Effect of spraying humic acid and salicylic acid on potato leaf area, yield and quality at two different levels of field capacity

**Authors Names**
a. Hadia Hassan  
b. Sawsan Suleiman  
c. Maher Ali Dais

**Article History**
Received on: 29/6/2021  
Revised on: 20/8/2021  
Accepted on: 11/1/2022

**Keywords:**
potato, humic acid, salicylic acid, water stress

**DOI:** https://doi.org/10.29350/jops.2022.27.1.1411

**ABSTRACT**
This study was carried out at Al-Hanadi Research Station - Agricultural Scientific Research Center in Lattakia during spring 2019, to study the effect of foliar spray with different concentrations of humic acid (500, 1000 and 1500 ppm) and salicylic acid (50, 100 and 150 ppm) on Spunta potato plants at two levels of irrigation 40% and 80% of field capacity. The experiment included 14 treatments and 3 replicates for each one. Experiment was designed according to a completely randomized design in split plots. Results showed a decrease in vegetative surface area and yield kg m^{-2} at irrigation level 40% of field capacity, while tubers content of proline and dry matter tubers increased. The highest tuber content of proline was in humic acid 1000 ppm at 40% of field capacity. At irrigation level 80% of field capacity, the treatment with salicylic acid 100 ppm showed the highest yield 40900 kg h^{-1} and the highest tuber content of sugars in salicylic acid 100 ppm at the same irrigation level. Salicylic acid treatment 50 ppm gave also the highest vegetative surface area (2278) cm^{2}.

1. Introduction

Potato, Solanum tuberosum, belongs to genus Solanum and Solanaceae family. This genus includes more than 2000 species spread in the most parts of the world. Potato plant is considered a drought-sensitive plant (59). Drought considered is one of the main factors affecting in potato yield and quality of tubers. (15) noticed that water stress causes a decrease in tubers weight, size and yield. It was found that 40% of the decrease in tuber inflation rate under stress conditions was caused by the decrease in tuber weight rate (29). Water stress causes a decrease in leaf area, plant height and coverage (60). Hassan (2002) found that tuber initiation stage is more sensitive to moisture deficiency than tuber growth and bulking stages. Water stress causes damage to pigments and plastids cells, lowers the content of chlorophyll and carotenoids content, and affects photosynthesis reactions in plants (65), which reduces plant growth and its production (22). Water stress leads to an increase in free oxygen forms (ROS) (free oxygen molecule O_{2}, hydrogen peroxide H_{2}O_{2} and hydroxyl radicals OH) resulting from incomplete reduction of oxygen upon exposure to stress (10). Free radicals cause lipid
oxidation (18), protein destroyed (43) and DNA damage (33) which causes weakness plant growth and development.

Plant tolerance drought by accumulation number of organic substances (74) such as proline, and sugars (72) which adjust osmotic pressure in cell also increase enzyme antioxidants that reduces damages caused by the increase of free radicals under stress conditions (39). Antioxidants has important role in neutralizing free radicals in plant cells and tissues, but these substances gradually decreased with increasing stress conditions.

Plant growth regulators are considered among the most important antioxidants that play an effective role in regulating biotic plant processes and affect in plants response and tolerance to environmental and biological stresses (45). Salicylic acid (SA) in appropriate concentrations considered as a good factor to control biotic and abiotic stresses in plants (28). Many studies showed that SA is used as a plant growth regulator to mitigate the harmful effects of drought (26). It improves Plant growth, enzyme activity, ions absorption and movement in plant (66) study showed that foliar spraying with SA and tryptophan had an effective role in reducing the negative effects on corn plants exposed to drought. Studies have also shown that humic acid (HA) can be used to effect in plant hormonal levels and improve the growth and development of agricultural crops under various stresses (79), including water stress (21). Several studies indicated the beneficial role of HA in increasing cell membrane permeability, rate of photosynthesis, evapotranspiration, protein and hormone assimilation, and root cells elongation (21). Treatment with HA improved potato plant growth, photosynthetic indicators and tuber yield at different levels of water deficit under greenhouse conditions (51). Other studies also showed that humic treatment improved biochemical indicators such as chlorophyll content, ascorbic acid, nitrogen content, starch, soluble solids, protein content and increases growth and productivity indicators of Spunta potatoes under stress conditions (69). The objective of this study was to enhance the tolerance of potato plants to water stress by using humic and salicylic acid.

Materials and methods:
The experiment were carried out at Al-Hanadi Research Station, Agricultural Scientific Research Center, Lattakia. Using potato Spunta tuber, a Dutch semi-late variety, it is considered one of the medium-ripened varieties (100-150 days from the date of planting), its tubers are elongated in shape and attractive. The variety is characterized by a good size of the vegetative group, the spunta yield is hight in the spring lug and good in the autumn (8). Spunta tubers for planting were secured from the General Organization for Seed Multiplication in Lattakia, and the tubers were planted in sandy loam soil with a good content of organic matter, a high content of phosphorous and medium content of potassium, Table (1).

Table 1: Verification of soil analysis values:

<table>
<thead>
<tr>
<th>Clay %</th>
<th>Silt %</th>
<th>Sand %</th>
<th>Available potassium mg.kg-1 soil</th>
<th>Available phosphorous mg.kg-1</th>
<th>mineral nitrogen mg.kg-1</th>
<th>Total calcium carbonate mg kg-1</th>
<th>Organic matter mg kg-1</th>
<th>EC ds/m</th>
<th>PH</th>
</tr>
</thead>
<tbody>
<tr>
<td>18</td>
<td>8.5</td>
<td>73.5</td>
<td>228.75</td>
<td>22.5</td>
<td>6.5</td>
<td>340</td>
<td>13.3</td>
<td>0.44</td>
<td>7.6</td>
</tr>
</tbody>
</table>

The Source: Soil, Plant and Fertilizer Analysis Methodologies Book - General Authority for Scientific and Agricultural Research.
Field soil was prepared for planting with two basic plowings at January 2019, followed by a third plowing, planning for planting in the first week of February. Tubers were planted in field at 10 cm of depth and 30 cm of distance between plants in the line and 70 cm between the lines. After planting, service practices included soil irrigation after planting, fertilizing (according to Agriculture Ministry recommendation to fertilize potatoes in sandy soils) with (369.5 kg ha\(^{-1}\)) urea fertilizer and (240 kg ha\(^{-1}\)) potassium sulfate fertilizer (7). After 35 days of planting, potato plants irrigated at two levels: 40% and 80% of field capacity. Field capacity was calculated basis on the volumetric moisture percentage in the soil and by membrane pressure device. Each irrigation level represents an independent piece including all treatments separated in completely randomized blocks at three replicates for each treatment. Potato plants were sprayed with salicylic acid and humic acid three times in interval ten days.

This research included following treatments:

<table>
<thead>
<tr>
<th>40% of Field capacity</th>
<th>80% of Field capacity</th>
</tr>
</thead>
<tbody>
<tr>
<td>8- C(_{40}): spray with water.</td>
<td></td>
</tr>
<tr>
<td>9- SA(_{1}): spray with salicylic acid 50 ppm</td>
<td></td>
</tr>
<tr>
<td>10- SA(_{2}): spray with salicylic acid 100 ppm</td>
<td></td>
</tr>
<tr>
<td>11- SA(_{3}): spray with salicylic acid 150 ppm</td>
<td></td>
</tr>
<tr>
<td>12- HA(_{1}): spray with humic acid 500 ppm</td>
<td></td>
</tr>
<tr>
<td>13- HA(_{2}): spray with humic acid 1000 ppm</td>
<td></td>
</tr>
<tr>
<td>14- HA(_{3}): spray with humic acid 1500 ppm</td>
<td></td>
</tr>
<tr>
<td>1- C(_{80}): spray with water.</td>
<td></td>
</tr>
<tr>
<td>2- SA(_{1}): spray with salicylic acid 50 ppm</td>
<td></td>
</tr>
<tr>
<td>3- SA(_{2}): spray with salicylic acid 100 ppm</td>
<td></td>
</tr>
<tr>
<td>4- SA(_{3}): spray with salicylic acid 150 ppm</td>
<td></td>
</tr>
<tr>
<td>5- HA(_{1}): spray with humic acid 500 ppm</td>
<td></td>
</tr>
<tr>
<td>6- HA(_{2}): spray with humic acid 1000 ppm</td>
<td></td>
</tr>
<tr>
<td>7- HA(_{3}): spray with humic acid 1500 ppm</td>
<td></td>
</tr>
</tbody>
</table>

In each treatment 15 plants were determined as samples for indicators. The leaf area and Vegetative surface area was determined as follows: leaf area was determined by disk method (6): Leaf area cm\(^2\) = (5 dry disks area * dry weight of 5 leaves) / dry weight of 5 dry disks. Vegetative surface area cm\(^2\) = leaf area * leaves number

**Yield kg m\(^{-2}\):** tubers were harvested after 120 days of planting, and tubers fresh weight was determined and the total yield (kg ha\(^{-1}\)).

**Dry matter (%):** many tubers from determined plants were taken and cut for many pieces and weighted (fresh weight), these pieces dried in an electric oven at 105 c for 48 hours for dried weight. Tubers dry matter (%) = (dry weight*100) / fresh weight.

**Tubers content of sugars (mg g\(^{-1}\)):** take 100 mg of fresh tubers, soak it in 3 ml of alcohol 80% in dark place for 48 hours, and remove it into warm water 30 C for 20 minutes. Add 20 ml of distilled water, take 1 ml of sample, and add 1 ml phenol (5%) and 5 ml of sulfuric acid (96%). Read the absorption (D) at 490 nm wavelength in spectrophotometer (23). Tuber Sugar content (mg g\(^{-1}\)) = D490* 1.657.

Tuber content of proline: proline was estimated according to (12), weight 100 mg of fresh tubers, put it in 10 ml salphosalicylic acid 3%, filter it, take 2 ml from the filtrate in a test tube with 2 ml acetic acid and 2 ml nenhydrin, place it in a hot water for half an hour after boiling. Cold samples directly at low temperatures, add 5ml toluene to each sample. Read absorption at 520 nm wavelength on spectrophotometer.

Experiment was designed as factorial experiment in a completely randomized design in split plots. Resulting data were analyzed using Costat program. The differences between the treatments averages were calculated using Duncan test at significance level 5% (24).
Experiment was designed as factorial experiment in a completely randomized design in split plots. Resulting data were analyzed using Costat program. The differences between the treatments averages were calculated using Duncan test at significance level 5% (24).

Results:

Vegetative surface area cm²: Results showed that irrigation level didn’t effect on potato vegetative surface area (Fig. 1). Potato vegetative surface area increased significantly when treated with humic acid by different concentrations, compared to other stress treatments, and control C₄₀. As well as vegetative surface area increased significantly in plants treated with salicylic acid 150 ppm compared to C₄₀ control treatment about 10.81%. At the irrigation level 80%, potato treated with humic acid and salicylic acid had a significant effect in increasing vegetative surface area compared to control C₈₀, especially humic acid 1000 and 1500ppm, which increased it by 44.08 and 42.09%, also salicylic acid treatment 50ppm by 44.25%.

![Graph showing leaf area of spunta potato treated by humic and salicylic acid at two irrigation level 40 and 80%](image)

**Fig 1: Leaf area of spunta potato treated by humic and salicylic acid at two irrigation level 40 and 80 % (LSD₀.₀₅=107.26)**

Where these treatments outperformed other irrigated treatments with significant differences.

Yield kg.h⁻¹: The results in the figure (2) shows a decrease in tuber yield of potato spunta by (32.40%) with water shortage increased. The yield increased significantly with increasing in humic and salicylic acid concentration at level 40%, SA 100 and 150 ppm treatments gave the highest yield of tubers in (23.19 and 24.15 %) compared with control C₄₀, and in significant differences.

Salicylic Acid Protects Potato Plants from Phytoplasma-associated Stress and Improves Tuber Photosynthetic Assimilation Salicylic Acid Protects Potato Plants from Phytoplasma-associated Stress and Improves Tuber Photosynthetic Assimilation
Yield kg h\(^{-1}\)

Fig.2: Tuber yield of potato spunta treated with humic and salicylic acid at two irrigation level 40 and 80 \(\%\) (LSD\(_{0.05}\)=0.23).

At irrigation level 80\% HA and SA treatment significantly increased spunta yield, and SA 100 and 150 ppm treatments gave the highest yield of tubers (4.09 and 3.39 kg/m\(^2\)) superior in significant differences over other treatments. Also HA treatment 500 and 1500 ppm increased tuber yield significantly by (10.15\% and 9.85\%) compare with control.

Dry matter \%: There was no significant difference of tuber content of dry matter at two irrigation level (Fig. 3). Also HA and SA did not have a significant effect in increasing dry matter\% in tubers under water stress, whereas dry matter \% increased significantly compared to dry matter\% under normal conditions. HA 1000 ppm gave the highest dry matter\% in spunta tubers (26.15\%) at level 40\%, while SA 100 ppm gave the highest dry matter (15.3\%) at level 80\%.

**Figure (3)** the tuber content of dry matter of potato spunta treated with humic acid and salicylic acid at two irrigation levels 40 and 80\% (LSD\(_{0.05}\)=2.74)

**Tubers content of sugars mg g\(^{-1}\):** the results in figure (4) showed no significant differences in tubers content of sugars at irrigation levels 40\% and 80\%. Also, potato treated with humic and salicylic acid had no significant differences in tuber content of sugars (mg g\(^{-1}\)) of stress plants compared with control.
C40%. Salicylic acid 150ppm reduced tubers content of sugars(mg g\(^{-1}\)) and gave the lowest tuber content of sugars(mg g\(^{-1}\)) compared to other stress treatments, and control C40%. At the irrigation level 80%, tubers content of sugars increased significantly in potato spunta plants treated with salicylic acid 100ppm (4.47 mg g\(^{-1}\)) compared to the other treatments and control C80. Salicylic acid treatment 150ppm was significantly superior in tuber content of sugars than humic acid 500 and 1000 ppm.

The tuber content of proline (ppm): The results in figure (5) shows that Spunta potato tubers at irrigation level 40% were superior in proline content by 73.87% compared to irrigation level 80%. HA and SA treatments causes an increasing in the tuber content of proline; HA 1000ppm and salicylic (50, 100 and 150ppm) treatments were superior with significant differences in the tuber.
Content of proline compared with control at irrigation level 40%. At irrigation level 80%, SA 100 and 150ppm treatment increased significantly tuber content of proline by 38.74 and 41.38% compared to the other irrigated treatment and control C80.

Discussion: Water stress directly effects photosynthetic pigments, transpiration, leaf surface area, root growth, element representation and hormonal balance, which affects plant growth and development (46). These results are consistent with results of (44), who indicated That water stress reduced yield, chlorophyll content, carotenoids, leaf area and relative water content of wheat plants exposed to stress. HA causes an increase in elements absorption, efficiency of utilization, and improve crop growth and production (34). HA is considered an effective factor to improve drought tolerance in plants because it affects on plant defense mechanism by stimulating antioxidant activity, some studies indicated that HA improved the resistance of corn and wheat plants by increasing antioxidants (5). HA directly increased photosynthesis under unsuitable conditions (51). SA plays an important role in protecting plants from stress by regulating the plant defense mechanism (antioxidants) (39). SA equilibriums H2O2 accumulation with substances that protect plant against stress (19). This is agree with results that indicated that treated with SA increased an antioxidant activity of Cucurbita pepo squash (63) and Cherry plants (78) to avoid Oxidative stress induced by pathological injury.

The sensitivity of potato crop to water lack in soil may due to a small size and mass of its roots and that 85% of root system is spread in the upper layer (up to 30 cm depth) of the soil, water stress causes plant growth weaken and small plant mass resulting in a decrease in tuber yield (53). Plants tend to decrease photosynthetic activity with water shortage increased (62) because of increasing off the stomata transduction sensitivity, photosynthesis system, transpiration and carbon dioxide concentration in plant tissue to stress conditions sensitive (16), which is negatively and directly reflected in yield (47). Water stress affects in cell water content and stomata closing, which reduces co2 fixation and decreases plant growth (45), thus reduces the optically manufactured materials amount and the transferred and stored materials in tubers, which reduces the resulting tubers weight. These results are similar to (4) results, who indicated to a decrease in tubers number and size, and a decrease in potato productivity when deficient water happened in tuber initiation stage. HA causes an increasing in elements absorption and the utilization efficiency, as well as improves also the crop growth and production (34). This agree with the (51) results reported that the HA treatment improved potato growth in greenhouse because it is improving photosynthetic system and improving fresh tubers productivity in lack irrigation conditions. (32) Was also found that HA treatment improved potato productivity of potato plants compared to untreated plants. Results of (48) showed that HA causes an increased in fresh weight, size and quality of tubers by increasing of the humic used concentration. The positive effect of salicylic acid is due to fact that it plays an important role in plant protection from biotic and abiotic stresses by regulating defense system (antioxidants) (39). SA improves transpiration rate, stomatal closure (64), membrane permeability (13), growth and photosynthesis (27). Also SA may enhance the production of H2O2 to levels that can help to reduce the oxidative damage caused by salinity stress in wheat plants (77). Similar results (40), found that SA treatment had the best effect compared to HA on three sweet pepper varieties, salicylic acid 1.5 g/l gave the best vegetative growth and the best productivity indicators including fruit number and diameter, the content of vitamin C, soluble solids and total sugars compared to the other treatments. Osmotic stress decreases carbon assimilation, increases stomata closure, determined photosynthetic rate and affects in the amount of carbohydrate which transported and stored in tubers (17), which reduces tubers weight. These results may agree with (1) reported, the highest values of tuber yield/plant and total yield/fed., N, P, starch, carbohydrates and dry matter % in tuber. Cell sap, bound water, osmotic pressure, proline content in leaf tissues and water use efficiency, increased with decreasing irrigation water quantity up to the (750 m3 / fed) level. Salicylic acid activate the consumption of dissolved carbohydrates to form new
young cells, which stimulates plant growth (54) also, increases photosynthesis rate and the transported and stored amount of carbohydrates into tubers, which reflected in an increase in dry matter. % Salicylic acid affects in the physiological and metabolic processes in plant (42) such as photosynthesis, transpiration, element uptake and transport (67) and mitigating the damages of growth inhibitors (free radicals and oxidants) in Stress conditions .SA stimulate an antioxidant enzymes activity and inhibit the free radicals in cell (70), which is reflected in an increase the yield of vegetable species (45). Similar results reported that SA has important role in increasing the tubers content of potassium, which increase osmotic pressure and an increase in water amount of tubers, and reduces tuber dry matter% (Soliman et al., 2018), however varying concentrations of SA increased tubers content of dry matter and starch in Spunta compared to un treated plants. Many studies reported that HA stimulates growth and production of plants and improves the crop quality (38). HA plays an important role in increasing cell membrane permeability, photosynthetic rate, evapotranspiration, proteins assimilation and hormones, and elongation of root cells ((21). This results agree with many studies indicated that HA increased tuber yield and improved quality of potato (2) and(68) ,also improved the tubers content of starch , soluble solids, protein and nutrients ( N, P and K) (68),(69) . Water deficient inhibits photosynthesis, dissolve chlorophyll pigment and causes damage to photosynthesis system (22). It also reduces stomata conductivity (36), and increase sugars and proline accumulation (57) soluble sugars play an osmotic- regulatory role that keeps the leaf cells full, maintains the integrity of the cell membrane and prevents proteins denaturation (55). (11) Results reported that the total soluble sugars increased and photosynthesis rate decreased as soon as leaf water potential decreased, but sugars, proline, and enzymatic activity of plants increased under stress condition (49). Salicylic acid improves cell membrane permeability, which facilitates elements absorption and transfer photosynthetic products and storage it in tubers (9). Also, these results (at irrigation level 80%) are similar to (50), which indicated that SA treatment increased roots content of sugars and carotene of sweet potato. SA improves plant growth indicators under various stress conditions, including water stress (70), and stimulated dissolved carbohydrates consumption to form new young cells, which motivates and increase plant growth (54). Also SA encourage the activity of many enzymes that inhibit free radicals. It is known that HA has an effective role in reducing the negative effect of stress (56), HA regulates hormonal levels in plants and increases synthesis of enzymes and growth hormones (52). Many studies (69) reported that HA improved chlorophyll content, ascorbic acid, nitrogen, starch, soluble solids, protein content, and enhanced growth and productivity indicators of Spunta potatoes under stress conditions. HA treatment increased the pea pod content of many organic compounds, including total sugars, and protein content (3).

Proline play an important role in increasing plant tolerance to water stress (76). Proline has important functions in adjusting osmotic pressure under water stress conditions and has a key role in protecting proteins and cell membrane stability in plants through its accumulation at high levels, which causes modulate the cell osmotic pressure (31). Several studies have reported an increase in proline accumulation in wheat plants grown under water stress (76). This is similar to (44) results that reported the high rate of proline accumulate in the plant exposed to water stress while its accumulation decreases under natural irrigation. Similar studies (49) found that proline content, soluble sugars, catalase enzyme activity and polyphenol oxidase, and peroxidase and superoxide dismutase increased under stress conditions. Also (57) results that reported the increasing in proline accumulation in potato tubers exposed to salinity and drought stress. SA improves plant growth indicators under different stress conditions, including water stress(70) also salicylic acid affect in increasing glutamate and ammonia accumulation in plants exposed to drought , which stimulates the proline accumulation increasing , otherwise proline accumulation may be due to absence of the enzymes proline oxidase and proline dehydrogenase activity (75) .(37) results showed an increase in relative water content, proline content and total chlorophyll content, and a decline in the electrolyte leakage of tomato plants.
grown under drought conditions when treated with salicylic acid. Also salicylic acid increased strawberry productivity under stress condition and increased proline fruits' content and soluble solids (41). Also (73) indicated that salicylic acid caused an increase in plant mass and protein content in chickpea leaves under drought conditions compared to untreated plants. HA acts as a biostimulor through its role in enhancing the activity of plant hormones and enzymes (52) and stimulating protein synthesis (61) which reduces the harmful effects of stress (56). Treatment with humic acid 120 kg/ha enhanced chlorophyll content, ascorbic acid, nitrogen content, starch, soluble solids, protein content, as well as growth and productivity indicators of Spunta potato under stress conditions (69). Similar results reported that humic acid increased the content of many organic compounds including total sugars and protein content in pea pods (3).

Conclusions:

Tubers content of dry matter, sugars and proline increased with a decrease in soil moisture, Humic and salicylic acid treatments effected in Spunta traits. Humic acid can play a positive role in improving spunta potato tolerance under stress water especially humic acid concentration 500ppm, and improve tuber content of dry matter and proline. Salicylic acid can increase potato productivity under different levels of field capacity, especially SA 50ppm.

Author Contributions: All authors contributed equally in writing this article. All authors read and approved the final manuscript.

Conflicts of Interest: The authors declare no conflict of interest.

References:


[34] Haider, G. et al, Biochar.,(2015), but not humic acid product amendment affected maize yields via improving plant-soil moisture relations, J. Plant Soil. 395, 141.


[40] Ibrahim,A; Abdel-Razzak,H; Wahb-Allah ,M; Alenazi ,M; Alsadon ,A; and Dewir ,H.Y. (2019). Improvement in Growth, Yield, and Fruit Quality of Three Red Sweet Pepper Cultivars by Foliar Application of Humic and Salicylic Acids, https://doi.org/10.21273/HORTTECH04263-18


