

## Physiological Parameters of Potato Explants as Affected by Silicon Nanoparticles under Drought Stress

Ihab El-tahan<sup>1</sup>; Enas Safaa Azab<sup>1</sup>; Eman Abdelrazik<sup>2</sup>; Mohamed Elhamahmy<sup>\*1</sup>

<sup>1</sup>Agricultural Botany Department, Faculty of Agriculture, Suez Canal University, Ismailia, Egypt

<sup>2</sup>Botany and Microbiology Department, Faculty of Science, Suez University, Suez, Egypt

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**Abstract:** Potential of growth and microtuburization of four potato cultivars (Hermes, Charlotte, Inova and Maris Peer) treated with two concentrations of Si-NPs (200 and 400 ppm) exposed to drought stress, induced by polyethylene glycol (PEG 10%) were studied. In general, Si-NPs at both concentrations enhanced most vegetative parameters of potato explants compared to PEG-stressed ones. Maximum stem length (16.9 cm), both number of leaves (18) and nuds (20)/explant were recorded in 200 ppm of Si-NPs treated explants of Inova cv. with synthesizing of the highest number of protein bands in microtuber (11) separated by SDS-PAGE and maximum width of sieve elements (6 $\mu$ m) of phloem tissue examined by TEM. Also, low concentration of Si-NPs treated Hermes cv. explants gave the highest values of FW of explant (0.76g), minimum period for microtuber initiation (71d), maximum number (2.3) and FW (2.3g) of microtuber/explant which attributed with the highest number of separated protein bands in shoots (11). Si-NPs at high concentration (400ppm) gave the maximum thickness of 5 layers of phelloderm (275  $\mu$ m) under phellem with large and abundant starch grains and maximum length of both sieve element (10 $\mu$ m) and companion cells (9  $\mu$ m) of phloem. It can recommend that, production of Si-NPs treated microtubers, under drought stress could be tolerant to drought and it must be evaluated under field conditions.

**Keywords:** *Solanum tuberosum* L., vegetative parameters, protein electrophoresis, histology, TEM

### INTRODUCTION

Potato (*Solanum tuberosum* L.) is an annual vegetable crop belongs to the family Solanaceae. Currently, it is the fourth largest stable food crop after maize, wheat, and rice. The world harvest area in 2019 was 17 million hectares (ha  $\approx$  2.38 feddan), produced about 370 million ton. China was the largest producer of potato followed by India, Russia, Ukraine, and USA. In Egypt, the cultivated area was 175161 hectares produced about 5 million ton with 28 ton as average of hectare. About 684735 tons of potatoes were annually exported from Egypt with a value of about 266 million dollars (FAO-STAT, 2019).

Potato asexually cultivated using tubers which annually imported from different European countries for summer plantation. Egypt annually imported about 152198 tons, with a value of about 103 million dollars. Tuberization is a highly complex developmental process, which can be induced under *in vitro* condition, for saving importation of tubers and produce disease free potato (Morais *et al.*, 2018). Microtubers are very appropriate and easy to transport, and it can be easily stored for long time. Therefore, it is necessary to establish a protocol for *in vitro* production of microtubers for rapid multiplication. Researchers used different materials such as plant growth regulators, sucrose, and nutrients for *in vitro* induction of microtuber in potato (Hossain and Sultana, 1998).

Potato is susceptible plant to drought stress, which attributed with negative effect on the physiological status of plant and indeed reduced the plant growth and tuber formation (Monneveux *et al.*, 2013). Plant under abiotic stress formed its reproductive parts as tuber, to save its offspring (Bundig *et al.*, 2016). Nowadays, nanoparticles (NPs) such as Si have been involved in multi-application of the plant biotechnology

which has positive effect on plant growth and productivity under normal and stress conditions (Shang *et al.*, 2019). Silicon has beneficial effects on different crops, mainly under biotic and abiotic stresses. Silicon can affect biochemical, physiological, and photosynthetic processes and, consequently, alleviates drought stress (Carlos *et al.*, 2009). Si-NPs was a synthesized amorphous silica powder containing silica particles with nano size-diameters ranged from 10–100 nm, commonly used as an antifungal agent, biopesticide and agro-fertilizer (Laane, 2018). Application of Si at 35 ppm was increased the leaf area and pigment concentration (chlorophylls and carotenoids), as well as the photosynthesis and transpiration rates in potato (Pilon *et al.*, 2013). Potato treated with 8–32 ppm of Si showed increment of yield by 20% and leaf size, chlorophyll content as well as nutrient status were improved significantly (Khan *et al.*, 2017). An increase in drought tolerance by selecting superior potato cultivars using new biotechnology approaches and application of nanomaterial could improve the productivity and profitability of potato (Gowayed *et al.*, 2017). Although, researchers have been carried out on microtuber production in potato, very little attention has been paid on the *in vitro* tuberization with nanoparticles under drought stress to establish suitable regeneration protocol. Drought-induced microtubers have to low water requirements as well as it will be valuable for human nutrition.

Therefore, this research aimed to study the effect of Si-NPs with two concentrations (200 and 400 ppm) on growth parameters and microtuberization of four potato cultivars (Hermes, Charlotte, Inova and Maris Peer) under drought stress induced by polyethylene glycol (PEG 10%). Protein profile of shoots and microtubers was also investigated. Macro and micro modification of microtubers tissues using light

\*Corresponding author e-mail: mhamahmy@yahoo.com

microscope and transmission electron microscope, respectively were evaluated.

## MATERIALS AND METHODS

### Potato Cultivars and *in Vitro* Culture:

Four potato cultivars (Hermes, Charlotte, Inova and Maris Peer) were obtained from the Agricultural Daltex corporation, El-Salhia El-gededa, El-Sharkia, Egypt. The experiment was conducted in January 2019 to March 2020, at the Laboratory of plant tissue culture, Department of Agricultural Botany, Faculty of Agriculture, Suez Canal University. Tubers were washed by Tap water to release dusts and soil particles, then washed by distilled water three times. Clean tubers were immersed in Clorox 10% (sodium hypochlorite 5%) for 3 min., then washed by distilled water 3 times, dipped in 70% of ethanol for 30 sec., washed by distilled water 3 times and finally air dried. To break tuber dormancy, tubers were soaked in 50 ppm of GA<sub>3</sub> (dissolved in 70% ethanol) for 3 hours, then air dried, incubate in a dark room for one week at room temperature (21-26°C) and humidity 60-70% to obtain sprouts with 3-4 mm length (Momena *et al.*, 2014).

Bud sprouts (3-4 mm long) were sterilized with distilled water 5 times, followed by dipping in 70% of ethanol for 30 sec. with continuous shaking, washed by distilled water 5 times, then 10% of Clorox with two drops of Tween-20 for 3 min., washed with sterile water for 5 times. After that it was dipped in HgCl<sub>2</sub> (0.1%) with two drops of Tween-20 for 5 min., washed with sterile water for 5 times. Five explants/jar were cultured in hormone-free MS medium (Murashige and Skoog, 1962). After pH adjustment at 5.7, the medium was supplemented with 30 g/ L of sucrose, 7 g/ L of agar and maintained at 24 ± 1°C, humidity 60-70% under light conditions (3000 lux, 16 h/ day). Explants were grown in 30 ml/ jar of MS for 70 d to reach maximum length (6-8 cm) or 8-10 nodes. Five explants/ jar were subculture, only one time for another 70 d to reach the same length.

### Drought Stress Induction, *in Vitro* Microtuberization and Nanoparticles Application: -

After 70 d from subculture, explants were exposed to Polyethylene glycol (PEG-6000, Sigma) at 10% to initiate osmotic potential -1.48 MPa, as described by (Michel and Kaufmann, 1973). Osmotic potential of each concentration was also measured by EC-meter (TDS-Digital Meter). PEG was dissolved in liquid MS (LMS) contained 8% of sucrose and 4 ppm of BAP (dissolved in KOH, 1N), to induced microtubers and used as control.

Nanoparticles (Sigma) with 100 nm size, Si Hydrophilic at 200 and 400 ppm were photographed using TEM (JEOL, Japan) to ensure its nano-dimensions and transportation inside potato cells as shown in Fig. (1).

NPs were added to LMS, warmed at 50°C for 3 min using water bath, then stirred with magnetic stirrer for 2 hours for complete dissolving. Each jar (150 ml) contained 30 ml of LMS with different concentration of

NPs, or not (control), and incubated in the dark conditions for 90 d at 18-20°C, 60-70% of humidity. Each treatment was replicated 10 times (10 Jar) and each Jar contained 5 explants. After 160 days from subculture, explants were harvested and washed with distilled water to remove residual agar and dried by filter paper. Some vegetative and molecular characters were estimated as follow: -

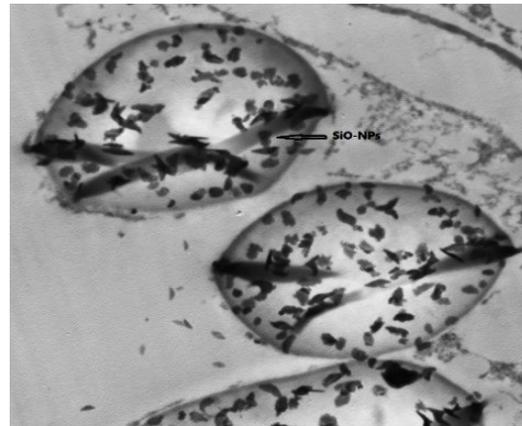


Fig. (1): Overall view of Si-NPs under TEM

### Vegetative and Microtubers Parameters:

Stem length (cm) of explant, number of shoots / explants, lateral shoots/ jar, leaves/ explant, nodules/ explant and fresh weight (g) of explant were measured. Period (d) required for tuber initiation, number of microtubers/explant and per jar, fresh weight (g) of microtubers/explant and per jar were also investigated.

### Histological Investigations by Light Microscope (LM) and Transmission Electron Microscope (TEM):

Potato cubes (2 x 2mm) were fixed in glutaraldehyde (GA) at 5 % dissolved in 0.1 M of cacodylate buffer (pH, 7.2), then washed by phosphate buffer solution. The cubes were refixed by osmic acid, washed by phosphate buffer, followed by dehydration in a graded ethanol series 30, 50, 70, 90 and absolute. Before polymerization using epoxy resin, it immersed in Aceton: ethyl (1:1), Acetone, Aceton: Risine (1:1). Thin sections, 4 µm, were cut (Ultramicrotome RMC, POWER TOME XL, USA) and stained with toluidine blue. Sections were photographed using LEICA DM500 microscope. Ultra-thin sections were stained with uranyl acetate and lead citrate. The sections were examined in a JEOL, JEM-1200 EX II transmission electron microscope (JEOL, Japan) at 80 KV (Sjoo *et al.*, 2009).

### Protein Profile by SDS-PAGE: -

One dimensional SDS-PAGE gel electrophoresis as described by Laemmli (1970) was used to separate the soluble proteins in shoots and microtubers of potato. Two hundred milligram of shoots or microtubers was detached in 1 ml of 10% of SDS with 100 µl β-mercapto ethanol for 15 min, followed by centrifuge at 11000 rpm for 10 min at 4°C. Then, 20 µl of the extract was mixed with 20 µl of SDS-loading sample buffer (SDS 4%, β-mercapto ethanol 3%, glycerol 20%, Tris HCl (50 mM

at pH 6.8) and bromophenol blue traces), warmed at 96°C for 3 min and 10 µl aliquot was used (10 µl of protein/lane). The resolving and stacking gels were prepared according to the standard protocol (Davis, 1964). The electrode buffer composed of 50 mM Tris, glycine (0.384 M) and SDS (0.1%). The protein bands were stained with Commassie Brilliant Blue R-250 dye (0.2% solution, freshly prepared in 45% methanol, 10% glacial acetic acid and 45% distilled water) at room temperature overnight. Gel was washed to remove the excess of staining solution in acetic acid (7%) and distilled water. The blue-stained protein bands were scanned densitometrically at 600 nm using standard marker protein (Pharmacia), ranged from 270 to 6.5 KDa.

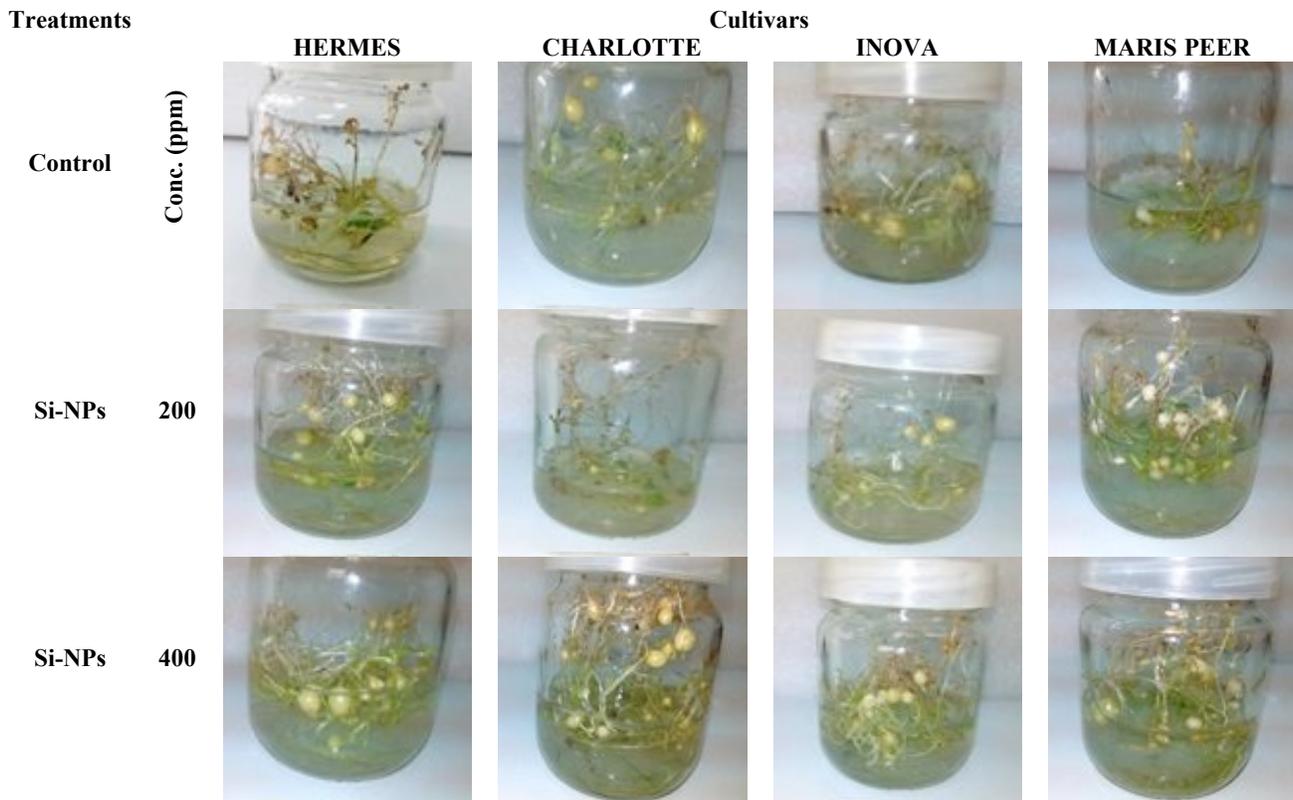
#### Statistical Analysis:

All treatments (Two concentrations of each Si-NPs as well as control of four potato cultivars) were replicated 10 times in a randomized complete blocks design. Analysis of variance (one-way analysis; ANOVA) and means comparisons (Duncan's multiple range tests, 5%) were performed using the MSTAT-C statistical package (Freed *et al.*, 1991).

## RESULTS AND DISCUSSION

### Effect of Si-NPs on Vegetative Characters:

Results revealed that Si-NPs at low concentration (200 ppm) recorded the maximum stem length (16.9 cm), both number of leaves/ explant (18) and nodules/ explant (20) under drought stress in Inova cv. explants (Table 1 and Fig. 2).



**Fig. 2:** Overall growth of potato of four cultivars after application of Si-NPs

PEG-stressed Inova cv. explants gave the highest number of shoots/ explant (2.3) and plantlets/ jar (11) compared to other treatments. Fresh weight (FW) reached to maximum values of both explant and explants/jar (0.76 and 5.83 g, respectively) in Hermes cv. explants treated with 200 ppm of Si-NPs. Results were agreed with Gowayed *et al.*, (2017) who found that, both shoot and root length, number of roots, callus fresh weight were increased in SiO<sub>2</sub>-NPs treatment at 50

ppm in both potato Proventa and Santecvcs. under salt conditions. Positive effect of Si on plant growth was due to increase GA<sub>3</sub> concentration which encourage the cell division and elongation as in *Salvia splendens* under high temperature as reported by Soundararajan *et al.*, (2014). The four cultivars were differed in response to drought, induced by PEG on explants from chlorosis to albino symptoms.

**Table (1).** Effect of Si-NPs application on vegetative characters of plantlets of potato cultivars under drought stress after 160 d from subculture

Cultivars	Treatments	Conc. (ppm)	Stem length (cm)/ plantlet	Average number of			Fresh weight (g) of		Drought symptoms of PEG on plantlets	
				Shoots/ explant	Plantlets/ jar	Leaves/ plantlet	Nods/ plantlet	Plantlet		Plantlets/ jar
Hermes	Control		9.4 bc	2 b	10.3 ab	8.7 c	10.3 c	0.23 c	2.40 ab	Chlorosis
	Si-NPs	200	15.4 ab	1.7 ab	7.7 cd	17 ab	18 ab	0.76 a	5.83 a	Albino
		400	15.5 ab	1.3 ab	7 cd	16.7 ab	18 ab	0.51 abc	3.43 ab	Chlorosis
Charlotte	Control		8.4 bc	1 b	5 e	8 c	9 c	0.47 abc	2.36 ab	Albino
	Si-NPs	200	10 bc	1 b	6.7 de	10.3 c	10.7 c	0.33 bc	2.19 ab	Chlorosis
		400	13.5 bc	1.3 ab	7 cde	14 abc	15.3 abc	0.66 a	4.58 ab	Chlorosis
Inova	Control		9.7 abc	2.3 a	11.3 a	11.3 bc	12 bc	0.30 bc	3.43 ab	Chlorosis
	Si-NPs	200	16.9 a	2 ab	9.3 bc	18 a	20 a	0.55 ab	5.15 ab	Albino
		400	11.7 abc	2 ab	8.3 cd	12.3 abc	13.3 bc	0.48 abc	4.0 ab	Albino
Maris peer	Control		7.4 c	1 b	5 e	9.3 c	10 c	0.24 c	1.19 b	Chlorosis
	Si-NPs	200	9 bc	1.7 ab	8 cd	9 c	10 c	0.32 bc	2.63 ab	Albino
		400	13 abc	1.3 ab	7 cde	14 abc	14.7 abc	0.66 a	4.91 ab	Chlorosis

**Table (2).** Effect of Si-NPs application on microtubers parameters of plantlets of potato cultivars under drought stress after 160 d from subculture

Cultivars	Treatments	Conc. (ppm)	Period (d) required for tuber initiation	Number of microtubers		Fresh weight (g) of microtubers		Tuber	
				/ Plantlet	/ Jar	/ Plantlet	/ Jar	Shape	Color
Hermes	Control		74 f	1 b	13 bcd	0.045 b	0.46 bcd	Ovate and circular	Reddish brown
	Si-NPs	200	71 g	2.3 a	18 abc	0.158 a	1.21 a		Cream
		400	74 f	2 ab	13.7 a-d	0.095 ab	0.63 bcd		Cream
Charlotte	Control		86 a	2 ab	9.7 cd	0.084 b	0.42 cd	Ovate and oblong	Cream
	Si-NPs	200	71 g	2 ab	12 bcd	0.078 b	0.51 bcd		Reddish brown
		400	71 g	2.3 a	16 a-d	0.093 ab	0.63 bcd		Cream
Inov	Control		81 b	2 ab	23 a	0.083 b	0.96 ab	Ovate and oblong	Reddish brown
	Si-NPs	200	77 d	2.3 a	21 ab	0.074 b	0.68 a-d		Cream
		400	73 f	2 ab	18 abc	0.078 b	0.65 bcd		Cream
Maris peer	Control		81 b	2 ab	8.3 d	0.048 b	0.24 d	Ovate and circular	Reddish brown
	Si-NPs	200	78 c	1 b	9.7 cd	0.063 b	0.52 bcd		Reddish brown
		400	75 e	1.7 ab	12 bcd	0.112 ab	0.86 abc		Cream

**Effect of Si-NPs on Microtubers Parameters:**

Si-NPs at low concentration-treated explants required the minimum periods (71 d) to initiate the microtubers in Hermes and Charlotte cvs. earlier by 15 d compared to PEG-stressed explants in Charlotte (Table 2 and Fig. 2). In general, Si-NPs at low concentration-treated explants recorded the highest values of microtubers numbers/explants (2.3), both microtubers fresh weight /explant (0.158 g) and fresh weight /jar (1.21g) in Hermes cv..High concentration of Si-NPs (400 ppm) delayed the pigmentation of microtubers by reddish brown color.These findings were agreed with Carlos *et al.* (2009) who found that, Si application at 284.4 mg /dm<sup>3</sup> of soil reduced stem and branches lodging and increased potato tuber weight under drought ranged from -0.020 to -0.050 MPa of soil water potential. Also, Saadatian *et al.* (2021) found that, foliar application of Si-NPs at concentration ranged from 0.8 and 3.2 mmol was higher effect than ionized-Si for improving the physiological characteristics and yield of mini-tuber of potato.

**Protein Profile in Shoot by SDS-PAGE:**

Table (3) and Fig. (3) showed different separated protein bands with molecular weight ranged from 96 to 14 KDa found in different potato cultivars. Response of cultivars were differed in presence and absence of specific protein bands according to Si-NPs concentration. Total of separated protein bands ranged from 6 to 11 bands and the maximum number of bands were recorded in 200 ppm of Si-NPs treated explants of Hermes and Charlotte cvs. Bands with molecular weight 34, 29, 24, 23 and 14 KDa were found in all treatments as well as PEG-stressed control of all four cultivars

under study. Both protein bands with MW 35 and 19KDa unfound in PEG-stressed explants but found in both low and high Si-NPs concentrations in all cultivars except Hermes cv.Bands with molecular weight about 40 KDa may be patatin and with 25KDa called protease inhibitors as described by Liu *et al.* (2003). However, Gowayed *et al.* (2017) detected a specific protein band, characterized sensitive cultivars of potato, which can be used as selective marker for tolerant strains. Elsadany *et al.* (2019) showed that, protein bands can be present or absent according to potato cultivar and exposure to gamma rays under salinity screening.

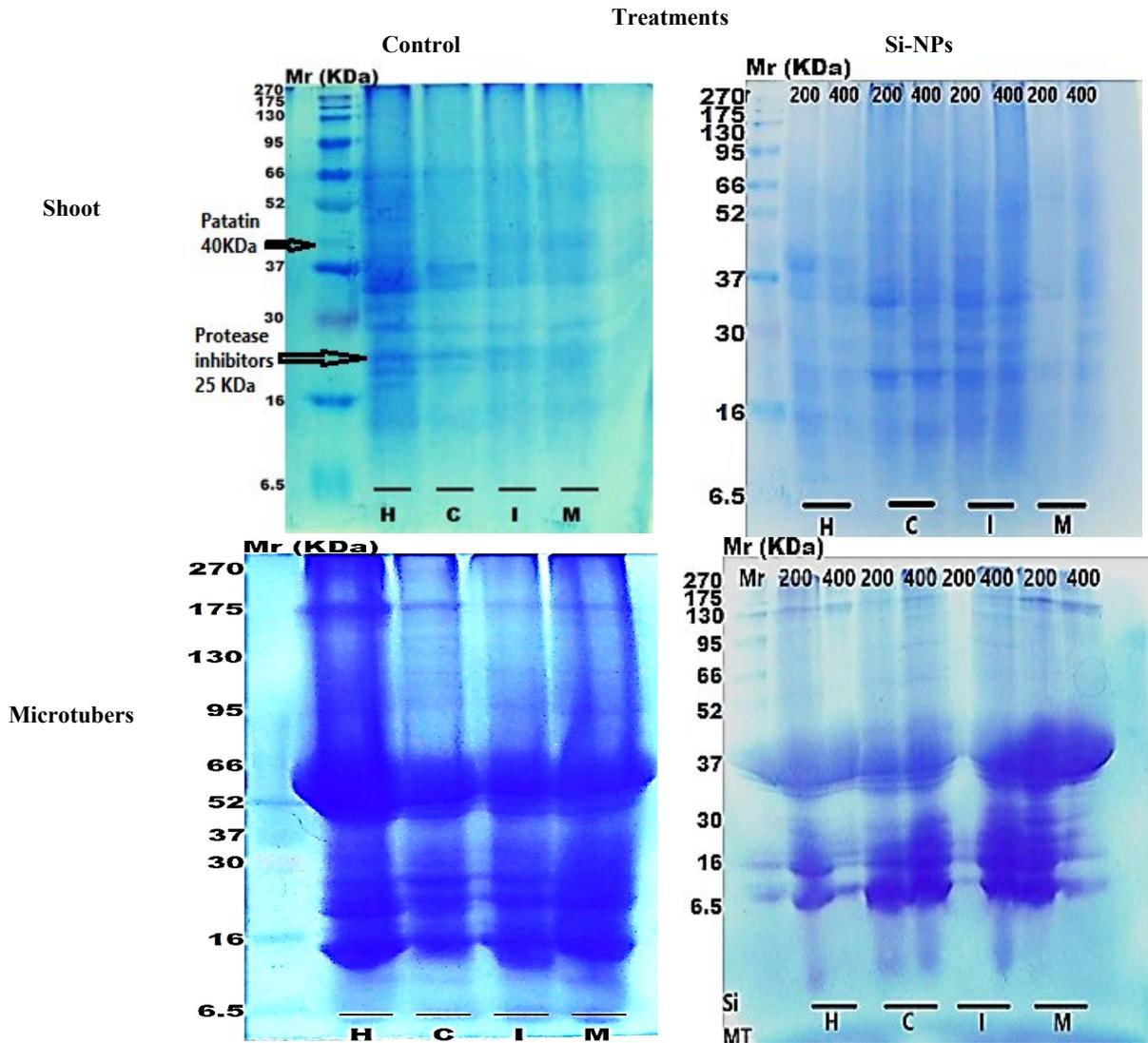
**Protein profile in microtubers by SDS-PAGE:**

Different protein bands with molecular weight ranged from 162 to 4.5 KDa were separated from microtubers in different potato cultivars as shown in Table (4) and Fig. (3). Total of separated protein bands ranged from 6 to 11 bands and the maximum number of bands were recorded in high concentration of Si-NPs treated-explants of Inova and Charlotte cvs. Bands with molecular weight 59, 31 and 25 KDa were found in all treatments as well as PEG-stressed control of all four cultivars under study. protein band with MW 105 KDa undetected in PEG-stressed explants but showed in low Si-NPs treatment in all cultivars except Inova cv. Protein bands with MW 34 and 4.5 KDa were found in PEG-stressed explants and unfound in low concentration of Si-NPs in all cultivars except Maris Peer cv. Carlos *et al.* (2009) found that, application of Si at 284.4 mg/dm<sup>3</sup> of soil decreased total sugars and soluble proteins concentrations in the leaves under drought regime ranged from -0.020 to -0.050 MPa of soil water potential.

**Table (3).** Ideogram of protein profile as affected by Si-NPs in shoot of potato explants under drought stress

Band No.	M.W (KDa)	Cultivars											
		Hermes			Charlotte			Inova			Maris peer		
		Treatments											
		Control	Si-NPs		Control	Si-NPs		Control	Si-NPs		Control	Si-NPs	
	200 ppm	400 ppm		200 ppm	400 ppm		200 ppm	400 ppm		200 ppm	400 ppm		
1	96	1	1	-	1	1	1	-	1	1	-	1	1
2	68	1	1	1	-	1	-	-	1	1	1	1	1
3	53	-	1	1	-	1	-	1	1	-	1	-	-
4	38	-	1	1	1	1	-	-	-	1	-	-	1
5	35	1	1	1	-	1	1	-	1	1	-	1	1
6	34	1	1	1	1	1	1	1	1	1	1	1	1
7	29	1	1	1	1	1	1	1	1	1	1	1	1
8	24	1	1	1	1	1	1	1	1	1	1	1	1
9	23	1	1	1	1	1	1	1	1	1	1	1	1
10	19	1	1	1	-	1	1	-	1	1	-	1	1
11	14	1	1	1	1	1	1	1	1	1	1	1	1
<b>Total</b>		<b>9</b>	<b>11</b>	<b>10</b>	<b>7</b>	<b>11</b>	<b>8</b>	<b>6</b>	<b>10</b>	<b>10</b>	<b>7</b>	<b>9</b>	<b>10</b>

[(-): Absent; (1): High intensity].



**Fig. (3):** Protein profile as affected by Si-NPs, in shoots and microtubers of four potato cultivars after 160 days of PEG (10%) exposure., [H (Hermes); C (Charlotte); I (Inova); M (Maris peer); Mr (Marker)].

**Table (4):** Ideogram of protein profile as affected by Si-NPs in microtubers of potato plantlets under drought stress

Band No.	M.W (KDa)	Cultivars											
		Hermes			Charlotte			Inova			Maris peer		
		Treatments											
		Control	Si-NPs		Control	Si-NPs		Control	Si-NPs		Control	Si-NPs	
	200 ppm	400 ppm		200 ppm	400 ppm		200 ppm	400 ppm		200 ppm	400 ppm		
1	162	1	1	1	1	1	1	1	-	1	1	1	1
2	105	-	1	-	-	1	1	-	-	1	-	1	-
3	74	1	1	1	-	1	1	1	1	1	1	1	1
4	59	1	1	1	1	1	1	1	1	1	1	1	1
5	54	1	1	1	1	1	1	1	-	1	1	1	1
6	34	1	-	1	1	-	1	1	-	1	1	1	1
7	31	1	1	1	1	1	1	1	1	1	1	1	1
8	25	1	1	1	1	1	1	1	1	1	1	1	1
9	22	1	1	1	1	1	1	1	1	1	1	-	-
10	14	1	1	1	-	1	1	1	1	1	1	1	1
11	4.5	1	-	1	1	-	1	1	-	1	1	1	-
<b>Total</b>		<b>10</b>	<b>9</b>	<b>10</b>	<b>8</b>	<b>9</b>	<b>11</b>	<b>10</b>	<b>6</b>	<b>11</b>	<b>10</b>	<b>10</b>	<b>8</b>

[-]: Absent; [1]: High intensity].

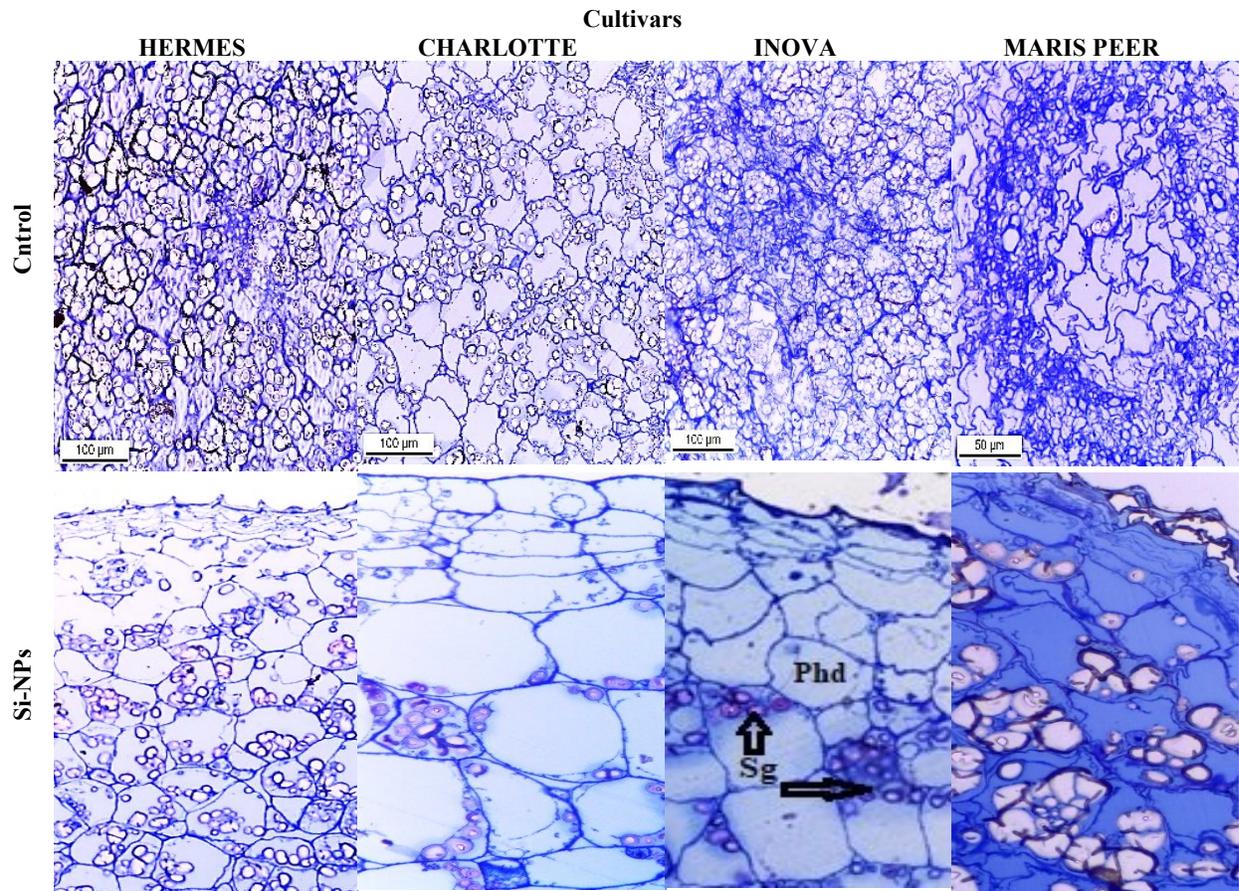


Fig. (4): Light microscope investigation of microtubers tissues of potato as affected by Si-NPs at high level of Si-NPs (400 ppm), [Ph (Phellem); Phd (phelloderm); Sg (Starch grains)].

Table (5): Effect of Si-NPs on histology of potato microtubers

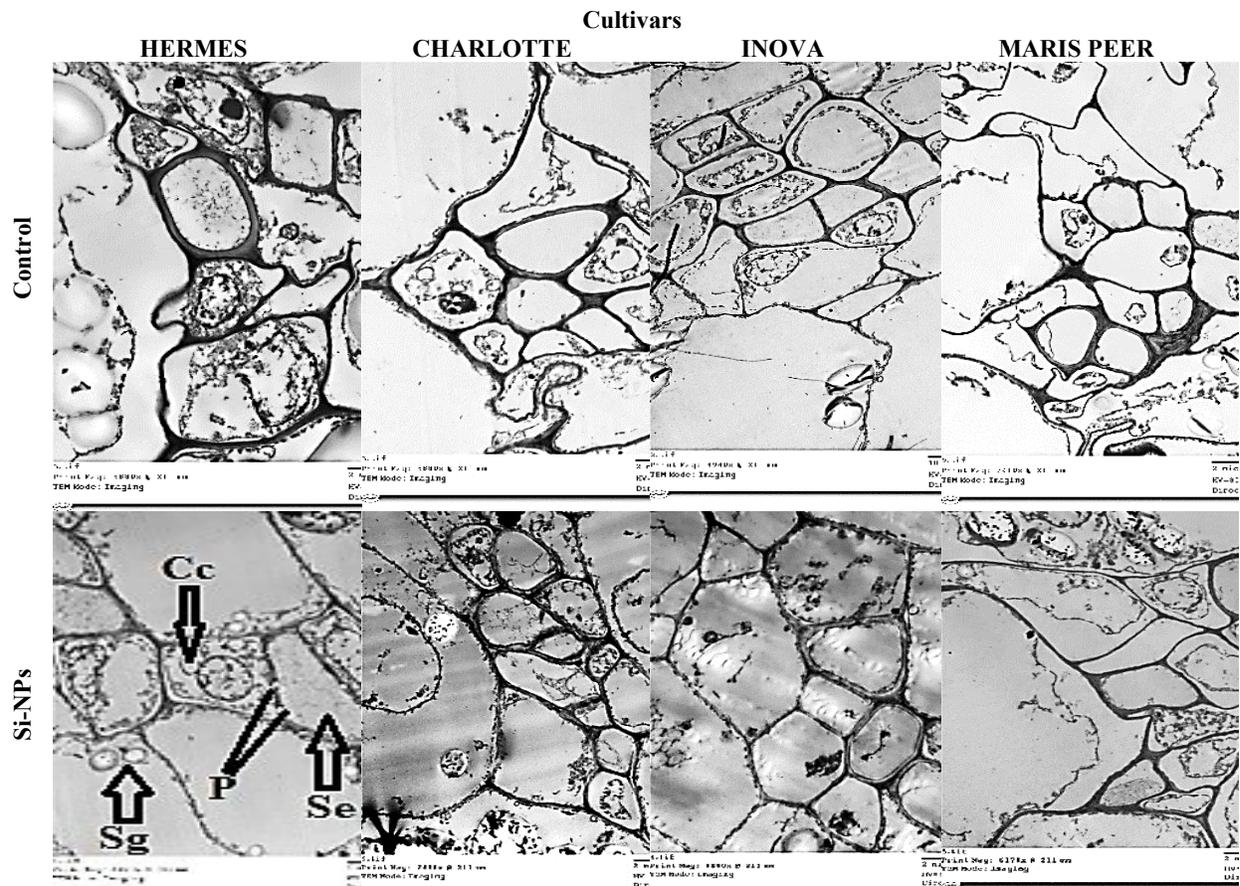
Cultivars	Treatments	Conc. (ppm)	Phellem		Phelloderm		Starch grains	
			Differentiation	Thickness (µm)	Cell status	Thickness of 5 layers (µm) under phellem	Abundancy	Shape
Hermes	Control		+	16.7 b	Turgid and symmetric	208.3	Low	Small
	Si-NPs	400	+	17.4 b	Turgid and symmetric	200 c	Abundant	Large
Charlotte	Control		+	17.4 b	Turgid and symmetric	208.7 b	Moderate	Medium
	Si-NPs	400	-	0 f	Turgid and symmetric	166.7 e	Abundant	Medium
Inova	Control		+	34.8 a	Turgid and symmetric	173.9 d	Abundant	Large
	Si-NPs	400	+	8.3 d	Turgid and symmetric	275 a	Abundant	Large
Maris peer	Control		+	4.3 e	Plasmolyzed and Asymmetric	130.4 g	Low	Small
	Si-NPs	400	+	12.5 c	Turgid and Asymmetric	133.3 f	Abundant	Large

### Histological Macro-modification of Microtubers:

Phellem thickness was recorded the maximum value (34.8 $\mu$ m) in PEG-stressed explants of Inova cv. However, Maris Peer cv. may be susceptible to drought due to its PEG-stressed cells were only plasmolyzed and asymmetric compared to other cultivars. Thickness of 5 layers of phelloderm (275  $\mu$ m) under phellem was in high value in 400 ppm of Si-NPs treated explants of Inova cv. Starch grains were low abundancy and small shape in PEG-stressed explants of both Hermes and Maris Peer cvs. but application of Si-NPs at 400 ppm increased its abundancy and shape. Observations were coordinated with **Reeve *et al.* (1969)** who found that, potato skin is composed of suberized phellem cells, the outer component of the tuber periderm. The periderm tissue consists of two additional cell types: a single-cell meristematic layer, the phellogen (cork cambium) that produces the phellem cells and is localized underneath them; and a parenchyma-like phelloderm that is derived from inward cell divisions of the phellogen.

### Histological Micro-modification of Phloem Tissue of Microtubers under TEM: -

Hermes explants of was more affected cultivar by Si-NPs at 400 ppm in length of sieve elements (10  $\mu$ m) and both length and width of companion cell (9 and 7  $\mu$ m, respectively). Explants of Inova cv. treated with 400 ppm of Si-NPs recorded the maximum width of sieve elements (6  $\mu$ m). The highest number of phloem units / vascular bundle (6) was found in explants of both Charlotte and Maris peer cvs. treated with 400 ppm of Si-NPs. Large sieve tube size and highly energy provided- companion cells were very important factors for high photo assimilates loading efficiency to the terminal sinks as tubers (Van Bel, 1996). Si increased the plant drought tolerance because of formation of a silica cuticle double layer under the leaf epidermis which reduces water losses during cuticular transpiration (Gong *et al.*, 2003). Also, it reduced stomatal conductance in relation to turgor loss of guard cells resulting from Si deposition and modified cell wall properties (Zhu and Gong, 2014). Si had strong abilities to extract water from the growth medium through promotion of root elongation (Hattori *et al.*, 2005) and up-regulation of aquaporin genes (Liu *et al.*, 2015).



**Fig. (5):** TEM investigation of microtubers tissues of potato as affected by Si-NPs at high level of NPs (400 ppm)., [Cc (Companion cell); Se (Sieve element); Sg (Starch grains); P (Plasmodesmata)]

**Table (6):** Effect of Si-NPs on phloem description using TEM of potato microtubers

Cultivars	Treatments	Conc. (ppm)	Sieve element ( $\mu\text{m}$ )		Companion cell ( $\mu\text{m}$ )		number of phloem units / vascular bundle
			Length	width	Length	width	
Hermes	Control		9 a	4 bc	5 b	2.3 c	5 ab
	Si-NPs	400	10 a	5.5 ab	9 a	7 a	4 ab
Charlotte	Control		10 a	3 c	6 b	3.5 bc	4 ab
	Si-NPs	400	4.4 c	4.8 ab	3.2 cd	2 c	6 a
Inova	Control		7.14 b	4.6 abc	2.6 d	1.73 c	4 ab
	Si-NPs	400	6.6 b	6 a	4.6 bc	4 b	5 ab
Maris peer	Control		4.3 c	4 bc	3.4 cd	1.73 c	3 b
	Si-NPs	400	6.5 b	5 ab	5.5 b	4 b	6 a

### CONCLUSION

Low concentration of Si-NPs (200 ppm) enhanced most vegetative parameters of Inova cv. explants under drought stress with induction of protein synthesis in microtubers and changing of fine structure of phloem tissue. Explants of Hermes cv. treated with 200 ppm of Si-NPs gave the maximum values of microtubers parameters with enhancement of protein synthesis in shoots and microtubers.

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## الصفات الفسيولوجية للنباتات الصغيرة للبطاطس المعاملة بجزيئات النانوسليكون تحت ظروف الجفاف

إيهاب محمود صالح الطحان<sup>1</sup>، إيناس صفاء إبراهيم عزب<sup>1</sup>، إيمان عبد الرازق محمد<sup>2</sup>، محمد علي محمود الحامح<sup>1</sup>

<sup>1</sup>قسم النبات الزراعي - كلية الزراعة - جامعة قناة السويس - الإسماعيلية - مصر  
<sup>2</sup>قسم النبات والميكروبيولوجي - كلية العلوم - جامعة السويس - السويس - مصر

تم دراسة قوة النمو وتكوين الدرنة الصغيرة في ٤ أصناف من البطاطس (هيرمس - شارلوت - أنوفا - مارس بير) والمعاملة بتركيزين من النانوسليكون ٢٠٠ و ٤٠٠ جزء في المليون تحت ظروف الجفاف والمستحث بواسطة اليولي ايثيلين جليكول بتركيز ١٠%. بصفة عامة أدت إضافة جزيئات النانوسليكون بكلما التركيزين إلى تحسين الصفات الخضريّة مقارنة بالنباتات المعرضة للجفاف بسجل أقصى طول للساق (١٦.٩سم) و عدد كلا من الأوراق (١٨) والعقد (٢٠) لكل نبات صغير في النباتات المعاملة بالتركيز المنخفض من النانوسليكون (٢٠٠ جزء في المليون) للصنف أنوفا مع تسجيل أكبر عدد من حزم البروتين (١١) في الدرنة الصغيرة المفصولة بواسطة التقريد الكهربائي للبروتين مع أقصى اتساع لخلايا الأنبوب الغربالي للحاء (٦ ميكروميتر) من خلال الفحص بالميكروسكوب الإلكتروني. كما سجلت النباتات الصغيرة لـ صنف الهرمس والمعاملة بالتركيز المنخفض من النانوسليكون أعلى قيم للوزن الطازج للنبات (٠.٧٦ جم) وأقل فترة مطلوبة لتكوين الدرنة الصغيرة على النبات (٧١ يوم) من إضافة البيئة السائلة وأقصى عدد من وزن الدرنة الصغيرة (٢.٣ جم) والذي ارتبط بأكبر عدد من حزم البروتين (١١) في الجزء الخضري للنبات. أعطى التركيز العالي من النانوسليكون (٤٠٠ جزء في المليون) أكبر سمك لعدد ٥ طبقات من القشرة الفلينية تحت الفلين (٢٧٥ ميكروميتر) مع غزارة وكبر حجم حبيبات النشا عند فحصها بالميكروسكوب الضوئي وكذلك أقصى طول لكلا من الأنبوبة الغربالية (١٠ ميكروميتر) والخلية المرافقة (٩ ميكروميتر) عند فحصها بالميكروسكوب الإلكتروني. يمكن التوصية بإمكانية إنتاج درنة صغيرة من البطاطس متحملة للجفاف نتيجة المعاملة بجزيئات النانوسليكون والتي يمكن إعادة تقييمها تحت ظروف الحقل.