

THE CHEMICAL COMPOSITION OF NEW OAT VARIETIES AND BREEDING LINES CREATED IN LATVIA

Vita Sterna, Sanita Zute, Inga Jansone, Linda Brunava, Inara Kantane

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Oat (*Avena sativa* L.) is a unique cereal due to its relatively high protein content and its distinct protein composition compared to other cereals. The chemical composition and nutrition value are influenced by genetic and environmental factors. For further breeding of oats and developing new products there is an urgent need to increase knowledge on variation in content of fibre and bioactive components in European oats and also on the importance of growing conditions. Therefore aim of investigation was to analyze and evaluate the chemical composition of new oat varieties and breeding lines grown with different nitrogen fertilizer rate. Investigations were performed at the State Stende Cereal Breeding Institute. Husked oat variety 'Lizete', naked oat variety 'Stendes Emilija' and naked oat breeding line '33793' were grown with different nitrogen fertilizer rate. In the studied samples content of protein, starch, dietary fibre, β -glucans, α -tocopherol content and amino acids composition were determined. The tested husked and naked oat grains have higher content of Valine, Leucine, Isoleucine, Histidine and Arginine as mentioned for other varieties. Determined higher protein, essential amino acids and β -glucan content in oat grains due to increased nitrogen fertilizer rate, although correlations were weak. The new oat varieties 'Lizete' and 'Stendes Darta' and perspective breeding line '33793' are able provide high protein amount in grain at lower nitrogen fertilizer rate.

Key words: Protein, amino acids, dietary fibre, β -glucans, nitrogen fertilizer rate.

Vita Sterna, Sanita Zute, Inga Jansone, Linda Brunava. State Stende Cereal Breeding Institute, „Dižzemes”, Dižstende, Lībagi parish, Talsi County, LV-3258, Latvia, vitasterna@inbox.lv
Inara Kantane. Faculty of Chemistry, Latvia University, Riga, Latvia.

INTRODUCTION

Among the cereal grain crops, oats were considered as protein source, they are also a good source of fibre, fat, unsaturated fatty acids, vitamins and minerals. Oat protein accounts for approximately 104.9 and 126.7 g kg⁻¹ reported by Biel et al. (2014). The nutritional quality of dietary protein is related to the concentration of essential amino acids in the protein. The chemical analysis of the oat hydrolysate indicated that the oat groat proteins have an excellent amino

acid balance, nutritionally superior to the other cereal grains (Arendt and Zannini, 2013). Oats are characterised by higher essential amino acid content compared to other cereals, because the major proteins of oats are globulins (Shewry & Halford 2002).

The oat grain contains high amount of non-starchy polysaccharides which are the main constituents of dietary fibre. As defined, dietary fibre is the edible parts of plants or analogous carbohydrates, that are resistant to digestion and absorption in

the human small intestine with complete or partial fermentation in the large intestine. Dietary fibre can be divided into soluble dietary fibre (SDF) and insoluble dietary fibre (IDF) where the SDF forms a solution when mixed with water.

Oat β -glucan has outstanding functional properties and is of huge importance in human nutrition. Several studies have shown the cholesterol level decreasing due to increase β -glucans in human diet. The β -glucans content in oats ranges from 2 to 8 g 100 g⁻¹ of oat groats (Welch 1995) and is seemingly influenced by genetic and environmental factors. In comparison to the other cereals, oat β -glucan are present in higher levels and are more readily soluble - 88 % which is much higher than barley (69 %) (Arendt & Zannini 2013).

Due to the increased consumer awareness regarding health and the role of various foods in the improvement of quality of life, oats hold many opportunities for development as foods, feeds, industrial and pharmaceutical products, which all add value to the oat crop. For further breeding of oats, there is an urgent need to increase knowledge on variation in content of fibre and bioactive components in European oats and also on the importance of growing conditions (Arendt & Zannini, 2013). Old and new types of oats need to be characterized on a broader scale. Therefore the aim of investigation was to analyze and evaluate the chemical composition of new oat grains' varieties and breeding lines using different nitrogen fertilizer rate during vegetation.

MATERIAL AND METHODS

The research was conducted at the State Stende Cereal Breeding Institute. The material consisted of husked oat varieties 'Laima and 'Lizete', naked oat variety Stendes Emilija (S-156) and naked oat breeding lines '33793' and '34170'.

Field experiments made for season 2013. The soil type was sod podzolized sandy loam Albeluvisol (Eutric), content of organic

substance 26.1 mg.kg⁻¹, soil pH KCl 5.6, available phosphorus P₂O₅ 194.0 mg.kg⁻¹, and potassium K₂O 176.2 mg.kg⁻¹. The experimental treatment consisted of three nitrogen (N) rates - N80, N120 and N160. Complex mineral fertilizer NPK 17:10:14 was used as a basic fertilizer at the rate 470 kg per ha (pure matter N - 80 kg ha⁻¹, P₂O₅ - 47 kg ha⁻¹, K₂O - 66 kg ha⁻¹) in the field trial. The N application was split, part at the end of the tillering stage (growing stage/GS 29) of the crops. Ammonium nitrate (N 34%) was used as a top-fertilizer in the amount 40 kg N ha⁻¹ (N120) and 80 kg N ha⁻¹ (N160). The field treatments were laid out in a randomized complete block design (the plot size 10 m², four replicates).

Mean grain samples from all replications (0.5 kg) were taken for testing with Infratec Analyser 1241 (test weight, protein, starch, β -glucans, and lipid content).

Amino acids

Dried, defatted oat grains samples are treated with constant boiling 6N hydrochloric acid in an oven at around 110°C for 23 h. Amino acids were detected using reversed-phase HPLC/MS (Waters Alliance 2695, Waters 3100, column XTerra MS C18 5 μ m, 1x100 mm). Mobile phase (90% acetonitrile: 10% deionised water) 0.5mlmin⁻¹, column temperature. 40°C.

The sum of essential amino acids was calculated as EAA = Thr + Val + Met + Ile + Leu + Phe + His + Lys (FAO/WHO).

Dietary fibre

Samples are incubated at ~ 95 °C for 30 min in a phosphate buffer (pH 8.2) solution containing 100 μ L alfa-amylase. The pH is then adjusted to 7.5 and 100 μ L protease is added. After incubation at 60 °C for 30 min the pH is adjusted to 4.5. Before the last incubation at 60 °C for 30 min, 200 μ L amyloglucosidase is added. Duplicate samples are always processed, allowing the subtraction of protein and ash for the calculation of the total dietary fibre (TDF) content. Soluble dietary fibre

Table 1. Protein, fat, starch and dietary fibre content of oat grains

Trait	Amount of N fertilizer rate, kg ha ²		
	80	120	160
	Mean ± SD		
Protein content, g kg ⁻¹	136.8 ± 24.1	145.2 ± 26.2	149.5 ± 28.4
Fat content, g kg ⁻¹	88.2 ± 25.2	87.7 ± 26.4	85.0 ± 25.1
Starch, g kg ⁻¹	380.9 ± 81.8	370.3 ± 8.51	370.6 ± 80.6
Total dietary fibre, g kg ⁻¹	230.0 ± 8.5	235.9 ± 6.8	203.9 ± 6.7
β- glucans, g kg ⁻¹	32.1 ± 3.9	33.4 ± 3.3	33.4 ± 2.6

(SDF) calculated through same procedure without using ethanol precipitation, where after filtration obtained amount of insoluble dietary fibre (IDF) subtracting from TDF.

The statistical analysis was performed using SPSS 17. Correlation defined as medium close if $0.5 < r < 0.79$. Statistical significance was declared at $p < 0.05$.

RESULTS

The chemical composition of tested oat grains' samples under different rates of nitrogen application is showed in the Table 1.

The results of research demonstrated that higher protein content was determined in samples which grown with higher nitrogen fertilizer rate (160 kg nitrogen per ha²). A weak positive correlation $r=0.31$ between protein amount in oat grains and nitrogen fertilizer rate was calculated, which is significant ($p=0.032$), but protein amount was not differed significantly among groups with different nitrogen fertilizer rate ($p=0.204$). Results of research showed the same increase of protein content as husked oat variety 'Lizete', as well as naked oats „Stendes Emilija” and '33793'. Increase of protein amount in connection with nitrogen fertilizer rate in different oat varieties and breeding line are showed in Fig. 1.

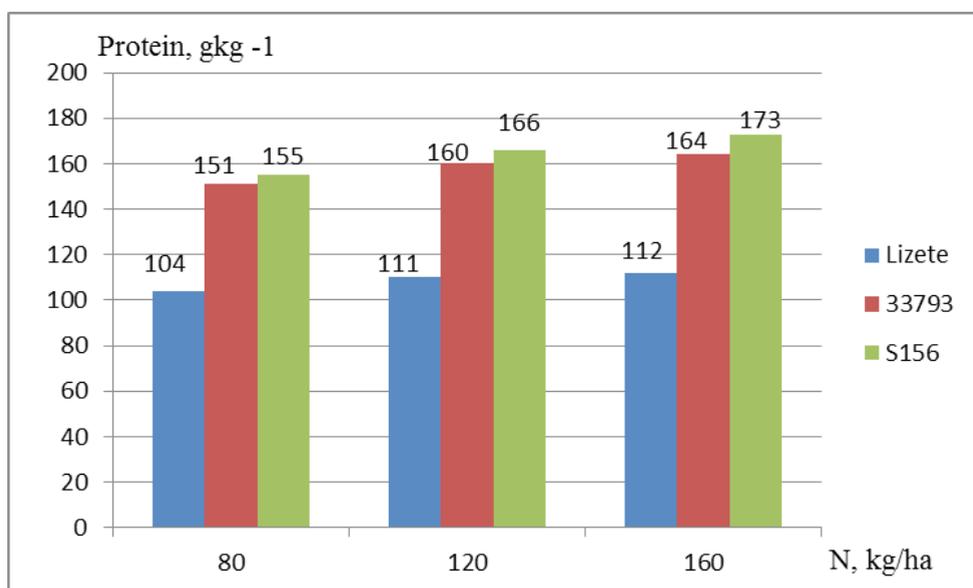


Fig. 1. Protein amount in oat grains grown in different nitrogen fertilizer rate.

Table 2. Composition of Amino Acids in oat grains grown in different conditions

Amino acids, g kg ⁻¹	Nitrogen fertilizer rate, kg ha ⁻¹			r*	Reference***
	80	120	160		
Val	6.65±0.48	6.95±0.71	7.32±0.95	0.414	4.5
Met	2.00±0.28	2.38±0.41	2.28±0.21	0.495	1.9
Ile	4.74±0.37	5.10±0.57	5.43±0.75	0.509	4.2
Leu	9.68±0.65	10.11±0.67	10.78±1.28	0.514	7.6
Lys	4.24±0.34	4.71±0.95	5.00±1.05	0.415	2.9
Thr	4.45±0.31	4.78±0.41	4.98±0.60	0.504	2.7
Phe	6.03±0.54	6.44±0.78	6.95±1.10	0.485	5.3
His	3.90±0.26	4.27±0.34	4.64±0.78	0.586	1.2
Tyr	4.02±0.31	4.30±0.74	4.48±0.77	0.340	3.3
Arg	10.22±0.60	10.93±0.98	11.42±1.28	0.518	4.8
Asp	9.78±0.80	10.34±1.25	10.58±1.94	0.363	5.4
Ser	6.05±0.68	6.28±0.59	6.50±1.00	0.278	3.5
Glu	27.31±3.40	29.34±3.37	31.28±5.31	0.434	32.5
Pro	7.99±0.57	8.48±0.78	8.88±1.13	0.459	10.8
Gly	6.41±0.50	6.60±0.51	6.87±0.64	0.384	4.4
Ala	5.99±0.51	6.31±0.56	6.57±0.88	0.391	3.8
EAA**	43.28±5.64	44.67±4.57	47.41±6.67	0.591	

* Correlation between amino acid and nitrogen fertilizer rate

** Sum of essential amino acids, g kg⁻¹

***Arendt, E.K. & Zannini E. (2013)

The fat content in tested oat varieties ranged from 5.2 to 12.1 g100g⁻¹, it was not differed significantly among groups with different nitrogen fertilizer rate and there was not observed correlation between oat grains fat content and nitrogen fertilizer rate. These differences are most likely due to genetic factors of varieties and breeding lines. The evaluation of starch amount and dietary fibre amount in oat grains grown in different conditions showed similar results. The results of research demonstrated that β- glucans amount in tested oat varieties and breeding lines ranged from 31 to 34 g kg⁻¹. There was observed a weak, positive correlation - r=0.32 between amount of β-glucans and nitrogen fertilizer rate, which is significant (p=0.029).

The sum of essential amino acids was not differed significantly in groups with different

nitrogen fertilizer rate ($p = 0.228$), but was observed positive medium close correlation ($r = 0.591$) between sum of essential amino acids and nitrogen fertilizer rate, which is statistically significant. Determined amino acids content of oat grains grown with different nitrogen fertilizer rate, total amino acid amount and correlation ratio reflected in Table 2. There were compared single amino acids content of oat grains from different varieties and breeding line in connection with nitrogen fertilizer rate in research (Fig. 2.) Higher amount of essential amino acids has variety '33793'.

DISCUSSION

The results of research demonstrated slightly higher protein content in comparison with other

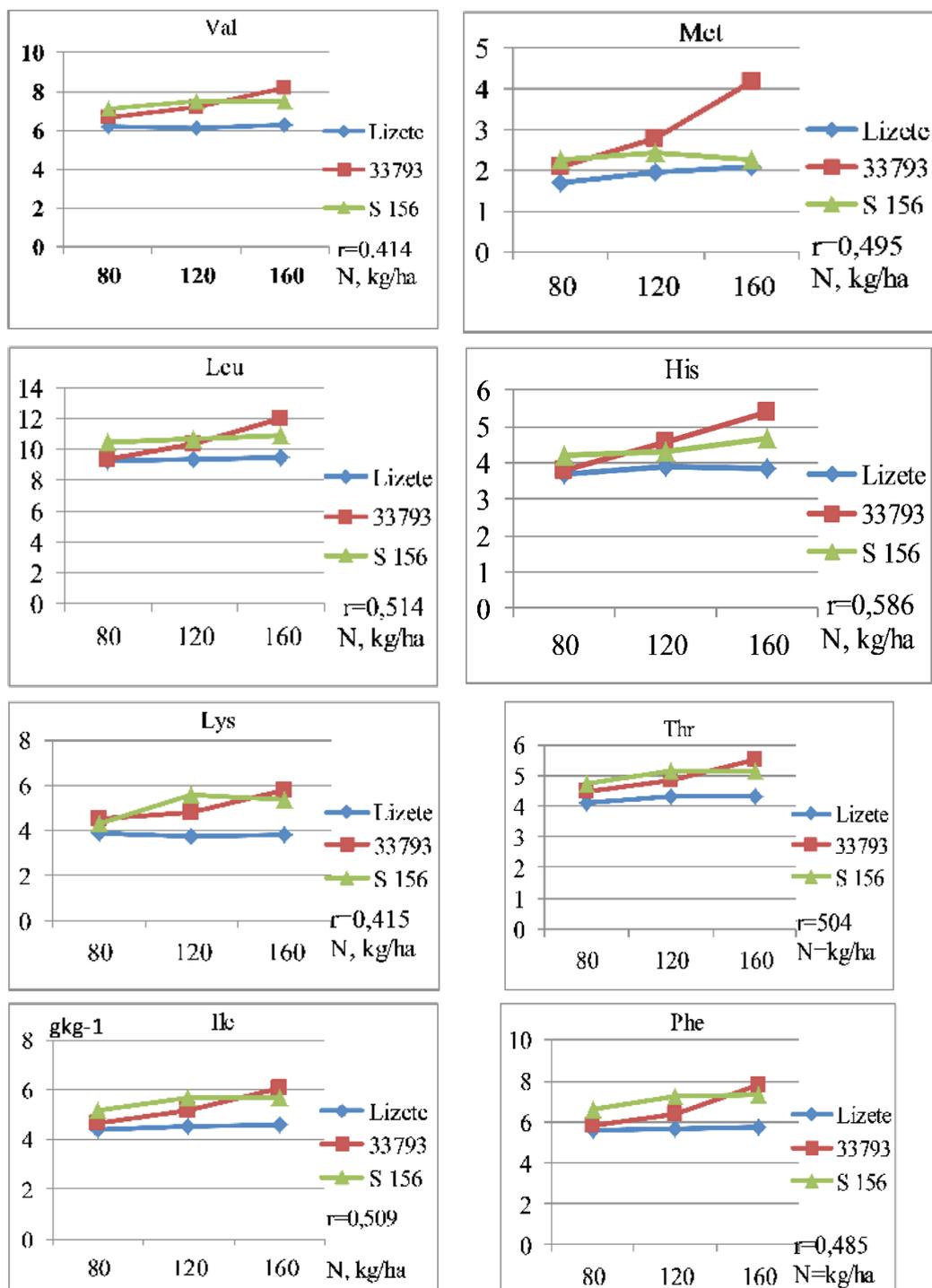


Fig. 2. Essential amino acids in oat samples, g Kg⁻¹

researchers findings, where protein content for husked oat was reported 115.0 g kg⁻¹, for naked oat - 143.4 g kg⁻¹ (Biel et. al., 2009). Givens et. al. (2004) reported that crude protein ranged 90 – 96 g kg⁻¹ for husked oats and 117 – 123 g kg⁻¹ for naked oats. Results of research assumed in figure 1, showed similar increase of protein amount as for husked oat variety ‘Lizete’, as well as naked oat variety ‘Stendes Emilija’ and naked oat breeding line ‘33793’. There is observed weak positive correlation between protein amount in oat grains and nitrogen fertilizer rate, but difference among groups grown in different conditions in same year is not significant. In turn, Givens et. al. (2004) concluded, that the optimum nitrogen fertiliser treatment (80-140 kg per ha) resulted in a consistent and significant (P<0.001) increase in crude protein for all varieties from a 95 to 118 g kg⁻¹ compared with zero nitrogen. The new oat varieties ‘Lizete’ and ‘Stendes Darta’ and perspective breeding line ‘33793’ are able provide high protein amount in grain at lower nitrogen fertilizer rate.

The fat content of tested oat grains varied from 5.20 to 12.40 g 100g⁻¹(5.2-12.4%). According to reported data (Zhou et al. 1999), the lipid content in the oat grain ranges from 3.1 to 11.8 %. There are not found data from literature about connection between fat content of oats and nitrogen fertilizer rate. Givens et. al. (2004) concluded that lipid content in oat is strongly dependent on meteorological conditions of sowing year. At the same time, optimum nitrogen treatment had a significant (P<0.001) reduction effect on starch levels from an average of 509 to 482 g kg⁻¹ DM (Givens et.al. 2004). The results of this study showed decrease of starch amount from 380.9 to 370.6 g kg⁻¹ and it was evaluated as not significant. The accumulation of starch is more sensitive to high temperature than the accumulation of nitrogen, which frequently determines increases in grain nitrogen proportion and thus results in higher protein content.

Oat fibre is the most variable constituent, with more than a five-fold variation across species. The results of research demonstrated high variation in total dietary fibre content in oat grain

– 136.6 g kg⁻¹ to 301.7 g kg⁻¹ (13.6-30.2%) and did not differ significantly by oat varieties or nitrogen fertilizer rate (p > 0.05). Data obtained in literature showed that oat caryopsis contains 10.2–12.1 % fibre, of which 4.1–4.9 % is soluble fibre and 6.0–7.1 % is insoluble fibre, depending on the oat genotype (Manthey et al. 1999).

The content of β-glucans of oat grain samples ranged 27 g kg⁻¹ to 35 g kg⁻¹ and it is lower in comparison with our previous results, where average content of β-glucans for naked oat varieties determined 49.9 g kg⁻¹ and 50.7 g kg⁻¹ (Brunava, 2014). Although observed weak significant correlation (r = 0.32) with nitrogen fertilizer rate, the difference between growing conditions in the same year was not significant (p = 0.174).

Not all proteins have the same nutritional value; protein quality strongly depends on its amino acid composition and digestibility. The sum of essential amino acids in this study ranged from 43.28 to 47.41 g kg⁻¹ Results of research demonstrated increase amino acid amount in oats grown with higher nitrogen fertilizer rate. As a whole is determined weak correlation, but Isoleucine, Leucine, Threonine, Histidine and Agrinine, the same as the sum of essential amino acids has medium close correlation with nitrogen fertilizer rate. The difference among oats grown using different nitrogen fertilizer rate was not significant. There are contradictions in scientific literature in connection with factors influenced amino acids composition. Some researchers consider, that amino acid composition of oat protein was unaffected by nitrogen, phosphorous and potassium fertilization although the effect of deficiencies of these nutrients decrease protein content (Arendt & Zannini, 2013), in turn, Givens et.al. (2004) concluded, that nitrogen fertilizer treatment significantly (P<0.001) increased the concentration of all amino acids. Results of single amino acid amount in different oat varieties in connection with nitrogen fertilizer rate (fig.2) showed that husked oat ‘Lizete’ has low amino acid increase, but naked oat breeding line ‘33793’ has strong increase of amino acid amount, when nitrogen fertilizer rate increased. Observed also,

that hulled oat variety „Stendes Dārta” and husked oat variety „Lizete” has similar response to nitrogen fertilizer rate.

CONCLUSIONS

Determined positive, medium close correlation between sum of essential amino acids, single amino acids Isoleucine, Leucine, Threonine, Histidine and Agrinine and nitrogen fertilizer rate. The new oat varieties ‘Lizete’ and ‘ Stendes Darta’ and perspective breeding line ‘33793’ are able provide high protein amount in grain at lower nitrogen fertilizer rate.

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