

ANTI-NUTRITIONAL FACTORS IN SOME GRAIN LEGUMES

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Abstract: Grain legumes, such as soybean (*Glycine max* (L.) Merr.), pea (*Pisum sativum* L.), faba bean (*Vicia faba* L.), lupins (*Lupinus* spp.), common vetch (*Vicia sativa* L.) and grass pea (*Lathyrus sativus* L.), represent one of the most quality and least expensive solutions for a long-term demand for plant protein in animal husbandry. One of the limitations to an increased use of grain legumes as feed is the presence of diverse compounds in their grain, commonly referred to as anti-nutritional factors, that both decrease nutritive value of grain legumes and, if taken in larger amounts, cause health problems that may be fatal for both human and the animals. By this reason, breeding programmes of all grain legumes is aimed at decreasing the content of anti-nutritional factors to a safe extent. Breeding of soybean cultivars for reduced amount of antinutritive factors resulted in the development of Kunitz-free soybean cultivars, which are suitable for thermal processing at lower temperatures and during a shorter period of time. This is the way of saving energy and preserving valuable nutritional composition of soybean grain. As for other species, the most significant progress has been made in protein pea, where all modern cultivars have either low or very low content of various anti-nutritional factors. Among the improvements are also ‘zero-tannin’ cultivars in faba bean, with a wide utilization in both animal feeding and bread industry, low-toxin common vetch cultivars, ‘sweet’ cultivars in lupins and low-ODAP cultivars in grass pea.

Key words: anti-nutritional factors, feed legumes, grain, pea, soybean

Grain legumes in animal feeding

Annual legumes are one of the most important crops on a global scale. In animal feeding, they can be used as green forage, forage dry matter, forage meal, silage, haylage, immature grain, mature grain and straw, while some species may be used for grazing too (Mikić *et al.*, 2006). The term *grain legumes* denotes exclusively annual crops cultivated for immature or mature grain, with a further

division into *food legumes*, also known as *edible legumes* or *pulses*, used for human consumption, and *feed* or *fodder legumes*, used in animal feeding (Mihailović et al., 2004). Unlike these, *forage legumes* are used in the form of forage and comprise both annual and perennial species, having an additional role as a source of biomass and green manure (Mihailović et al., 2007b).

Globally, the most important feed grain legume is soybean (*Glycine max* (L.) Merr.), with a total production of 216,144,262 t and harvested area of 94,899,216 ha (Faostat, 2009). There follows pea (*Pisum sativum* L.), with a global production of 10,128,486 t and a harvested area of 6,896,172 ha, and faba bean (*Vicia faba* L.), with a total production of 4,868,681 t and harvested area of 2,626,809 ha. Common (*Vicia sativa* L.) and other vetches are cultivated on 782,318 ha, while lupins (*Lupinus* spp.) are grown on 679,999 ha on a global scale. Species such as grass pea (*Lathyrus sativus* L.) have more local importance. In Serbia, soybean is cultivated on about 150,000 ha, while feed pea and vetches are grown on about 30,000 ha (Mihailović et al., 2005b).

Table 1. Crude protein content (g kg⁻¹) and amino acid content (g kg⁻¹) in grain dry matter of some feed grain legumes (Mihailović et al., 2007a; Viveros et al., 2007)

Species	Soybean meal	Pea	Faba bean	Common vetch	White lupin	Grass pea
Crude protein	533.0	267.0	274.0	313.0	388.0	290.0
Alanine	30.2	4.4	3.3	4.4	13.3	13.5
Arginine	40.2	16.7	13.2	6.6	37.1	22.4
Aspartic acid	76.0	32.2	33.1	25.4	48.0	34.5
Cystine				4.5	4.2	5.0
Glutamic acid	35.8	17.8	28.7	22.1	78.0	49.8
Glycine	23.5	13.3	11.0	9.9	16.8	
Histidine	12.3	10.0	7.7	16.6	9.1	8.4
Isoleucine	29.1	13.3	11.0	14.3	13.9	10.8
Leucine	44.7	17.7	18.7	21.0	28.3	18.3
Lysine	36.9	13.3	16.5	16.6	14.1	19.4
Methionine	7.8	4.4	3.3	4.4	2.9	2.5
Phenylalanine	31.3	16.7	14.3	14.3	15.5	12.1
Proline	8.9	6.7	4.4	4.4	16.3	
Serine	64.8	31.1	28.7	19.9	21.3	13.7
Threonine	17.9	6.7	7.7	12.1	13.6	11.1
Tryptophan				2.4		2.1
Tyrosine	20.1	8.9	8.8	9.9	15.9	8.1
Valine	33.5	18.9	15.4		14.4	

Feed legumes have a great significance in animal feeding as one of the most quality and least expensive solutions for a long-term demand for plant protein in animal husbandry. Generally, feed legumes are characterised by a rather high crude protein content, ranging from about 270 g kg⁻¹ in pea and faba bean grain dry

matter, over nearly 400 g kg⁻¹ in white lupin (*Lupinus albus* L.) to more than 500 kg⁻¹ in soybean meal (Table 1). Due to a different amino acid composition between species, each of the feed legume species may easily find its useful place in animal feeding, supplementing and replacing each other.

One of the limitations to an increased use of grain legumes as feed is the presence of diverse compounds, commonly referred to as anti-nutritional factors, that both decrease nutritive value of grain legumes and, if taken in larger amounts, cause health problems that may be fatal for both human and the animals. By this reason, breeding programmes of all grain legumes is aimed at decreasing the content of anti-nutritional factors to a safe extent, leading to an increased proportion of grain legumes in diets for all species and categories of animals (Matić *et al.*, 2005).

Anti-nutritional factors in soybean

Among the anti-nutritional factors present in soybean seed, the main ones are protease inhibitors – Kunitz trypsin inhibitor (KTI) and Bowman-Birk inhibitor, and lectins. Protease inhibitors represent 6% of the protein present in soybean seed. Approximately, 80% of the trypsin inhibition is caused by KTI, which strongly inhibits trypsin and therefore reduces food intake by diminishing their digestion and absorption. Another effect of KTI is the induction of pancreatic enzyme, hyper secretion and the fast stimulation of pancreas growth, hypertrophy and hyperplasia. Due to this, raw soybean can not be used for feeding monogastric animals. Heat treatment doesn't completely eliminate these factors and may decrease protein solubility. Despite the efficiency of thermal treatment to reduce protease inhibitors, residual inhibition (10-20%) is maintained (Carvalho *et al.*, 1998). By this reason, a part of the breeding program of the Maize Research Institute Zemun Polje is aimed at developing soybean cultivars with reduced trypsin inhibitors content. As a result, two cultivars lacking KTI, Lana and Laura, were released Srebrić *et al.* (2008). The trypsin inhibitor content in these cultivars ranges from 15.01 mg g⁻¹ in Laura to 15.35 mg g⁻¹ in Lana, which is about 50% reduced as compared to the genotypes with standard grain type (Perić *et al.*, 2009). The utilization of these cultivars is a great opportunity for saving energy and preserving valuable nutritional composition of soybean grain, which is of interest in industrial processing. This trait makes them also suitable for direct feeding in adult non-ruminant animals without previous thermal processing.

Lectins are proteins that are widely distributed in plant kingdom and have unique property of binding carbohydrate-containing molecules with a high specificity, causing agglutination of red blood cells. Soybean agglutinin (SBA) causes the atrophy of the microvilli, reduces the viability of the epithelial cells and increases the weight of small intestine because of hyperplasia of crypt cells (Grant

et al., 1987; Puzstai *et al.*, 1990). The inactivation of soybean lectin by a moist heat treatment is parallel with the destruction of trypsin inhibitors. Soybean lectin is quite resistant to inactivation by dry heat treatment. The soybean genotypes lacking SBA were found Pull *et al.* (1978). The comparison of relative contribution of KTI and SBA on chicken growth revealed a greater anti-nutritional effect of KTI than of SBA (Douglas *et al.*, 1999).

Soybean contain high amount of oligosaccharides, consisting mainly of raffinose and stachyose. These oligosaccharides are poorly digested and have been implicated as causes for the poor utilization of energy from soybean meal fed to poultry (Lesake *et al.*, 1995). Raffinose and stachyose are heat stable; the attempts have been made to eliminate them by enzymatic action and selecting desirable soybean varieties (Neus *et al.*, 2005).

Phytic acid is present in soybean seed and products to the extent of 1-1.5% of DM. It is able to chelate mineral elements, such as zinc, magnesium, iron, calcium and potassium and makes these elements longer absorbed from intestines. About two thirds of the total phosphorus from soybean seed is bound to phytic acid (Nelson *et al.*, 1968). Several soybean genotypes have been developed with a low phytic acid content, often featured with lower grain yield and seed viability. More breeding cycles are needed to improve a cultivar performance and keep phytic acid at a low level (Spear and Fehr., 2007).

Beside mentioned anti-nutritional factors, soybean contains physiologically active compounds with small or unknown effects, such as tannins, saponins, antivitamin and isoflavones.

Anti-nutritional factors in feed pea

Main anti-nutritional factors in pea are trypsin inhibitors and lectins (Mihailović *et al.*, 2005c). As in other grain legumes, these factors have a proteinaceous nature, belonging to albumins, and are inactivated by high temperatures or soaking in formaldehyde. According to their trypsin inhibitor activity, expressed by trypsin inhibitor unit (TIU) per dry matter (DM), feed pea cultivars are classified into four groups: 1) very low activity (2-4 TIU mg⁻¹ DM), low activity (4-7 TIU mg⁻¹ DM), medium activity (7-10 TIU mg⁻¹ DM) and fairly high activity (10-13 TIU mg⁻¹ DM). Due to achievements in breeding, responding to the demands by animal husbandry, all modern feed pea cultivars have low or very low trypsin inhibitor activity, making farmers independent from processing industry and providing them with an excellent source of quality plant protein (Mikić *et al.*, 2003). Although it is confirmed that environment may influence trypsin inhibitor activity, it is certain that genotype remains the most important factor in its expression and thus underlines the role of breeding.

Among other anti-nutritional factors in pea, there are tannins and lectins. Tannins are present in coloured-flowered cultivars, that may be used for both forage and grain production, although often with decreased digestibility of grain crude protein in comparison with typical feed pea cultivars. The content of lectins is generally considered as low and thus of less importance.

Anti-nutritional factors in faba bean and other feed legumes

The main anti-nutritional factors in faba bean are tannins. Like in pea, there is a strong positive correlation between white colour of flowers and reduction of tannin content, being controlled by at least two recessive genes (*Duc, 1997*). The cultivars without tannins, commonly known as *zero-tannin* or *tannin-free* faba bean, have found a wide application for both human consumption and in animal feeding and essentially ensured the place of faba bean in feed production. The combination of the absence of tannins and winter hardiness in faba bean proved as possible and have resulted in the development of winter tannin-free cultivars (*Link et al., 2008*).

Among the poisonous matters that make common vetch unsuitable for monogastric animals and humans, the most important are γ -glutamyl- β -cyanoalanine, that has a bad influence on metabolism of sulphuric amino acids, and vicine and convicine, responsible for a disease called favism (*Mihailović et al., 2005a*). Among the most important achievements of the contemporary common vetch breeding are potential for a hay crude protein content up to 260 g kg⁻¹, a dry grain crude protein content of about 300 g kg⁻¹ and less than 0.60 % of grain toxins (*Matić et al., 2007*).

The main anti-nutritional factors in lupins are alkaloids, together with phytates, protease inhibitors and lectins. The development of so-called *sweet* types of white and other lupins, meaning the selection for low-alkaloid content, have been one of the major achievements in lupin domestication. Modern sweet lupin cultivars usually have an alkaloid content of less than 200 mg kg⁻¹ (*Cowling et al., 1998*), with a strict regulation in great lupin producers such as Australia and Poland. However, the cultivars with high alkaloid content, called *bitter lupins*, have their role as forage or green manure crops.

A neurotoxin, β -N-oxalyl-L-a, β -diaminopropionoc acid (ODAP) play the main anti-nutritional role in grass pea and causes a disease called lathyrism. The ODAP content depends on both genotype and environment, offering the development of cultivars for specific regions as the best solution (*Campbell, 1997*).

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Antinutritivni činioci nekih zrnenih mahunarki

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Rezime

Zrne mahunarke, poput soje (*Glycine max* (L.) Merr.), graška (*Pisum sativum* L.), boba (*Vicia faba* L.), lupina (*Lupinus* spp.), obične grahorice (*Vicia sativa* L.) i sastrice (*Lathyrus sativus* L.) predstavljaju jedno od najkvalitetnijih i najjeftinijih rešenja za dugoročni nedostatak biljnih proteina u stočarstvu. Jedno od ograničenja povećanju korišćenja zrnenih mahunarki u ishrani domaćih životinja jeste prisustvo različitih sastojaka u zrnu, tzv. antinutritivnih činilaca, koji umanjuju njihovu hranljivu vrednost i, u slučaju da se unesu u većoj količini, mogu da dovedu do teških posledica po ljude i životinje. Iz tog razloga, oplemenjivanje svih zrnenih mahunarki usmereno je i ka snižavanju sadržaja antinutritivnih činilaca na bezopasan nivo. Oplemenjivanje soje na smanjeni sadržaj antinutritivnih činilaca dovelo je do stvaranja *Kunitz-free* sorti soje, pogodnih za termičku obradu na nižim temperaturama i kraćeg trajanja, što je način uštede energije i očuvanja vrednih hranljivih sastojaka sojinog zrna. Što se tiče drugih zrnenih mahunarki, najveći napredak ostvaren je kod proteinskog graška, kod kojeg sve savremene sorte imaju nizak ili vrlo nizak sadržaj antinutritivnih činilaca. Među dostignućima su i *zero-tannin* sorte boba, sa širokom upotrebom u stočarstvu i industriji hleba, sorte obične grahorice sa niskim sadržajem toksina, tzv. slatke sorte lupina i sorte sastrice sa niskim sadržajem ODAP.

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