



## The nutritional value of narrow-leaved lupin (*Lupinus angustifolius*) for broilers

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**ABSTRACT.** The nutritional value of four cultivars of *Lupinus angustifolius* L. seeds was investigated. The experiment was conducted on 50 male 16-day-old Ross 308 chickens. The birds were randomly allocated to five dietary treatments (10 replications each). Digestibility was calculated using the difference method. The basal diet and four diets composed of ground lupin seeds (cv. Sonet, Boruta, Graf or Neptun) and the basal diet at a proportion of 25:75 (w/w) were prepared. The birds were fed the diets starting from day 16; on days 19 and 20 of life, excreta were individually collected, then all chickens were sacrificed and ileal digesta was sampled for determination of ileal digestibility. Ileal dry matter digestibility ranged between 56.8% and 62.5% ( $P < 0.05$ ). The apparent ileal digestibility of aspartic acid, proline, alanine, valine, histidine and lysine was negatively correlated ( $P < 0.05$ ) with the *in vitro* water extract viscosity (WEV) of lupin seeds. The apparent metabolizable energy (AME<sub>N</sub>) ranged from 7.91 to 9.27 MJ · kg<sup>-1</sup>, the lowest was determined for cv. Graf, the highest for Boruta and Sonet. The Graf cultivar was also characterized by the lowest dry matter ileal and total tract digestibility ( $P < 0.05$ ). The AME<sub>N</sub> of lupin seeds was negatively correlated with WEV ( $r = -0.67$ ;  $P < 0.05$ ) and raffinose level ( $r = -0.72$ ;  $P < 0.05$ ). The relationship between raffinose content and AME<sub>N</sub> was linear ( $P < 0.05$ ).

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### Introduction

Lupines are legume crops quite common all over the world; the annual production of these legumes reached 1.2 mln tons in 2011 (FAOSTAT, 2013). According to earlier research, lupin protein is utilized to the same degree as soyabean meal protein (Steenfeldt et al., 2003; Mieczkowska et al., 2005). In the past, the use of narrow-leaved lupin (*Lupinus angustifolius*) as a valuable source of protein for poultry was limited due to high alkaloid and

non-starch polysaccharide (NSP) contents that negatively affect growth, feed intake and nutrient utilization (Olver and Jonker, 1997; Hughes et al., 2000). During recent decades, however, plant breeders have succeeded in developing lupin cultivars with very low alkaloid contents (Jezierny et al., 2010; COBORU, 2011), which improved their nutritional value for monogastric animals. On the other hand, whole lupin seeds still contain approximately 35% NSP, of which over 5% are water soluble, limiting their use in poultry feeding (Choct and Hughes, 1999).

It is well known that nutrient composition and antinutritional factors in narrow-leaved lupines generally depend on the cultivar and growing conditions. Published data on nutrient and amino acid utilization and apparent metabolizable energy (AME) of narrow-leaved lupines for broilers are limited and inconsistent. Nalle et al. (2011) reported that narrow-leaved lupines could be a good source of most amino acids, but a poor source of sulphur-containing amino acids and AME. The above authors showed that narrow-leaved lupin AME ranged from 6.38 to 7.12 MJ · kg<sup>-1</sup> dry matter. According to Smulikowska and Rutkowski (2005), lupin-seed AME can reach 7.22 MJ · kg<sup>-1</sup> for broilers. On the other hand, Choct and Hughes (1999) found that the AME of narrow-leaved lupines ranged from 8.0 to 12.3 MJ · kg<sup>-1</sup>. In contrast to Nalle et al. (2011), Choct and Hughes (1999) observed differences in AME between cultivars.

The aim of this study was to explain the causes of variations in nutrient digestibility and metabolizable energy (AME<sub>N</sub>) content of Polish cultivars of narrow-leaved lupines for broiler chickens.

## Material and methods

### Lupin seeds

Lupin (*Lupinus angustifolius* L.) seeds of four cultivars, Sonet, Boruta, Graf and Neptun, registered in 1999, 2002, 2004 and 2009, respectively (COBORU, 2011), were harvested in 2011. Seeds were obtained from the Plant Breeding Stations in Przebędowo and Wiatrowo (Poland).

### Diets

The basal diet was prepared as shown in Table 1 and was mixed in a proportion of 75:25 (w/w) with corresponding lupin meals. To allow digestibility to be determined, 3 g · kg<sup>-1</sup> titanium dioxide was included as a non-absorbable marker. All diets were offered in mash form *ad libitum*.

### Animals and sample collection

The experiment complied with the guidelines of the Local Ethics Commission with respect to animal experimentation and care of animals under study.

The experiment was conducted on 50 male 16-day-old Ross 308 chickens. The birds were randomly allocated to five dietary treatments (10 replications each) and kept in individual cages. In four treatments, diets containing a lupin meal (cv. Sonet, Boruta, Graf or Neptun) and the basal diet at a proportion of 25:75 were fed. The birds in one treatment were fed only the basal diet.

**Table 1.** Basal diet composition and nutritive value

Indices	g · kg <sup>-1</sup>
<b>Ingredients</b>	
maize (9% CP)	600.0
soyabean meal (45% CP)	292.68
soya oil	41.59
fish meal (65% CP)	29.35
monocalcium phosphate	10.31
limestone	5.06
Premix <sup>1</sup>	10
TiO <sub>2</sub>	3
NaCl	2.89
NaHCO <sub>3</sub>	0.13
Met	2.7
Lys	1.7
Thr	0.59
<b>Calculated</b>	
ME, MJ · kg <sup>-1</sup>	12.56
<b>Analysed, g · kg<sup>-1</sup>.<sup>2</sup></b>	
GE, MJ · kg <sup>-1</sup>	17.64
crude protein	21.2
ether extract	7.18
Ca	0.92

<sup>1</sup> provides per kg diet: IU: vit. A 11250, cholecalciferol 2500; mg: vit. E 80, menadione 2.50, vit. B<sub>12</sub> 0.02, folic acid 1.17, choline 379, D-pantothenic acid 12.5, riboflavin 7.0, niacin 41.67, thiamin 2.17, D-biotin 0.18, pyridoxine 4.0, ethoxyquin 0.09, Mn 73, Zn 55, Fe 45, Cu 20, I 0.62, Se 0.3, salinomycin 60; <sup>2</sup> n=10

Each bird had unlimited access to water and feed. The experimental diets were fed for a five-day adaptation period. On days 19 and 20, excreta were individually collected twice per day and immediately frozen for future analysis (n = 10). On day 21 of the experiment, all chickens from each group were sacrificed by cervical dislocation and the ileum was removed. Digesta were flushed from the terminal ileum (15 cm adjacent to the ileo-caecal junction) and pooled (2 birds/sample) to provide sufficient material for chemical analysis (n = 5).

### Chemical analysis

For chemical analysis, representative samples of seeds were ground to pass through a 0.5 mm sieve. Seeds were analysed (n = 4) for dry matter (DM), crude protein (CP), ether extract (EE), acid detergent fibre (ADF) and neutral detergent fibre (NDF) using methods 934.01, 976.05, 920.39, 942.05 and 973.18, respectively, according to AOAC (2007).

The amino acid (AA) content was determined with an AAA-400 Automatic Amino Acid Analyser using ninhydrin for post-column derivatization. Before analysis the samples were hydrolysed with 6 N HCl for 24 h at 110°C (procedure 994.12; AOAC, 2007). Gross energy (GE) was determined using an adiabatic bomb calorimeter (KL 12Mn,

Precyzja-Bit PPHU, Poland) standardized with benzoic acid. Titanium dioxide was determined according to Short et al. (1996), the samples were prepared in accordance with the procedure proposed by Myers et al. (2004).

Lupin alkaloids were extracted from the meal with trichloroacetic acid and methylene chloride and determined by gas chromatography (Shimadzu GC17A) using a capillary column (Phenomenex). Raffinose family oligosaccharides (RFO) were extracted and analysed by high-resolution gas chromatography as described previously by Zalewski et al. (2001). The water extract viscosity (WEV) of lupin seeds was measured *in vitro*. Prior to the determination of WEV, lupin samples were ground in a mill using a sieve with 0.5 mm mesh and then 1 g of each of the examined cultivars was mixed with 5 ml distilled water for 1 h at 40°C. The samples were centrifuged at 10 000 g for 10 min at 4°C, the supernatant was withdrawn and viscosity was determined in a Brookfield Digital DV-II+ cone/plate viscometer (Brookfield Engineering Laboratories Inc., Stoughton, MA, USA) maintained at 40°C at a shear rate of 12 · s<sup>-1</sup>. WEV units are mPas·s (mPas·s = cP = 1 × 100 dyne s cm<sup>-2</sup>).

### Calculations and statistical analysis

Using calculation of crude protein as an example, the following equation was used to calculate the apparent ileal digestibility (AID) and apparent total tract digestibility (ATTD) of various dietary components of the basal and experimental diets:

$$DC (\%) = \left\{ 1 - \left[ \frac{TiO_{2\% \text{ digesta/excreta}}}{TiO_{2\% \text{ diet}}} \right] \times \left( \frac{CP_{\% \text{ digesta/excreta}}}{CP_{\% \text{ diet}}} \right) \right\}$$

The following equation was used to calculate the digestibility coefficients of various dietary components and the AME<sub>N</sub> level of lupin seeds:

$$DC_{CP} = \frac{(DC_{CP \text{ diet}} \times C_{CP \text{ diet}} - DC_{CP \text{ basal}} \times C_{CP \text{ basal}})}{(C_{CP \text{ diet}} - C_{CP \text{ basal}}) \times 0.25}$$

where: DC<sub>CP</sub> – digestibility coefficient of CP; DC<sub>diet</sub> – digestibility coefficient of CP in a diet; C<sub>diet</sub> – concentration of CP in a diet; DC<sub>basal</sub> – digestibility coefficient of CP in the basal diet; C<sub>basal</sub> – concentration of CP in the basal diet.

The AME<sub>N</sub> of basal and experimental diets was calculated using the above equations and was corrected to zero nitrogen balance using 34.4 kJ · g<sup>-1</sup> N retained (Hill and Anderson, 1958).

All data were explored earlier to discard any possible outliers. The analyses were performed using the appropriate procedures of SAS Software (distribution analyses; outliers were defined as observa-

tions whose distance to the location estimate exceeds 3 times the standard deviation). The obtained results were subjected to one-factorial analysis of variance (ANOVA). Means from experiments were compared using the Duncan test and differences were assumed to be significant at the level of *P* < 0.05.

The correlation between lupin seed composition and nutritional value was assessed using Person's correlation calculated on the basis of the following formula:

$$r_{xy} = \text{cov}(x,y) / S_x S_y$$

where: *r*<sub>xy</sub> – Person's correlation coefficient, cov(*x*,*y*) – co-variance (covariation between *x* and *y*), *S*<sub>*x*</sub>, *S*<sub>*y*</sub> – standard deviations of the *x* and *y* cultivar.

To predict the AME<sub>N</sub> of lupin seeds from the raffinose content, linear regression analysis was performed according to the following formula (raffinose content as an example):

$$y_i = \text{min}_i + m_i \times g$$

where: *y*<sub>*i*</sub> – the response variable, *min*<sub>*i*</sub> – the intercept, *m*<sub>*i*</sub> – the slope, *g* – of the raffinose intercept is a theoretical value that does not necessarily coincide with the zero raffinose content in seeds.

In the performed experiment, standard error of the mean (SEM) was adopted as a measure of error.

## Results

The chemical composition of lupin seeds is presented in Table 2. Narrow-leaved lupin cv. Boruta contained more crude protein than other cultivars, the ether extract content was the highest in cv. Neptun. The cv. Graf seeds were characterized by the highest NDF and ADF contents. The total oligosaccharide content ranged between 79.9 and 97.4 g · kg<sup>-1</sup> and

**Table 2.** Chemical composition of narrow-leaved lupin cultivars (n=4)

Contents	Units	Cultivars			
		Boruta	Neptun	Sonet	Graf
Crude protein		36.0	34.0	31.0	35.0
Ether extract		5.8	6.2	5.7	5.4
NDF	g · 100 g <sup>-1</sup>	25.9	23.4	25.4	30.4
ADF		21.4	20.6	22.8	26.4
Total oligosaccharides		87.7	97.4	79.9	88.0
Raffinose	g · kg <sup>-1</sup>	12.0	13.7	11.2	14.5
Stachyose		56.0	64.0	52.0	57.0
Verbaskose		20	19.1	16.0	17.0
Total alkaloids		44.0	13.0	39.0	4.0
Angustifoline		5.52	1.35	4.18	0.369
Izolupanine	mg · kg <sup>-1</sup>	2.01	0.766	1.81	0.256
Lupanine		24.7	7.30	24.3	1.80
130H lupanine		11.8	3.59	8.68	1.57
WEV	mPas·s	1.21	1.65	1.40	1.81

WEV – water extract viscosity

**Table 3.** Apparent ileal digestibility of dry matter, crude protein and amino acids in four cultivars of narrow-leaved lupins for broilers

Nutrient, %	Cultivar				SEM	P
	Boruta	Neptun	Sonet	Graf		
Dry mater <sup>1</sup>	62.5 <sup>a</sup>	61.7 <sup>a</sup>	61.5 <sup>a</sup>	56.8 <sup>b</sup>	0.72	0.011
Crude protein	81.7	82.3	80.3	79.4	0.58	0.287
Asp	82.4 <sup>a</sup>	80.1 <sup>ab</sup>	76.5 <sup>bc</sup>	73.3 <sup>c</sup>	1.13	0.012
Thr	78.8 <sup>a</sup>	78.5 <sup>a</sup>	75.1 <sup>ab</sup>	71.9 <sup>b</sup>	0.98	0.028
Ser	80.4 <sup>a</sup>	79.5 <sup>a</sup>	73.7 <sup>b</sup>	72.5 <sup>b</sup>	1.04	0.004
Glu	89.8 <sup>a</sup>	91.2 <sup>a</sup>	83.5 <sup>b</sup>	83.6 <sup>b</sup>	0.97	<0.001
Pro	88.0 <sup>a</sup>	81.0 <sup>b</sup>	76.7 <sup>bc</sup>	74.7 <sup>c</sup>	1.35	<0.001
Gly	78.4 <sup>a</sup>	77.3 <sup>a</sup>	74.1 <sup>ab</sup>	71.3 <sup>b</sup>	1.02	0.047
Ala	84.0 <sup>a</sup>	82.7 <sup>a</sup>	79.9 <sup>ab</sup>	75.6 <sup>b</sup>	1.08	0.019
Val	83.6 <sup>a</sup>	81.9 <sup>a</sup>	78.6 <sup>ab</sup>	75.0 <sup>b</sup>	1.15	0.028
Ile	86.2	85.3	81.4	78.5	1.17	0.056
Leu	87.8 <sup>a</sup>	87.2 <sup>a</sup>	83.0 <sup>ab</sup>	79.4 <sup>b</sup>	1.22	0.037
Tyr	80.5 <sup>a</sup>	80.7 <sup>a</sup>	75.6 <sup>ab</sup>	74.6 <sup>b</sup>	0.94	0.025
Phe	86.1	85.4	81.7	78.8	1.09	0.051
His	83.5 <sup>a</sup>	81.3 <sup>a</sup>	78.3 <sup>a</sup>	62.6 <sup>b</sup>	1.95	<0.001
Lys	87.0 <sup>a</sup>	86.8 <sup>a</sup>	83.3 <sup>a</sup>	72.0 <sup>b</sup>	1.48	<0.001
Arg	85.9 <sup>ab</sup>	89.4 <sup>a</sup>	81.2 <sup>bc</sup>	79.1 <sup>c</sup>	1.24	0.010

<sup>1</sup>n=5, <sup>abc</sup> means with different superscripts within a row are significantly different at  $P \leq 0.05$ , respectively; SEM – standard error of the mean

the dominating oligosaccharide was stachyose (on average, over 65% total oligosaccharides). The WEV of seeds was different across cultivars. The highest viscosity was determined for cv. Graf, the lowest, for cv. Boruta. The alkaloid level was low and ranged from 4 (cv. Neptun) to 44 mg · kg<sup>-1</sup> (cv. Boruta). Lupanine was the dominating alkaloid in all seeds and ranged from 24.7 mg · kg<sup>-1</sup> in cv. Boruta to 1.8 mg · kg<sup>-1</sup> in cv. Graf.

Ileal digestibility results are presented in Table 3. There were differences in DM digestibility among cultivars at the ileum level ( $P < 0.05$ ) ranging between 56.8% and 62.5%. The ileal digestibility of amino acids, except isoleucine and phenylalanine, depended on the cultivar ( $P < 0.05$ ). The apparent digestibility of aspartic acid, proline, alanine, valine, histidine and lysine was negatively correlated with WEV ( $P < 0.05$ ).

**Table 4.** Total tract apparent crude fat digestibility, dry mater retention, AME<sub>N</sub> value and metabolizability of GE of narrow-leaved lupin seeds

Indices	Cultivar				SEM	P
	Boruta	Neptun	Sonet	Graf		
AME <sub>N</sub> , MJ · kg <sup>-1</sup>	9.27 <sup>a</sup>	8.67 <sup>b</sup>	9.16 <sup>a</sup>	7.91 <sup>c</sup>	0.101	<0.001
AME <sub>N</sub> /GE	48.6 <sup>ab</sup>	51.0 <sup>a</sup>	46.8 <sup>b</sup>	41.8 <sup>c</sup>	0.76	<0.001
Crude fat	74.5 <sup>a</sup>	74.0 <sup>b</sup>	80.4 <sup>a</sup>	69.6 <sup>c</sup>	0.86	<0.001
Dry mater	60.3 <sup>a</sup>	59.0 <sup>a</sup>	57.3 <sup>a</sup>	49.4 <sup>b</sup>	0.90	<0.001

<sup>1</sup>n=10; <sup>abc</sup> means with different superscripts within a row are significantly different at  $P \leq 0.05$ , respectively; SEM – standard error of the mean

Total tract crude fat digestibility varied among cultivars (Table 4). Graf was characterized by the lowest AME<sub>N</sub> level. The average metabolizable energy for all cultivars was 8.76 MJ · kg<sup>-1</sup> of seeds, the highest was determined for cv. Boruta and Sonet, and the lowest, for cv. Graf. The AME<sub>N</sub> of lupin seeds was negatively correlated with WEV ( $r = -0.67$ ;  $P < 0.05$ ) and raffinose content ( $r = -0.72$ ;  $P < 0.05$ ). The relationship between raffinose content and AME<sub>N</sub> was linear ( $P < 0.05$ ; Table 5). The obtained  $r^2$  for AME<sub>N</sub> explained over 51% of variability ( $r^2 = 0.51$ ; Table 6). The highest metabolizability of gross energy (GE) was in cv. Neptun.

**Table 5.** Pearson's correlation coefficients between lupin seed traits and digestibility and AME<sub>N</sub> for broiler chickens (AID, n=24; AME<sub>N</sub> and ATTD; n=48)

Lupin traits	Response	Correlation coefficient (r)	P
Raffinose content	EE ATTD	-0.59	<0.001
	DM ATTD	-0.45	0.001
	DM AID	-0.48	0.019
Oligosaccharides content	EE ATTD	-0.30	0.036
WEV	AMEN	-0.67	<0.001
	EE ATTD	-0.36	0.013
	DM ATTD	-0.46	0.001
	DM AID	-0.52	0.009
	AID of selected amino acids		
	Asp	-0.46	0.024
	Pro	-0.59	0.002
	Ala	-0.45	0.028
	Val	-0.42	0.040
	His	-0.67	<0.001
	Lys	-0.61	0.002

AID – apparent ileal digestibility; ATTD – apparent total tract digestibility; WEV – water extract viscosity; EE – ether extract; DM – dry matter

**Table 6.** Regression parameters for the equation  $y = a + bx$  estimating AME<sub>N</sub> (MJ · kg<sup>-1</sup>), (y) from raffinose content in narrow-leaved lupin (x)

	Intercept (a)	Slope (b)	r <sup>2</sup>	Significance
AME <sub>N</sub>	13.89	-3.982	0.5119	<0.0001

## Discussion

During recent decades, modifications of lupin genotypes were focused on the improvement of agronomical traits, as well as on lowering the alkaloid content and increasing CP levels. The investigated seed nutrients were within the range determined previously for the older Polish cultivars of narrow-leaved lupines (Gdala and Buraczewska, 1996; Sujak et al., 2006). In comparison with the results of Alloui et al. (1994), the CP, EE, NDF and ADF contents in Polish narrow-leaved lupin seeds

were also similar to the results from our laboratory. On the other hand, Australian cultivars (Nalle et al., 2011) were characterized by lower CP and similar EE contents in comparison with our results. Nonetheless, the presented results show that the total alkaloid content decreased at the turn of the 20<sup>th</sup> and 21<sup>st</sup> centuries. Older Polish cultivars of narrow-leaved lupin were characterized by at least ten-fold higher alkaloid contents (Alloui et al., 1994; Gdala and Buraczewska, 1996) than the ones discussed in this study. The total oligosaccharide content determined in this trial was higher than that measured by Gdala and Buraczewska (1996) and the difference was over  $10 \text{ g} \cdot \text{kg}^{-1} \text{ DM}$ . According to earlier research (Górecki et al., 1997; Lahuta, 2011), the content of oligosaccharides in lupin seeds increases under drought conditions, so the observed differences could be a result of different environmental conditions during growth.

The  $\text{AME}_N$  of protein feeds is crucial in the process of diet formulation. A low  $\text{AME}_N$  of the protein source necessitates adding large amounts of oil, thus increasing the final price of a feed mixture. The investigated narrow-leaved lupin seeds differed with respect to  $\text{AME}_N$  levels (from  $7.91$  to  $9.27 \text{ MJ} \cdot \text{kg}^{-1}$ ) in contrast to the results of Nalle et al. (2011), who investigated three Australian cultivars and obtained  $\text{AME}_N$  levels ranging from  $5.35$  to  $6.18 \text{ MJ} \cdot \text{kg}^{-1}$ . Alloui et al. (1994) reported that the  $\text{AME}_N$  values of Polish narrow-leaved lupines (cv. Emir and Sur) ranged from  $8.4$  to  $8.8 \text{ MJ} \cdot \text{kg}^{-1} \text{ DM}$ . Choct and Hughes (1999) found that  $\text{AME}$  values of Australian narrow-leaved lupin cotyledons could reach even  $10.8 \text{ MJ} \cdot \text{kg}^{-1}$  after hull removal. Recently, Roura et al. (2013) showed that chickens have well-developed bitter taste receptors, which may affect voluntary intake of bitter raw materials and, ultimately, depress  $\text{AME}$ . The narrow-leaved lupin seeds used in the present study were characterized by very low alkaloid contents and could be classified as sweet lupine for that reason; the  $\text{AME}_N$  value and the digestibility of seed nutrients did not correlate with the alkaloid content. Narrow-leaved lupines contain NSP that can lower nutrient digestibility,  $\text{AME}_N$ , and performance (Jezierny et al., 2010). It is also known that the concentration of antinutritional factors in narrow-leaved lupines varies depending on the cultivar and growing season (Choct and Hughes, 1999). In this experiment, the WEV of seeds was measured *in vitro* (Table 2) and ranged from  $1.21$  to  $1.81 \text{ mP} \cdot \text{s}$ . It is possible that the variations in  $\text{AME}_N$  content between cultivars were caused by high digesta viscosity, but this parameter was not measured. It is believed that NSP in lupines affects feed intake and

digestibility (Carré et al., 1985). The mechanism leading to the changes in the digestive tract is unclear, but it has been suggested that these negative effects of NSP may be attributed to their viscous nature and complicated interactions with digestive tract microbes (Smits and Annison, 1996). Since most modern cultivars of *L. angustifolius* contain low concentrations of alkaloids, the main problem in using lupines seems to be the level of NSP. On the other hand, narrow-leaved lupin seeds contain less than 4% soluble NSP and 46% insoluble NSP, according to Smits and Annison (1996), Nalle et al. (2011), and/or even 15% of soluble NSP, according to Bach Knudsen (1997). According to Gdala and Buraczewska (1996), the total NSP content for Polish cultivars was 40%. This high inherent variation in soluble NSP content in *L. angustifolius* seeds may partially explain differences in the  $\text{AME}_N$  content across different samples. In our experiment, the  $\text{AME}_N$  of seeds correlated well ( $r = 0.67$ ) with the WEV value ( $P < 0.001$ ). Seeds with the highest WEV were also characterized by the lowest  $\text{AME}_N$ . At the same time, WEV influenced ether extract digestibility, which decreased as WEV increased ( $r = 0.36$ ). According to earlier studies, soluble NSP could affect metabolizable energy due to depression of ether extract digestibility. Soluble NSP may increase bacterial activity through digesta flow reduction and by providing extra nutrients for fermentation. As indicated by Smits and Annison (1996), increased bacterial activity causes an increase in bile salt deconjugation, resulting in poor ether extract digestion.

Legumes are plants rich in  $\alpha$ -galactosides (GAL): raffinose, stachyose and verbascose. The GAL account for approximately  $60\text{--}70 \text{ g} \cdot \text{kg}^{-1}$  of DM (Gdala and Buraczewska, 1996; Bach Knudsen, 1997). In our study, the average GAL content was over  $88 \text{ g} \cdot \text{kg}^{-1}$ , which is in agreement with Bach Knudsen (1997). These low molecular weight sugars are not hydrolysed because poultry lack endogenous  $\alpha$ -1,6 galactosidase activity. Previous research demonstrated that stachyose and raffinose contents affect the  $\text{AME}_N$  of soya protein concentrate and soyabean meal (SBM) (Coon et al., 1990; Leske et al., 1993). When SBM low in oligosaccharides was used in broiler chickens (Leske et al., 1991, 1993; Baker et al., 2011; Chen et al., 2013; Perryman et al., 2013), or SBM isolate in turkeys (Jankowski et al., 2009), better nutrient utilization and growth performance were observed. In agreement with the above results, the  $\text{AME}_N$  of the investigated *L. angustifolius* seeds depended on the raffinose level ( $r^2 = 0.512$ ;  $P < 0.05$ ). Lupin seeds having the highest raffinose content ( $14.5 \text{ g} \cdot \text{kg}^{-1}$ )

were also characterized by the lowest AME<sub>N</sub> level (7.91 MJ · kg<sup>-1</sup>,  $r = -0.72$ ). As suggested by Leske et al. (1991), high concentrations of these oligosaccharides in the digestive tract may cause water retention and, consequently, increase the digesta flow rate, leading to adverse effects on nutrient absorption and utilization and low AME. Generally, it is suggested that legume oligosaccharides have hygroscopic properties and may affect transit of gastrointestinal tract (GIT) digesta (Bedford, 1996) by elevating intestinal osmolarity (Smits and Annison, 1996). In our study, ether extract digestibility was also depressed as the raffinose content in seeds increased ( $r = -0.59$ ;  $P < 0.001$ ), which could partially explain the negative influence of raffinose content on AME<sub>N</sub> levels. Some authors have speculated that a high GAL content can depress ether extract digestibility due to overstimulation of GIT microflora. Coon et al. (1990) found that ileal digestibility of soyabean meal GAL was less than 1%, but its total tract digestibility could be over 90%, which suggests that raffinose and stachyose could be fermented by gut microflora.

The investigated narrow-leafed lupin cultivars were relatively good sources of lysine and arginine, which is characteristic of legume seeds in general (Gatel, 1994). Apparent amino acid digestibility ranged from 71.3% to 91.2%, and average crude protein digestibility was over 80%. In the case of all AA, two cultivars, Sonet and Graf, were characterized by lower apparent amino acid digestibility. We found that the apparent AA ileal digestibility of some amino acids was, however, negatively affected by seed WEV (Table 5). There have been only a few studies examining the effects of NSP on amino acid digestibility in poultry. Angkanaporn et al. (1994) suggested that wheat NSP increased endogenous amino acid losses, which may partially explain the differences among lupin cultivars in AA digestibility in our study. On the other hand, Boguhn and Rodehutsord (2010) did not find a significant improvement of AA digestibility after adding NSP-hydrolysing enzymes to wheat-SBM diets.

## Conclusions

The obtained results suggest that narrow-leafed lupines are good sources of protein, but poor sources of AME for chickens. On the other hand, our results show that the AME value for narrow-leafed lupin seeds presented by Smulikowska and Rutkowski (2005) could have been underestimated. It seems that the nutritional value of narrow-leafed lupin seeds for broilers depends, to a considerable extent, on raffinose content and on viscosity. Therefore, it could be concluded that *L. angustifolius* seeds characterized

by low water extract viscosity and a raffinose content below 1.2% have a high nutritional value.

It should be emphasised that current alkaloid levels in Polish *L. angustifolius* cultivars are low and they have a negligible impact on broiler performance.

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