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EFFECT OF NANOFERTILIZERS AND BIOFERTILIZERS ON YIELD OF MAIZE: BIPLOT ANALYSIS

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Abstract

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One of the newest and most interesting fields of science is nanotechnology, which is exploiting many areas such as life sciences; however, its application in agriculture is rarely studied. In order to examine the environment-friendly fertilizers (nano-sized and biological fertilizers), and control the agricultural nonpoint source pollution from the source, a field experiment was arranged to study the effects of bulk fertilizers (NPK, nitrogen biofer-tilizer, and phosphorus biofertilizer), nanofertilizers (nanoboron, nanozinc and nanocomplete) and no fertilizer treatment (control) on morphological traits of maize. The first two Principal Components (PCs) were used to create a biplot, which accounted for 72% of the variance of the treatment × trait interaction. The nanozinc fertilizers following nanoboron fertilizer were the best in most of the biological yield, seed yield, harvest index, and 100-grain weight. The vector-view biplot revealed a strong positive association between chlorophyll and protein content, seed yield with the number of kernels per ears and the number of rows per kernel. Nanocomplete fertilizer was the best treatment for chlorophyll content, protein percent and straw yield, while NPK did not high increase in the most traits of maize. This investigation indicated that treatment × trait biplot can graphically show the interrelationships among traits and facilitate visual comparison of fertilizers. In conclusion, it was found that zinc and boron nanofertilizers increased the production of most of the traits in maize. This study indicated that nano types of fertilizers could promote the studied traits in maize plant.

Keywords: nanoboron, nanocomplete, nanozinc, treatment × trait interaction.

INTRODUCTION

Maize (*Zea mays* L.) is one of the most important crops both in the food and the feed industry. This crop is frequently cultivated all around the world and environmental stresses may significantly affect its production (SoNG et al., 2010). Today, plants tolerant to the environmental stresses supply the food security in the world (MORADI et al., 2012). Meanwhile, selecting the tolerant genotypes due to the environmental interactions is really hard, which restricts the knowledge about the function and role of tolerance mechanisms (MESSMER et al., 2009). Stresses have negative effects on physiological indices such as photosynthesis, water use efficiency and leaf water content as well as season length, assimilate remobilization, canopy height, leaf area index and root growth in maize, which finally reduces the yield in arid and semiarid areas (EGILLA et al., 2005; PAYERO et al., 2006).

Yield component characteristics are affected and controlled by a complex of genes with different flexibility, whereas the response of maize yield depending on the severity of stresses, time and stage of occurrence vary up to 76% (FARRE et al., 2000). OKTEM (2008) has reported that fresh ear yield is reduced by about 40% due to reduction of kernel weight and quantity. Fertilization is one of the most effective factors. SZMIGIEL et al. (2013) have indicated the positive effects of mineral fertilization. The effectiveness of fertilization depends on the environmental conditions such as climatic factors or environmental stresses. The general fertilizer recommended for maize is NPK (nitrogen, phosphorus and potassium), but most farmers in semiarid areas are not endowed to invest on fertilizer application.

In plants, zinc and boron play important roles in various metabolic processes, whereas zinc as an integral component of many enzyme structures is known as an essential nutrient required for plant growth (AULD, 2001). Boron also is needed for plant growth at a normal dose as it is generally toxic at levels slightly above (CLARK et al., 1999). This microelement improves the quality of maize and its availability decrease in soils having high pH (ISMAIL, 2003). Micronutrients are important elements in fertilizers, but considering the energy needed in their synthesis, the fertilizers have a high monetary value. For increasing the efficiency of fertilizer utilization, the emerging nanostrategies as nanofertilizers are expected to be far more effective than conventional bulk fertilizers (DE ROSA et al., 2010). Nanofertilizers have high surface area and significantly improve physico-chemical and biological properties of soil (TARAFDAR et al., 2014). Despite the information available on foliar application of some nanomicronutrient fertilizers on some crops, there is less sufficient information about efficiency of nanofertilizers. Therefore, this study aimed to evaluate the impact of biofertilizers and nanofertilizers on yield, its components and some morphological traits of maize.

MATERIALS AND METHODS

Field experiment

The experiment was conducted in the field of the Department of Plant Genetics and Production, Faculty of Agriculture of Maragheh University. The location (46°16' E, 37°24' N, altitude: 1477 m) is characterized by semiarid climate, rainfall occurring in spring from April to May. All plots were sowed by hand and desired plant population was maintained by thinning the extra plants. Maize was sowed 65 cm apart and within-row spacing of 20 cm and weed control was regularly done by hand. The deficit irrigation (soil water content at field capacity is 25%) was performed during the initiation of reproductive growth until maturity stage. The area is a well-drained clay loam soil, at least 100 cm depth; soil water content, after the soil was saturated and allowed to drain freely for about 24 to 48 hours (field capacity), was at 33% and wilting point at 16% by volume for the surface to 100-cm soil layer. The clay loam pH 7.5, overlying heavy clay, containing 0.03% N, 0.01% P and 0.02% K.

Fertilizer treatments were as follows: nofertilizer was used as control (NF), biofertilizer of nitrogen (BioN), biofertilizer of phosphorous (BioP), nanochelated boron (NanoB), nanochelated zinc (NanoZn), complete nanofertilizer (NanoC) (all nanofertilizers were used in oxide form), and chemical fertilizer (NPK). The NPK fertilizers were 180 kg N ha⁻¹, 100 kg P_2O_5 ha⁻¹, and 50 kg K_2O ha⁻¹ in form of urea, super phosphate and potassium sulphate, respectively. NPK fertilizer was applied in two stages before sowing and top dressing one month after sowing. Biofertilizers were applied as seed inoculation just before planting. Nano-chelate fertilizers were applied three times by foliar spry at concentration of 2000 ppm at nine-leaf stage, stem elongation, heading. Synthesized nanoparticles were obtained from the Fanavar Sepehr Parmis Company, Iran. Weed control was carried out manually twice: 20 and 40 days after the sowing date, respectively. All necessary cultural practices were also carried out regularly.

Measurement of traits

Ten random plants were harvested from each plot for determination of leaf area (LA), and were divided into leaf, stem, cobs and pods as well. At the final harvest, three rows of maize four meters length were harvested from the centre of each plot for determination of seed yield (SY), straw yield (STY) and harvest index (HI). Also, these traits were measured from ten random plants: plant height (PH), hundred-grain weight (HGW), number of kernels per ear (NKE), number of rows per ear (NRE), ear length (EL), stem diameter (STD) and leaf area (LA).

Protein content (PRO) and oil percent (OIL) of seed were measured using a near-infrared seed analyser (Zeltex zx-50). Chlorophyll content (CHL) was measured as well on ten plant leaves per plot, using the Minolta SPAD-502 in fully expanded upper leaves at the flowering stage. Relative water content (RWC) was measured in leaves adjusted to ear at the beginning of grain development stage. The leaves were placed in polythene bags and transported to the laboratory as quickly as possible in order to minimize water losses due to evaporation. The samples were also weighed immediately as fresh weight (FW), then sliced into 2 cm sections and floated on distilled water for 4 h. The turgid leaf discs were then rapidly blotted to remove surface water and weighed to obtain turgid weight (TW). The leaf discs were dried in the oven at 60°C for 24 h and then dry weight (DW) obtained. The RWC was calculated by the formula given by BARRS (1968):

RWC (%) = [FWDW) / (TW-DW] * 100

The biplots were generated using GGE biplot software as the standardized values of the trait averages (YAN, 2001) and via the treatment \times trait biplot equation as follows:

$$\frac{\alpha_{ij} - \beta_j}{\sigma_j} = \sum_{n=1}^2 \lambda_n \xi_{in} \eta_{jn} + \varepsilon_{ij} = \sum_{n=1}^2 \xi_{in}^* \eta_{jn}^* + \varepsilon_{ij}$$

where α_{ij} is the mean value of treatment *i* for trait *j*, β_j is the mean value of all treatments in trait *j*, σ_{ij} is the standard deviation of trait *j* among the treatment means, λ_n is the singular value for principal component n (PCn), ξ_{in} and η_{jn} are scores for treatment *i* and trait *j* on PCn, respectively, and ε_{ij} is the residual associated with treatment *i* in trait *j*.

RESULTS AND DISCUSSION

The treatment \times trait biplot of mean performance of maize genotype across different fertilizer treatments explained 72% of the total variance (PC1 accounted 45% and PC2 27%) of the standardized data set (Fig. 1). The relatively high percentage variation indicates the simple relationships among the meas-



Fig. 1. Polygon view of treatment × trait biplot showing the effect of fertilization treatment on trait values. Fertilization treatments: control (NF), biofertilizer of nitrogen (BioN), biofertilizer of phosphorus (BioP), nanochelated boron (NanoB), nanochelated zinc (NanoZn), complete nanofertilizer (NanoC), and NPK. Traits: seed yield (SY), straw yield (STY), harvest index (HI), plant height (PH), hundred-grain weight (HGW), number of kernels per ear (NKE), number of rows per ear (NRE), ear length (EL), stem diameter (STD), leaf area (LA), protein content (PRO), oil percent (OIL), chlorophyll content (CHL), relative water content (RWC).

ured traits across different fertilizer treatments (YAN & RAJCAN, 2002; KAYA et al., 2006). According to SABAGHNIA & JANMOHAMMADI (2014), the fundamental patterns among the various traits should be captured by the biplots and the criterion of success of the treatment × trait biplot model is the identification of the first two principal component (PC) axes in the model. However, in accordance with SABAGHNIA et al. (2015), ADMASSU et al. (2008) have proposed two PC axes for any two-way dataset analysis like treatment × trait biplot model, which were sufficient for predictive model, thus, the interaction of seven fertilizer treatments with fourteen measured traits of this study was well estimated by the first two interaction PCs of fertilizer treatments and measured traits.

Polygon-view biplot is showing the treatment \times trait analysis on the traits based on first two PC scores (Fig. 1). The traits were considered as entries for the tester and the treatments, and this figure shows, which treatment(s) were the best in an individual trait. The treatment(s) for each vertex of the polygon in the biplot were the best or the worst in terms of the characteristics found in the sector by any two lines that meet at the origin of the polygon. According to Fig. 1, nanozinc fertilizer (NanoZn) treatment was the best in terms of seed yield, harvest index, hundred-grain weight, the number of kernels per ear, the number of rows per ear, ear length, stem diameter, leaf area, relative water content, indicating that it can be used as the best fertilizer in the maize production that are outstanding in these traits. KANWAL et al. (2010) have demonstrated that application of Zn can improve maize grain yield as well as other yield components and morphological traits. Also, according to POTARZYCKI (2011), grain vield of maize increases with Zn fertilization and its application significantly increase both total nitrogen uptake and grain yield performance. Based on Fig. 1, nanocomplete fertilizer was the best fertilizer treatment for chlorophyll content, protein percent and straw yield, while biofertilizer nitrogen was the best fertilizer treatment for oil percent, and NPK was the best treatment for plant height. Even though both of nanofertilizers (NanoZn following to NanoB) were identified for good yield and yield components traits, although they were not the best for some important traits such as oil and protein percentages, and estimated that the yield property characters for oil and

protein percentages may not be good. According to KIANI et al. (2013), nitrogen biofertilizer improves oil percent, but the application of phosphorus biofertilizer increases protein content of maize, which is in good agreement with our findings.

In the treatment × trait biplot vector view, a vector is drawn to each trait marker from the biplot origin to facilitate the visualization of the interrelationships and those between the traits. Provided that a sufficient amount of total variation was explained by the biplot, the correlation coefficient between any two traits is approximated by the angle cosine between their vectors (YAN & RAJCAN, 2002). As biplot showed, the largest variation resulted from all of the measured traits except plant height as indicated by their vector's relative length (Fig. 2).

The most prominent interrelationships revealed by this biplot were: (i) a strong positive association between CHL and PRO, between STY, EL and LA, and between NKE, NRE and SY as indicated by the angles between their vectors; (ii) a near zero correlation between OIL and STD, as well as CHL and PRO with NKE, NRE and SY, which was shown by the near perpendicular vectors; and (iii) large obtuse angles, which showed a negative association between OIL with CHL and PRO (Fig. 2). Although most of the current predictions above can be verified by the Pearson correlation coefficients (Table 1), but some others are not consistent with the original correlation coefficients, and these discrepancies are observed, because the treatment × trait biplot method explained less than 100% (in this study, 72%) of the total variation.

Although all of the above conclusions have some minor errors, treatment × trait biplot shows predictions of the general pattern of the entire dataset and the predictions are probably more reliable than the individual observations (YAN & RAJCAN, 2002). Significant positive correlation between yield and kernel weight has also been reported by SRECKOV et al. (2010). KUMAR et al. (2011) have reported that grain yield has highly significant positive correlation with ten traits viz., the number of kernels per row, cob length, cob girth and 100-seed weight, plant height, leaf area index, tassel length, the number of branches per tassel, the number of kernel rows per ear and chlorophyll content, and similar results also have been reported for the number of kernels per row and 100seed weight (CHINNADURAI & NAGARAJAN, 2011).



Fig. 2. Vector view of treatment × trait biplot showing the relationship between measured traits under different fertilizer treatments. Fertilization treatments: control, (NF), biofertilizer of nitrogen (BioN), biofertilizer of phosphorus (BioP), nanochelated boron (NanoB), nanochelated zinc (NanoZn), complete nanofertilizer (NanoC), and NPK. Traits: seed yield (SY), straw yield (STY), harvest index (HI), plant height (PH), hundred-grain weight (HGW), number of kernels per ear (NKE), number of rows per ear (NRE), ear length (EL), stem diameter (STD), leaf area (LA), protein content (PRO), oil percent (OIL), chlorophyll content (CHL), relative water content (RWC).

	PH	HGW	NKE	NRE	CHL	EL	RWC	STD	LA	STY	SY	HI	PRO
HGW	-0.03												
NKE	0.11	0.58											
NRE	0.14	0.41	0.89										
CHL	-0.22	-0.46	0.21	0.05									
EL	-0.47	-0.11	0.50	0.37	0.82								
RWC	-0.58	0.24	0.51	0.36	0.56	0.87							
STD	-0.29	0.33	0.87	0.83	0.40	0.76	0.70						
LA	0.12	-0.15	0.64	0.72	0.55	0.51	0.22	0.68					
STY	0.23	-0.28	0.55	0.51	0.70	0.54	0.15	0.56	0.93				
SY	-0.07	0.76	0.95	0.78	0.12	0.46	0.60	0.82	0.44	0.32			
HI	-0.13	0.91	0.81	0.63	-0.12	0.28	0.56	0.65	0.14	0.00	0.94		
PRO	0.14	-0.19	0.18	-0.07	0.71	0.58	0.52	0.13	0.09	0.30	0.14	0.06	
OIL	-0.16	0.51	0.04	0.29	-0.70	-0.41	-0.02	-0.01	-0.21	-0.56	0.17	0.37	-0.60

Table 1. Pearson's simple correlation coefficients between the studied traits of maize

Traits: seed yield (SY), straw yield (STY), harvest index (HI), plant height (PH), hundred-grain weight (HGW), number of kernels per ear (NKE), number of rows per ear (NRE), ear length (EL), stem diameter (STD), leaf area (LA), protein content (PRO), oil percent (OIL), chlorophyll content (CHL), relative water content (RWC).

An ideal treatment is defined as the treatment that combines several good features in its performance in order to identify an ideal treatment and in the context of treatment analysis. In the biplot shown in Fig. 3, the single-arrow line passing through the biplot origin is referred to the average-tester axis abscissa, on which the treatments are classified according to their performance characteristics and the doublearrow line (average-tester axis ordinate) divides the average-tester axis abscissa into two at the middle axis (YAN et al., 2007). The portion of the averagetester axis towards the right shows the above average treatments, and to the left shows below average treatments based on this biplot (Fig. 3). The treatments performed above average were as follows: NanoZn, NanoB, and BioP; while NanoC, BioN, NF and NPK performed below average in terms of traits.

NPK poor performance in this study is in line with POTARZYCKI (2011) findings, who has reported micronutrient (like zinc) application performed better than NPK. An ideal treatment should have the highest mean performance across traits (i.e. the longest projection onto the average tester axis (average-tester axis abscissa) and the shortest entry-vector, so, it should be close to the ideal treatment represented by the innermost focused circle with an arrow pointing to it (YAN & KANG, 2003). Therefore, such ideal treatment can be used as a reference check in subsequent tests, where the set of morphological characteristics will be measured. According to Fig. 3, nanoZn and nanoB treatments are the closest to the position of an ideal treatment.

It is classified as the highest in terms of morphological performance, because it is desirable for most of the characteristics, and these treatments could serve as good fertilizer requirements of which better than conventional fertilizer application are such as NPK or other macro-particle fertilizes. This result is in line with the report of SABAGHNIA (2015), who has mentioned that nanoFe application is the most superior fertilizer in many agronomic and yield trait performances of lentil, when evaluated in a field trial.

Treatments suitable for obtaining good seed yield of maize could be seen in the biplot of Fig. 4, which is a vector-view function of treatment \times trait biplot model and shows treatments that are closely related to the target characteristic among other characteristics.



Fig. 3. Ideal fertilizer treatment view of treatment × trait biplot and comparison of the fertilizer treatments with the ideal fertilizer treatment. Fertilization treatments: control (NF), biofertilizer of nitrogen (BioN), biofertilizer of phosphorus (BioP), nanochelated boron (NanoB), nanochelated zinc (NanoZn), complete nanofertilizer (NanoC), and NPK.

According to this biplot, nanoZn and nanoB were identified as fertilizer treatments suitable for obtaining high grain yield and application of above treatments is expected to lead to improved grain yield. This revealed that using nano-sized micronutrient fertilizers will not only result in the development of high seed yield, but also with other desirable agronomic traits as well as yield components that enhance wide use of such treatments. BALA et al. (2014) have shown beneficial role of nanofertilizer application in seed germination and plant growth in chickpea due to increase in the activity of growth hormone gibberellin; and AMIRNIA et al. (2014) have highlighted the positive effects of nanofertilizers (Fe, P and K) on saffron feature improvement. RICO et al. (2011) have stated that nanotechnology has positive effect on plants, including the increase of percentage and rate of germination as well as root and shoot lengths in many crops. LIU et al. (2010) have indicated that nanocomposites could be safe for wheat seed germination, seedling development and growth, and also conclude that use of nano-sized fertilizers is useful in crop production beside the economic benefits.

Several studies have shown that the use of nanofertilizers increase nutrient use efficiency, reduce soil toxicity, minimize the potential negative effects associated with over dosage and reduce the application frequency, because nanotechnology has a high potential for sustainable agriculture, particularly in developing countries (Naderi & Danesh-Shahraki, 2013). The advent of nanotechnology has provided a wealth of various engineered nanoparticles with new physical, chemical and biological features, while one of these new facilities is the encapsulation of fertilizers in a nanoparticle, which is done in some ways: (1) encapsulation inside nanoporous, (2) coated with thin polymer film, and (3) delivering as emulsion of nanoscale dimensions (CHEN et al., 2012; RAI et al., 2012). Nano-fertilizers will combine nano-devices to synchronize fertilizer release and crop uptake and prevent undesirable nutrient losses to the soil (DE Rosa et al., 2010). Undoubtedly, treatment \times trait biplot model is an excellent tool for visual data analysis, and this approach has some advantages compared to conventional data analysis methods: (i) Graphical presentation enhancing the ability to understand data



Fig. 4. Vector view of treatment × trait biplot showing the relationships of different fertilizer treatments with target trait seed yield. Fertilization treatments: control (NF), biofertilizer of nitrogen (BioN), biofertilizer of phosphorus (BioP), nanochelated boron (NanoB), nanochelated zinc (NanoZn), complete nanofertilizer (NanoC), and NPK.

set patterns, (ii) more interpretative, facilitating pairspecific comparisons of treatments. A potential limitation of the biplot method is that it may not explain most of the variation in some cases, so it does not display all data patterns. Even if this is the case, the biplot of the first two main components can still display the data's most important patterns. Even when this is the case, it is possible to ensure that the biplot of the first two main components still shows the most important data patterns.

CONCLUSION

Our findings indicated significant differences among various nano-, bio- and bulk fertilizers in terms of yield, yield components and other traits of maize. It was found that nanozinc and nanoboron compared to other fertilizers increased the productivity of the most traits in maize. Therefore, a suitable choice of nanofertilizer can be considered the most crucial factor in maize farm management.

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NANOTRĄŠŲ IR BIOTRĄŠŲ POVEIKIS KUKURŪZŲ DERLIUI: BIPLOT ANALIZĖ

Mojtaba Nouraein

Santrauka

Viena iš naujausių mokslo taikymo sričių yra nanotechnologijos, kurios plačiai naudojamos gyvybės mokslų srityje, tačiau jų taikymas žemės ūkyje yra ribotas. Siekiant ištirti aplinkai nekenksmingas nano ir biologines trąšas, lauko eksperimentiniu metodu buvo nustatyta birių (NPK ir biotrąšų) ir nanotrąšų (nanoboro, nanocinko bei nanotrąšų komplekso) įtaka kukurūzų derliaus savybėms. Pagrindinių komponenčių analizės *biplot* diagramos atskleidė, kad didžiausią įtaką biologiniam ir sėklų derliui, derliaus indeksui bei grūdų masei turėjo nanocinko ir nanoboro trąšos. *Biplot* analizė parodė, kad nanotrąšos turėjo teigiamą poveikį chlorofilo ir baltymų kiekiui bei šiaudų derliui, o NPK trąšos neturėjo reikšmės daugumai tirtų kukurūzų derliaus savybių. Apibendrinant, buvo nustatyta, kad cinko ir boro nanotrąšos pagerino kukurūzų derliaus savybes. Šis tyrimas parodė, kad *biplot* analizė gali būti naudojama trąšų ir derliaus komponentų koreliacinių ryšių grafinei išraiškai, palengvinančiai tiriamų trąšų palyginimą.