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The Influence of nano-TiO_2 and Nano-Silica Particles Effects on Yield and Morphological Traits of Sunflower

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Abstract: Present study is performed to evaluate the effects of foliar application of salicylic acid, glycine betaine, ascorbic acid, nano-silica and nano titanium dioxide on yield and yield component of sunflower. Chlorophyll content, leaf length, leaf width, days to 50 % flowering, day to maturity, plant height, husk percentage, number of seeds per head, head number per plant, percentage of empty achenes, 1000-seed weight, kernel weight, grain length, straw yield, harvest index, grain yield and oil percent were measured. Results showed that the first two principal components accounted 49 % and 19 %, respectively of sums of squares of the TT interaction and were used to create a two-dimensional treatment by trait (TT) biplot. The vertex treatments in polygon of TT biplot were ascorbic acid, glycine betaine, nano-TiO_2 and control which Nano-Ti_2 treatment indicated high performance in chlorophyll content, day to maturity, number of seeds per head, head number per plant, kernel weight, grain length, straw yield, harvest index, grain yield and oil percent. The identification of ideal treatment, the treatment that is most favorable treatment among all treatments, showed that the nano-TiO_2 might be used in selecting superior traits and it can be considered as the candidate treatment. Treatments suitable for obtaining of high seed yield were identified in the vector-view function of TT biplot and displayed nano-silica and nano titanium dioxide as the best treatments suitable for obtaining of high seed yield. In short, nano-fertilizer could increase crop yields and improve the fertilizer efficiency.

Keywords: nano-silicon dioxide, oil content, drought stress, TiO_2 nanoparticles

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Introduction

Sunflower (*Helianthus annuus* L.) is an oilseed crop is grown throughout the semiarid regions in many areas of the world and its oil represents a share of 7.3 % of the plant oil annual global production (Velasco *et al.*, 2015). Its annual production in Iran is about 40 thousand tones which achieved from 41 thousand hectares while the average yield is relatively low and it has been recorded about 970 kg ha^{-1} (FAOSTAT, 2016). It belonging to the Asteraceae, and characterized by a strong taproot, which enables it to thrive in dry climates and allows utilization of nutrients from below the root zone of cereals (Diepenbrock and Pasda, 1995). In semiarid areas, where the proportion of less fertile soils is high, it may be difficult to fulfill the nutritional needs of crops and fertilizer application represents a suitable tool to compensate nutrient deficiencies and to replace elements removed in the products harvested. The importance of oil crops such as sunflower has increased in recent years, especially with the interest in the production of biofuels because it is a crop which is well adapted to dry conditions (Weiss, 2000). Moreover, it is a well deep-rooted crop which can meet its water needs by exploring a big volume of soil in comparison to the most crops like cereal crops.

Utilization of exogenous compatible solutes and growth regulators in semiarid regions can be an effective practice for improving crop performance which is generally applied to mitigate the adverse effect of environmental stress (Wang *et al.*, 2010). Compatible solutes are molecules that protects cells from desiccation by maintaining a high intracellular osmolality and among different materials, glycine betaine (GB) has been further investigated and suggested that GB has very imperative role in crops protection under extreme environmental conditions (Wani *et al.*, 2013). Also, salicylic acid (SA) is naturally occurring plant hormone, influences various physiological and biochemical functions in plants because it can play role as a regulatory signal mediating plant response to abiotic stresses such as drought (Wang *et al.*, 2010). Ascorbic acid (AA) has been used to as a cofactor for enzymes involved in regulating photosynthesis, hormone biosynthesis, and regenerating other antioxidants; and it is essential to many aspects of plant growth and responses.

In recent years, nanotechnology has been expanded to agriculture and the numerous benefits of nanoparticles applications has been reported which can be used to increase the supply of nutrition elements to crop, like nano-silica ($nSiO_2$) and nano-titanium dioxide (TiO_2) which have exceptional biological characteristics. Nano-titanium dioxide particles are promising as efficient factor for plants to improve biomass production by enhancing metabolic activities and conversion of light energy (Gao *et al.*, 2008), and also, it increase the activity of the enzymatic antioxidant factors such as superoxide dismutase peroxidase (Hong *et al.*, 2005). According to Raliya *et al.* (2015), foliar application of nano-titanium dioxide particles on mung bean could significantly improve morphological traits, chlorophyll content and soluble leaf protein and according to Siddiqui *et al.* (2015), foliar application of nano-silica has gained greater consideration during growth period. Also, foliar spray of nano-silica on plants improve the plant growth and performance by enhancing the accumulation of antioxidant enzymes and increase the efficiency of photosynthetic apparatus (Kalteh *et al.*, 2014; Xie *et al.*, 2011). Exogenous application of nanoparticles can improve the plant resistance against environmental stress in semiarid regions, but despite recent progress in understanding the some environmental consequences of nanoparticles, little research is available about the influence of these materials on oil and seed yield performance of sunflower. Therefore, the aim of the this investigation was to determine the possible role of glycine betaine, salicylic acid and ascorbic acid growth regulators and nano-silica ($nSiO_2$) and nano-titanium dioxide (TiO_2) particles on morphological traits, yield components, seed yield and oil content of sunflower.

Material and methods

A field experiment was conducted during 2014–2015 growing season at Maragheh, Iran (37°23′ N, 46°16′ E and altitude 1485 m) with the average annual temperature 11.2 °C, and the rainfall average 353 mm. The soil type was a sandy loam with pH 7.6. The experimental field was ploughed once in early fall and harrowed twice before planting and 150 kg urea with 100 kg P_2O_5 ha^{-1} fertilizers were applied in the form of urea and triple superphosphate at the field preparation. The experiment was performed based on randomised complete block design in four replicates with Azargol sunflower hybrid sowed was at 28 March. Each experimental plot was consisting of 8 rows, 4.5 m length, and at 60 × 20 cm inter-plant and inter-row, respectively. Weeds were controlled by hand-hoeing. Treatments were Control: water spray, Nano-Si: foliar application of nano-silica suspension (2 mM), G-B: foliar spray of glycine betaine (100 mM), S-A: foliar spray of salicylic acid (1 mM), A-A: foliar application of ascorbic acid (1 mM), and Nano-Ti: spray of nano-titanium dioxide suspension (2 mM) which were applied at stem elongation, head determination and seed filing steps. Nanomaterial were prepared from the Pishgaman Nano Company, Iran.

Chlorophyll content (CHL) was measured with a hand-held dual wavelength meter (SPAD 502, Chlorophyll meter, Japan) in fully expanded upper leaves at the flowering stage. Leaf length (LL) and leaf width (LW) were measured at the end of flowering stage on ten randomly selected plants. Also, traits days to 50 % flowering (DF) and day to maturity (DM) were recorded. The morphological traits plant height (PH), husk percentage (HP), number of seeds per head (NSH), head number per plant (HNP), percentage of empty achenes (PEA) which was the ration of empty achenes to all achenes of each head, kernel weight (KW) and grain length (GL) were measured based on 10 random plants. The 1000-seed weight (TSW), straw yield (STY), harvest index (HI) and grain yield (GY) were recored for each plot. Oil percent (OIL) of seed were measured using a Near-infrared seed analyzers analyzer (Zeltex). Treatment by trait (TT) analysis using biplot (Yan and Kang, 2003) was used to determine which treatment was best and for what trait, which were generated using the standardized values of the traits means. The biplot analysis was based on Model 2. The polygon-view was based on treatment-focused singular value partitioning, while the vector views were based on the trait-focused singular value partitioning and is, therefore, appropriate for visualizing the relationships among traits and genotypes (Yan and Kang, 2003). All analyses reported in this study were conducted by using the GGEbiplot software (Yan, 2001).

Results and discussion

Figure 1 shows the "which-won-where" view of biplot and the outmost treatments (four in this case) formed a four-side polygon and the biplot was divided into four sectors delimited by the lines perpendicular to each side of the polygon. The traits fell into each of the four sectors of polygon. For traits of each sector, the nominal "winner" is at the vertex and so, Control and A-A (ascorbic acid) were treatments the wining treatments only for percentage of empty achenes (PEA) and plant height (PH), respectively (Figure 1). However, G-B (glycine betaine) was the winning treatments in the sector containing the husk percentage (HP), days to 50 % flowering (DF), leaf length (LL), 1000-seed weight (TSW) and leaf width (LW) traits. Finally, Nano-Ti was the winning treatments in the sector containing the other 10 traits including chlorophyll content (CHL), day to maturity (DM), number of seeds per head (NSH), head number per plant (HNP), kernel weight (KW), grain length (GL), straw yield (STY), harvest index (HI), grain yield (GY) and oil percent (OIL). The results shown in Figure 1 suggested that there might be distinct groups of traits (four in this case) in compliance with the large magnitude of treatment by trait interaction and the high value of PC1 and PC2 contributions to the total sum of squares in TT biplot.

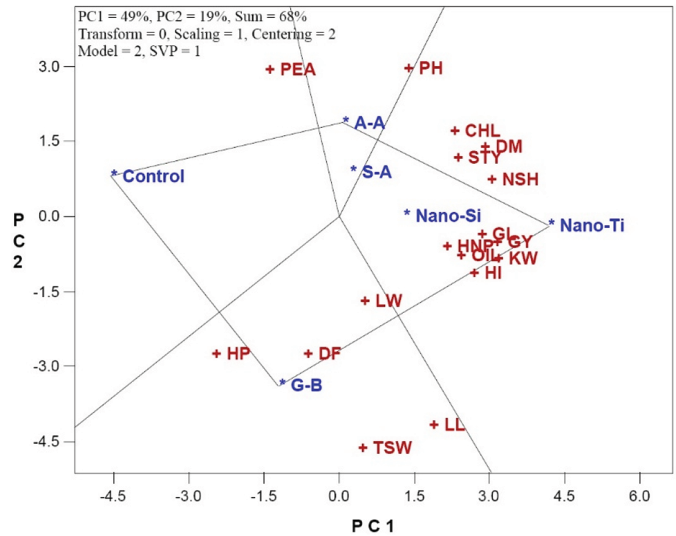

Figure 1: Polygon-view of treatment by trait (TT) biplot showing which treatment had the highest values for which traits.
Traits are chlorophyll content (CHL), leaf length (LL), leaf width (LW), days to 50 % flowering (DF), day to maturity (DM), plant height (PH), husk percentage (HP), number of seeds per head (NSH), head number per plant (HNP), percentage of empty achenes (PEA), 1000-seed weight (TSW), kernel weight (KW), grain length (GL), straw yield (STY), harvest index (HI), grain yield (GY) and oil percent (OIL).

Application of nano-scale TiO_2 at low concentration improved the traits of linseed under drought stress conditions, leading to better plant performance and similar to our results, chlorophyll content of linseed was found in leaves of nano-TiO_2-treated (Baiazidi-Aghdam *et al.*, 2015).

Based on the cosine of angles of traits vectors, there were positive correlation among CHL, STY, DM and NSH as well as among GY, GL, KW, HNP, OIL and HI (Figure 2). Also, there was near zero correlation between LL with PH, between LL with HP, between PEA with PH, and between TSW with CHL, STY, DM and NSH traits (Figure 2; Table 1). The presence of wide obtuse angles i. e. strong negative correlations among the traits (between PEA with LL, between PH with DF and between HP with CHL, STY, DM and NSH) is indication of strong crossover TT interactions (Yan and Tinker, 2006). According to literature, such results have also been reported for positive correlation between seed with 1000-seed weight (Singh and Labana, 1990) and oil content (Jhagirdhar, 1986).

As depicted in Figure 3 the single-arrowed line called average–tester coordination abscissa points to higher performance across traits to identification of

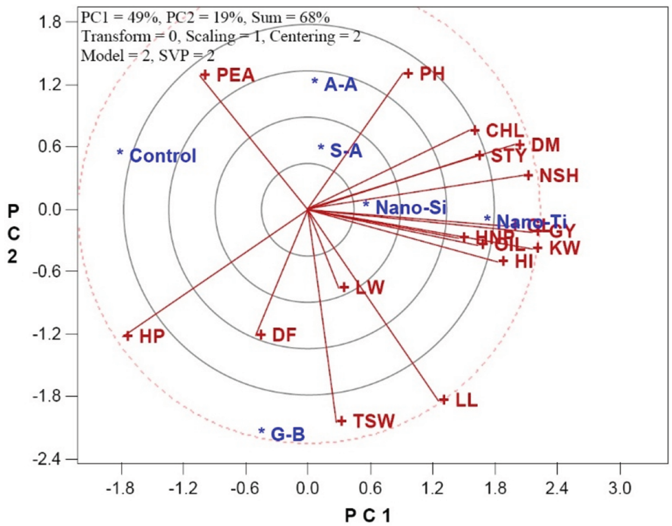

Figure 2: Vector view of treatment by trait (TT) biplot showing the interrelationship among measured traits under different treatments.
Traits are chlorophyll content (CHL), leaf length (LL), leaf width (LW), days to 50 % flowering (DF), day to maturity (DM), plant height (PH), husk percentage (HP), number of seeds per head (NSH), head number per plant (HNP), percentage of empty achenes (PEA), 1000-seed weight (TSW), kernel weight (KW), grain length (GL), straw yield (STY), harvest index (HI), grain yield (GY) and oil percent (OIL).

the ideal treatment which has high performance in most or all measured traits. When we rank traits across treatments it should be done with respect to an ideal trait that lies on average–tester coordination abscissa (absolutely stable) in the positive direction and has a vector length equal to the longest vector of the traits on the positive side of this abscissa i. e. highest performance, therefore, traits which are closer to “ideal trait” are more desirable than others (Yan and Tinker, 2006) and thus, GL, GY, KW, HNP, OIL and HI were high performance with consistent performance across the treatments (Figure 3). Also, CHL, DM, STY and NSH following to PH, LL and LW traits could be regarded similar to the above mentioned traits. TT biplot instability distance from the ideal entry is the measure of determination of the best entry or treatment and among the four highest performing entries, Nano-Ti following to Nano-Si treatments were identified as the most favorable treatment (Figure 4). Furthermore, TT biplot analysis indicated that G-B treatment performed better than Control, A-A and S-A treatments (Figure 4).

Table 1: Simple correlation coefficients among sunflower traits.

	PH	LL	LW	CHL	NSH	TSW	DF	DM	HP	KW	PEA	GL	HNP	GY	STY	HI
LL	−0.13															
LW	0.07	0.07														
CHL	0.28	0.05	−0.02													
NSH	0.29	0.34	0.14	0.57												
TSW	−0.03	0.41	0.05	−0.31	−0.04											
DF	−0.12	0.18	0.25	0.23	−0.16	0.02										
DM	0.10	0.22	−0.10	0.57	0.74	−0.08	−0.25									
HP	−0.41	0.09	−0.01	−0.47	−0.61	0.15	0.29	−0.45								
KW	0.33	0.44	0.10	0.28	0.59	0.42	−0.16	0.49	−0.51							
PEA	−0.26	−0.50	0.00	−0.13	−0.17	−0.21	−0.21	−0.01	0.26	−0.29						
GL	0.34	0.45	0.37	0.24	0.64	0.05	−0.25	0.33	−0.32	0.47	−0.45					
HNP	0.22	0.20	−0.01	0.13	0.37	−0.12	0.07	0.31	−0.02	0.34	−0.02	0.16				
GY	0.30	0.37	0.06	0.31	0.74	0.06	−0.02	0.57	−0.30	0.60	−0.13	0.42	0.88			
STY	0.13	0.16	0.24	0.09	0.27	−0.06	−0.55	0.28	−0.33	0.18	−0.17	0.52	−0.25	−0.04		
HI	0.23	0.30	−0.02	0.27	0.61	0.07	0.15	0.45	−0.17	0.50	−0.07	0.23	0.90	0.95	−0.36	
OIL	0.01	0.49	−0.05	0.07	0.51	0.35	−0.48	0.60	−0.30	0.58	−0.08	0.42	0.07	0.37	0.56	0.17

Critical values of correlation $P < 0.05$ and $P < 0.01$ (df 4) are 0.81 and 0.92, respectively.
Traits are chlorophyll content (CHL), leaf length (LL), leaf width (LW), days to 50 % flowering (DF), day to maturity (DM), plant height (PH), husk percentage (HP), number of seeds per head (NSH), head number per plant (HNP), percentage of empty achenes (PEA), 1000-seed weight (TSW), kernel weight (KW), grain length (GL), straw yield (STY), harvest index (HI), grain yield (GY) and oil percent (OIL).

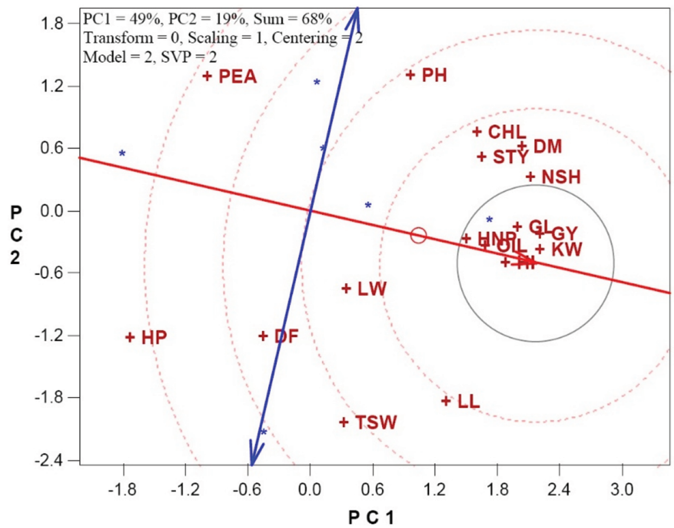

Figure 3: Ideal tester view of treatment by trait (TT) biplot, showing the relationships of different traits with ideal tester.
Traits are chlorophyll content (CHL), leaf length (LL), leaf width (LW), days to 50 % flowering (DF), day to maturity (DM), plant height (PH), husk percentage (HP), number of seeds per head (NSH), head number per plant (HNP), percentage of empty achenes (PEA), 1000-seed weight (TSW), kernel weight (KW), grain length (GL), straw yield (STY), harvest index (HI), grain yield (GY) and oil percent (OIL).

The best treatment for obtaining of high grain yield (GY) and high oil content (OIL) could be find in the special vector-view of TT biplot (Figure 5) which shows treatments that have close association with a target trait among other traits. According to these biplots of Figure 5, Nano-Ti following to Nano-Si treatments were the best treatments suitable for obtaining of high performance of grain yield as well as high oil content. Thus, application of this treatment combination is expected to lead to improve the target trait under rainfed growing conditions in semi-arid region. This suggests that using nano-titanium dioxide suspension (2 mM) or nano-silica suspension (2 mM) will not only result in the development of high grain yield but also cause to obtain the other desirable agronomic traits which are associated to grain yield such as oil content. Therefore, it seems that application of Ti and Si nanoparticles had good effect on sunflower yield performance, yield components and morphological traits in comparison to application of glycine betaine (100 mM), salicylic acid (1 mM), ascorbic acid (1 mM) compounds or water spray (Control). Nano-titanium dioxide has been studied as photocatalysts (Ullah and Dutta, 2008), and Lei *et al.* (2007) found that, nano-titanium dioxide increases photosynthesis

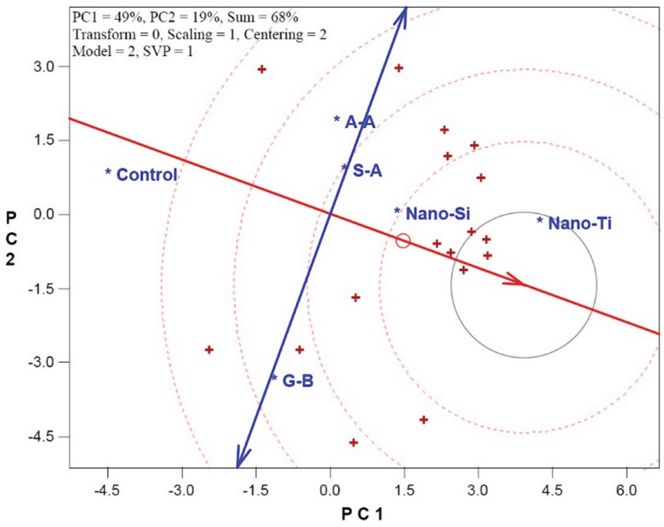

Figure 4: Ideal entry view of treatment by trait (TT) biplot, showing the relationships of different treatments with ideal entry.
Traits are chlorophyll content (CHL), leaf length (LL), leaf width (LW), days to 50 % flowering (DF), day to maturity (DM), plant height (PH), husk percentage (HP), number of seeds per head (NSH), head number per plant (HNP), percentage of empty achenes (PEA), 1000-seed weight (TSW), kernel weight (KW), grain length (GL), straw yield (STY), harvest index (HI), grain yield (GY) and oil percent (OIL).

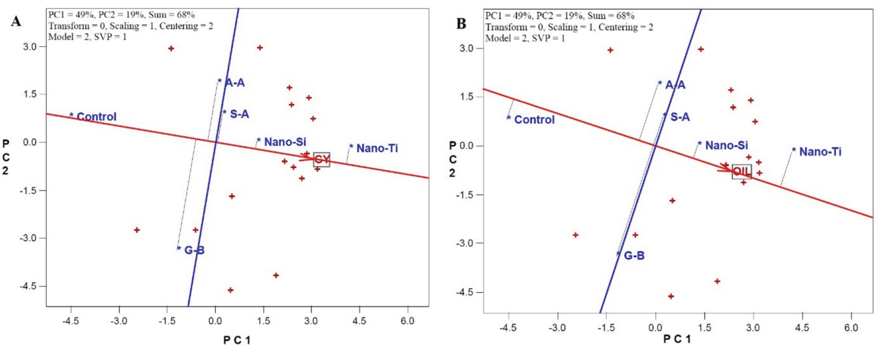

Figure 5: Vector view of treatment by trait (TT) biplot, showing the relationships of different treatments with grain yield (GY) and oil content (OIL) traits.
Traits are chlorophyll content (CHL), leaf length (LL), leaf width (LW), days to 50 % flowering (DF), day to maturity (DM), plant height (PH), husk percentage (HP), number of seeds per head (NSH), head number per plant (HNP), percentage of empty achenes (PEA), 1000-seed weight (TSW), kernel weight (KW), grain length (GL), straw yield (STY), harvest index (HI), grain yield (GY) and oil percent (OIL).

and plant growth of spinach and enhances absorption of the sun's energy. Zheng *et al.* (2005), treated spinach old seeds with the nano-titanium dioxide and observed that most traits including germination rate and vigor index increased in seeds treated while Lu *et al.* (2002) reported that evaluations for super oxide dismutase and catalase increased in soybean that had been treated with nano-titanium dioxide. Also, Owolade *et al.* (2008), reported that nano-titanium dioxide application on some traits of cowpea such as number of seeds per pod, number of pod per plant, 1000 seed weight and grain yield performance were better than of these traits compared to the control. Some treatments like as nano-titanium dioxide and nano-silica increased the plant growth characteristics and partly expanded the source size, thus, it revealed that nanoparticles improved the both vegetative and reproductive traits. These findings further support the idea of Jaberzadeh *et al.* (2013) who reported that application of nano-titanium dioxide increased wheat growth and significantly improved grain yield under drought stress condition. Our finding about increment of oil percentage in yield performance by application of beneficial nanoparticles corroborates these earlier findings. Davar-Zareii *et al.* (2014) found that foliar application of nanoparticles during reproductive growth stage significantly improved the oil percentage in safflower. However, the findings of the current study do not support the results of Ahmed (2013) who suggested that salicylic acid and ascorbic acid would be highly helpful for increasing oil yield and fatty acid contents in sunflower. Theresults of present study suggested that foliar application of nano-titanium dioxide can be suitable option for improving sunflower production.

Our study results indicate that nano-titanium dioxide and nano-silica play a significant role on increasing of sunflower yield and other traits, so that higher amounts of traits obtained with spraying of nanoparticles in compared with spraying distilled water (Control), A-S, A-A and G-B. The studies on improving photosynthesis suggested that nano-TiO_2 could increase this process and protect chloroplasts (Gao *et al.*, 2008; Lei *et al.*, 2007; Yang *et al.*, 2006). Nano-TiO_2 particles enhanced seed germination and growth of canola seedlings (Mahmoodzadeh *et al.*, 2013), as well as improving plant growth and yielded components of wheat under drought stress conditions (Jaberzadeh *et al.*, 2013). Effectiveness of nanoparticles depends on their concentration and varies among plants, therefore different concentrations needs further evaluation and may provide a more comprehensive interpretation. The lower concentrations of nano-SiO_2 improved seed germination of maize (Suriyaprabha *et al.*, 2012); and seedling growth of Changbai larch while nano-SiO_2 enhances the plant growth and development by increasing chlorophyll parameters, such as net photosynthetic rate, transpiration rate and photochemical quench (Xie *et al.*, 2011). Shah and Belozerova (2009) tested nano-silica on lettuce and found that these nanoparticles have a significant influence on seeds. This research reveals that the research on nano-titanium dioxide and nano-silica particles on sunflower, is in the beginning; more rigorous works are needed to understand physiological mechanisms of plants in relation to these nanoparticles as well as studies are needed to explore the mode of action of nanoparticles, their interaction with biomolecules.

Conclusions

Foliar spray of nano-titanium dioxide following nano-silica nanoparticles considerably enhanced the yield and yield components of sunflower performance in semi-arid region. In contrast application of glycine betaine, salicylic acid and ascorbic acid had not any positive effect on sunflower.

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