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Research Article

Foliar spray of Si and Ti nanoparticles affected enzymatic antioxidants in rapeseed (*Brassica napus* L.)

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Abstract

Some beneficial nanoparticles such as silicon and titanium are encouraging in foliar feeding due to their large surface area and biocompatible properties. In current experiment, seedling of rapeseed varieties (‘Hydromel’, ‘Nathalie’, ‘Alonso’) was sprayed with titanium dioxide and silicon dioxide nanoparticles (Ti and Si-NPs) and then responses of some enzymatic antioxidant and plasma membrane integrity were evaluated. Foliar application of Si-NPs significantly increased the activity of ascorbate peroxidase compared to the control, in all three varieties. Spray of both Ti and Si-NPs increased guaiacol peroxidase activity in ‘Nathalie’ and ‘Alonso’ varieties while in ‘Hydromel’ only Si-NPs spray had positive effects on activity this antioxidant. Behaviors of superoxide dismutase (SOD) isozymes under different foliar treatments and in varieties were different, but the highest activity of total SOD isozyme in ‘Hydromel’ and ‘Alonso’ varieties was obtained by foliar application of Si-NPs and Ti-NPs, respectively. The lowest amount of hydrogen peroxide was obtained by foliar application of Ti-NPs. Examination of malondialdehyde (MDA) also showed that the most vulnerable membrane belonged to ‘Hydromel’ variety and foliar application of Si-NPs and Ti-NPs could improve membrane integrity. These results suggest that foliar application of both Si-NPs and Ti-NPs improves the scavenging capacity of reactive oxygen species (ROS), although the response of the enzymatic antioxidants was largely influenced by the variety.

Keywords: ascorbate peroxidase; catalase; hydrogen peroxide; membrane integrity; ROS scavenging

Introduction

Brassicaceae includes of several economically important species broadly used as sources of oil and food, and as ornamental plants. Rapeseed (*Brassica napus* subsp. *napus*), also known as rape, oilseed rape colza. A

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modified variant of rapeseed developed in Canada has been named “canola” or “double low” variety (Raboanatahiry *et al*., 2021), for its low content in erucic acid (less than 2%) and glucosinolates (less than 30 $\mu molg^{-1}$). However, the production of this plant is strongly affected by environmental constraints in central region of Iran. Drought and high temperature stresses are the most important abiotic stresses limiting rapeseed yield potential in semi-arid regions, leading to food and nutritional insecurity in Iran. Therefore, any crop management that can improve plant defenses and strengthen plant growth has great importance.

The foliar application of stimulants defined here as ‘a substance or material which has the capability to positively adjust plant growth’ has increased meaningfully over the past decade (Popović *et al*., 2013; 2020a; 2020b; Saa *et al*., 2015; Lakić *et al*., 2020; Kolarić *et al*., 2021). The mechanisms by which foliar applied materials penetrate the leaf surface is complex and will be influenced by size and polar nature of the applied molecules (Fernandez and Brown, 2013). It appears that nanotechnology has a potential to provide solutions to fundamental agricultural; problems caused by environmental stress (Fattahi *et al*., 2018). Nanoparticles (NPs) are unit resources with an individual dimension from 1 to 100 nm and inconceivable structural and physicochemical properties due to their large surface area and biocompatible characteristics (Ahmed *et al*., 2021). Nanoparticles associate with plants, causing a variety of morphological and physiological changes depending on their properties. Both ameliorative and negative impacts of NPs have been studied at different developmental stages of plant and the response varies upon NPs types, sizes, exposure time, duration and crop species (Munir *et al*., 2018; Rizwan *et al*., 2019). In recent years, the application of some effective nanoparticles in agriculture has become widespread. There has been a special focus on SiO_2 and TiO_2 nano-particles. Although silicon is not an essential element for plants, its partial uptake by many plants plays a positive role in the plant's defense as well as abiotic stresses. Our previous results showed that foliar application of SiO_2 and TiO_2 nano-particles invigorated growth of safflower and barley plants in semi-arid region (Janmohammadi *et al.,* 2016; 2017). Rizwan *et al.* (2019) depicted that SiO_2 and TiO_2 nanoparticles were able in diminishing cadmium stress in rice seedlings. So that The NPs decreased electrolyte leakage, and malondialdehyde content and improved the activities of superoxide dismutase, preoxidase, catalase, and ascorbate peroxidase in rice shoots over the control. Also, Asgari *et al.* (2018) found that Si nano-particles significantly increased the antioxidants enzymes activities (peroxidase, catalase, and superoxide dismutase) in oat (*Avena sativa* L.) seedlings. Silicon increases rigidity of cell wall. Si nano-particles binds with pectins, polyphenols and hemicellulose in cell walls and improves the mechanical strength of the plant cell wall (Rajput *et al.,* 2021). Also, in foliar application of nano-silicon it may reduce plant transpiration by creating a layer on leaf surface and, thus, make plants more resistant to drought and high temperature (Strout *et al*., 2013). TiO_2 nanoparticles are also used as an essential nutrient for plant growth and development and also improves the photosynthetic pigments in plants such as chlorophylls, carotenoids and anthocyanins contents, therefore, enhanced the crop growth and yield (Chaudhary and Singh, 2020). Modern management of companies, regardless of the activity in which the company operates, requires top management to apply and introduce internal control mechanisms (Popović *et al*., 2020c; 2021). Despite the plenty of information available on foliar application of some nanoparticles on plant growth and development, there is no sufficient information about efficiency of Si and Ti nanoparticles under natural farm conditions in semi-arid regions where water scarcity and high temperature have negative impacts on plants. The present study was carried out to evaluate the effects of foliar spray of Si and Ti nanoparticles on reactive oxygen species (ROS) scavenging capacity in rapeseed varieties in the northwest of Iran.

Materials and Methods

Site of study

A field experiment was carried out during 2016-2017 growing season in semiarid region of Qazvin (latitude: 36°08′N; longitude: 49°85′E; altitude: 1278 m), in northwest of Iran. The yearly average of precipitation (30 years long-term period) was 301 mm that typically concentrated during the autumn and winter months (November to February). In August, precipitation was at its lowest, with an average of 1 mm. The greatest amount of precipitation occurred in April, with an average of 75 mm. The climate of location based on Köppen climate classification is BSK; semi-arid moderate (Peel *et al.,* 2007). Soil samples (0-30 cm) were collected at sowing and analyzed for some physic-chemical properties. The soil texture of the studied area was clay loam with 0.31% organic matter content and pH of 7.6. Total N content was 0.04%, available P was 9.13 $mgkg^{-1}$, available K was 121.0 $mgkg^{-1}$, available Fe was 1.71 $mgkg^{-1}$, available Zn was 0.58 $mgkg^{-1}$ and available Mn was 2.28 $mgkg^{-1}$.

Experimental design

The trial was laid out in randomized complete block design with three replications in split plot arrangement (3 × 3). Seeds of three rapeseed varieties ('Hydromel', 'Nathalie', 'Alonso') were obtained from Seed and Plant Research Improvement Institute, Karaj, Iran. Seed sowing was done with hand during the third decade of September in 3 cm depth. Prior to sowing, the field was thoroughly ploughed to ensure adequate soil aeration. The field was irrigated prior to sowing to maintain proper moisture content in the subsurface of the soil. Plot size of was 16 m^2 (4 × 4) which was allocated to 9 combined treatments so that foliar application of nano-particles assigned to main plots and varieties distributed in the sub-plots. Each subplot contains eight plant row, 4 m long with spacing of 0.5 m between rows, and at a rate of 20 seeds per meter of row. Foliar treatment included control (check; spray of distilled water), nano-Ti (20 ppm), nano-Si (20 ppm). A drip system was applied for irrigation. Nano-particles were obtained from the Iranian Nanomaterials Pioneers Company. The synthesized nano particles were characterized morphologically by transmission electron microscope (Zeiss EM 10 C, Merck, Darmstadt, Germany). Foliar spray carried out three times during the vegetative (stem elongation, early lateral growth) and reproductive growth stages (inflorescence emergence).

Biochemical analysis

Leaf samples was collected at initiation of seed development stage for biochemical analysis. Leaf samples (0.5g) were homogenized in ice cold 0.1 M phosphate buffer (pH=7.5) containing 0.5 mM EDTA with pre-chilled pestle and mortar for evaluating the activity of ROS scavenging enzymes such as catalase (CAT), ascorbate peroxidase (APX), superoxide dismutase (SOD) and guaiacol-peroxidase (GPX). Each homogenate was transferred to centrifuge tubes and was centrifuged at 4°C in refrigerated centrifuge for 15 min at 15000×g. The supernatant was used for enzyme activity assay as described previously by Janmohammadi (2012). SOD activity in the gels was detected using activity staining as described earlier (Beauchamp and Fridovich, 1971). The three types of SOD, Fe-SOD, Mn-SOD, and Cu-Zn SOD were identified using inhibitors. Mn-SOD was visualized by its insensitivity to 5 mM H_2O_2 and 2 mM cyanure de potassium (KCN), while Cu-Zn SOD was sensitive to 2 mM KCN. Fe-SOD was inhibited by 5 mM H_2O_2 (Navari-Izzo *et al*., 1998). CAT activity was measured according to Aebi (1984). About 3 ml reaction mixture containing 1.5 ml of 100 mM potassium phosphate buffer (pH=7), 0.5 ml of 75 mM H_2O_2, 0.05 ml enzyme extraction and distilled water to make up the volume to 3 ml. Reaction started by adding H_2O_2 and decrease in absorbance recorded at 240 nm for 1 min. Enzyme activity was computed by calculating the amount of H_2O_2 decomposed. Guaiacol peroxidase (GPX) was determined by measuring the oxidation of guaiacol. The assay mixture contained 10 $mmolL^{-1}$ potassium phosphate (pH 6.4), 8 $mmol.L^{-1}$ guaiacol, and 2.75 $mmol.L^{-1}$ H_2O_2. The increase in absorbance was recorded at 470 nm within 2 min (linear phase) after the addition of H_2O_2 using a spectrophotometer (UV-1800, Shimadzu, Japan). Malondialdehyde (MDA) is one of the most commonly reported biomarkers of lipid peroxidation. MDA content was estimated in leaf as described by Heath and Packer (1968). For assessment of MDA content, a total of 0.1 g of rapeseed fresh leaves (the youngest fully expanded leaves) from each treatment was ground with a mortar and pestle in 4 mL of 50 mM phosphate buffer (pH 7.6) and centrifuged at 14,000 g for 20 min. After separation, the supernatant from the leaf residue, 0.4 mL of each sample added to 1.2 mL of reaction mixture including 0.5% of 2-thiobarbituric acid (TBA) and 20% (v/v) of trichloroacetic acid (TCA). The mixture was incubated for 30 min at 95 °C and then quickly cooled with ice to stop the reaction. The cooled mixture was re-centrifuged at 10,000 g for 10 min and finally the absorbance was measured at 532 nm. The extinction coefficient of 155 mM^{-1} cm^{-1} was used to measure the MDA concentration.

Statistical analysis

The collected data were analyzed using SAS 9.1 software. Mean values were compared according to Steel and Torrie (1980) by least significant difference (LSD) at $p < 0.05$.

Principal component analysis (PCA) was used to study the relationship among the verities and foliar treatments and to identify the important combined treatments. Cluster analysis was used to group the physio-biochemical parameters according to similarity in response of foliar treatments. PCA and cluster analysis was done using SPSS.

Results and Discussion

The constant increase in the need for food and raw materials for processing, are the reasons for increasing the yield and require constant study of rapeseed genetics. The application of nanoparticles can significantly reduce the effects of adverse environmental factors on the yield and quality of rapeseed. Analysis of variance (ANOVA) showed that catalase (CAT) activity affected by both factors (foliar spray and variety). The highest CAT activity was recorded for Nathalie variety treated with Ti-NPs (Table 1).

Table 1. Effect of foliar spry of silicon and titanium nanoparticles on physio-biochemical parameters in rapeseed (*Brassica napus* L.) varieties, grown in semi-arid region of Qazvin, Iran

Treatment		CAT	TSOD	Cu/Zn-SOD	Mn-SOD	Pr	MDA
Hydromel	Ti-NPs	0.017^{c}	2.45^{cd}	0.008^{d}	0.005^{c}	0.29^{ab}	85.35^{d}
	Si-NPs	0.022^{b}	3.99^{a}	1.64^{a}	0.004^{c}	0.26^{bc}	183.59^{ab}
	Control	0.014^{cd}	1.33^{de}	0.548^{bc}	0.085^{c}	0.24^{c}	185.17^{a}
Nathalie	Ti-NPs	0.031^{a}	1.82^{cde}	0.011^{d}	0.36^{c}	0.32^{a}	118.44^{cd}
	Si-NPs	0.022^{b}	2.81^{bc}	0.64^{bc}	0.85^{b}	0.29^{ab}	140.86^{abc}
	Control	0.016^{c}	1.14^{e}	0.34^{cd}	0.035^{c}	0.26^{bc}	183.11^{ab}
Alonso	Ti-NPs	0.015^{cd}	3.64^{ab}	0.004^{d}	1.67^{a}	0.31^{a}	160.75^{abc}
	Si-NPs	0.022^{b}	2.07^{cde}	0.716^{b}	0.003^{c}	0.24^{c}	134.64^{bc}
	Control	0.011^{d}	1.70^{cde}	0.40^{bc}	0.006^{c}	0.25^{bc}	181.67^{ab}
LSD		0.0049	1.16	0.35	0.162	0.045	49.14
		Significance level					
Variety (V)		**	ns	**	**	**	ns
Nano-particle Spray (S)		**	**	**	**	**	**
V×S		**	**	**	**	ns	*

CAT: catalase activity (Units mg $protein^{-1}.min^{-1}$), TSOD: total superoxide dismutase activity, Cu/Zn-SOD: activity of copper/zinc superoxide dismutase isoezyme, Mn-SOD: activity of manganese superoxide dismutase, Pr: total protein (%), MDA: malondialdehyde content ($nmol.gFW^{-1}$). In each column if the difference between the means is greater than the LSD (Least Significant Difference) value, then the means are significantly different (P ≤ 0.05). **: NS: significant at 0.01 level, *: significant at 0.05 level, not statistically significant.

Although Ti-NPs in Nathalie variety had a greater effect on CAT activity, in Hydromel and Alonso variety the highest activity of this enzyme was obtained by exogenous foliar application of Si-NPs. Assessment of ascorbate peroxidase (APX) showed that foliar application of nanoparticles and variety affected the activity of this enzymes. The highest APX activity was recorded in Nathalie and Alonso variety sprayed with Si-NPs. The lowest level of APX activity was observed in Hydromel variety. However, in all varieties under no-application of nanoparticles, the activity of ascorbate peroxidase was significantly lower than other foliar treatments. However, the effect of Si-NPs on APX was greater than that of Ti-NPs in all varieties (Figure 1). Guaiacol peroxidase (GPX) activity of three rapeseed varieties under foliar application of nanoparticles is presented in Figure 2. The response of this enzyme in Nathalie and Alonso varieties to foliar treatments was very similar to each other, so that the foliar application of both nanoparticles (Si and Ti) significantly increased the activity of this enzyme. While in hydromel variety, only the application of Si-NPs could increase the activity of this enzyme.

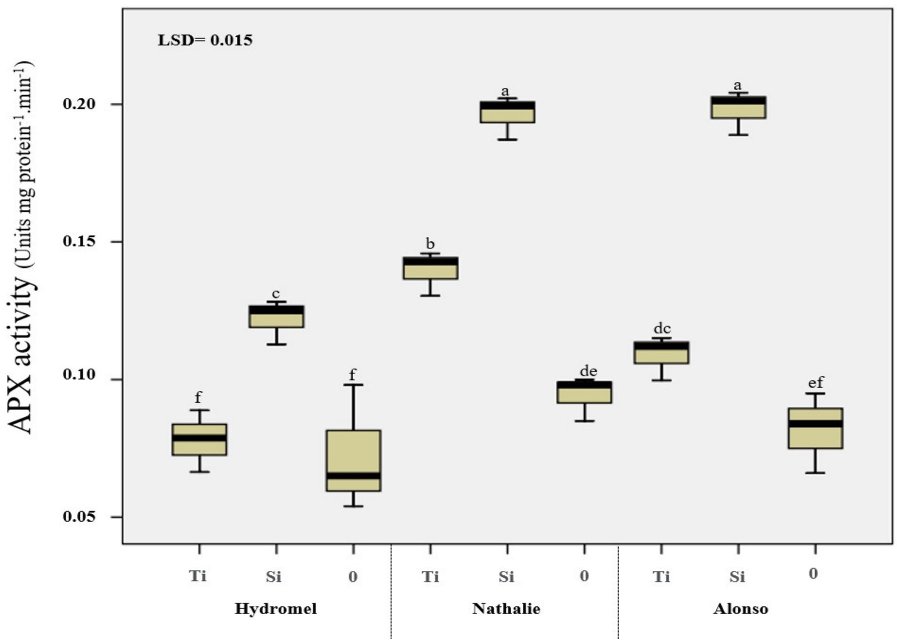

Figure 1. Effect of foliar application of nanoparticles (silicon and titanium) on Ascorbate peroxidase (APX) activity in three rapeseed varieties, grown under semi-arid region of Qazvin, Iran
Different uppercase letters in columns indicate a statistical difference according to LSD test ($p < 0.05$).

According to the active site metal, the multiple SOD isoforms are classified into three major groups (types): Fe-SOD (iron cofactor), MnSOD (manganese cofactor), and Cu/ZnSOD (copper and zinc as cofactors; copper is the redox active catalytic metal). Examination of the total activity of superoxide dismutase isoezymes (TSOD) showed that the foliar application of Si-NPs in Hydromel variety and the application of Ti-NPs in Alonso variety increased the total activity of superoxide dismutase enzyme by three times over the control (no-application of nanoparticles). Mean comparison of Fe-SOD activity between combined treatments (variety × foliar spray) revealed that response of Fe-SOD to foliar application treatments was significantly different in varieties, so that the highest activity was recorded for Nathalie using Si-NPs, which was about two and a half times higher than plants not treated with nanoparticles. The lowest level of Fe-SOD activity was observed in Alonso variety without the foliar application of nanoparticles (Figure 3). Interestingly, in Hydromel variety, application of both nanoparticles significantly increased Fe-SOD activity when compared with control (no-application of nanoparticles). Evaluation of Cu/ZnSOD showed that response of this enzyme to foliar treatments was very similar to TSOD. The highest activity of Cu/ZnSOD was recorded for Hydromel plants treated with Si-NPs. The application of Si-NPs had a greater effect on the activity of this isozyme compared to Ti-NPs. Evaluation of Mn-SOD showed that the only foliar application of Ti-NPs in Alonso variety and Si-NPs in Nathalie variety had the greatest effect (Table 1).

Estimation of leaf protein content showed that the application of Ti-NPs in all three varieties significantly increased the amount of this biomolecule.

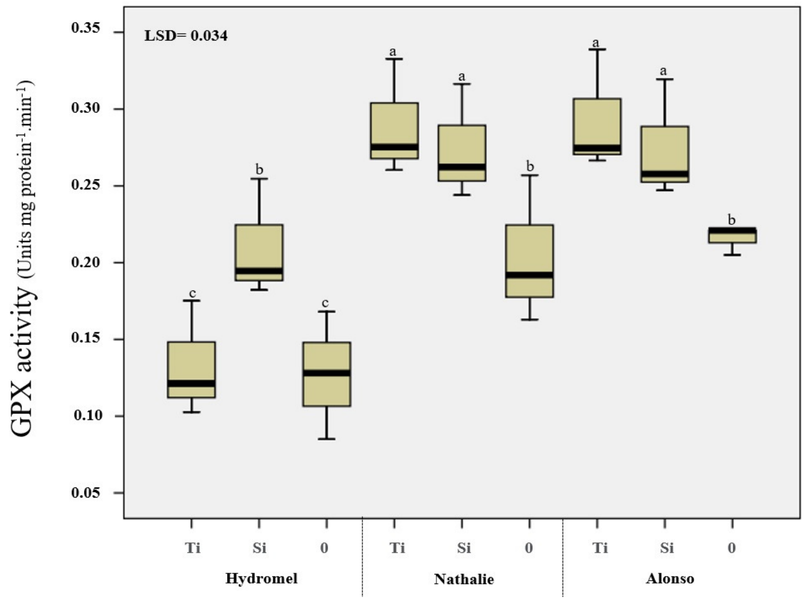

Figure 2. Guaiacol peroxidase (GPX) activity of three rapeseed varieties affected by of exogenous spray of nanoparticles (silicon and titanium) under semi-arid region of Qazvin, Iran

Columns denoted by different letters indicated significant ($p < 0.05$) differences among different synthesis conditions.

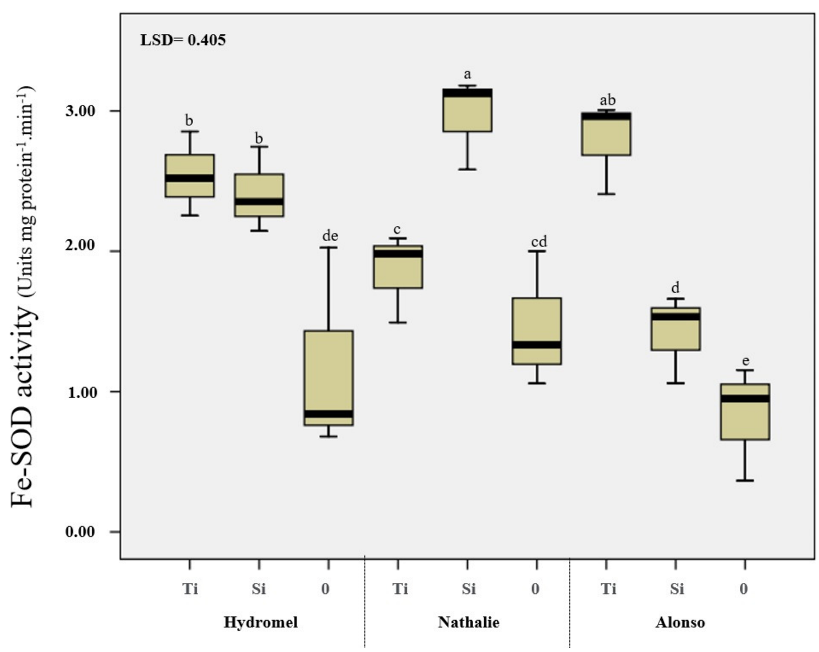

Figure 3. Effect of foliar application of nanoparticles (silicon and titanium) on iron superoxide dismutase (FeSOD) activity in three rapeseed varieties, grown under semi-arid region of Qazvin, Iran

Different uppercase letters in columns indicate a statistical difference according to LSD test ($p < 0.05$).

The most prominent positive effect of foliar application of nanoparticles on protein content recorded in Nathalie variety. Malondialdehyde (MDA) is one of the most commonly used biomarkers for lipid peroxidation. The lowest amount of MDA was recorded in Hydromel and Nathalie variety by foliar application of Ti-NPs (Table 1). The lowest effect of foliar application of nanoparticle on MDA content was related to Alonso variety, which seems that evaluated antioxidant system have not critical role in inhibition of membrane peroxidation. Assessment of hydrogen peroxides concentration revealed that foliar application of both nanoparticles significantly reduced the concentration of hydrogen peroxide, however the greatest reduction was achieved in 'Nathalie' and 'Hydromel' varieties with the foliar application of Ti-NPs (Figure 4).

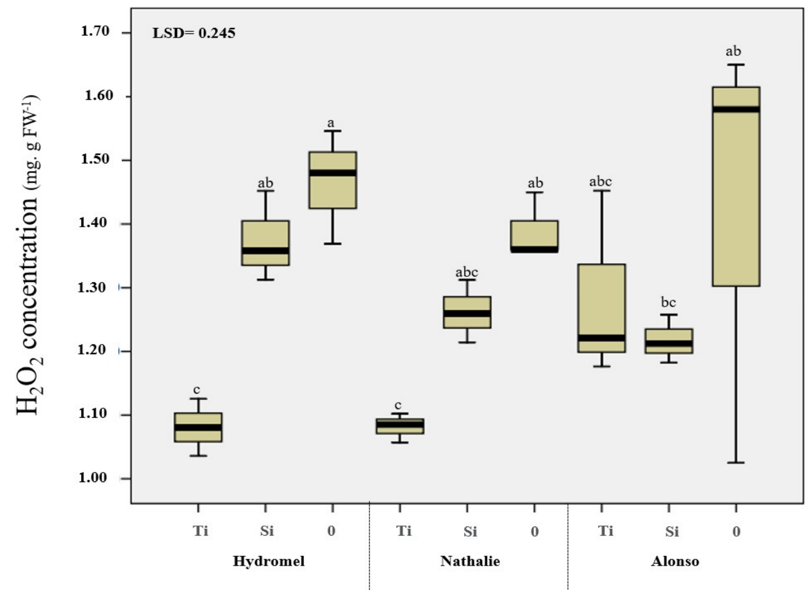

Figure 4. Impact of exogenous spray of nanoparticles (silicon and titanium) in three rapeseed varieties on hydrogen peroxide concentration

Columns denoted by different letters indicated significant (p < 0.05) differences among different synthesis conditions.

Cluster analysis of physio-biochemical traits under in different varieties and under various foliar treatments showed that the dendrogram was divided into three clusters (Figure 5). Cluster I consisted of APX, GPX, and CAT which significantly increased by the foliar spray of Si-NPs and their response was approximately similar in Nathalie and Alonso. Cluster II contained the TSOD, Fe-SOD, Mn-SOD and protein content which showed highest value by foliar spray of Si-NPs in Nathalie and Ti-NPs in Alonso. Cluster III contained the hydrogen peroxide content and MDA content which both are membrane degrading agents as well as lipid peroxidation residues and their highest values obtained in Alonso and Natalie varieties without nanoparticles foliar application. Interestingly, the use of Ti-NPs in the Hydromel variety caused to the lowest MDA and H_2O_2.

Figure 6 displays a biplot in the dimension of the first and second PCs. On the plot, two main groups of combined treatments were separated so that for factor 1 there were no-application of nanoparticles in all three varieties and 'Hydromel' with Si-NPs, the lowest amount of antioxidant enzyme activities was recorded in these combined treatments. Besides, factor 2 separated foliar spray of Ti-NPs in all three varieties and spray of Si-NPs in 'Alonso' and 'Nathalie' which showed the highest activity of antioxidant enzymes.

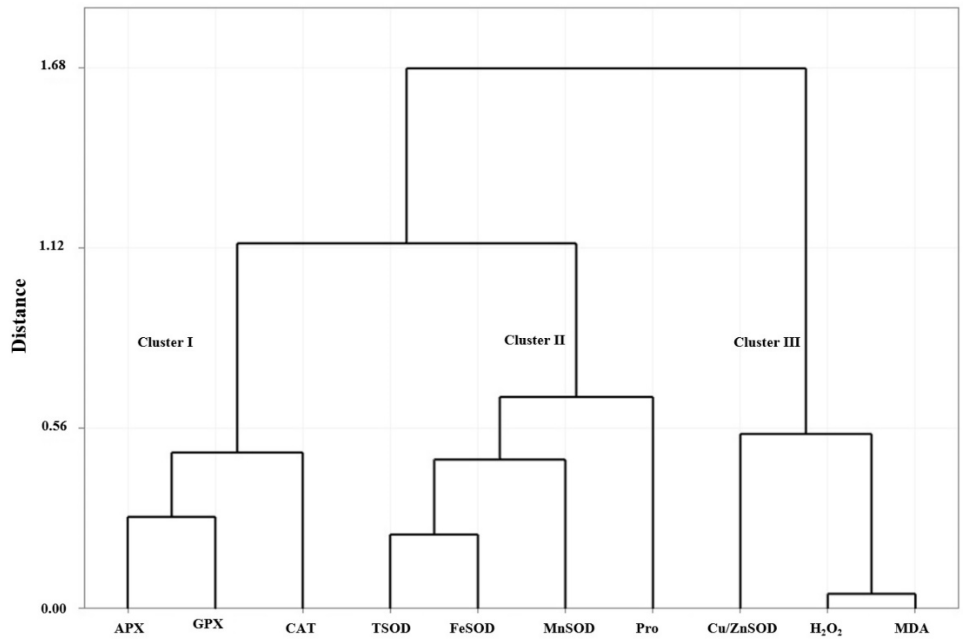

Figure 5. Cluster analysis of ROS scavenging enzymes and physio-biochemical parameters of rapeseed according to similarity in response to foliar treatments. APX: ascorbate peroxidase, GPX: guaiacol peroxidase, CAT: catalase activity, TSOD: total superoxide dismutase, Fe-SOD: iron- superoxide dismutase, Cu/Zn-SOD: copper/zinc superoxide dismutase isoezyme, Mn-SOD: manganese superoxide dismutase, Pr: total protein, H_2O_2: Hydrogen peroxides concentration, MDA: malondialdehyde content.

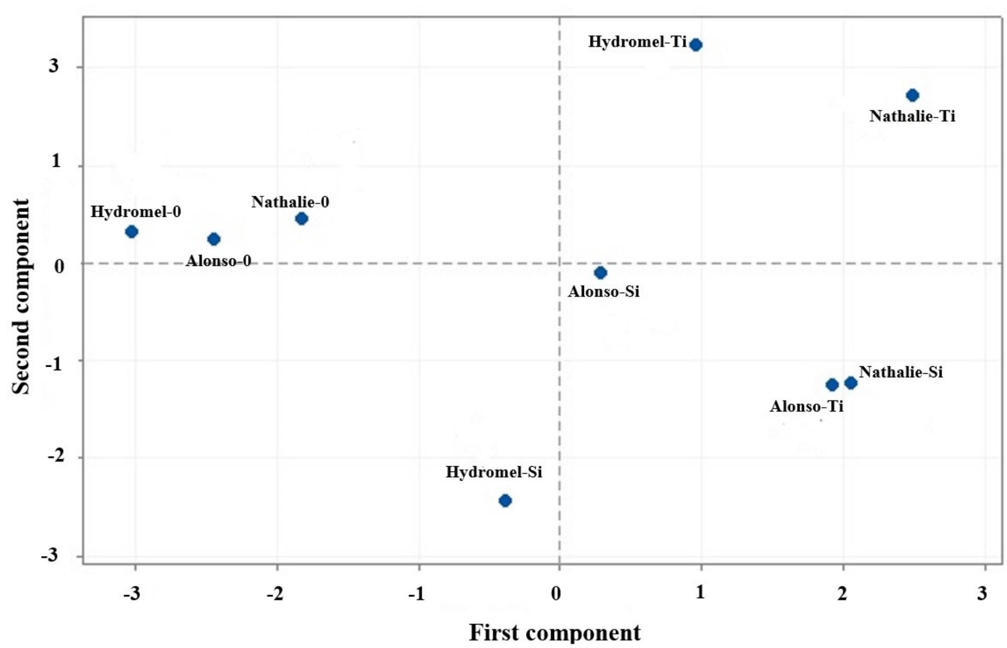

Figure 6. Principal component analysis (PCA) based on combined treatments (foliar spry and rapeseed varieties) which measured for evaluated physio-biochemical traits

Our results showed that although the activity of antioxidant enzymes in Hydromel was lower than other varieties, but the activity of total SOD isozymes as well as the activity of iron-SOD isozyme was strongly stimulated by the use of nanoparticles, especially Si-NPs. SOD catalyzes the dismutation of superoxide anion radical ($O_2\cdot^-$) to less reactive products i.e., O_2 and H_2O_2. SOD constitutes the front-line of defense against reactive oxygen species and oxidative stress in plant cells. Changes in SOD encoding gene transcript level and SOD activity are regarded as indicators of the level of ROS production and oxidative stress. However, SOD seems to have dual roles in controlling ROS damage and regulating ROS signaling (Wang *et al.*, 2018). Given that the behavior SOD isozymes activities of rapeseed varieties against different foliar application of nanoparticles was different and even so this difference was obvious under control condition (without foliar application), it probably indicates that the mechanisms and pathways involved in reactive oxygen species production is different in various cultivars. Depending on the metal bound in the catalytic active site, three SOD classes in plants were identified: manganese SOD (Mn-SOD), copper/zinc SOD (Cu/Zn-SOD) and iron SOD (Fe-SOD). Although Cu/Zn-SOD is the most plentiful SOD form in plant cells (are available in chloroplasts, mitochondria, cytosol, peroxisomes, as well as in the apoplast), this isozyme was not necessarily affected by foliar application of nanoparticles in all varieties, and it appears that other SOD isozymes may compensate Cu/Zn-SOD roles. Fe-SOD is mainly regarded as a chloroplastic enzyme was more responsive to foliar treatment in all varieties. The results strongly imply that Fe-SOD can be introduced as suitable biomarker for evaluating the nanoparticles effects on enzymatic antioxidant system. Mn-SOD is localized in mitochondria and peroxisomes (Leonowicz *et al*., 2018) and it was less responsive to nanoparticles. These results partly suggest that the activity of chloroplast isozymes is more affected by the applications of nano-stimulants. In our view, the most compelling explanation for the present set of findings is that foliar application of Si-NPs in Hydromel and utilization of Ti-NPs in 'Alonso' significantly affected redox status by increasing the activity of copper/zinc SOD and iron SOD and dismutation of superoxide anion radical, receptivity. Although the present results clearly support effects on NPs effects on SOD activity, the role of antioxidant enzymes in redox signaling complicates interpretations. Catalases are efficient scavengers of H_2O_2 and are complementary for antioxidant effect of superoxide dismutase (Martins and English, 2014). Our finding showed that the effect of Si-NPs on CAT was incremental in all varieties. The present results are consistent with Siddiqui *et al.* (2014) who revealed that Nano-silicon dioxide mitigates the adverse effects of salt stress on *Cucurbita pepo* L. by increase the activity of some antioxidant enzyme such as CAT. The silicon within the plant, in addition to increase CAT activity directly also increases the lignification of the cell wall in xylems and can results in xylem openings and strengthen the plant's nutrient transport system and indirectly supports the antioxidant status of the plant (Asghari *et al*., 2018). Our finding revealed that foliar application of Si and Ti-NPs increased guaiacol peroxidases (GPX) in more responsive varieties ('Nathalie' and 'Alonso'). A possible explanation for this might be that GPX is ubiquitous haem-containing enzyme participating in many physiological processes such as ROS scavenging. Some authors have speculated that nanoparticles treatments triggered the accumulation of reactive oxygen species, increased production of stress enzymes that ultimately lead to a better and more stable redox status (Basahi, 2021). Despite the stimulation of the antioxidant system in the rapeseeds plants initiated by nanoparticles spray, it appears that there was a significant lipid peroxidation activity and MDA production especially in Alonso variety. Ti-NPs foliar spray decreased Hydrogen peroxide (H_2O_2) significantly in all varieties. H_2O_2 is produced from superoxide anion ($O_2\bullet^-$) by SOD through a dismutation reaction then can be scavenged by CAT. However, this is not the end of the road and it is possible that hydroxyl radical ($\bullet$OH), the most reactive and perilous ROS, is formed from H_2O_2 in the presence of metal ions. The hydroxyl radical plays an important role in the reactions of lipid peroxidation (Ito *et al.*, 2019). Although the activity of some key enzymes, such as CAT, GPX, and APX increased by NPs treatments, it is likely that other group of ROS scavengers are still faced with some restrictions and have not been affected by NPs and could not alleviate the lipid peroxidation. The ROS scavenging system is a large and complex network in which, in many cases, there is a close cross talking between enzymatic and non-enzymatic antioxidants. To accurately identify the effect of nanoparticles, it is necessary to study a large part of antioxidant systems.

Conclusions

Results showed that the Si and Ti-NPs had varying impact on the enzymatic antioxidant status of *Brassica napus* varieties. Balanced and timely nutrition of oilseed rape gives positive effects both on the yield and on its oil content. In order to increase resistance to drought, the application of Si and Ti-NPs stimulates faster plant development, improves flowering, and prevents abortion of formed flowers, enables better flowering and pod setting. APX activity increased by Si- NPs in all evaluated varieties. Our results showed that genetic differences between varieties play a very important role in how response to nanoparticle treatments. Natalie and Alonso varieties showed a more noticeable response to foliar spraying treatments. Superoxide dismutase isozymes and protein content significantly stimulated by Ti-NPs. Foliar application of Si- NPs had a relatively stable and improving effect on the activity of CAT, APX and GPX in all cultivars. The foliar application of NPs minimized the oxidative stress, which was evidenced by the low level of MDA. Our result suggested that both nanoparticles can be used as suitable choices for designing the ROS scavenging stimulants for rapeseeds fields in semi-arid region. Further research should be focused on the effect of genetic differences between varieties on response of antioxidant systems to nanoparticle foliar application.

Authors' Contributions

Conceptualization, V.S., B.D., S.E., M.J., N.S.; methodology, V.S., J.M.; software, V.S., B.D.; validation, J.M., V.S., B.D.; formal analysis, V.S., S.E., M.J.; investigation, V.S., J.M., N.S.; resources, J.M.; data curation, V.S.; writing–original draft V.S., B.D., S.E., M.J., N.S., preparation, J.M.; V.S; B.D..; writing–review and editing, V.S., B.D., S.E., M.J., N.S.; visualization, V.S., M.J.; supervision, B.D., V.S., S.E.; project administration, J.M.; B.D.; funding acquisition, V.S., D.B. All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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