

Compositional and nutritional evaluation of several lupin seeds

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Abstract

Lupin seeds of different species representing diverse varieties of sweet lupin grown in Poland were investigated. The chemical compositions of lupin isolates and amino acid composition of the proteins, as well as the nutritive values were estimated. No significant differences ($P \geq 0.05$) were observed among lupin isolates in their dry matter, crude fibre or alkaloid contents. The highest protein content (465 ± 11 g/kg d.m.) was found in seeds from lupins belonging to *Lupinus luteus* ($P \leq 0.01$), while the highest oil content (ca. 115 g/kg d.m.) was found in *Lupinus albus* ($P \leq 0.05$).

All the species examined were characterised by a shortage of methionine, lysine, tryptophan and valine while the level of leucine was satisfactory for most of the species. Yellow lupin was deficient in isoleucine. White lupin was found to be a nutritionally more valuable crop than other species by the standards of nutrition for mature human and animals. Apart from the highest level of amino acids within the crude protein (AA – 97.7 g/16 gN, $P \leq 0.01$), it was found to have a better and nutritionally more beneficial amino acid composition and the highest essential amino acids level (EAA). White lupin was characterised by a higher essential amino acid index (EAAI) as well as chemical score (CS) of restrictive amino acids, and the highest protein efficiency ratio (PER), expressed in terms of the availability of leucine and tyrosine as compared to blue and yellow lupin varieties. White lupin, followed by blue and yellow lupin, was found to be suitable for animal feeding as well as for the production of high-protein concentrates for further food processing and use in animal and human nutrition.

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1. Introduction

Lupin is an economically and agriculturally valuable plant which is able to grow in different soils and climates. Interest in lupin production is increasing, due to its potential as a source of protein, or for pharmaceutical purposes, a green manure or, due to the high alkaloid content, as a natural component of plant pesticides (Farrell, Perez-Maldonado, & Mannion, 1999; Gaultier et al., 2003; James, Panter, Gaffield, & Molyneux, 2004; Lam-

part-Szczapa et al., 2003; Perez-Maldonado, Mannion, & Farrell, 1999; Smart, Foster, Rothenberg, Higgins, & Hogan, 2003). Although lupin has been well-known, widely grown and utilised by people in the Mediterranean area and Andean highlands, in Europe its cultivation remains far behind that of other leguminous plants. In Poland, on fertile soils, mainly horse-bean, broad-bean and pea, competitive to white and narrow-leaved lupin (*Lupinus albus* and *Lupinus angustifolius*), are cultivated but, on exhausted or heavy soils, yellow lupin (*Lupinus luteus*) is the only highly productive plant which can be used for food and fodder production (Jasińska & Kotecki, 1993; Święcicki, 1987). Apart from the high protein content, lupin has a strong capability for

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nitrogen fixation and organic phosphorus release from soil and can be used in crop rotation during intensive grain production (Fan, Tang, & Rengel, 2002; Honeycutt, 1998). The utilisation of this plant can be extended to the production of protein concentrates, which – when added to other food products or fodder – can enrich their nutritional values, thus giving functional food (Archer, Johnson, Devereux, & Baxter, 2004; Batterham, Andersen, Lowe, & Darnell, 1986; Dijkstra, Linnemann, & van Boekel, 2003; Guillaume, Otterby, Linn, Stern, & Johnson, 1987; Linnemann & Dijkstra, 2002; Marrs, 1996). Lupin seeds may also be a potential source of alimentary cellulose for the production of dietetic food. The high-protein fraction (25–40%, Erickson, 1985) could be used as a substance for enriching different kinds of products, such as pastries, breads, chips and milk substitutes and also be a main food component when animal proteins are eliminated. Efforts to obtain lupin protein concentrates containing 60–70% of crude protein have been made in many laboratories (Chapleau & Lamballerie-Anton, 2003; Mubarak, 2001; Wasche, Müller, & Knauf, 2001).

The main anti-nutritional substances found in lupin seeds are various alkaloids of the quinolizidine group. These bitter compounds make the seed unpalatable and sometimes toxic (Michael, 2002, 2003; Torres, Quintos, Necha, & Wink, 2002; Wysocka & Brukwicki, 1998). The task of plant breeders is to produce an alkaloid-free lupin (sweet lupin) which can be consumed by humans after soaking in running water, or directly by animals and, which can be easily converted into protein-rich food. Although over recent decades, a growing body of research on sweet lupin has begun, mainly to produce species characterised by a low alkaloid content and short vegetation period, in Poland the level of cultivation of this plant is still considered to be low (Święcicki, Buirchell, & Cowling, 2000).

The aim of this paper is to analyse the compositional and nutritional profiles of a number of popular Polish cultivars belonging to three species of sweet lupins collected in the year 2003.

2. Materials and methods

2.1. Materials

2.1.1. Raw material

Lupin cultivars were chosen from the European species most popular and accessible in Poland. Lupin seeds of *L. luteus* (Idol, Juno, Kroton, Legat, Markiz, Mister, Parys, Polo and Taper), *L. angustifolius* (Baron, Cezar, Elf, Graf, Polonez, Sonet, Wersal and Zeus,) and *L. albus* (Boros and Butan) were used. Lupin seeds from Baron, Boros, Butan, Cezar, Elf Graf, Parys, Wersal and Zeus cvs were provided by Smolice Plant Breeding

and Acclimatisation – Przebędowo, Poznań, Poland. Seeds of Idol, Juno, Kroton, Legat, Markiz, Mister, Polonez, Sonet and Taper cvs were provided by Plant Breeding and Acclimatisation SHR Wiatrowo, Poland. The seeds were collected in the year 2003. The seeds were cleaned and rendered free of dust, then stored in tightly closed glass jars at room temperature until used.

2.1.2. Chemicals

Chemicals used in the analysis of dry matter, crude ash, crude protein, oil (ether extract), crude fibre, N-free extract and alkaloids (analytical grade) were purchased from POCh, Gliwice, Poland. Amino acid standards (analytical grade, apart from DL-tryptophan) were purchased from ZMBD CHEMIK, Prague, Czech Republic. DL-tryptophan standard was purchased from SERWA Feinbiochemica Heidelberg.

2.2. Methods

2.2.1. Preparation of samples

Ground lupin grain was produced by using a laboratory KNIFETEC 1095 sample mill, Foss Tecator.

2.2.2. Chemical composition

Dry matter, crude protein, oil, crude fibre, crude ash and N-free extracts were determined by the method described by Skulmowski (1974). All determinations were expressed on a dry matter basis.

2.2.3. Amino acids

Amino acids were determined using an AAA 400 automatic amino acid analyser (INGOS, Czech Republic). Prior to analysis, samples were subjected to acid hydrolysis in the presence of 6 M HCl at 105 °C for 24 hours. Sulphur-containing amino acids were determined separately in 6 M HCl after oxidative hydrolysis (formic acid + hydrogen peroxide, 9:1 v/v, 20 h at 4 °C). Tryptophan was determined according to the method described in the Official Methods of Analysis of the Association of Analytical Chemists (1990).

2.2.4. Anti-nutritional factors

Alkaloids were extracted and determined by the method of Wiewiórowski, Bratek, and Drzewiecka (1958).

2.2.5. Estimation of nutritional values of lupin protein isolates

The quality of protein was estimated by determination of total amino acids (AA), as well as the fraction of the exogenous amino acids (EAA). The nitrogen content in human food and fodder varies between 16 and 18 g/100 g of protein isolate (16 g/100 g for leguminous plants; FAO/WHO/UNU, 1985; FAO/WHO, 1991). Because the nutritional significance of much of the

non-peptide nitrogen is unclear, nitrogen analysis of foods is much more precise than the single amino acid analysis and nutritional significance than can be attached to it. Amino acid determinations were expressed on a g/16 gN basis, equivalent to g/100 g of protein.

The chemical score (CS) was calculated on the basis of the procedure described previously by Rakowska, Szkiłładziowa, and Kunachowicz (1978), based on comparison of the concentration ratio of the amino acid having the shortest supply a_i (restrictive amino acid) to the concentration of this amino acid in the standard a_s ($CS = (a_i/a_s) \times 100$). Two standards were used: amino acids of food protein composition appropriate for a mature human (MH) (FAO/WHO/UNU, 1985; FAO/WHO, 1991) and amino acid composition of the whole egg protein (WE) (Hidvégi & Békés, 1984), considered a complete and balanced food and fodder protein. The recommended levels of exogenous amino acids were as follows: Lys – 5.5 and 7.0 g/16 gN, Met + Cys – 3.5 and 5.7 g/16 gN, Thr – 4.0 and 4.7 g/16 gN, Ile – 4.0 and 5.4 g/16 gN, Trp – 1.0 and 1.7 g/16 gN, Val – 5.0 and 6.6 g/16 gN, Leu – 7.0 and 8.6 g/16 gN, His – 0 and 2.2 g/16 gN, Phe + Tyr – 6.0 and 9.3 g/16 gN, respectively, for mature human and whole egg protein standards. The exogenous amino acids (EAA) were estimated according to Oser (1959) in terms of geometric mean of all the concentrations of participating exogenous amino acids compared to a concentration of corresponding standard (in g/16 gN):

$$EAA = \sqrt[n]{a_1/a_{1s} \times 100 \times \dots \times a_n/a_{ns} \times 100},$$

where n is the number of participating amino acids, ns is the number of corresponding amino acids in standard.

In the classical method of Oser (1951, 1959), concentrations of Lys, sum of Met + Cys, Thr, Ile, Trp, Val, Leu, His and Phe + Tyr were considered, whereas the standard for mature human (MH) excludes histidine.

The essential amino acid index (EAAI) was calculated as follows:

$$EAAI = 10^{\log EAA},$$

where $\log EAA$ has the description (after Rakowska et al., 1978):

$$\log EAA = \frac{1}{10} \left(\log \frac{a_1}{a_{1s}} \times 100 + \log \frac{a_2}{a_{2s}} \times 100 + \dots + \log \frac{a_n}{a_{ns}} \times 100 \right).$$

Protein efficiency ratio (PER) was expressed traditionally as the ratio of the weight gain to the amount of the protein consumed in rat. According to Alsmeyer, Cunningham, and Happich (1974), this method cannot be applied to humans, mainly because it measures organism growth but not maintenance. These authors proposed an equation predicting protein usability which

is expressed in terms of concentrations of only two amino acids – leucine and tyrosine, based on experiments on their availability/digestibility:

$$PER = -0.468 + 0.454Leu - 0.105Tyr,$$

where Leu and Tyr are concentrations of these amino acids expressed in g/16 gN.

2.2.6. Statistical analysis

The admissible error for the determinations of chemical components was 5% while, in determination of amino acids and alkaloids, it was 10%.

One-way analysis of variance was carried out on the experimental results using species as an independent variable. The significance of differences between means was compared by Duncan's multiple range test. All calculations were performed using an ANOVA package from STATISTICA.pl.6.0.

3. Results and discussion

Table 1 shows the chemical composition of the lupin isolates reported on a dry matter basis. Similar results for dry matter/moisture have been reported previously (Cox, 1998; Dervas, Doxastakis, Zinoviadi, & Triandafylkos, 1999; El-Adawy, Rahma, El-Bedewey, & Gafar, 2001; Erbaş, Certel, & Uslu, 2005 and Petterson, 1998). Protein contents of the lupin seeds examined were higher than those of a lot of legumes. The highest protein content was found in the lupin varieties belonging to *L. luteus* (ca. 465 g/kg), followed by *L. albus* (ca. 360 g/kg) and *L. angustifolius* (ca. 330 g/kg). Significant differences between all the species examined were found ($P \leq 0.01$). In the case of *L. luteus*, protein content was higher than that reported for soy bean (41% d.m. – Favier, Ripert, Toque, & Feinberg, 1995). Crude protein in *L. angustifolius* and *L. albus* was lower than that found for *L. luteus* but higher than that reported previously (Fernández & Batterham, 1995) and also higher than the protein levels in haricot bean and lentil (29% and 27%, respectively) (Favier et al., 1995). On the other hand, the values obtained for the white lupin were lower than those reported for its other cultivars grown in Europe (Roth-Meier & Kirchgessner, 1993).

The species examined varied in oil contents. In white lupin the amount of oil was found to be significantly higher than those in yellow and white lupins ($P \leq 0.01$). The statistical difference between *L. luteus* and *L. angustifolius* was lower ($P \leq 0.05$). The quantity of oil found for white lupin (104 and 126 g/kg d.m. for the cvs Butan and Boros, respectively) was twice higher than those found for blue and yellow lupin and is close to the value of 112 g/kg reported by Becker, Marquard, and Gross (1989) for wild white lupin cultivars grown in Brazil. Oil content found for yellow lupin (55 ± 4 g/kg)

Table 1
Chemical compositions of lupin seeds (g/kg dry matter)

| Specification | Dry matter (g/ kg) | Crude protein | Oil | Crude fibre | Crude ash | N-free extract | Alkaloids |
|-----------------------------------|--------------------|------------------|------------------|-------------|------------------|------------------|-----------|
| <i>L. luteus</i> varieties | | | | | | | |
| Idol | 918 | 468 | 54 | 137 | 49 | 281 | 0.50 |
| Juno | 917 | 454 | 57 | 126 | 45 | 306 | 0.49 |
| Kroton | 919 | 482 | 56 | 118 | 45 | 288 | 2.06 |
| Legat | 918 | 464 | 45 | 128 | 45 | 307 | 0.72 |
| Markiz | 912 | 471 | 60 | 131 | 48 | 278 | 0.58 |
| Mister | 914 | 447 | 54 | 152 | 43 | 304 | 0.65 |
| Parys | 913 | 465 | 58 | 125 | 45 | 295 | 2.38 |
| Polo | 921 | 462 | 56 | 130 | 43 | 298 | 2.35 |
| Taper | 917 | 474 | 56 | 117 | 46 | 297 | 0.88 |
| Mean value | 917 | 465 ^A | 55 ^{Aa} | 139 | 46 ^A | 295 ^A | 1.18 |
| SD | ±3 | ±11 | ±4 | ±8 | ±2 | ±11 | |
| <i>L. angustifolius</i> varieties | | | | | | | |
| Baron | 907 | 330 | 55 | 123 | 39 | 434 | 0.33 |
| Cezar | 906 | 356 | 86 | 129 | 35 | 380 | 0.74 |
| Elf | 906 | 354 | 62 | 141 | 39 | 389 | 0.67 |
| Graf | 904 | 322 | 71 | 133 | 35 | 426 | 0.25 |
| Polonez | 909 | 318 | 64 | 126 | 40 | 440 | 0.94 |
| Sonet | 913 | 295 | 62 | 127 | 37 | 467 | 0.33 |
| Wersal | 903 | 340 | 66 | 119 | 35 | 426 | 1.00 |
| Zeus | 903 | 329 | 70 | 116 | 34 | 439 | 0.91 |
| Mean value | 906 | 330 ^B | 68 ^{Ab} | 140 | 37 ^B | 425 ^B | 0.65 |
| SD | ±3 | ±19 | ±8 | ±9 | ±2 | ±26 | |
| <i>L. albus</i> varieties | | | | | | | |
| Boros | 912 | 351 | 126 | 150 | 37 | 335 | 0.40 |
| Butan | 895 | 376 | 104 | 137 | 41 | 343 | 0.37 |
| Mean value | 904 | 363 ^C | 115 ^C | 144 | 39 ^{BC} | 339 ^C | 0.39 |

Means in the same column with different letters are significantly different; A–C – $P \leq 0.01$; a,b – $P \leq 0.05$.

was lower than that reported by Becker et al. (1989) but similar to that reported by Lubowicki, Petkov, Kotlarz, and Jaskowska (2000) for other popular cultivars grown in Poland in the years 1992–1996. In the case of blue lupin, the value of 68 ± 8 g/kg was similar to that reported previously for lupin varieties cultivated in Poland.

All the lupin extracts had medium/high amounts of crude fibre (ca. 140 g/kg). No statistical difference was observed between species concerning fibre content ($P \geq 0.05$). Crude fibre has many desirable functional properties, such as facilitating alimentary functions, helping in micro-component delivery and glucose metabolism and also slowing down the processes of re-absorption of undesirable dietary components, such as cholesterol (Chapleau & Lamballerie-Anton, 2003; Hall, Johnson, Baxter, & Ball, 2005; Sirtori et al., 2004). It also has a high water-holding capacity (7.1 g H₂O/g) (Huyghe, 1997), which potentially makes the lupin flour a good component of dietary products.

Ash content was highest in *L. luteus* (46 ± 2 g/kg), followed by *L. albus* (ca. 39 g/kg) and *L. angustifolius* (37 ± 2 g/kg). Yellow lupin had significantly more ash ($P \leq 0.01$) than other species while no statistical difference was observed between white and blue lupin ($P \geq 0.05$).

Nitrogen-free extract differed significantly between species ($P \leq 0.01$) and was as follows: *L. angustifolius* 425 ± 26 , *L. albus* ca. 340 g/kg and *L. luteus* 295 ± 11 g/kg. Apart from starch, sugars and pectin, this fraction contains water-soluble non-starch polysaccharides (NSP) as well as oligosaccharides. Erbaş et al. (2005) reported that the high content of sugars, especially mono- and disaccharides, is an advantage when lupin flour is used in production of different fermented products (such as bread and pastry additives). Experiments on young piglets show that, although they can negatively affect digestibility and nutrient absorption and act as anti-nutritional factors, their presence may also have beneficial dietary effects, such as helping casein digestibility and therefore, in the case of humans, helping in reduction of the prevalence of allergies (Gdala & Buraczewska, 1996; Gdala, 1998; Zduńczyk, Juśkiewicz, Frejnagel, & Gulewicz, 1998).

Alkaloids in some yellow lupin species exceeded that in blue and white lupins examined and varied between 0.5 and 2.4 g/kg dry matter, which are values higher than the recommended level. According to Erickson (1985) and Godfrey, Mercy, Emms, and Payne (1995), alkaloids in fodder and protein concentrates should not exceed 0.02–0.04% as a high alkaloid content can cause a significant decrease in protein digestibility and

may also result in neurological disorders, such as convulsions, unsettled balance or breathing disturbances (Agid, Pertuiset, & Dubois, 1988; Martinez, Loarca-Pina, & Ortiz, 2003; Pothier, Cheav, Galand, Dormeau, & Viel, 1998). Zduńczyk et al. (1998) reported that, although lupin species contain similar amounts of oligosaccharides, the concentrations of alkaloids can vary considerably. These authors discovered that the high ratio of oligosaccharides, accompanied by a low alkaloid content, may have a negative influence on protein digestibility, mainly in the case of animal feeding. This is, however, a matter of debate.

Tables 2–4 show the amino acid composition of the lupin seeds of *L. luteus*, *L. angustifolius* and *L. albus*, respectively. In contrast to plants, humans and animals are able to synthesise only 9 amino acids used in protein synthesis (non-essential amino acids). The biosynthesis of the remaining (essential) amino acids, thereby the protein synthesis, is not possible without their continuous supply through food consumption. In the case of low-protein diets, symptoms, such as delay in growth,

negative nitrogen uptake or disturbances in protein synthesis, can take place. Therefore foods and fodder rich in exogenous amino acids are desirable. The protein demand of different organisms depends on their physiological state stipulated mainly by age. For example, young and growing mammals (up to approximately two years in humans) need proteins rich in amino acids, such as arginine and histidine, as such amino acids are the source of the active centres of many enzymes. In contrast, adults show almost no physiological demand for these amino acids. Protein quantity, as well as composition, is the limitation of protein quality (Tabé & Higgins, 1998). For humans, adequate quantities of lysine, methionine and tryptophan are considered necessary in food of high nutritive value (FAO/WHO/UNU, 1985; FAO/WHO, 1991; Molvig et al., 1997). A number of approaches, based on the analysis of amino acids, have been considered for the estimation of protein quality in human and fodder foods. According to Alsmeyer et al. (1974), the nutritional value of food should be expressed in terms of leucine and tyrosine contents,

Table 2
Amino acid compositions and nutritional values of the seeds from *L. luteus*

| Specification | Idol | Juno | Kroton | Legat | Markiz | Mister | Parys | Polo | Taper | Mean value | MH ^a (%) | WE ^a (%) |
|--|------|------|--------|-------|--------|--------|-------|------|-------|------------|---------------------|---------------------|
| <i>Essential amino acids (g/16 gN)</i> | | | | | | | | | | | | |
| Lys | 4.3 | 4.6 | 4.4 | 4.5 | 4.6 | 4.2 | 4.5 | 4.6 | 4.6 | 4.5 | 81 | 64 |
| Met + Cys | 2.8 | 3.1 | 2.4 | 3.0 | 2.8 | 2.5 | 2.8 | 2.8 | 2.9 | 2.8 | 80 | 49 |
| Cys | 2.1 | 2.5 | 1.8 | 2.3 | 2.2 | 2.1 | 2.2 | 2.2 | 2.3 | 2.2 | | |
| Thr | 2.9 | 2.9 | 2.7 | 2.8 | 2.8 | 2.6 | 3.2 | 3.2 | 2.9 | 2.9 | 72 | 61 |
| Ile | 3.6 | 3.6 | 3.5 | 3.6 | 3.6 | 2.9 | 3.6 | 3.5 | 3.5 | 3.5 | 88 | 65 |
| Trp | 0.6 | 0.6 | 0.6 | 0.6 | 0.6 | 0.6 | 0.6 | 0.6 | 0.6 | 0.6 | 60 | 35 |
| Val | 3.2 | 3.1 | 3.3 | 3.3 | 3.4 | 2.9 | 3.1 | 3.3 | 3.3 | 3.2 | 64 | 49 |
| Leu | 6.6 | 6.8 | 6.9 | 7.0 | 7.3 | 6.1 | 6.6 | 6.5 | 7.2 | 6.8 | 96 | 79 |
| His | 2.5 | 2.6 | 3.0 | 3.0 | 2.8 | 2.8 | 2.4 | 2.4 | 2.4 | 2.7 | | 100 |
| Phe + Tyr | 4.4 | 5.0 | 4.9 | 4.9 | 5.1 | 4.2 | 4.8 | 5.3 | 5.4 | 4.9 | 82 | 53 |
| Tyr | 1.0 | 1.4 | 1.3 | 1.4 | 1.5 | 1.0 | 1.5 | 1.8 | 2.0 | 1.4 | | |
| <i>Non-essential amino acids (g/16 gN)</i> | | | | | | | | | | | | |
| Arg | 9.8 | 9.8 | 10.1 | 9.8 | 9.5 | 11.7 | 9.8 | 9.8 | 9.6 | 10.0 | | |
| Asp | 10.0 | 9.7 | 9.7 | 9.7 | 9.8 | 8.4 | 9.9 | 10.0 | 9.9 | 9.7 | | |
| Ser | 4.6 | 4.3 | 4.5 | 4.6 | 4.5 | 3.6 | 4.0 | 3.9 | 4.6 | 4.3 | | |
| Glu | 22.0 | 21.6 | 21.5 | 21.8 | 22.1 | 23.3 | 22.1 | 21.7 | 21.9 | 22.0 | | |
| Pro | 3.4 | 3.3 | 3.3 | 3.6 | 3.4 | 2.1 | 3.5 | 3.5 | 3.1 | 3.2 | | |
| Gly | 3.8 | 3.7 | 3.5 | 3.6 | 3.8 | 3.2 | 3.7 | 3.8 | 3.7 | 3.7 | | |
| Ala | 3.0 | 2.8 | 2.6 | 3.1 | 2.8 | 2.3 | 3.2 | 3.0 | 3.0 | 2.9 | | |
| <i>Nutritional values^b</i> | | | | | | | | | | | | |
| AA (g/16 gN) | 87.4 | 87.4 | 87.0 | 88.8 | 88.8 | 83.6 | 87.8 | 87.9 | 88.9 | 87.5 | | |
| EAA (g/16 gN) | 28.4 | 29.8 | 28.8 | 29.6 | 30.1 | 26.2 | 29.2 | 29.8 | 30.5 | 29.1 | + ^c | |
| CS | 60.5 | 59.4 | 57.3 | 59.7 | 58.3 | 59.0 | 62.4 | 63.4 | 59.4 | 59.9 | + ^c | |
| EAAI | 75.7 | 78.6 | 75.2 | 78.1 | 78.2 | 69.9 | 78.2 | 79.5 | 79.5 | 77.0 | + ^c | |
| EAA (g/16 gN) | 30.9 | 32.3 | 31.7 | 32.6 | 32.9 | 29.0 | 31.6 | 32.2 | 32.9 | 31.8 | | + ^c |
| CS | 35.6 | 34.9 | 33.7 | 35.1 | 34.3 | 34.7 | 36.9 | 37.3 | 34.9 | 35.2 | | + ^c |
| EAAI | 58.2 | 60.2 | 57.9 | 59.9 | 60.2 | 54.2 | 59.9 | 60.8 | 61.0 | 59.1 | | + ^c |
| PER | 2.39 | 2.47 | 2.49 | 2.54 | 2.67 | 2.17 | 2.36 | 2.28 | 2.59 | 2.40 | | |

^a Amino acid levels expressed as % of standards; MH-mature human, WE-whole egg protein standards.

^b AA, amino acid participation; EAA, essential amino acid participation; CS, chemical score of restrictive amino acid(s); EAAI, essential amino acid index; PER, protein efficiency ratio.

^c Calculated on the basis of MH or WE standard.

Table 3
Amino acid compositions and nutritional values of the seeds from *L. angustifolius*

| Specification | Baron | Cezar | Elf | Graf | Polonez | Sonet | Wersal | Zeus | Mean value | MH (%) | WE (%) |
|--|-------|-------|------|------|---------|-------|--------|------|------------|--------|--------|
| <i>Essential amino acids (g/16 gN)</i> | | | | | | | | | | | |
| Lys | 5.0 | 4.7 | 4.8 | 4.7 | 4.5 | 4.5 | 4.7 | 4.5 | 4.7 | 85 | 67 |
| Met + Cys | 2.2 | 2.0 | 2.0 | 2.3 | 2.3 | 2.3 | 2.1 | 1.9 | 2.1 | 60 | 37 |
| Cys | 1.5 | 1.3 | 1.3 | 1.6 | 1.6 | 1.6 | 1.5 | 1.3 | 1.4 | | |
| Thr | 3.0 | 3.0 | 3.2 | 3.1 | 3.3 | 3.3 | 3.0 | 3.1 | 3.1 | 78 | 66 |
| Ile | 4.0 | 3.9 | 4.0 | 3.7 | 3.5 | 3.7 | 3.6 | 3.8 | 3.8 | 94 | 70 |
| Trp | 0.7 | 0.7 | 0.7 | 0.7 | 0.7 | 0.7 | 0.7 | 0.7 | 0.7 | 71 | 42 |
| Val | 3.9 | 3.8 | 4.0 | 3.8 | 3.7 | 3.8 | 3.8 | 3.9 | 3.8 | 77 | 58 |
| Leu | 6.8 | 7.5 | 7.6 | 6.6 | 6.0 | 6.2 | 6.2 | 6.2 | 6.6 | 93 | 77 |
| His | 3.2 | 3.2 | 3.2 | 3.0 | 2.9 | 3.0 | 2.9 | 3.0 | 3.1 | | 100 |
| Phe + Tyr | 5.4 | 5.3 | 5.6 | 5.4 | 5.0 | 5.4 | 5.1 | 5.1 | 5.3 | 88 | 57 |
| Tyr | 1.6 | 1.5 | 1.7 | 1.6 | 1.5 | 1.9 | 1.5 | 1.4 | 1.6 | | |
| <i>Non-essential amino acids (g/16 gN)</i> | | | | | | | | | | | |
| Arg | 11.7 | 11.2 | 11.0 | 10.9 | 10.6 | 10.1 | 10.4 | 10.5 | 10.8 | | |
| Asp | 10.3 | 9.9 | 10.0 | 10.2 | 9.7 | 9.6 | 10.2 | 9.8 | 10.0 | | |
| Ser | 3.9 | 3.9 | 4.2 | 4.2 | 4.2 | 4.3 | 3.8 | 3.8 | 4.0 | | |
| Glu | 23.9 | 23.6 | 23.9 | 23.8 | 23.0 | 22.6 | 22.0 | 22.3 | 23.1 | | |
| Pro | 4.0 | 3.4 | 3.7 | 3.3 | 3.3 | 3.8 | 3.3 | 3.3 | 3.5 | | |
| Gly | 4.2 | 4.1 | 4.2 | 4.1 | 4.0 | 4.0 | 4.1 | 3.9 | 4.1 | | |
| Ala | 3.1 | 3.0 | 3.2 | 3.1 | 3.0 | 3.1 | 3.0 | 3.1 | 3.1 | | |
| AA (g/16 gN) | 95.4 | 93.3 | 95.3 | 92.9 | 89.6 | 90.5 | 89.0 | 88.9 | 91.9 | | |
| EAA (g/16 gN) | 31.2 | 30.9 | 31.8 | 30.4 | 28.9 | 30.0 | 29.2 | 29.2 | 30.2 | + | |
| CS | 61.1 | 54.6 | 55.1 | 65.4 | 63.1 | 64.2 | 61.1 | 56.6 | 60.2 | + | |
| EAAI | 82.6 | 79.4 | 81.8 | 81.4 | 77.9 | 80.9 | 78.2 | 77.8 | 80.0 | + | |
| EAA (g/16 gN) | 34.4 | 34.1 | 35.1 | 33.3 | 31.7 | 33.0 | 32.1 | 32.2 | 33.2 | | + |
| CS | 37.5 | 33.6 | 33.8 | 40.1 | 38.8 | 39.4 | 37.5 | 34.8 | 36.9 | | + |
| EAAI | 63.0 | 61.2 | 62.9 | 62.1 | 59.7 | 61.8 | 59.9 | 59.7 | 61.3 | | + |
| PER | 2.42 | 2.76 | 2.77 | 2.36 | 2.10 | 2.13 | 2.18 | 2.18 | 2.36 | | |

while other classifications are based on the chemical scores for 9–11 amino acids considered essential (Oser, 1959). Of great importance is the presence of sulphur-containing amino acids, mainly methionine, which is necessary for the synthesis of cysteine, as well as phenylalanine needed for the synthesis of tyrosine (James & Hove, 1980; Molvig et al., 1997). Methionine deficiency in the lupin species cultivated in Poland has been reported previously by Lubowicki et al., 2000. All the examined species manifest a large deficiency of sulphur-containing amino acids, for which the recommended level is 3.5 g/16 gN (Molvig et al., 1997). Methionine levels of 0.6–0.7 g/16 gN, found for the species examined, were low but comparable to results reported previously for other lupins (El-Adawy et al., 2001; Lubowicki et al., 2000; Petterson, 1998; Tabe & Higgins, 1998). The recommended level of methionine in animal feed is between 1.6 and 1.9 g/16 gN (Tabé & Higgins, 1998). As compared to standards for human and animal foods (MH & WE, respectively, see Tables 2–4), apart from methionine, all isolates were poor in lysine, tryptophan and valine, while the level of leucine was satisfactory for most of the species. In the case of isoleucine, the yellow lupin had the lowest content, while blue lupin was characterised by a small deficiency and

white lupin had a satisfactory level of this amino acid. Glutamic acid and aspartic acid were the major non-essential amino acids in all lupin protein isolates. For the statistical analysis see Table 5.

Although all the species differed in the amounts of crude protein, white lupin showed much higher amounts of total amino acids (AA) than did blue lupin and the yellow lupin ($P \leq 0.01$). The essential amino acids content (EAA) was calculated, as described previously in Section 2, on the basis of mature human (MH) and whole egg standards (WE) (see Tables 2–4). For all the species examined EAA were below the 36 g/16 gN recommended by Favier et al. (1995), based on nine exogenous amino acids (Lys, Met, Cys, Thr, Ile, Trp, Val, Leu and His). *L. albus* cvs contained more essential amino acids than *L. angustifolius* and *L. luteus*. No statistical difference was found for EAA in yellow and blue lupin while that of white lupin differed considerably ($P \leq 0.01$).

Nutritional values of lupin protein isolates were estimated. The chemical protein scores (CS) were calculated from the comparison of concentrations of less abundant amino acid(s) to a standard. Sulphur-containing amino acids were found restrictive for *L. angustifolius* and *L. albus* while tryptophan was for *L. luteus*. The highest

Table 4
Amino acid compositions and nutritional values of the seeds from *L. albus*

| Specification | Boros | Butan | Mean value | MH (%) | WE (%) |
|--|-------|-------|------------|--------|--------|
| <i>Essential amino acids (g/16 gN)</i> | | | | | |
| Lys | 5.1 | 4.8 | 4.9 | 90 | 71 |
| Met + Cys | 2.5 | 2.7 | 2.5 | 74 | 45 |
| Cys | 1.8 | 2.1 | 1.9 | | |
| Thr | 3.1 | 4.0 | 3.5 | 88 | 76 |
| Ile | 4.1 | 4.5 | 4.3 | 100 | 79 |
| Trp | 0.7 | 0.5 | 0.6 | 61 | 36 |
| Val | 3.8 | 4.3 | 4.1 | 82 | 62 |
| Leu | 8.2 | 7.5 | 7.8 | 100 | 91 |
| His | 3.1 | 3.5 | 3.3 | | 100 |
| Phe + Tyr | 5.5 | 5.9 | 5.6 | 95 | 61 |
| Tyr | 1.5 | 2.0 | 1.7 | | |
| <i>Non-essential amino acids (g/16 gN)</i> | | | | | |
| Arg | 11.1 | 11.7 | 11.4 | | |
| Asp | 9.9 | 11.1 | 10.5 | | |
| Ser | 4.1 | 4.9 | 4.5 | | |
| Glu | 24.2 | 22.9 | 23.5 | | |
| Pro | 3.8 | 3.1 | 3.5 | | |
| Gly | 4.3 | 4.4 | 4.3 | | |
| Ala | 3.1 | 3.2 | 3.2 | | |
| AA (g/16 gN) | 96.5 | 98.9 | 97.7 | | |
| EAA (g/16 gN) | 32.9 | 34.1 | 33.5 | + | |
| CS | 70.9 | 76.7 | 73.8 | + | |
| EAAI | 83.9 | 86.2 | 85.0 | + | |
| EAA (g/16 gN) | 36.0 | 37.7 | 36.8 | | + |
| CS | 43.5 | 47.1 | 45.3 | | + |
| EAAI | 65.0 | 66.6 | 65.8 | | + |
| PER | 3.06 | 2.69 | 2.87 | | |

chemical scores were calculated for white lupin, followed by blue and yellow lupins. No significant difference was found between yellow and blue lupins in their CS values while white lupin differed considerably ($P \leq 0.01$) from these species. White lupin had a higher essential amino acid index (EAAI) and also protein efficiency ratio (PER), based on lysine and tyrosine availability than had blue and yellow lupins.

In summary, no significant differences ($P \geq 0.05$) were observed among lupin isolates in their dry matter, crude fibre on alkaloid contents, which varied between species and varieties. White lupin was found to be a nutritionally most valuable crop as it had the highest oil content ($P \leq 0.01$) as well as less alkaloids than other species. It was also found to have a better and nutritionally more beneficial amino acid composition than had the blue and yellow lupin varieties. All the species were deficient in methionine. Also, the levels of lysine, tryptophan and valine were found to be below the standards of nutrition. Yellow lupin was deficient in isoleucine. The level of leucine was satisfactory for most of the species. The white lupin, followed by blue and yellow lupin, was found to be suitable for animal and human nutrition and also for the production of protein supplements and high-protein concentrates for further food processing.

Table 5
The results of analysis of variance carried out on the experimental results using species as an independent variable

| Specification | <i>L. luteus</i> | <i>L. angustifolius</i> | <i>L. albus</i> |
|--|--------------------|-------------------------|-------------------|
| <i>Essential amino acids (g/16 gN)</i> | | | |
| Lys | 4.5 ^A | 4.7 ^{Aa} | 4.9 ^{Bb} |
| Met + Cys | 2.8 ^A | 2.1 ^B | 2.5 ^A |
| Cys | 2.2 ^{Aa} | 1.4 ^B | 1.9 ^{Ab} |
| Thr | 2.9 | 3.1 | 3.5 |
| Ile | 3.5 ^A | 3.8 ^A | 4.3 ^B |
| Trp | 0.6 | 0.7 | 0.6 |
| Val | 3.2 ^A | 3.8 ^{Ba} | 4.1 ^{Bb} |
| Leu | 6.8 ^A | 6.6 ^A | 7.8 ^B |
| His ¹ | 2.7 ^{Aa} | 3.1 ^{Bb} | 3.3 ^B |
| Phe + Tyr | 4.9 ^A | 5.3 ^{AB} | 5.6 ^B |
| Tyr | 1.4 ^A | 1.6 ^A | 1.7 ^A |
| <i>Non-essential amino acids (g/16 gN)</i> | | | |
| Arg | 10.0 ^A | 10.8 ^{AB} | 11.4 ^B |
| Asp | 9.7 ^a | 10.0 ^{ab} | 10.5 ^b |
| Ser | 4.3 ^A | 4.0 ^A | 4.5 ^A |
| Glu | 22.0 ^{Aa} | 23.1 ^{Bb} | 23.5 ^B |
| Pro | 3.2 ^A | 3.5 ^A | 3.5 ^A |
| Gly | 3.7 ^A | 4.1 ^{Ba} | 4.3 ^{Bb} |
| Ala | 2.9 | 3.1 | 3.2 |
| AA (g/16 gN) | | 87.5 ^A | 91.9 ^B |
| Standard | MH | WE | |
| EAA (g/16 gN) | + | 29.1 ^A | 30.2 ^A |
| CS | + | 59.9 ^A | 60.2 ^A |
| EAAI | + | 77.0 ^A | 80.0 ^A |
| EAA (g/16gN) | | + | 31.8 ^A |
| CS | | + | 35.2 |
| EAAI | | + | 59.1 ^A |
| PER | | | 2.40 ^A |
| | | | 2.36 ^A |
| | | | 2.87 ^B |

Means in the same row with different letters are significantly different A–C, $P \leq 0.01$; a,b, $P \leq 0.05$.

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