

Evaluation of the digestibility of *Lathyrus sativus* in growing pigs

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ABSTRACT

The EU ban on meals of animal origin and the rejection by some States, including Italy, of genetically modified organisms (e.g. soybean), have resulted in the need for novel protein sources. The present study was undertaken to evaluate the digestibility of rations where soybean was partially replaced with *Lathyrus sativus* L. Twelve hybrid growing pigs were fed three diets formulated to be isoenergetic and isoproteic according to a Latin square design, each for 22 days. In the two experimental diets soybean was replaced by 10% or 20% *L. sativus*. Subjects were about 78 kg live weight at baseline. After 10 days of adaptation to each diet, faeces samples were collected from the rectum at the same time of day for 3 days. At the end of the administration of each diet, animals were weighed and blood was collected to determine some protein metabolism parameters. Feed and faeces were analysed for nutrients and AIA in order to calculate the coefficients of digestible utilisation of nutrients. Results were analysed by ANOVA using JMP (SAS system). The growth performance data (ADG and FCR) did not demonstrate differences attributable to the substitution of soybean with *L. sativus*. The coefficients of digestible utilisation indicate that soybean can be replaced with this legume seed. Low plasma urea in animals receiving 20% *L. sativus* confirmed the good metabolic efficiency of protein.

Key words: Pigs, Digestibility, Legumes, Lathyrus sativus.

RIASSUNTO

STIMA DEL VALORE NUTRITIVO DELLA CICERCHIA (LATHYRUS SATIVUS L.) IN RAZIONI PER SUINI

Il reperimento di fonti proteiche ad elevato valore biologico costituisce un aspetto centrale nella formulazione delle razioni soprattutto per gli animali monogastrici. In seguito al divieto dell'uso di farine di origine animale e alla posizione cautelativa che ha portato ad impedire in alcuni paesi, tra cui l'Italia, l'uso in alimentazione zootecnica di semi geneticamente modificati (Direttiva UE 18/2001), scaturisce la necessità di trovare fonti proteiche valide ed alternative alla soia. Sulla base delle motivazioni fin qui addotte con il presente lavoro si è voluta valutare la digeribilità, in suini in accrescimento, di una razione nella cui composizione la soia era stata sostituita dalla cicerchia in ragione del 10 e 20%. A tale scopo sono stati utilizzati 12 suini ibridi alimentati con tre diete isoenergetiche ed isoproteiche somministrate secondo uno schema a quadrato latino. La dieta di Controllo conteneva soia mentre nelle diete 10% e 20% la soia era stata sostituita in ragione del 10 e 20 % con Lathyrus sativus. I suini sono stati pesati all'inizio della prova di digeribilità per costituire gruppi omogenei; dopo un periodo di adattamento alla dieta (10 d), sono stati sottoposti a prelievi di feci, dall'ampolla rettale, per tre giorni consecutivi alla stessa ora. Alla fine di ogni prova si effettuava il controllo del peso e si faceva un prelievo di sangue per la determinazione di alcuni parametri ematici legati al metabolismo proteico. Sul mangime e sulle feci, previamente sottoposte a liofilizzazione, sono state effettuate le analisi per la determinazione dei principi nutritivi e ceneri acido insolubili. Sulla base dei risultati ottenuti è stata quindi calcolata la digeribilità dei principi nutritivi. I dati sono stati sottoposti ad analisi della varianza considerando il quadrato latino. I risultati ottenuti relativi alle performance di allevamento: accrescimento medio giornaliero ed indice di conversione non hanno evidenziato differenze attribuibili alla parziale sostituzione della soia con la cicerchia (AMG: 0,84 g vs 0,83 g vs 0,91 g e ICA: 3,46 vs 3,71 vs 3,13). I coefficienti di utilizzazione digestiva della sostanza organica (87,59 vs 88,70 e 87,98) e della proteina grezza (85,64 vs 85,51 e 84,02) confermano la possibilità di sostituire la soia con questa leguminosa e anche il livello dell'urea ematica nella Dieta 20% (4,7 mmol/l vs 5,7 mmol/l dieta di Controllo) conferma una migliore utilizzazione metabolica della proteina.

Parole chiave: Suini, Digeribilità, Leguminose, Lathyrus sativus.

Introduction

The 2001 ban on meat and bone meals in animal feeds (European Commission, 2001) has created a need for alternative protein sources. Demand for protein rose from 61 million tons in 1990 to 64 million tons in 2000. It is important to find proteinrich crops, especially for monogastric animals. Before the ban, protein was supplied by soybean (60%), colza and sunflower by-products (16%), and meat and fish meals (7%); legume grains accounted for less than 2% of consumption and 6% of all protein feeds (Battini et al., 2003). The EU estimates that the protein deficit due to the ban is about 3 million tons in terms of soybean (Battini et al., 2003), which is the most widely used protein source in swine breeding. This is due to several factors: its 44% protein content, high digestibility (about 90%) and biological value, and a content in essential amino acids that is similar to that of animal proteins. Heat treatment, to which soybean is usually subjected, improves the availability of some amino acids, particularly methionine. The EU

imports 90% of its soybean requirements from the USA and Brazil. Since most soybean is genetically modified (European Commission, 2001), some countries, like Italy, have taken a cautious stance towards these organisms (GMOs) and have banned their use in animal feeds (European Commission, 2001).

Lathyrus sativus has a good protein content, particularly in lysine, an amino acid important in the swine diet; it is low in fat but its starch content supplies abundant energy, which Hanbury et al. (2000) estimated to be 11.30 MJ/kg of Metabolic Energy (Table 1). Like other legumes, L. sativus contains few sulphur amino acids, therefore in mixed diets it is complementary to cereals, which provide high levels of these amino acids. Like other legume seeds, it also contains anti-nutritional factors (ANF) whereas, unlike other legume grains, it contains a non-protein amino acid, ODAP (ß-n-oxalyl-L- α , β -diaminopropanoic acid). Large quantities of L. sativus, e.g. 30% of calorie consumption, for 3-4 months (Campbell, 1994) can cause neurolathyrism. In the human and animal nervous sys-

composition (modified from Martiliotti <i>et al.</i> , 1996)							
Composition		L. sativus	Soybean meal	Composition		L. sativus	Soybean meal
Dry matter	%	91.3	90.0	Starch	% DM	41.2	3.7
Crude protein	% DM	29.4	48.2	Ca	w w	0.16	0.34
Ether extract	" "	1.6	1.2	Р	w w	0.42	0.62
Crude fibre	" "	8.0	7.7	Lysine	g/16 gN	6.8	6.1
NDF	" "	15.6	15.8	Methionine	w w	0.7	1.4
ADF	" "	9.3	10.5	Threonine	w w	3.5	3.9
ADL	" "	1.2	0.8	Cysteine	w w	1.4	1.5
Ash	" "	2.6	6.7				

Table 1.Lathyrus sativus L. (modified from Hanbury *et al.*, 2000) and soybean meal
composition (modified from Martillotti *et al.*, 1996)

tems ODAP works like glutamate, binding to specific receptors and causing permanent damage due to neuronal degeneration and neurotoxic effects. The high concentration of this neurotoxin observed in the early phases of seed maturation eventually decreases; however, heat treatment such as extrusion inactivates ODAP (Harrison, 1997; Akalu et al., 1998). The present study was undertaken to evaluate in growing pigs the digestibility of a ration in which soybean meal was replaced with 10% or 20% *L. sativus*.

Material and methods

Twelve hybrid pigs (Large White x Suffolk), 9 females and 3 males, born and reared in the open air until the time of the study (about 70 kg live weight; LW), were housed in three pens (Box 1, 2 and 3) provided with an outdoor area of about 1.5 m²/head. Average LW was 78.1 kg before the trial and 138 kg at slaughter. The three isoproteic, isoenergetic and isoaminoacidic (for lysine, methionine and threonine) diets devised for the study were a Control Diet (C Diet) and two experimental diets in which extruded L. sativus replaced 10% (10% Diet) or 20% (20% Diet) soybean (see Tables 2 and 3). The diets were administered consecutively according to a Latin square design. The daily ration was calculated as 8% of LW^{0.75} (Ricci Bitti et al., 1975). Pigs were weighed on an Omega electronic scale before and after

receiving each diet (given for 22 days) after 12 hours of fasting. The rations were weighed daily and administered twice a day. After 10 days of adaptation to each diet, faeces samples were collected from the rectum of each subject at the same time of day on days 10, 11, and 12, and then on days 20, 21 and 22. The faeces of three days from each subject were stored at 4° C and finally pooled, thus obtaining 72 samples. They were weighed, frozen, lyophilised and placed in a stove at 65°C to remove all water, then weighed again to calculate the proportion of dry matter (DM). After grinding, ration and faeces samples were analysed using the ASPA method to determine chemical composition and the acid insoluble ash method (Martillotti et al., 1987) to determine digestibility. For all nutrients, we calculated the coefficient of digestibility (DUC) (Borgioli, 1991) and the digestible coefficient of energy (DUC E) according to Noblet et al. (1989) (DUC $E_1 = 97.5 - 0.116$ *NDF), Perez *et al.* (1984) (DUC E₂ = 93.8 - 0.128*NDF - 0.064*(NDF-ADF) as well as using acid insoluble ash (DUC E_{AIA}). Average daily gain (ADG) and the feed conversion rate (FCR) / box were calculated based on pig weight. Upon completion of each diet, blood from the jugular vein was collected in test tubes containing Li-heparin. Plasma obtained by centrifugation was stored at -20°C for the determination of some parameters of protein metabolism: total proteins (using the biuret method modified by Henry), urea (using the enzymatic method) and

Table 2. Composition	Composition of diets (%)					
	C Diet	10% Diet	20% Diet			
Corn	36.0	38.5	40.0			
Barley	30.5	22.0	15.0			
Soybean meal	25.0	20.0	15.5			
Bran	5.0	5.0	5.0			
L. sativus		10	20			
Animal fat	0.5	1.5	1.5			
CaCO3	1.1	1.1	1.1			
$Ca(H_2PO_4)$	1.0	1.0	1.0			
NaCL	0.4	0.4	0.4			
Vit-min supplement	0.5	0.5	0.5			

Table 3.	Chemical com	npositio	on of diets		
			C Diet	10% Diet	20% Diet
Dry matter	%		86.6	86.0	86.2
Crude protein	% D	М	17.0	17.0	17.1
Ether extract	w	w	2.70	2.60	2.60
Crude fibre	w	w	4.8	4.7	4.6
NDF	w	w	14.5	14.0	13.6
ADF	w	w	5.0	4.9	4.9
Ash	w	"	7.0	6.7	6.5
Ca	w	w	0.7	0.7	0.7
Р	w	w	0.6	0.6	0.6
Lysine*	w	w	0.90	0.91	0.91
Methionine*	w	w	0.28	0.26	0.25
Threonine*	w	w	0.66	0.65	0.65
DE*	MJ/kg	DM	13.29	13.59	13.71

* estimated value

albumin (using the bromocresol purple dye-binding method) were determined with Du Pont's Photometric Auto Analyser SMS to evaluate the diet's influence. Data were analysed by ANOVA using the GLM procedure (JMP, SAS system)

where:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \gamma_k + \varepsilon_{ijk}$$

 Y_{iik} = dependent variable μ = general mean α_i = diet effect (i = 1, 3) $\beta_i = \text{box effect } (j = 1, 3)$ γ_{k} = period effect within each row (k = 1, 3) ε_{ijk} = residual error

The differences were analysed using a post hoc test.

Results and discussion

The growth performance over the whole experimental period is reported in Table 4. Statistical analysis did not demonstrate significant differences due to the diet. The ADG was highest and the FCR best in pigs receiving the 20% Diet. The ADG obtained in this study was overall higher than the one reported by Trombetta et al. (2001) in a trial where soybean had partially been replaced with corn germ meal, the one described by Martelli et al. (2000) for pigs fed beet-pulp, and the trial of Castaing and Noblet (1997) with growing pigs fed sepiolite-containing diets. The high ADG obtained with all three diets may be ascribed to the change in the rearing system (from open air to box), which may have made up for weight not gained in the open air. A box effect was not found (0.872 vs 0.867 vs 0.831, respectively, for Box 1, 2, and 3).

The coefficients of digestibility of organic matter (DOM), crude protein (DCP) and extract ether (DEE) are reported in Table 5 according to the diet effect. The DOM was similar to the total digestibility coefficient of OM reported by van der Poel et al. (1997) for processed diets. The statistical analysis demonstrated significant differences for DCP (P< 0.047) and DEE (P< 0.003). The DCP was similar in the Control and the 10% Diet (85.64% and 85.51%, respectively) and was lower in the 20% Diet (84.02%). It was similar to the one reported by Noblet et al. (1998), and was in between the experimental and control values obtained in the same genetic type by Trombetta et al. (2001) in a

Table 4.	Grow	th performance			
		C Diet	10% Diet	20% Diet	Р
ADG	kg/d	0.84	0.83	0.91	0.523
FCR		3.46	3.71	3.13	0.158

* estimated value

Table 5. Coefficient of digestible utilisation (DUC) of some nutritional parameters							
		C Diet	10% Diet	20% Diet	SE	Р	
Samples DUC (%) of:	n.	36	36	36			
Organic matter	r	87.59	88.70	87.98	0.344	0.086	
Crude protein		85.64	85.51	84.02	0.489	0.047	
Ether extract		75.98	76.37	72.38	0.835	0.003	
Crude fibre		55.55	59.95	60.54	1.639	0.079	
NDF		80.05	80.77	80.82	0.530	0.522	
ADF		67.61	70.50	77.63	0.775	0.000	
Hemicellulose		84.79	84.94	82.75	0.641	0.039	

previous trial. The DEE exhibited a trend similar to that of protein, although the animals receiving the 20% Diet showed a larger decrease. The values measured with the these diets were lower than those of Trombetta *et al.* (2001), but better than those reported by Noblet *et al.* (1998) for Pietrain x Large White pigs receiving a feed containing *Lupinus angustifolius.* Statistical analysis for the box effect showed significant differences in DOM, DCP and DEE (P< 0.001) for Box 2.

The analysis of DUC of crude fibre (DCF), NDF (DNDF) and ADF (DADF) (Table 5) highlighted significant differences due to the diet for DADF (P< 0.000) and Hemicellulose (P< 0.039), while for DCF and DNDF it was not significant. The coefficients of digestibility of ADF were higher in pigs receiving 20% *L. sativus*. The digestibility data for CF were 10-15% higher than those reported by Noblet *et al.* (1998) and similar to those obtained in pigs receiving corn germ meal (Trombetta *et al.*, 2001) and in sows fed maize fibre (Le Goff and Noblet, 2001). The higher values of DCF and

DADF for diets 10% and 20% could be ascribed to the extrusion treatment applied to inhibit ANF activity, which may have broken complex chemical links, improving fibre compound digestibility. The DCF was different from the CF coefficient reported by van der Poel et al. (1997), who found that feed processing reduced CF digestibility. Also for these parameters, Box 2 had significantly better values (P< 0.01) than the other two boxes.

The estimated value of the digestible coefficients of energy calculated according to Noblet *et al.* (1989) (DUC $E_1 = 97.5 - 0.116*NDF$), Perez *et al.* (1984) (DUC $E_2 = 93.8 - 0.128*NDF - 0.064*(NDF-ADF)$ and using acid insoluble ash (DUC E_{AIA}) are reported in Table 6. The use of different equations to calculate the energy digestibility yielded different values for the same diet; in particular, the energy DUCs obtained according to Perez *et al.* and the AIA method are similar, though lower, than those obtained according to Noblet *et al.*. The DUC E_{AIA} was significantly lower (P< 0.019) in Diet C. Statistical analysis gave bet-

Table 6.	Coefficient of digestible utilisation of energy (DUC E)						
		C Diet	10% Diet	20% Diet	SE	Р	
Samples	n.	36	36	36			
DUC E1	%	93.3	93.6	93.3			
DUC E ₂	"	90.8	91.0	90.7			
DUC E _{AIA}	n	90.88	91.94	91.63	0.254	0.019	

E₁: according to Noblet et al. (1989); E₂: according to Perez et al. (1984); E_{AIA}: using AIA data.

Table 7	Blood parameters of protein profile							
	blood parameter							
Items		C Diet	10% Diet	20% Diet	Р			
Samples	n.	12	12	12				
Total Protein	g/l	59.0	59.0	61.0	0.7349			
Urea	mmol/l	5.7	5.0	4.7	0.1264			
Albumin	g/l	34.0	34.0	35.0	0.7956			

ter values for Boxes 2 and 3 than for Box 1 (P< 0.01). The better DNDF and DADF of diets 10 and 20% and Boxes 2 and 3 explain the improvement in the energy coefficient of digestibility, in line with the observation Le Goff and Noblet (2001).

The protein profile parameters are reported in Table 7. The statistical analysis did not highlight significant differences related to the diet or the box effect. However, plasma urea was lower in pigs receiving the experimental diets, especially the 20% diet (4.7 mmol/l), suggesting a better use of *L. sativus* and confirming previous work by Eggum (1970), Malmlöf *et al.* (1988), Falaschini *et al.* (1994), Trombetta and Falaschini (2001), and Trombetta (2004). Total protein and albumin showed slightly higher values in pigs receiving the 20% Diet.

Conclusions

The fully-grown pig has a greater ability than younger animals to utilise dietary fibre thanks to more intense hindgut microbial activity. Diet quality is therefore due to an interaction among chemical composition, feed processing, age and gender. The growth performance data and the coefficients of digestible utilisation of the different components obtained in the present study suggest that soybean in the swine diet can be replaced with legume grains such as *L. sativus*. Moreover, the low urea levels measured in pigs receiving the 20% Diet confirms a good protein metabolic efficiency also in diets containing feeds alternative to soybean meal. These results need to be confirmed, and further research is warranted to test other alternative protein sources to be used in swine feed formulation.

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