



## Growth, Yield and Resistance Responses of Three Cultivars on True Seed Shallots to Twisted Disease with Salicylic Acid Application

Rachmanto Bambang Wijoyo<sup>1</sup>, Endang Sulistyaningsih<sup>1\*</sup> and Arif Wibowo<sup>2</sup>

<sup>1</sup>Department of Agronomy, Faculty of Agriculture, Universitas Gadjah Mada, Yogyakarta, Indonesia;

<sup>2</sup>Department of Plant Protection, Faculty of Agriculture, Universitas Gadjah Mada, Yogyakarta, Indonesia

\*Corresponding author: [endangsih@ugm.ac.id](mailto:endangsih@ugm.ac.id)

### Abstract

Twisted disease is one of the main diseases on shallot caused by *Fusarium* sp. One method to increase the resistance of shallot cultivars to the twisted disease can be performed by using True Seed Shallot (TSS) combining with a salicylic acid application. TSS is known as free pathogen. The objective of the research was to examine the effects of a salicylic acid application to the growth, yield and resistance responses of TSS on Tuktuk, Sanren and Lokananta cultivars to the twisted disease. The research was arranged in factorial Completely Randomized Design (CRD), consisting of two factors with three replications. The first factor included Tuktuk, Sanren and Lokananta cultivars and the second factor comprised salicylic acid applications (without salicylic acid and inoculation, without salicylic acid with inoculation, immersion treatment of salicylic acid with inoculation, spray treatment with inoculation as well as immersion and spray treatment with inoculation). The results showed that the application of salicylic acid in different treatments had the same effects on the growth, yield and resistance of TSS. An interesting result is found, in which the application of salicylic acid by immersing and spraying could reduce the incidence of twisted disease by 12.5% in TSS. Therefore, the application of salicylic acid by immersing and spraying can be one of the recommended twisted disease management.

**Keywords:** induced systemic resistance; salicylic acid; true seed shallots

**Cite this as:** Wijoyo, R. B., Sulistyaningsih, E., & Wibowo, A. (2020). Growth, Yield and Resistance Responses of Three Cultivars on True Seed Shallots to Twisted Disease with Salicylic Acid Application. *Caraka Tani: Journal of Sustainable Agriculture*, 35(1), 1-11. doi: <http://dx.doi.org/10.20961/carakatani.v35i1.30174>

### INTRODUCTION

Shallots (*Allium cepa* L. Aggregatum group) is one of the important horticultural commodities for Indonesian people. Shallots are often used as a condiment to enhance the flavor on other foods because it contains nutrients and compounds that are classified as non-nutritional substances and enzymes that are useful for improving and maintaining the health of the human body (Minh, 2019). The demand for shallots increases every year and tends to be evenly distributed at any time, while the production of shallots is seasonal (Astuti et al., 2019). The decrease of shallots productivity is caused by the attack of plant-disturbing

organisms that make cultivars susceptible to pathogens (Yusidah and Istifadah, 2018). The disease can reduce the quality and quantity of shallot yield. Twisted disease caused by *Fusarium acutatum* is one of the main diseases of shallot plant that caused significant yield loss (Lestiyani, 2015). During 2003-2005, cumulative infection of twisted disease was noted increasing from 48.1 ha to 268.1 ha (BPS, 2017). Typical symptoms of the disease are yellowing of leaves, twisting on the leaves and drying on roots (Wiyatiningsih, 2011).

Agrochemical firms have been actively influencing farmers to continue using chemical pesticides. The use of chemical pesticides tends to

\* Received for publication May 18, 2019

Accepted after corrections September 25, 2019

cause serious environmental degradation and poses a threat to human health, hence it may not be sustainable (Kamarulzaman et al., 2012). The application of pesticides is the most frequently used method for disease control. The utilization of synthetic pesticides can cause negative impacts on the environment, whereas, the residue can not be decomposed by organisms except certain microorganisms. Moreover, pesticide residues also accumulate in cells or tissues of organisms (Soemirat and Ariesyadi, 2017). Due to the use of synthetic pesticides, therefore an eco-friendly-alternative is needed (Chang et al., 2016). One of the ways to reduce the detrimental effects of chemical pesticides is to apply salicylic acid as a resistance inducer in plants (Dihazi et al., 2011). Induction of resistance is one of the ways that can be applied to control the disease. Induction causes the physiological conditions to activate the resilience system and stimulate the natural resistance mechanism of the host through the application of external inducers, i.e. biological, chemical and physical agents (Agrios, 2005).

In general, farmers cultivate shallots using bulbs seeds (vegetatively). Using seeds from the same shallot bulb cultivar continuously often decreases the quality of bulbs and yields because the quality of the bulbs is less-guaranteed due to the accumulation of pathogens in bulbs (Shimeles, 2014). One of the efforts to increase the resistance of shallot cultivars to the twisted disease can be performed by using TSS (True Seed Shallot) since TSS is known free of pathogens (Pangestuti and Sulistyaningsih, 2011). The use of TSS is one alternative that can be developed to improve the quality of shallot seeds. PT. East West Seed Indonesia is one of the TSS producers that has introduced Tuktuk, Sanren and Lokananta TSS cultivar seeds. The advantages of growing shallots using seeds include reducing production costs both in the provision of planting material and transportation, being free from tubular infectious pathogens and requiring more efficient handling (Sumarni et al., 2016).

Tuktuk cultivars are superior varieties of shallots that can adapt well in the lowlands with a height of 20-220 meters above sea level and are well-planted in the dry season. Tuktuk yields can reach 20-30 tons ha<sup>-1</sup>, while conventional bulb seed yields range from 8-12 tons ha<sup>-1</sup>. This contributes to an increase in the production of 12-18 tons ha<sup>-1</sup> (Buda et al., 2018). Sanren cultivar is a shallot variety that can produce well when

planted during the dry and the rainy seasons and has a high production level and medium-size bulbs. This variety is resistant to twisted and anthracnose diseases which often attack during the rainy season (Askari-khorasgani and Pessarakli, 2019). Lokananta cultivar is a shallot variety that is resistant to twisted disease, has a high production and is suitable for planting in the lowlands. The yield of bulbs per hectare is  $\pm$  18.49-24.58 tons ha<sup>-1</sup> (Saidah et al., 2019).

Moreover, the resistance of shallots to twisted disease can also be increased by inducing the resistance using salicylic acid on TSS through immersion or spray technique (Mandal et al., 2009). Accumulation of salicylic acid in plant tissue is involved to activate a signal of Pathogenesis-Related Proteins (PR-proteins) formation and induce systemic resistance mechanism when pathogen infects plant (Juwanda et al., 2016). In addition, the salicylic acid has an important role as a phytohormone that regulates plant growth, especially the physiological activities such as photosynthesis, ethylene-producing nitrate metabolism, regulation to abiotic stresses and signal molecules that play a role in the resistance to pathogens (Muthulakshmi and Lingakumar, 2017). The objective of the present study is to examine the effects of salicylic acid application to the growth, yield and resistance responses of TSS on Tuktuk, Sanren and Lokananta cultivars to twisted disease.

## MATERIALS AND METHOD

The research was conducted on February-August 2018 in the experimental farm, Control Technology Laboratory and Plant Production Management Laboratory, Faculty of Agriculture, Universitas Gadjah Mada, Yogyakarta. The research was arranged in a factorial Completely Randomized Design (CRD), consisting of two factors with three replications. The first factor included TSS cultivars (Tuktuk, Sanren and Lokananta) and the second comprised the application method of salicylic acid (without salicylic acid and inoculation, without salicylic acid with inoculation, immersion treatment of salicylic acid with inoculation, spray treatment with inoculation as well as immersion and spray treatment with inoculation). The materials consist of TSS of Tuktuk, Sanren and Lokananta cultivars, pure *Fusarium acutatum* culture collection from Control Technology Laboratory,

sterile soil, manure, NPK fertilizer 15-15-15, SP-36 fertilizer, KCl fertilizer, NPK fertilizer 15-9-20, aquades, FeCl<sub>3</sub>, liquid nitrogen, potato dextrose agar (PDA) medium and peptone-pentachloronitrobenzene (PCNB) medium.

The treatment of salicylic acid using immersion treatment was carried out by immersing TSS in a 15 ppm salicylic acid solution for 30 minutes following Khotimah et al. (2017). The spray treatment was performed by spraying 15 ppm salicylic acid solution on the seedlings from the seeds as much as 50 ml per 500 cm<sup>2</sup> two days before planting (38 days after seedling) (Krishardianto and Sukma, 2017). The planting medium was sterilized soil prepared by steaming in an autoclave for approximately two hours. Sterilized soil was mixed with manure with a dose of 4 tons ha<sup>-1</sup> (Suwandi et al., 2016). The mixture of planting media was put into a tray of 32 cm x 45 cm x 16 cm. *Fusarium* sp. was inoculated in the planting medium by drenching method at one week after the transplantation (WAT). The planting medium was poured with 240 ml per tray conidia suspension with a density of 10<sup>6</sup> conidia ml<sup>-1</sup> with distilled water (Cahyaningrum et al., 2017).

In the observation, the incubation period was observed, particularly when the symptoms of the

twisted disease first appeared in plants in each treatment combination in units of weeks. Disease incidence was observed by counting the number of plants that showed twisted disease symptoms.

$$\text{Disease incidence} = \frac{a}{b} \times 100 \%$$

Note:

a = Total number of plants showing twisted disease symptoms

b = Total number of plant population

The total population of *Fusarium* in the soil was observed by taking a 3 g soil sample per treatment diluted with 27 ml of aquabidest and then homogenized with a vortex. The suspension was diluted again by taking 1 ml of suspense dissolved with 9 ml of aquabidest and shaken until homogeneous. Dilution was carried out up to 10<sup>-3</sup>. Then, the suspension at 10<sup>-3</sup> dilution was taken as much as 100 µl and poured on PCNB media and flattened with L-glass and then incubated for 7-14 days. Colony density was calculated by counting the number of fungal colonies in each petri dish and then putting them into the formula (Sinta, 2018).

$$\text{Total population of } Fusarium \text{ in the soil} = \frac{\Sigma \text{Fusarium colony}}{\text{dilution factor} \times \text{ml suspension}}$$

Salicylic acid content was observed by using spectrophotometry (Warrier et al., 2013). The first step to do this measurement was making a standard solution using distilled water as the solvent to obtain the standard curve equation. After that, 50 mg of fresh leaves were crushed with liquid nitrogen and added with 1 ml of distilled water. The leaf extract was then centrifuged at a speed of 10,000 g for 10 minutes to take 100 µl supernatant and mixed with 0.1% ferric chloride solution up to 3 ml. The complex formation between Fe<sup>3+</sup> ions and salicylic acid was violet. The complex formation was determined by spectrophotometry at A540 nm. The absorbance value was entered into the standard curve equation to obtain endogenous salicylic acid content (Warrier et al., 2013).

The data of the surface area and length of roots, leaf area index (LAI), plant growth rate, total dry weight, harvest index and fresh weight of bulbs

were obtained. All the observation data were analyzed using variance analysis with α 5% followed by DMRT analysis if there were significant differences between treatments.

## RESULTS AND DISCUSSION

The results showed that there was no interaction between cultivars and salicylic acid application methods to the incubation period, disease incidence, total *Fusarium* population in the soil, root surface area, root length, LAI, plant growth rate, total dry weight, harvest index and natural dry weight of shallot per clump, but there were interactions between cultivars and salicylic acid application methods to the endogenous salicylic acid content. Shallot plants that were inoculated by *Fusarium* fungus caused twisted disease in the field. Symptoms on the infected plants showed yellowing and twisting on the leaves (Figure 1).

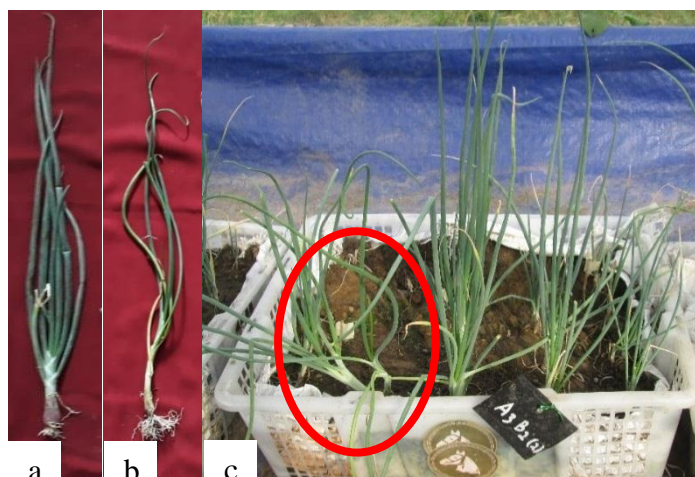


Figure 1. Twisted disease symptoms on shallots, (a) healthy plant, (b) infected plant, (c) yellowing and twisting

The incubation period illustrated the first time of the symptoms of the twisted disease appeared on the shallots. Tuktuk, Sanren and Lokananta cultivars had the same incubation periods (Table 1). Salicylic acid in different application methods did not show any effect on the incubation period. However, the presence of inoculum on the planting medium accelerated the incubation period until five days faster than incubation without fungal inoculation. Tuktuk cultivar had a

higher disease incidence than Sanren and Lokananta cultivars. This indicates that Tuktuk cultivars are more susceptible to twisted disease compared to Sanren and Lokananta cultivars. The treatment of salicylic acid by immersing and spraying had reduced the incidence of disease by 12.5%. This result has confirmed that this treatment can significantly reduce disease incidence.

Table 1. The incubation period (the week after inoculation/WAI), disease incidence (%) at 7 WAT and population of *Fusarium* in the soil at 16 WAT

Treatments	Incubation period (WAI)	Disease incidence (%)	Total population of <i>Fusarium</i> in the soil (cfu g <sup>-1</sup> of soil)
<b>Cultivars</b>			
Tuktuk	4.13a	28.33a	9.06 x 10 <sup>3</sup> b
Sanren	4.47a	22.92b	17.13 x 10 <sup>3</sup> a
Lokananta	4.33a	20.83b	8.60 x 10 <sup>3</sup> b
<b>Salicylic acid applications</b>			
Without salicylic acid, without inoculation	8.78p	5.56r	5.44 x 10 <sup>3</sup> r
Without salicylic acid + inoculation	3.22q	34.03p	11.00 x 10 <sup>3</sup> pq
Immersion treatment of salicylic acid + inoculation	3.11q	31.25pq	17.33 x 10 <sup>3</sup> p
Spray treatment of salicylic acid + inoculation	3.44q	27.78pq	14.56 x 10 <sup>3</sup> pq
Immersion and spray treatment of salicylic acid + inoculation	3.00q	21.53q	9.67 x 10 <sup>3</sup> q
CV (%)	17.64	5.29	16.77

Note: The numbers followed by the same letters in the same column do not show significant differences based on the DMRT at 5% confidence level

There were differences in the number of total *Fusarium* population in soil samples (Table 1). The population of fungus on inoculated soil

showed significantly greater results (9.67 x 10<sup>3</sup> - 17.33 x 10<sup>3</sup>) compared to those of the population that were not inoculated using *Fusarium* (5.44 x

10<sup>3</sup>) in the soil. *Fusarium* population in the soil demonstrated that they could live in the same conditions in the soil. The twisted disease that had been treated with salicylic acid was different from those without treatment with salicylic acid. Furthermore, the high number of *Fusarium* at the end of harvest had increased the incidence of plant diseases and shortened the incubation period in shallots. The different application methods of salicylic acid showed no significant results in decreasing *Fusarium* population in the soil. This indicates that the application of exogenous salicylic acid by immersing the seeds and

spraying the nursery does not reduce the *Fusarium* population in the soil.

The three cultivars of shallots have different responses to the content of endogenous salicylic acid at 10 days after the inoculation (Table 2). Lokananta has significantly higher endogenous salicylic acid content than Sanren and Tuktuk on fungal-inoculated media treatment. However, the same content of endogenous salicylic acid is observed on both Lokananta and Sanren cultivars in immersing and spraying treatments. Abbaspour and Ehsanpour (2016) reported that pathogen inoculation in plants induced plant resistance through the content of endogenous salicylic acid.

Table 2. Endogenous salicylic acid content (ppm) in the leaf at 10 days after inoculation (DAI)

Cultivars	Application methods					Average
	Without salicylic acid, without inoculation	Without salicylic acid + inoculation	Immersion treatment of salicylic acid + inoculation	Spray treatment of salicylic acid + inoculation	Immersion and spray treatment of salicylic acid + inoculation	
Tuktuk	19.87de	27.20de	13.20e	19.20de	32.53d	22.40
Sanren	18.53de	20.53de	9.87e	15.87de	57.87c	24.53
Lokananta	16.20de	66.53ab	77.87a	74.53ab	59.87bc	59.00
Average	18.20	38.09	33.64	36.53	50.09	(+)
CV (%)	25.95					

Note: The numbers followed by the same letters in the same column do not show any significant differences based on the DMRT at 5% confidence level. The sign (+) shows there is an interaction between factors

Tuktuk cultivars, both with and without fungus inoculation, have the same endogenous salicylic acid. The endogenous salicylic acid content on Tuktuk inoculated with fungus treatment, was not significantly different from that without salicylic acid application. However, salicylic acid application by immersing and spraying increased endogenous salicylic acid content (59.42%), significantly higher than the increase in content when using immersing, but not different from then increase when using spraying application. Similar endogenous salicylic acid content was observed on Sanren cultivars, both with inoculation of fungus and salicylic acid by immersing and spraying treatments and without inoculation of fungus and salicylic acid. However, application of salicylic acid by immersing and spraying on the shallots inoculated with fungus appeared to have significantly higher endogenous salicylic acid compared to the plants inoculated with fungus which were treated by immersing or spraying. Lokananta cultivars inoculated with fungus had a

significantly higher endogenous salicylic acid compared to plants that were not inoculated with fungus. This cultivar, inoculated with fungus and treated with salicylic acid in different application methods, had the same endogenous salicylic acid with that inoculated with fungus in the planting media without salicylic acid. Salicylic acid treatment by immersing did not have any significant effects on the content of endogenous salicylic acid compared to salicylic acid treatment by spraying or without salicylic acid. However, the content was significantly higher than salicylic acid treatment by immersing and spraying.

Tuktuk cultivar was more susceptible to the twisted disease than both Sanren and Lokananta cultivars. Tuktuk cultivar had lower endogenous salicylic acid content than Lokananta cultivars. Tuktuk cultivars contained endogenous salicylic acid which was the same as the content of endogenous salicylic acid in Sanren cultivars, but it had a different response to the treatment of exogenous salicylic acid. The treatment of

exogenous salicylic acid in Tuktuk cultivars increased the content of endogenous salicylic acid by 16.38%, whereas the treatment of exogenous salicylic acid in Sanren cultivars increased the content of endogenous salicylic acid to 64.42%. It confirms that Sanren is the most responsive cultivar to the treatment of exogenous salicylic acid. Salicylic acid as an endogenous component of plants is synthesized in response to local and systemic infections produced in uninfected tissue and contributes to systemic expression and systemic resistance induced (Hayat et al., 2013).

Roots are the first part of the plant affected by

*Fusarium* infection, so observations of root morphology are important (Table 3). Tuktuk, Sanren and Lokananta cultivars had the same length and surface area of roots at 3 and 7 WAT. The treatment of salicylic acid in different application methods gave the same effects on the length and surface area of roots at 3 and 7 WAT. Twisted disease caused by *Fusarium* sp. is a soil-borne pathogen where the initial attack of this pathogen infects the roots or leaves contact directly to the soil surface. Fadhillah et al. (2014) reported that *Fusarium*-affected plants showed disturbed root growth.

Table 3. The width and length of roots at 3 and 7 WAT

Treatments	Surface area of roots (cm <sup>2</sup> )		Length of roots (cm)	
	3 WAT	7 WAT	3 WAT	7 WAT
<b>Cultivars</b>				
Tuktuk	2.84a	13.39a	6.79a	23.23a
Sanren	3.11a	14.41a	10.50a	23.59a
Lokananta	3.43a	15.53a	8.29a	18.93a
<b>Salicylic acid applications</b>				
Without salicylic acid, without inoculation	2.94p	8.79p	7.89p	15.82p
Without salicylic acid + inoculation	3.84p	16.35p	9.42p	22.89p
Immersion treatment of salicylic acid + inoculation	2.53p	19.38p	7.30p	29.44p
Spray treatment of salicylic acid + inoculation	2.85p	16.02p	9.52p	23.19p
Immersion and spray treatment of salicylic acid + inoculation	3.49p	11.67p	8.50p	18.24p
CV (%)	23.71	25.85	28.36	28.60

Note: The numbers followed by the same letters in the same column do not show any significant differences based on the DMRT at 5% confidence level

The LAI is the ratio of the overall leaf area to the land area. The LAI values at 3 and 7 WAT were not significantly different in TSS cultivars and the application method of salicylic acid (Table 4). The application of salicylic acid by immersing, spraying and combination of immersing and spraying on shallots inoculated with fungus showed the LAI values that were not significantly different from the shallots which were not treated with salicylic acid. The shoot also has an important role in the accumulation of dry weight determined by the results of photosynthesis, where the process occurs in the shoot of plants, especially leaves in which the capture of sunlight by the leaves will determine the process of photosynthesis (Zakariyya, 2016).

The growth rate of Lokananta cultivars was 54.66% and 40.51% significantly higher than the

rates of Tuktuk and Sanren cultivars, respectively (Table 5). The different application methods of salicylic acid gave the effect that was not significantly different from the rate of plant growth. Application of salicylic acid by immersing, spraying and the combination of immersing and spraying which had been inoculated with fungus did not show significant effects on the rate of plant growth compared to the shallots which were not treated with salicylic acid. The growth rate rises by the increase in leaf area of the plant so that much sunlight will be captured and can be used for photosynthesis. The increase in plant growth rate plays an important role in escalating the accumulation of assimilates in parts of the plant which further support the formation of crop yields (Gardner et al., 2008).

Table 4. LAI at 3 and 7 WAT

Treatments	LAI	
	3 WAT	7 WAT
<b>Cultivars</b>		
Tuktuk	0.81a	1.55a
Sanren	0.98a	1.82a
Lokananta	1.07a	2.39a
<b>Salicylic acid applications</b>		
Without salicylic acid, without inoculation	0.99p	2.00p
Without salicylic acid + inoculation	1.07p	1.90p
Immersion treatment of salicylic acid + inoculation	0.88p	1.73p
Spray treatment of salicylic acid + inoculation	0.96p	2.33p
Immersion and spray treatment of salicylic acid + inoculation	0.86p	1.65p
CV (%)	16.10	16.45

Note: The numbers followed by the same letters in the same column do not show any significant differences based on the DMRT at 5% confidence level

Table 5. Plant growth rate ( $\text{mg cm}^2 \text{ week}^{-1}$ ) for the period of 3-7 WAT

Treatments	Plant growth rate ( $\text{mg cm}^2 \text{ week}^{-1}$ )
<b>Cultivars</b>	
Tuktuk	1.41b
Sanren	1.85b
Lokananta	3.11a
<b>Salicylic acid applications</b>	
Without salicylic acid, without inoculation	1.96p
Without salicylic acid + inoculation	1.82p
Immersion treatment of salicylic acid + inoculation	1.60p
Spray treatment of salicylic acid + inoculation	2.60p
Immersion and spray treatment of salicylic acid + inoculation	2.65p
CV (%)	0.15

Note: The numbers followed by the same letters in the same column do not show significant differences based on the DMRT at 5% confidence level

In general, the application of salicylic acid by immersing, spraying, and immersing and spraying on the three cultivars indicated that accumulation of total dry weight was not different from the total weight of the cultivars that did not receive treatment with salicylic acid (Table 6). The result showed that the total dry weights of both of on three cultivars and in the different salicylic acid applications at 3 WAT were not significantly different. Lokananta cultivar at 7 WAT had a significantly higher total dry weight of plants than Tuktuk and Sanren cultivars. The response of total dry weight accumulation of plants at 7 WAT to the different application methods of salicylic acid was directly proportional to the rate of plant growth. Gardner et al. (2008) reported that total dry weight is influenced by plant growth rate.

The different application methods of salicylic acid have the same effect on the index value of shallot harvest (Table 7). Harvest index shows the proportion of economic dry weight (bulbs) to the total dry weight of shallot. Higher harvest index value indicates that economical dry weight produced by assimilates is high. Lokananta cultivar had a higher total dry weight at 7 WAT than Tuktuk and Sanren. Generally, salicylic acid applications by immersing, spraying, and immersing and spraying on inoculated shallot plants gave the same effect on plants without salicylic acid treatment and without inoculation of fungus on the growing media. This shows that all treatments have the same role in translocating assimilates to bulbs.

Table 6. Total dry weight (g) at 3 and 7 WAT

Treatments	Total dry weight (g)	
	3 WAT	7 WAT
<b>Cultivars</b>		
Tuktuk	0.13a	0.54b
Sanren	0.15a	0.68b
Lokananta	0.17a	1.07a
<b>Salicylic acid applications</b>		
Without salicylic acid, without inoculation	0.16p	0.73p
Without salicylic acid + inoculation	0.18p	0.70p
Immersion treatment of salicylic acid + inoculation	0.12p	0.58p
Spray treatment of salicylic acid + inoculation	0.14p	0.89p
Immersion and spray treatment of salicylic acid + inoculation	0.14p	0.90p
CV (%)	10.85	16.95

Note: The numbers followed by the same letters in the same column do not show significant differences based on the DMRT at 5% confidence level

Table 7. Harvest index of three TSS cultivars with different salicylic acid applications

Treatments	Harvest index
<b>Cultivars</b>	
Tuktuk	0.50a
Sanren	0.42a
Lokananta	0.48a
<b>Salicylic acid applications</b>	
Without salicylic acid, without inoculation	0.48p
Without salicylic acid + inoculation	0.43p
Immersion treatment of salicylic acid + inoculation	0.51p
Spray treatment of salicylic acid + inoculation	0.48p
Immersion and spray treatment of salicylic acid + inoculation	0.45p
CV (%)	27.04

Note: The numbers followed by the same letters in the same column do not show significant differences based on the DMRT at 5% confidence level

Lokananta cultivar had fresh bulbs weight of 19.44% which was significantly higher than the fresh bulb weight of Tuktuk and Sanren (Table 8). *Fusarium* non-inoculated plants on the planting media had the same effects as plants inoculated with *Fusarium*. Furthermore, salicylic acid treatment in different application methods on the inoculated plants with *Fusarium* gave the same effects as the plants which were neither treated with salicylic acid nor inoculated.

*Fusarium* infection on shallot triggers a resistance mechanism through the accumulation of endogenous salicylic acid, especially on the leaves. Salicylic acid application by immersing and spraying can increase the resistance of shallots by decreasing 12.5% of twisted disease incidence. Successful inducing resistance to the

diseases is also influenced by several factors, such as incompatibility between chemicals and plants, appropriate dosage, induction method and period between induction treatment and inoculation (Hayat et al., 2010). Abo-Elyousr et al. (2009) reported that the highest content of endogenous salicylic acid on onion plants is observed at ten days after salicylic acid application and inoculation of *S. vesicarium*. A similar result is also exposed; endogenous salicylic acid content is also found on Tuktuk and Sanren cultivars treated with salicylic acid by immersing and spraying at ten DAI, as well as the salicylic acid on Lokananta cultivar by immersing. However, this application is not significantly different from the plants without inoculation and salicylic acid application on Tuktuk and Lokananta cultivars.



Table 8. The natural dry weight of TSS per clump

Treatments	Natural dry weight of shallots per clump (g)
<b>Cultivars</b>	
Tuktuk	15.02b
Sanren	15.01b
Lokananta	18.62a
<b>Salicylic acid applications</b>	
Without salicylic acid, without inoculation	14.59p
Without salicylic acid + inoculation	15.81p
Immersion treatment of salicylic acid + inoculation	17.82p
Spray treatment of salicylic acid + inoculation	16.78p
Immersion and spray treatment of salicylic acid + inoculation	16.06p
CV (%)	23.14

Note: The numbers followed by the same letters in the same column do not show significant differences based on the DMRT at 5% confidence level

Induction of resistance in a plant is closely related to the content of salicylic acid as the receptor that will activate the formation of PR-Protein (Murphy et al., 2001). In this study, salicylic acid treatment had no significant effects on the incidence of *Fusarium* in shallots. A similar result was also reported by Khotimah et al. (2017) that exogenous treatment of salicylic acid had no significant effects on the incubation period and the incidence of disease. Disease incidence is more influenced by the genetic structure of plant cultivar than the addition of exogenous salicylic acid. This is in line with the statement of Leino et al. (2018) that each cultivar is affected by different genetic traits. Salicylic acid can support the plant resistance by reducing the percentage of disease incidence, but there does not appear any relation to the plant growth rate, accumulation of total dry weight and fresh weight of bulbs.

## CONCLUSIONS

This study showed that the application of salicylic acid in different application methods gave the same effects on the growth and yield of TSS and the treatment of salicylic acid by immersing and spraying could reduce 12.5% of the twisted disease incidence in TSS by accumulating its endogenous salicylic acid.

## ACKNOWLEDGMENT

The present study was fully funded by the Australian Center for International Agricultural Research (ACIAR) project HORT 2009-056.

## REFERENCES

- Abbaspour, J., & Ehsanpour, A. (2016). The impact of salicylic acid on some physiological responses of *Artemisia aucheri* Boiss. under in vitro drought stress. *Acta Agriculturae Slovenica*, 107(2), 287. <https://doi.org/10.14720/aas.2016.107.2.03>
- Abo-Elyousr, K. A. M., Hussein, M. A. M., Allam, A. D. A., & Hassan, M. H. (2009). Salicylic acid induced systemic resistance on onion plants against *Stemphylium vesicarium*. *Archives of Phytopathology and Plant Protection*, 42(11), 1042–1050. <https://doi.org/10.1080/03235400701621719>
- Agrios, G. (2005). Plant pathology: Fifth edition. In *Plant Pathology: Fifth Edition*. <https://doi.org/10.1016/C2009-0-02037-6>
- Askari-khorasgani, O., & Pessarackli, M. (2019). Agricultural management and environmental requirements for production of true shallot seeds. *Advances in Plants & Agriculture Research*, 9(2), 318–322. <https://doi.org/10.15406/apar.2019.09.00441>
- Astuti, L. T. W., Daryanto, A., Syaukat, Y., & Daryanto, H. K. (2019). Technical Efficiency of Shallot Farming in Central Java Province: Stochastic Frontier Modelling. *International Journal of Progressive Sciences and Technologies*, 13(2), 222–232. Retrieved from <http://ijpsat.ijsh-t-journals.org/index.php/ijpsat/article/view/800>
- BPS. (2017). *Statistik Tanaman Sayuran dan*

- Buah-buahan Semusim Indonesia 2017*. Retrieved from <https://www.bps.go.id/publication/2018/10/05/bbd90b867a6ee372e7f51c43/statistik-tanaman-sayuran-dan-buah---buah-an-semusim-indonesia-2017.html>
- Buda, I., Agung, I., & Ardhana, I. (2018). Nitrogen Fertilizer Increased Bulb Diameter and Yields of True Seed and Bulb –propagated Shallot Varieties. *International Journal of Innovative Research in Science, Engineering and Technologi*, 7(1), 80–86. <https://doi.org/10.15680/IJRSET.2018.0701007>
- Cahyaningrum, H., Prihatiningsih, N., & Soedarmono, S. (2017). Intensitas dan Luas Serangan Beberapa Isolat *Fusarium oxysporum* f.sp. zingiberi pada Jahe Gajah. *Jurnal Perlindungan Tanaman Indonesia*, 21(1), 16-22. <https://doi.org/10.22146/jpti.17743>
- Chang, P. L., Hsieh, M. M., & Chiu, T. C. (2016). Recent advances in the determination of pesticides in environmental samples by capillary electrophoresis. *International Journal of Environmental Research and Public Health*, 13(4), 1–20. <https://doi.org/10.3390/ijerph13040409>
- Dihazi, A., Serghini, M. A., Jaiti, F., Daayf, F., Driouich, A., Dihazi, H., & El Hadrami, I. (2011). Structural and Biochemical Changes in Salicylic-Acid-Treated Date Palm Roots Challenged with *Fusarium oxysporum* f. sp. albedinis. *Journal of Pathogens*, 2011, 1–9. <https://doi.org/10.4061/2011/280481>
- Fadhilah, S., Wiyono, S., & Surahman, M. (2014). Pengembangan Teknik Deteksi Fusarium Patogen Pada Umbi Benih Bawang Merah (*Allium ascalonicum*) di Laboratorium [Development of Detection Technique for Fusarium Pathogen on Seedling Shallot (*Allium ascalonicum*) Bulb at Laboratorium]. *Hortikultura*, 24(2), 171–178. <http://dx.doi.org/10.21082/jhort.v24n2.2014.p171-178>
- Gardner, F. P., Pearce, R. B., & Mitchell, R. L. (2008). Fisiologi Tanaman Budidaya. In *UI Press*. Jakarta: UI Press.
- Hayat, Q., Hayat, S., Irfan, M., & Ahmad, A. (2010). Effect of exogenous salicylic acid under changing environment: A review. *Environmental and Experimental Botany*, 68(1), 14–25. <https://doi.org/10.1016/j.envexpbot.2009.08.005>
- Hayat, S., Ahmad, A., & Alyemeni, M. N. (2013). Salicylic acid. Plant growth and development. In *Reactions Weekly*. <https://doi.org/10.2165/00128415-201013250-00111>
- Juwanda, M., Khotimah, K., & dan Amin, M. (2016). Peningkatan Ketahanan Bawang Merah Terhadap Penyakit Layu Fusarium Melalui Induksi Ketahanan Dengan Asam Salisilat Secara Invitro. *Agrin*, 20(1), 15–28. Retrieved from <http://lib.ui.ac.id/file?file=digital/2017-5/20451161-310-528-1-SM.pdf>
- Kamarulzaman, N. H., Mazlan, N., Rajendran, S. D., & Mohayidin, M. G. (2012). Role of biopesticides in developing a sustainable vegetable industry in Malaysia. *International Journal of Green Economics*, 6(3), 243–259. <https://doi.org/10.1504/ijge.2012.050973>
- Khotimah, K., Sulistyaningsih, E., & Wibowo, A. (2017). In Vitro Induced Resistance of Fusarium Wilt Disease (*Fusarium oxysporum* f.sp. cepae) by Salicylic Acid in Shallot CV ‘Bima Brebes.’ *Ilmu Pertanian (Agricultural Science)*, 2(1), 001–008. <https://doi.org/10.22146/ipas.12840>
- Krishardianto, A., & Sukma, D. (2017). Morphological Characterization and Effects of Treatments Fertilization and Gift Silica (Si) on Genotype Hybrid Cattleya Orchids. *Bul. Agrohorti*, 5(2), 167–175. <https://doi.org/10.29244/agrob.5.2.167-175>
- Leino, M. W., Solberg, S., Tunset, H. M., Fogelholm, J., Strese, E. M. K., & Hagenblad, J. (2018). Patterns of Exchange of Multiplying Onion (*Allium cepa* L. Aggregatum-Group) in Fennoscandian Home Gardens. *Economic Botany*, 72(3), 346–356. <https://doi.org/10.1007/s12231-018-9426-2>
- Lestiyani, A. (2015). *Identifikasi, patogenisitas dan variabilitas penyebab penyakit moler pada bawang merah* (Universitas Gadjah Mada). Retrieved from [http://etd.repository.ugm.ac.id/index.php?mod=penelitian\\_detail&sub=PenelitianDetail&act=view&typ=html&buku\\_id=81416&obyek\\_id=4](http://etd.repository.ugm.ac.id/index.php?mod=penelitian_detail&sub=PenelitianDetail&act=view&typ=html&buku_id=81416&obyek_id=4)
- Mandal, S., Mallick, N., & Mitra, A. (2009). Salicylic acid-induced resistance to *Fusarium oxysporum* f. sp. lycopersici in tomato. *Plant*

- Physiology and Biochemistry*, 47(7), 642–649. <https://doi.org/10.1016/j.plaphy.2009.03.001>
- Minh, N. P. (2019). Technical Factors Affecting To Pickle Shallot (*Allium Ascalonicum*) Fermentation. *J. Pharm. Sci. & Res*, 11(3), 879–881. Retrieved from <https://www.jpsr.pharmainfo.in/Documents/Volumes/vol11issue03/jpsr11031938.pdf>
- Murphy, A. M., Gilliland, A., Eng Wong, C., West, J., Davinder, D. P., & Carr, J. P. (2001). Signal transduction in resistance to plant viruses. *European Journal of Plant Pathology*, 107(1), 121–128. <https://doi.org/10.1023/A:1008732123834>
- Muthulakshmi, S., & Lingakumar, K. (2017). Role of salicylic acid (SA) in plants – A review. *International Journal of Applied Research*, 3(3), 33–37. Retrieved from <http://www.allresearchjournal.com/archives/2017/vol3issue3/PartA/3-2-94-381.pdf>
- Pangestuti, R., & Sulistyaningsih, E. (2011). Potensi Penggunaan True Seed Shallot (TSS) Sebagai Sumber Benih Bawang Merah Di Indonesia. *Semiloka Nasional “Dukungan Agro-Inovasi Untuk Pemberdayaan Petani” Kerjasama UNDIP, BPTP Jateng Dan Pemprov Jawa Tengah*, (August 2011), 258–266. Retrieved from <https://www.researchgate.net/publication/308120605%0APOTENS I>
- Saidah, Muchtar, Syafruddin, & Pangestuti, R. (2019). Pertumbuhan dan hasil panen dua varietas tanaman bawang merah asal biji di Kabupaten Sigi, Sulawesi Tengah. *Pros Sem Nas Masy Biodiv Indon*, 5(1), 213–216. <https://doi.org/10.13057/psnmbi/m050212>
- Shimeles, A. (2014). The performance of true seed shallot lines under different environments of Ethiopia. *Journal of Agricultural Sciences*, 59(2), 129–139. <https://doi.org/10.2298/jas1402129s>
- Sinta, A. (2018). *Aplikasi Bahan Organik dan Trichoderma sp. Untuk Menekan Perkembangan Penyakit Moler pada Bawang Merah* (Gadjah Mada University). Retrieved from [http://etd.repository.ugm.ac.id/index.php?act=view&buku\\_id=165091&mod=penelitian\\_detail&sub=PenelitianDetail&typ=html](http://etd.repository.ugm.ac.id/index.php?act=view&buku_id=165091&mod=penelitian_detail&sub=PenelitianDetail&typ=html)
- Soemirat, J., & Ariesyadi, H. D. (2017). *Toksikologi Lingkungan* (Vol. 00). Retrieved from <http://ugmpress.ugm.ac.id/id/product/lingkungan/toksikologi-lingkungan>
- Sumarni, N., Sopha, G. A., & Gaswanto, R. (2016). Respons Tanaman Bawang Merah Asal Biji True Shallot Seeds terhadap Kerapatan Tanaman pada Musim Hujan. *Jurnal Hortikultura*, 22(1), 23. <https://doi.org/10.21082/jhort.v22n1.2012.p23-28>
- Suwandi, Sopha, G. A., & Hermanto, C. (2016). *Petunjuk Teknis (Juknis) Proliga Bawang Merah 40 T/Ha Asal TSS (= True Shallot Seed)*. Balai Penelitian Tanaman Sayuran Puslitbang Hortikultura, Badan Litbang Pertanian. Retrieved from <http://sumbar.litbang.pertanian.go.id/images/pdf/JUKNIS-PROLIGA.pdf>
- Warrier, R. R., Paul, M., & Vineetha, M. V. (2013). Estimation of salicylic acid in Eucalyptus leaves using spectrophotometric methods. *Genetics and Plant Physiology*, 3(1–2), 90–97. Retrieved from [https://www.researchgate.net/publication/279819825\\_Estimation\\_of\\_salicylic\\_acid\\_in\\_Eucalyptus\\_leaves\\_using\\_spectrophotometric\\_methods](https://www.researchgate.net/publication/279819825_Estimation_of_salicylic_acid_in_Eucalyptus_leaves_using_spectrophotometric_methods)
- Wiyatiningsih, S. (2011). *Populasi Fusarium oxysporum f.sp. cepae, Intensitas Penyakit Moler, dan Hasil Umbi Bawang Merah di Tiga Daerah Sentra Produksi*. Retrieved from <http://eprints.upnjatim.ac.id/id/eprint/3249>
- Yusidah, I., & Istifadah, N. (2018). The abilities of spent mushroom substrate to suppress basal rot disease (*Fusarium oxysporum f.sp cepae*) in shallot. *International Journal of Biosciences*, 13(1), 440–448. <http://dx.doi.org/10.12692/ijb/13.1.440-448>
- Zakariyya, F. (2016). Menimbang indeks luas daun sebagai variabel penting pertumbuhan tanaman kakao. *Warta*, (3), 8–12. Retrieved from <https://docplayer.info/54696186-Menimbang-indeks-luas-daun-sebagai-variabel-penting-pertumbuhan-tanaman-kakao-fakhrusy-zakariyya-1.html>