



Original article

Non-destructive determination of protein and amino acids concentration and their relationship with grain yield affected by different irrigation, sowing date, cultivar and nitrogen treatments

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Extended abstract

Introduction

Maize (*Zea mays* L.) is an important component of human, livestock and poultry diet and its quality is increasingly being considered by the world market. In maize grain, protein and amino acids are important nutritional parameters that in addition to genetics, can be greatly affected by environmental factors and crop management practices. On the other hand, the increasing need for quality parameters such as protein requires the development of rapid and accurate measurement tools. Near-infrared (NIR) is a proven method for measuring the concentration of protein and amino acids, as it is described as a fast, low-cost, non-destructive and green method.

Materials and methods

In order to determine the effect of some agronomic factors on protein, total essential (EAA) and non-essential amino acids (NAA) and their relationships with maize grain yield, a field experiment was conducted in 2018 in a split-split plot factorial arrangement and as Randomized Complete Block Design (RCBD) with three replications where was considered irrigation intervals as main factor (12 days and 6 days intervals), planting date as sub factor (21 June and 22 July), maize cultivars (KSC704 and KSC260) and nitrogen fertilizer rate (zero and 400 kg ha⁻¹ Urea) as factorial treatments. Irrigation with 6 days intervals, 400 kg ha⁻¹ Urea and planting date 21 June were considered as typical non-stress conditions and common cultivation system of the region. At the physiological maturity stage, plants harvested from the second and third row of each plot were used to determine grain yield. The ears were shelled and the kernels were dried in a laboratory oven at the temperature of 60 °C until completely dry. Then grains were ground and the percentage of grain protein, EAA and NAA, were measured using a near infrared (NIR) spectrometer (model FOSS NIRS XDS – Denmark) in the wavelength range of 1100 to 2500 nm with five intervals. Data obtained were examined by analysis of variance using the F test by SAS (version 9). The F values were considered significant at the error probability level of 5% (p<0.05). Minitab software (version 17) was used to calculate the correlation and regression coefficients.

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Results

The results revealed that higher irrigation, nitrogen, early planting date, and KSC260 cultivar increased grain yield by 18.26, 8.03, 29.06 and 7.70%, respectively. The interaction effect of irrigation and nitrogen on protein concentration was significant ($P \leq 0.05$). Lower irrigation and higher nitrogen fertilizer increased the protein by 0.78% compared with non-fertilizer treatment. The protein concentration in the KSC260 was higher than KSC 704 by 1.06%. The effect of irrigation on all amino acids excluding valine, lysine and tryptophan was significant and they were increased as irrigation intervals increased. Applying urea increased all amino acids except lysine. The effect of planting date on some amino acids was significant and they often were higher by late planting date. The effect of cultivar on all amino acids excluding asparagine, lysine and tryptophan was significant and was higher in KSC260 than KSC 704. Higher irrigation and nitrogen application decreased EAA to NAA ratio by 0.20 and 0.09%, respectively ($P \leq 0.05$). Grain yield was positively correlated with protein, TAA and NAA concentration (Pearson's $r = 0.37, 0.35, 0.40$, respectively). There was a positive and significant relationship between protein with EAA and NAA and the highest relationship was observed between protein and NAA ($a=0.1545, R^2 = 0.32\%, P \leq 0.001$).

Conclusions

Findings suggested that using higher irrigation and nitrogen fertilizer increased maize grain yield and also essential amino acids concentration, and decreased the ratio of essential amino acids to non-essential ones. Due to the high complexity and sensitivity of measuring qualitative characteristics such as protein concentration and composition, replicating this experiment in different years and locations is recommended, taking into account the higher levels of influential factors.

Keywords: Amino Acid, Protein, Maize, Near-Infrared (NIR) method, Quality

Table 1. Effects of irrigation (I), planting date (PD), nitrogen fertilizer (N) and cultivar (C) on protein and amino acid concentration in maize grain.

Treatment	Essential Amino Acids (EAA) (%)									
	Met	Thr	Val	Ile	Leu	Phe	His	Arg	Lys	Trp
Irrigation (I)										
I1	0.161 ^a	0.269 ^a	0.356 ^a	0.258 ^a	0.882 ^a	0.362 ^a	0.228 ^a	0.369 ^a	0.231 ^a	0.060 ^a
I2	0.159 ^b	0.265 ^b	0.359 ^a	0.256 ^b	0.876 ^b	0.355 ^b	0.222 ^b	0.361 ^b	0.231 ^a	0.060 ^a
<i>P-value</i>	0.0014	<0.0001	0.1998	0.0016	0.0013	0.0004	<0.0001	<0.0001	0.7006	0.3700
Planting Date (PD)										
PD1	0.159 ^b	0.263 ^b	0.352 ^b	0.251 ^b	0.874 ^b	0.355 ^b	0.224 ^a	0.363 ^b	0.230 ^b	0.0601 ^b
PD2	0.160 ^a	0.268 ^a	0.363 ^a	0.262 ^a	0.883 ^a	0.362 ^a	0.226 ^a	0.367 ^a	0.232 ^a	0.0609 ^a
<i>P-value</i>	0.0167	0.0001	<0.0001	<0.0001	<0.0001	0.0002	0.0594	0.0022	0.0095	0.0315
Cultivar (C)										
C1	0.161 ^a	0.265 ^b	0.353 ^b	0.253 ^b	0.868 ^b	0.354 ^b	0.221 ^b	0.358 ^b	0.230 ^a	0.062 ^a
C2	0.163 ^a	0.269 ^a	0.361 ^a	0.260 ^a	0.889 ^a	0.363 ^a	0.229 ^a	0.373 ^a	0.234 ^a	0.061 ^a
<i>P-value</i>	0.1032	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.1148	0.1121
Nitrogen (N)										
N1	0.156 ^b	0.265 ^b	0.351 ^b	0.248 ^b	0.871 ^b	0.352 ^b	0.223 ^b	0.361 ^b	0.232 ^a	0.059 ^b
N2	0.164 ^a	0.269 ^a	0.363 ^a	0.265 ^a	0.886 ^a	0.364 ^a	0.228 ^a	0.370 ^a	0.230 ^a	0.061 ^a
<i>P-value</i>	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0002	<0.0001	0.1112	<0.0001

Table 1. Continued

Treatment	Protein (%)	(%.) Nonessential Amino Acids (NAA)						
		Cys	Asp	Ser	Glu	Pro	Gly	Ala
Irrigation (I)								
I1	10.500 ^a	0.176 ^a	0.552 ^a	0.365 ^a	1.567 ^a	0.687 ^a	0.315 ^a	0.585 ^a
I2	9.749 ^b	0.152 ^b	0.488 ^b	0.347 ^b	1.393 ^b	0.359 ^b	0.301 ^b	0.555 ^b
<i>P-value</i>	0.0017	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Planting Date (PD)								
PD1	10.145 ^a	0.163 ^a	0.504 ^a	0.355 ^a	1.476 ^a	0.669 ^b	0.302 ^b	0.568 ^a
PD2	10.104 ^a	0.165 ^a	0.507 ^a	0.357 ^a	1.485 ^a	0.678 ^a	0.314 ^a	0.572 ^a
<i>P-value</i>	0.8493	0.2977	0.4590	0.5336	0.5290	0.0012	<0.0001	0.2412
Cultivar (C)								
C1	9.915 ^b	0.161 ^b	0.502 ^a	0.348 ^b	1.457 ^b	0.667 ^b	0.305 ^b	0.559 ^b
C2	10.335 ^a	0.166 ^a	0.508 ^a	0.364 ^a	1.503 ^a	0.679 ^a	0.312 ^a	0.581 ^a
<i>P-value</i>	0.0598	0.0057	0.1059	<0.0001	0.0029	<0.0001	0.0016	<0.0001
Nitrogen (N)								
N1	9.808 ^b	0.155 ^b	0.467 ^b	0.346 ^b	1.407 ^b	0.666 ^b	0.301 ^b	0.546 ^b
N2	10.441 ^a	0.172 ^a	0.543 ^a	0.366 ^a	1.554 ^a	0.680 ^a	0.315 ^a	0.595 ^a
<i>P-value</i>	0.0065	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

I1; Irrigation with 12 day intervals, I2; Irrigation with 6 day intervals, PD1; Planting Date June 21, PD2; Planting Date July 22, C1; cultivar KSC704, C2; cultivar KSC260, N1; without N fertilizer, N2; N fertilizer at optimal level. Means sharing the same case letter do not differ significantly at P < 0.05 based on LSD test

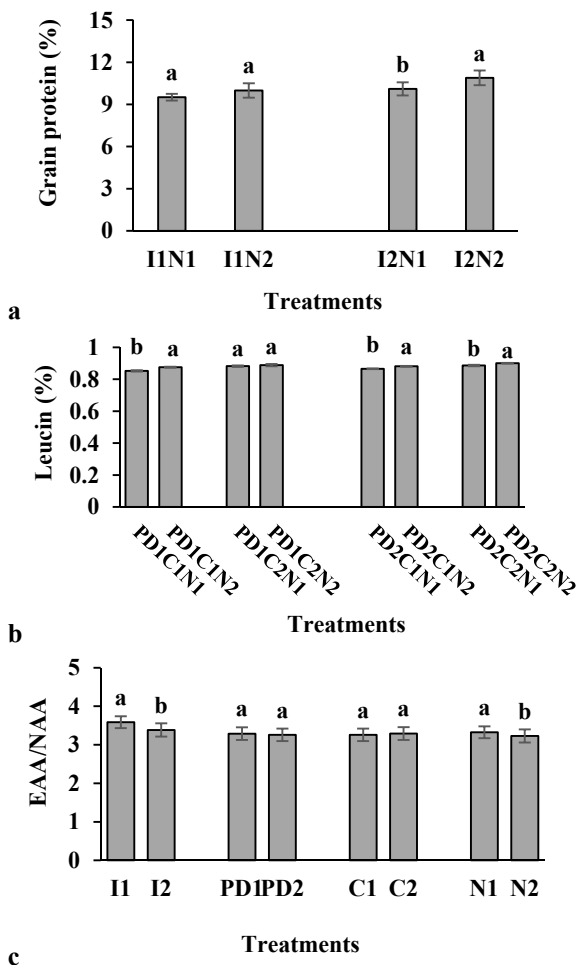


Fig. 1. Interaction effect of irrigation and nitrogen on protein concentration (a), interaction effect of planting date, cultivar and nitrogen on amount of Leucine amino acid (b), ratio of essential amino acids (EAA) to non-essential amino acids (NAA) at different irrigation levels, planting date, cultivar and nitrogen fertilizer (d). (I1; Irrigation with 12 day intervals, I2; Irrigation with 6 day intervals, PD1; Planting Date June 21, PD2; Planting Date July 22, C1; cultivar KSC704, C2; cultivar KSC260, N1; without N fertilizer, N2; N fertilizer at optimal level. Means sharing the same case letter do not differ significantly at P < 0.05 based on LSD test).

Table 2. Correlations among grain yield, concentration of protein, total amino acids, essential amino acids, and non-essential amino acids

	1	2	3	4	5
1 Grain yield	1				
2 Protein	0.37**	1			
3 Total amino acids	0.34*	0.56**	1		
4 Essential amino acids	0.23 ^{ns}	0.43**	0.83**	1	
5 Non-essential amino acids	0.40**	0.56**	0.98**	0.74**	1

ns: non-significant, *: significant at $P < 0.05$, **: significant at $P < 0.01$

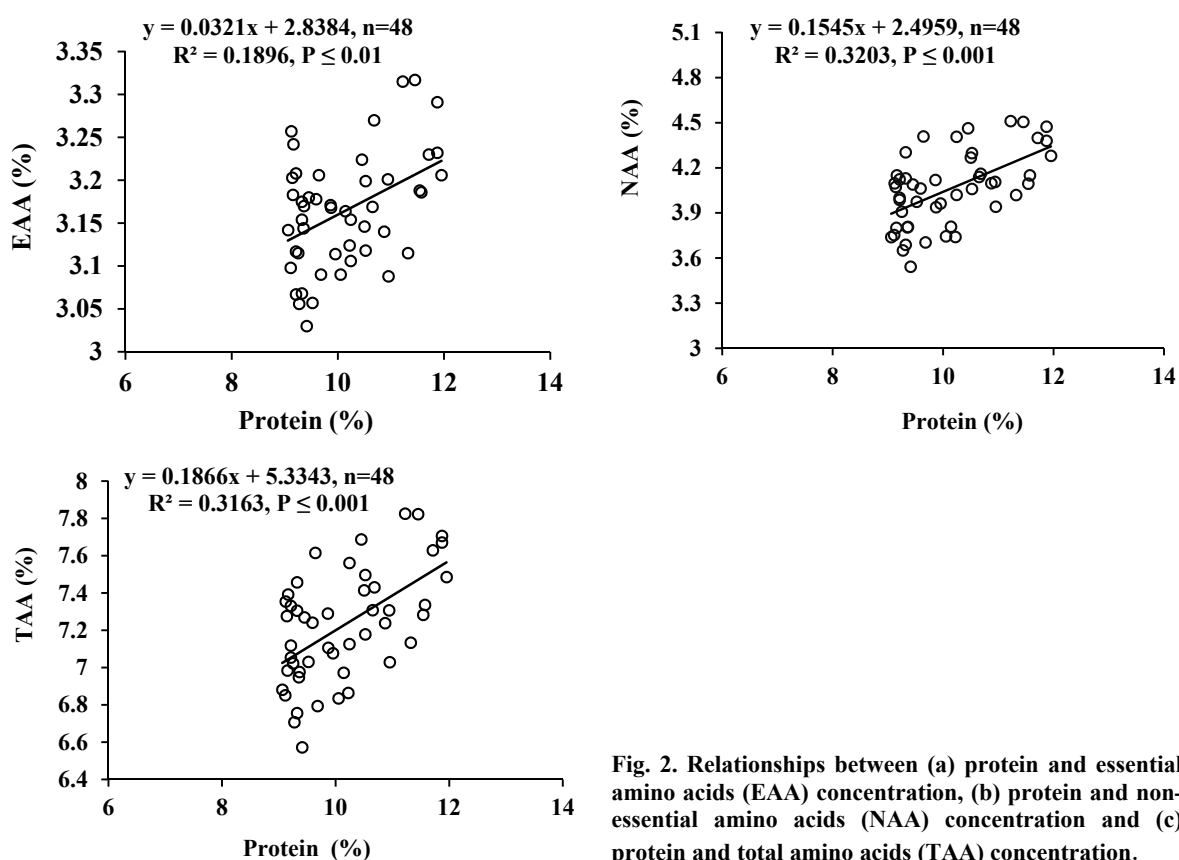


Fig. 2. Relationships between (a) protein and essential amino acids (EAA) concentration, (b) protein and non-essential amino acids (NAA) concentration and (c) protein and total amino acids (TAA) concentration.