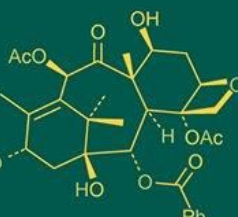
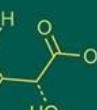
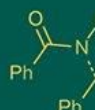
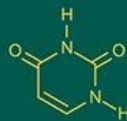
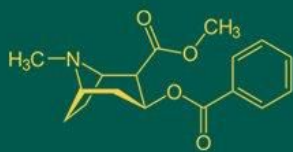


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Survey and *in vitro* efficacy of fungicides and bioproducts against Turcicum leaf blight disease of maize caused by *exserohilum turcicum*

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Abstract

A survey was conducted during the year 2021-2022, in five districts of Manipur, viz Imphal East, Imphal West, Bishnupur, Senapati, and Kangpokpi, to find out the major diseases of maize, and an *in vitro* experiment was also conducted to evaluate the efficacy of different fungicides and botanicals against the major disease, Turcicum leaf blight disease of maize by poisoned food technique. The survey results revealed the widespread occurrence of the disease, with the highest mean disease incidence of 77% and severity of 70% recorded in Senapati district, and the lowest in Imphal West, at 40% and 44%, respectively. *In vitro* evaluation showed that Azoxystrobin 18.2% + Difenconazole 11.4% SC at 0.10% resulted in 100% inhibition of mycelial growth, followed by mancozeb 75 WP at 0.25% (95.23%). Among the bioproducts, *Allium sativum* (Garlic) bulb extract @ 10% was most effective (75.80% inhibition), followed by *Azadirachta indica* (Neem) leaf extract @ 10% (70.86%) and *Trichoderma asperellum* (62.59%). The results indicate that Azoxystrobin + Difenconazole is the most effective chemical option, while garlic and neem extracts offer promising eco-friendly alternatives for integration into sustainable TLB management strategies.

Keywords: *Exserohilum Turcicum*, Turcicum leaf blight, maize, fungicides, bioproducts

Introduction

Maize (*Zea mays L.*) is the third most important cereal crop after wheat and rice in India. It is one of the most versatile emerging crops, having wider adaptability under varied agro-climatic conditions. It is known as the 'Queen of cereal' and staple food crop for human beings and basic raw material for food, feed and industrial products such as oil, protein, starch, beverages, food sweeteners, pharmaceuticals, cosmetics, etc. This crop is being grown throughout the season, i.e. in spring, kharif and Rabi seasons. Due to continuous cultivation of maize, the pressure of insect pests and disease is also increasing due to the continuous availability of the host plant.

It can be grown in various agro-climatic conditions in many parts of India. In India, it occupies the 5th rank in area and 3rd in production and productivity among cereal crops. It is cultivated in an area of 10.04 million ha, with a total annual production of 33.62 million tonnes, giving an average yield of 3.19 tonnes/ha (Agristat, 2022)^[17].

The growing of maize is totally rainfed in the North-east region of India for its food security. It is used for direct consumption, and used as feed, fodder for livestock and for industrial purposes also. In the NER region, the production and production is very low as compared to other states of India; it needs expansion in area, dependent on rainfall, as well as adoption of new hybrids which is resistant or tolerant to pests and diseases. Diseases are one of the major constraints for the reduction in yield. Amongst the diseases such as Turcicum Leaf blight, Maydis Leaf blight, Banded Leaf and Sheath Blight, and Common rust. The Turcicum Leaf Blight of Maize, which is also known as Northern corn leaf blight, caused by *Exserohilum turcicum*, is a major disease of maize that affects the production and productivity of maize (Nongmaithem *et al.*). It can reduce the yield up to 90% under conducive environmental conditions. Therefore, a survey and *in vitro* evaluation of fungicides and a bioproduct study

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were conducted for sustainable management and for incorporation in the Integrated disease management of Turcicum Leaf Blight diseases of Maize.

Materials and Methods

Survey for Incidence and Severity of Turcicum Leaf Blight Disease of Maize

A random survey was conducted in five districts of Manipur, namely Imphal East, Imphal West, Senapati, Bishnupur and Kangpokpi districts to identify the major diseases occurring in the state of Manipur. Before conducting the survey, preliminary information was gathered from the Krishi Vigyan Kendra about the major maize-growing district in Manipur. And the survey was conducted for three consecutive years during the kharif season from 2021-2023.

The survey was conducted randomly by selecting the field at an interval of 10 km in all the districts. In each district, 3

villages were selected and identified, and five fields were surveyed by selecting 100 plants randomly, varying the stage of crop from knee high to grain filling stage. The incidence of the disease was calculated by using the formula given by Nawanosike *et al.* (2015).

$$\text{Disease incidence} = \frac{\text{Number of diseased plant}}{\text{Total number of plant examined}} \times 100$$

The disease severity was calculated by using a 1 to 9 disease rating scale, given by Chung *et al.* (2010)^[3]; Mitiku *et al.* (2014)^[14] (Table 1). The Percent Disease Index (PDI) was calculated by using the formula given by Wheeler (1969). The individual scoring of each plant was converted into PDI as described by Hooda *et al.* (2018)^[8].

$$\text{PDI} = \frac{\text{Sum of individual ratings}}{\text{No. of plants examined}} \times \frac{100}{\text{Maximum disease rating}}$$

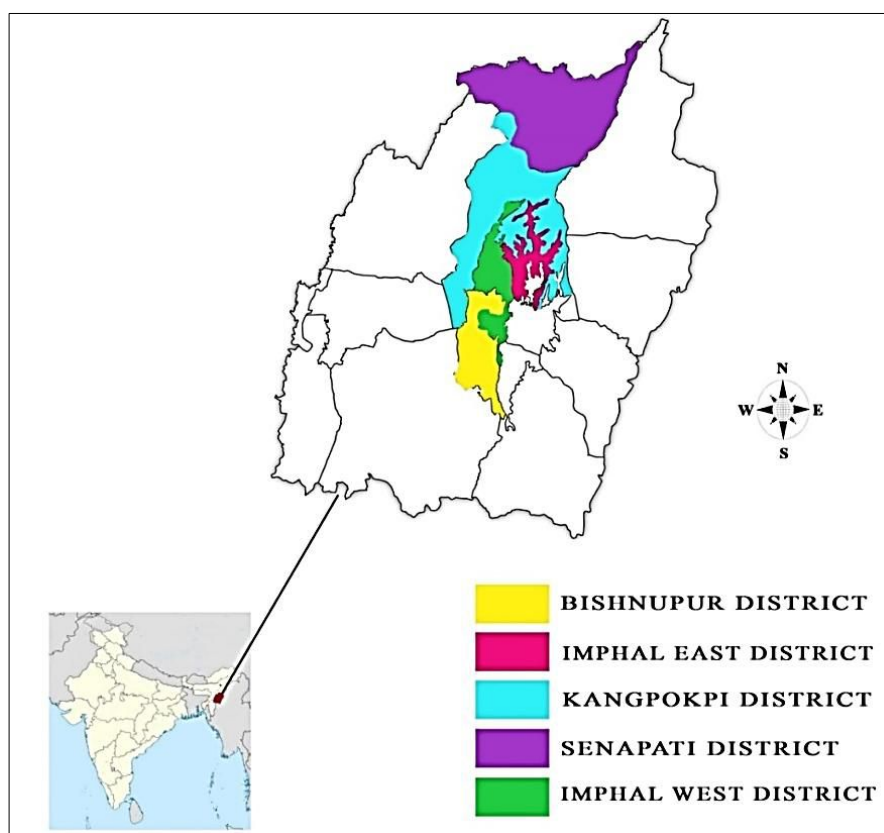


Fig 1: Survey district of Manipur

Isolation and purification of the pathogen

The isolation of the plant pathogen was done by using the method described by Manamgoda *et al.* (2012)^[12]. Initially, the leaf samples were cut into small pieces of 0.5x0.5cm in diameter containing both healthy and infected-looking tissues and sterilized in 1% sodium hypochlorite solution, followed by washing in three changes of sterile distilled water. After removing the excess water, the cut portions were aseptically transferred to sterile petriplates containing 20 ml of solidified potato dextrose agar and kept in an incubator at 25±1 °C for 5 days. After 5 days, a bit of mycelium from the actively growing portion was transferred aseptically to new, fresh PDA petridishes. After purifying, the culture was transferred to a PDA test tube for further use.

In-vitro evaluation of fungicides and bioproducts

The *in vitro* efficacy of fungicides and bioproducts was performed by using the poisoned food technique on Potato agar medium described by Vincent (1947)^[15]. For *Trichoderma asperellum*, the *in vitro* evaluation was conducted by the dual culture method described by Dennis and Webster (1971)^[5]. The efficacy was evaluated by calculating the percent inhibition of mycelial growth over the control.

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

Where

I = Fungal growth inhibition in per cent

C=Colony diameter in control plates in cm

T=Colony diameter in Treatment plates in cm

Table 1: The details of treatment for *in vitro* studies

Sl. No.	Treatment
1.	Mancozeb 75 WP@ 0.25%
2.	Azoxystrobin 18.2w/w + Difenoconazole 11.4% w/w SC@ 0.10%
3.	<i>Azadirachta indica</i> (Neem) leaves @ 10%
4.	<i>Allium sativum</i> (Garlic) bulb@ 10% @
5.	<i>Lantana camara</i> @ 10%
6.	Cow urine @ 20% @
7.	<i>Trichoderma asperellum</i> @ 1%
8.	Control

Poisoned Food Technique

To obtain the required concentration of fungicides and bioproduct, the required quantity of fungicides and bioproduct was added to molten, lukewarm potato dextrose agar medium. After that, 20 ml of poisoned medium was poured into sterile petri dishes and allowed it solidify. with the help of a sterile 6mm corkborer, a mycelial disc was cut from the periphery of culture plates of the fungus and then kept in the centre of 90mm Petri plates. Plates with no fungicides or bioproducts act as control plates. Three replications were maintained for each treatment. And kept at an incubator at a temperature of $28 \pm 1^\circ\text{C}$. The radial growth

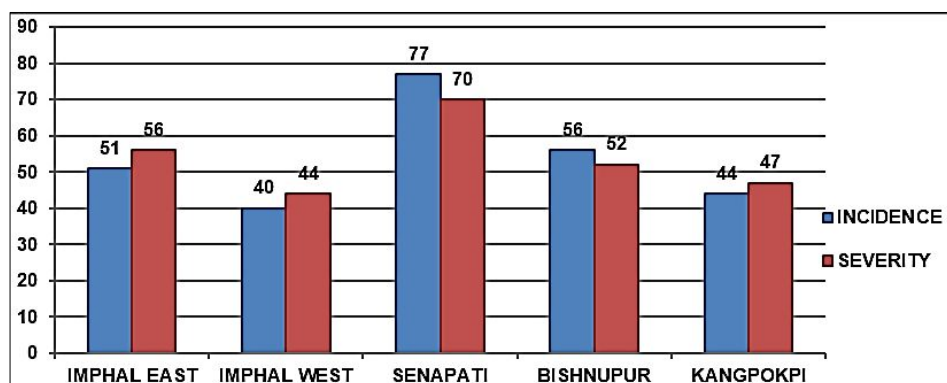
of each treatment was recorded when the control plates were fully grown.

Results and Discussion

Survey for Incidence and Severity of Turcicum Leaf Blight Diseases of Maize

A random survey was conducted in five major maize-growing districts of Manipur to know the incidence and severity of Turcicum leaf blight disease of maize, namely Imphal East, Imphal West, Senapati, Bishnupur and Kangpokpi districts of Manipur for three consecutive years during the kharif season from 2021-2023.

During the survey, a total of 50 maize fields were surveyed, 10 from each district. The results of the study indicated that Turcicum leaf blight diseases were widespread in all the districts surveyed. The mean highest disease incidence (77%) and severity (70%) were found in the Senapati district. The disease incidence was followed by Bishnupur (56%), Imphal East (51%), and Kangpokpi district with 44%. The lowest mean disease incidence was recorded in Imphal West district, with 40%. Similarly, the disease severity by Imphal East (56%), Bishnupur (52%), and Kangpokpi with 47%. The lowest mean disease incidence was found in Imphal West District with 44% disease severity (Graph 1).

**Graph 1:** Showing the mean disease incidence and severity of Turcicum leaf blight disease of maize in five surveyed districts.

In-vitro Screening of Fungicides and Bioproducts

The *in vitro* Screening of Fungicides and Bioproducts was Done by the Poison Food Technique. The efficacy was evaluated by calculating the percentage inhibition of the mycelial growth over the control. The results showing the percentage inhibition of growth of *Exserohilum turcicum* by fungicides and bioproducts are shown in Table 2. Among the fungicides and bioproducts tested, Azoxystrobin 18.2 w/w + Difenoconazole 11.4% w/w SC @ 0.1% was found to be the most effective and recorded 100% radial growth inhibition of the test pathogen. *Allium sativum* (Garlic) bulb @ 10% was also recorded highly promising with 75.80% inhibition of mycelial growth of test pathogen, i.e. *Exserohilum turcicum*, followed by *Azadirachta indica*

(Neem) leaves @ 10% (70.86%) and *Trichoderma asperellum* (62.59%). The lowest inhibition of mycelial growth was recorded in *Lantana camara* @10% (55.85%). These findings are in corroboration with Manu *et al.* (2017) [11], who studied that under *in-vitro* efficacy against *Exserohilum turcicum*, the mean highest per cent inhibition of mycelial growth was recorded in Garlic bulb extract, with 79.63% growth inhibition.

Bhatt and Kumar (2019) [12] reported that 8 fungicides were tested under *in vitro* conditions against *Exserohilum turcicum* of Maize and found that Azoxystrobin 23%, showed the highest inhibition of 92.22% mycelial growth at a 5.0 ppm concentration.

Table 2: *In vitro* efficacy of fungicides and bioproducts on the mycelial growth inhibition of *Exserohilum turcicum*

Treatment	Per cent inhibition of mycelial growth (cm)
Mancozeb 75 WP@ 0.25%	95.23
Azoxystrobin 18.2w/w + Difenoconazole 11.4% w/w SC@ 0.10%	100.00
<i>Azadirachta indica</i> (Neem) leaves @ 10%	70.86
<i>Allium sativum</i> (Garlic) bulb@ 10% @	75.80
<i>Lantana camara</i> @ 10%	55.85
Cow urine @ 20% @	56.91
<i>Trichoderma asperellum</i> @ 1%	62.59
Control	0.00
SEm±	0.13
CD(P=0.05)	0.38
CV%	1.39

Conclusion

The survey confirmed the widespread occurrence of Turcicum leaf blight across Manipur, with the highest incidence and severity in the Senapati district. *In vitro* studies revealed that Azoxystrobin + Difenconazole was the most promising fungicide, which found 100 per cent mycelial growth inhibition of *E. turcicum*, causing Turcicum leaf blight disease of maize. Among bioproducts, garlic and neem extracts exhibited strong antifungal activity and can be incorporated into integrated disease management strategies to reduce chemical dependence. Further field evaluation of these promising treatments is warranted to validate their effectiveness under natural conditions.

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