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RESEARCH PAPER

Efficacy of selected fungicides and antibacterial chemicals against *Fusarium verticillioides*, *Macrophomina phaseolina* and *Dickeya zeae* associated with maize stalk rot

Mudunuri Ramabhadra Raju • Battina Deepthi Priya • Vemana Sujatha • Injeti Sudhir Kumar

Abstract: Maize stalk rot is a devastating disease complex caused by the co-infection of fungal and bacterial pathogens, severely threatening global maize production. This study evaluated the *in vitro* efficacy of eleven fungicides (at 100%, 50%, and 25% of the recommended field rates) against *Fusarium verticillioides* and *Macrophomina phaseolina* using the poisoned food technique. Concurrently, five antibacterial agents were screened against *Dickeya zeae* using a disc diffusion assay under a Completely Randomized Design (CRD). *In vitro* screening revealed that tebuconazole 38.39 SC, thiophanate-methyl 70 WP, azoxystrobin 18.3 + difenoconazole 11.4 SC, trifloxystrobin 25 + tebuconazole 50 WG, and captan 70 + hexaconazole 5 WP formulations exhibited superior antifungal activity, achieving over 90% mycelial growth inhibition against the key post-flowering stalk rot pathogens *Fusarium verticillioides* and *Macrophomina phaseolina* even at 50% of their recommended field dosages. Probit analysis corroborated this, with EC50 values of the top-performing fungicides against both pathogens consistently falling below 25% of the recommended field dose, indicating scope for effective disease control at reduced dosages. Against the bacterial pathogen *D. zeae*, copper oxychloride 50 WP at 0.3% emerged as the most effective non-antibiotic treatment, achieving 71.0% relative efficacy relative to positive control, Plantomycin @ 250 ppm (Streptomycin Sulphate 9% + Tetracycline Hydrochloride 1% SP), followed by bleaching powder at 3% (66.1%) and copper hydroxide 46.1 WG at 0.2% (41.9%). These baseline *in vitro* findings identify highly potent, broad-spectrum chemical candidates for managing the maize stalk rot complex. Further validation under greenhouse and natural field conditions is warranted to incorporate these molecules into sustainable integrated disease management (IDM) strategies.

Keywords: Maize • Stalk rot • *Fusarium verticillioides* • *Macrophomina phaseolina* • *Dickeya zeae* • Bacterial stalk rot • Fungicides • Antibiotics

Introduction

Maize (*Zea mays* L.) is one of the most important cereal crops worldwide, serving as a vital source of food, feed, and industrial raw material. Global maize production during 2025–26 is projected at approximately 1.296 billion tonnes, while India recorded a historic production of 55.09 million tonnes during the same period (USDA, 2026; MoA and FW, 2026). In India, maize is known to be affected by more than 61 diseases with seedling blights, leaf and sheath blights, post-flowering stalk rots and grain infections being most prevalent and destructive (Payak and Sharma, 1985). Post-flowering stalk rots (PFSR) are destructive diseases affecting maize production in tropical and subtropical regions. Yield losses due to the stalk rot complex have been reported to range from 10 to 42 per cent under natural field conditions and may become considerably higher under favourable

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environmental conditions (Khokhar *et al*., 2014). The severity of the PFSR is often aggravated by drought stress, nutrient imbalance, high plant population and other physiological stresses during the reproductive phase of crop growth (Dodd, 1980). Among the stalk rot pathogens of maize, *Fusarium verticillioides* is the most prevalent pathogen capable of infecting roots, stalks, ears and kernels and causing substantial yield losses and deterioration of grain quality. Charcoal rot caused by *Macrophomina phaseolina* is another important component of the maize stalk rot complex. The pathogen possesses an exceptionally wide host range and capable of surviving in soil through microsclerotia for prolonged periods (Su *et al*., 2001). Infection results in shredding of vascular tissues, pith disintegration, premature drying and lodging of affected plants (Gupta *et al*., 2012; Marquez and Giachero, 2021). Long term survival ability of the PFSR pathogens and their adaptability to adverse environmental conditions often limit the effectiveness of cultural management practices. In recent years, bacterial stalk rot caused by *Dickeya zeae* has emerged as a serious threat to maize cultivation in several maize-growing regions, particularly in kharif-sown crops. The disease is characterized by soft rot and maceration of stalk tissues, accompanied by a characteristic foul odour, ultimately leading to lodging and collapse of affected plants. Under severe disease pressure, yield losses may range from 21 to 98% (Thind and Payak, 1978).

Among the available management interventions for minimizing losses due to PFSR, fungicides continue to play an important role owing to their rapid action and effectiveness against a broad spectrum of pathogens. Fungicides belonging to different chemical groups have been reported to effectively suppress *Macrophomina phaseolina* and other stalk rot pathogens, highlighting their potential role in disease management (Iqbal and Mukhtar, 2020; Lokesh *et al*., 2020; Patil *et al*., 2024). Similarly, Solorzano and Malvick (2011) reported improved maize germination, plant stand and grain yield following fungicidal seed treatment against Fusarium-associated diseases, while Kumar *et al*. (2016) demonstrated the effectiveness of chemical treatments in suppressing bacterial stalk rot caused by *Dickeya zeae*. Despite the availability of several fungicidal and antibacterial molecules, comparative information regarding efficacy of specific fungicides/ bactericides against the major fungal and bacterial pathogens associated with the maize stalk rot complex remains limited. Therefore, the present investigation was undertaken to evaluate the *In vitro* efficacy of selected chemical treatments against *Fusarium verticillioides*, *Macrophomina phaseolina* and *Dickeya zeae,* major stalk rot pathogens of maize.

Materials and methods

Experimental location

The present investigation was conducted in the Department of Plant Pathology, Agricultural Research Station, Peddapuram, Kakinada district – 533437, Andhra Pradesh, India, during 2025–26. The study was undertaken to evaluate the *in vitro* efficacy of selected fungicides against major stalk rot pathogens of maize, namely *Fusarium verticillioides*, *Macrophomina phaseolina* and *Dickeya zeae*.

Isolation and maintenance of pathogens

Freshly infected maize stalk samples exhibiting typical stalk rot symptoms were collected from the field and washed thoroughly under running tap water to remove adhering soil and debris. Small pieces consisting of both healthy and diseased sections were excised from the advancing margin of infection, surface sterilised with 0.1% sodium hypochlorite solution for 1–2 min, followed by three washings with sterile distilled water under aseptic conditions. The surface sterilised pieces were blotted dry on sterile filter paper and placed on Petri plates containing potato dextrose agar (PDA) medium and incubated at 28 ± 2°C for four days. Pure cultures of the fungal pathogens were obtained by hyphal tip method and maintained through periodic subculturing on Potato dextrose agar medium. The fungal pathogens were identified based on morphological characteristics, including conidial shape and size, hyphal pattern, and colony colour, and their identities were subsequently confirmed through the identification services of the Indian Type Culture Collection (ITCC), New Delhi, as *Fusarium verticillioides* and *Macrophomina phaseolina*.

For isolation of the bacterial pathogen, infected stalks were surface sterilised and cut into small pieces. The pieces were immersed in sterile distilled water for 10 min to allow bacterial ooze to diffuse into the suspension. The suspension was streaked onto nutrient agar (NA)

plates and incubated at 28 ± 2°C for 24–48 h. Pure cultures were obtained by repeated subculturing of individual colonies and maintained at 4°C for further experimentation. The bacterial isolate was tentatively identified as *Dickeya zeae* based on its colony morphology, biochemical characteristics, including the KOH, oxidase, and catalase tests, and pectolytic activity on potato and carrot tissues. The identity of the isolate was further supported by successful pathogenicity testing on maize.

Chemicals evaluated

A total of eleven chemicals with fungicidal/fungistatic properties (eight fungicides, two copper compounds and bleaching powder) were evaluated at three different doses – 100% recommended dose of formulation (RDF), 50% RDF and 25% RDF – against the fungal stalk rot pathogens. Five chemicals, namely copper oxychloride 50 WP, copper hydroxide 46.1 WG, bleaching powder 30%, streptomycin sulphate 9% + tetracycline hydrochloride 1% SP (standard antibiotic check, positive control), and 2-bromo-2-nitropropane-1,3-diol, were evaluated against the bacterial stalk rot pathogen.

Evaluation of fungicides against fungal pathogens

The efficacy of fungicides against *Fusarium verticillioides* and *Macrophomina phaseolina* was evaluated by the poisoned food technique (Nene and Thapliyal, 1979). The required quantity of each fungicide was incorporated into sterilised molten Potato Dextrose Agar (PDA) medium maintained at 45–50°C to obtain the desired concentrations corresponding to 25, 50 and 100 per cent of the recommended dose of formulation. The amended medium was thoroughly mixed and poured into sterilised Petri plates (90 mm diameter). After solidification a 5 mm diameter mycelial disc from the five days old actively growing culture of each stalk rot pathogen was placed at the centre of the Petri plate. PDA plates without fungicide served as untreated controls. Each treatment was replicated three times and arranged in a Completely Randomized Design (CRD). The inoculated plates were incubated at 28 ± 1°C until the fungal growth in the control plates reached the edge of the Petri dish.

The per cent inhibition of mycelial growth over control for fungi and relative efficacy relative to positive control for bacteria were calculated using the formula proposed by Vincent (1947):

$$P = ((C - T)/C) \times 100$$

where, P = Per cent inhibition of mycelial growth/ Relative efficacy relative to positive control; C = Radial growth of pathogen in control (mm)/Inhibition annulus in positive control cm² (Plantomycin); T = Radial growth of pathogen in fungicide-treated plates (mm)/Inhibition annulus in chemical treatment cm².

Table 1. Fungicides and antibacterial chemicals evaluated against the maize stalk rot pathogens

Chemical name	Trade name	100% RDF	a.i. at 100% field dose (µg/mL)
Propiconazole 25 EC	Tilt	1.0 ml/L	250.00
Tebuconazole 38.39 SC	Buonos	1.5 ml/L	575.85
Copper oxychloride 50 WP	Blitox	3.0 g/L	1500.00
Copper hydroxide 46.1 WG	Kocide	2.0 g/L	922.00
Bleaching powder 30%	–	3.0 g/L	300.00
Thiophanate methyl 70% WP	Topsin-M	1.5 g/L	1050.00
Azoxystrobin 18.2% + Difenoconazole 11.4% SC	Amistar Top	1.25 ml/L	370.00
Trifloxystrobin 25% + Tebuconazole 50% WG	Nativo	0.4 g/L	300.00
Captan 70% + Hexaconazole 5% WP	Taqat	2.0 g/L	1500.00
Metiram 55% + Pyraclostrobin 5% WG	Cabrio Top	1.5 g/L	900.00
Fluxapyroxad 250 g/L + Pyraclostrobin 250 g/L SC	Priaxor	1.0 ml/L	500.00
Streptomycin Sulphate 9% + Tetracycline Hydrochloride 1% SP	Plantomycin	1.0 g/L	100.00
2-Bromo-2-nitropropane-1,3-diol	Bronopol	0.33 g/L	330.00

RDC – Recommended dose of formulation; a.i.- active ingredient

In vitro evaluation against Dickeya zeae by disc diffusion assay

The efficacy of selected antibacterial chemicals against *Dickeya zeae* was evaluated using the disc diffusion assay (Bauer *et al*., 1966). Nutrient agar plates were seeded with a bacterial suspension adjusted to approximately 10^8 CFU mL^{-1}. Whatman No. 4 filter paper discs (5 mm diameter) were impregnated with the respective chemical solutions at 25, 50 and 100% of the recommended concentrations and placed on the surface of nutrient agar plates previously seeded with the bacterial suspension. The plates were incubated at 28 ± 1°C for 24–48 h and the diameter of the inhibition zone surrounding each disc was recorded. The Inhibition annulus (cm^2) was calculated and used for comparison among treatments. Relative efficacy of the tested chemicals was calculated in comparison with the check treatment, plantomycin (Streptomycin Sulphate 9% + Tetracycline Hydrochloride 1% SP) at 250 ppm.

Inhibition annulus (IA) = $\pi (R_1 - R_2) \times (R_1 + R_2)$

where, IA = Area of the inhibition annulus (cm^2); R_1 = Radius of the inhibition zone + radius of the filter paper disc/well; R_2 = Radius of the filter paper disc or agar well alone; π = Constant approximately equal to 3.14.

Statistical analysis

The experiment was conducted in a Completely Randomized Design (CRD) with three replications. Data on radial growth, percent inhibition, inhibition annulus and relative efficacy were subjected to analysis of variance (ANOVA). Percentage data were transformed using angular (arcsine) transformation wherever necessary before analysis. Treatment means were compared using the Critical Difference (CD) test at the 5% level of significance and were further subjected to Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$ for mean separation into significant groups.

For fungicides evaluated against the maize fungal stalk rot pathogens, the concentration of active ingredient (a.i.) incorporated into the poisoned medium at 25, 50 and 100% of the recommended field dose was calculated in µg/mL (ppm) from the field application rate and the declared a.i. content of each formulation. Per cent inhibition values were subjected to probit analysis, regressing the probit of proportional inhibition against $\log_{10}$ (a.i. concentration), to estimate the half-maximal (EC50) and 90% (EC90) effective concentrations for each fungicide against *F. verticillioides* and *M. phaseolina*. Where the fitted EC50/EC90 fell outside the concentration range actually tested, it is reported as below (<) or above (>) the nearest tested concentration rather than as an extrapolated value, since only three concentration levels spanning a four-fold range were bioassayed.

Results and discussion

In vitro efficacy of fungicides against Fusarium verticillioides

Differential efficacy among fungicides in suppressing the growth of *Fusarium verticillioides* was observed under *in vitro* conditions. Propiconazole 25 EC, tebuconazole 38.39 SC, thiophanate methyl 70 WP and trifloxystrobin 25% + tebuconazole 50% WG completely suppressed the growth of *F. verticillioides* at different concentrations under evaluation (Figure 1, Plate 1). Whereas, azoxystrobin 18.2% + difenoconazole 11.4% SC exhibited comparable efficacy with 94.19% inhibition of growth of *F. verticillioides*. The superior performance of these fungicides may be attributed to the inhibition of ergosterol biosynthesis by triazoles and disruption of mitochondrial respiration by strobilurins, resulting in effective suppression of fungal growth (Bartlett *et al*., 2002). Similar observations on the efficacy of triazole- and strobilurin-based fungicides against stalk rot and other fungal pathogens have been reported by Thori *et al*. (2012), Meena *et al*. (2021) and Shin *et al*. (2014). Thiophanate methyl 70 WP also exhibited complete inhibition of the pathogen, which may be associated with its systemic fungitoxic activity and protective as well as curative properties against fungal pathogens (Rekanovic *et al*., 2010). Copper hydroxide 46.1 WG and bleaching powder 30% were comparatively less effective, recording inhibition values of 23.58 and 7.41%, respectively over control. Increasing fungicide concentration significantly enhanced pathogen suppression, with mean inhibition increasing from 55.08% at 25% concentration to 65.19% at 100%, indicating a positive concentration-dependent response. The effectiveness of combination fungicides may be attributed to the presence of active ingredients with different modes of action, which enhance fungitoxic activity and broaden the spectrum of pathogen control.

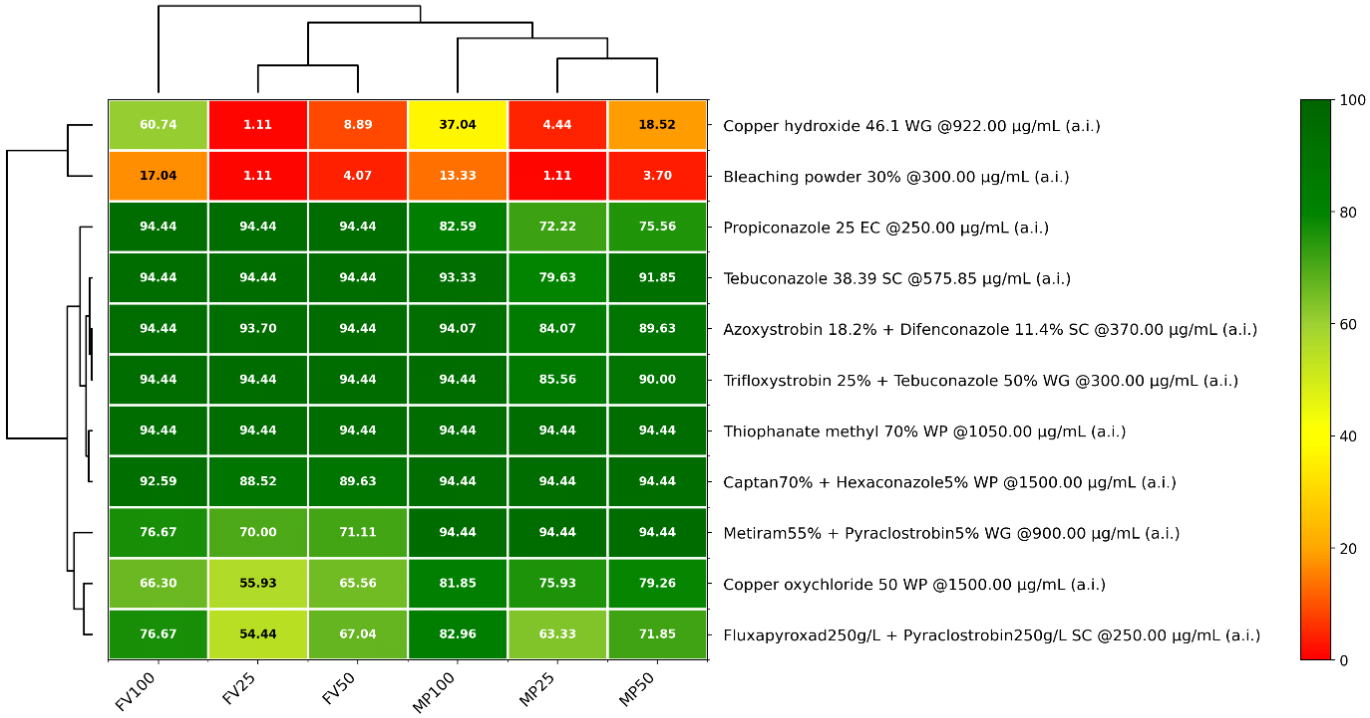
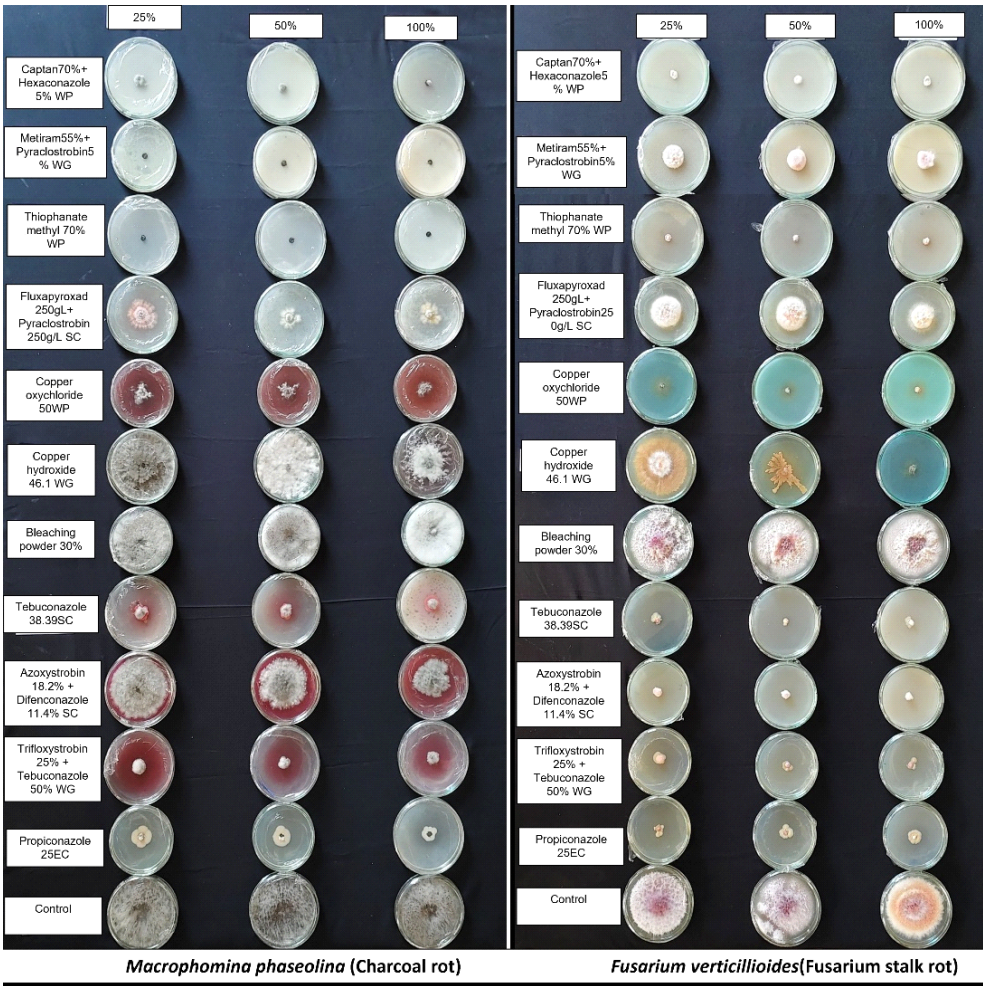


Figure 1. Clustered heat map with hierarchical clustering of fungicides based on inhibitory activity against *Fusarium verticillioides* and *Macrophomina phaseolina*

Plate 1. Relative efficacy of fungicides at three doses (100% RDF, 50% RDF and 25% RDF) on growth of *Fusarium verticillioides* (Fusarium stalk rot pathogen) and *Macrophomina phaseolina* (Charcoal rot pathogen) under *in vitro* conditions (Poisoned food technique)



In vitro efficacy of fungicides against Macrophomina phaseolina

Among the fungicides, thiophanate methyl 70 WP, captan 70% + hexaconazole 5% WP and metiram 55% + pyraclostrobin 5% WG completely inhibited mycelial growth of charcoal rot pathogen, *Macrophomina phaseolina* at all concentrations (Figure 1, Plate 1). Trifloxystrobin 25% + tebuconazole 50% WG, azoxystrobin 18.2% + difenoconazole 11.4% SC and tebuconazole 38.39 SC were also effective, recording inhibition values above 88% and significantly restricting fungal growth. Similar findings were reported by Patel *et al*. (2017) and Hegde *et al*. (2018), who observed substantial suppression of *M. phaseolina* by systemic fungicides and fungicide combinations under *in vitro* conditions. Likewise, Chouhan *et al*. (2025) reported strong inhibitory activity of metiram 55% + pyraclostrobin 5% WG against *M. phaseolina*, with efficacy increasing at higher concentrations. The effectiveness of thiophanate methyl may be attributed to its conversion into carbendazim, which interferes with fungal cell division through disruption of β-tubulin assembly (Davidse and Flach, 1978). Copper oxychloride 50 WP and propiconazole 25 EC provided moderate suppression, whereas fluxapyroxad 250 g/L + pyraclostrobin 250 g/L SC recorded intermediate efficacy. In contrast, copper hydroxide and bleaching powder were the least effective treatments, recording only 20.00% and 6.05% inhibition, respectively.

Probit-based EC50 and EC90 values, expressed as µg/mL (ppm) of active ingredient, are presented in Table 3 for the eleven fungicidal formulations tested against *F. verticillioides* and *M. phaseolina*. Propiconazole 25 EC, tebuconazole 38.39 SC, thiophanate methyl 70 WP and trifloxystrobin 25% + tebuconazole 50% WG required the lowest a.i. concentrations to achieve 50% inhibition of both pathogens, consistent with their complete suppression of mycelial growth even at the lowest dose tested. In contrast, copper hydroxide 46.1 WG and bleaching powder 30% required substantially higher concentrations, with EC50 values exceeding or approaching the highest concentration tested, reflecting their weaker, contact-based fungitoxic activity. Because the bioassay evaluated only three concentrations spanning a four-fold range, EC50 values for the most active fungicides and EC90 values for several treatments could not be precisely interpolated within the tested range and are reported as bounds; these should be interpreted as indicative baseline comparisons rather than precise toxicological endpoints, and confirmation with a wider, lower-concentration dose series is recommended for the most active compounds.

Clustered heatmap depicting *in vitro* efficacy (mean per cent mycelial growth inhibition of three replications) by poisoned food technique for eleven fungicides evaluated at three concentrations — 25%, 50%, and 100% of the field dose (indicated alongside of chemical name) against *Fusarium verticillioides* (FV) and *Macrophomina phaseolina* (MP). Colour intensity, ranging from red (low inhibition) to dark green (high inhibition), represents the mean per cent inhibition value for three replications in each cell. Hierarchical clustering (Euclidean distance, average linkage) was applied to both rows (fungicides) and columns (pathogen–concentration combinations) to group treatments by similarity in efficacy pattern. Mean values were subjected to arcsine (angular) transformation prior to statistical analysis and critical difference (CD at P = 0.05) was 1.31 and 1.67 for fungicide (Factor A), and 0.66 and 0.83 for concentration (Factor B), with corresponding coefficients of variation (CV) of 2.54% and 3.21%, against *F. verticillioides* and *M. phaseolina,* respectively.

In vitro antibacterial efficacy of various chemical agents against Dickeya zeae

Antibacterial chemicals differed significantly in their efficacy against *Dickeya zeae*. Streptomycin sulphate 9% + tetracycline hydrochloride 1% SP (Plantomycin) @ 250 ppm exhibited significantly superior activity as the standard antibiotic control, recording the maximum inhibition annulus of 2.95 cm² (Table 2), followed by copper oxychloride, which showed the highest relative efficacy among the non-antibiotic treatments at 71.0% of the positive control (Plantomycin) at 100% concentration, followed by bleaching powder (66.1% relative efficacy), copper hydroxide (41.9% relative efficacy) and 2-bromo-2-nitropropane-1,3-diol (Figure 2). The antibacterial activity of copper-based compounds may be attributed to membrane disruption, protein denaturation and enzyme inactivation induced by Cu^{2}z ions, whereas bleaching powder acts through oxidative damage mediated by active chlorine release. Similar

findings on the effectiveness of copper-based compounds and oxidative disinfectants against phytopathogenic bacteria have been reported by Lal (1970), Abeer and Raheb (2014), Kumar *et al*. (2016) and Nair and Rashid (2024), supporting their role in bacterial disease suppression. Irrespective of treatment, antibacterial activity increased significantly with increasing concentration. Mean relative efficacy increased from 26.5% at 25% concentration to 62.9% at 100% concentration, while the mean inhibition annulus increased from 0.78 to 1.85 cm², indicating a clear concentration-dependent enhancement in antibacterial activity (Table 2 and Figure 2). The results indicate that copper-based compounds are comparatively more effective in suppressing *D. zeae* growth than the other non-antibiotic treatments.

Hierarchical clustering grouped fungicides according to their inhibitory activity against *F. verticillioides* and *M. phaseolina* at different concentrations, as shown in Figure 1. Systemic fungicides are clustered together, exhibiting consistently high inhibition (>90%) against both pathogens. In contrast, copper hydroxide and bleaching powder formed a separate cluster characterized by comparatively low inhibitory activity. The clustering pattern further revealed a close association between the responses

Table 2. *In vitro* evaluation of antibacterial chemicals for efficacy against *Dickeya zeae* (disc diffusion assay)

Treatment (antibacterials)	Inhibition Annulus (cm²) % Recommended dose of chemical			Mean
	25%	50%	100%	
Copper oxychloride 50 WP	0.19 (0.83)d	0.94 (1.20)c	2.07 (1.60)ab	1.07 (1.21)b
Copper hydroxide 46.1% WG	0.19 (0.83)d	0.84 (1.16)c	1.24 (1.32)bc	0.76 (1.10)c
Bleaching powder 30%	0.19 (0.83)d	0.41 (0.95)d	1.94 (1.56)ab	0.85 (1.11)c
Streptomycin Sulphate 9% + Tetracycline Hydrochloride 1% SP (250 ppm)	2.95 (1.86)a	2.95 (1.86)a	2.95 (1.86)a	2.95 (1.86)a
2-Bromo-2-nitropropane-1,3-diol	0.37 (0.93)d	0.75 (1.12)c	1.03 (1.24)c	0.72 (1.09)c
Mean	0.78 (1.06)c	1.18 (1.26)b	1.85 (1.52)a	

SE(m)±: Factor A (Chemical) 0.02, Factor B (Concentration) 0.01, Interaction (A × B) 0.05. CD (P = 0.05): Factor A 0.04, Factor B 0.09. CV (%): 4.95.

Values in parentheses indicate square-root transformed values used for statistical analysis. Means followed by the same letter(s) are not significantly different according to Duncan's Multiple Range Test (DMRT) at P ≤ .05. Streptomycin Sulphate 9% + Tetracycline Hydrochloride 1% SP (250 ppm) was used as positive control for the treatments.

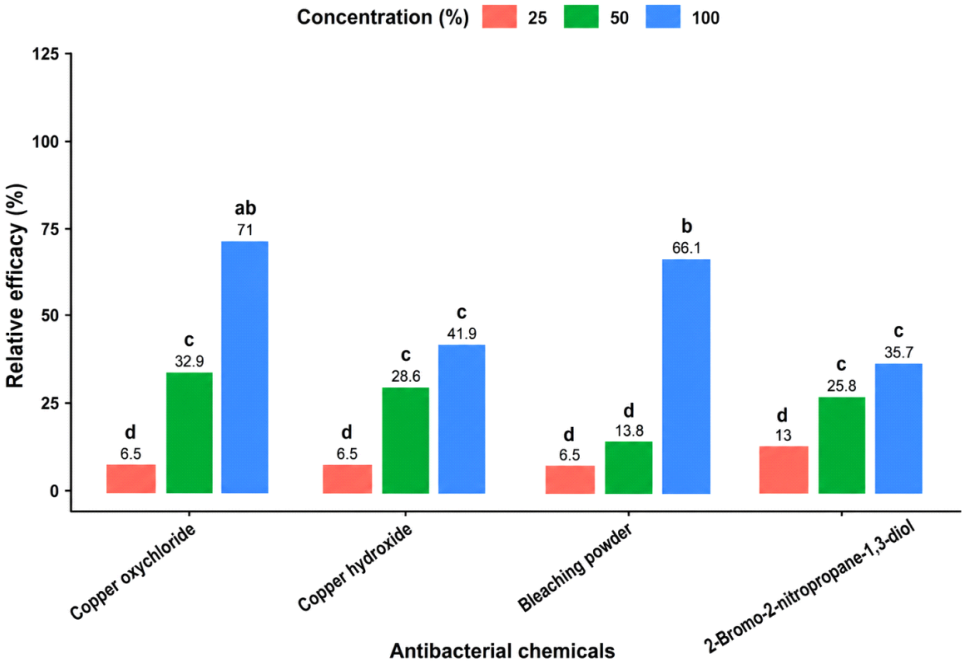


Figure 2. Grouped bar chart showing the relative efficacy of antibacterial chemicals relative to plantomycin @ 250 ppm against *Dickeya zeae* at different concentrations.
Bars represent mean relative efficacy (%) of antibacterial chemicals relative to positive control, plantomycin @ 250 ppm on *Dickeya zeae*. Different lowercase letters indicate significant differences among treatments according to Duncan's Multiple Range Test (DMRT) at P ≤ 0.05

Table 3. Probit EC50 and EC90 values of fungicides (µg/mL/ppm a.i.) against *Fusarium verticillioides* and *Macrophomina phaseolina*

Fungicide	a. i. at field dose (µg/mL)	*F. verticillioides* EC50 (µg/mL)	*F. verticillioides* EC90 (µg/mL)	*M. phaseolina* EC50 (µg/mL)	*M. phaseolina* EC90 (µg/mL)
Propiconazole 25 EC	250.00	<62.5	<62.5	<62.5	>250.0
Tebuconazole 38.39 SC	575.85	<144.0	<144.0	<144.0	312.74
Copper oxychloride 50 WP	1500.00	<375.0	>1500.0	<375.0	>1500.0
Copper hydroxide 46.1 WG	922.00	858.25	>922.0	>922.0	>922.0
Bleaching powder 30%	300.00	>300.0	>300.0	>300.0	>300.0
Thiophanate methyl 70% WP	1050.00	<262.5	<262.5	<262.5	<262.5
Azoxystrobin 18.2% + Difenoconazole 11.4% SC	370.00	<92.5	<92.5	<92.5	188.97
Trifloxystrobin 25% + Tebuconazole 50% WG	300.00	<75.0	<75.0	<75.0	138.66
Captan 70% + Hexaconazole 5% WP	1500.00	<375.0	665.32	<375.0	<375.0
Metiram 55% + Pyraclostrobin 5% WG	900.00	<225.0	>900.0	<225.0	<225.0
Fluxapyroxad 250 g/L + Pyraclostrobin 250 g/L SC	250.00	<62.5	>250.0	<62.5	>250.0

Values preceded by '<' indicate that the fitted EC value lies below the lowest concentration tested (25% of recommended field dose); values preceded by '>' indicate that it lies above the highest concentration tested (100% of recommended field dose). Untagged values fall within the tested concentration range and were interpolated by probit regression (probit of proportional inhibition vs. $\log_{10}$ concentration).

observed at 50 and 100% concentrations, indicating a similar efficacy trend across both pathogens at higher fungicide doses.

Conclusion

Present study successfully identified highly effective chemical treatments against the major pathogens responsible for the post-flowering stalk rot (PFSR) complex in maize under *in vitro* conditions. Among the evaluated fungicides, tebuconazole (38.39 SC @ 1.5 ml/L), thiophanate methyl (70 WP @ 1.5 g/L), azoxystrobin + difenoconazole (18.3 + 11.4 SC @ 1.25 ml/L), trifloxystrobin + tebuconazole (25 + 50 WG @ 0.4 g/L), and captan + hexaconazole (70 + 5 WP @ 2.0 g/L) demonstrated superior antifungal activity and achieved more than 90% mycelial growth inhibition against both *Fusarium verticillioides* and *Macrophomina phaseolina*, even when tested at 50% of their recommended field dosages under *in vitro* conditions. This was further substantiated by probit analysis, wherein EC50 values of these five fungicides against both pathogens consistently fell below 25% of the recommended field dose, confirming their high intrinsic toxicity even at reduced dosages. Beyond confirming potency, this finding carries a practical advantage for growers: it demonstrates that these fungicides retain effective disease-suppressing activity well below their labelled rates, offering scope for dose optimisation that can lower treatment costs, reduce the overall chemical load on the crop and surrounding environment, and help delay the onset of fungicide resistance when deployed within a broader IDM strategy.

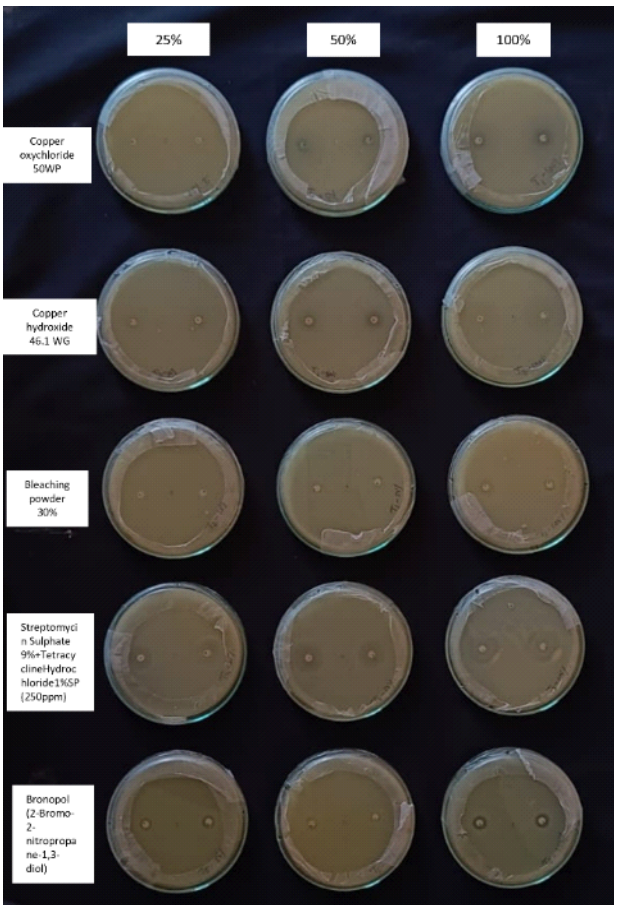


Plate 2. *In vitro* efficacy of select antibacterial chemicals against *Dickeya zeae* (bacterial stalk rot pathogen) by disc diffusion assay

Conversely, for the bacterial pathogen *Dickeya zeae*, copper oxychloride (50 WP @ 3.0 g/L) emerged as the most effective non-antibiotic treatment, achieving 71.0% relative efficacy compared to positive control Plantomycin, Bleaching powder and other evaluated copper compounds proved less effective.

Ultimately, these baseline laboratory findings establish a robust framework for selecting targeted chemical molecules to manage the maize stalk rot complex. However, because *in vitro* efficacy does not always translate directly to the field, further validation under greenhouse and natural field conditions is essential to confirm their performance under ambient disease pressures and to establish optimized, cost-effective integrated disease management (IDM) strategies. As the present findings are based solely on poisoned food and disc diffusion assays, preliminary detached-stalk or ex-vivo tissue assays are recommended prior to field-level deployment recommendations, to additionally assess phytotoxicity and host-tissue penetration of the chemical molecules.

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