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## **QUANTITATIVE AMINO ACID ANALYSIS OF SEEDS FROM COMMON BEAN UNIVERSITY COLLECTION**

### **SUMMARY**

Current study is concentrated on the abundance of Kazakhstan food as heirloom beans and pumpkins varieties and lines based on the extending germplasm including domestic as international accessions and specimens received from Asian countries, European sources, Russia, Turkey and USA. A range of introduced common bean varieties have indicated high seed germination and maturation rates as well as resistance to water deficit, whereas domestic varieties have been demonstrated to surpass certain foreign accessions and varieties by seed weight and other seed parameters. Data on bean collection, grown in steppe and mountain zones of Almaty region presented in that paper are completed by quantitative and qualitative amino acid evaluation.

Domestic and external varieties and lines (cvs “Aktatti”, “Bijchanka”, “Zuzka”, “Camelia”, “Katka”, “Luna”, “Nazym”, “Red Goya”, “Talgat” and “Ufinskaya”) have been clustered by using the data on amino acid composition of bean seeds analyzed by liquid chromatography. Essential amino acids have been shown to achieve 27.5 – 29.8% of total amino acids content in domestic lines. Apparently, Lys-Thr couple of essential amino acids known to be significant for plant growth. If tyrosinylation index (Phe/Tyr ration) for local lines is around 0.91 – 0.94, similar index for external varieties is of 0.88-0.89. Thus, it may mean that local lines contain membrane proteins possessing complex (mechanic, thermal and chemical) stability when compared with international bean analogues under investigation. By the content of individual amino acids (Glu, Asp, Ala and Pro) domestic bean lines have been shown to exceed external varieties 2.0-2.4 times.

**Keywords:** beans, variety, amino acid composition

### **INTRODUCTION**

The breeding outcome of new cultivars for common bean, *Phaseolus vulgaris* L. may be predicted from natural hybrids that are adapted to varying

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climate conditions within its growing zone. Consequently, the crop's ability to grow in other areas can be predicted. Transformation of wild species became especially extensive with human intervention, when forms with desirable nutritional quality and agronomic traits had been sought for (Bodnar and Lavrinenko, 1977).

During the course of evolution, bushy, large-leaf, early maturity forms with determinant type of growth, large number of flowers, and non-dehiscent pods have been selected (Zhukovsky, 1971).

In comparison to other legumes, common bean is more capricious in that it prefers fertilized sandy soils or light clay-containing soils. Cold clay soils with high moisture content are not considered to be appropriate for common bean. In addition, turf formation and soil compaction, caused by high acidification, may also decrease crop yield (Popov and Martynov, 2001). Due to acidic soils, the growth of nitrogen-fixing bacteria is reduced resulting in suppression of nitrogen fixation (Zerfus et al., 1997). Neutralization or lime addition (calcification) to acidic soils leads to increasing yields of common bean. Interestingly, various landraces of common bean are cultivated all over Kosovo, in combination with maize or as a monoculture in small gardens (Fetahu et al., 2013). It was determined that the stress has low impact on photosynthetic rates in common bean (Shekari et al., 2014).

Under mountain and steppe (plain) conditions of the Almaty Region, morphogenetic traits of 37 cultivars of common bean from different soil and climatic zones (Kazakhstan, American, Chinese, Polish, Russian, Turkish, and Czech collections) have been evaluated.

This study was carried out under crop rotation in mountain and steppe (plain) zones of the Almaty Region in 2011-2013. Apart from food and heirloom pumpkins, thirty-seven cultivars of common bean and its relatives were planted for: i, generation and study on domestic cultivars of common bean; ii, setting up the collection so as to be processed by the students under the supervision of researchers; iii, development of field and seed research capability at new "Zhangal Talap" Agrobiocenter of al-Farabi Kazakh National University; iv, analysis of amino acid composition in different bean seeds.

## MATERIAL AND METHODS

Part of stock varieties after preliminary propagation and introduction has been registered as the State Certificate on the subject of author rights No. 612 of 14 May, 2012 entitled: "Distribution and exchange of bean specimens".

Investigation on the varieties of this collection has been performed according to the Vavilov Institute and Awassa Agricultural Research Center protocols (Korsakov, 1975; Asfaw et al., 2009). Seeds were sown on plots of 2 x 10 meters, using double-row sowing with wide inter-row spacing (40-60 cm) and at least two replicates. The cultivar "Aktatti" was used as a standard for the Almaty Region. To provide computer aid to planning of the work and planting of

cultivars our own software entitled “Planting manager” (the State Certificate on the Subject of Property Rights No. 1034 of August, 1, 2012) was used.

Seventeen cultivars and lines of common bean, *Phaseolus vulgaris* L. were planted in the mountain zone (9 - at the field of the Institute of Botany and Phytointroduction; 8 - mountain plot in the Almarasan Gorge). Twenty cultivars and lines of common bean and its relatives (broad bean, *Vicia faba* L. and Turkish beans, *Phaseolus coccineus* L.) were planted in the steppe zone (“Zhang Talap” Agrobiocenter).

The current paper is focused on percentage of emergence and pod length along with the content of seed amino acids analyzed by liquid chromatography. Domestic bioorganomineral fertilizer, provided by the Faculty of Chemistry and Chemical Technologies, KazNU, was introduced into soil after 25-35 days post-planting in amounts of 25-30g per plot with subsequent moderate watering. Statistic treatment of the data obtained was performed by the methods of analysis of variance (Dospekhov, 1985; Bisgaard, 2008).

## RESULTS AND DISCUSSION

In the case of Czech collection of introduced cultivars, it was observed that the cv. “Zuzka” showed the highest percentage of emergence (53.0%) at the 30-th day after sowing under mountain conditions. Two other Czech cultivars showed the emergence of 23.3% (cv. “Katka”) and 16.6% (cv. “Luna”) under the same conditions. The Cv. “Zuzka” surpassed other cultivars in leaf size (11.2 x 8.0 cm), whereas these parameters for cvs. “Katka” and “Luna” were 6.5 x 4.5 and 9.3 x 6.4 cm, respectively. Furthermore, it was also noted that the cv. “Zuzka” was much earlier to flower than other cultivars.

Morphogenetic studies of genetic stocks for breeding and phenological observations over the process of sprout emergence indicated that local lines were superior to introduced Czech cultivars with respect to the percentage of emergence (see Table 1).

The percent emergence value for the cv. “Zuzka” was much greater than those for other Czech cultivars that were included in the study. This data are of theoretical and evolutionary significance as pod size, germination, seed size and lower stem length for warm-season legumes are considered as domestication-related traits (Isemura et al., 2007). It has been shown that common bean leaves possess egg-like or transitional to wide egg-like form under local conditions. In addition, some cultivars and lines had silver-polished stipules and variations in leaf colour, which is known to be a characteristic genetic trait intrinsic for the cultivar. This trait is dependent on the vegetative stage of the plant, soil quality and amounts of fertilizer applied.

One of the tasks of this study was to determine superior lines by examining phenotypic data for percentage of emergence obtained from a mountain location in 2012 in comparison with elite Czech common bean cultivars. This kind of cluster analyses based on other phenotypic characteristics (branch angle, height, hypocotyls diameter, lodging, maturity, upper pods, pods per plant, and yield)

was performed by Canadian researchers (Beattie et al., 2003) As shown in Fig. 1, with respect to the percentage of emergence, which was processed using computational cluster analysis, the local line “Nazym” is closer to the cv. “Zuzka”, as is another local line “Talgat”, which is more distant from two other Czech cultivars, “Katka” and “Luna”. This suggests that the line “Nazym” is unique by this morphogenetic character. This graph includes local and Czech lines only as Czech cultivars were introduced into the mountain zone in 2012. Noteworthy, similar approach was used for demonstrating genetic resemblance of the European and the North African faba bean germplasms which were closely associated with their geographical origins and ecological habits (Wang et al., 2012).

Table 1. Results of phenological observations: intermediate evaluation of percentages of emergence for Kazakhstan and Czech cultivars of common bean (30-th day after sowing)

No	Cultivar or line	Emergence, %%
1	Zuzka	53.33± 0.15***
2	Katka	23.33± 0.10***
3	Luna	16.67± 0.10
4	Nazym	78.26± 0.25***
5	Talgat	50.00± 0.20***

Footnote: \*\*\* P<0.001

Propagation of common bean collection in Kazakhstan is in progress. Similar research towards the enrichment and analysis of national bean collections is being done by other researchers (Asfaw et al., 2009, Kumar et al., 2008). The objective of the Chinese investigation (Zhang et al., 2008) was to evaluate a collection of domestic landraces for the genetic variability, genepool identity and relationships within and between the groups identified among the genotypes. The landraces were clustered into two genepools. Polish researchers evaluated the genetic diversity among commercial varieties and local landraces of the *Phaseolus* dwarf common bean and the *Phaseolus* runner bean to reveal a considerable polymorphism of *P. vulgaris* and *P. coccineus* accessions which formed distinct groups (Nowosielski et al., 2002). One of the positive outcomes of present study is the amount of polymorphism in stocks and possibility of introducing foreign cultivars, the Czech collection in particular. In the mountain zone of the Almaty Region (mountain plot is in the Almarasan Gorge) it was established that three cultivars, cvs. “Zuzka”, “Katka” and “Luna” have a highly desirable traits such as high yield and early maturity deserving further study. However, all Czech cultivars have shown high susceptibility to bean weevil (*Acanthoscelides obtectus* Sav.) at room temperature. Cv. “Luna” was observed to be the earliest to reach maturity with a maturation period of 80 days after planting. Other cultivars reached the same stage of maturity 10-12 days later.

The data showed that the maximal size of mature pod was attained on the 92-th day after planting, and it belonged to cv. “Zuzka” ( $13.3 + 0.1$  cm). The pod size values for cvs. “Katka” and “Luna” were  $12.0 + 0.2$  cm and  $10.8 + 0.1$  cm, respectively. The local line “Aktatti” had similar pod lengths in the range of  $11.0 + 0.1$  cm, whereas other local lines, “Nazym” and “Talgat” had pod sizes of  $12.4 + 0.1$  and  $9.0 + 0.2$  cm, respectively, at their technical maturity stage.

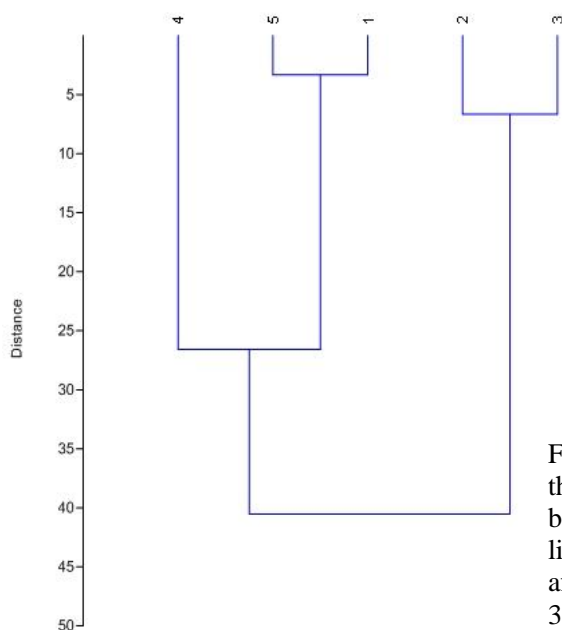


Figure 1. Cluster analysis presenting the germination rate differences between Kazakhstan and Czech lines of common bean (30-th day after sowing) 1. Zuzka 2. Katka 3. Luna 4. Nazym and 5. Talgat

Pod length values for cv. “Zuzka” and line “Talgat” were clearly much greater than those of other cultivars and lines investigated.

Essential amino acids have been indicated to compose in domestic lines 27.5 – 29.8% of total amino acids quantities. As Lys and Thr are significant for plant growth, it has been shown that the tyrosinylation index (Phe/Tyr ration) of local lines is around 0.91 – 0.94, whereas that index attributed to external varieties is of 0.88-0.89. This may emphasize that the quality of membrane proteins in local lines assists in acquiring a complex (mechanic, thermal and chemical) stability comparing to international common bean analogues under investigation.

By the amount of some important amino acids (Glu, Asp, Ala and Pro) domestic bean lines have been revealed to exceed external varieties 2.0-2.4 times. As shown in Fig. 2, if estimated by using the same computational attitude (see Fig. 1), the local line “Nazym” is unique by the amount of essential amino acids, taking over other cultivars and lines. In the steppe zone (“Zhanga Talap” Agrobiocenter) American, Polish and Russian common bean cultivars have been successfully propagated. These observations indicate that cvs. “Bijchanka”, “Camelia”, “Red Goya” and “Ufimskaya” would be most adapted to the steppe zone upon their introduction into that zone.

Using local “Aktatti” line, we investigated the effect of new domestic bioorganomineral fertilizer on morphogenetic traits of common bean plants. The results show that, the yield of this line can be increased by 19-25%, irrespective of climate conditions.

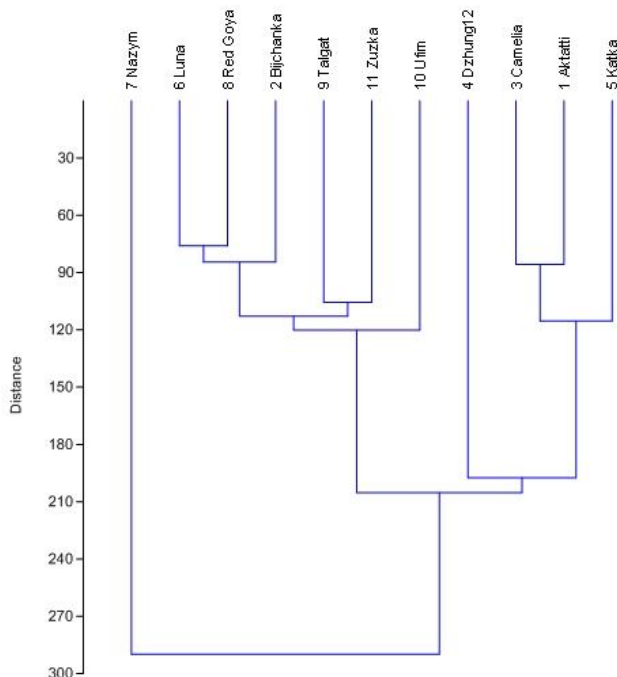


Fig. 2. Cluster analysis outlining the differences in quantities of essential amino acids between Kazakhstan, Czech and other cultivars and lines of common bean

## CONCLUSIONS

Based on the survey of morphogenetic traits of available seed stocks, a catalogue of main parental cultivars for common bean has been developed. It includes approximately 40 parental specimens of common bean and its relatives of diverse geographic origin. Out of the Czech bean collection introduced in the mountain zone, the variety noticed to reach maturity earliest was cv. “Luna” (80 days of maturation), whereas other varieties achieved their technical maturity 10-12 days later than “Luna”. As for germination rates, tested by computational cluster analysis, the local line “Nazym” being closer by maturity date to cv. “Zuzka” and other local bean line “Talgat”, appears to be more promising to be grown commercially in southeast regions of Kazakhstan on the basis of this and its other desirable traits. Local line “Nazym” is also referred to be advanced by the content of essential amino acids, taking over other cultivars and lines under the study. In addition to Czech and local cultivars and lines, six French cultivars of bush and liana common beans (“Argus”, “Coco nain blanc precoce”, “Triomphe de Farcy”, “Merveille de Venise”, “Mistica”, and “Phenomene” manufactured by “Truffaut” and “Vilmorin” companies), are currently being

investigated. Five of these cultivars (except cv. “Coco nain blanc precoce”) show high or average productivity. In addition, cv. “Argus” is distinguishable by its hybridization ability (the data are in progress).

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