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Research Article

Vegetative Performance of a Crossed-Shallot (*Allium fistulosum* L. x *Allium cepa* L. var. *aggregatum*) Applied with Dark Septate Endophyte

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ABSTRACT

Dark septate endophyte (DSE) application on shallot is still quite uncommon. This study investigated DSE application on crossed-shallot (*Allium fistulosum* L. x *Allium cepa* L. var. *aggregatum*) in the highlands of Lembang, West Java, Indonesia. Four treatments (*Dendrothyrium* sp. strain CPP 1.1.44, *Curvularia* sp. strain TKC 22, and *Cladosporium* sp. strain KSP.1 and control) with four replications on crossed shallot were investigated. Fresh weight, number of bulbs, number of pseudo-stems, number of leaves, plant height, pseudo-stem diameter, and leaf diameter were measured at 5, 7, 9, 11, and 13 weeks after planting (WAP). All treatments were freshly picked and weighed at 16 WAP. The results showed that crossed-shallots have different responses among treatments. However, all treatments showed the same response for maximum growth time. The highest values for the number of pseudo-stems, number of leaves, plant height, pseudo-stem diameter, and leaves diameter were reached at 13, 11, 9, 9, and 9 WAP, respectively. All treatments were harvested at 16 WAP, 3–8 weeks longer than shallot parental plants (*A. cepa* L. var. *aggregatum*). Applying three DSEs was likely to reduce the number of pseudo-stems from crossing lines of shallots and bunching onions. DSE KSP.1 treatment demonstrated an increase in plant height and the number of bulbs. Meanwhile, TKC 22 treatment showed to increase plant height, pseudo-stem, leaf diameter, and fresh weight.

Keywords: dark septate endophyte, shallot, harvest time, pseudo-stem

Introduction

Shallot (*Allium cepa* L. var. *aggregatum*) and bunching onion (*Allium fistulosum* L.) are two of the five crucial edible species in *Allium* (van der Meer, 1997). Both are important because of their extensive and diverse distribution and use. They are consumed as fresh ingredients in cooking, i.e., fried (Song et al., 2023), powdered (Dewayani et al., 2019), or pasted (Ndruru & Herawati, 2021). Shallot is rich in flavonoids and anthocyanins (Shigyo et al., 1997), whereas onion bunching has many carotenoids (Kopsell et al., 2010) and saponins (Zolfaghari et al., 2016). Because of the content of antioxidants and antimicrobial activities (Zolfaghari et al., 2020), bunching onions have many functions, including anti-inflammatory (Wang et al., 2013), antiobesity (Sung et al., 2018), antiallergen (Jippo et al., 2022), increasing body immunity (Hirayama et al., 2019), anti-influenza (Lee et al., 2012), and antitumor (Țigu et al., 2021). They can even be used as bioethanol (Robati, 2013) or phytoremediation (Alikasturi et al., 2020), likewise with shallots (Major et al., 2022).

The extensive number of diseases that attack shallots and bunching onions causes the search for resistance genes to biotic stress to be increasingly prioritized. These genes can be collected through interspecies crosses, such as between *A. fistulosum* L. and *A. cepa* L. var. *aggregatum*. Crosses between these two species were carried out for the first time in 1931 and continued with researches related to hybrids (Chuda & Adamus, 2009), diversity of bulb forms of offspring (Yaguchiet et al., 2009), QTL various traits (Tsukazaki et al., 2017), formation of cytoplasmic male sterile (CMS) (Liu et al., 2019), biotic resistance of offspring (Kudryavtseva et al., 2019), chromosome maps (Tsukazaki et al., 2008), mitochondrial genome (Xing et al., 2023) to the formation of AlliumMap (McCallum et al., 2012). Although crosses between the two species have also been carried out in Indonesia, publications related to the cross results are rarely found (Saadah et al., 2023).

Biostimulants like dark septate endophyte (DSE) can induce plant resistance and performance. DSE can suppress *Fusarium* disease (Surono & Narisawa, 2018), white root rot caused by *Rigidoporus microporus* in rubber plants (Dalimunthe et al., 2023), blast disease caused by *Pyricularia oryzae* (*Po*) in rice (Yuliani et al., 2020), basal stem rot caused by *Ganoderma boninense* in oil palm, and several other diseases in shallots (Krestini et al., 2023). DSE can also improve plant performance with various mechanisms (Azmi et al., 2022; Surono & Narisawa, 2017). DSE can even serve as a phytoremediation (Melati et al., 2023).

The use of DSE is still infrequent on horticultural crops, particularly on the crossed line between *A. fistulosum* and *A. cepa* var. *aggregatum*. Therefore, this study aimed to evaluate the application of DSE on crossed shallots (*A. fistulosum* x *A. cepa* var. *aggregatum*).

Materials and Methods

The research was conducted in May–September 2022 in the highlands of Lembang (1,250 m asl), West Bandung, West Java, Indonesia. The materials used in this research consisted of one accession of crossed shallots (*A. fistulosum* L. x *A. cepa* L. var. *aggregatum*), manure, NPK fertilizer, KCl fertilizer, SP-36, pesticides, and three types of DSE: P1 (*Dendrothyrium* sp. strain CPP 1.1.44) (Tawfeeq Al-Ani et al., 2021), P2 (*Curvularia* sp. strain TKC 22) (Yuliani et al., 2020), and P3 (*Cladosporium* sp. strain KSP.1) (Tawfeeq Al-Ani et al., 2021), which were the collections of Indonesian Soil Research Institute, Bogor, West Java. P1, P2, P3, and control P0 (without DSE treatment) were used as treatment, with four replications.

All the materials were planted with spacing of 20 cm × 15 cm. Tillage was carried out 2 weeks before planting, making beds, and applying manure and SP-36. NPK fertilizer was applied 2–3 times at 2, 4, and 6 weeks after planting (WAP). DSE application was carried out at 3 WAP by dripping as much as 1 ml at the growing point. Plant pests and diseases were intensively controlled during the research.

Growth parameters were observed at 5, 7, 9, 11, and 13 WAP, including number of pseudo-stems, number of leaves, plant height, pseudo-stem diameter, and leaf diameter. Harvest was carried out at 16 WAP, and fresh weight and number of bulbs were recorded. The data collected was tabulated and analyzed for variance using the SPSS program and further tested using Duncan test if significant differences existed in the treatments.

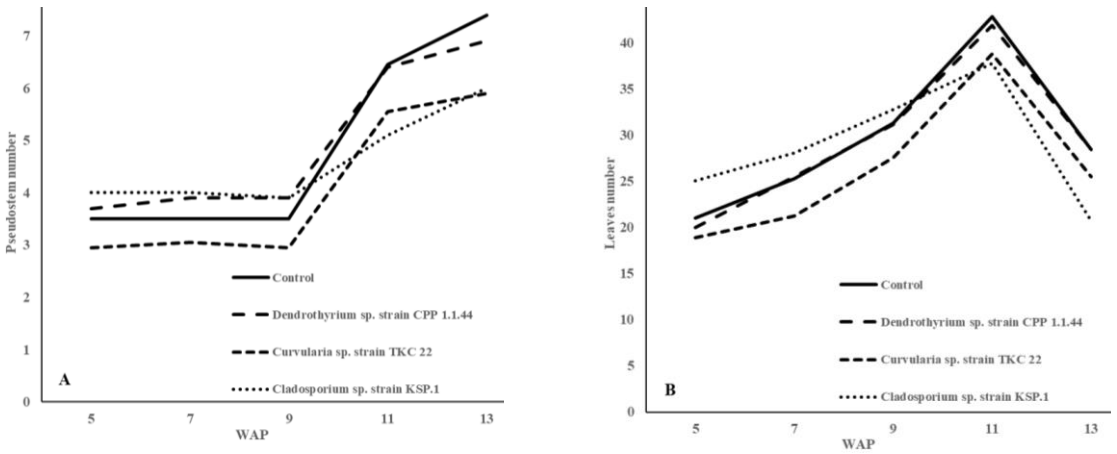

Fig. 1. Number of pseudo-stem (A) and leaves (B) of crossed shallot plants treated with control, *Dendrothyrium* sp. strain CPP 1.1.44, *Curvularia* sp. strain TKC 22, and *Cladosporium* sp. strain KSP.1

Results and Discussion

The analysis of variance showed that all treatments have no significant effect on number of pseudo-stem numbers, number of leaves, plant height, pseudo-stem diameter, leaves diameter, fresh weight, and number of bulbs. The pseudo-stems number of all treatments in this study reached a maximum at 13 WAP in all treatments, ranging from 6.0 to 7.4. The number of pseudo-stems treated with P1 and P3 tended to be higher than that treated with P0 from the start of observation (5 WAP) to the beginning of 9 WAP. However, the number of pseudo-stems P0 treatments rose higher, up to 13 MST (12.47), compared to other treatments (Figure 1A). Application of three DSEs reduced the number of pseudo-stems of crossing line (between bunching onions and shallots).

Leaves numbers of all samples in this study increased to a maximum of 11 WAP with a range of 37.75–42.80 and sloped to the end of the observation (13 WAP). The number of P3 leaves went up higher than all other samples to 9 WAP. Furthermore, up to 11 WAP, the control had the highest number of leaves (42.80) compared to other treatments (Figure 1B).

Plant height of all samples in this study rised to a maximum at 9 WAP with a range of 72.95–78.20 cm and sloped to the end of the observation. The plant height of P2 and P3 tended to be higher than P0 and P1 from the beginning to the end of the observation (Figure 2A). DSEs *Curvularia* sp. strain TKC 22 (P2) and *Cladosporium sp.* strain KSP.1 (P3) demonstrated to increase the plant height of crossed-shallot plants. This is in line with the results of previous studies which stated that DSEs *Curvularia* sp. strain TKC 22 and *Cladosporium sp.* strain KSP.1 could increase the height of rice plants (Tawfeeq Al-Ani et al., 2021; Yuliani et al., 2020). In another case, applying *Cladosporium* sp. on tomatoes could increase plant height, number of leaves, and length of roots of tomato plants (Răut et al., 2021). The increase in growth was due to the ability of DSE *Curvularia* sp. to dissolve aluminum phosphate (Spagnoletti et al., 2017), and iron and IAA contents of *Cladosporium* sp. (Răut et al., 2021).

Pseudo-stem diameter of all treatments tested tended to rise to a maximum at 9 WAP with a range of 19.55–22.17 mm and sloped to the end of the observation. P2 pseudo-stem graphs tended to be higher than all other treatments until the end of the observation (Figure 2B). It was noticed that DSE *Curvularia* sp. strain TKC 22 (P2) increased the diameter of the pseudo-stem of the crossed-shallot plants. Other studies showed that variety, temperature, day length, and interaction between temperature and day length influenced garlic pseudo-stems diameter (Atif et al., 2019). The pseudo-stem diameter of bunching onion positively correlated with leaf length, leaf diameter, pseudo-stem length, plant height, weight per tiller, and number of leaves (Azmi & Kirana, 2011).

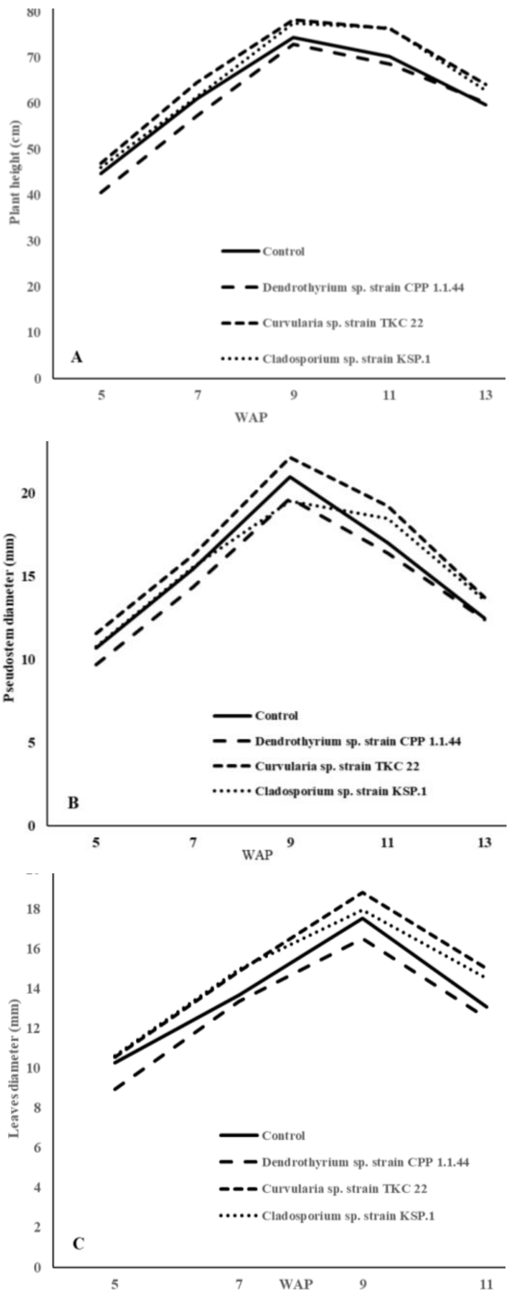

Fig. 2. Plant height (A), pseudo-stem diameter (B), and leaves diameter (C) of crossed shallot plants treated with control, *Dendrothyrium* sp. strain CPP 1.1.44, *Curvularia* sp. strain TKC 22, and *Cladosporium* sp. strain KSP.1

Leaf diameter of all samples tended to rise to a maximum of 9 WAP with a range of 16.51–18.83 mm and sloped to the end of the observation (11 WAP). The P2 leaf diameter graph tended to be higher than all treatments until the end of the observation (11 WAP) (Figure 2C). There was an indication that DSE *Curvularia* sp. strain TKC 22 (P2) increased the leaf diameter of the crossed-shallot plants. Until now, no reports have been found regarding the use of DSE in crossbreeding materials between bunching onions and shallots. However, reports have been found regarding the use of other microorganisms/microbes in bunching onion, specifically bunching onions inoculated with mycorrhizal fungi (*Glomus* sp.), which exhibited a larger shooth length and leaf sheath diameter compared to non-inoculated plants (Tawaraya et al., 2012).

In this study, the crossed-shallot plants were harvested at 16 WAP, 2.5 months longer than bunching onions and shallots planted in highland (8–13 WAP, depending on variety) (Saadah et al., 2023; Saputri & Jonni, 2020; Waluyo et al., 2021). It is likely that the crossed-shallot plants have a longer life than common shallots.

The fresh weight of P0 (215.15 g) was the highest compared to other treatments. This is contrary to the results of research on rice, *Curvularia* sp. strain TKC 22 (P2) could increase rice plant wet and dry weight (Yuliani et al., 2020). The highest number of bulbs was shown by the P3 treatment (5 bulbs), compared to the P0, P1, and P2 treatments (4, 4, and 3, respectively) (Table 1). Despite having the lowest weight, P3 produced the highest number of bulbs compared to other treatments. These crossed-shallot plants can be harvested as a bunching onion when the leaves are still green or harvested bulbs when it is used as seed (propagation material). For propagation, many bulbs take precedence over the weight of the harvest because the greater the number of pseudo-stems, the more planting material they have. The characteristics of crossed-shallot plants follow the genetic character of annual bunching onion more than shallots, which are annual plants (Kołota et al., 2012).

Table 1. Fresh weight and number of bulbs of crossed-shallot plants treated with three types of dark septate endophyte.

Treatment	Fresh weight (g)	Number of bulbs
P0	215.15	4
P1	197.85	4
P2	190.35	3
P3	171.75	5
HSD 5%	-	-

P0 = control (without DSE treatment), P1 = *Dendrothyrium* sp. strain CPP 1.1.44, P2 = *Curvularia* sp. strain TKC 22, and P3 = *Cladosporium* sp. strain KSP.1.

Conversely, for bunching onions, a high fresh harvest weight is more important than the number of pseudo-stems/bulbs for consumption because the crop is sold fresh based on the harvest weight. The data shows that if it is sold as a bunching onion, crossed-shallot plants should not be treated with DSE. Conversely, if it is used as the following propagation material, *Cladosporium* sp. strain KSP.1 (P3) can be recommended because it has been demonstrated to increase the number of bulbs.

Endophytes can actively or passively trigger plant growth through various mechanisms. Endophytes provide a variety of metabolites that improve host plant fitness by increasing plant resistance to biotic and abiotic stress and promoting plant growth (Sudha et al., 2016). Plant growth increases because endophytes help access elements C, N, and P from the rhizosphere, sending them to the host plant so that they are easily absorbed and used by the plant (He et al., 2019; Vergara et al., 2018). The P content in bunching onions increased compared to control with endophytic administration (*Glomus* sp.) (Tawaraya et al., 2012). In sorghum, *Curvularia* sp. increased aerial biomass and radicle length (Barresi et al., 2022).

Conclusion

Crossed shallot plants showed different responses among different DSE treatments. However, all treatments showed the same response for maximum growth time. The highest values for the number of pseudo-stems, number of leaves, plant height, pseudo-stem diameter, and leaf diameter were reached at 13, 11, 9, 9, and 9 WAP, respectively. At 16 WAP, all treatments were harvested, which was 3–8 weeks later than the harvest age of common shallot plants (*A. cepa* var. *aggregatum*). Applying three types of DSEs reduced the number of pseudo-stems from crossing line of shallots and bunching onions. Treatment of DSE KSP.1 could increase plant height and the number of bulbs. Meanwhile, treatment of TKC 22 could increase plant height, pseudo-stem, leaf diameter, and fresh weight.

Conflict of Interest

We confirm that we have no conflict of interest regarding any financial, personal, or other affiliations with individuals or organizations related to the subject matter discussed in the manuscript.

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