



Original Article

Effect of Foliar Application of Micronutrients on Antioxidants and Pungency in Onion

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Abstract

The aim of the present work was to study the effect of foliar application of micronutrients [Zinc: $Zn_1(0\%)$; $Zn_2(0.5\%)$; $Zn_3(1.0\%)$ and Boron: $B_1(0\%)$; $B_2(0.25\%)$; $B_3(0.5\%)$] on the antioxidants and pungency of onion cv. "Sukhsagar" (*Allium cepa* L.). In this experiment, it was suggested that the highest contents of total and free phenol was obtained by the highest dose of Zn (1%) in combination with 0% B, and single dose of Zn (0.5%) in combination with 0% B respectively. The superoxide dismutase (SOD) activity was found to increase with respect to control following the highest dose of B (0.5%) alone. However, the peroxidase (POD) activity increased more with respect to control following the single dose of B (0.25%) rather than the double dose (0.5%). The highest Molybdate reducing antioxidant potential (MRAP) was observed in 0.5% Zn in combination with 0% B, whereas that of 2, 2-Diphenyl- β -picrylhydrazyl radical scavenging activity (DPPHRAC) was found in the interaction effect of the double doses of both Zn (1%) and B (0.5%), which also offered the lowest lipid peroxidation. The highest pyruvic acid development was observed by the interaction effect of 0% Zn and 0.25% B. Based on the average values of the biochemical parameters and the results of PCA, the treatment with Zn_1B_3 (0% of Zn in combination with double dose of 0.5%) of B) was proved to be most promising with respect to antioxidant properties.

Keywords: Allium cepa L., antioxidants, foliar application, micronutrients, pyruvic acid

Introduction

From time immemorial, onion (*Allium cepa* L.) has been used as common food and also for the treatment of many diseases. Apart from providing basic nutrition, onion is well known for its health benefits; numerous therapeutic properties have been reported in onion viz., anticarcinogenic, antibiotic, antibacterial, antifungal and antioxidant properties (Benkeblia, 2005). Antioxidants rich food play an important role in prevention of cardiovascular diseases and cancers (Garber *et al.*, 2002), neurodegenerative diseases (Di Matteo and Esposito, 2003), as well as prevention of inflammation and problems causes by cell and cutaneous aging (Ames *et al.*, 1993).

Micronutrients have received a great deal of importance in crop production during recent years because of the widespread occurrences of their deficiencies from different parts of the country. Researchers from almost all the states in the country have also reported significant responses of many crops to micronutrient fertilization. Zinc (Zn) and boron (B) is an essential trace element for plants, being involved in many enzymatic reactions and is necessary for their good growth and development. Some studies estimates indicate that a large number of diverse materials can serve as sources of plant nutrients. The majority of nutrient input to agriculture comes from commercial mineral fertilizers. Mineral fertilizers are considered to play a significant but lesser role in nutrient contribution, leaving aside their beneficial effects on soil physicochemical and biological properties. Foliar feeding is a relatively new and controversial technique of feeding plants by applying liquid fertilizer directly to their leaves.

Zinc (Zn) is one of the essential micronutrients playing a significant role in many vital metabolic processes (Rout and Das, 2003; Aravind and Prasad, 2005a; Aravind and Prasad, 2005b). An increase in the level of reactive O_2 species (ROS) may appear in Zn-deficient plants. Zn deficiency enhances O2. generation by enhancing NADPH-dependent oxidase activity (Cakmak, 2000). Moreover, as it is an integral constituent of Cu/Zn superoxide dismutase (Cu/Zn SOD), Zn play an important role in the detoxification of the $O_2^{\bullet-}$ (Apel and Hirt, 2004). Besides Zn deficiency, increased in activity of POD (Kosesakal and Unal, 2009). Hajiboland and Amirazad (2010) also reported that under deficiency of Zn caused the activities of POX and CAT to decrease. Excess Zn can also affect the uptake of other nutrient elements. Thus, deficiency of the other elements may cause oxidative stress (del-Rio et al., 1991; Bonnet et al., 2000). These oxygen species are highly reactive

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and cause the death of plants by damaging membrane lipids, proteins, pigments and nucleic acids (Chaoui *et al.*, 1997; Weckx and Clijsters, 1997; Bonnet *et al.*, 2000; Cuypers *et al.*, 2002).

There is evidence that B is one of the nutrients responsible for the changes in concentration and a number of metabolic pathways such as carbohydrate metabolism, nitrogen metabolism, phenol metabolism and ascorbate metabolism in plants (Marschner, 1995; Dordas and Brown, 2005; Luaszewski and Blevins, 1996). In fact, it is well known that B deficiency causes an accumulation of phenolics through the stimulation of the enzyme phenylalanine-ammonium lyase (PAL) (Cakmak et al., 1995; Ruiz et al., 1998b; Camacho-Cristobal et al., 2002). Other reports have shown that B deficiency not only induced quantitative changes but also qualitative changes in the phenolic pool of plants (Camacho-Cristobal et al., 2002 Camacho-Cristobal et al., 2004; Karioti et al., 2006). However, B deprivation also increased the activity of polyphenol oxidase (PPO) (Pfeffer et al., 1998; Camacho-Cristobal et al., 2002), enzyme that catalyses the oxidation of phenolic compounds into quinones. Besides yield increase a relationship also exists between vitamin C content and boron treatment in different vegetables like summer squash, beet, tomato and potato (Luaszewski and Blevins, 1996; Govindan et al., 1950; Mondy and Mucshi, 1993).

Aerobic organisms are constantly exposed to one or more systems that generate reactive oxygen species (ROS), including the superoxide radical anion (O2.-), hydroxyl radical (OH), hydrogen peroxide (H_2O_2) , various peroxyl radicals (ROO[•]), and singlet oxygen $(^1O_2)$. These ROS are highly reactive and can damage living cells if formed in significant amounts. To avoid cellular damage by ROS, most biological systems have developed defense mechanisms, antioxidants that convert ROS to unreactive derivatives. Recently, Denre et al. (2011) reported that the onion is rich in antioxidants. The authors also concluded that the green pungent pepper contains a few chain breaking antioxidants (e.g. Vitamin C and phenolics etc. that scavenge oxygen radicals and thereby break radical chain sequences) and few preventive antioxidants (e.g. SOD, POD and CAT etc. which prevent or inhibit the formation of ROS). Therefore, the aim of the present study was to investigate the effects of foliar application of micronutrients (Zn and B) with respect to level of antioxidant constituents and antioxidant activities along with pyruvic acid development of onion in order to identify the best treatment that could be used to develop improved quality.

Materials and Methods

Field experiment

The seedling was grown in nursery beds prepared in a sandy loam soil. The beds were 1.5 m long and 1.0 m wide. Weathered cow dung manure, 4 kg m², was mixed into the beds. Beds were drenched with formaldehyde (4%) and covered with polythene sheets for ten days to avoid damping off disease. Seedling was treated with Thiram (3 g kg⁻¹ of seed) (Gujarat Pesticides Private Limited, Gandhinagar, Gujarat, India) prior to sowing. Onion seeds of the cv. 'Sukhsagar' the most popular cultivar in West Bengal was obtained from AICRP on Vegetable Crops and were sown at 0.5 cm depth and 5 cm apart and covered with finely sieved leaf mold. After sowing, beds were covered with straw until germination and hand watered regularly. Seedlings were raised as before and hardened by withholding water 4 days before transplanting.

The main field was of sandy loam soil. Land preparation was done with 3-4 ploughings and beds were prepared. Fertilizer at the rate of 60N-60P-60K kg ha-1 was applied before transplanting and incorporated well into the soil. The source of nitrogen (N) was urea, that of phosphate (P) was single superphosphate and Potash (K) was from muriate of potash. Additional nitrogen fertilizer at the rate of 60 N kg ha-1 was applied 21 days after transplanting. The 40 days old seedlings were transplanted on muddy flat surface soil in 5 cm deep furrows with 15 cm spacing between rows and 10 cm within plants in plots. The plot size was 2.25 m \times 2.0 m. The experiment consisting of nine treatments including control (only water sprayed) were arranged in a factorial randomized block design with three replications of each treatment at the In-check Research Farm, Bidhan Chandra Krishi Viswavidyalaya, Kalyani, Nadia, West Bengal, India. The detail treatments are summarized in Table 1.

As because onion is a shallow rooted crop, supplementary irrigations (12 weekly irrigations providing approximately 5 cm of water were applied with the first being before transplanting) were given. There were four light hoeings within 9 weeks of transplanting. The application of fungicide Chlorothalonil at 2 g L^{-1} with sticker Sandovit at 1 ml L^{-1} was applied four to five times beginning 15 days after transplanting and at 15 days intervals to control purple blotch disease [*Alternaria porri* (Ellis) Cif.].

Onion bulbs from each plot were harvested at 127-128 days after transplanting (when more than 80 % plant leaves were dried). Sample (15-20 pieces of bulbs) from each plot was collected after harvesting. Fresh materials were washed, dried with soft tissue, and chopped with a sharp knife into small pieces before analysis of the content of non-enzymatic antioxidants, activity of enzymatic antioxidants and antioxidant activity under different systems of assay [molybdate reducing antioxidant potential (MRAP), DPPH radical scavenging activity and lipid peroxidation] as well as pyruvic acid development (PAD) which is a measure of pungency.

Chemical analysis

Non-enzymatic antioxidants

Analysis of ascorbic acid content (AAC)

One gram of finely chopped onion tissue was extracted with 20 ml of 4% oxalic acid following maceration in a pestle and mortar and the material centrifuged for 30 min at 10,000 g. Ascorbic acid content was determined using the dichlorophenol indophenol titration procedure (Casanas *et al.*, 2002).

Analysis of total phenol content (TPC) and free phenol content (FPC)

Total phenol was extracted in 50% methanolic 1.2 N HCl and Free phenol was extracted in 50% aqueous methanol by boiling one gram of finely chopped tissue for 1.5 h at 80-90 °C following the method of Vinson *et al.* (1995) and subsequent analysis was with the Folin-Ciocalteau reagent using Gallic acid as standard.

Enzymatic antioxidants

Analysis of super oxide dismutase activity (SOD)

One gram finely chopped bulb tissue extracts to inhibit photochemical reduction of nitroblue tetrazolium (NBT) in riboflavin light NBT system (Beauchamp and Fridovich, 1971)

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Table 1. Different doses of micronutrients (Zn and B) used as foliar spray for onion							
Treatments	Zinc (Zn) (%)	Boron (B) (%)					
$T_1(Zn_1B_1)$	0.0	0.0					
$T_2(Zn_1B_2)$	0.0	0.25					
$T_3(Zn_1B_3)$	0.0	0.5					
$T_4(Zn_2B_1)$	0.5	0.0					
$T_5(Zn_2B_2)$	0.5	0.25					
$T_6(Zn_2B_3)$	0.5	0.5					
$T_7 (Zn_3B_1)$	1.0	0.0					
$T_8(Zn_3B_2)$	1.0	0.25					
$T_9 (Zn_3B_3)$	1.0	0.5					
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Where, the source of Zn was supplied from ZnSO2.7H2O, and B from Na2B4O7.10H2O. Each spraying was done four times with sticker starting from 30 days after transplanting and subsequent ones at an interval of 10 days during vegetative stage.

was used to determine SOD activity. Absorbance at 560 nm was recorded and percentage inhibitory activity was calculated as $[(A_0-A_c)/A_0] \times 100$ where A_0 = absorbance without extract and A_c = absorbance with extract.

Analysis of peroxidase activity (POD)

One gram of freshly harvested onion tissue from each treatment was macerated in a pestle and mortar and extracted with 10 ml tris-HCL buffer (pH 7.6) to determine POD activity. Triturated samples were centrifuged at 10,000 g for 30 min at 4°C and supernatants were assessed for enzyme activity. The POD was estimated by mixing 0.1 ml chilled enzyme extract with 2.8 ml reaction mixture (0.5% o-dianisidine dissolved in methanol, 0.28 ml Na-acetate buffer, 2.4 ml water). The reaction was initiated by adding 0.1 ml H₂O₂ (30%). Changes in absorbance at 430 nm were monitored at 1 min intervals for up to 3 min. POD activity was measured using *o*-dianisidine as the substrate (Bhattacharya *et al.*, 2010) and expressed as mM o-dianisidine oxidized•min⁻¹•g⁻¹ of fresh tissue using the extinction coefficient 1.13×10^4 •M⁻¹•cm⁻¹.

Analysis of catalase activity (CAT)

An assay mixture of 3 ml of phosphate buffer, 2 ml of H_2O_2 , and 1 ml of enzyme source were pipette into a porcelain crucible and incubated for 1 min at 20°C to estimate CAT activity. The reaction was stopped with addition of 10 ml of 0.7 N H_2SO_4 and the reaction mixture titrated against 0.01 N KMnO4 to determine residual H_2O_2 until a faint pink color that persisted for at least 15 s. One unit of CAT activity was defined as amount of enzyme that destroyed 1 µmol of H_2O_2 •min⁻¹•g⁻¹ of fresh tissue. Changes in activity were measured using the method of Kar and Mishra (1976).

Antioxidant activity

Analysis of total antioxidant activity

A 0.5 gram freshly chopped onion tissue was extracted by macerating with 10 ml distilled water for estimation of total antioxidant activity. Tubes containing extract and reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate, and 4 mM ammonium molybdate) were incubated at 95 °C for 90 min. After incubation the mixture cooled to room temperature, absorbance of each solution was measured at 695 nm against a blank (Prieto *et al.*, 1999). Antioxidant capacity was expressed as gallic acid (mg/g) equivalent.

Analysis of 2, 2-Diphenyl-β-picrylhydrazyl radical scavenging activity (DPPHRAC)

The scavenging effect of onion for stable 2, 2-diphenyl-β-

picrylhydrazyl (DPPH) radical was monitored according to the method of Braca *et al.* (2001). One gram of fresh, finely chopped onion tissue was extracted with 10 ml distilled water. A 0.2 ml aqueous extract was added to 6 ml of a 0.004% methanolic solution of DPPH. Absorbance at 517 nm was recorded after 30 min and percentage inhibitory activity was calculated as $[(A_0 - A_c)/A_0] \times 100$ where A_0 = absorbance without extract and A_c = absorbance with extract.

Analysis of lipid peroxidation (LP)

Lipid peroxidation was measured as the amount of malondialdehyde (MDA) produced by the thiobarbituric acid (TBA) reaction as described by Dhindsa *et al.* (1981). A one gram sample of chopped fresh onion tissue was homogenized in 2 ml of 20% trichloroacetic acid (TCA) followed by centrifugation at 5,000 g. A 20% TCA solution containing 0.5% TBA and 4% butylated hydroxytoluene was added to a 1 ml aliquot before heating at 95 °C for 30 min and then cooling in an ice bath. The contents were centrifuged at 10,000 g and absorbance read at 532 nm. The concentration of MDA was calculated using an extinction coefficient of 155•mM⁻¹•cm⁻¹.

Analysis of pyruvic acid development (PAD)

Pungency of onion bulbs was determined as pyruvic acid (Schwimmer and Weston, 1961; Anthon and Barrett, 2003). Pyruvic acid concentration was determined using the method of Schwimmer and Weston (1961). A one gram sample of each genotype was crushed in a pestle and mortar and incubated with 2, 4-dinitrophenylhydrazine and the absorbance read at 420 nm on a spectrophotometer for total pyruvic acid concentration, determined against a sodium pyruvate standard curve.

Statistical analyses

Data were subjected to ANOVA of a factorial randomized block design and by Duncan's multiple range tests, to determine differences among means. Principal component analysis (PCA), as the method of identifying the factor dimension of the data, was used to summarize the treatment informing in a reduced number of factors for selection of the best performing treatment. Statistical analyses were done using SPSS Professional Statistics ver. 7.5 (SPSS Inc., Irvine, California).

Results and Discussion

Non-enzymatic antioxidants Ascorbic acid and phenol

Boron application is known to elevate the ascorbate concentration in plant tissues as compared to B-deficient

conditions (Blevins and Lukazewski, 1998). Increased ascorbate concentration by foliar applications of B was reported in potato (Mondy and Munshi, 1993). Keles *et al.* (2004) observed a significant rise in ascorbate level in orange under conditions of excess B. In our experiment neither Zn nor B application could produce any significant effect on AAC (Table 2). However, AAC increased, though not significantly, in both Zn and B treatments alone than the untreated controls (Zn₁ and B₁). The highest AAC was obtained in the combination Zn₃B₂ followed by Zn₂B₃.

In contrast to AAC, the phenol contents were significantly different among the treatments. TPC, following Zn application alone, produced significantly higher value in Zn_3 compared to control (Zn_1). In case of FPC, Zn₂ alone produced significantly higher value as compared to Zn_1 and Zn_3 , both of which produced no significant differences between their corresponding values. In B treatments alone, TPC decreased with increasing doses of B producing significantly different values between B1 and B2. The values of TPC exhibited by B_2 and B_3 were not significantly different. A decreasing trend in FPC values with respect to control was also observed. The values of FPC significantly decreased in B_2 while it increased significantly in B_3 . However, the value of FPC in latter was quite less than the control (B_1) . The decrease in phenols effectuated by B application supports the fact that phenols accumulate under conditions of B deficiency (Marschner, 1995). The highest TPC was obtained in Zn₃B₁ whereas the highest FPC was obtained in Zn_2B_1 .

Enzymatic antioxidants SOD, POD and CAT

Among the antioxidative enzymes, SOD and CAT exhibited values, which are not significantly different. However, SOD activity increased in Zn₂ following a decrease in Zn3 with respect to control, whereas the trend was reverse in B_2 and B_3 respectively. There are reports in favour of increase in SOD activity following application of excessive B (Garcia et al., 2001; Molassiotis et al., 2006; Cervilla et al., 2007). In case of CAT both Zn and B treatments alone resulted in almost similar values and the same trend of increase in single dose followed by a decrease in the double dose. The treatments Zn₂B₁ and Zn₂B₂ exhibited the highest SOD and CAT activities respectively. Zn is supposed to increase SOD and CAT activity (Luo et al., 2010) in Jatropha seedlings upto certain concentrations of Zn. The CAT activity in response to B application varies according to plant species e.g., while increased B concentration reduced CAT activity in citrus (Keles et al., 2004), it induced the activity of this enzyme in sunflower (Dube et al., 2000), grapevine (Gunes et al., 2006) and tomato (Cervilla et al., 2007).

Single dose of Zn alone resulted in no significant differences in POD activity with respect to control, whereas the double dose (Zn_2) significantly decreased the POD activity, which is contrary to the observation that POD activity in Jatropha seedlings was induced gradually by increasing Zn concentrations (Luo *et al.*, 2010). In case of B treatments alone, the POD activity increased

significantly with respect to control, but more increment was observed in B_2 (single dose) rather than B_3 (double dose). This observation on POD activity conforms to the report of Molassiotis *et al.* (2006), who also noticed an increase in POD activity in leaves of apple rootstocks under high B.

Antioxidant activity

MRAP, DPPHRAC and LP

The MRAP values under Zn alone showed an increase in Zn₁ followed by a decrease in Zn₂ with respect to control. However, the differences were not significant. In case of B alone the values assumed a clear significantly decreasing trend with respect to control. A similar trend was observed in case of total phenol content, which reinforces the fact that phenols contribute largely to MRAP assay. The maximum value of MRAP was observed in Zn₂B₁.

The DPPHRAC assay revealed significantly increasing scavenging activity under different doses of Zn alone with respect to control. As because ascorbate contributes largely to DPPHRAC assay, a similar trend like AAC was observed in this assay. In case of B application alone, however, the scavenging activity decreased significantly in B_2 and increased significantly in B_3 . The highest DPPHRAC (lowest IC₅₀ value) was observed in the treatment Zn₃B₃.

The values of LP are actually the values of concentrations of MDA produced by lipid peroxidation. Under Zn treatments alone, the value of LP increased significantly in Zn₂, while that decreased significantly in Zn₃ than that of control Zn₁. In B application alone, there was no significant differences between the values of B₂ and B₃, but the values showed an increasing trend. However, these values were significantly lower than that of the control (B₁). Increase in MDA concentration in relation to excess B application was observed in apple rootstock (Molassiotis *et al.*, 2006), grape (Gunes *et al.*, 2006) and tomato (Cervilla *et al.*, 2007). The highest MDA concentration was exhibited by the combination Zn₂B₁.

Pungency

PAD

PAD, under Zn treatments alone, produced significantly lower values than the control, though there were no significant differences between the PAD values exhibited by Zn_2 and Zn_3 . Again, the B applications alone led to values with no significant differences between them. However, the values were higher than the control (B₁) and decreased in the double dose (B₃) than that of the single dose (B₂). The highest PAD was observed in the treatment Zn_1B_2 .

Principal component analysis (PCA)

In this experiment also PCA was used to summarize the treatment information in a reduced number of components, where a total of three components were chosen (PC1, PC2 and PC3) due to their Eigen value being greater than 1.0 and they together explained 76% of total variance (Table 3).

The first component alone explained 34% of total variance where all the variables other than TPC and SOD are positively loaded meaning that all other variables restrict TPC and SOD, in which the former is not desirable. The positive loading of DPPHRAC and LP is also not desirable. Therefore, on the basis of the first component, the treatments Zn_1B_2 , Zn_1B_3 , Zn_3B_1 etc. can

Treatment	AAC (mg/100 g)	TPC (mg/100 g GAE)	FPC (mg/100 g GAE)	SOD (IC50: mg/ml)	POD (mM <i>o</i> -dianisidine oxidized/g/min)	CAT (units)	MRAP (mg/g GAE)	DPPHR AC (IC50: mg/ml)	LP (nmol/ 100 g)	PAD (µmol/g)
Zn					0 /					
1	12.52	112.60b ^a	48.76b	5.12	0.33a	1.04	41.12	15.18a	54.56b	10.62a
2	13.47	113.35b	60.52a	4.80	0.33a	1.06	41.96	11.63b	60.00a	8.74b
3	13.71	123.13a	51.67b	5.00	0.29b	0.96	40.32	10.44c	45.89c	8.31b
В										
1	12.29	126.23a	68.24a	4.79	0.27c	1.03	48.95a	11.73b	58.11a	8.81
2	13.71	113.34b	42.78c	5.16	0.36a	1.08	37.49b	15.58a	50.78b	9.54
3	13.71	109.51b	49.92b	4.97	0.31b	0.96	36.96b	9.94c	51.56b	9.31
Zn ₁										
B ₁	11.34	122.46	49.67	5.04	0.14	0.98	31.88	12.18	49.67	8.04
B ₂	13.47	116.57	49.36	4.87	0.42	1.10	46.15	21.05	53.00	13.31
B ₃	12.76	98.76	47.24	5.44	0.43	1.06	45.32	12.32	61.00	10.51
Zn ₂										
B1	13.47	101.72	92.94	3.70	0.37	1.07	73.65	9.83	73.00	9.41
B ₂	12.05	118.23	40.84	5.40	0.45	1.11	24.53	14.73	54.33	7.42
B ₃	14.90	120.10	47.77	5.30	0.18	1.01	27.70	10.33	52.67	9.38
Zn ₃										
B1	12.05	154.51	62.11	5.62	0.31	1.06	41.32	13.18	51.67	8.99
B ₂	15.60	105.21	38.15	5.22	0.23	1.02	41.78	10.97	45.00	7.89
B ₃	13.47	109.66	54.75	4.16	0.33	0.80	37.84	7.17	41.00	8.04
Analysis of v	ariance (F valu	es)								
Zn	NS	4.968 [*]	14.444**	NS	16.384"	NS	NS	49.422 ^{**}	35.378**	26.650**
В	NS	11.053	66.387**	NS	60.946**	NS	8.551	67.497**	11.335"	NS

Zn X B	NS	18.420**	27.947**	16.389**	212.329"	NS	21.311"	23.529**	16.500**	24.093**
[*] cionificant at 50	** cionifica	stat 1% NS no	n cionificant							

*significant at 5%, **significant at 1%, NS - non significant "Values followed by the same letter in a column are not significant at *P* < 0.05, Duncan's Multiple Range Test (DMRT)

Table 3. Results of principal component analysis (PCA) for effect of Zn and B on antioxidants content and antioxidant activity along with pyruvic acid development in onion

Principle component	Eigen value	% v:	ariance	% Cumulative variance				
Eigen values and variance accounted for (%) by PCA based on correlation matrix								
1	3.40	3	4.01	34.01				
2	2.73	2	7.27	61.28				
3	1.43	1	4.33	75.61				
Variance		PC1	PC2	PC3				
Factor loadings due to PCs with Eigen values greater than 1								
Ascorbic acid		0.02	-0.34	0.77				
Total Phenol(GAE)		-0.40	0.36	-0.65				
Free Phenol(GAE)		0.80	-0.32	-0.45				
SOD		-0.64	0.63	-0.01				
POD		0.52	0.44	0.11				
CAT		0.36	0.78	-0.01				
MRAP(GAE)		0.91	-0.21	-0.01				
DPPHRAC		0.11	0.91	0.15				
LP		0.86	0.16	-0.19				
PAD		0.48	0.54	0.38				

be selected as having all desirable traits (Fig. 1).

PC2, on the other hand, explained another 27% of total variance in which AAC, FPC, and MRAP are negatively loaded as opposed by all other variables which are positively loaded. But positive loading of SOD, DPPHRAC, and LP is not desirable. So, considering PC2, the performing treatments

would be Zn_1B_3 , Zn_2B_3 and Zn_3B_2 .

PC3 explained an additional 14% of total variance in which AAC, POD, DPPHRAC and PAD restrict the values of TPC, FPC, and LP. Considering PC3 the performing treatment combinations are Zn_1B_3 , Zn_3B_3 , Zn_2B_2 (Fig. 2).



Fig. 1. Scatter diagram of the regression factor scores for the first and second components produced by PCA

Conclusions

Considering all the parameters and the results of three principal component analyses it can be suggested that the promising treatment combination for onion crop might be Zn_1B_3 followed by Zn_3B_3 , and Zn_2B_3 , which may bring about the proper value addition by enhancing the antioxidants content and antioxidant activity along with pungency development.

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Fig. 2. Scatter diagram of the regression factor scores for the first and third components produced by PCA

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