# Uptake and utilization efficiency of nitrogen and phosphorus in barley genotypes

Svetla KOSTADINOVA, Nevenka GANUSHEVA and Marina MARCHEVA\*

Agricultural University – Plovdiv, 12 Mendeleev str, 4000 Plovdiv, Bulgaria \*correspondence: marcheva.marina@gmail.com

# Abstract

Field experiment was carried out in order to study the genotypic variation in nitrogen and phosphorus uptake and utilization by barley. The study included nine Bulgarian genotypes of two-rowed barley: three cultivars Krami, Krasen, Kristi and six perspectives breading lines. The genotypes were sown in a randomized, complete block design with four replications after maize as predecessor with the size of experimental plots of 7 m<sup>2</sup>. It was found that barley genotypes differed in the uptake efficiency of both elements - nitrogen and phosphorus. Genotypes were divided into two groups in regard to nitrogen. Cultivars Krami, Krasen, Kristi and lines 2390300, 24102400, 22506999 uptake 170-186 kg N\*ha<sup>-1</sup>, and lines 704112296, 24201900, 689069970 demonstrated higher nitrogen uptake efficiency 213-241 kg N\*ha<sup>-1</sup>. Most of the new lines had higher phosphorus uptake efficiency in a range 90-111 kg  $P_2O_5$ ha<sup>-1</sup> compared to the varieties. The ratio total N/total  $P_2O_5$  uptake was close to 2.0 for the most of genotypes. The obtained mean values of nitrogen utilization efficiency were 81.6 kg biomass and 34.0 kg grain per kg N, respectively. The mean values of phosphorus utilization efficiency were 171.3 kg biomass and 71.4 kg grain per kg  $P_2O_5$ , respectively. The barley genotypes utilized more efficient phosphorus then total uptake nitrogen for producing grain and straw. Line 24102400 demonstrated higher nitrogen utilization efficiency for biomass and grain from all studied genotypes, followed by variety Krami. These two genotypes could be recommended as perspective in future improves of nitrogen efficiency in barley. Uptake efficiency of nitrogen and phosphorus positively correlated with the barley productivity and nutrients content in the grain and straw. In contrast, the relationship between total uptake of both elements at maturity and utilization efficiency of nitrogen or phosphorus for grain formation was negative.

Keywords: barley, genotype, nitrogen, phosphorus, utilization efficiency

### Introduction

Global food security requires yield improvements or an expansion of land area used for agriculture (Smil, 2011). World population growth and economic development make increasing productivity a high priority for all agricultural systems and suggest that nutrient removal in crop harvest is likely to continue to increase. In addition optimum resource use efficiency is a prerequisite for sustainability (Hawkesford, 2012). The use of more nutrient-efficient crops is important for maintaining yields while enhancing environmental sustainability (Weih et al., 2011; Wignarajah, K., 1995). Nutrient use efficiency is a function of multiple interacting genetic and environmental factors and despite its importance, a clear understanding of the major mechanisms and inheritance of nutrient efficiency is lacking (Ladha et al., 2005). Agricultural breeding programs are conducted mainly in environments where inputs such as nitrogen fertilizers are highly regulated to ensure that crop deficiencies are minimized (Basra and Goval. 2001: Foulkes et al., 2009). In contrast, low-input and organic farming systems often have limited pools of mineral nitrogen and a greater reliance on organic sources and internal cycling of nitrogen. Consequently, traits related to nitrogen use efficiency (NUE) in an evolutionary context may be more important for low-input and organic systems than in conventional systems (Dawson et al., 2008). The current crop cultivars in temperate regions need better understanding of all the inefficiencies and in finding appropriate genetical or other innovations that will increase nutrient efficiency without slowing improvements in crop productivity (Anbessa et al., 2009; Fageria et al., 2008). There are many interpretations of nutrient (specifically of NUE) and its improvement (Glass, 2003; Hirel et al., 2007; Parry and Reynolds, 2007; Dawson et al., 2008; Fageria et al., 2008; Sylvester-Bradley et al., 2009). Fertilizer use efficiency reflects the recovery of applied fertilizer by the crop, however from the crop perspective, N (or other nutrient) use efficiency is a measure of biomass produced as a function of the N (or other nutrient) available to that crop (Dobermann, 2007; Snyder and Bruulsema, 2007). In cereals NUE is the product of two definable and independent major sub-traits - N uptake efficiency (NUpE) and N utilization efficiency (NUtE) (Moll et al., 1982). NUpE is the total N taken up by the crop as a fraction of the total N available; as such it is a measure of the ability of the crop to capture available N and is principally determined by rootassociated traits such as root depth proliferation and activity (Gerloff, 1987; Hawkesford, 2012). Total N-uptake may be affected by sink size, in the form of above ground biomass, but also in turn, directly determines the size of this biomass. NUtE reflects the functionality of the aboveground biomass, and for wheat is defined as the grain vield as a function of the total amount of N taken up (grain + straw). Canopy architecture, function and longevity determine the production of carbohydrate for grain filling and hence yield. A complication is the need for N by the grain during grain filling, a requirement fulfilled mainly by remobilization from the senescing (and hence decreasingly functionally active) canopy (Abeledo et al., 2008). Genotypic specificity of mineral nutrition has been demonstrated in cereals but studies have been conducted mainly with wheat and nitrogen from main nutrients.

The aim of the present study was to establish the specificity of uptake and use of nitrogen and phosphorus in new varieties and breading lines of malting barley for possible application in the breading program of this crop.

# Material and methods

The experiment was performed at the experimental field of Agricultural University – Plovdiv, Bulgaria (42°9' N, 24°45" E, 160 m altitude) during growing season 2008-2009. The study included nine Bulgarian genotypes of two-rowed barley: three cultivars Krami, Krasen, Kristi and six perspectives breeding lines. All genotypes were selected in the Department of Genetic and Plant breeding of Agricultural University – Plovdiv, The genotypes were sown in a randomized, complete block design with four replications after maize as predecessor with the size of experimental plots of 7 m<sup>2</sup>. Standard farming practices for the region of Southern Bulgaria were applied. Pre-sowing fertilization was done with 60 kg N\*ha<sup>-1</sup>. The soil type is alluvialmeadow (Mollic Fluvisols) with neutral reaction  $pH_{H2O}=7.20$ . It is poorly soloncovac, sand-clay. The subsoil waters are found at depth of 100-200 cm. Some of the more important agrochemical indexes are: humus content - 3.72%, total nitrogen 0.28%, total phosphorus – 0.32%, total potassium – 2.81%. Horizon A1 has a depth of up to 28 cm, brown-black, friable, with well distinguished crumb-granular structure, with roots and worm paths. The content of physical clay in the upper horizons reaches up to 50% (it is 33% in Horizon A). The soil contains a certain amount of calcium carbonate (1.63 – 3.00%), which adds to it favorable physical-chemical and water properties. The content of available nutrients in the soil before sowing were mineral N - 43.3 mg N min\*kg<sup>-1</sup>; phosphorus (Egner-Ream) - 228 mg  $P_2O_5$ \*kg<sup>-1</sup>; and exchangeable potassium - 380 mg K<sub>2</sub>O\*kg<sup>-1</sup>. Growing season 2008 - 2009 was characterized as favorable for barley growth and development in regard to mean temperatures and rainfall.

At physiological maturity from each plot plants from 1 m<sup>2</sup> area were harvested. The collected samples were separated to grain and straw (leaves+stems+glumes+awns), oven-dried to constant weight at 70 °C and ground for nitrogen and phosphorus analyses following Kjeldahl digestion with  $H_2SO_4$ – $H_2O_2$ . The concentration of total nitrogen was determined by distillation, and total phosphorus concentration was determined by the vanadomolybdate yellow colorimetric method. The amount of nitrogen and phosphorus in the grain and straw was calculated as multiplying dry mass of the plant part by nutrients concentration. On this basis the uptake efficiency of nitrogen and phosphorus in barley genotypes was determined (Gerloff, 1987). The utilization efficiency of uptake nutrients was defined as efficiency for biomass formation and efficiency of grain yield (Gerloff, 1987; Yang et al., 2003; Dawson et al., 2008). Harvest indexes of nitrogen and phosphorus were used for characterization of distributed nutrients to the grain. They were calculated in percentage as ratio grain N or P/ total aboveground plant N or P at maturity.

The obtained data were subjected to analysis of variance (ANOVA) and Duncan's multiple comparison range test procedure was employed to denote significant differences between the genotypes using the SPSS package (SAS Institute, 1990). Only differences at  $\alpha$  = 0.95 were accepted for proven.

#### Results and discussion

The average grain yield of genotypes was high, as most of the studied lines proved superior varieties Krasen and Kristi in this index and did not differ significantly from the standard variety Krami (Figure 1). Harvest index of the yields varied from 0.388 at line 24201900 to 0.461 at cultivar Krasen.

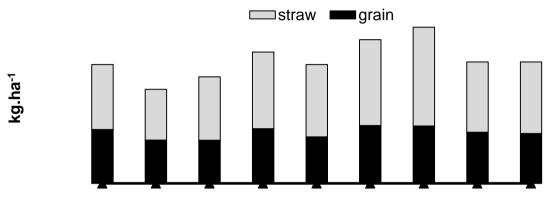


Figure 1. Grain and straw yields, and harvest index (HI, right axis y)

At physiological maturity (full ripeness) the concentrations of nitrogen and phosphorus in the grain and straw at the varieties and lines differed significantly between genotypes (Table 1).

Genotypes	Nitroge	n (N %)	Phosphorus (P <sub>2</sub> O <sub>5</sub> %)		
	Grain	Straw	Grain	Straw	
Krami	1.74 <sup>e</sup>	0.71 <sup>d</sup>	0.81 <sup>abc</sup>	0.40 <sup>c</sup>	
Krasen	2.06 <sup>a</sup>	0.84 <sup>b</sup>	0.86 <sup>a</sup>	0.43 <sup>abc</sup>	
Kristi	2.02 <sup>abc</sup>	0.89 <sup>a</sup>	0.73 <sup>d</sup>	0.39 <sup>c</sup>	
704112296	2.05 <sup>ab</sup>	0.76 <sup>c</sup>	0.85 <sup>a</sup>	0.42 <sup>bc</sup>	
2390300	1.97 <sup>bc</sup>	0.68 <sup>e</sup>	0.83 <sup>ab</sup>	0.50 <sup>a</sup>	
24102400	1.28 <sup>f</sup>	0.79 <sup>c</sup>	0.77 <sup>bcd</sup>	0.49 <sup>ab</sup>	
24201900	1.94 <sup>cd</sup>	0.83 <sup>b</sup>	0.83 <sup>ab</sup>	0.40 <sup>c</sup>	
689069970	2.10 <sup>a</sup>	0.81 <sup>b</sup>	0.83 <sup>ab</sup>	0.39 <sup>c</sup>	
22506999	1.87 <sup>d</sup>	0.71 <sup>d</sup>	0.76 <sup>cd</sup>	0.41 <sup>bc</sup>	

Table 1. Concentrations of nitrogen and phosphorus in barley grain and straw

The rank of arrangement of genotypes on concentration of nitrogen and phosphorus in the analysed plants parts was different and not unidirectional. The straw and chaff were approximately two times lower percentage of nitrogen and phosphorus compared to the grain. The concentration of nitrogen in the grain varied from 1.28% (line 24102400) to 2.10% (line 689069970). The concentration of phosphorus in the grain was changed in a more narrow range 0.73 - 0.85%  $P_2O_5$ .

The barley genotypes differ by absorbed amount nitrogen and phosphorus in the grain and straw (Table 2). By accumulated nitrogen in the grain the genotypes can be divided into three groups. Four genotypes accumulated comparatively less nitrogen in the grain (up to 12 kg N\*ha<sup>-1</sup>), three genotypes differ with more nitrogen in the grain (over 14 kg N\*ha<sup>-1</sup>), and the third group can be assigned the standard variety Krami and line 22506999 with accumulated in the grain 12.1 kg N\*ha<sup>-1</sup>. The studied cultivars and lines assimilated from 4.08 to 6.15 kg P<sub>2</sub>O<sub>5</sub>\*ha<sup>-1</sup> in the grain. Correlation between the amount of nitrogen or phosphorus in the grain and straw was not demonstrated. Exception was line 24201900 which accumulated the most nitrogen together in the grain and straw. Most of the studied barley genotypes allocated about 65% of the total accumulated nitrogen in the grain.

Genotypes	Nitrogen uptake, kg N*ha <sup>-1</sup>		NHI (%)	Phosphorus uptake, kg P <sub>2</sub> O <sub>5</sub> *ha <sup>-1</sup>		PHI (%)
	Grain	Straw		Grain	Straw	
Krami	12.1 <sup>bc</sup>	6.03 <sup>e</sup>	67.1 <sup>a</sup>	5.66 <sup>ab</sup>	3.37 <sup>de</sup>	62.8 <sup>ab</sup>
Krasen	11.5 <sup>c</sup>	5.50 <sup>f</sup>	67.7 <sup>a</sup>	4.82 <sup>c</sup>	2.85 <sup>e</sup>	62.9 <sup>ª</sup>
Kristi	11.3 <sup>c</sup>	7.31 <sup>c</sup>	60.5 <sup>bc</sup>	4.08 <sup>d</sup>	3.22 <sup>de</sup>	55.6 <sup>cd</sup>
704112296	14.5 <sup>a</sup>	7.50 <sup>c</sup>	65.8 <sup>a</sup>	5.98 <sup>a</sup>	4.16 <sup>bc</sup>	59.0 <sup>abc</sup>
2390300	11.8 <sup>c</sup>	6.12 <sup>e</sup>	65.8 <sup>a</sup>	4.96 <sup>bc</sup>	4.51 <sup>b</sup>	52.4 <sup>d</sup>
24102400	8.84 <sup>d</sup>	8.71 <sup>b</sup>	50.2 <sup>d</sup>	5.73 <sup>ab</sup>	5.37 <sup>a</sup>	51.6 <sup>d</sup>
24201900	14.4 <sup>a</sup>	9.70 <sup>a</sup>	59.7 <sup>c</sup>	6.15 <sup>a</sup>	4.75 <sup>ab</sup>	56.4 <sup>bcd</sup>
689069970	13.9 <sup>ab</sup>	7.32 <sup>c</sup>	65.4 <sup>a</sup>	5.47 <sup>abc</sup>	3.56 <sup>cde</sup>	60.5 <sup>abc</sup>
22506999	12.1 <sup>bc</sup>	6.50 <sup>d</sup>	64.8 <sup>ab</sup>	4.92 <sup>bc</sup>	3.78 <sup>cd</sup>	56.5 <sup>bcd</sup>

Table 2. Uptake and allocation of nitrogen and phosphorus in barley grain and straw

The allocation of total removed phosphorus in maturity to the grain was in the range 55.0 - 62.0%. The line 24102400 was characterized with the lowest nitrogen and phosphorus harvest indexes - 50.2% and 51.6%, respectively.

The efficiency of nutrients absorption often determined as the ability of the plants to absorb a certain element at low level of soil stocks or the nutrient medium (Dawson et al., 2008). Usually however the selection of the cultures were performed in agrochemical conditions not limited their growth and productivity (Abeledo et al., 2008). In the present study was proved genotypic reaction in the efficiency of absorption of both elements - nitrogen and phosphorus (Table 3).

Genotypes	Total uptake		Expense for 100 kg grain		$N : P_2O_5$
	kg N*ha⁻¹	kg P <sub>2</sub> O <sub>5</sub> *ha <sup>-1</sup>	kg N	kg $P_2O_5$	ratio
Krami	181 <sup>c</sup>	90 <sup>c</sup>	2.60 <sup>e</sup>	1.29 <sup>ns</sup>	2.00
Krasen	170 <sup>c</sup>	77 <sup>d</sup>	3.04 <sup>bcd</sup>	1.37	2.22
Kristi	186 <sup>c</sup>	73 <sup>d</sup>	3.35 <sup>a</sup>	1.32	2.54
704112296	220 <sup>b</sup>	101 <sup>b</sup>	3.12 abcd	1.44	2.17
2390300	180 <sup>c</sup>	95 <sup>bc</sup>	3.00 <sup>cd</sup>	1.58	1.90
24102400	176 <sup>c</sup>	111 <sup>a</sup>	2.36 <sup>e</sup>	1.49	1.58
24201900	241 <sup>a</sup>	109 <sup>a</sup>	3.25 <sup>ab</sup>	1.47	2.22
689069970	213 <sup>b</sup>	90 <sup>c</sup>	3.21 <sup>abc</sup>	1.37	2.36
22506999	186 <sup>c</sup>	87 <sup>c</sup>	2.89 <sup>de</sup>	1.35	2.14

Table 3. Total uptake of nitrogen and phosphorus in aboveground dry mass at maturity and expense for 100 kg barley grain

By a total nitrogen uptake amount from the soil barley accessions were divided into two groups. The first group included varieties Krami, Krasen, Kristi and breeding lines 2390300, 24102400 and 22506999, which uptake from the soil from 170 to 186 kg N\*ha<sup>-1</sup>. The second group included three of the studied lines (704112296, 24201900 and 689069970), which was characterized with higher nitrogen uptake from 213 to 241 kg N\*ha<sup>-1</sup>. The uptake of phosphorus for producing a 100 kg of barley grain or expense of P, was slightly changed in dependence of genotype with an average value of 1.41 kg P<sub>2</sub>O<sub>5</sub>. The results demonstrated a genotypic specific reaction of barley to expense of nitrogen to produce 100 kilograms of grain. Variety Krami and breeding line 24102400 used the least nitrogen for forming 100 kg of barley grain – 2.60 and 2.36 kg N, respectively. The highest N uptake for 100 kg grain was observed at variety Kristi – 3.35 kg N. The ratio total N/total P<sub>2</sub>O<sub>5</sub> uptake was close to 2.00 for the most of genotypes, excluding variety Kristi and line 24102400. Breeding line 24102400 was high efficient in respect to total phosphorus accumulation. At the same time that line allocated at least of the total removed nitrogen and phosphorus to the grain.

The obtained mean values of nitrogen utilization efficiency were 81.6 kg biomass per kg N and 34.0 kg grain per kg N (Table 4).

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Genotypes	NUEb	NUEg	PUEb	PUEg
Krami	84.9 b	38.5 b	170.4 bc	77.3 a
Krasen	71.3 e	32.9 cd	158.8 c	73.1 ab
Kristi	74.3 de	29.9 e	189.1 a	76.2 ab
704112296	77.1 cd	32.1 cde	167.6 bc	69.7 bc
2390300	83.6 b	33.4 cd	158.7 c	63.5 c
24102400	105.6 a	42.5 a	167.4 bc	67.5 bc
24201900	79.4 c	30.8 de	176.3 ab	68.3 bc
689069970	73.9 de	31.1 de	173.7 b	73.2ab
22506999	84.3 b	34.7 c	180.1 ab	74.2 ab

Table 4. Nitrogen and phosphorus utilization efficiency for biomass (NUEb and PUEb) and for grain (NUEg and PUEg) at barley genotypes

The mean values of phosphorus utilization efficiency were 171.3 kg biomass per kg total accumulated  $P_2O_5$  and 71.4 kg grain per kg  $P_2O_5$ . Therefore, the barley genotypes utilized more efficient total removed phosphorus in maturity then total uptake nitrogen for producing grain and straw. The breeding line 24102400 had the significant highest nitrogen utilization efficiency for biomass and for grain from all studied accession, followed by variety Krami. Kristi demonstrated lower nitrogen utilization efficiency for grain and for biomass formation - 29.9 74.3 kg per kg N, respectively. Variety Krasen was characterized as less effective genotype with respect to nitrogen and phosphorus utilization for the formation of biomass. Line 2390300 differed from the rest of genotypes in phosphorus utilization efficiency as less effective.

Total N or P uptake at maturity positively correlated with the productivity of barley genotypes (Table 5). Total N or P uptake at maturity (Uptake efficiency of nutrient) positively correlated with the productivity of barley genotypes and the nutrient content of grain and straw (Table 5). The negative relationship was observed between total accumulated N/P and utilization efficiency for grain formation (NUEg/PUEg).

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Coefficient of correlation	Grain yield	Straw yield	Total yield	N/P <sub>2</sub> O <sub>5</sub> in grain	N/P <sub>2</sub> O <sub>5</sub> in straw	NUEg/PUEg
r for N	0.517	0.666	0.650	0.831	0.679	-0.514
r for $P_2O_5$	0.894	0.884	0.925	0.914	0.854	-0.667

Table 5. Correlation between the total uptake of nitrogen (N) or phosphorus (P<sub>2</sub>O<sub>5</sub>) and some parameters at barley genotypes

# Conclusions

Barley genotypes were divided into two groups in regard to nitrogen uptake efficiency, Cultivars Krami, Krasen, Kristi and lines 2390300, 24102400, 22506999 uptake 170-186 kg N\*ha<sup>-1</sup>. Second group of lines 704112296, 24201900, 689069970 demonstrated higher nitrogen uptake efficiency of 213-241 kg N\*ha<sup>-1</sup>. Most of the new breeding barley lines had higher phosphorus uptake efficiency in a range 90-111 kg  $P_2O_5$ \*ha<sup>-1</sup>compared to the varieties. The ratio total N/total  $P_2O_5$  uptake was close to 2.0 for the most of genotypes. The obtained mean values of nitrogen utilization efficiency were 81.6 kg biomass and 34.0 kg grain per kg N, respectively. The mean values of phosphorus utilization efficiency were 171.3 kg biomass and 71.4 kg grain per kg  $P_2O_5$ , respectively. The barley genotypes utilized more efficient phosphorus then total uptake nitrogen for producing grain and straw. Line 24102400 was characterized with higher nitrogen utilization efficiency for biomass and grain from all studied genotypes, followed by variety Krami. These two genotypes could be recommended as perspective in future improves of nitrogen efficiency in barley. Uptake efficiency of nitrogen and phosphorus positively correlated with the barley productivity and nutrients content of grain and straw. In contrast, the relationship between total uptake of both elements at maturity and utilization efficiency of nitrogen or phosphorus for grain formation was negative.

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