AGRIVITA
Journal of Agricultural Science
www.agrivita.ub.ac.id

Synergistic Action of PGPR and Biofungicide with Active Ingredient of *Cladosporium clasporioides* to Control White Rust on Chrysanthemum

Wakiah Nuryani[1*], Hanudin[1], Indijarto Budi Rahardjo[1] and Kurniawan Budiarto[2]

[1] Indonesian Ornamental Crops Research Institute, Cianjur, West Java, Indonesia
[2] Indonesian Citrus and Subtropical Fruits Research Institute, Batu, East Java, Indonesia

ARTICLE INFO

Keywords:
Antagonist microbes
Chrysanthemum
Consortium
White rust

Article History:
Received: October 7, 2021
Accepted: November 17, 2021

*) Corresponding author:
E-mail: wnuryani@yahoo.com

ABSTRACT

The application of antagonist microbes to control significant diseases on crops is an essential issue in the eco-friendly and sustainable agriculture of the chrysanthemum production system. The application of antagonist consortiums is expected to synergistically suppress the pathogen more effectively than a single microbe, thus increasing the marketable flower yield. The research is carried out to evaluate the single and combined application of antagonists *C. ladosporioides* and PGPRs, *B. subtilis*, and *P. flourescens* to control white rust in Chrysanthemum. The results show that there is no cumulative effect from the combination of biofungicide and the PGPR on disease suppression, disease incidence, and plant growth improvement than single antagonist treatments. Compared to synthetic fungicide, biofungicide and the PGPR treatments give higher parasitism intensity, though the values were negligible among the treatments. The lowest disease intensity is recorded from synthetic fungicide treatment. The improvement of flower quality due to biofungicide, PGPR, and synthetic fungicide treatments was found only on the longer vase life than untreated plants. Observation on the compatibility of antagonists with PGPRs are still needed to increase the effectiveness in controlling white rust in Chrysanthemum.

INTRODUCTION

Chrysanthemum (*Dendranthema grandiflora Tzvelev syn. Chrysanthemum morifolium* [Ramat.] Kitam) is one of the most commercially cultivated cut flowers worldwide, including in Indonesia. In the country, the production centers are located in Berastagi (North Sumatera), Solok (West Sumatera), Cipanas and Lembang (West Java), Ungaran (Central Java), Sleman and Kulonprogo (DI Yogyakarta), Batu and Pasuruan (East Java), Tomohon (North Sulawesi) and Malino (South Sulawesi) (Hayati et al., 2019). Chrysanthemum ranks as the most marketed cut flower in the domestic market, in line with the production increase in the last five years. The production was recorded 427.5 million stalks in 2014 and increased to 488.2 million stalks in 2018 following the increment of harvesting and planting area (Helmiatin & Susanty, 2019).

However, these positive trends have been still constrained by the poor physical quality of the flowers, especially for the traditional farmers. The pest and pathogen attacks, especially white rust, still become one of the main problems in improving cut flower productivity and quality (Nuryani et al., 2018) The disease is caused by the pathogenic fungus *Puccinia horiana* P. Henn (Basidiomycetes), limitedly hosted in 12 species, including Chrystanthemum and Nipponanthemum and Leucanthemella (O'Keefe & Davis, 2015; Zeng et al., 2013). The pathogen might infect the plant in most growth stages from the rooting process, young plants, and the flowering stage. The disease intensity and severity are depended on the plant genotype and environmental condition (Marwoto, 2012)). The high disease incidence is usually related to warm temperature, high humidity and occurs in susceptible genotypes (De Backer et al.,

ISSN: 0126-0537 Accredited First Grade by Ministry of Research, Technology and Higher Education of The Republic of Indonesia, Decree No: 30/E/KPT/2018

Cite this as: Nuryani, W., Hanudin., Rahardjo, I. B., & Budiarto, K. (2022). Synergistic action of PGPR and biofungicide with active ingredient of *Cladosporium clasporioides* to control white rust on chrysanthemum. *AGRIVITA Journal of Agricultural Science*, *44*(1), 31–39. http://doi.org/10.17503/agrivita.v44i1.3379

2011; Hanudin et al., 2017; Wang et al., 2020). In severe attacks, the production loss might reach 80-100% without marketable flowers (Yusuf & Suhardi, 2016)

The use of synthetic chemicals still becomes the leading grower choice in handling the pathogen. However, in several countries, including Indonesia, there has no registered synthetic pesticide to handle specifically for white rust in Chrysanthemum (Yusuf et al., 2014). To cope with these situations, growers then tend to use various kinds of fungicide with inappropriate dosages, expecting the reduction of damages. The chemicals employ even in the absence of the symptoms and intensity of the diseases to assure the marketable flower quality. In long and frequent applications, these costly practices increased production cost, induced pathogen resistance, and made the chemicals no longer effective (Mahish & Ghritlahare, 2017; Rahardjo et al., 2019; Singh & Vijay, 2011).

Efforts have been made to make the chrysanthemum production system more efficient and profitable. The use of natural enemies like mycoparasite is considered an alternative way to minimize the economic losses due to white rust and reduce the use of chemicals. Cladosporium Link is one of the most common genera of fungi that might compete to control that nutrient source or other environmental factors. It can also exhibit saprophytic fungi to exploit dead substrates or even enable to act as pathogenic fungi to penetrate their fungal hosts (Herrera et al., 2016; Traquair et al., 1984). On *Puccinia horiana* only *Cladosporium uredinicola*, *C. sphaerospermum* and *Cladosporium* sp. have been reported that inhibited the disease develop (Silber et al., 2014; Torres et al., 2017). The parasitic mechanism of Cladosporium was through the ability of Cladosporium hypha to envelop the teliospore of Puccinia and resulted in the morphological malformation and dysfunction of the spores (Torres et al., 2017).

The effectiveness of antagonists in suppressing the disease increases when they combine consortium with other synergistic antagonists (Köhl et al., 2019). Several reports indicated the success of using microbe consortium, such as *Bacillus megaterium* and *Burkholderia cepacia* to control root rot caused by *Fusarium oxysporum f.sp. radicis-lycopersici* in tomato (Omar et al., 2006), a combination of *B. firmus* E65 and *P. aeruginosa* C32b to control bacterial blight disease in rice (Suryadi et al., 2013), and combination of *P. flourescens* and *B. subtilis* to control leaf blight in coconut (Johnson et al., 2017). Screening on *C. cladosporioides* isolates has been carried out and revealed several promising potentials with a degree of suppression to white rust in an average of 35.8% (Amirmijani et al., 2014). On the other hand, Lengai & Muthomi (2018) also reported that several PGPRs, such as *P. flourenscens* and *B. subtilis* isolates, have been evaluated and selected to reduce white rust attacks in chrysanthemums. The combined application of these antagonist microbes is expected to merely increase the effectivity of the antagonists to suppress the disease development and intensity compared to a single application. The research is conducted to evaluate the combination of *C. cladosporioides* and PGPRs in controlling white rust disease in Chrysanthemum.

MATERIALS AND METHODS

The research was conducted from January to December 2019, covering laboratory and greenhouse works. The experiments were carried out at the Indonesian Ornamental Crops Research Institute (IOCRI) with an altitude of 1100 masl. The chrysanthemum variety used was Puspita Nusantara which was known as a susceptible variety to white rust. The selected Cladosporium and PGPR isolates gave the highest suppression to white rust based on Yusuf et al.(2019) and Hanudin et al. (2017), respectively. The selected biofungicide with active ingredient Cladosporium and PGPR isolates has been tested in vitro for compatibility. The combination of Cladosporium and PGPR treatments for in vivo evaluation was presented in Table 1.

Soil Preparation, Planting, and Plant Maintenance

The area inside the plastic houses was hoed to clean the planting sites from weeds and other substances. Then, the soil was mixed adequately with 30 t/ha manure and 300 kg/ha NPK (15:15:15) fertilizers. The planting sites were then organized into 1 x 1.5 m beds, and the distance among beds was 60 cm. The planting beds were then poured with water to facilitate humidity before planting. Rooted cuttings were planted in beds with a density of 100 plants/m^2. All the plants were maintained in long-day conditions using 100 lux artificial lighting for 4 h every night until 30 days after planting. The plants were sprayed using Abamectin (Syngenta Co.

Ltd, Indonesia) once a week at the recommended dosage for insect pest prevention. Additional fertilizers were supplemented via top dressing using NPK (16:16:16) with the rate of 200, 300 dan 100 kg/ha 21, 42 dan 63 days after planting (DAP). Flower harvest was carried out when at least 70% of the total flowers on each bed were fully opened.

Treatment Application, Data Gathering, and Data Analysis

The suspension of biofungicide, PGPR, and combination of biofungicide and PGPR were made following (Hanudin et al., 2017). The experiment was arranged in randomized complete block design with three replicates. The biofungicide treatments were applied at 14 DAP at noontime (05.00 pm) by spraying the plants with suspension treatments using a semi-automated hand sprayer. The volume of suspension was arranged at 1000 ml per bed. The volume was increased to 1.500 ml per bed at 48 DAP. The treatment was applied weekly until 83 DAP.

The experimental parameters included plant growth and lower quality, disease intensity, pustule number, disease development, the intensity of parasitism. Disease intensity was determined based on 10% plant samples of each bed and carried out at 21, 35, 49, 56, 63 dan 77 DAP. The scale and damage criteria for determining disease intensity were following Yusuf & Suhardi (2013) as presented in Table 2.

The disease intensity was calculated using the formula 1.

$$I = \frac{\Sigma(v \times n)}{(Z \times N)} 100\% \qquad 1)$$

Where : I = Intensity of white rust (%); v = Scale of the observed damage; n = number of infected plants categorized in the respected damage scale; Z = highest scale of the observed damage

Table 1. Combination of Cladosporium and PGPR treatments to control white rust in Chrysanthemum under plastic house condition.

Treatments code(s)	Description	Concentration (g/ml/l)
CT	Biofungicide with an active ingredient of *Cladosporium* sp applied solely	2 g
CB	Biofungicide with an active ingredient of *Cladosporium* sp applied in combination with *B. subtilis*	2 g
CP	Biofungicide with an active ingredient of *Cladosporium* sp applied in combination with *P. florescens* (Pf)	2 g
CBP	Biofungicide with an active ingredient of *Cladosporium* sp applied in combination with *B. subtilis* and *P. florescens* (Pf)	2 g
BT	Biofungicide with an active ingredient of *B. subtilis* applied solely	10 ml
PT	Biofungicide with an active ingredient of *P. florescens* (Pf) applied solely	10 ml
FSP	Synthetic fungicide with an active ingredient of Pyraclostrobin 250 EC	1 g
K	Water (control)	

Table 2. Scale and damage criteria of white rust (*Puccinia horiana* Henn) infection on Chrysanthemum.

Scale	Damage Criteria
0	Not infected (symptomless)
1	Very low, infection detected only on lower plant leaves, and the intensity not exceed 5% of the total leaf area.
2	Low, infection detected on lower plant leaves, and the intensity ranges 5-10% from the total leaf area.
3	Medium damage, infection detected on middle and lower plant leaves, and the intensity ranges 10-20% from total leaf area.
4	Heavy damage, infection detected on upper, middle and lower plant leaves and the intensity ranges 20-40% from total leaf area.
5	Very heavy damage, infection detected on upper, middle and lower plant leaves and the intensity was more 40% from total leaf area.

Several pustules were observed at 21, 42, 63, and 84 DAP on the 5 leaves at the middle of the stem. A number of pustules represented the dynamic disease development. The intensity of parasitism was observed on 10% plant samples per bed. Five leaves per plant were randomly selected, and the number of parasitized pustules was observed. The parasitized pustules were characterized by the existence of growing whitish-grey Cladosporium hypha covering the pustule dome. The observation was carried out at 21, 35, 49, 63, 77, and 91 DAP. In every observation, the leaves from different plants were selected for the basis of measurement. Intensity of parasitism was calculated using formula 2.

$$P = \frac{a}{b} \times 100\% \quad \text{.......... 2)}$$

Where : P = intensity of parasitism (%); a = number of parasitized pustules; b = number of obsevable pustules

Flower quality was determined through flower diameter, flower thickness, vase life, and flower stalk length. The grade criteria of Chrysanthemum cut flower from the basis of stalk length were categorized as: A (stalk length > 80cm), B (60-80 cm), C (40-60 cm), and D (stalk length < 40 cm). All the gathered data were analyzed using ANOVA, and mean comparisons were carried out based on LSD (α ≤ 5%).

RESULTS AND DISCUSSION

Disease Intensity

The intensity of white rust on chrysanthemum plants was varied among the biofungicide, PGPR, biofungicide-PGPR combinations, and synthetic fungicide treatments at 21 DAP (Table 3). The lowest disease intensity was observed at combined Cladosporium + Pf, and the highest was at single Pf. The disease intensity increased at 35 and 49 DAP in almost all treatments. During this period, the highest disease intensity was observed at the control (water) treatment, and the value showed negligible differences with the rest treatments. The disease intensity on the biofungicide Cladosporium treatments on both single and combination with PGPR was relatively less varied at 49, 63, 77 up to 91 DAP. The lower and stable disease intensity during 63 to 91 DAP was detected at synthetic fungicide treatment.

In terms of disease suppression compared to control, the treatment of synthetic fungicide showed the most effectiveness (Table 3). While among the biofungicide and PGPR treatments, the degree of suppression was varied. The expected better disease suppression on the combination of biofungicide Cladosporium + PGPR was not detected. Among the single and combination treatments of biofungicide Cladosporium and PGPR, the highest suppression on white rust was detected on the single biofungicide Cladosporium and PGPR treatments. This condition indicated that higher synergistic effects of Cladosporium and PGPR were less detected when applied in combination. A similar situation was also reported by Prayitno (2017) when applying bacterial consortium on the bioremediation of oil-polluted soil.

The Average Number of Pustules and Disease Development

The dynamic disease development represented by the number of pustules was also varied among the treatments in every 14 days observation. In general, the number of pustules increased from 21 to 42 DAP (Table 4). The increment continued up to 63 DAP in most all treatments with different rates. The highest increment was detected on the treatments of single biofungicide Cladosporium (CT), and the value was even higher than the control. The decrease rate in pustule formation was found only on the treatment of synthetic fungicide (FSP). Up to 84 DAP, the rate of pustule formation decreased in all treatments.

Based on the dynamic disease development, the treatments of biofungicide Cladosporium, PGPR, and combination of biofungicide Cladosporium + PGPR were less effective in suppressing disease development during 21 to 63 DAP compared to synthetic fungicide, which had the lowest pustule formation rates. These conditions inferred that the applied antagonists were not able to inhibit the development pathogenic fungus *P. horiana*. Aside from the growing period needed, the capability of the antagonists in supressing the disease was determined by the adaptation of the antagonists on the targeted plant parts. When the antagonists could not adapt to the targeted sites, the antagonists could not utilize the source of environmental factors to grow optimally to parasitize the pathogenic fungus (Heydari & Pessarakli, 2010).

Table 3. White rust intensity on chrysanthemum cv. Puspita Nusantara under biofungicide Cladosporium, PGPR, a combination of Cladosporium + PGPR and synthetic fungicide treatments.

Treatment code(s)	White rust intensity (%)*) at the growing period of Chrysanthemum (DAP)						Disease suppression (%) compared to control
	21	35	49	63	77	91	
CT	13.0 abc	33.5 b	34.0	37 ab	40.5 ab	50 b	23.0
CB	9.5 bc	35 ab	37.5	36.5 ab	40.5 ab	53 b	18.0
CP	6.0 c	35 ab	35.5	35.5 ab	38.5 ab	51 b	21.5
CBP	13.5 abc	39.5 ab	31.0	33 ab	36 b	50 b	23.0
BT	11.5 abc	38.5 ab	35.0	41 ab	44.5 ab	54 b	13.8
PT	19.0 a	40 ab	38.0	33 ab	41 ab	49.5 b	23.8
FSP	15.0 ab	36.5 ab	28.5	20 c	21 c	31 c	52.0
K	13.0 abc	45 a	40.0	45 a	51 a	65 a	-
CV (%)	22.21	16.79	25.88	21.97	19.95	14.25	

Remarks: *) Values in the same column followed by different letters differ significantly under LSD (α ≤ 5%).

Table 4. Dynamic white rust disease development on chrysanthemum cv. Puspita Nusantara under biofungicide *Cladosporium*, PGPR, a combination of *Cladosporium* + PGPR and synthetic fungicide treatments

Treatment code(s)	Number of newly developed pustules after...DAP*)			
	21	42	63	84
CT	0.47 a	3.15 a	14.02 a	4.22 a
CB	0.42 a	2.25 a	5.85 ab	3.87 ab
CP	0.3 a	5.55 a	7.12 ab	4.25 a
CBP	0.6 a	1.70 a	7.12 ab	4.27 a
BT	0.35 a	2.22 a	7.77 ab	4.27 a
PT	0.4 a	6.62 a	7.12 ab	3.05 ab
FSP	0.57 a	4.27 a	3.62 b	1.47 b
K	0.85 a	2.80 a	6.67 ab	4.92 a
CV (%)	32.18	24.7	37.93	22.79

Remarks: *) Values in the same column followed by different letters differ significantly under LSD (α ≤ 5%).

Intensity of Parasitism

The capability of antagonists *Cladosporium* sp. and PGPR (*B. subtilis* and *P. flourescens*) on suppressing white rust disease was represented in the intensity of parasitism as presented in Fig. 1. In general, the intensity of parasitism on each treatment was varied along with the plant growth period. During 21 to 35 DAP, the intensity of parasitisms in most treatments was decreased, except in a single application of biofungicide Cladosporium (CT) and *P. flourescens* (PT). The intensity of parasitisms increased at 49 DAP and then sharply decreased at 63 and 77 DAP in all treatments. In the harvesting period (91 DAP), the intensity of parasitism increased in all treatments with different rates.

In general, the single and combined applications of biofungicide Cladosporium and PGPR showed higher intensity of parasitism than synthetic fungicide at 49 DAP. These conditions inferred that though the synergistic effects of biofungicide and PGPR were less detected, the single parasitic effect of the antagonist was still naturally occurred. The phenomena were observed on the lower intensity of parasitism in the treatment of combined biofungicide and PGPR, yet higher than synthetic fungicide when applied in a single treatment up to 91 DAP (Fig. 1). Any author has never reported the combined application of *Cladosporium* sp. with *B.*

subtilis and *P. flourescens* in any crop so far. Thus, these findings are considered the first report related to these antagonists' compatibility characteristic when applied in combination to control white rust in chrysanthemums. Different effectivity of antagonists in suppressing the disease when applied in single and combination was also reported Rosyidah et al. (2013)) when applying *Trichoderma viride*, *Streptomyces* sp. and *P. fluorescens* ton control bacterial wilt *R. solanacearum* in potato.

Plant Growth and Flower Quality

Plant height, flower stalk lenght, flower diameter and vase life of chrysantemum cv. Puspita Nusantara treated by various treatments of biofungcide, PGPR and synthetic fungicide are presented in Table 5. In general, the treatment of biofungicide Cladosporum, PGPR, combined biofungicide Cladosporium + PGPR, and synthetic fungicide have given negligible effect on plant height, flower stalk length, and flower diameter, except on vase life. On vase life, the treatment of single Cladosporium (CT), a combination of biofungicide Cladosporium + *P. flourescens* (CP), and a combination of biofungicide Cladosporium + *B. subtilis* + *P. flourescens* (CBP) gave significant longer vase life than control.

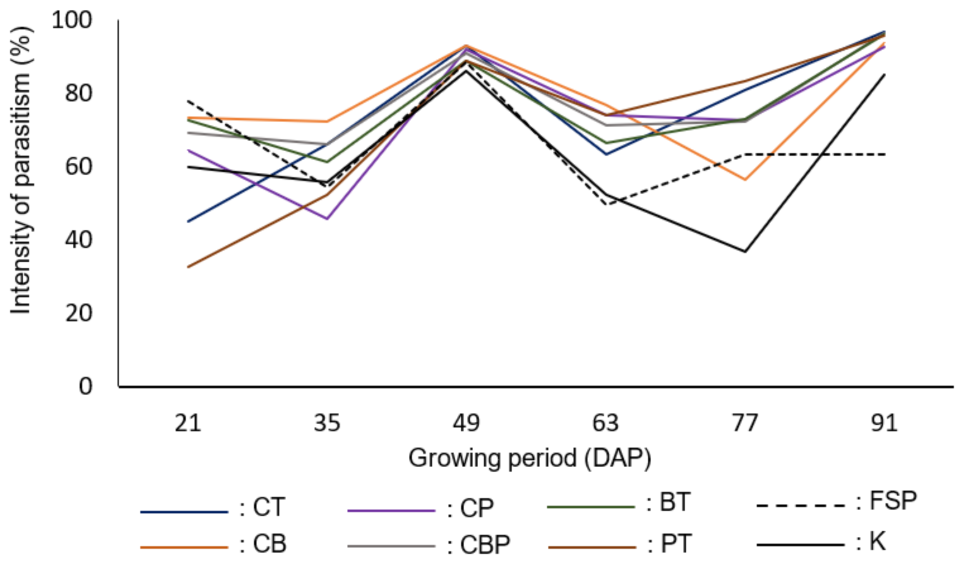

Fig. 1. Intensity of parasitism of biofungicide, PGPR, combination biofungicide + PGPR, and synthetic fungicide on white rust disease in Chrysanthemum.

Table 5. Plant growth and flower quality of chrysantyemum cv. Puspita Nusantara is treated by various biofungcide, PGPR, and synthetic fungicide treatments

Treatment code(s)	Plant height*) (cm)	Flower stalk lenght*) (cm)	Flower diameter*) (cm)	Vase life*) (days)
CT	81.65	86.28	6.02	9.5 a
CB	84.10	90.13	5.85	9.25 ab
CP	80.75	83.70	6.12	9.65 a
CBP	83.52	111.2	6.12	9.65 a
BT	81.20	86.05	6.77	9.05 ab
PT	81.42	85.75	6.12	9.4 ab
FSP	84.47	89.85	6.62	9.2 ab
K	78.45	86.65	6.67	8.85 b
CV (%)	5.54	18.92	6.44	4.15

Remarks: *) Values in the same column and different letters differ significantly under LSD (α ≤ 5%).

Compared to control, fewer variations on plant growth performance and flower quality on Chrysanthemum under different biofungicide, PGPR, and synthetic fungicide treatments indicated that the applied treatments gave minimum direct effect on the plant growth improvement. The expected chain effect of biofungicide and PGPR application to plant growth performance through the minimum white rust attacks was not confidently established. The effects of biofungicide and PGPR application on white rust intensity, disease development, and parasitism as shown in Table 3, Table 4 and Fig. 1, respectively, were insignificant compared to control. The trends were unstable until 91 DAP. With biofungicide and PGPR, the plant growth improvement was expectedly acheived from the less physical damage on the plant part, especially the leaves due to less pathogen attack and development (Harman et al., 2021)on, and within them. Some of these endophytically colonize plant roots. The colonization of roots by certain symbiotic strains of plant-associated bacteria and fungi results in these plants performing better than plants whose roots are colonized by only the wild populations of microbes. We consider here crop plants whose roots are inhabited by introduced organisms, referring to them as Enhanced Plant Holobionts (EPHs, thus increasing the physiological integrity of the plant to grow optimally. The plant growth improvement can also be derived from the symbiotic relationship between PGPR (*B. subtilis* and *P. flourescens*) and the host plant (Ranadev et al., 2019). While so far, antagonist *Cladosporium* sp. has been reported effective only in disease suppression, yet no evidence on its symbiotic and other direct action on plant growth promotion (Chaibub et al., 2020).

CONCLUSION

Single and combined application of biofungicide *Cladosporium* sp. and PGPR (*B. subtilis* and *P. flourescens*) give negligible effects on white rust attacks. The highest disease suppression of this treatment is 23.8% and lower than that of synthetic fungicide treatments that reached 52%. The application of synthetic fungicide also has lower pustule formation rates during the entire plant growth period. Only in the intensity of parasitism, the biofungicide and PGPR treatments give apparent effects than a synthetic fungicide. Biofungicide and PGPR treatments also have fewer effects on plant growth and flower quality improvement. Only the treatment of single bioactive fungicide of Cladosporium (CT), bioactive consortium of biofungicide Cladosporium + *P. flourescens* (CP), and a consortium of biofungicide Cladosporium + *B. subtilis* + *P. flourescens* (CBP) give longer vase life than control. Observation on antagonists' compatibility with PGPRs is still needed to increase the effectiveness in controlling white rust in Chrysanthemum.

ACKNOWLEDGEMENT

The authors would like to express their thank and high appreciation to the Director of The Indonesian Agency for Agricultural Research and Development (IAARD). Through the Center for Horticultural Research and Development (ICHORD), the Indonesian Ornamental Crops Research Institute (IOCRI) that financed, gave suggestions, criticisms in the planning and implementation of research. The authors also wish to thank the following personals; Mr. Muhidin, Ade Sulaiman, and all those who helped and worked during the research and report.

REFERENCES

Amirmijani, A., Khodaparast, S. A., & Zare, R. (2014). Contribution to the identification of *Cladosporium* species in the North of Iran. *Rostaniha*, *15*(2), 133–145. https://doi.org/10.22092/botany.2014.101237

Chaibub, A. A., de Sousa, T. P., de Oliveira, M. I. S., Arriel-Elias, M. T., de Araújo, L. G., & de Filippi, M. C. C. (2020). Efficacy of *Cladosporium cladosporioides* C24G as a multifunctional agent in upland rice in agroecological systems. *International Journal of Plant Production*, *14*(3), 463–474. https://doi.org/10.1007/s42106-020-00097-2

De Backer, M., Alaei, H., Van Bockstaele, E., Roldan-Ruiz, I., van der Lee, T., Maes, M., & Heungens, K. (2011). Identification and characterization of pathotypes in *Puccinia horiana*, a rust pathogen of Chrysanthemum x morifolium. *European Journal of Plant Pathology*, *130*(3), 325–338. https://doi.org/10.1007/s10658-011-9756-8

Hanudin, H., Budiarto, K., & Marwoto, B. (2017). Application of PGPR and Antagonist fungi-based biofungicide for white rust disease control and its economyc analysis in chrysanthemum production. *AGRIVITA Journal of Agricultural Science*, *39*(3), 266–278. https://doi.org/10.17503/agrivita.v39i3.1326

Harman, G., Khadka, R., Doni, F., & Uphoff, N. (2021). Benefits to plant health and productivity from enhancing plant microbial symbionts. *Frontiers in Plant Science*, *11*, 610065. https://doi.org/10.3389/fpls.2020.610065

Hayati, N. Q., Nurmalinda, N., & Marwoto, B. (2019). Inovasi teknologi tanaman krisan yang dibutuhkan pelaku usaha (Technology innovation of chrysanthemum needed by stakeholders). *Jurnal Hortikultura*, *28*(1), 147–162. https://doi.org/10.21082/jhort.v28n1.2018.p147-162

Helmiatin, H., & Susanty, E. (2019). The SWOT analysis for chrysanthemum farmers business development strategies for fresh chrysanthemum farmers. *GATR Journal of Business and Economics Review*, *4*(3), 137–146. https://doi.org/10.35609/jber.2019.4.3(3)

Herrera, C. S., Hirooka, Y., & Chaverri, P. (2016). Pseudocospeciation of the mycoparasite Cosmospora with their fungal hosts. *Ecology and Evolution*, *6*(5), 1504–1514. https://doi.org/10.1002/ece3.1967

Heydari, A., & Pessarakli, M. (2010). A review on Biological Control of fungal plant pathogens using microbial antagonists. *Journal of Biological Sciences*, *10*(4), 273–290. https://doi.org/10.3923/jbs.2010.273.290

Johnson, I., Ramjegathesh, R., Sheela, J., Shoba, N., & Maheshwarappa, H. P. (2017). Development of microbial consortia for the management of leaf blight disease of coconut. *Acta Phytopathologica et Entomologica Hungarica*, *52*(1), 1–14. https://doi.org/10.1556/038.52.2017.007

Köhl, J., Kolnaar, R., & Ravensberg, W. J. (2019). Mode of action of microbial biological control agents against plant diseases: relevance beyond efficacy. *Frontiers in Plant Science*, *10*(845), 1–18. https://doi.org/10.3389/fpls.2019.00845

Lengai, G. M. W., & Muthomi, J. W. (2018). Biopesticides and their role in sustainable agricultural production. *Journal of Biosciences and Medicines*, *6*(6), 7–41. https://doi.org/10.4236/jbm.2018.66002

Mahish, P. K., & Ghritlahare, A. (2017). Pathogenicity of phoma chrysanthemicola to chrysanthemum plants (asteraceae family) and control of pathogen by chemical and biological approach. *Biosciences, Biotechnology Research Asia*, *14*(3), 1191–1200. https://doi.org/10.13005/bbra/2561

Marwoto, B. (2012). Penyakit karat putih pada krisan dan upaya pengendaliannya. *Jurnal Penelitian Dan Pengembangan Pertanian*, *31*(2), 51–57. http://repository.pertanian.go.id/handle/123456789/1172

Nuryani, W., Silvia, E., Hanudin, H., & Budiarto, K. (2018). Aplikasi biofungisida berbahan aktif corynebacterium sp. ramah lingkungan dalam pengendalian penyakit karat putih pada krisan. *Jurnal Teknologi Lingkungan*, *19*(1), 23–32. https://doi.org/10.29122/jtl.v19i1.1905

O'Keefe, G., & Davis, D. D. (2015). Morphology of puccinia horiana, causal agent of chrysanthemum white rust, sampled from naturally infected plants. *Plant Disease*, *99*(12), 1738–1743. https://doi.org/10.1094/PDIS-02-15-0239-RE

Omar, I., O'Neill, T. M., & Rossall, S. (2006). Biological control of fusarium crown and root rot of tomato with antagonistic bacteria and integrated control when combined with the fungicide carbendazim. *Plant Pathology*, *55*(1), 92–99. https://doi.org/10.1111/j.1365-3059.2005.01315.x

Prayitno, J. (2017). Ujicoba konsorsium mikroba dalam upaya bioremediasi tanah tercemar minyak dengan menggunakan teknik landfarming skala bangku. *Jurnal Teknologi Lingkungan*, *18*(2), 208–215. https://ejurnal.bppt.go.id/index.php/JTL/article/view/2050

Rahardjo, I. B., Budiarto, K., & Marwoto, B. (2019). Evaluation of biological and chemical fungicides to control white rust in chrysanthemum grown under open condition. *AGRIVITA Journal of Agricultural Science*, *41*(1), 55–63. https://doi.org/10.17503/agrivita.v41i1.1787

Ranadev, P., Nagaraju, K., Muthuraju, R., & Kumari, R. V. (2019). Studies on isolation, characterization and in-vitro screening of plant growth promoting rhizobacteria from rhizospheric soil of chrysanthemum (Dendranthema grandiflora Tzvelev.). *International Journal of Current Microbiology and Applied Sciences*, *8*(6), 790–803. https://doi.org/10.20546/ijcmas.2019.806.096

Rosyidah, A., Wardiyati, T., Abadi, A. L., & Maghfoer, M. D. (2013). Enhancement in effectiveness of antagonistic microbe by means of microbial combination to control Ralstonia solanacearum On potato planted in middle latitude. *AGRIVITA Journal of Agricultural Science*, *35*(2), 174–183. https://doi.org/10.17503/Agrivita-2013-35-2-p174-183

Silber, J., Ohlendorf, B., Labes, A., Wenzel-Storjohann, A., NÃther, C., & Imhoff, J. F. (2014). Malettinin E, an antibacterial and antifungal tropolone produced by a marine Cladosporium strain. *Frontiers in Marine Science*, *1*(35). https://doi.org/10.3389/fmars.2014.00035

Singh, P. K., & Vijay, K. (2011). Biological control of fusarium wilt of chrysanthemum with trichoderma and botanicals. *Journal of Agricultural Technology*, *7*(6), 1603–1613. http://www.ijat-aatsea.com/pdf/v7_n6_11_November/13_IJAT_2011_7_6__Pawan_Kumar_Singh_FX_confirmed.pdf

Suryadi, Y., Susilowati, D. N., Kadir, T. S., Zaffan, Z. R., Hikmawati, N., & Mubarik, N. R. (2013). Bioformulation of antagonistic bacterial consortium for controlling blast, sheath blight and bacterial blight diseases on rice. *Asian Journal of Plant Pathology*, *7*(3), 92–108. https://doi.org/10.3923/ajppaj.2013.92.108

Torres, D. E., Rojas-Martínez, R. I., Zavaleta-Mejía, E., Guevara-Fefer, P., Márquez-Guzmán, G. J., & Pérez-Martínez, C. (2017). Cladosporium cladosporioides and Cladosporium pseudocladosporioides as potential new fungal antagonists of Puccinia horiana Henn., the causal agent of chrysanthemum white rust. *PLOS ONE*, *12*(1), e0170782. https://doi.org/10.1371/journal.pone.0170782

Traquair, J. A., Meloche, R. B., Jarvis, W. R., & Baker, K. W. (1984). Hyperparasitism of Puccinia violae by Cladosporium uredinicola. *Canadian Journal of Botany*, *62*(1), 181–184. https://doi.org/10.1139/b84-030

Wang, Y., Zeng, J., Xia, X., Xu, Y., Sun, J., Gu, J., Sun, H., Lei, H., Chen, F., Jiang, J., Fang, W., & Chen, S. (2020). Comparative analysis of leaf trichomes, epidermal wax and defense enzymes activities in response to puccinia horiana in chrysanthemum and ajania species. *Horticultural Plant Journal*, *6*(3), 191–198. https://doi.org/10.1016/j.hpj.2020.03.006

Yusuf, E. S., Budiarto, K., & Rahardjo, I. B. (2019). Evaluation of Cladosporium sp. Mycoparacites as biocontrol agents of white rust disease on chrysanthemum. *AGRIVITA Journal of Agricultural Science*, *41*(3), 405–415. https://doi.org/10.17503/agrivita.v41i3.1864

Yusuf, E. S., Djatnika, I., & Suhardi, S. (2014). Koleksi dan Karakterisasi Mikoparasit Asal Karat Putih Pada Krisan. *Jurnal Hortikultura*, *24*(1), 56–64. http://ejurnal.litbang.pertanian.go.id/index.php/jhort/article/view/3336

Yusuf, E. S., & Suhardi, -. (2016). pengaruh varietas, perompesan daun, dan penyemprotan fungisida terhadap intensitas penyakit karat (Puccinia horiana P. Henn.) pada tanaman krisan (Dendranthema grandiflora Tzvelev). *Jurnal Hortikultura*, *25*(1), 217–222. https://doi.org/10.24246/agric.2013.v25.i1.p19-25

Yusuf, E. S., & Suhardi, S. (2013). Pengaruh varietas, perompesan daun, dan penyemprotan fungisida terhadap intensitas penyakit karat (Puccinia horiana P. Henn.) pada tanaman krisan (Dendranthema grandiflora Tzvelev). *Agric*, *25*(1), 19–25. https://doi.org/10.24246/agric.2013.v25.i1.p19-25

Zeng, J., Sun, J., Xu, Y., Chen, F., Jiang, J., Fang, W., & Chen, S. (2013). Variation for resistance to white rust (Puccinia horiana) among Ajania and Chrysanthemum Species. *HortScience*, *48*(10), 1231–1234. https://doi.org/10.21273/HORTSCI.48.10.1231