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REGULAR ARTICLE

NUTRITIONAL QUALITY OF GRAIN AMARANTHS (*AMARANTHUS* L.) COMPARED TO PUTATIVE MUTANT LINES

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ABSTRACT

Amaranth, an alternative crop, is attracting researcher's attention mainly because of high nutritional quality of seeds, which can be slightly changed also by induced mutagenesis. In this paper 15 grain amaranth genotypes including 8 putative mutation-derived lines have been studied. The average total seed protein content in mutation-derived amaranth lines was about 2.00 % higher than in non-treated amaranth genotypes. The significantly highest content of albumins and globulins was detected in line C 26 and C 27. The highest coefficient of nutritional quality had *Amaranthus hypochondriacus* Plainsman and putative mutant lines C 82 and C 15. Detailed analyses of biochemical traits in mutation-derived lines showed improved nutritional quality over the non-treated control varietes, therefore this plant material might be a good matrix in the amaranth further breeding programme.

Keywords: *Amaranthus*, seed proteins, γ radiation, nutritive value

INTRODUCTION

Grain amaranth (*Amaranthus* spp.) is an ancient crop originating from Central and South America. The high nutritional value of amaranth seeds, functional potential, brief growth cycle, adaptability to unfavorable climate and soil condition and the food use of the entire plant is the reason for increasing research interest in this pseudocereal **(Pospišil *et al.*, 2006; Capriles *et al.*, 2008)**. The cultivated amaranths are utilized as food grains, leafy vegetables, and forage crops in diverse geographic areas. For use in food products and human consumption, three varieties are of interest, namely *Amaranthus cruentus*, *A. caudatus* and *A. hypochondriacus*.

Amaranthus cruentus L. seed contain aproximately 4 % ash, 8 % fat, protein concentration between 12.5 % to 17.6 %, 13 % dietary fiber and 60 % starch **(Capriles *et al.*, 2008; Caselato-Suosa and Amaya-Farfán, 2012)**. It is an attractive protein source due to high content of essential amino acids, especially lysine and methionine, which are presented in limited quantities in cereal grains **(Gamel *et al.*, 2004; Capriles *et al.*, 2008; Caselato-Sousa and Amaya-Farfán, 2012)**.

Favorable composition of grain amaranth flour helps prevent certain diseases (heart condition, diabetes, brain stroke), and its high content of fiber and starch has a positive effect on digestion disorder **(Peterka *et al.*, 2001)**. Moreover, amaranth has been recognized as a source of beneficial micro-nutrients content like vitamins, minerals and antioxidants, such as riboflavin, niacin, ascorbic acid, calcium and magnesium contents. In the last few years, the properties of amaranth in cholesterol reduction as an anti-oxidant, anticancer, anti-allergic, and antihypertensive agent; and as a food for patients with celiac disease and immunodeficiences, have been assessed in clinical studies **(Castelano-Sousa and Amaya-Farfán, 2012).**

Conventional plant breeding is based on the use of genetic variation and selection of desired genotypes, what requires the screening of relatively large population. The simple and efficient techniques for inducing genetic variation, such as use of radiation for inducing of mutations and selection for desired traits is an essential component of any plant breeding programme **(Gajdošová et al., 2008)**. Mutagenesis is widely used method in plant breeding for improving characters and properties of plants and to increase genetic polymorphism in plant genomes **(Labajová *et al*., 2012)**.

The objective of our study was to compare the grain amaranths to mutagenesis-derived amaranth lines on the base of nutritional quality.

MATERIAL AND METHODS

Plant material

Two spieces of grain amaranth, *Amaranthus cruentus* (genotype Bergundy, Ficha and Olpir) and *Amaranthus hypochondriacus* (genotype Koniz, 1008, and Plainsman) and product of interspecific hybridization *Amaranthus hypochondriacus x Amaranthus hybridus* (K-433), were obtained from the Gene Bank of the Slovak Republic in the Plant Production Research Center Piešťany.

The seeds of *Amaranthus cruentus* genotype Ficha and hybrid K-433 were irradiated by γ radiation dose 175 Gy and positively selected during 14 mutant generations (1998-2012). Finally, five putative mutant lines of *A. cruentus* Ficha (C 26, C 27, C 82, C 236, C 279) and three lines of hybrid K-433 (D 54, D 279, D 282) with significantly increased weight of thousand seeds (WTS) were selected **(Gajdošová *et al*., 2007).**

In this work 15 grain amaranth genotypes including 8 putative mutation-derived lines from the year 2006 were analysed.

Biochemical characterization

The total nitrogen content was determined by improved Kjeldahl method using a Velp Scientifica system (DK 6 heating digester and UDK 127 basic destillation unit; Velp Scientifica, Italy) followed by titration with H_2SO_4. The fractional composition of protein was done using the Golenkov method **(Michalík, 2002)**, the nitrogen content in each fraction was determined by the micro-Kjeldahl method.

The content of crude protein from total nitrogen was calculated by the conversion rate (% N x 5,7). Coefficient of nutritional quality was calculated on the base of the protein fractions by the formula: ((albumins + globulins + rest) / prolamins)) x 100.

Statistical analysis

Results were analyzed using one way analysis of variance (ANOVA) and the test of Fisher´s least signifficant diference at a significance level of 0.01. These tests were realized in STATISTICA (data analysis software system) version 10. (StatSoft, Inc. 2011; **www.statsoft.com**).

RESULTS AND DISCUSSION

Crude protein content

Amaranth belongs to pseudocereals and has a reported protein concentration from 14.0 % to 17.0 %, according to **Bressani *et al.* (1987),** or from 13.2 % to 18.2 %, according to **Gorinstein *et al.* (2001).** The percentage of protein content in *A. cruentus* is about 16.2 % **(Mendoza and Bressani, 1987)** and 17.9 % in *A. hypochondriacus* seeds **(Cai *et al.*, 2004)**.

The protein contents with statistically significant differences in investigated amaranth genotypes and putative mutant lines are presented in Table 1.

Table 1 The percentage of crude protein content and fractional composition of protein in amaranth seeds

Genotype	Crude protein, %	Alb + Glo, %	Pro + Glu, %
Bergundy	10.795[a]	55.261[e]	32.445[d,e]
Ficha	11.994[c]	57.337[f]	30.935[b,c]
Olpir	10.795[a]	52.444[b,c]	29.481[a,b]
Koniz	12.794[d,e]	53.750[c,d,e]	29.125[a]
1008	11.994[c]	53.333[c,d]	30.000[a,b]
Plainsman	11.354[b]	51.417[b]	31.560[c,d]
K-433	12.953[e]	53.587[c,d,e]	30.252[a,b,c]
C 15	12.794[d,e]	52.508[b,c]	30.750[b,c]
C 26	10.875[a]	59.570[g]	30.891[b,c]
C 27	11.194[a,b]	58.571[f,g]	30.286[a,b,c]
C 82	12.474[d]	54.232[c,d,e]	30.770[b,c]
C 236	11.434[b]	54.829[d,e]	33.846[e,f]
D 54	13.593[f]	48.949[a]	34.473[f]
D 279	11.594[b,c]	50.764[a,b]	32.422[d,e]
D 282	13.033[e]	53.997[c,d,e]	37.429[g]

Legend: alb+glo (albumins+globulins), pro+glu (prolamins+glutelins); Different letters in the same colums indicate a significant difference between means at 1 % level according to Fisher LSD test.

The average total protein content in seeds of non-treated amaranth genotypes was 11.8±0.9 % of dry matter, which was about 2 % less than in irradiated putative mutant lines (12.0±1.0 %). This is in agreement with **Palenčárová and Gálová (2009)**, who detected the crude protein content in amaranth genotypes 11.4 %.

The highest protein content was determined in putative mutant line D 54 (13.6 %), which was statistically different to all other samples, on the other hand the lowest content was detected in amaranth genotype Bergundy (10.8 %). Putative mutant lines C 15 and C 82 had significantly higher content of crude protein to their reference sample genotype Ficha.

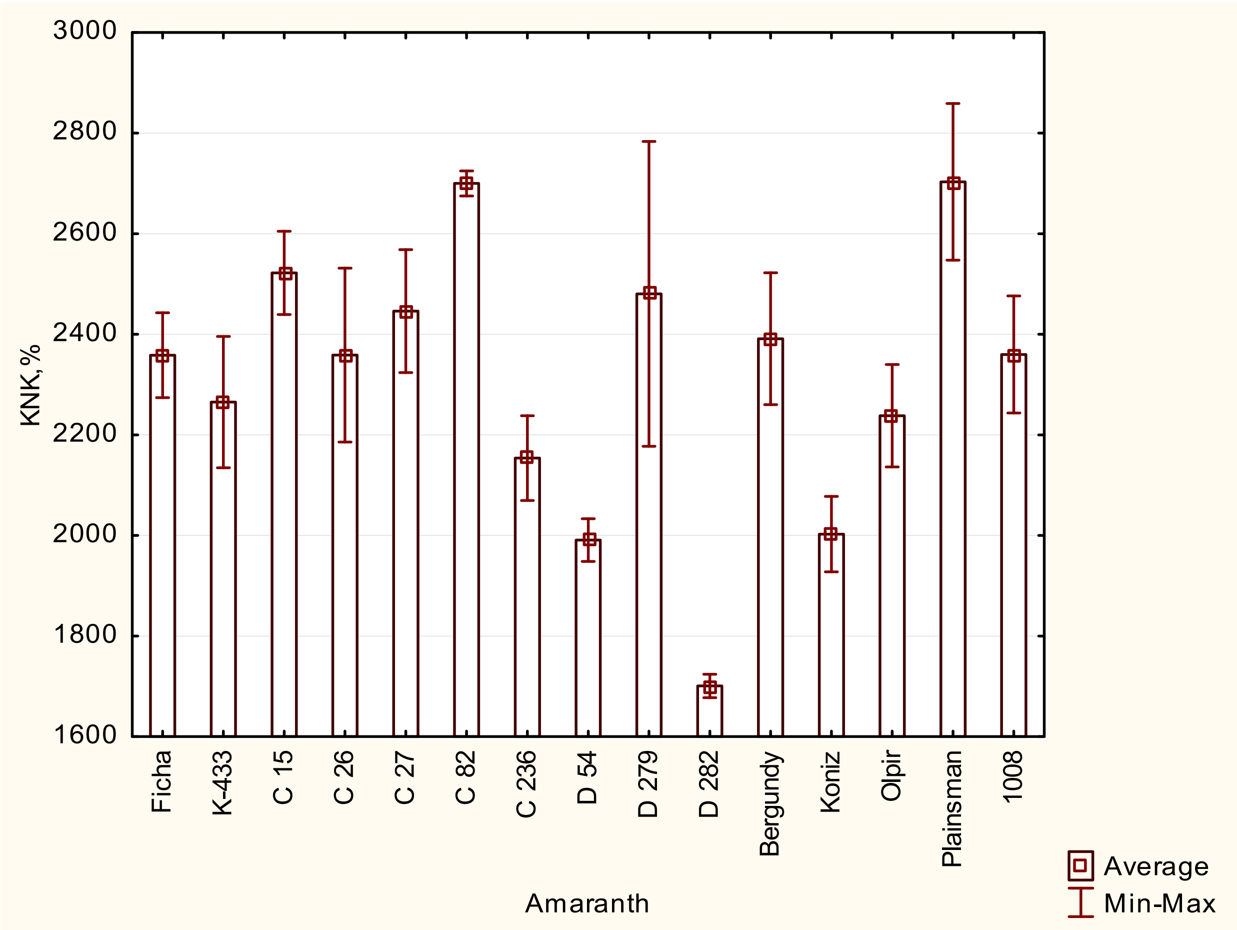
Fractional composition of proteins

Mandal and Mandal (2000) reported that recent classification divides seed proteins into storage, structural and biologically active proteins. The seed storage proteins are non-enzymatic and have the sole purpose of providing proteins (nitrogen and sulphur source) required during germination and establishment of a new plant. Seed proteins are generally classified by **Osborne (1924)** into four types based on solubility: albumins, globulins, prolamins and glutelins.

Most studies on amaranth proteins suggested that albumins are the major fraction (48.9-65.0 %), followed by glutelins (22.4-42.3 %), globulins (13.7-18.1 %) and prolamins (1.0-3.2 %) **(Cai *et al.*, 2004)**. The fractional composition of amaranth proteins indicates their high digestibility, since well digestable proteins, albumins and globulins with counterbalanced amino acid composition comprise more than 50 % of total fractional composition **(Zheleznov *et al.*, 1997)**.

The average content of cytoplasmic proteins in amaranth seeds was 54.1±2.8 %, what is in agreement with **Zheleznov *et al.* (1997)** results. The significantly highest value of albumins and globulins was detected in putative mutant line C 26 (59.6 %) and C 27 (58.6 %), the lowest content had line D 54 (49.0 %) (Tab. 1). The content of storage proteins (prolamins and glutelins) in non-treated amaranth genotypes ranged from 29.1 % to 32.4 %, while putative mutant lines contained from 30.28 % to 37.4 % prolamins and glutelins. Mutation-derived line D 282 indicated the highest content (37.4 %) of prolamins and glutelins and was statistically different to all other samples. Prolamines, alkali-soluble proteins with imbalanced amino acid composition and low content of essential amino acids, represented only 3.0 % in our samples (data not showed), whereas prolamines in wheat comprise about 37.4 % and in barley about 32.7 % of total seed proteins **(Gálová *et al.*, 2012).**

Coefficient of nutritional quality of pseudocereals is characteristic by high values, because of low content of storage proteins and higher content of albumins and globulins **(Gálová *et al.*, 2012)**. Mutation-derived amarant lines had in average about 1.6 % lower coefficient of nutritional quality compared to non-treated genotypes.



Picture 1 The coefficient of nutritional quality of amaranth genotypes

Legend: KNK- coefficient of nutritional quality.

On the other hand, putative mutant line C82 had the highest coefficient (2700.0 %) from mutation-derived samples and it was about 12.7 % when compared to reference sample Ficha (Picture 1). Putative mutant line D 282 had the significantly lowest coefficient of nutritional quality compared to all amaranth genotypes.

Several studies of the same collection of mutation-derived amaranth lines focused on enzyme polymorphism and DNA analysis have been done **(Labajová *et al.*, 2011a,b Múdry *et al.*, 2011; Kečkešová *et al.*, 2012;)**. These authors concluded, that from the results of eleven frequently studied enzymes in plant breeding and seed improvement, there were no significant differences between seeds, seedlings and leaves of putative mutants compared to untreated reference samples. **Labajová et al. (2011b)** reported, that amaranth accessions were analysed using microsatellite markers (ISSR techniqe) of which both produced reproducible polymorphic banding patterns and were able to distinguish both, the species specifity and mutant lines specifity. The calculated Jaccard similarity coefficient was in all cases higher than 0.5 what shows the high level of the same intergenic fragments in their intergenic space as the species-specific fingerprinting pattern.

CONCLUSION

The goal of presented work was to compare the protein content, fractions and coefficient of nutritional quality in amaranth putative mutant seeds with grain amaranth genotypes obtained from the Gene Bank of Slovak Republic in the Plant Production Research Center Piešťany. The nutritive value of amaranth putative mutant lines was comparable and even better than commercial genotypes including reference sampes. On the basis of results, we can predict, that mutation-derived amaranth lines could be considered as valuable matrix useful in further amaranth breeding programme.

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