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Compatibility of *Trichoderma harzianum* with Different Concentrations of Copper Oxychloride Fungicide

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Abstract

Trichoderma harzianum is a widely used biocontrol agent known for its antagonistic activity against various plant pathogens. Copper oxychloride is a common fungicide used to combat fungal diseases in agriculture. The present research aims to investigate the compatibility of *Trichoderma harzianum* with different concentrations of copper oxychloride fungicide. The study was conducted to assess the potential impact different concentrations of copper oxychloride viz., 200 ppm, 400 ppm, 600 ppm, 800 ppm and 1000 ppm on the growth, survival, and antagonistic capabilities of *Trichoderma harzianum*. The results obtained from this research can provide valuable insights into the feasibility of integrating *Trichoderma harzianum* with copper oxychloride for sustainable and effective disease management in agriculture.

Keywords: Antagonist activity, Biocontrol agent, Copper oxychloride, Fungal diseases, Fungicide, *Trichoderma harzianum*

Introduction

Trichoderma harzianum is a highly effective biocontrol agent widely utilized in agriculture and horticulture to combat various plant pathogens. This filamentous fungus belongs to the *Trichoderma* genus and is renowned for its exceptional antagonistic properties against a range of plant pathogens, including fungi, bacteria, and nematodes.[1] As a biocontrol agent, *T. harzianum* works through multiple mechanisms. It produces an array of bioactive compounds such as chitinases, glucanases, and proteases that effectively degrade the cell walls of pathogenic organisms, leading to their suppression. Additionally, it competes for nutrients and space with harmful pathogens, further limiting their growth and establishment. Moreover, the fungus stimulates the plant's defense mechanisms, inducing systemic resistance, and enhancing the plant's ability to fend off diseases. The use of *T. harzianum* offers several advantages over chemical pesticides. It is eco-friendly, non-toxic to humans and animals, and does not leave

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harmful residues in the environment. Moreover, its compatibility with various crops and its ability to persist in the rhizosphere make it a sustainable and cost-effective solution for disease management. Overall, *Trichoderma harzianum* has proven to be a promising alternative to synthetic pesticides, promoting environmentally sustainable agriculture while effectively protecting crops from devastating pathogens. Continuous research and development in the field of biocontrol are essential to harness its full potential and ensure food security while safeguarding the environment.[2,3,4,5]

Copper oxychloride is a widely used fungicide in agriculture and horticulture. It is an inorganic compound that contains copper, oxygen, and chlorine. The chemical formula for copper oxychloride is Cu(OH)Cl, and its molecular structure consists of copper ions (Cu2+), hydroxyl groups (OH-), and chlorine ions (Cl-). Copper oxychloride is typically available in the form of a light green powder or granules. The active ingredient, copper oxychloride, is often mixed with inert carriers and adjuvants to facilitate application and improve its efficacy. Copper oxychloride is usually applied as a foliar spray or as a seed treatment. It is used to control various fungal diseases in crops, such as powdery mildew, downy mildew, anthracnose, and other fungal infections. The frequency and timing of applications depend on the crop, the target fungal pathogen, and environmental conditions.[6,7]

Copper oxychloride acts on fungi through multiple mechanisms such as disruption of cell membranes: copper ions interfere with the functioning of fungal cell membranes. They can penetrate the cell walls and disturb the permeability of the cell membrane, leading to the leakage of essential cellular contents and causing the cell to die, generation of Reactive Oxygen Species (ROS): inside the fungal cells, copper ions can catalyze the formation of ROS such as superoxide radicals and hydroxyl radicals. ROS cause oxidative stress and damage various cellular components, ultimately leading to the death of the fungus, inhibition of enzymes: copper oxychloride can inhibit key enzymes involved in vital metabolic pathways of the fungi, disrupting their growth and reproduction, prevention of spore germination: copper oxychloride can prevent spore germination in fungi, which is crucial for their propagation and spread and Systemic Acquired Resistance (SAR) Induction: in plants treated with copper oxychloride, the compound can trigger the plant's defense mechanisms, inducing systemic acquired resistance (SAR). SAR helps the plant become more resistant to fungal infections by activating defense-related genes and producing antifungal compounds.[6,8,9,10,11]

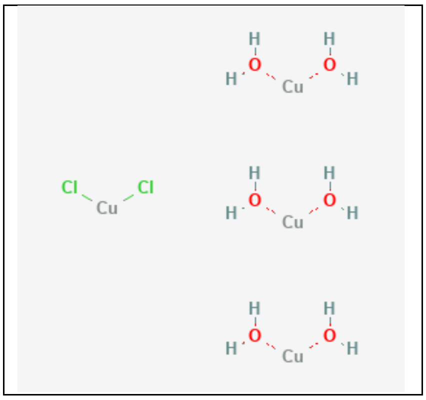

Fig 1: Structure of copper oxychloride [11]

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Trichoderma harzianum is a well-known biocontrol agent widely used in sustainable agriculture practices due to its potential in controlling a wide range of plant pathogens. On the other hand, copper oxychloride is a commonly used fungicide for the control of fungal diseases in various crops. The co-application of biocontrol agents and fungicides has garnered interest in integrated disease management strategies. This research aims to evaluate the compatibility of *Trichoderma harzianum* with different concentrations of copper oxychloride fungicide, with a focus on their combined effects on plant health and the antagonistic activity of *Trichoderma harzianum*.

Materials and Methods

Sample collection:

- Collection of *Trichoderma harzianum* sample

Trichoderma harzianum was collected from Microbiology laboratory, Department of Microbiology, Maharani Cluster University, Bengaluru, Karnataka, India and cultured on Potato Dextrose Agar (PDA) medium. The pure culture of *Trichoderma harzianum* was maintained on PDA slants.

- Copper Oxychloride fungicide

Copper oxychloride fungicide was obtained commercially from the local markets.

Compatibility Assay:

Poisoned Food Technique was used to assess the compatibility of *Trichoderma harzianum* with different concentrations of copper oxychloride viz., 200 ppm, 400 ppm, 600 ppm, 800 ppm and 1000 ppm. Potato dextrose agar was prepared, autoclaved and copper oxychloride was incorporated with potato dextrose agar media at different concentrations. Plates without copper oxychloride served as the control. After inoculation of *Trichoderma harzianum* on agar plates, plates were incubated at 27°C for 7 days. The growth inhibition zone and mycelial density of *Trichoderma harzianum* were measured after incubation. Experiments were performed in triplicates.

Results

The in vitro compatibility assay revealed that *Trichoderma harzianum* exhibited tolerance to different concentrations of copper oxychloride at all concentrations tested viz., 200 ppm, 400 ppm, 600 ppm, 800 ppm and 1000 ppm.

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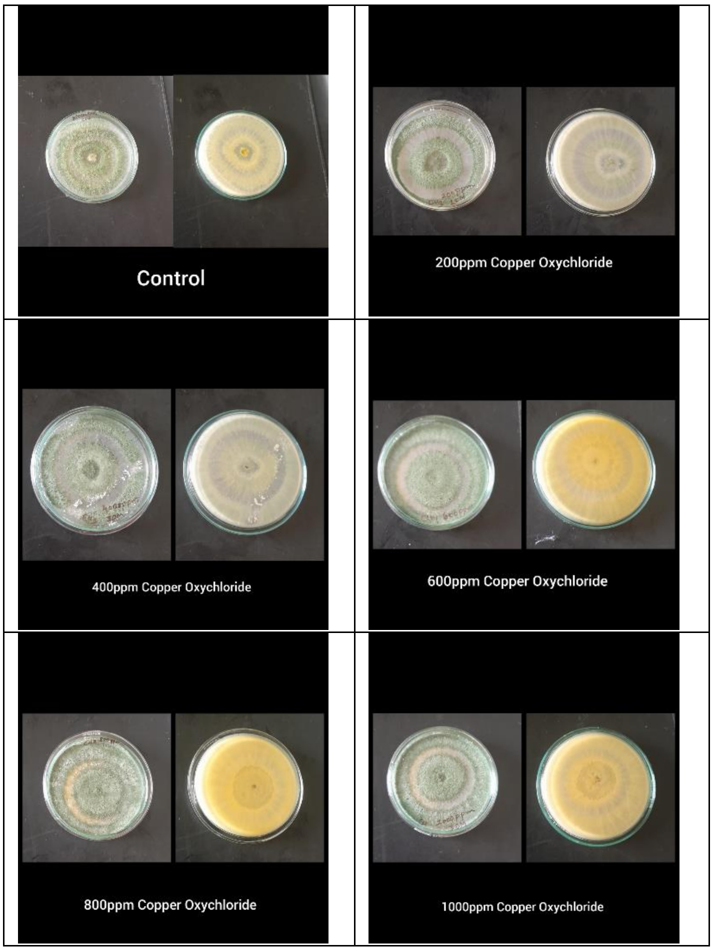

Fig 2: Compatibility of *Trichoderma harzianum* with Copper oxychloride

Table 1: Compatibility of *Trichoderma harzianum* with Copper oxychloride

Fungicide	Concentrations	Zone of inhibition (mm)	Mean colony diameter (mm)
Control	00 ppm	00	90
Copper oxychloride	200 ppm	00	90
Copper oxychloride	400 ppm	00	90
Copper oxychloride	600 ppm	00	90
Copper oxychloride	800 ppm	00	90
Copper oxychloride	1000 ppm	00	90

Discussion

The results of this research indicate that *Trichoderma harzianum* is completely compatible with different concentrations of copper oxychloride, making them suitable for co-application in integrated disease management strategies.

From the study of Bhale, Udhav et al., compatibility of fungicides viz. Mancozeb and Captan with *Trichoderma spp (T. viride, T. harzianum, T. koningii, T. pseudokoningii and T. virens)* at

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different concentrations was revealed. The results indicated that lower concentrations of Mancozeb and Captan was not effective in inhibiting the radial growth of *Trichoderma spp*. However, concentration of Mancozeb above 5000 µg/ml and of Captan above 500 µg/ml significantly reduced the radial growth of *Trichoderma spp*. *Trichoderma viride, T. harzianum, T. virens and T. koningii*.[12] These results are partially similar and partially in contrast with our result where *Trichoderma harzianum* was fully compatible with all concentrations of copper oxychloride viz., 200 ppm, 400 ppm, 600 ppm, 800 ppm and 1000 ppm. Amoghavarsha Chittaragi et al., had studied the compatibility of *Trichoderma asperellum* with captan fungicide. Results revealed that *Trichoderma asperellum* was not compatible with captan.[13] These results are in contrast with our results where *Trichoderma harzianum* is compatible with copper oxychloride fungicide. G. Bindu Madhavi et al., had studied the compatibility of *Trichoderma viride* with contact fungicides viz., pencycuron and propineb. Results revealed that *Trichoderma viride* was compatible with pencycuron and propineb.[14] Similar to that study, in the present study *Trichoderma harzianum* was compatible with copper oxychloride fungicide.

The present study explores the compatibility of *Trichoderma harzianum*, a biocontrol agent widely used in agriculture, with different concentrations of Copper Oxychloride fungicide. The study employed the poisoned food technique to evaluate the potential interactions between the biocontrol agent and the fungicide. The findings reveal crucial insights into the effects of Copper Oxychloride on the viability and growth of *Trichoderma harzianum*. The results indicate that all concentrations of the fungicide has supported the growth of *Trichoderma harzianum*. This research has significant implications for sustainable agricultural practices, as it highlights the importance of understanding the compatibility of biocontrol agents and chemical fungicides. Farmers and researchers can use this information to make informed decisions about integrated pest management strategies that maximize the benefits of both biocontrol agents and chemical interventions while minimizing potential adverse effects. Further studies could focus on investigating the underlying mechanisms behind the observed interactions and exploring alternative combinations of biocontrol agents and fungicides to optimize their compatibility and enhance overall disease management strategies in agriculture.

Conclusion

In conclusion, *Trichoderma harzianum* can be effectively integrated with different concentrations of copper oxychloride for enhanced disease management without compromising its antagonistic potential. This integrated approach holds promise for sustainable and environmentally friendly disease control in agriculture. However, further research is required to determine the optimal concentrations and application methods for achieving the best results in different crop systems.

References:

1. Felipe M. Rivera, Elisabete B. Paula Barros, Alcilucia Oliveira, Claudia M. Rezende, Selma G.F. Leite, Chapter 113 – Production of 6-Pentyl-Î±-Pyrone by Trichoderma harzianum using Brazilian Espresso Coffee Grounds, Editor(s): Vicente Ferreira,

ISSN PRINT 2319 1775 Online 2320 7876

Research paper © 2012 IJFANS. All Rights Reserved, UGC CARE Listed (Group -I) Journal Volume 12, Iss 1, Jan 2023

Ricardo Lopez, Flavour Science, Academic Press, 2014, Pages 619-622, ISBN 9780123985491, https://doi.org/10.1016/B978-0-12-398549-1.00113-6. (https://www.sciencedirect.com/science/article/pii/B9780123985491001136)

2. WALTER GAMS, PAUL DIEDERICH, KADRI PÃ•LDMAA, 17 – FUNGICOLOUS FUNGI, Editor(s): GREGORY M. MUELLER, GERALD F. BILLS, MERCEDES S. FOSTER, Biodiversity of Fungi, Academic Press, 2004, Pages 343-392, ISBN 9780125095518, https://doi.org/10.1016/B978-012509551-8/50020-9. (https://www.sciencedirect.com/science/article/pii/B9780125095518500209)
3. GEORGE N. AGRIOS, Chapter nine – CONTROL OF PLANT DISEASES, Editor(s): GEORGE N. AGRIOS, Plant Pathology (Fifth Edition), Academic Press, 2005, Pages 293-353, ISBN 9780120445653, https://doi.org/10.1016/B978-0-08-047378-9.50015-4.(https://www.sciencedirect.com/science/article/pii/B9780080473789500154)
4. RIVKA BARKAI-GOLAN, CHAPTER 11 – BIOLOGICAL CONTROL, Editor(s): RIVKA BARKAI-GOLAN, Postharvest Diseases of Fruits and Vegetables, Elsevier, 2001, Pages 221-251, ISBN 9780444505842, https://doi.org/10.1016/B978-044450584-2/50011-3. (https://www.sciencedirect.com/science/article/pii/B9780444505842500113)
5. Sriyanka Lahiri, David Orr, Chapter 11 – Biological Control in Tomato Production Systems: Theory and Practice, Editor(s): Waqas Wakil, Gerald E. Brust, Thomas M. Perring, Sustainable Management of Arthropod Pests of Tomato, Academic Press, 2018, Pages 253-267, ISBN 9780128024416, https://doi.org/10.1016/B978-0-12-802441-6.00011-5. (https://www.sciencedirect.com/science/article/pii/B9780128024416000115)
6. Bayram S, Genc A, Buyukleyla M, Rencuzogullari E. Genotoxicity and cytotoxicity of copper oxychloride in cultured human lymphocytes using cytogenetic and molecular tests. Cytotechnology. 2016 Oct;68(5):2027-36. Doi: 10.1007/s10616-016-9943-8. Epub 2016 Jan 11. PMID: 26754841; PMCID: PMC5023576.
7. Ferreira LC, Scavroni J, da Silva JR, Cataneo AC, Martins D, Boaro CS. Copper oxychloride fungicide and its effect on growth and oxidative stress of potato plants. Pestic Biochem Physiol. 2014 Jun;112:63-9. Doi: 10.1016/j.pestbp.2014.04.010. Epub 2014 May 8. PMID: 24974119.
8. Mathew P, Austin RD, Varghese SS, Manojkumar AD. Effect of copper-based fungicide (bordeaux mixture) spray on the total copper content of areca nut: Implications in increasing prevalence of oral submucous fibrosis. J Int Soc Prev Community Dent. 2015 Jul-Aug;5(4):283-9. Doi: 10.4103/2231-0762.161755. PMID: 26312227; PMCID: PMC4547442.
9. Ottesen AR, Gorham S, Pettengill JB, Rideout S, Evans P, Brown E. The impact of systemic and copper pesticide applications on the phyllosphere microflora of tomatoes. J Sci Food Agric. 2015 Mar 30;95(5):1116-25. Doi: 10.1002/jsfa.7010. Epub 2014 Dec 11. PMID: 25410588; PMCID: PMC4368374.
10. European Food Safety Authority (EFSA); Arena M, Auteri D, Barmaz S, Bellisai G, Brancato A, Brocca D, Bura L, Byers H, Chiusolo A, Court Marques D, Crivellente F, De Lentdecker C, Egsmose M, Erdos Z, Fait G, Ferreira L, Goumenou M, Greco L, Ippolito A, Istace F, Jarrah S, Kardassi D, Leuschner R, Lythgo C, Magrans JO, Medina P, Miron I, Molnar T, Nougadere A, Padovani L, Parra Morte JM, Pedersen R, Reich

ISSN PRINT 2319 1775 Online 2320 7876

Research paper UGC CARE Listed (Group -I) Journal Volume 12, Iss 1, Jan 2023

H, Sacchi A, Santos M, Serafimova R, Sharp R, Stanek A, Streissl F, Sturma J, Szentes C, Tarazona J, Terron A, Theobald A, Vagenende B, Verani A, Villamar-Bouza L. Peer review of the pesticide risk assessment of the active substance copper compounds copper(I), copper(II) variants namely copper hydroxide, copper oxychloride, tribasic copper sulfate, copper(I) oxide, Bordeaux mixture. EFSA J. 2018 Jan 16;16(1):e05152. Doi: 10.2903/j.efsa.2018.5152. PMID: 32625696; PMCID: PMC7009614.

11. National Center for Biotechnology Information (2023). PubChem Compound Summary for CID 44135802, copper oxychloride, AldrichCPR. Retrieved July 27, 2023 from https://pubchem.ncbi.nlm.nih.gov/compound/copper-oxychloride_-AldrichCPR.
12. Bhale, Udhav. (2016). Compatibility of Fungicides and Antagonistic Activity of Trichoderma spp against plant Pathogens. Bioscience Methods. 6. 1-9.
13. Maheshwary NP, Gangadhara Naik B, Amoghavarsha Chittaragi, MK Naik, Satish KM and Nandish MS, Compatibility of Trichoderma asperellum with fungicides, The Pharma Innovation Journal 2020; 9(8): 136-140, ISSN (E): 2277- 7695, ISSN (P): 2349-8242, www.thepharmajournal.com
14. G. Bindu Madhavi, S.L. Bhattiprolu and V. Bali Reddy, Compatibility of biocontrol agent Trichoderma viride with various pesticides, J. Hortl. Sci., Vol. 6(1):71-73, 2011