INTRODUCTION

Drought stress drastically reduces photosynthesis in leaves due to a reduction in the number of leaf pores and reduced levels of Rubisco (Hussain, Nazir, & Fariduddin, 2019). The effect of reduced water uptake under drought stress conditions on plant growth can be due to reduced water uptake pressure, reduced cell turgidity, or reduced nitrogen uptake (Reddy, Chaitanya, & Vivekanandan, 2004), all of which can greatly affect the rate of cell division and cell growth. In drought stress, the rate of photosynthesis in the plant is reduced due to the accumulation of toxic compounds in plant cells, especially active oxygen. In addition, oxidative stress can trigger changes in proteins and DNA which reduce growth (Ghasemi Pirbalouti, Rahmani Samani, Hashemi, & Zeinali, 2014; Reddy, Sekhar, Sreeharsha, & Reddy, 2019). Moreover, drought stress increases the amount of abscisic acid in the plant, which reduces the growth rate (Siebeneichler et al., 2020).

Research has shown that chitosan consumption is common in living organisms, including plants (Ngo et al., 2015). Chitosan boosts plants’ defenses and can even increase resistance to plant pathogens (Sharma et al., 2019). Chitosan is derived from chitin, which is an antitoxic substance. Especially in low doses, chitosan can improve a plant’s resistance to pathogens by strengthening its immune system (Ngo et al., 2015). Chitosan protects cells against plant pathogens through lipid.
peroxidation. It can be said that chitosan maintains normal plant conditions against some forms of stress (Yin, Fretté, Christensen, & Grevsen, 2012). Chitosan has found applications in plant genetics due to its protective properties, its compatibility with plants and the environment, and its stable chemical structure. The suitable chemical properties of chitosan as well as its numerous applications in increasing resistance to various stresses, including resistance to plant pathogens, have led to its widespread use for crop production in areas facing environmental stresses (Kashyap, Xiang, & Heiden, 2015). Chitosan treatment increases the germination percentage of seeds and therefore improves their establishment in the field, leading to increased competitiveness with weeds, increased water use efficiency, and improved resistance to stress in seedlings (Xuan Tham et al., 2001). Therefore, based on previous and ongoing global research on the effects of chitosan and salicylic acid on stressed plants, the effects of foliar application of these compounds were studied in potato plants under drought conditions.

MATERIALS AND METHODS

Experimental Sites

The experiment was conducted in 2019 (10 June – 01 November) at an agricultural research station in the city of Fariman, Iran (36°29’ N, 59°17’ E, at 1,176 m above sea level). The region has a cold and dry climate according to Koppen’s climatic classification.

Experimental Design and Treatments

The experiment was implemented as a completely randomized split-plot design with three replications. Experimental treatments included irrigation levels and the application of chitosan and salicylic acid. The main plots were 12 by 6 m and the sub-plots were 6 by 3 m. The main plots represented three levels of irrigation (100%, 80%, and 60% of available soil water). The treatments in sub-plots included control treatment, 0.5 g/l salicylic acid, 2 g/l chitosan, and combined treatment with chitosan and salicylic acids.

Irrigation Treatments

Irrigation deficit treatments were applied 20 days after plant emergence and at each irrigation period. The irrigation period was determined according to the temperature conditions of the region at intervals of 3 to 5 days. Crop water need was determined using OPTIWAT.

Chitosan and Salicylic Acid Application

Foliar application of chitosan and salicylic acid was performed in two stages during the growing season: at the flowering stage (beginning of tuber development), and at the mid-growth stage of tubers (approximately 60 days after plant emergence).

Soil Physical and Chemical Properties

Before planting, a suitable amount of soil was taken from a depth of 0 to 30 cm to check the physical and chemical condition of the soil. Total nitrogen was measured by the micro-Kjeldahl apparatus through the distillation process. Phosphorus was extracted with sodium bicarbonate and measured by the blue dye method using molybdenum phosphate (Olsen, Cole, Watandbe, & Dean, 1954). Potassium was extracted by ammonium acetate and evaluated using flame light. Soil texture was silty clay and the physicochemical properties of the soil were as follows: organic matter 0.42%, pH 7.6, total nitrogen 200 mg/kg, available phosphorus 5.1 mg/kg, electrical conductivity (EC) 0.43 dS/m, and available potassium 189 mg/kg. To homogenize soil chemical properties, potassium sulfate fertilizer and triple superphosphate fertilizer were applied one month before planting according to the results of the soil test. Urea fertilizer was also applied based on soil tests in several stages during irrigation.

Measurements

Laboratory Section

In the laboratory, we measured the relative water content of leaves, cell membrane stability, total chlorophyll content, chlorophyll a and b content, free phenol percent, proline content, soluble carbohydrates, leaf area, and dry leaf weight.

Relative Water Content (RWC)

The Coleman method (Coleman, 2008) was used to determine the relative water content of leaves in 6 stages (three times from plant emergence to tuber formation, and three times from tuber formation to harvest). In each stage, sampling was done 48 hours after irrigation. Between six to twelve lateral and terminal leaflets per replicate were cut from three bushes and placed in distilled water after weighting. Leaves were kept in distilled water for 24 hours in the laboratory at 25°C, after which the leaves were dried and turgid leaf weight
was measured. Dry leaf weight was measured after baking the leaflets for 48 hours in an oven at 80°C. Relative water content was evaluated by the following formula:

$$RWC = \left( \frac{FW - DW}{TW - DW} \right) \times 100$$  \hspace{1cm} 1)

Where: FW, DW, and TW are fresh leaf weight, dry weight, and turgid weight, respectively.

### Cell Membrane Stability

To evaluate cell membrane stability under stress conditions, ten leaves were cut from three different potato shrubs. The surface of the leaves was washed three times with distilled water to remove solutes. Leaf samples from different treatments were placed in bottles containing 50 cm$^3$ of distilled water for 20 hours. Then, the leakage rate was measured by an EC meter at 25°C. To determine the maximum leakage rate, the samples were autoclaved for 21 minutes at 110°C and 1.5 atmospheres. After the samples were cooled to room temperature, their electrical conductivity was measured again. The rate of cell leakage under stress conditions is obtained using the following equation:

$$Cell\ leakage = \left( \frac{EC_1}{EC_2} \right) \times 100$$  \hspace{1cm} 2)

Where: EC1 and EC2 are the electrical conductivity of samples in the first stage and the second stage (maximum leakage) respectively.

The percentage of damage to leaf cells compared to non-stress conditions was calculated by the following equation (Arvin & Donnelly, 2008).

$$\%\text{Injury} = 1 - \left[ 1 - \left( \frac{T_1}{T_2} \right) - \left( \frac{C_1}{C_2} \right) \right] \times 100$$  \hspace{1cm} 3)

Where: T and C are electrical conductivity in stress and no-stress conditions, respectively.

### Leaf Chlorophyll Content

Chlorophyll a and b content of leaves was measured using the 80% acetone extraction method (Grzesiak, Grzesiak, Filek, & Stabryla, 2003). For this purpose, 36 samples of leaves were harvested in the middle stage of tuber development. The samples were immediately placed in a special freezer at -70°C, after being rapidly frozen by liquid nitrogen. In the laboratory, 0.1 g of the leaf was sampled and rubbed into liquid nitrogen. The rubbing process was continued using 80% acetone to create a colorless solution. The volume of the acetone solution was then increased to 25 ml and centrifuged at 4,000 rpm for 10 minutes. After separating the supernatant, the absorption of the clear solution was read at 645, 652, and 663 nm. To measure carotenoid content, absorbance was also obtained at 470 nm. The following equation was used to calculate total chlorophyll, chlorophyll a, and chlorophyll b content.

$$Chlorophyll\ total = \frac{A(625)}{345} \times \frac{v}{w}$$  \hspace{1cm} 4)

$$Chlorophyll\ a = \left( 12.7(663) - 2.69A(645) \right) \times \frac{v}{w} \times 1000$$  \hspace{1cm} 5)

$$Chlorophyll\ b = \left( 22.9(645) - 2.68A(663) \right) \times \frac{v}{w} \times 1000$$  \hspace{1cm} 6)

$$Carotenoid = \left( \frac{1000A(470) - 1.0\ Chlorophyll\ a - 85.02\ Chlorophyll\ b}{198} \right)$$  \hspace{1cm} 7)

Where: A, v, and w are the amount of absorbed light, sample size, and leaf weight, respectively.

### Soluble Carbohydrate Content

The number of leaf carbohydrates was evaluated and determined using standard phenol sulfuric acid and glucose (Dubois, Gilles, Hamilton, Rebers, & Smith, 1956). For this purpose, 100 mg of fresh leaf samples were stored in 70% ethanol at 4°C for 20 hours in a homogenizer. Insoluble solids were separated using centrifugation at 3,500 rpm for 10 minutes. Appropriate amounts were extracted from the supernatant at volume ratios of 1, 2, and 2, respectively, and mixed with distilled water. Then, with a volume ratio of 2 to 1, the solution was mixed with phenol and finally brought to a specific volume with 98% sulfuric acid. Ultimately, after remaining in 100°C water for 30 minutes, the absorption rate was determined at 480 nm.

### Leaf Proline Content

The amount of proline in leaf tissue was measured based on the Bates method (Bates, Waldren, & Teare, 1973). Leaf samples that were harvested at the mid-stage of tuber development were homogenized by a microtube homogenizer in 1 ml of 3% sulfuric acid. Insoluble solids were separated using centrifugation at 3,500 rpm for 10 minutes. Two hundred ml of glycolic acetic acid and 200 ml of ninhydrin were added to 200 ml of the extract. The resulting mixture was placed in warm water at 100°C for 30 minutes, and after cooling, 600 ml of toluene was added to the mixture. Proline concentration was read at 520 nm using the top solution. The frequency of proline was determined using its standard curve.
Glutathione Peroxidase

Drought stress affects almost all vital cell processes (Yang, Hu, Li, Wu, & Qian, 2009). Drought stress increases the relative concentration of oxygen free radicals, especially hydrogen peroxide (Xu, Burgess, Zhang, & Huang, 2016). The results of some studies have shown that protein-rich enzymes play an essential role in reacting with active oxygen radicals. One of the most important of these enzymes is glutathione peroxidase. This enzyme plays a vital role in counteracting oxidative stress and balancing oxygen-free radicals. Such a function may be mediated directly by an intermediate reaction or by an enzymatic mechanism that causes a decrease in the concentration of hydrogen peroxide (Petriccione et al., 2015). Another activity of glutathione peroxidase is its role in inducing the operation of the glycolic acid cycle in the process of converting fatty acids into sugar and producing cellular energy. Also, the movement of glutathione peroxidase has a significant role in the protection of cell membranes, especially in stressful conditions (El Hadrami, Adam, El Hadrami, & Daayf, 2010). Due to the role of cell membranes in the selective exchange of substances and ions, the protective mechanisms and the role of glutathione peroxidase in the protection of cell membranes and prevention of membrane lipid peroxidation are very important (Jung et al., 2002). Accordingly, measuring the peroxidation of membrane fats under stress conditions is known as one of the crucial indicators of resistance (Kolupaev, Karpets, & Kosakovska, 2008).

Total Phenol Percent

The Folin-Ciocalteu method was utilized to measure total phenol concentration in leaf samples (Singleton & Rossi, 1965). To this aim, 100 mg of fresh leaf samples were stored in 70% ethanol at 4°C for 20 hours in a homogenizer. Insoluble solids were separated using centrifugation at 3,500 rpm for 10 minutes. One ml of distilled water and 20 ml of Folin-Ciocalteu reagent were added to 20 ml of solution. After 5 minutes, 120 ml of sodium carbonate was added and the solution was placed at room temperature for 30 minutes. Total phenol content was measured at 765 nm as mg/g of dry matter.

Leaf Surface and Leaf Dry Weight

From each treatment, two plants were randomly selected during the budding and mid-growth (one month after flowering) stages of tuber development. Plants were cut off at the soil surface and the aerial parts of the plant were taken to the laboratory. To measure the dry weight of leaves, leaves were removed from the plant, placed in a 75°C oven for 48 hours, and weighed using a digital scale. Leaf area was also calculated using a Licore 3100 leaf surface measuring device.

Farm Section (Yield and yield components of potato)

The yield was determined based on plants in the center of each sub-plot (30 plants). Plants in the first and last 50 cm of each subplot and those in the two side rows of each sub-plot were not included in the analysis to minimize the impact of treatments applied to adjacent sub-plots. To measure yield components, dry weight was measured for the 10 plants growing in the central 1.5 m² of each sub-plot. The number of tubers per plant, wet weight and dry weight of the tubers, and the average weight of the tubers were also determined. Deformed and green tubers, as well as tubers with a diameter of less than 35 mm (Bol et al. 2020), were not included in the study since such tubers have little market value.

RESULTS AND DISCUSSION

Yield and Yield Components of Potato

Potato yield for factor A (irrigation percentage) showed a direct relationship between irrigation percentage and yield. The highest yield in the treatment without drought stress was 42.64 t/ha. The lowest yield for this index was observed in the severe drought treatment (60% full irrigation) at 30.47 t/ha (Fig. 1). Due to the effect of irrigation on other yield components such as fresh weight of tubers, the number of tubers per plant, the average weight of tubers, the weight of tubers per plant, shoot weight, dry weight of each tuber, and total biomass, these components showed trends which were similar to that of yield. Therefore, the highest value for each of these indicators was recorded in the 100% irrigation treatment, and the lowest value was seen in the severe drought stress treatment (Table 1). Based on the data, the treatment of 0.5 g of salicylate was significantly different from the treatment of a salicylates and chitosan mixture and it was the best treatment (Fig. 2).
Table 1. Interactive effects of drought stress and chitosan and salicylic acid application on yield and yield components of potato.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Yield (t/ha)</th>
<th>Fresh weight of tuber (g)</th>
<th>Number of tuber per plant</th>
<th>Mean weight of tuber (g)</th>
<th>Weight tuber in plant (g)</th>
<th>Shoot dry weight (g)</th>
<th>Tuber dry weight (g/m²)</th>
<th>Total biomass (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Drought stress</strong></td>
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<td></td>
</tr>
<tr>
<td><strong>No stress</strong></td>
<td>42.64a</td>
<td>4.53a</td>
<td>94.92a</td>
<td>49.326a</td>
<td>453a</td>
<td>309.4a</td>
<td>968.2a</td>
<td>1277.6a</td>
</tr>
<tr>
<td><strong>Medium stress</strong></td>
<td>36.3ab</td>
<td>4a</td>
<td>88.92a</td>
<td>44.925a</td>
<td>400a</td>
<td>242.53a</td>
<td>865.7a</td>
<td>1108.2a</td>
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<tr>
<td><strong>High stress</strong></td>
<td>30.47b</td>
<td>3.375ab</td>
<td>80.75a</td>
<td>43.435a</td>
<td>337.5a</td>
<td>189.23a</td>
<td>745a</td>
<td>934.2a</td>
</tr>
<tr>
<td><strong>Factor B</strong></td>
<td></td>
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</tr>
<tr>
<td><strong>Control</strong></td>
<td>35.76a</td>
<td>4.1778a</td>
<td>89.22a</td>
<td>49.138a</td>
<td>417.78a</td>
<td>261.36a</td>
<td>899.5a</td>
<td>1160.8a</td>
</tr>
<tr>
<td><strong>0.5 g/l Salicylic</strong></td>
<td>39.24a</td>
<td>4.2289a</td>
<td>97.11a</td>
<td>47.702a</td>
<td>422.89a</td>
<td>271.82a</td>
<td>924.4a</td>
<td>1196.2a</td>
</tr>
<tr>
<td><strong>2 g/l Chitosan</strong></td>
<td>35.91a</td>
<td>3.4022a</td>
<td>73.67a</td>
<td>43.909a</td>
<td>403.33a</td>
<td>225.61a</td>
<td>726.5a</td>
<td>952.2a</td>
</tr>
<tr>
<td><strong>0.5 g/l + 2 g/l</strong></td>
<td>34.95a</td>
<td>4.0644a</td>
<td>92.78a</td>
<td>42.832a</td>
<td>406.44a</td>
<td>229.42a</td>
<td>888.2a</td>
<td>1117.6a</td>
</tr>
<tr>
<td><strong>No stress</strong></td>
<td></td>
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<td></td>
</tr>
<tr>
<td><strong>Control</strong></td>
<td>40.28abc</td>
<td>4.5933a</td>
<td>99.33a</td>
<td>46.847a</td>
<td>459.33a</td>
<td>326.13a</td>
<td>989.4a</td>
<td>1315.5a</td>
</tr>
<tr>
<td><strong>0.5 g/l Salicylic</strong></td>
<td>45.71a</td>
<td>4.3067a</td>
<td>103.33a</td>
<td>41.657a</td>
<td>430.67a</td>
<td>304.9abc</td>
<td>895.4a</td>
<td>1200.3a</td>
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<tr>
<td><strong>2 g/l Chitosan</strong></td>
<td>45.68a</td>
<td>4.0333a</td>
<td>73.33a</td>
<td>58.963a</td>
<td>403.33a</td>
<td>289.47abcd</td>
<td>855.9a</td>
<td>1145.4a</td>
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<tr>
<td><strong>0.5 g/l + 2 g/l</strong></td>
<td>38.89abc</td>
<td>5.1867a</td>
<td>103.67a</td>
<td>49.837a</td>
<td>518.67a</td>
<td>317.1abc</td>
<td>1132a</td>
<td>1449.1a</td>
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<td></td>
<td></td>
</tr>
<tr>
<td><strong>Control</strong></td>
<td>40.65ab</td>
<td>5.06a</td>
<td>97a</td>
<td>52.69a</td>
<td>506a</td>
<td>266.53abcd</td>
<td>1064.5a</td>
<td>1331.1a</td>
</tr>
<tr>
<td><strong>0.5 g/l Salicylic</strong></td>
<td>36.22abcd</td>
<td>4.1667a</td>
<td>87a</td>
<td>45.39a</td>
<td>416.67a</td>
<td>271.97abcd</td>
<td>938.5a</td>
<td>1210.5a</td>
</tr>
<tr>
<td><strong>2 g/l Chitosan</strong></td>
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<td>2.9733a</td>
<td>77a</td>
<td>39.123a</td>
<td>297.33a</td>
<td>201.4bcd</td>
<td>634.2a</td>
<td>835.6a</td>
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<tr>
<td><strong>0.5 g/l + 2 g/l</strong></td>
<td>35.42abcd</td>
<td>3.8a</td>
<td>94.67a</td>
<td>42.497a</td>
<td>380a</td>
<td>230.23abcd</td>
<td>825.6a</td>
<td>1055.9a</td>
</tr>
<tr>
<td><strong>High stress</strong></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td><strong>Control</strong></td>
<td>26.35d</td>
<td>2.88a</td>
<td>71.33a</td>
<td>47.877a</td>
<td>288a</td>
<td>191.4cde</td>
<td>644.6a</td>
<td>836a</td>
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<td><strong>0.5 g/l Salicylic</strong></td>
<td>35.80abcd</td>
<td>4.2133a</td>
<td>101a</td>
<td>41.45a</td>
<td>421.67a</td>
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<td>939.2a</td>
<td>1177.8a</td>
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<tr>
<td><strong>2 g/l Chitosan</strong></td>
<td>29.18cd</td>
<td>3.2a</td>
<td>70.67a</td>
<td>45.02</td>
<td>320a</td>
<td>185.97de</td>
<td>689.5a</td>
<td>875.5a</td>
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<td><strong>0.5 g/l + 2 g/l</strong></td>
<td>30.53bcd</td>
<td>3.2067a</td>
<td>80a</td>
<td>39.393a</td>
<td>320.67a</td>
<td>140.93e</td>
<td>706.8a</td>
<td>847.8a</td>
</tr>
</tbody>
</table>
Fig. 1. Effects of drought stress on dry matter yield of potato

Fig. 2. Effects of foliar application treatments on yield
Due to the importance of studying the interaction effects of foliar application of different treatments used in the study and the status of irrigation percentage on the yield of the potato crop, we analyze the results based on the application of sub-treatments:

a. Salicylic acid: The trend of changes in salicylic acid treatment on potato yield showed a significant effect on plant moisture supply status. However, the effect of salicylic acid on maintaining the yield of potatoes in 60 and 80% irrigation treatments indicates the ability of this treatment to improve yield in more severe drought stress conditions. The best yield treatment was salicylic acid treatment with 100% irrigation (Fig. 3).

b. Chitosan: The trend of changes in potato yield under different irrigation conditions using chitosan achieved individual results. The highest yield of potatoes was obtained in chitosan treatment and 100% irrigation. Also, chitosan treatments with 80 and 60% irrigation had significant differences with each other and with chitosan and 100% irrigation. The trend of performance in this treatment was directly related to reducing irrigation percentage (Fig. 3).

c. Control: No significant difference in yield in 80 and 100% irrigation treatments was observed in potato yield. While in the control treatment with 60% irrigation, the lowest yield of potatoes was obtained. This indicates the high effect of moisture status on the yield of the potato crop and the high need of this crop to use foliar materials to deal with the noticeable reduction of yield of this crop in conditions of drought stress (Fig. 3).

d. Chitosan and salicylic acid: Potato yield was reduced by reducing the moisture content (main treatment) and the simultaneous use of chitosan and salicylic acid. The yield of potatoes in 100% irrigation and application of chitosan and salicylic acid was the highest in this treatment and 60% of irrigation and application of chitosan and salicylic acid were the lowest (Fig. 3).

Fig. 3. Interaction effects of irrigation percentage and foliar application treatments on potato yield
Significantly different results were obtained for yield and shoot weight, while no significant differences were seen between other indices in reciprocal traits (Table 1). The results obtained in the performance section were very similar to those of Emami Bistgani, Siadat, Bakhshandeh, Ghasemi Pirbalouti, & Hashemi (2017) and Kulak, Ozkan, & Bindak (2019).

In drought stress, one of the main problems affecting leaves is the reduction of water content, which causes leaves to dehydrate and sometimes wither. Chitosan reduces the effects of drought stress on plants by preventing water from escaping the leaves, closing the pores, and increasing their efficiency, which can produce favorable effects on yield in the face of drought stress (Kulak, Ozkan, & Bindak, 2019). Foliar application of chitosan reduces the negative effects of drought stress on the plant by controlling the opening and closing of pores (Khan, Singha, & Panda, 2002). Although the main mechanisms of the effect of chitosan on plant growth and development have not been determined yet, its physiological effects on various plant organs have been reported (Yang, Hu, Li, Wu, & Qian, 2009). A study on the effects of chitosan as a foliar spray on potato leaves showed that the use of chitosan solution can significantly reduce the negative effects of drought stress on plants (Jiao et al., 2012). The results of the present study are similar to other studies on the foliar application of chitosan and its effects on crop growth and yield indices. However, more research is needed on the effects of salicylic acid on growth and yield characteristics.

**Photosynthetic Pigments**

Given its large leaf area, the potato plant extracts a large part of the available water in the soil, which later transpires through stomata. Availability of water is very important for growth and photosynthesis in the plant (Yang, Dong, & Liu, 2006). Photosynthesis is one of the first processes affected by drought stress (Chaves, Flexas, & Pinheiro, 2009). The change in photosynthesis is mainly due to the disruption of the opening and closing process of the pores as a result of drought stress (Li et al. 2020). Drought stress also reduces leaf chlorophyll content (Aranjuelo, Irigoyen, & Sánchez-Díaz, 2007). At the same time, drought stress conditions increase the number of active oxygen radicals, disturbing the cellular oxidative balance. Continuation of drought stress can cause oxidative stress and change enzymatic regulation in cells (Choi, Kim, Lee, Hong, & Hwang, 2007).

In our study, chlorophyll concentrations decreased with increasing intensity of drought stress (Table 2). The 100% irrigation treatment had the highest concentration of chlorophyll and showed a significant difference ($P < 0.05$) with the other two irrigation treatments. However, despite the higher concentration of chlorophyll in the 80% irrigation treatment, there was no significant difference between this treatment and the 60% treatment in terms of chlorophyll concentration. The absence of drought stress contributes to the health of the plant and the development of leaves, which are two of the main reasons for the increase in chlorophyll pigments.

In terms of total chlorophyll and chlorophyll concentration, the 2 g/l treatment with chitosan produced the best results. The 0.5 g/l salicylic acid treatments and the combined treatment with chitosan and salicylic acid were not significantly different from the chitosan treatment. At all irrigation deficit levels, the control treatment had the lowest concentration of total chlorophyll and chlorophyll $a$, and was significantly different from the other three treatments. With respect to chlorophyll $b$ concentration in potato leaves, the combination of salicylic acid and chitosan produced the best results. The chitosan treatment performed better in terms of increasing chlorophyll concentration in the leaves compared to the salicylic acid treatment and was significantly better than the control treatment. It appears that chlorophyll stability, especially in conditions of mild drought stress, can be considered as an indicator of drought resistance (Dzung, Khanh, & Dzung, 2011).

**Leaf Proline Content**

The concentration of proline in leaves is one of the crucial indicators of stress in plants. We expect that proline concentration leaves will increase with stress. However, based on the results obtained (Table 2), the concentration of proline in potato leaves does not show a significant difference between different treatments (Nohong & Syamsuddin, 2015). There were no significant differences between the control treatment, the chitosan treatment, the salicylic acid treatment, and the combined treatment in terms of leaf proline content.
Table 2. Effect of drought stress and chitosan and salicylic acid on potato physiological characteristics

<table>
<thead>
<tr>
<th>Treatment</th>
<th>C (mg/g)</th>
<th>GP (DA/min*mg)</th>
<th>TC (mg/g)</th>
<th>Ca (mg/g)</th>
<th>Cb (mg/g)</th>
<th>FP (%)</th>
<th>P (μmole/g)</th>
<th>SS (%)</th>
<th>RWC</th>
<th>LA</th>
<th>LEC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Drought stress</td>
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</tr>
<tr>
<td>No stress</td>
<td>671.54a</td>
<td>0.19a</td>
<td>148.61a</td>
<td>61.74a</td>
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<td>149.56a</td>
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<td>69.24a</td>
<td>19.56a</td>
<td>1.1a</td>
<td>89.69a</td>
<td>21.1a</td>
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<td>0.02b</td>
<td>149.56a</td>
<td>69.24a</td>
<td>19.56a</td>
<td>1.1a</td>
<td>89.69a</td>
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<td></td>
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<tr>
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<td>0.02b</td>
<td>149.56a</td>
<td>69.24a</td>
<td>19.56a</td>
<td>1.1a</td>
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<td>0.02b</td>
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<td>69.24a</td>
<td>19.56a</td>
<td>1.1a</td>
<td>89.69a</td>
<td>21.1a</td>
<td>61.5a</td>
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</table>

Remarks: Carotenoids (C), Glutathione peroxidase (GP), Total chlorophyll, Chlorophyll a (Ca), Chlorophyll b (Cb), free phenol (FP), Proline (P), Soluble sugars percent (SS), Relative water content (RWC), Leaf dry weight (LD), Leaf area (LA), Leaf electrical conductivity (LEC).

Table 3. Interactive effects of drought stress and chitosan and salicylic acid application on physiological characteristics of potato

<table>
<thead>
<tr>
<th>Treatment</th>
<th>C (mg/g)</th>
<th>GP (DA/min*mg)</th>
<th>TC (mg/g)</th>
<th>Ca (mg/g)</th>
<th>Cb (mg/g)</th>
<th>FP (%)</th>
<th>P (μmole/g)</th>
<th>SS (%)</th>
<th>RWC</th>
<th>LA</th>
<th>LEC</th>
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<tr>
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<tr>
<td>2 g/l Chitosan</td>
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<td>19.56a</td>
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Remarks: Carotenoids (C), Glutathione peroxidase (GP), Total chlorophyll, Chlorophyll a (Ca), Chlorophyll b (Cb), free phenol (FP), Proline (P), Soluble sugars percent (SS), Relative water content (RWC), Leaf dry weight (LD), Leaf area (LA), Leaf electrical conductivity (LEC).
Glutathione Peroxidase
The highest concentration of glutathione peroxidase was observed in stress-free (100% irrigation) conditions. However, under conditions of low and severe drought stress, the amount of glutathione peroxidase was significantly lower. The highest concentration of glutathione peroxidase was recorded for the chitosan treatment and the combination of chitosan and salicylic acid (Herbette et al., 2002). However, treatment with salicylic acid alone led to significantly lower glutathione peroxidase concentrations compared to other treatments.

Carotenoids
The concentration of carotenoids, similar to other plant pigments, varies depending on the intensity of stress. Based on the results presented in Table 2, the highest concentration of carotenoids was seen in the treatment without drought stress (558.42 mg/g). The lowest concentrations of carotenoids were obtained in the 60% irrigation treatment (severe drought stress), which is significantly different from the stress-free treatment. The application of salicylic acid, chitosan, and treatment with a combination of both failed to produce a significant difference in carotenoid concentrations compared to the control treatment. These results indicate that salicylic acid and chitosan do not influence the concentration of carotenoids in leaves. Water stress, however, significantly affects the concentration of this pigment in leaves.

Free Phenol Percent
Phenolic compounds are found in free radical scavenging inhibitors. These compounds are also involved in chelating the metal ions active in regenerative reactions capable of catalyzing lipid peroxidation. Phenolic compounds are one of the largest groups of natural flavonoids (Yin, Fretté, Christensen, & Grevsen, 2012) and have extensive antioxidant and antimicrobial effects, in addition to a diversity of other biological activities. Based on the results, the total phenol content was highest in stress-free conditions and lowest under extreme drought stress (60% irrigation), showing a significant difference. However, total phenol content in plants under moderate drought stress was not significantly different from plants in the stress-free treatment. Except for the salicylic acid treatment, foliar applications were not significantly different from the control treatment (Yin, Fretté, Christensen, & Grevsen, 2012). In the salicylic acid treatment, the amount of total phenol was significantly lower than the control treatment.

Soluble Sugar Percent
The number of soluble carbohydrates in leaves increased significantly with increasing drought stress (Table 3). In conditions of severe drought stress, the number of soluble sugars in the leaves was significantly higher than in stress-free conditions. Medium drought stress also increased the concentration of soluble sugars, but not significantly. Application of chitosan slightly increased the number of soluble sugars in the leaves compared to the control treatment, but this difference was not significant. Also, the use of salicylic acid alone and the combined use of chitosan and salicylic acid did not significantly change the amount of soluble sugars compared to the control treatment (Nohong & Syamsuddin, 2015).

Drought stress is one of the most critical factors in reducing agricultural yields worldwide. In the present study, we investigated the effect of foliar application of salicylic acid and chitosan on yield, yield components, and physiological characteristics of potato plants under drought and non-drought conditions. The results clearly showed the negative effects of stress, especially severe drought, on the yield characteristics and growth indicators of potatoes. However, the impact of chitosan and salicylic acid, although considerable in a limited number of indicators, did not generally lead to significant differences in the most important indicators. Therefore, based on the results of this study, the best way to deal with reduced potato yield in dry areas is to irrigate the field or change the planting date to avoid extreme heat and the higher water requirements of the plants.

CONCLUSION AND SUGGESTION
Drought stress is one of the most critical factors in reducing agricultural yield worldwide. Arid regions cover about half of Earth’s land area, with Africa, Asia, Latin America, and Europe accounting for 66%, 40%, 24%, and 15% of arid areas, respectively. Approximately 72% of arid regions and 100% of very arid regions are located in developing countries. Therefore, many studies are underway to address the effects of drought stress on crop yields, especially in arid regions. In
In this research, we investigated the effect of foliar application of salicylic acid and chitosan on yield, yield components, and physiological characteristics of potato plants under drought and non-drought conditions. The results showed the negative effect of drought stress, especially severe drought, on the yield characteristics and growth indicators of the potato plant. The impact of spraying chitosan and salicylic acid, although considerable for a limited number of indicators, did not lead to a significant difference in the important indicators overall. Therefore, the best way to deal with reduced potato yield in dry areas is to irrigate the field completely or to change the planting date to avoid extreme heat and the higher water requirements of the potato crop. Due to the prevalence of drought stress worldwide and the positive effects of chitosan and salicylic acid on reducing the effects of stress on crops, it is recommended that similar studies be conducted on other major crops in arid and semi-arid regions of the world. The results of such studies should be made available to countries with arid and semi-arid climates by FAO in order to encourage farmers to use these novel solutions to reduce the effects of stress and increase yield.

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Seyyed Ali Morovvat et al.: Optimal Effects of Chitosan and Salicylic Acid on Drought Stress


