kept in a closed air-conditioned building maintained at 18-23°C and with a day/night cycle of 12/12 h (light from 07:00 to 19:00) five weeks, from 35 days (weaning) to 69 days (slaughter). Rabbits were housed in groups of 2 and randomly divided into 3 groups according to diet (ten replicates per treatment). Three diets were formulated to be nearly isonitrogenous and isoenergetic, according to the De Blas and Mateos (2010) recommendations for growing rabbits. The control diet (CTD), containing 15% of SBM as the main protein source, and the other two diets with complete replacement of SBM by seeds of LA (LAD) and LL (LLD) (Table 1).

At the end of the trail, at 69 days age, ten rabbits from each treatment rabbits were slaughtered through cervical dislocation, and the carcasses were refrigerated for 24h before sample analysis and collection. The head, liver, hind leg and dissectible fat were weighed to estimate each percentage in relation to the whole chilled carcass. The *Longissimus dorsi* muscle was extracted and used to determine colour and pH and stored for later analysis. Colour analysis was determined with a portable colourimeter and pH was measured using a portable pH meter with a penetrating electrode.

### Chemical Analyses

Dried samples of the experimental diets were ground to pass a 1mm screen and were analysed, according to the AOAC (1990) procedures. For dry matter (DM), crude protein (CP), ether extract (EE), neutral detergent fibre expressed exclusive of residual ash (NDFom), acid detergent fibre expressed exclusive of residual ash (ADFom), sulphuric acid lignin (Lignin (sa)) and starch. Fatty acid composition and cholesterol content were determined according to Quiñones *et al.* (2019) and Lorenzo and Pateiro (2013).

**Table 1:** Ingredients and chemical composition (g/kg as-fed basis) of the experimental diets

	CTD	LAD	LLD
<b>Ingredients</b>			
Soybean meal	150	-	-
White lupin seed	-	150	-
Yellow lupin seed	-	-	150
Wheat	149	21	92
Wheat bran	155	350	350
Sugar cane molasses	10	10	10
Lucerne	189	263	127
Grape seed meal	50	50	50
Wheat straw	120	32	107
Beet pulp	150	80	80
Mineral-vitamin premix <sup>1</sup>	10	10	10
<b>Chemical composition</b>			
Dry matter	916	913	910
Crude protein	161	164	163
Ether extract	35	34	42
NDFom	346	344	364
ADFom	217	227	212
Lignin (sa)	52	64	55
Starch	124	81	120

<sup>1</sup> Mineral and vitamin mixture (per kg of feed): calcium more sal 16; vitamin A, 10,000 IU; vitamin D3, 1080 U; vitamin E, 36 mg; niacinamide, 50 mg; Ca- pantothenate, 20 mg; folic acid, 5 mg; Fe, 78 mg; Cu, 14 mg; Co, 0.5 mg; Mn, 20 mg; Zn, 60 mg; Se, 0.05 mg; I, 1.1 mg; choline chloride, 260 mg. b 0.5% lysine, 0.1% methionine, 0.16% Methionine + Cysteine, 0.8% threonine, 0.04% tryptophan, 0.57% arginine, 0.37% de valine, 0.3% isoleucine; 0.49% leucine.

### Statistical Analysis

Statistical analysis was performed with JMP version 14 program (SAS Institute, Cary, NC, USA). Data were submitted to one-way analysis of variance (ANOVA) with the diet as the main factor, using the General Linear Model procedure. Tukey's multiple comparison test was used to compare the means values when a significant effect is observed. Statistical significance was accepted at  $P < 0.05$ .

## RESULTS AND DISCUSSION

The results showed that total replacement of soybean meal by lupine seeds had no significant effect ( $p > 0.05$ ) on the contribution of the dressing out percentage, organ weight, hind leg weight, dissectible fat, pH and colour parameters (Table 2). Several authors reported similar results when incorporating *L. albus* in the finishing diet of rabbits (Gugotek *et al.*, 2018; Uhlřřov *et al.*, 2018) although there are no published studies, to our knowledge, involving *L. luteus* incorporation in rabbits' diets.

Rabbits fed with LLD presented a similar ( $p < 0.05$ ) final weigh to those fed with CTD; however, LAD had a negative effect on weight at slaughter. Unlike Volek *et al.* (2014), which found no differences ( $p > 0.05$ ) on performances of rabbits fed with a diet with 15% of *L. albus* inclusion, on this study the LAD diet presented lower values for live weight at slaughter. Nevertheless, the authors used a different variety of *L. albus*, which could justify these differences.

**Table 2:** Effect of lupines incorporation in diets on carcass characteristics of growing rabbits (n=10)

	CTD	LAD	LLD	SEM*	P
Slaughter live weight, g	2739.0 <sup>a</sup>	2605.8 <sup>b</sup>	2731.1 <sup>ab</sup>	40.37	0.047
Dressing out percentage (%)	60.5	60.6	59.4	0.42	0.104
As refrigerd carcass yield (%)					
Head	7.98	7.97	7.96	0.18	0.996
Liver	7.69	7.52	7.05	0.27	0.244
Dissecable fat	2.89	2.83	2.91	0.15	0.923
Hind leg	29.7	29.7	29.9	0.32	0.940
pHu (refrigered carcass)	6.0	6.0	5.9	0.07	0.909
Colour of <i>longissimus dorsi</i>					
lightness, L*	46.6	45.7	44.3	1.01	0.293
redness, a*	3.5	3.8	4.5	0.69	0.569
yellowness, b*	11.0	11.2	10.9	0.37	0.781

CTD, control diet (soybean meal); LAD, *Lupinus albus* diet; LLD, *Lupinus luteus* diet; SEM, standard error of mean.

<sup>a, b</sup> Means with different letters in the same row differed ( $P < 0.05$ ).

Diets containing lupine seeds seem to have had a negative effect on the amount of SFA content compared to the control diet (Table 3), which was also observed by Volek and Marounek (2011). The significantly lower content of PUFA was found by the same authors when comparing a diet supplemented with *L. albus* with another containing sunflower meal. In another study, the amount of PUFA was similar in soybean meal diets and lupine ones (Volek *et al.*, 2018), however in the present study, both LAD and LLD diets presented higher PUFA values. The results of PUFA n-6/ PUFA n-3 ratio were higher for LLD, although both Volek and Marounek (2011) and Volek *et al.* (2018) showed similar results in terms of PUFA n-6/ PUFA n-3 ratio for diets with *L. albus* to those found in this study. The low ratio found in the meat of rabbits fed with LAD presents a positive result since it might contribute to human health when consumed (Ulbricht *et al.*, 1991). Cholesterol content was significantly lower in the *longissimus* muscle of rabbits fed with diets in which *Lupinus luteus* was incorporated.

**Table 3:** Effect of lupines incorporation in diets on fatty acids composition of fattening growing rabbits (n=10)

	CTD	LAD	LLD	SEM	P
Cholesterol (mg/100mg)	28.60 <sup>a</sup>	28.46 <sup>a</sup>	25.56 <sup>b</sup>	0.470	0.0072
Total SFA (mg/g of fat)	44.21 <sup>a</sup>	38.67 <sup>c</sup>	40.47 <sup>b</sup>	0.502	<0.0001
Total MUFA (mg/g of fat)	31.66 <sup>b</sup>	33.88 <sup>a</sup>	30.54 <sup>b</sup>	0.380	0.0003
Total PUFA (mg/g of fat)	24.12 <sup>b</sup>	27.44 <sup>a</sup>	28.98 <sup>a</sup>	0.496	<0.0001
PUFA n-6/ PUFA n-3 ratio	7.08 <sup>b</sup>	5.40 <sup>c</sup>	8.75 <sup>a</sup>	0.261	<0.0001

CTD, control diet (soybean meal); LAD, *Lupinus albus* diet; LLD, *Lupinus luteus* diet; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; SEM, standard error of mean. <sup>a, b, c</sup> Means with different letters in the same row differed (P<0.05).

## CONCLUSIONS

The results of this study show that it is possible to replace soybean meal for lupine seeds in rabbits feed with small differences in carcass characteristics. However, the replacement affected the fatty acid profile of fattening growing rabbits and cholesterol levels, but it is necessary to study further the impact of including *L. luteus* in rabbit feed. Different levels of incorporation need to be studied, as well as the inclusion of lupines as a prebiotic and its effect on villi health and cecal microbiota.

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# Effect of Total Replacement of Soybean Meal by Lupine Seeds (*L. albus* and *L. luteus*) on Carcass Characteristics and Meat Fatty Acids Composition of Growing Rabbits

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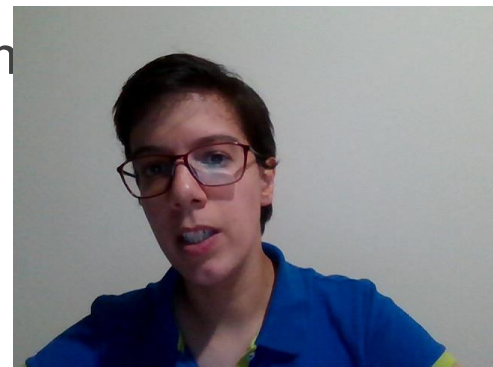
- ▶ EU dependence on the import of soybean meal (SBM): alternative protein sources.
- ▶ Lupines:
  - ▶ No impact on performances of growing rabbits and positive impact of the fatty acid profile of rabbit meat.
- ▶ **Aim: effect of total replacement of SBM by white and yellow lupins (*L. albus* and *L. luteus*) on carcass characteristics and fatty acids composition.**

# Introduction



# Materials and Methods

- ▶ 60 weaned hybrid rabbits (New Zealand x Californian) from 35 days (weaning) to 69 days.
- ▶ Groups of 2 rabbits randomly divided into 3 diets (ten replicates per treatment).
- ▶ 3 diets nearly isonitrogenous and isoenergetic: control diet (CTD: 15% of SBM), two diets (LAD e LLD: complete replacement of SBM by seeds *L. albus*, cv. Nacional and *L. luteus*, cv. Mister)
- ▶ Slaughter: 69 days age, ten rabbits from treatment.





# Materials and Methods

- ▶ Carcasses: refrigerated for 24h before sample analysis and collection.
- ▶ Head, liver, hind leg and dissectible fat were weighed: estimation of percentage in relation to the whole chilled carcass.
- ▶ *Longissimus dorsi* muscle: extracted to determine colour and pH and stored for fatty acid composition and cholesterol analysis.



	CTD	LAD	LLD	SEM*	P
Slaughter live weight, g	2739.0 <sup>a</sup>	2605.8 <sup>b</sup>	2731.1 <sup>ab</sup>	40.37	0.047
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Colour of <i>longissimus dorsi</i>					
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	CTD	LAD	LLD	SEM	P
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PUFA n-6/ PUFA n-3 ratio	7.08 <sup>b</sup>	5.40 <sup>c</sup>	8.75 <sup>a</sup>	0.261	<0.0001



# Conclusions

It is possible to replace soybean meal for lupine seeds in rabbits feed with small differences in carcass characteristics.

The replacement affected the fatty acid profile of fattening growing rabbits and cholesterol levels.

Further the impact of including *L. luteus* in rabbit feed.

Future studies: prebiotic, villi health and cecal microbiota.





- ▶ The authors acknowledge financial support through project INTERACT - “Integrative Research in Environment, Agro-Chains and Technology”, reference NORTE-01-0145-FEDER-000017, in its line of research entitled ISAC, co-financed by the European Regional Development Fund through NORTE 2020 (North Regional Operational Program 2014/2020).
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