

NUTRITIVE VALUE OF CITRUS CO-PRODUCTS IN RABBIT FEEDING

DE BLAS J.C.*, FERRER P.†, RODRÍGUEZ C.A.*, CERISUELO A.‡, GARCÍA REBOLLAR P.*,
CALVET, S.†, FARIAS C.*

*Departamento de Producción Agraria, ETSI AAB, UPM, 28040, MADRID, Spain.

†Instituto de Ciencia y Tecnología Animal. Universitat Politècnica de València. Camino de Vera s.n. 46022 VALENCIA, Spain.

‡Centro de Investigación y Tecnología Animal, Instituto Valenciano de Investigaciones Agrarias, Pol. La Esperanza 100, 12400
SEGORBE, Castellón, Spain.

Abstract: Pulps from different citrus fruits are relevant agro-industrial co-products in the Mediterranean area in terms of amounts produced and availability. Moreover, part of the product is dehydrated, which increases its interest in monogastric species such as rabbits. Seventy eight samples from various Spanish producers using several types of fresh fruits (orange, tangerine, lemon and pomelo) and different processing methods of orange and tangerine samples (either fresh or dried after adding $\text{Ca}(\text{OH})_2$) were analysed for their chemical composition and *in vitro* digestibility. Average dry matter (DM) contents of ash, neutral detergent fibre, acid detergent fibre, acid detergent lignin (ADL), soluble fibre, crude protein (CP), insoluble neutral and acid detergent CP, ether extract and gross energy were 49.0, 226, 139, 12.1, 213, 71.2, 13.1, 4.2, 30.5 g and 17.8 MJ/kg DM, respectively. Mean DM and CP *in vitro* digestibility were 86.7 and 95.6%, respectively. Digestible energy was estimated to be 15.1 MJ/kg DM. A high variability (coefficient of variation from 17% for CP to 60% for ADL) was observed among the samples for most of the traits studied, which was partially explained by the effects of type of fruit and processing. Lemon pulps had on average higher ash and fibre but lower sugar contents than the other pulps. Dehydration processes increased ash content (almost double than for fresh pulp) due to lime addition. As regards the current results, citrus pulp has potential for use in rabbit diets as a source of energy and soluble fibre.

Key Words: citrus pulp, type of fruit, processing, nutritive value, rabbits.

INTRODUCTION

The utilisation of agro-industrial co-products as animal feeds can be expected to have a positive economic impact and a reduction of the environmental burden. Citrus production is a major sector worldwide, covering a relatively stable surface of almost 10 million hectares and output of roughly 150 million tons. From the different citrus fruits, orange (50% of total production) is the most important, followed by tangerines (30% of total), lemons (12%) and pomelo (8%). China and Brazil are the main producers, but countries in the Mediterranean area, and particularly Spain, are also relevant (Faostat, 2017).

An important part of citrus production is transformed. In Spain, around 20% of total citrus production is transformed (MAPAMA, 2017), while more than 50% is processed in large producers such as United States or Brazil (Braddock, 1999; USDA, 2016). Moreover, a part of the citrus pulp production is dehydrated, which facilitates its transport and inclusion in feed formulations. Pulps obtained from citrus processing are therefore relevant co-products in terms of amounts produced and availability. Accordingly, the use of citrus pulps is increasingly interesting as an energy source for ruminants, but also for some monogastric species such as rabbits.

The main operations for citrus juice extraction and pulp production are reviewed by Braddock (1999) and include extraction, finishing and centrifugation. Additionally, essential oils and essences may be recovered. Citrus pulp is

Correspondence: J.C. de Blas, c.deblas@upm.es. Received May 2017 - Accepted November 2017.
<https://doi.org/10.4995/wrs.2018.7699>

produced as a final by-product, and are composed of peels, seeds and fruit pulp. The amount of this by-product accounts for one half of the wet mass of the whole fruit, and can be dried to facilitate its transport and use. Lime ($\text{Ca}[\text{OH}]_2$) is normally added to aid the dehydration, which also requires an important energy input. Finally, the dehydrated citrus pulp is usually pelleted, adding in some cases citrus molasses to the lime before the drying operation. Citrus molasses are produced by concentration of some of the water and sugars mechanically recovered by pressing the residue before the dehydration.

The chemical composition of citrus pulp varies widely according to the type of fruit, growing conditions, ripeness, climate and the manufacturing process used (Martínez-Pascual, 1977; Bampidis and Robinson, 2006). According to several databases (INRA, 2004; Bampidis and Robinson, 2006; FEDNA, 2010; CVB, 2016; SIA, 2017 and Heuzé *et al.*, 2017), citrus pulp is characterised by a high proportion of sugars, although the different sources vary in the proportion assigned to this component (from 20 to 40%). The concentration of insoluble (NDF) and soluble fibre (SF, mostly pectins) is also relevant, accounting for around 19 and 30%, respectively, as average. There is also a significant ash content (about 6.5%) with a high proportion of Ca but low in P. Instead, both the crude protein (CP) and the essential amino acid levels (especially methionine and total sulphur amino acids) are low (about 2.8 and 2.3% of the total CP content, respectively) compared to rabbit requirements (4.9 and 3.5% for growing rabbits; De Blas and Mateos, 2010).

The aim of the current study was to measure the nutrient composition and *in vitro* digestibility of different citrus pulp co-products obtained from commercial industries using different manufacturing procedures, in order to evaluate the potential nutritive value for rabbits.

MATERIALS AND METHODS

Samples

A total of 78 samples of citrus pulps were surveyed during the citrus harvest season 2015/2016. Samples were provided by different commercial citrus plants in Spain, considering various times of sampling during the production season and diverse types of fruits. Samples were collected from the industry either fresh or dried. Information on pulp dehydration was provided by the industries supplying the samples. The drying process consisted of adding lime at a variable proportion to enable partial mechanical dehydration and final heat drying at 135-155°C.

Chemical analysis

Fresh samples were frozen and freeze dried before analyses. All samples were ground to 1 mm pore size. Dry matter, ash, crude fibre and ether extract were determined using procedures 934.01, 942.05, 978.10 and 920.39, respectively, from AOAC (2000). Total N was measured by combustion, using Leco equipment (model FP-528, Leco Corporation, St. Joseph, MI, USA) and CP content was estimated as an N content 6.25 (AOAC procedure 986.06). Total sugars were analysed according to the method of Yemm and Willis (1954). Total dietary fibre (TDF) was measured following procedure 985.29 (AOAC, 2000). Neutral detergent fibre, acid detergent fibre and acid detergent lignin (NDF, ADF and ADL, respectively) were determined sequentially using the filter bag system (Ankom Technology Corp., Macedon, NY, USA) according to Mertens (2002), AOAC (2000; procedure 973.187) and Van Soest *et al.* (1991), using heat stable amylase (42FAA, Ankom Technology Corp., Macedon, NY, USA) and expressed without residual ash. The SF concentration was calculated by difference as TDF–NDF (Van Soest *et al.*, 1991; Abad *et al.*, 2013). This method was compared with others by Abad *et al.* (2013), tending to render a higher value of soluble fibre. Otherwise, a European Ring-Test performed using this estimation in several laboratories (Trocino *et al.*, 2013) showed a high relation with digestive and health traits in rabbits, which is also supported by Gidenne (2015). Hemicelluloses and cellulose contents were estimated from the difference between NDFADF and ADFADL, respectively. The proportion of neutral and acid detergent insoluble CP (NDICP and ADICP, respectively) was determined following the standardised procedures of Licitra *et al.* (1996), by analysing the N content (combustion method) in the NDF and ADF residues, respectively. Calcium was determined by atomic absorption spectrophotometric and colorimetric methods (procedure 991.25, AOAC 2000). Gross energy concentration was measured in an isoperibolic bomb calorimeter (Parr 6400, Parr Instruments Co., Moline, IL, USA).

***In vitro* analysis**

From the 78 samples chemically analysed, a group of 33 was selected searching for a wide variation in SF, NDF and sugars concentrations, and were analysed for *in vitro* DM and CP digestibility in rabbits. *In vitro* analyses were performed following the method proposed by Ramos *et al.* (1992) based on that of Boisen *et al.* (1991), and using Ankom bags following the modification proposed by Abad *et al.* (2013). Samples, previously ground to 1 mm pore size, were weighed (0.5 g) into filter bags (Ankom Technology Corp., Macedon, NY, USA) and placed in a Daisy II incubator jar (3.5 L and 30 filter bags/jar). A magnetic rod, 25 mL of phosphate buffer (0.1 M, pH 6.0) and 10 mL of 0.2 M HCl solution were added for each bag (750 mL of phosphate buffer and 300 mL of HCl solution for each jar with 30 bags). The solutions were mixed carefully by gentle magnetic stirring, the pH was measured and then adjusted to pH 2 with 1 M HCl or 1 M NaOH solutions. Then, a 1 mL/bag (30 mL/jar) of fresh pepsin solution (25 mg of pepsin (pepsin from porcine, 2000 FIP-Units/g protein, Merck n 7190)/mL 0.2 M HCl) was added and mixed. The jar was closed and the samples incubated in an oven at 40°C for 1.5 h. After this incubation, 10 mL/bag (300 mL/jar) of fresh pancreatin solution (100 mg of pancreatin [pancreatin from porcine, grade VI]/mL phosphate buffer pH 6.8) was added, the jar closed and the samples incubated at 40°C for 3.5 h. After the second incubation, the pH was adjusted to 4.8 by adding acetic acid and the 0.5 mL/bag (15 mL/jar) of Viscozyme (Viscozyme 120 L, 120 FBG/G, Novo Nordisk) was added, mixed and incubated at 40°C for 16 h. The entire soluble residue was then removed and the bags were washed in the jar with water at 40°C 3 times and mixed carefully (the first one for 30 min at 40°C with agitation in the incubator), followed by rinsing with acetone to prevent adherence of any residue in the bags. Finally, the bags were dried at 103°C for 24 h. Two bags without any sample were used as blanks in each jar. *In vitro* DM digestibility (DMd_{iv}) was calculated as:

$$DMd = 1 - \frac{W_3 - (W_1 \times C_1)}{W_2}$$

Where W_1 is the bag tare weight, W_2 is the dry sample weight, W_3 is the final oven dry weight of the bag after digestive process and C_1 is the blank bag correction (final oven dry weight of the blank bag after digestion/original blank bag weight). Additionally, the CP concentration was measured in the dry residue to determine the *in vitro* CP digestibility (CPd_{inv}).

Statistical analysis

The effects studied were analysed using the MIXED procedure from SAS (2008), considering the supplier as a random effect and type of fruit and its processing as main fixed effects. Interactions were not measured because the sample collection was not structured, as only orange and tangerine samples were available to study the effects of processing. Most of the dried samples ($n=17$) were obtained from one of the processing plants surveyed, and used to compare the effects of processing against fresh samples ($n=15$) from the same origin. The Tukey test was used for multiple mean comparisons among types of fruit. Proc CORR of SAS (2008) was performed to determine correlations among chemical variables. A stepwise regression analysis was used to predict the *in vitro* DM and CP digestibility from the chemical composition, using PROC REG of SAS (2008). The stepwise procedure only introduces variables in the model when they contribute to a significant improvement ($P<0.05$) in the estimation of the dependent variable.

RESULTS

The mean values for each analytical component and its variability (expressed from its range, standard deviation and coefficient of variation) for the whole set of data samples surveyed are presented in Table 1. The results indicate that sugars (40.8%), soluble fibre (21.3%) and neutral detergent fibre (NDF, 22.6%) were the main citrus pulp components, with a relevant variation among samples ($CV=25-32\%$). Variability was even greater for other feed constituents such as ash, ADL or NDICP and ADICP, ($CV=49.2-60.5$). In contrast, GE content or *in vitro* digestibilities of DM and CP varied in a lesser range ($CV=3-7\%$).

The correlation matrix among the chemical components is shown in Table 2. Neither CP or ether extract (EE) contents were significantly correlated to any of the other components studied. Otherwise, there was a high negative relation between sugars and all the fibrous traits, especially for TDF ($r=-0.89$) and ADF ($r=-0.84$). Likewise, it can be

Table 1: Mean and variability of the chemical composition (%DM) and *in vitro* digestibility (%) of 78 samples of citrus pulps.

Traits	n	Mean	Minimum	Maximum	SD	CV
Ash	78	4.90	3.0	20.3	2.88	58.8
Crude protein	78	7.12	5.12	11.2	1.2	16.9
Ether extract	78	3.05	1.0	8.3	1.4	46
Crude fibre	78	12.3	6.4	28.6	3.98	32.4
Neutral detergent fibre	78	22.6	11.8	43.1	5.8	25.7
Acid detergent fibre	78	13.9	8.3	23.2	3.0	21.6
Acid detergent lignin	78	1.21	0.34	5.18	0.72	60
Hemicelluloses	78	8.70	3.0	24.8	4.0	46
Cellulose	78	12.7	7.4	18.2	2.7	21.3
Total dietary fibre	78	43.9	22.3	70	9.2	21.1
Soluble fibre	78	21.3	6.8	42.2	6.8	32
NDICP	78	1.31	0.31	2.9	0.64	49.2
ADICP	78	0.42	0	1.07	0.24	60
Sugars	67	40.8	17.6	64.9	10.5	25.7
Gross energy (MJ/Kg DM)	78	17.8	15.6	19.2	0.6	3.37
DMd _{iv}	33	86.7	78.2	97.4	5.7	6.59
CPd _{iv}	10	95.6	89.4	97.7	3.1	3.3

NDICP: Neutral detergent insoluble crude protein; ADICP: Acid detergent insoluble crude protein; DMd_{iv}: Dry matter digestibility *in vitro*; CPd_{iv}: Crude protein digestibility *in vitro*.

observed that SF was positively associated with several insoluble fibre traits, as CF and NDF. The correlation between total and soluble fibre contents was very high ($r=+0.79$).

The effects of type of fruit (orange, tangerine, lemon and pomelo) and processing type (fresh or dried+Ca[OH]₂) on the chemical composition and *in vitro* digestibility of DM and CP are presented in Table 3. All the dried samples in the survey corresponded to orange and tangerine. Accordingly, the effect of type of fruit was measured using only fresh samples. The results indicate that fresh lemon pulp had a lower content of sugars and GE together to a generally

Table 2: Correlation (Pearson's coefficient) matrix¹ among chemical composition in 78 samples of citrus pulps.

Traits	CF	NDF	ADF	ADL	Hem	Cell	TDF	SF	CP	NDICP	ADICP	EE	Sugars
NDF	0.57												
ADF	0.73	0.57											
ADL	0.37	0.37	0.5										
Hem	0.28	0.28	0.35	NS									
Cell	0.71	0.71	0.97	0.29	0.40								
TDF	0.69	0.69	0.81	0.49	0.36	0.76							
SF	0.45	0.45	0.45	0.48	-0.24	0.37	0.79						
CP	NS												
NDICP	NS	NS	NS	0.30	0.39	NS	NS	NS	NS				
ADICP	NS	NS	0.29	0.54	NS	NS	NS	NS	0.26	0.76			
EE	NS	NS	NS	NS									
Sugars	-0.68	-0.67	-0.84	-0.54	-0.34	-0.81	-0.89	-0.63	NS	-0.26	-0.38	-0.23	
GE	-0.42	-0.42	-0.28	NS	NS	-0.27	-0.35	-0.25	0.47	-0.28	NS	NS	0.37

¹Only significant ($P<0.05$) correlation coefficients are shown.

CF: Crude fibre; CP: Crude protein; NDF: Neutral detergent fibre; ADF: Acid detergent fibre; ADL: Acid detergent lignin; Hem: Hemicellulose=NDF-ADF; Cell: Cellulose= ADF-ADL; TDF: Total dietary fibre; SF: Soluble fibre; NDICP: Neutral detergent insoluble crude protein; ADICP: Acid detergent insoluble crude protein; EE: Ether extract.

Table 3: Effect of type of fruit (in fresh samples) and type of processing (within orange and tangerine samples) on chemical composition of citrus pulps (%DM).

	n	Traits												
		Moisture	Ash	CF	NDF	ADF	ADL	SF	Sugars	CP	NDICP	ADICP	EE	GE
Type of fresh fruit														
Orange	34	10.7	3.53 ^a	10.4 ^a	20.5	12.6 ^a	0.88	19.8 ^{ab}	46.8 ^b	7.01	0.97	0.23	3.20	18.0 ^b
Tangerine	10	10.3	3.74 ^a	10.9 ^a	21.1	12.9 ^a	1.11	18.9 ^{ab}	45.8 ^b	7.95	1.07	0.34	2.40	18.3 ^b
Lemon	9	10.0	4.47 ^b	16.3 ^b	31.2	17.4 ^b	1.33	25.5 ^b	28.8 ^a	7.15	1.34	0.31	3.90	17.3 ^a
Pomelo	4	14.5	4.23 ^{ab}	14.3 ^{ab}	25.2	16.5 ^b	0.89	14.9 ^a	44.6 ^b	7.48	1.18	0.23	3.45	17.8 ^{ab}
Type of processing (orange and tangerine samples) within the same processing plant														
Fresh	15	11.0	3.75	12.5	21.7	14.5	1.14	21.0	43.7	7.68	1.02	0.32	2.76	18.3
Dried	17	12.5	6.68	11.2	21.8	13.2	1.50	22.2	41.0	6.96	2.23	0.62	2.36	17.4
SD		3.17	0.55	1.70	3.51	1.65	0.57	5.39	6.64	0.89	0.32	0.20	1.04	0.39
<i>P</i> -value														
Type of fruit		0.32	<0.001	<0.001	<0.001	<0.001	0.075	0.035	<0.001	0.22	0.20	0.18	0.18	0.001
Processing		0.21	<0.001	0.039	0.95	0.040	0.090	0.46	<0.001	0.18	<0.001	<0.001	0.30	<0.001

CF: Crude fibre; NDF: Neutral detergent fibre; ADF: Acid detergent fibre; ADL: Acid detergent lignin; SF: Soluble fibre; CP: Crude protein; NDICP: Neutral detergent insoluble crude protein; ADICP: Acid detergent insoluble crude protein; EE: ether extract; GE: Gross energy (MJ/kg DM). n: Number of samples

^{a, b}Means with different superscripts in the same column differ ($P < 0.05$).

higher level of soluble and insoluble fibrous components than the other citrus pulps. Neither CP, NDICP, ADICP or EE concentrations were affected by type of fruit. Orange and tangerine pulps did not differ for any chemical constituent, so they were analysed together to test for the effects of processing. As shown in Table 3, the addition of Ca(OH)₂ before drying led to a decrease in sugar and GE concentration and to an increase of ash and some fibrous traits (ADF, ADL, NDICP and ADICP) compared to fresh samples.

In vitro digestibilities of DM (DMd_{iv}) and CP (CPd_{iv}) were high (respectively 86.7 and 95.6% on av.) with little variation among samples (standard deviation [SD]=5.86 and 3.44). A stepwise regression analysis, using as independent variables all the chemical traits measured, included only CF in the prediction equations of DM and CP *in vitro* digestibility:

DMd_{iv} (%) = 93.9 (±2.65, standard error of mean [SEM]) - 0.61 (±0.20) CF (n=33); R²=0.22; P=0.005

CPd_{iv} (%) = 102.3 (±2.69) - 0.67 (±0.023) CF (n=10); R²=0.52; P=0.02

The regression equation derived by Villamide *et al.* (2009) was used to estimate the digestible energy (DE) content for rabbits of the citrus pulp samples from its measured DMd_{iv} and EE concentration:

DE (MJ/kg DM) = -1.51 + 0.180 DMd_{iv} (%) + 0.32 EE (%DM); R²=0.69; residual standard deviation=0.46.

The calculated values averaged 15.1 MJ/kg DM with a range between 13.2 and 17.0 MJ/kg DM.

DISCUSSION

The results of this study show that sugars are the main component in the citrus pulps studied, averaging 40.8% DM. This value is higher than most of those published in the literature (e.g. Bampidis and Robinson, 2006), which might be related to the systematic re-addition of molasses in most current Spanish commercial dried citrus pulp systems. The proportion of sugars was lower than average both in lemon and Ca(OH)₂ processed samples (28.8 and 41.0% DM, respectively). These changes were parallel to inversely higher contents in ADF concentration. A relatively low sugar and high ADF content in lemon pulp was also reported by Bampidis and Robinson (2006), whereas Martínez-Pascual and Fernández-Carmona (1980a) found that the proportion of ADF increased in direct relation to the amount of Ca(OH)₂ added to the pulp before drying.

The concentration of insoluble fibre (estimated by the NDF content) was also important (22.6% DM as average), with higher levels in lemon (31.2% DM) and pomelo (25.2% DM) samples and no significant differences among

processing methods. The fibrous fraction tends to be poorly digested in rabbits (De Blas *et al.*, 1992; Wiseman *et al.*, 1992), due to the relatively short fermentation time in this species. However, the extension of fibre digestion is inversely related to the proportion of lignin (García *et al.*, 2000; De Blas, 2013), so digestibility of NDF in citrus pulp should be appreciable because of its low degree of lignification (5.35% ADL on NDF, as average in the present study).

There is little previous information on the SF concentration in citrus pulps. The mean value obtained in the current data set was quite relevant (21.3% DM), with the highest value corresponding to the lemon pulp (25.5% DM). Soluble fibre has importance in rabbit nutrition, as several recent reviews on the use of beet and apple pulp in rabbit diets (De Blas, 2013; Trocino *et al.*, 2013; Abad, 2015) have proven a high digestion efficiency for this component in rabbits, in addition to a positive effect on mortality prevention in growing animals. There is no former information published on the effects of the dietary inclusion of citrus pulp on these traits, but it might be expected that they would be similar to those observed for other sources of SF.

Citrus pulp does not promote gut motility, increasing retention time in the gut (Fraga *et al.*, 1991), probably because of the low proportion of long-sized particles in this type of fibre and the presence of soluble fibre that forms gels in the digesta. Consequently, its inclusion in substitution for traditional sources of fibre (such as alfalfa hay) resulted in a decrease of feed intake and growing performance (Pérez de Ayala *et al.*, 1991). Besides, this result may be related to the water soluble pectins contained in citrus pulp, which form gels in the gut, increasing the viscosity and promoting satiety. Citrus pulp also contains essential oils (such as limonin) characterised by a bitter taste. However, several studies reviewed by Heuzé *et al.* (2017) have not observed a decrease of rabbit performance at levels of inclusion up to 25%, when overall diets provided a sufficient level of long sized insoluble fibre. Similar conclusions have been obtained in recent work with growing pigs fed balanced diets containing 15 or 20% of citrus pulp (Antezana *et al.*, 2015; Beccaccia *et al.*, 2015, respectively).

Ash content in citrus pulp was variable, with the highest concentration (6.68% DM as average) observed for samples dried after $\text{Ca}(\text{OH})_2$ addition and higher levels in lemon than in other fresh pulps. Consequently, GE concentration was lower in these samples than for the average of the data set studied. In addition, most of the total ash content (38%) corresponded to Ca, which can lead to problems when trying to control the dietary Ca:P ratio.

The CP content in citrus pulp was low and was not affected by treatments, averaging 7.12% DM and constituting a major limit for its inclusion in rabbit diets, as current CP recommendations in rabbit diets range from 16.7% DM in growing diets to 19.4% DM in breeding animals (De Blas and Mateos, 2010). For dried citrus pulp, the high temperatures (135-155°C) used might explain the higher values of NDICP, ADICP and ADL observed in this group of samples. As demonstrated by Garau *et al.* (2007), temperature during dehydration modifies physico-chemical properties of dietary fibre, as well as the antioxidant effect of citrus by-products. Quantifying the effect of dehydration conditions on citrus pulp composition was outside the scope of this study, as samples were taken from industrial plants with specific conditions. However, technological alternatives considering lower temperatures could influence the chemical composition found in our study.

The DE content (15.1 MJ/kg DM) estimated in the current study from *in vitro* DM digestibility was relatively high. This prediction should be considered as a potential energy value, as it does not take into account the short fermentation time allowed in the rabbit digestion of a balanced diet. In this way, our estimation is close to the 15.9 MJ DE/kg DM determined by Martínez-Pascual and Fernández-Carmona (1980b); in this study, citrus pulp was given as the sole source of feed, which implied a low DM consumption (69.8 g/day), probably because of a high retention time in the gut that led to satiety and a lower intake. However, for diets used in practice with higher levels of long sized particles, the caecal retention time is normally shorter (Gidenne *et al.*, 2008; De Blas, 2013), which ensures a faster rate of passage and thus a higher feed intake and better performance. Consequently, de Blas and Villamide (1990) observed a decrease (from 13.1 to 11.3 MJ/kg DM) in the DE of citrus pulp for rabbits when increasing the ADF level in the basal diet from 13.5 to 24.6% DM. Accordingly, both INRA (2004) and FEDNA (2010) assigned to this ingredient a DE concentration of respectively 12.6 and 12.4 MJ/kg DM, lower than the value predicted in the present work (15.1 MJ/kg DM), but still higher than the recommendations of DE in diets for growing and breeding rabbits (11.3 and 11.9 MJ/kg DM, respectively; de Blas and Mateos, 2010).

Overall, commercial samples of citrus pulps currently available in Spain can be considered a good source of energy and soluble fibre for rabbit diets according to their analytical composition and *in vitro* determinations. However, its

inclusion in rabbit feeds is limited by its low supply of indigestible fibre and crude protein and by the high level of Ca in the case of Ca(OH)₂ processed samples. The main variation factors were the type of fruit (lemon being different from the others) and processing (dried or fresh). The dehydration method, particularly lime addition and dehydration temperature, could affect the composition of dried citrus pulp.

Acknowledgements: This project was funded by the Spanish Ministry of Science and Innovation (AGL2014-56653). Authors are grateful to FEDNA for the grant obtained by Mr. Farias.

REFERENCES

- Abad R., Ibáñez, M.A., Carabaño, R., García, J. 2013. Quantification of soluble fibre in feedstuffs for rabbits and evaluation of the interference between the determinations of soluble fibre and intestinal mucin. *Anim. Feed Sci. Technol.*, 182: 61-70. <https://doi.org/10.1016/j.anifeedsci.2013.04.001>
- Aba, R. 2015. Identification of the method to quantify soluble fibre and the effect of the source of fibre on the ileal and faecal digestibility of soluble and insoluble fibre in rabbits. *PhD Thesis, UP Madrid*.
- Antezana, W., Calvet, S., Beccaccia, A., Ferrer, P., De Blas, C., García-Rebollar, P., Cerisuelo, A. 2015. Effects of nutrition on digestion efficiency and gaseous emissions from slurry in growing pigs: III. Influence of varying the dietary level of calcium soap of palm fatty acids distillate with or without orange pulp supplementation. *Anim. Feed Sci. Technol.*, 209: 128-136. <https://doi.org/10.1016/j.anifeedsci.2015.07.022>
- AOAC. 2000. Official Methods of Analysis, 17th ed. Association of Official Analytical Chemists, Washington, DC.
- Bampidis V.A., Robinson P.H. 2006. Citrus by-products as ruminant feeds: A review. *Anim. Feed Sci. Technol.*, 128: 175-217. <https://doi.org/10.1016/j.anifeedsci.2005.12.002>
- Beccaccia A., Calvet S., Cerisuelo A., Ferrer P., García-Rebollar P., De Blas C. 2015. Effects of nutrition on digestion efficiency and gaseous emissions from slurry in growing pigs: I. Influence of the inclusion of two levels of orange pulp and carob meal in isofibrous diets. *Anim. Feed Sci. Technol.*, 208: 158-169. <https://doi.org/10.1016/j.anifeedsci.2015.07.008>
- Boisen, S. 1991. A model for feed evaluation based on *in vitro* digestibility dry matter and protein. In: Fuller, M.F. (Ed.), *In vitro Digestion for Pigs and Poultry*. CAB International, Wallington, 135-145.
- Braddock R.J. 1999. Handbook of citrus by-products and processing technology. *John Wiley & Sons Inc.*
- CVB. 2016. Veevoedertabel (Livestock Feed Table) Centraal Veevoeder Bureau, Lelystad. The Netherlands.
- De Blas J.C., Villamide M.J. 1990. Nutritive value of beet and citrus pulps for rabbits. *Anim. Feed Sci. Technol.*, 31: 239-246. [https://doi.org/10.1016/0377-8401\(90\)90128-U](https://doi.org/10.1016/0377-8401(90)90128-U)
- De Blas J.C., Wiseman J., Fraga M.J., Villamide M.J. 1992. Prediction of the digestible energy and digestibility of gross energy of feeds for rabbits. 2. Mixed diets. *Anim. Feed Sci. Technol.* 39: 39-59. [https://doi.org/10.1016/0377-8401\(92\)90030-A](https://doi.org/10.1016/0377-8401(92)90030-A)
- De Blas J.C., Mateos G.G. 2010. Feed formulation. In: J.C. de Blas and J. Wiseman (eds.) *The nutrition of the rabbit (2nd ed)*, 222-232. CABI Publishing CAB International, Wallingford, UK. <https://doi.org/10.1079/9781845936693.0222>
- De Blas J.C., Chamorro S., García-Alonso J., García-Rebollar P., García-Ruiz A.L., Gómez-Conde M.S., Menoyo D., Nicodemus N., Romero C., Carabaño R. 2012. Nutritional digestive disturbances in weaner rabbits. *Anim. Feed Sci. Technol.*, 173: 102-110.
- De Blas, J.C. 2013. Nutritional impact on health and performance in intensively reared rabbits. *Animal*, 7: 102-111. <https://doi.org/10.1017/S1751731112000213>
- FAOSTAT. 2017. Food and agriculture data. Food and Agriculture Organization of the United Nations. Available at <http://www.fao.org/faostat/en/>. Accessed May 2017.
- FEDNA. 2010. Tablas Fedna de composición y valor nutritivo de alimentos para la fabricación de piensos compuestos. 3^a ed. C. de Blas, G.G. Mateos, P. García-Rebollar (eds), 502.
- Fraga M.J., Pérez de Ayala P., Carabaño R., de Blas C. 1991. Effect of type of fiber on the rate of passage and on the contribution of soft feces to nutrient intake of finishing rabbits. *J. Anim. Sci.*, 69: 1566-1574. <https://doi.org/10.2527/1991.6941566x>
- Garau M.C., Simal S., Rossello C., Femenia A. 2007. Effect of air-drying temperature on physico-chemical properties of dietary fibre and antioxidant capacity of orange (*Citrus aurantium* v. Canoneta) by-products. *Food chemistry*, 104: 1014-1024. <https://doi.org/10.1016/j.foodchem.2007.01.009>
- García J., Carabaño R., Pérez Alba L., De Blas J.C. 2000. Effect of fiber source on cecal fermentation and nitrogen recycled through cecotrophy in rabbits. *J. Anim. Sci.*, 78: 638-646. <https://doi.org/10.2527/2000.783638x>
- Gidenne T., Carabaño R., García J., de Blas C. 2008. Fibre digestion. In: J.C. de Blas and J. Wiseman (eds.) *The nutrition of the rabbit (2nd ed)*, 66-82. CABI Publishing CAB International, Wallingford, UK. <http://dx.doi.org/10.1079/9781845936693.0066>
- Gidenne T. 2015. Dietary fibres in the nutrition of the growing rabbit and recommendations to preserve digestive health: a review. *Animal*, 9: 227-242. <https://doi.org/10.1017/S1751731114002729>
- Heuzé V., Tran G., Hassoun P., Lebas F. 2017. Citrus pulp, dried. Feedipedia, a programme by INRA, CIRAD, AFZ and FAO, Available at <http://www.feedipedia.org/node/680> Last updated on January 1, 2017, 17:52.
- Licitra, G., Hernández, T.M., Van Soest, P.J. 1996. Standardization of procedures for nitrogen fractionation of ruminant feed. *Anim. Feed Sci. Technol.* 57, 347-358. [https://doi.org/10.1016/0377-8401\(95\)00837-3](https://doi.org/10.1016/0377-8401(95)00837-3)
- MAPAMA. 2017. Anuario de Estadística 2015. Ministerio de Agricultura, Pesca, Alimentación y Medio Ambiente. Available at <http://www.mapama.gob.es/es/estadistica/temas/publicaciones/anuario-de-estadistica>. Accessed May 2017.
- Martínez-Pascual J.L. 1977. Utilización de la pulpa de cítricos en la alimentación de terneros, corderos y conejos. *PhD Thesis, ETSIA, Valencia*.

- Martínez-Pascual, J.L and Fernández-Carmona, J. 1980a. Citrus pulp in diets for fattening rabbits. *Anim. Feed Sci. Technol.*, 5: 23-31. [https://doi.org/10.1016/0377-8401\(80\)90007-3](https://doi.org/10.1016/0377-8401(80)90007-3)
- Martínez-Pascual J.L., Fernández-Carmona J. 1980b. Composition, digestibility, nutritive value and relations among them of several feeds for rabbits. In *Proc.: II Rabbit World Congress, Barcelona, 214-223*.
- Mertens D.R., Allen M., Carmany J., Clegg J., Davidowicz A., Drouches M., Frank K., Gambin D., Garkie M., Gildemeister B., Jeffress D., Jeon C.S., Jones D., Kaplan D., Kim G.N., Kobata S., Main D., Moua X., Paul B., Robertson J., Taysom D., Thiex N., Williams J., Wolf M. 2002. Gravimetric determination of amylase-treated neutral detergent fiber in feeds with refluxing in beakers or crucibles: Collaborative study. *J. AOAC Int.*, 85: 1217-1240.
- Pérez de Ayala P., Fraga M.J., Carabaño R., de Blas J.C. 1991. Effect of fiber source on diet digestibility and growth in fattening rabbits. *J. Appl. Rabbit Res.*, 14: 159-165.
- Ramos M.A., Carabaño R., Boisen S. 1992. An *in vitro* method for estimating digestibility in rabbits. *J. Appl. Rabbit Res.*, 15: 938-946.
- SAS Institute. 2008. SAS/STAT® User's guide, version 9.3. SAS Institute Inc., Cary, NC.
- SIA. 2017. Servicio de Información sobre alimentos. Available at <http://www.uco.es/sia/> Accessed January 2017.
- Trocino A., García J., Carabaño R., Xiccato G. 2013. A meta-analysis on the role of soluble fibre in diets for growing rabbits. *World Rabbit Sci.*, 21: 1-15. <https://doi.org/10.4995/wrs.2013.1285>
- USDA. 2016. Citrus Fruits 2016 Summary. *National Agricultural Statistics Service. United States Department of Agriculture*.
- Van Soest P.J., Robertson J.B., Lewis B.A. 1991. Methods for dietary fibre, neutral detergent fiber, and non starch polysaccharides in relation to animal nutrition. *J. Dairy Sci.*, 74: 3583-3597. [https://doi.org/10.3168/jds.S0022-0302\(91\)78551-2](https://doi.org/10.3168/jds.S0022-0302(91)78551-2)
- Villamide M.J., Carabaño R., Maertens L., Pascual J., Gidenne T., Falcão-E-Cunha L., Xiccato, G. 2009. Prediction of the nutritional value of European compound feeds for rabbits by chemical components and *in vitro* analysis. *Anim. Feed Sci. Technol.*, 150: 283-294. <https://doi.org/10.1016/j.anifeedsci.2008.09.007>
- Wiseman J., Villamide M.J., de Blas J.C., Carabaño R. 1992. Prediction of the digestible energy and digestibility of gross energy of feeds for rabbits. 1. Individual classes of feeds. *Anim. Feed Sci. Technol.*, 39: 27-38. [https://doi.org/10.1016/0377-8401\(92\)90029-6](https://doi.org/10.1016/0377-8401(92)90029-6)
- Yemm E.W., Willis A.U. 1954. The estimation of carbohydrates in plant extracts by anthrone. *Biochem. J.*, 57: 508-514. <https://doi.org/10.1042/bj0570508>
-