Integrative effects of biostimulants and salinity on vegetables: Contribution of bioumik and Lithovit®-urea50 to improve salt-tolerance of tomato

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Abstract. The separate and combined effect of lithovit-urea50 and bioumik was tested on salt-stressed tomato crops. Salinity was induced using three different NaCl solutions (2, 4 and 8 dS m⁻¹). Under the salinity effect, all aspects of plant growth were inhibited. Total chlorophyll and carotenoids reduced from mg g⁻¹ FW and 1.1 mg g⁻¹ FW at 2 dS m⁻¹ to reach 1.01 mg g⁻¹ FW and 0.66 mg g⁻¹ FW at 8 dS m⁻¹ in control plants. Plants treated by the combination of both products had the highest chlorophyll and carotenoids content with 2.24 mg g⁻¹ FW and $1.34 \text{ mg g}^{-1} \text{ FW}$, $1.88 \text{ mg g}^{-1} \text{ FW}$ and $1.05 \text{ mg g}^{-1} \text{ FW}$, and $1.39 \text{ mg g}^{-1} \text{ FW}$ and $0.86 \text{ mg g}^{-1} \text{ FW}$ respectively at 2, 4 and 8 dS m⁻¹. Treating plants by this combination maximized flower number, fruit weight, yield and fruit diameter at 2 dS m⁻¹ (17 flowers, 47.93 g, 431.1 g plant⁻¹ and 3.23 cm respectively) and 4 dS m⁻¹ (15flowers, 36.45 g, 291.85 g plant⁻¹ and 2.8 cm respectively). The separate application of bioumik minimized cell electrolyte leakage at 2 dS m⁻¹ (8.82%) compared to control (11.43%). Additionally, plants treated by lithovit-urea and bioumik had the highest relative water content with 107.3%, 96.5% and 91.2% respectively at 2, 4 and 8 dS m⁻¹. N, Ca and Mg in roots were significantly the highest at 2 dS m⁻¹ (4.5%, 2.6% and 0.5% respectively), at 4 dS m⁻¹ (3.74%, 2.49% and 0.48% respectively) and at 8 dS m⁻¹ (3.21%, 2.61% and 0.32% respectively). K content in roots was maximized following the separate application of biournik with 3.21% at 2 dS m⁻¹ and 2.55% at 8 dS m⁻¹. Conclusively, lithovit-urea and bioumik helped plants in tolerating salt-stress with an optimal effect obtained after their combination.

Key words: biostimulants, growth, physiology, salinity, small scale fertilizers, tomato.

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INTRODUCTION

Tomato crop is one of the most important horticultural crops with a total production volume of 180 million tons and a cultivated area of 5 million ha. China, USA and India are the top producers worldwide with 36.6, 12.8 and 11.7 million tons produced annually, respectively (FAOSTAT, 2021). This crop is considered as moderately sensitive to salinity tolerating a nutrient solution not exceeding 2.4 to 4 dS m⁻¹. Above this level, all aspects of plant growth and production is affected (Cuartero & Fernández-Muñoz, 1998; Bustomi et al., 2014). Salinity inhibits the development of roots and aerial parts and causes a drastic reduction in the yielding capacity of stressed tomato plants. The primary adverse effect caused by salinity is the osmotic stress due to the hyperaccumulation of sodium in the root zone and plant parts. This stress inhibits water and vital nutrient movement in the plant and causes subsequent alteration in physiological and metabolic processes including photosynthesis and cell division (Shimul et al., 2014; Zhang et al., 2016; Zhang et al., 2017). Plants have developed various means to withstand salinity such as the endogenous accumulation of organic solutes including sucrose, sorbitol, and mannitol (Flowers & Colmer, 2015) and, enzymatic (ascorbate peroxidase, peroxidase, catalase) and non-enzymatic (glutathione and glutathione reductase) antioxidants (Blokhina et al., 2003). In the same context, researchers have tried to apply exogenously different compounds, similar to those accumulated naturally in stressed-plants, as a method to improve the salt-tolerance of the crop. In previous studies, foliar spraying of osmoprotectans (glycine betaine), auxin-like substances (acetyl salicylic acid), nutrient-rich fertilizers (monopotassiumphosphate) and sugar alcohols was highly efficient (Gul et al., 2017; Sajyan et al., 2018, 2019a, 2019b, 2019c; Issa et al., 2020). Such implementation counteracted the negative impacts of salinity through an improvement in nutrient content, photosynthetic pigments and plant physiology. Consequently, the use of these components maximized the vegetative growth and yielding capacity of stressed plants compared to the non-treated ones.

Other methods adopted included the use of biostimulants as priming material or through direct application on plants. In the agricultural industry, the manufacturing of biostimulants is a witnessing a rapid growth as an efficient tool of yield promoters as well as pre-stress conditioners (Yakhin et al., 2017). The application of plant derived protein hydrolysate increased vegetative performance and boosted the photosynthetic apparatus of salt-stress lettuce crops (Lucini et al., 2015). Similar ameliorative effects were reported following the use of licorice root extract on *Phaseolus vulgaris* under salinity stress (Rady et al., 2019). Furthermore, the use of *Moringa oleifera* extracts enhanced hormones and nutrient content in stressed sorghum, and promoted enzymatic and non-enzymatic antioxidants (Desoky et al., 2018).

Other groups of biostimulants including small sized-biofertilizers are bein g Lately tested on crops subjected to salinity. Such components include lithovit-standard and lithovit-guano 25 which were previously sprayed on tomato, eggplant and pepper under saline conditions, and alleviated the negative effect induced by this abiotic stress (Sajyan et al., 2019d, 2019e; Issa et al., 2020; Sajyan et al., 2020). Products such as lithovit-urea50, a nitrogenous rich fertilizer, were not previously tested under environmental stress. However, previous studies implemented this product in mushroom production mainly *Pleurotus ostreatus*. As a result, lithovit-urea50 applied at different

timing maximized production and qualitative attributes of the produced mushroom (Naim et al., 2020; Sassine et al., 2021). Bioumik is another similar product that has been less applied on vegetables under abiotic stress. Bioumik is a balanced biological method combining microorganisms and micronutrients (iron, zinc, manganese, magnesium, molybdenum, and calcium) of humic and folic acid compounds with amino acids. Accordingly, soil fertility is enhanced and the ability of the plant to absorb vital elements is ameliorated with this product (Biozar, 2021).

Based on their composition, lithovit-urea50 and bioumik seemed to be highly beneficial when applied to tomato crops irrigated with saline solutions. The current trial aimed to study for the first time the separate and combined effect of lithovit-urea50 and bioumik on the performance of salt-stressed tomato crop.

MATERIALS AND METHODS

Treatments and measurements

Tomato seeds (var. Sila) were sterilized in 0.1% sodium hypochlorite for 30 min, washed with distilled water and sown in plastic trays. After 30 days, uniform seedlings having 3 to 4 true leaves were transplanted in pots containing a mixture of peat and soil. Plants were kept in open-field conditions with a temperature 20 ± 5 °C and a relative humidity of 70% during the growing period which lasted 100 days (from May to August/ 2020). Two products were applied through foliar spraying: lithovit-urea (5 g L⁻¹) and bioumik (5 g L⁻¹). These products were applied in a separate or a combined form on saltstressed tomato irrigated by three different NaCl solutions (2, 4 and 8 dS m⁻¹). Lithoviturea is manufactured by Tribodyn/Germany and has the following composition: 33% CaCO₃, 21% N total nitrogen, 18.5% CaO, 6.5% SiO₂, 1.2% MgO, 0.5% Fe and 0.01% Mn. Bioumik is manufactured by Biozar/Iran and has the following composition: 5% Fe, 3% Zn, 0.36% Ca, 2% Mn, 0.36% Mg, 3% K, 0.1% Mo, 10% humic acids. Products were applied three times during plant growth cycle at 15, 30 and 45 days after transplantation (DAT). Salinity was induced using three different NaCl solutions with different EC levels; 2, 4 and 8 dS m⁻¹. Saline irrigation started at 20 DAT with an interval of 2 days. The EC of the saline solution was continuously monitored. At each salinity level, enough drainage was allowed until obtaining an EC_{water drainage} = EC_{irrigation solution}. Adjustment of ECwater drainage was done using the corresponding saline solution. The experiment was arranged as randomized completely block design with three replications. Two experimental factors with different levels were studied; 'Salinity' including three levels (three NaCl solutions: 2, 4 and 8 dS m⁻¹) and product application including 4 levels (control: no application, lithovit-urea, bioumik and lithovit-urea + bioumik).

During the growth cycle, several measurements were taken to study the effect of salinity and treatments. Vegetative traits included, plant height, leaf number, stem diameter and weight of plants parts. For the determination of fresh weights plants were removed from pots, washed to removed adherent soil, then, separated into roots, stems and leaves. First, fresh weight of different part was measured using a digital balance. Afterwards, plant parts were oven dried at 70 °C until a constant weight was obtained. Additionally, photosynthetic pigments including chlorophyll content and carotenoids were also determined in leaves as described by Sassine et al. (2020). Cell electrolyte leakage and relative water content were measured also on leaves as described by Sajyan et al. (2020). Nutrient content in roots and shoots were measured on ash of roots and

shoots ground, heated to 550 °C and dissolved in diluted HCl with a few drops of nitric acid as described by Cottenie et al. (1982). Relative water content in leaves was measured as described by Mata & Lamattina (2001). Fruit traits included flower number, cluster number, fruit number, fruit fresh weight, yield and fruit diameter.

Statistical analysis

Data was subjected to analysis of variance using Statistical Package for Social Sciences (SPSS) software version 25® software. Means were compared by Duncan's multiple range tests at $P \le 0.05$.

RESULTS

Vegetative traits

Vegetative growth on stressed tomato was inhibited under the effect of salinity. Plant height, leaf number and stem diameter were the lowest at 8 dS m⁻¹ in all plants

(Table 1). Treating plants with lithovit-urea, bioumik and their combination helped in reducing the adverse effect of salinity. In specific, spraying plants with a combination of both products maximized plant height and stem diameter with 70.48 cm and 1.41 cm, 48.63 cm and 1.2 cm respectively at 2 and 4 dS m⁻¹. A separate application of bioumik maximized leaf number under all EC levels.

Fresh and dry weights of plants parts (Table 2) was similarly inhibited with salinity and improved by different treatments. It was observed that spraying tomato with a combination of bioumik and lithovit-urea maximized fresh and dry weight of roots, stems and leaves almost under all EC levels. For instance, fresh weights of roots of plants treated by the combination

Table 1. Vegetative traits of tomato as affected by salinity and treatments

	PH		SD
	(cm)	LN	(cm)
2 dS m ⁻¹ / Control	48.10de	10.00d	1.00c
2 dS m ⁻¹ / lithovit-urea	56.90c	11.75b	1.30ab
2 dS m ⁻¹ / bioumik	61.53b	15.25a	1.20abc
2 dS m ⁻¹ /lithovit-	70.48a	12.00b	1.41a
urea+bioumik			
4 dS m ⁻¹ / Control	40.15h	10.00d	1.07c
4 dS m ⁻¹ / lithovit-urea	47.65de	11.25bc	1.10bc
4 dS m ⁻¹ / bioumik	44.55fg	11.75b	1.20abc
4 dS m ⁻¹ /lithovit-	48.63d	10.00d	1.20abc
urea+bioumik			
8 dS m ⁻¹ / Control	32.50i	8.75e	0.80d
8 dS m ⁻¹ / lithovit-urea	46.40ef	10.00d	1.10bc
8 dS m ⁻¹ / bioumik	44.00g	10.25cd	1.10bc
8 dS m ⁻¹ /lithovit-	43.25g	9.75de	1.20abc
urea+bioumik			

Means (n = 9) followed by the same letter within each column are not significantly different according to Duncan's multiple range tests. PH – plant height; LN – leaf number; SD – stem diameter.

significantly higher significantly compared to control at all EC levels. This ameliorative effect was similarly observed with a less extent in the remaining treatments compared to control at all EC levels. For instance, spraying plants by both products separately or in combination doubled fresh weight of leaves compared to control at 4 and 8 dS m⁻¹.

Table 2. Vegetative traits of tomato as affected by salinity and treatments

	FWR	DWR	FWS	DWS	FWL	DWL
	(g)	(g)	(g)	(g)	(g)	(g)
2 dS m ⁻¹ / Control	6.83de	1.70de	27.47c	6.62bcd	57.33f	12.30d
2 dS m ⁻¹ / lithovit-urea	8.00bc	2.00bc	34.37a	7.62a	95.67ab	13.67cd
2 dS m ⁻¹ / bioumik	8.65b	2.15ab	33.53a	7.52ab	96.30ab	14.30bc
2 dS m ⁻¹ /lithovit-urea+bioumik	10.23a	2.30a	36.00a	6.76abcd	100.33a	16.39a
4 dS m ⁻¹ / Control	6.00fg	1.66de	20.87ef	5.79d	40.47g	10.10e
4 dS m ⁻¹ / lithovit-urea	7.06de	1.83cd	24.20d	6.48cd	84.33de	13.33cd
4 dS m ⁻¹ / bioumik	7.45cd	1.89cd	24.33d	6.01cd	93.33b	14.30bc
4 dS m ⁻¹ /lithovit-urea+bioumik	8.27b	2.20ab	30.63b	6.77abc	91.67bc	15.73ab
8 dS m ⁻¹ / Control	5.40g	1.70de	19.50f	5.88cd	40.00g	10.07e
8 dS m ⁻¹ / lithovit-urea	6.00fg	1.66de	24.33d	6.35cd	80.20e	14.00bcd
8 dS m ⁻¹ / bioumik	6.06fg	1.58e	23.20de	6.08cd	87.17cd	13.40cd
8 dS m ⁻¹ /lithovit-urea+bioumik	6.60ef	1.76de	28.60bc	6.57cd	85.67d	15.00abc

Means (n = 9) followed by the same letter within each column are not significantly different according to Duncan's multiple range tests. FWR – fresh weight of roots; DWR – dry weight of roots; FWS – fresh weight of stems; DWS – dry weight of stems; FWL – fresh weight of leaves; DWL – dry weight of leaves.

Photosynthetic pigments, cell electrolyte leakage and relative water content

Under salinity effect, photosynthetic pigments including chlorophyll a, b and carotenoids were significantly reduced (Table 3). In control plants, total chlorophyll and carotenoids reduced from 1.6 mg g⁻¹ FW and 1.1 mg g⁻¹ FW, respectively at 2 dS m⁻¹ to a reach minimum of 1.01 mg g⁻¹ FW and 0.66 mg g⁻¹ FW, respectively at 8 dS m⁻¹.

Table 3. Photosynthetic pigments of tomato as affected by salinity and treatments

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	Chl a	Chl b	T Chl	Car
	(mg g ⁻¹ FW)			
2 dS m ⁻¹ / Control	0.98de	0.62c	1.60c	1.10b
2 dS m ⁻¹ / lithovit-urea	1.21bc	0.71b	1.92b	1.11b
2 dS m ⁻¹ / bioumik	1.31ab	0.66bc	1.97b	1.22ab
2 dS m ⁻¹ /lithovit-urea+bioumik	1.42a	0.82a	2.24a	1.34a
4 dS m ⁻¹ / Control	0.87ef	0.41e	1.28de	0.80de
4 dS m ⁻¹ / lithovit-urea	1.15bcd	0.52d	1.67c	0.85d
4 dS m ⁻¹ / bioumik	1.11cd	0.51d	1.62c	0.90cd
4 dS m ⁻¹ /lithovit-urea+bioumik	1.22bc	0.66bc	1.88b	1.05bc
8 dS m ⁻¹ / Control	0.69f	0.32f	1.01f	0.66e
8 dS m ⁻¹ / lithovit-urea	0.90e	0.43e	1.33de	0.80de
8 dS m ⁻¹ / bioumik	0.85ef	0.32f	1.17ef	0.85d
8 dS m ⁻¹ /lithovit-urea+bioumik	0.86ef	0.53d	1.39d	0.86d

Means (n = 3) followed by the same letter within each column are not significantly different according to Duncan's tests. Chl a – chlorophyll a; Chl b – chlorophyll b; T Chl – total chlorophyll; Car – carotenoids.

Reductions caused by salinity were lowered following the application of different products in separate or combined form. These treatments increased significantly the content in photosynthetic pigments of stressed tomato compared to control at all EC levels. A maximum ameliorative effect was detected after spraying bioumik and lithovit-urea in combination. Plants treated by this combination had significantly higher total chlorophyll and carotenoids content compared to all the remaining treatments

including control at all EC levels with 2.24 mg g^{-1} FW and 1.34 mg g^{-1} FW, 1.88 mg g^{-1} FW and 1.05 mg g^{-1} FW, and 1.39 mg g^{-1} FW and 0.86 mg g^{-1} FW respectively at 2, 4 and 8 dS m⁻¹. On the contrary, cell electrolyte leakage was improved by increasing in salinity peaking in control plants at 8 dS m⁻¹ (35.43%) (Table 4). The separate application of

bioumik minimized cell electrolyte leakage at 2 dS m⁻¹ (8.82%) compared to control (11.43%). Additionally, the application of bioumik and lithoviturea in combination minimized this trait at 4 and 8 dS m⁻¹ with 13.4% and 26.6% respectively. Finally, although relative water content was lowered with salinity, however, treating plants by both products improved significantly such trait. comparing between treatments, it was obvious that the combination of both products was optimal on this trait at all EC levels. Plants treated by lithovit-urea and bioumik the highest relative water content with 107.3%, 96.5% and 91.2% respectively at 2, 4 and 8 dS m⁻¹. In general, all treatments induced a significant improvement in relative water content except at 8 dS m⁻¹.

Table 4. Cell electrolyte leakage and relative water content of tomato as affected by salinity and treatments

	CEL (%)	RWC (%)
2 dS m ⁻¹ / Control	11.43fg	91.97cd
2 dS m ⁻¹ / lithovit-urea	9.49g	101.57b
2 dS m ⁻¹ / bioumik	8.82g	100.53b
2 dS m ⁻¹ /lithovit-	8.89g	107.30a
urea+bioumik		
4 dS m ⁻¹ / Control	19.43c	80.87g
4 dS m ⁻¹ / lithovit-urea	15.23de	89.47de
4 dS m ⁻¹ / bioumik	17.47cd	87.43def
4 dS m ⁻¹ /lithovit-	13.40ef	96.50bc
urea+bioumik		
8 dS m ⁻¹ / Control	35.43a	82.60fg
8 dS m ⁻¹ / lithovit-urea	28.40b	84.40efg
8 dS m ⁻¹ / bioumik	26.67b	88.23def
8 dS m ⁻¹ /lithovit-	26.60b	91.20cd
urea+bioumik		

Means (n = 3) followed by the same letter within each column are not significantly different according to Duncan's multiple range tests. CEL – cell electrolyte leakage: RWC – relative water content.

Nutrient content in roots and shoots

As shown in Table 5, the increase in salt-stress from 2 to 8 dS m⁻¹ caused a significant reduction in nutrient content including N, P, K, Ca and Mg in both treated and non-treated plants. On the other hand, a significant increase almost in all macronutrients content was observed following the application of different treatments. The increase peaked following foliar spraying of the combination of lithovit-urea and bioumik. For instance, N, Ca and Mg in roots were significantly the highest at 2 dS m⁻¹ (4.5%, 2.6% and 0.5% respectively), at 4 dS m⁻¹ (3.74%, 2.49% and 0.48% respectively) and at 8 dS m⁻¹ (3.21%, 2.61% and 0.32% respectively). Additionally, K content in roots was maximized following the separate application of bioumik with 3.21% and 2.55% respectively at 2 and 8 dS m⁻¹. P content in roots was significantly improved by product application only at 8 dS m⁻¹ following the application of bioumik alone or in combination with lithovit-urea. When comparing between both products applied in a separate form, it was observed that application of bioumik was slightly better than lithovit-urea. N, K, Ca and Mg content in roots of plants treated by bioumik were higher than those of plants treated by lithovit-urea. The application of different treatments reduced significantly Na accumulation in roots. Na content was minimized following bioumik application at 2 (0.28%) and 4 dS m⁻¹ (0.31%), and lithovit-urea application at 8 dS m⁻¹ (0.45%).

Table 5. Nutrient content in roots of tomato as affected by salinity and treatments

	N (%)	P (%)	K (%)	Ca (%)	Na (%)	Mg (%)
2 dS m ⁻¹ / Control	3.02d	0.63ab	3.04ab	2.21de	0.61c	0.22de
2 dS m ⁻¹ / lithovit-urea	4.00b	0.66a	3.07ab	2.42abcd	0.32g	0.44abc
2 dS m ⁻¹ / bioumik	4.42a	0.59abcd	3.21a	2.45abc	0.28g	0.46ab
2 dS m ⁻¹ /lithovit-urea+bioumik	4.50a	0.50def	2.98bc	2.60a	0.30g	0.50a
4 dS m ⁻¹ / Control	2.55e	0.51cdef	2.59ef	2.11e	0.82b	0.16e
4 dS m ⁻¹ / lithovit-urea	3.22d	0.50def	2.57ef	2.37bcd	0.51def	0.32cd
4 dS m ⁻¹ / bioumik	3.66c	0.56bcde	2.75de	2.45abc	0.31g	0.34bcd
4 dS m ⁻¹ /lithovit-urea+bioumik	3.74c	0.51cdef	2.81cd	2.49ab	0.44f	0.48a
8 dS m ⁻¹ / Control	1.70g	0.45f	2.50f	1.60f	0.96a	0.16e
8 dS m ⁻¹ / lithovit-urea	2.20f	0.49ef	2.52f	2.03e	0.45ef	0.23de
8 dS m ⁻¹ / bioumik	3.00d	0.60abc	2.55ef	2.24cde	0.60cd	0.29de
8 dS m ⁻¹ /lithovit-urea+bioumik	3.21d	0.55bcde	2.47f	2.61a	0.54cde	0.32cd

Means (n = 3) followed by the same letter within each column are not significantly different according to Duncan's multiple range tests.

The content of nutrient in shoots (Table 6) was similarly influenced by salinity and product application. N and Ca in shoots were significantly the highest after the application of a combination of lithovit-urea and bioumik at 2 dS m⁻¹ (4.5% and 3.1% respectively) and at 4 dS m⁻¹ (4.31% and 2.8% respectively). P content did not significantly improve following the application of any treatment at all EC levels.

Table 6. Nutrient content in shoots of tomato as affected by salinity and treatments

			•	•		
	N (%)	P (%)	K (%)	Ca (%)	Na (%)	Mg (%)
2 dS m ⁻¹ / Control	3.44d	0.88a	4.72bc	2.50de	0.41c	0.40cd
2 dS m ⁻¹ / lithovit-urea	4.25bc	0.81a	4.74bc	3.00ab	0.31de	0.58b
2 dS m ⁻¹ / bioumik	4.40ab	0.91a	4.85ab	2.95ab	0.25e	0.60ab
2 dS m ⁻¹ /lithovit-urea+bioumik	4.54a	0.79ab	5.00a	3.10a	0.26e	0.70a
4 dS m ⁻¹ / Control	3.01e	0.66bc	4.50c	2.40e	0.56b	0.32de
4 dS m ⁻¹ / lithovit-urea	4.00c	0.79ab	4.57c	2.66cd	0.35cd	0.43cd
4 dS m ⁻¹ / bioumik	4.25bc	0.79ab	4.70bc	2.77bc	0.40c	0.50bc
4 dS m ⁻¹ /lithovit-urea+bioumik	4.31ab	0.80ab	4.60bc	2.80bc	0.30de	0.50bc
8 dS m ⁻¹ / Control	2.00f	0.55c	3.37e	1.90f	0.66a	0.28e
8 dS m ⁻¹ / lithovit-urea	3.19de	0.59c	3.72d	2.30e	0.52b	0.40cd
8 dS m ⁻¹ / bioumik	3.43d	0.61c	3.80d	2.40e	0.50b	0.45c
8 dS m ⁻¹ /lithovit-urea+bioumik	3.40d	0.57c	3.60d	2.43e	0.53b	0.41cd

Means (n = 3) followed by the same letter within each column are not significantly different according to Duncan's multiple range tests.

K content responded differently according to EC level; it was maximized by the combination of both products at 2 dS m⁻¹ (5%) and by the separate application of bioumik at 4 (4.7%) and 8 dS m⁻¹ (3.8%). Mg was affected similarly by product application. Finally, Na content was significantly reduced compared to control following the application of all treatments. It was the lowest in plants sprayed by bioumik at 2 (0.25%) and 8 dS m⁻¹ (0.5%) and by the combination at 4 dS m⁻¹ (0.3%).

Fruit traits

As shown in Table 7, salinity caused a significant reduction in fruit traits. In control plants, number of flowers, number of fruits, fruit weight, yield and fruit diameter were reduced from 11 flowers, 6fruits, 29.37 g, 176.12 g plant⁻¹ and 2.21 cm, respectively at 2 dS m⁻¹ to reach a minimum of 9 flowers, 4.25 clusters, 25.38 g, 107.95 g plant⁻¹ and 2.02 cm respectively at 8 dS m⁻¹. All treatments applied helped in reducing the adverse effect caused by salinity on these traits. For instance, the number of clusters was increased by 1 to 2 clusters in treated plants compared to control at all EC levels. Additionally, the application of lithovit-urea and bioumik in combination maximized flower number, fruit weight, yield and fruit diameter at 2 dS m⁻¹ (Table 7). Fruit number was maximized by the application of bioumik at 2 (8.25 fruits), 4 (8 fruits) and 8 dS m⁻¹ (7.25 fruits).

Table 7. Fruit traits of tomato as affected by salinity and treatments

	CN	Fl N	Fr N	FW	Yield	FD
	CN	ΓIIN	LI IA	(g)	(g plant ⁻¹)	(cm)
2 dS m ⁻¹ / Control	3.00bc	11.00fg	6.00de	29.37ef	176.12ef	2.21g
2 dS m ⁻¹ / lithovit-urea	5.00a	14.00cd	7.00cd	40.48b	284.10c	2.70bc
2 dS m ⁻¹ / bioumik	4.75a	16.00ab	8.25ab	41.95b	345.25b	3.08a
2 dS m ⁻¹ /lithovit-urea+bioumik	5.00a	17.00a	9.00a	47.93a	431.10a	3.23a
4 dS m ⁻¹ / Control	3.25bc	10.00gh	5.00ef	28.40f	141.85fg	2.30fg
4 dS m ⁻¹ / lithovit-urea	4.00ab	12.00ef	6.00de	31.00e	186.00e	2.60cd
4 dS m ⁻¹ / bioumik	4.00ab	14.00cd	8.00abc	35.00cd	280.13c	2.50de
4 dS m ⁻¹ /lithovit-urea+bioumik	4.75a	15.00bc	8.00abc	36.45c	291.85c	2.80b
8 dS m ⁻¹ / Control	2.50c	9.00h	4.25f	25.38g	107.95g	2.02h
8 dS m ⁻¹ / lithovit-urea	4.00ab	12.00ef	7.00cd	30.63e	214.53de	2.31fg
8 dS m ⁻¹ / bioumik	4.00ab	12.00ef	7.25bc	33.48d	241.35d	2.50de
8 dS m ⁻¹ /lithovit-urea+bioumik	4.00ab	13.00de	6.00de	34.23d	205.93de	2.40ef

Means (n = 9) followed by the same letter within each column are not significantly different according to Duncan's multiple range tests. CN – cluster number; FlN – flower number; FrN – fruit number; FW – fruit weight; FD – fruit diameter.

DISCUSSION

Increasing in salinity had inhibitory effects on all traits. It caused a significant reduction in plant height, leaf number and stem diameter. In previous studies, similar inhibitory effects caused by saline conditions were reported (Sajyan et al., 2018; Mahmoud et al. 2020). As mentioned, such effect is due primarly to an osmotic stress upcoming from salt accumulation in the roots, reducing shoot growth rate and inhibiting cell division and expansion (Liang et al., 1996). High Na+ content in plants reduces the absorption of fundamental nutrient including K, Ca, Mg and others. On pepper crop, an increase in salinity up to 6 dS m⁻¹ caused a reduction in nutrient uptake and, an accumulation in Na content up to two-fold (Sajyan et al., 2020). Abdeldym et al. (2020) reported an inhibition in water movement reducing fresh weights of plants and increasing dry matter. In the current study, comparable negative results were observed including reduction, in fresh weights of plant parts and in macronutrients accumulation (Table 2, 5 and 6).

On the other hand, lithovit-urea50 and bioumik applied separately or in combination induced tolerance of tomato to salinity especially when comparing to non-treated plants. Regarding the separate effect of these products, it was observed that bioumik was better than lithovit-urea. Plants treated with the former product had more or less higher values for almost all the studied traits with some exceptions. In fact, both products include in their composition many nutrients such as nitrogen and others. However, bioumik contains humic, folic and amino acids which are not found in lithovit-urea50.

In fact, bioumik was less applied previously on crops. Therefore, the exact mechanism behind the ameliorative effect is still poorly understood. However, based on the current results, it was observed that the separate use of this product rich in humic, fulvic and amino acids and in nutrients was significantly efficient. Foliar spray of humic acid to leaves promoted growth in cucumber (Canellas et al., 2015; De Hita et al., 2020). According to these reports, an increase in hormonal concentration mainly auxins in roots and shoots was associated with the application of humic substances which might enhanced the rate of growth in all plant parts including roots, leaves and stems. These results were translated in the current study following the application of bioumik especially when observing fresh and dry weights of plant parts. Additionally, increasing in humic substances in plant parts was associated with a decrease in abscisic acid (ABA) contributing also in the stimulation of plant growth. ABA is well known to be associated with leaf senescence and inhibition in shoot growth (Ghanem et al., 2008; Vysotskaya et al., 2018). Other reports also coupled the application of humic substances with an increase in nutrient content (N, K, Ca, Mg, and P absorption) and a decrease in toxic elements including Na in maize and garden cress under salt stress (Elmongy et al., 2018; Kaya et al., 2018). Bioumik containing both organic substances (fulvic and humic acids) and nutrients maximized all nutrient content in roots and shoots of tomato crop subjected to salinity (Table 5 and 6) especially when combined to lithovit-urea50.

Furthermore, the application of bioumik caused a significant increment in photosynthetic pigments which were inhibited by salinity. The possible reason behind this effect is the combination of organic acids with nutrient in plant foliage and roots. This combination was maximized when bioumik and lithovit-urea 50 were combined. In other terms, organic acids found in bioumik might have cause reduction in pH, and a release for cationic element such as Fe required for photosynthesis (Latif & Mohamed, 2016). Similar effects were observed in many previous studies (Akladious & Mohamed, 2018; Kaya et al., 2018). In this way, the enrichment in organic acids increased salt-tolerance of crops through an increase in rubisco enzyme (Latif & Mohamed, 2016). The already discussed effect was boosted after the addition of lithovit-urea50 containing vital elements required for chlorophyll formation such as Fe and Mg. The combination of bioumik with lithovit-urea50 boosted the immunity system of tomato plants by minimizing sodium accumulation starting from roots to shoots, and by maximizing nutrient uptake. This combination also boosted relative water content in leaves reflecting a better water movement in the plant under salt-stress compared to non-treated plants. This stimulatory effect was translated by an increase in weight, size and number of tomato fruits. Consequently, yielding capacity of tomato crop was maximized. Basically, as mentioned in previous sections, the presence of organic acids and nutrients (found in both products) promoted fruit set and growth through a decrease in ABA which cause fruit abscission under salinity. The presence of vital elements in both products such as

calcium and phosphorus ensured good conditions for fruit set and for fruit growth. Similar stimulatory effects were observed following the implementation of lithovit-urea50 in mushroom production at different timings; yields produced were maximized at all flushes. Additionally, the accumulation of nutrient and amino acids was promoted after the use of this product (Naim et al., 2020; Sassine et al., 2021).

The implementation of these products helped plants in withstanding salinity by ensuring a balanced nutritional status coupled with a protected cell membrane reducing electrolyte leakage from cells. In this way, Na uptake was inhibited under saline conditions, and photosynthetic machinery of the plants was re-established.

CONCLUSIONS

From the current study, it was revealed the combination of bioumik with lithoviturea50 maximized almost all the studied traits including vegetative, physiological and reproductive attributes of tomato crops compared to non-treated plants. Additionally, in this combination an adjuvant rather than antagonistic effect was evidenced between lithovit-urea50 and bioumik.

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REFERENCES

- Abdeldym, E.A., El-Mogy, M.M., Abdellateaf, H.R. & Atia, M.A. 2020. Genetic characterization, agro-morphological and physiological evaluation of grafted tomato under salinity stress conditions. *Agronomy* **10**(12), 1948. https://doi.org/10.3390/agronomy10121948
- Akladious, S.A. & Mohamed, H.I. 2018. Ameliorative effects of calcium nitrate and humic acid on the growth, yield component and biochemical attribute of pepper (*Capsicum annuum* L.) plants grown under salt stress. *Scientia Horticulturae* **236**, 244–250. https://doi.org/10.1016/j.scienta.2018.03.047
- Biozar. 2021. http://biozarco.ir/en/portfolio/bioumik-2/
- Blokhina, O., Virolainen, E. & Fagerstedt, K.V. 2003. Antioxidants, oxidative damage and oxygen deprivation stress: A review. *Annals of Botany* **91**, 179–194. https://doi.org/10.1093/aob/mcf118
- Bustomi, R.A., Senge, M.S.T.R., Suhandy, D. & Tusi, A. 2014. The effect of EC levels of nutrient solution on the growth, yield, and quality of tomatoes (*Solanum lycopersicum*) under the hydroponic system. *Journal of Agricultural Engineering and Biotechnology* **2**(1), 7–12.
- Canellas, L.P., Dobbss, L.B., Oliveira, A.L., Chagas, J.G., Aguiar, N.O., Rumjanek, V.M., Novotny, E.H., Olivares, F.L. Spaccini, R. & Piccolo, A. 2012. Chemical properties of humic matter as related to induction of plant lateral roots. *European Journal of Soil Science* **63**, 315–324. https://doi.org/10.1111/j.1365-2389.2012. 01439.x
- Cottenie, A., Verloo, M., Kiekens, L., Velghe, G. & Camerlynck, R. 1982. *Chemical Analysis of Plant and Soils*. Laboratory of Analytical and Agrochemistry. State University of Gent, Belgium, pp. 63.
- Cuartero, J. & Fernández-Muñoz, R. 1998. Tomato and salinity. *Scientia Horticulturae* **78**, 83–125. https://doi.org/10.1016/S0304-4238(98)00191-5

- De Hita, D., Fuentes, M., Fernández, V., Zamarreño, A.M., Olaetxea, M. & García-Mina, J.M. 2020. Discriminating the short-term action of root and foliar application of humic acids on plant growth: emerging role of jasmonic acid. *Frontiers in plant science* 11, 493. https://doi.org/10.3389/fpls.2020.00493
- Desoky, E.S.M., Merwad, A.R.M. & Rady, M.M. 2018. Natural biostimulants improve saline soil characteristics and salt stressed-sorghum performance. *Communications in Soil Science and Plant Analysis* **49**, 967–983. https://doi.org/10.1080/00103624.2018.1448861
- Elmongy, M.S., Zhou, H., Cao, Y., Liu, B. & Xia, Y. 2018. The effect of humic acid on endogenous hormone levels and antioxidant enzyme activity during in vitro rooting of evergreen azalea. *Scientia Horticulturae* 227, 234–243. https://doi.org/ 10.1016/j.scienta.2017.09.027
- Food and Agriculture Organization of the United Nations (FAO). FAOSTAT: Food and Agriculture Data; Food and Agriculture Organization of the United Nations: Rome, Italy, 2021. Available online: http://www.fao.org/faostat/en/ (accessed on 5 May 2021).
- Flowers, T.J. & Colmer, T.D. 2015. Plant salt tolerance: adaptations in halophytes. *Annals of Botany* **115**(3), 327–331. https://doi.org/10.1093/aob/mcu267
- Ghanem, M.E., Albacete, A., Martínez-Andújar, C., Acosta, M., Romero-Aranda, R., Dodd, I.C. Stanley Lutts, S. & Pérez-Alfocea, F.2008. Hormonal changes during salinity-induced leaf senescence in tomato (*Solanum lycopersicum* L.). *Journal of Experimental Botany* **59**, 3039–3050. https://doi.org/10.1093/jxb/ern153
- Gul, H., Farman, M., Hussain, A., Irshad, L.M. & Azeem, M. 2017. Exogenously applied sorbitol alleviates the salt stress by improving some biochemical parameters in spinach (*Spinacia oleracea* 1.). *International Journal of Biology and Biotechnology* **14**(4), 677–686.
- Issa, D.B., Alturki, S.M., Sajyan, T.K. & Sassine, Y.N. 2020. Sorbitol and lithovit-guano25 mitigates the adverse effects of salinity on eggplant grown in pot experiment. *Agronomy Research* **18**(1), 113–126. https://doi.org/10.15159/ar.20.075
- Kaya, C., Akram, N.A., Ashraf, M. & Sonmez, O. 2018. Exogenous application of humic acid mitigates salinity stress in maize (*Zea mays* L.) plants by improving some key physicobiochemical attributes. *Cereal Research Communications* **46**, 67–78. https://doi.org/10.1556/0806.45.2017.064
- Latif, H.H. & Mohamed, H.I. 2016. Exogenous applications of moringa leaf extract effect on retrotransposon, ultrastructural and biochemical contents of common bean plants under environmental stresses. *South African Journal of Botany* **106**, 221–231. https://doi.org/10.1016/j.sajb.2016.07.010
- Liang, Y., Shen, Q., Shen, Z. & Ma, T. 1996. Effects of silicon on salinity tolerance of two barley cultivars. *Journal of Plant Nutrition* 19, 173–183. https://doi.org/10.1080/01904169609365115
- Lucini, L., Rouphael, Y., Cardarelli, M., Canaguier, R., Kumar, P. & Colla, G. 2015. The effect of a plant-derived biostimulant on metabolic profiling and crop performance of lettuce grown under saline conditions. *Scientia Horticulturae* **182**, 124–133. https://doi.org/10.1016/j. scienta.2014.11.022
- Mahmoud, A.W.M., Abdeldaym, E.A., Abdelaziz, S.M., El-Sawy, M.B. & Mottaleb, S.A. 2020. Synergetic effects of zinc, boron, silicon, and zeolite nanoparticles on confer tolerance in potato plants subjected to salinity. *Agronomy* **10**, 19. https://doi.org/10.3390/agronomy10010019
- Mata, C.G. & Lamattina, L. 2001. Nitric oxide induces stomatal closure and enhances the adaptive plant responses against drought stress. *Plant Physiology* **126**(3), 1196–1204. https://doi.org/10.1104/pp.126.3.1196
- Naim, L., Alsanad, M.A., El Sebaaly, Z., Shaban, N., Abou Fayssal, S. & Sassine, Y.N. 2020. Variation of *Pleurotus ostreatus* (Jacq. Ex Fr.) P. Kumm. (1871) performance subjected to different doses and timings of nano-urea. *Saudi Journal of Biological Sciences* **27**(6), 1573–1579. https://doi.org/10.1016/j.sjbs.2020.03.019

- Rady, M.M., Desoky, E.-S.M., Elrys, A.S. & Boghdady, M.S. 2019. Can licorice root extract be used as an effective natural biostimulant for salt-stressed common bean plants? *South African Journal of Botany* **121**, 294–305. https://doi.org/10.1016/j.sajb.2018.11.019
- Sajyan, T.K., Shaban, N., Rizakallah, J. & Sassine, Y.N. 2018. Effects of monopotassium phosphate, nano-calcium fertilizer, acetyl salicylic acid and glycinebetaine application on growth and production of tomato (*solanum lycopersicum*) crop under salt-stress. *Agronomy research* **16**(3), 872–88. https://doi.org/10.15159/ar.18.079
- Sajyan, T.K., Allaw, W., Shaban, N. & Sassine, Y.N. 2019a. Effect of exogenous application of glycine betaine on tomato plants subjected to salt-stress. *Acta Horticulturae* **1253**, 41–48. https://doi.org/10.17660/ActaHortic.2019.1253.6
- Sajyan, T.K., Chokor, M., Shaban, N. & Sassine, Y.N. 2019b. Enhancing salt tolerance of tomato (*Solanum lycopersicum*) by foliar application of aspirin (acetyl salicylic acid). *Acta Horticulturae* **1253**, 49–54. https://doi.org/10.17660/ActaHortic.2019.1253.7
- Sajyan, T.K., Rizkallah, J., Sebaaly, Z., Shaban, N. & Sassine, Y.N. 2019c. Investigating the potential use of mono-potassium phosphate (MKP: 0-52-34) applied through fertigation as a method to improve salinity tolerance of tomato plants. *Acta Horticulturae* **1253**, 1–8. https://doi.org/10.17660/ActaHortic.2019.1253.7
- Sajyan, T.K., Shaban, N., Rizakallah, J. & Sassine, Y.N. 2019d. Performance of salt-stressed tomato crop as affected by nano-CaCO₃, glycine betaine, MKP fertilizer and aspirin application. *Agriculture and Forestry* **65**(1), 19–27. https://doi.org/10.17707/AgricultForest.65.1.02
- Sajyan, T.K., Naim, L., Sebaaly, Z., Rizkallah, J., Shaban, N. Sassine, Y.N. 2019e. Alleviating the adverse effects of salinity stress on tomato crop (*solanum lycopersicum*) using nano-fertilizer as foliar application. *Acta Horticulturae* 1253, 33–40. https://doi.org/10.17660/ActaHortic.2019.1253.5
- Sajyan, T.K., Alturki, S.M. & Sassine, Y.N. 2020. Nano-fertilizers and their impact on vegetables: contribution of Nano-chelate Super Plus ZFM and Lithovit®-standard to improve salt-tolerance of pepper. *Annals of Agricultural Sciences* **65**(2), 200–208. https://doi.org/10.1016/j.aoas.2020.11.001
- Sassine, Y.N., Alturki, S.M., Germanos, M., Shaban, N., Sattar, M.N. & Sajyan, T.K. 2020. Mitigation of salt stress on tomato crop by using foliar spraying or fertigation of various products. *Journal of Plant Nutrition* **43**(16), 2493–2507. https://doi.org/10.1080/01904167.2020.1771587
- Sassine, Y.N., Naim, L., El Sebaaly, Z., Abou Fayssal, S., Alsanad, M.A. & Yordanova, M.H. 2021. Nano urea effects on *Pleurotus ostreatus* nutritional value depending on the dose and timing of application. *Scientific Reports* 11, 5588. https://doi.org/10.1038/s41598-021-85191-9
- Shimul, M.A.H, Ito, S.I.C., Sadia, S., Roni, M.Z.K. & Jamal Uddin, A.F.M. 2014. Response of tomato (*Lycopersicon esculentum*) to salinity in hydroponic study. *Bangladesh Journal of Scientific Research* **10**(3), 249–254.
- Vysotskaya, I.V., Arkhipova, T.N., Kudoyarova, G.R. & Veselov, S.Y. 2018. Dependence of growth inhibiting action of increased planting density on capacity of lettuce plants to synthesize ABA. *Journal of Plant Physiology* **220**, 69–73. https://doi.org/10.1016/j.jplph.2017.09.011
- Yakhin, O.I., Lubyanov, A.A., Yakhin, I.A. & Brown, P.H. 2017. Biostimulants in plant science: A global perspective. *Frontiers in Plant Science* 7, 1–32. https://doi.org/10.3389/fpls.2016.02049
- Zhang, P., Senge, M. & Dai, Y. 2016. Effects of salinity stress on growth, yield, fruit quality and water use efficiency of tomato under hydroponics system. *Reviews in Agricultural Science* **4**, 46–55. *https://doi.org/*10.7831/ras.4.46
- Zhang, P., Senge, M. & Dai, Y. 2017. Effects of salinity stress at different growth stages on tomato growth, yield and water use efficiency. *Communications in Soil Science and Plant Analysis* **48**(6), 624–634. https://doi.org/10.1080/00103624.2016.1269803