RVSKVV / Thesis / M.Sc. . No. 1575

Study and management of Pearl millet blast incited by *Pyricularia grisea* (Cooke) Sacc.

THESIS

Submitted to the

Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya

In partial fulfillment of the requirements for the Degree of

MASTER OF SCIENCE

In

AGRICULTURE

PLANT PATHOLOGY

By

PRERANA PARIHAR

Department of Plant Pathology
Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya
College of Agriculture, Gwalior (M.P.)

2018

CERTIFICATE – I

This is to certify that the thesis entitled, **"Study and management of Pearl millet blast incited by *Pyricularia grisea* (Cooke) Sacc."** *submitted in partial fulfillment of the requirements for the Degree of* **MASTER OF SCIENCE** *in* **Agriculture** *department of* **Plant Pathology** *of* **Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior** *is a record of the bona-fide research work carried out by* **Miss Prerana Parihar**, **ID. No. 16111807** *under my guidance and supervision. The subject of the thesis has been approved by the Student's Advisory Committee and the Director of Instruction.*

No part of the thesis has been submitted for any other degree or diploma or has been published. All the assistance and help received during the course of the investigations has been acknowledged by the scholar.

Place : Gwalior **Dr. R.K. Pandya**

Date : …….. **Chairman of Advisory Committee**

MEMBER OF STUDENT'S ADVISORY COMMITTEE

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Member (Mrs. Rajni Singh Sasode) : …….……

Member (Dr. A.K. Singh) : …….....…..……..

Member (Dr. V. B. Singh) : …….....……..

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CERTIFICATE –II

This is to certify that thesis the entitled, **"Study and management of Pearl millet blast incited by *Pyricularia grisea* (Cooke) Sacc."** *submitted by* **Miss Prerana Parihar, ID.No. 16111807** *to the* **Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior** *in partial fulfillment of the requirements for the degree of* **Master of Science** *in* **Agriculture** *in the Department of* **Plant Pathology** *has been accepted after evaluation by the external examiner and approved by the Student's Advisory Committee after an oral examination of the same.*

Place : Gwalior **Dr. R.K. Pandya**

Date : …….. **Chairman of Advisory Committee**

MEMBERS OF THE ADVISORY COMMITTEE

Chairman (Dr. R.K. Pandya) : ……..…….……

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Member (Dr. V. B. Singh) : …….....……..

Head of the Department : ……..…....

Dean, Collage of Agriculture, Gwalior : …..……...

Director of Instruction : ……

Contents

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

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Place: **Gwalior**

Date :

(Prerana Parihar)

Chapter - I

INTRODUCTION

Pearl millet (*Pennisetum glaucum*) belongs to the family *Poaceae,* subfamily *Panicoideae* (ICRISAT, 2006). Millet ranks as the sixth most important grain in the world and sustains one third of the world's population and it is an important part of the diet in former Soviet Union, Africa, India and Egypt (Railey, 2006). 100 grams of bajra has the following nutritional values: energy 360 calories, moisture 12 g, protein 12 g, fat 5 g, mineral 2 g, fiber 1 g, carbohydrate 67g, Calcium 42mg, phosphorus 242mg, and iron 8mg. It is an important grain and fodder crop of Africa and Indian subcontinent. In India it is popularly known as bajra grown in the area having an average rainfall of 150-1000 mm. Our country is the largest producer of this crop in Asia occupies an area of 6983 thousand ha with the production of 8057 million tones and the productivity is 1154 kg/ha during 2015-16. (Anon.2015-16). Rajasthan, Gujarat, Maharashtra, Madhya Pradesh, Uttar Pradesh, Haryana, Karnataka, Andhra Pradesh and Telangana are the major Pearl millet growing states of the country. In Madhya Pradesh during 2015-16 the crop was cultivated in an area of 267 thousand ha with an annual production 618 million tones and productivity is 2315 kg/ha.(Anon. 2015-16). Northern region of Madhya Pradesh comprising Morena, Bhind and Gwalior is the major Pearl millet growing area of the state; as these three districts jointly contribute more than 70 % share in area and production of this crop in the state.

The crop is affected by number of diseases such as blast, downy mildew, smut, rust, and ergot etc. Out of these blast has become a major biotic constraint of Pearl millet particularly in the crop cultivated for fodder purposes. Blast incited by *Pyricularia grisea* (Cooke) Sacc. was first identified in Uganda in 1933 (Emechebe,1975) and was first reported in India from Kanpur 1953 (Mehta et al.,1953) although blast was considered a minor disease of pearl millet in India, the disease incidence has increased alarmingly during the recent years (Lukose *et.al.*, 2007, AICPMIP 2009). The disease appears as grayish, water-soaked lesions on foliage that enlarge and

become necrotic, resulting in extensive chlorosis and premature drying of young leaves.

The lesion size varies from small, roundish, elliptical, diamond shaped to elongated, measuring 1-2 mm to 20 mm. Lesions are often surrounded by a chlorotic halo, which turns necrotic, giving the appearance of concentric rings. Leaf blast on pearl millet has been found to be negatively correlated with green-plot yield, dry matter yield and digestive dry matter (Wilson and Gates, 1993) thus affecting the productivity and quality of the crop (Thakur et al., 2011).

Several management strategies have been proposed and evaluated to minimize the blast disease incidence. Cultural practices, host plant resistance, use of fungicides and botanicals are the four strategies adopted to control pearl millet blast. The disease can be managed by the cultivation of resistant/ tolerant genotypes as the use of resistant cultivars is the most economical and eco-friendly method for the management of disease. The management of the pathogen can also be managed by the use of chemicals. A number of botanicals possessing fungicidal properties hence they may be tested under *in vitro* and thereafter under *in vivo* condition with the possibilities to get alternative to the hazardous chemicals in the direction of an environment friendly management.

Keeping the above facts in mind the present studies are proposed of following lines:

1. To find out the status of Pearl millet blast in Northern Madhya Pradesh.
2. *In vitro* evaluation of culture media to find out the most suitable medium for the growth of *Pyricularia grisea.*
3. *In vitro* evaluation of botanicals and chemicals against *Pyricularia grisea*.
4. Evaluation of hybrids, varieties, locally collected and other pearl millet material against blast.
5. Field evaluation of selected chemicals and botanicals for the management of Pearl millet blast.

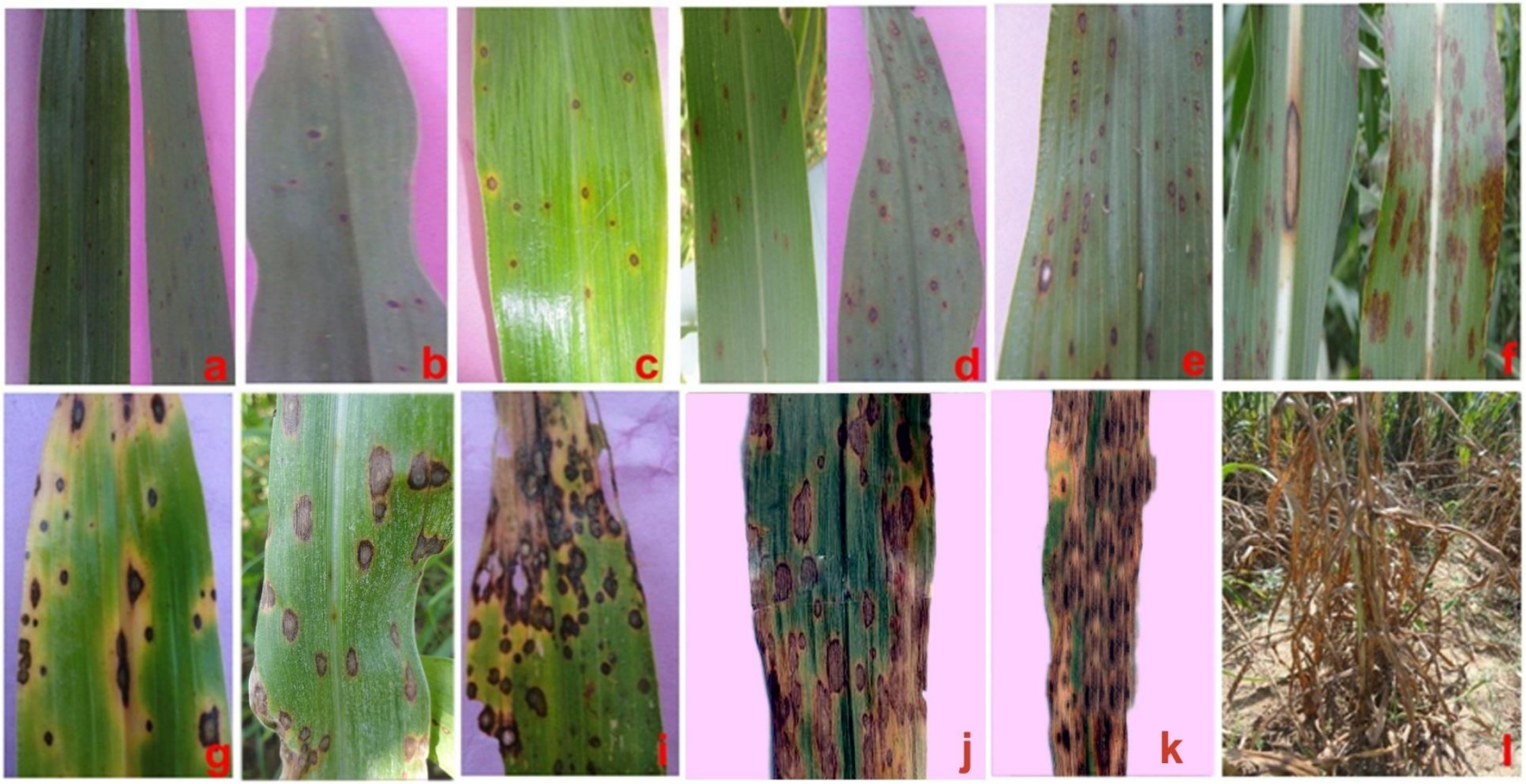

Plate1-(a)Minute circular to slightly linear dark brown specks on the leaf surface,(b-e) Dark brown specks gradually increase in no. and size and become elliptical to irregular.(f-h) Lesion with dark brown margin and light greyish centre.(i-j) Overlapping of lesion with necrotic holes.(k) Lesion collapse and cover whole leaf. (l) Severely infected plant .

Plate 1 - Progressive development of blast on leaves of pearl millet.

Chapter - II

REVIEW OF LITERATURE

The available literature of work done on blast disease of Pearl millet and it's management strategies have been reviewed in this chapter. The review of literature pertaining to the respective objectives are presented in the following headings and sub-headings.

2.1. CAUSAL ORGANISM

The genus *Pyricularia grisea* (Cooke) Sacc. was established by Saccardo (1880) which was originally described from crabgrass (*Digiteria sanguinalis* L.). The name "*Pyricularia*" refers to the pyriform shape of the conidia.

The fungus *Pyricularia grisea* (Cooke.) Sacc. (formerly *Pyricularia oryzae* Cavara.) anamorph of *Magnaporthe grisea* (Hebert) Barr. is a heterothallic, filamentous fungus pathogenic to almost 50 plant species in 137 members of Poaceae (Ou, 1980; Choi *et al.,* 2013; Murakami *et al.,* 2000; Inukai *et al.,* 2006) including *Eleusine*. The perfect stage of *Pyricularia grisea* was earlier named as *Ceratosphaeria grisea* (Hebert, 1971). Later Yaegashi and Nishihara (1976) suggested the genus *Magnaporthe*. Yaegashi and Udagawa (1978) finally proposed *M. grisea* as the perfect stage of *P. grisea* (Cke.) Sacc. instead of *Ceratosphaeria grisea*.

Mycelium in cultures is aerial or submerged, hyaline or olivaceous, 1.5 – 6.0 µm in width, septate branched, conidiophores one to many, fasciculate, simple or rarely branched.

The fungus *M. grisea* produces three-celled, pyriform macro conidia in the imperfect stage and four celled, spindle-shaped ascospores in the perfect stage (Barr, 1977; Hebert, 1971; Kato *et al.,* 1976).

SYSTEMATIC POSITION:

Kingdom	**:**	**Fungi**
Phylum	**:**	**Ascomycota**
Class	**:**	**Sordariomycetes**
Order	**:**	**Magnaporthales**
Family	**:**	**Magnaporthaceae**
Genus	**:**	***Pyricularia*(Anamorph)**
		Magnaporthe (Teleomorph)
Species	**:**	**G*risea***

2.2 SYMPTOMS

Padmanabhan (1974) reported that the lesions or spots first appear as minute brown specks, then grow to become spindle shaped pointed at both ends, several cm long and about 0.5 – 1.0 cm wide. The centre is greenish grey often showing a brownish margin. The size, colour and shape of the lesions, however, vary with different climatic conditions and also varietal response. Under favourable conditions on a susceptible cultivar several greyish spots may appear, become larger and broader and coalesce, leading to withering of the whole leaf. (Manibhushan Rao,1994)

2.3 Disease survey

Devda (2009) observed pearl millet blast as an important biotic constraint in Northern Madhya Pradesh and reported that the average severity of blast in Morena, Bhind and Gwalior was 5.5, 3.8 and 5.4 percent respectively.

Yadav *et al.* (2012) surveyed the pearl millet fields of Morena, Bhind and Gwalior and reported 11.53, 13.40 and 11.28 per cent severity of blast respectively.

2.4 Growth of the pathogen in different media.

Sun *et al.,* (1989) studied the effects of 17 media on 41 isolates of *P. oryzae*. They found that, corn meal and rice straw agar media were most conducive for sporulation.

Awoderu *et al.* (1991) observed that linear growth of *P. oryzae* in Potato dextrose agar medium, while conidial production was greatest on 1 per cent soluble starch yeast extract agar.

Arun kumar and Singh (1995) found that, maximum colony diameter of rice isolate of *P. grisea (M. grisea)* occurred on malt extract agar and Leonin agar. Du Xinfa *et al.* (1995) stated that, Pyricularia isolates from hosts including rice and common weeds in paddy fields sporulated abundantly on sterilized barley or sorghum grains.

Mijan Hossain (2000) observed that among the non synthetic media, potato dextrose agar supported maximum radial growth (85.00 mm), next was host extract + 2 per cent sucrose agar medium (80.33 mm) followed by oat meal agar (75.00 mm). Cruz *et al.* (2009) observed the higher sporulation on wheat meal culture medium in alternate light, dark regime.

Srivastava *et al.* (2009) conducted an experiment to develop and evaluate cultural characteristics of *P. grisea* isolates from *Eleusine coracana* (finger millet) on media derived from rice and finger millet viz. rice leaf agar, ragi leaf agar and ragi flour agar. Results revealed that ragi flour media recorded better growth and sporulation of *P. grisea* at 28 C and pH 7.5 than other three media.

Mahdieh S (2013) reported that PDA medium could provide the best medium for *P.oryzae* vegetative growth, regardless of light condition. However, *P. oryzae* could sporulate when light was provided either continuously or at intervals. A combination of 16/8 hr light/darkness intervals and adding rice materials to culture media could induce *P. oryzae* for better sporulation.

Malviya R. (2014) tested four media namely Potato dextrose agar, Czapek’s Dox Agar, Rechard Agar medium and Corn meal agar. However,

PDA was found to be highly suitable for the growth and sporulation of *P.grisea*.

2.5 Evaluation of botanicals and chemicals against *Pyricularia grisea*.

Rodriguez *et al.* (1993) found that seed treatment with Beam 75% WP (tricyclozole) reduced leaf blast at least 50 per cent compared to control.

Ram Singh and Dodan (1994) reported that, tricyclazole and propiconozole (both @0.1%) were the most effective in reducing neck blast and increasing the yield.

Saifulla (1994) achieved best disease control and yield with tricyclozole and blasticidin.

Thangavelu *et al.* (1995) studied the effect of different neem-based formulations on rice blast disease both *in vitro* and *in vivo*. Replin RD9 was found superior to all the test compounds that reduced the per cent germination of spores *in vitro*. In field also replien RD9 was found superior to all other test compounds.

Enyinnia (1996) observed that, tricyclazole (Beam) and benomyl (Benlate) suppressed foliar and neck blast development, but tricyclazole was superior to benomyl.

Ganguly (1994) studied nine botanicals viz. *Vinca rosea, Catharanthus roseus, Latana camara, Ocimum tenuiflorum (O. sanctum), Solanum melongena (Aubergines), Azadirachta indica, Polyalthia longifolia, Aegle marmelos* and *Datura metel* showed antifungal activity against *Pyricularia oryzae(Mangnaporthe grisea)* and *Heliminthosporium oryzae (Cochliobolus miyabeanus)* under laboratory condition and found that *P. longifolia* and *A. indica* were the most effective.

Tripathi (2000) reported that seed treatment with Carbendazim @ 4g./kg. followed by one foliar spray with this fungicide @ 0.05 at tillering and Corotop 205 G @ 30 kg/ha at panicle initiation stage, was found to be, the best for blast control (39.20%) and increasing the yield (31.81%).

Amadioha (2000) reported that water and ethanol leaf extracts and oil extract of seeds of *Azadirachta indica* (Neem) significantly reduced the *in vitro* radial growth of *P. oryzae* and the development and spread of blast disease in rice plants in the greenhouse.

Garcia Fernandez (2001) found that yield losses caused by *Pyricularia oryzae* can be managed by the use of resistant cultivar and the application of Tricyclazole, Tebuconazole, Isoprothiolane, Kasugamycin, Prochloroz, and Carbendazim+Flusilazole.

Joshi (2002) conducted field experiment in Maharashtra, India during the seasons of 1998-2000 to determine the efficacy of Tricyclazole in controlling *Pyricularia grisea* causing blast disease in rice and evaluated its effect on rice yield. Beginning one month after planting, 3-week old seedling of rice cv. RTN-711 were sprayed with 0.05,0.06 and 0.12% Tricyclazole at fortnightly intervals, along with Mancozeb or Carbendazim and no spray as control. All 3 concentration of Tricyclazole were significantly superior in reducing disease intensity. There was a liner relationship between disease intensity and yield.

Prajapati *et al.*, (2004) found that Tricyclazole proved significantly superior in decreasing the leaf and neck blast by 62.9 and 64.1% respectively, with corresponding increase of 72.3% in grain yield over the control and was at par with Carbendazim 50 WP.

Sundravadana *et al.*, (2008) *in vitro* study reveals that there was no phytotoxicity effect at different concentrations of Azoxystrobin. The reduction of blast incidence and yield increased curve obtained showed flattening between the range 125, 250 and 500 g a. i/ha , hence the optimum rate of Azoxystrobin was fixed to be at 125 g a. i/ha for the control of blast disease.

Varma and Santha Kumari (2012) *in vitro* study was conducted to evaluate the efficacy of five fungicides against rice blast pathogen, *Magnaporthe oryzae*, by poisoned food technique. The fungicides were Isoprothiolane 40% EC (Fuji-one) at 1, 1.5 and 2.0 ml, carpropamid 27.8% SC (Protiga) at 0.5, 1.0 and 2.0 ml/l, Carbendazim 50% WP (Bavistin) at 0.75, 1.0 and 1.5 g/l, Tricyclazole 75% WP (Beam) at 0.1, 0.6 and 1.0 g/l and

Propiconazole 25% EC (Tilt) at 0.5, 0.75 and 1.0 ml/l. Isoprothiolane at 1.5 ml/l recorded the maximum inhibition of mycelial growth (94.85%), followed by Carpropamid at 1.0 ml/l (91.48%).

Hajano Jamul-U-Ddin *et al.* (2012) evaluated five fungicides, three botanicals and six bio-agents. Only Mancozeb appeared as the highly effective fungicide that completely inhibited the mycelial growth of the fungus. Among botanical only higher dose of garlic (*Allium sativum* L.) completely inhibited the mycelial growth of the test fungus. Among biocontrol agents maximum mycelial inhibition of *P. oryzae* was provided by *Paecilomyces lilacinus* followed by *Trichoderma* spp.

Kunova *et al.* (2013) reported that the mycelial growth of *Magnaporthe oryzae* was inhibited at lower concentrations of azoxystrobin and relatively high concentrations of tricyclazole, while sporulation was more sensitive to both fungicides and was affected at similarly low doses. This study presents for the first time detailed azoxystrobin and tricyclazole growth response curves for *M. oryzae* mycelium growth and sporulation. The maximum inhibition (93.30%) of *P.grisea* was obtained in the treatment of mancozeb 75 WP .

Sireesha and Venkateswarlu (2013) evaluated five plant part extracts namely Neem seed kernel extract, Neem oil, *Asafoetida spp.* and *Pongamia sp*. extracts and Panchagavya against blast of rice in *in vitro* conditions. The results concluded that the Neem seed kernel showed a significantly more mean suppression value.

Nasruddin and Amin (2013) reported that Difenoconazole and Difenoconazole+Propiconazole were evaluated against the rice blast disease and found effective in suppressing blast and protecting yield as compared to the other tested fungicides.

Naik *et al.* (2014) reported that, mancozeb was on par with another combiproduct; captan 70 + hexaconazole 5 (93.17%) as well as copper oxychloride (89.41%) whereas it was found to be significantly superior over the combi-product; tricyclozole18 + mancozeb 62 (87.38%) and chlorothalonil 75 WP (83.81%) across different concentrations.

Netam *et al.*, (2014) reported that Complete inhibition of mycelial growth was given by Tricyclazole followed by Ediphenphos and Mancozeb 65 per cent WP+Carbendazim12 per cent WP under laboratory condition. *In vivo*, Ediphenphos and Tricyclazole were found effective in controlling leaf, neck and finger blast.

Pal (2014) studied six fungicides like Kresoxim methyl, Azoxystrobin, Propiconiazole, Trifloxystrobin +Tebuconazole (Nativo), Difeconazole, and Tricyclazole to control the leaf blast of rice. Among them Trifloxystrobin + Tebuconazole (Nativo) was found to be a highly effective.

Hubert *et al.,* (2015) evaluated the effect of aqueous extracts of *Aloe vera*, *Allium sativum*, *Annona muricata*, *Azadirachta indica*, *Bidenspilosa*, *Camellia sinensis*, *Chrysanthemum coccineum*, processed *Coffee arabica*, *Datura stramonium, Nicotiana tabacum* and *Zingiber officinalis* for control of rice blast disease (*Pyricularia grisea*) *in-vitro* and *in-vivo*. The results indicate that processed *C. Arabica* at 10% and 25% (v/v) had the highest (81.12%) and (89.40%) inhibitory effect, respectively, against *P. grisea*. Aqueous extract from *N. tabacum* at 10% concentration ranked third (80.35%) in inhibiting *P. grisea*. These were followed by extracts from 25% *A. vera* (79.45%) and 25% *C. coccineum* flower (78.83%).

Joshi and Gohel (2015) carried out an *in vitro* study using poison food technique by evaluating nine fungicides and two antibiotics at three concentrations against *Pyricularia grisea.* Among the tested fungicides, propiconazole, mancozeb, tricyclazole and carbendazim (12%) + mancozeb (63%) at all the three concentrations completely inhibited the mycelial growth of the pathogen against blast of pearl millet. Experiment under field condition revealed that two foliar sprays at an interval of 15 days commencing from the first initiation of disease with tricyclazole, 0.05 per cent or iprobenfos, 0.1 per cent or isoprothiolane, 0.05 per cent was most effective in reducing blast intensity.

Pandey (2015) reported that Aqueous extract of leaves of *Azadirachta indica, Emblica officinalis, Pongamia glabra* and *Acacia nilotca* were tested *in-vitro* at 0.2% and 0.5% concentration using poisoned food technique for

antimicrobial activity against mycelial growth of *Magnaporthe oryzae* causing leaf blast and *Bipolaris oryzae* causing brown spot in rice. The leaf extracts were found significantly effective in reducing mycelial growth of the pathogens. The result reveals that *A. indica* leaf extract @ 0.5% was found most effective in minimizing the mycelial growth of both the pathogens 28.35 mm and 27.12 mm, closely followed by *P.glabra* leaf extract 29.57 and 30.10 mm in the same concentration, 96 hrs after incubation.

2.6 Evaluation of hybrids varieties, locally collected and other pearl millet material against blast.

One thirty five pearl millet lines were evaluated against blast at Gwalior and reported blast severity in the range of 0 - 47.5%. The result reveals that only one entry MH 1541 remained completely free from blast while its maximum severity i.e. 47.5% was recorded in MH 1513. Devda (2009)

Thakur *et al.* (2009) developed a field screening technique for blast disease caused by *P. grisea.* The field screening technique involved the use of a highly susceptible line as an infector row grown after every four test rows, artificial spray inoculation of 30-days-old plants using *P. grisea* spore suspension and maintaining high humidity through irrigation for 2 weeks following inoculation. In all, 211 elite hybrid parental lines, including 126 designated B-lines, 20 designated R-lines and 65 potential R lines were evaluated for blast resistance in the disease nursery. Fourty-five line identified as blast resistant (score ≤3.0 on 1–9 scale) were further screened through greenhouse screening technique.

Nagaraja *et al.* (2010) evaluated core set of 520 finger millet accessions for blast resistance under prevailing weather conditions in field revealed that the incidence of neck and finger blast decreased significantly with increased temperature from 23.9 to 27.0°C and reduced rainfall from 303 to 83.4 mm during flowering period, however, the RH remained almost constant (88.34 to 88.90 %).

Gupta *et al.* (2012) conducted tests under field condition. on the basis of foliar blast reaction he selected two resistant restorer lines (ICMR 06222 and ICMR 07555) and two susceptible maintainer lines (ICMB 95444 and

ICMB 89111). Each of the two resistant parents was crossed with two susceptible parents to generate 4 sets of F1s, F2s and their backcrosses with both resistant and susceptible parental lines. These were evaluated for disease reaction with artificial inoculation under field conditions. The disease reaction of the F1s, and the segregation patterns of resistance in the F2s and backcross generations, showed that resistance to foliar blast in pearl millet is controlled by a single dominant gene.

To know the virulence pattern of pearl millet blast pathogen *M. grisea* Sharma *et al.* (2013) 25 isolates were collected from four major pearl millet growing states in India i.e., Rajasthan, Haryana, Maharashtra and Uttar Pradesh on ten pearl millet genotypes viz., ICMB 02444, ICMB 02777, ICMB 06444, ICMB 93333, ICMB 96666, ICMB 97222, ICMB 99444, 863B, ICMR 06222 and ICMB 95444 under green house conditions. Differential reactions to the test isolates were recorded on ICMB 02444, ICMB 93333, ICMB 97222, 863B and ICMR 06222.

Khadka *et al.*,(2013) tested five landraces Acc4337, Acc6202, Acc456, KLB184, Kabre and four exotic lines, PR202, HR911, SPFMK2, and GPU48 against *Pyricularia grisea* which have been used in national finger millet research programme by Hill Crop Research Programme (HCRP), Kabre, Dolakha. Among the tested lines Gpu48, showed resistant reaction and Acc4337 showed susceptible reaction in artificial inoculation.

Yadav *et al.* (2013) evaluated twenty five promising pearl millet hybrids and varieties against blast. The minimum severity of 7.5% was recorded in PB 106, GHB-744, and GHB-732,while its maximum severity (32.50%) was recorded in B-2301, PB 106, GHB 744 and GHB-732.

G. Prakash *et. Al.* (2016) evaluated 15 inbred lines in which three entries were highly resistant viz., PPMI 1087, PPMI 1089 and PPMI 660 and two entries (PPMI 1084 and J 108) were identified as resistant. This method will be useful for large scale screening of pearl millet entries against blast pathogen.

Chapter – III

MATERIAL AND METHODS

The investigation has been carried out in the Department of Plant Pathology R.V.S.K.V.V. Gwalior (M.P.) during kharif 2017. The present studies on pearl millet blast were focused on screening of material /cultivars and management aspects. The material used and methods followed are described below:-

Climate

The climate of Gwalior is subtropical. The rainy season normally starts from middle of June after commencement of south-west monsoon and last up to September. Maximum precipitation of rains occurred in the month of July and August. Winter season runs from November to mid-February and hot summer season from April to mid-June. October is the transitory month between rainy and winter season.

Soil

The soil of experimental site was alluvial clay loam texture. The available nitrogen, phosphorus and potassium content of soil was 200.7 kg, 10.1 kg and 260.9 kg per hectare, respectively low, medium low to medium, respectively. The water holding capacity of soils is also medium.

Experimental site

The present studies were conducted at Research Farm, College of Agriculture, Gwalior during kharif season of 2017. Gwalior is situated in Northern part of Madhya Pradesh at an elevation of 211.52 meters from mean sea level and lies between latitude and longitude of 26º14' North and 78º15' East, respectively.

Material

3.1 Equipments and apparatus

The equipments and apparatus which have been used in the study are given below:- Laminar air flow, BOD incubator, Refrigerator, Autoclave, Glassware, Microscope, Hot air oven, pH meter, Electronic balance, Forceps, Inoculation Needle, Cork borer, Blade etc.

3.1.1 Chemicals

The chemicals which have been used in the study are given below:-

Agar–Agar, Dextrose, Di Potassium hydrogen Phosphate, Magnesium sulphate, Sodium nitrate, Potassium chloride, Potassium nitrate, Ferrous sulphate, Sucrose, Potassium monobasic phosphate, Ferric chloride.

3.2 Cleaning and sterilization of equipments

Corning make glassware were used during the period of investigation. All the glassware were cleaned with chronic acid, followed by thorough washing with detergent powder and then rinsing tap water before use. The sterilization of media was done at 15lbs, pressure for 20 min. Petriplates were sterilized in hot air sterilizer at 180°C for 2 hrs. The plastic petriplates used in bio control study, were sterilized by alcohol. The isolation chamber was sterilized by alcohol, followed by ultraviolet exposure for 20 min. The other equipments used in isolation chamber like forceps, inoculation needle, cork borer, blade, etc. were sterilized by dipping them in alcohol, followed by heating on flame.

3.2.1 Sterilization procedure

A. Sterilization of glass wares

Glass wares were washed in liquid detergent under running tap water, and rinsed with distilled water 2-3 times. These were air-dried and then kept in oven for sterilization at 180°C for at least 2 hrs. Plastic wares were autoclaved at 121°C, 15 psi for 15 min.

B. Sterilization of inoculating needles, forceps, cork – borer and working table

Clean inoculating needle was sterilized by dipping the loop of needle in spirit and heating over the flame until red – hot. The process was repeated 2 – 3 times. Forceps and cork – borer were also sterilized in the way of needle. The working table of laminar air flow was disinfected by sweeping with cotton soaked in absolute alcohol and exposing it to UV light for 30 minutes.

C. Sterilization of media and distilled water

Sterilized glassware and plastic wares were used for dispensing media and distilled water. All media were autoclaved at 121°C, 15 psi pressure for 15-30 min.

D. Sterilization of laminar air flow

Prior to the day of inoculation of fungus sample, the laminar air flow was saturated with alcohol vapors. At the time of inoculation the laminar air flow chamber was wiped with 70% alcohol or general spirit. Then only required instruments were kept in the chamber and exposed to UV rays for 15-20 min. All the operation viz., transfer, inoculation etc. were done over a gas burner flame.

E. Culture media

All the solid media were sterilized in an autoclave at 15 lbs pressure (p.s.i) for 15 min. Liquid media sterilized at 10 lbs p.s.i. for 10 min. and process was repeated after 24 hrs.

3.3 Isolation of pathogen

3.3.1 Preparation of culture medium

For isolation of target pathogen in vitro condition, potato dextrose agar (PDA) medium was used. For preparation of PDA, 250 g peeled potatoes were cut into slices and boiled in 500ml of distilled water in conical flask. The extract was strained through a piece of muslin cloth and 20 g dextrose was added in it. 20 g agar – agar was melted in 500 ml of distilled water separately and was mixed in potato dextrose solution and the volume was made upto

1000 ml by adding distilled water. PDA was poured in flasks, plugged with non– absorbent cotton plugs and sterilized in an autoclave.

3.3.2 Isolation and purification of the pathogen

Small pieces of infected tissues 1 – 2 mm dimension from the advancing margin of the spot, adjacent to healthy portions were cut with blade, washed well in distilled water to remove dust adhered to the infected pieces. Pieces were dipped in 0.1 per cent mercuric chloride solution for 30 seconds and finally washed well in three changes of sterilized distilled water. The bits were then transferred to PDA slants with the help of inoculating needle under aseptic condition and incubated at 28±1ºc. After 72 hrs, fragments of hyphal growth from the growing tips were transferred to fresh PDA slants. Pure culture was made, following repeated hyphal tip transfer. Pure culture was maintained on PDA slants by sub culturing it at 30 days intervals. For preservation of cultures the plugged end of the culture tubes were dipped in melted wax and stored in a refrigerator at 5±1ºC.

3.4 Culture media

Potato dextrose agar medium (PDA) containing following ingredients was used during the course of investigation.

Peeled and sliced potato	:	200 g
Dextrose	:	20 g
Agar-agar	:	20 g
Distilled water	:	1000 ml

For the preparation of PDA, sliced potato was boiled in 500 ml of distilled water until they become soft. The supernatant was filtered. The supernatant and melted agar was then mixed measured and the volume was restored to 100 ml with hot water. It was again boiled for five minutes and then dextrose was added. The medium was poured into the flask and culture tubes. The flasks and culture tubes were plugged with non-absorbent cotton plug and sterilized in an autoclave at a pressure of 1.05 kg/cm² at 121.6°C. After this the medium was used under aseptic condition for bioassay of the test fungus.

3.4.1 Methods of inoculation

For inoculating different solid media in Petri- plates, 7 days old culture grown on potato dextrose agar medium was used. The small size of the inoculum was cut and placed at the centre of the plate in an inverted position, so that it came in direct contact with the surface of the medium. For

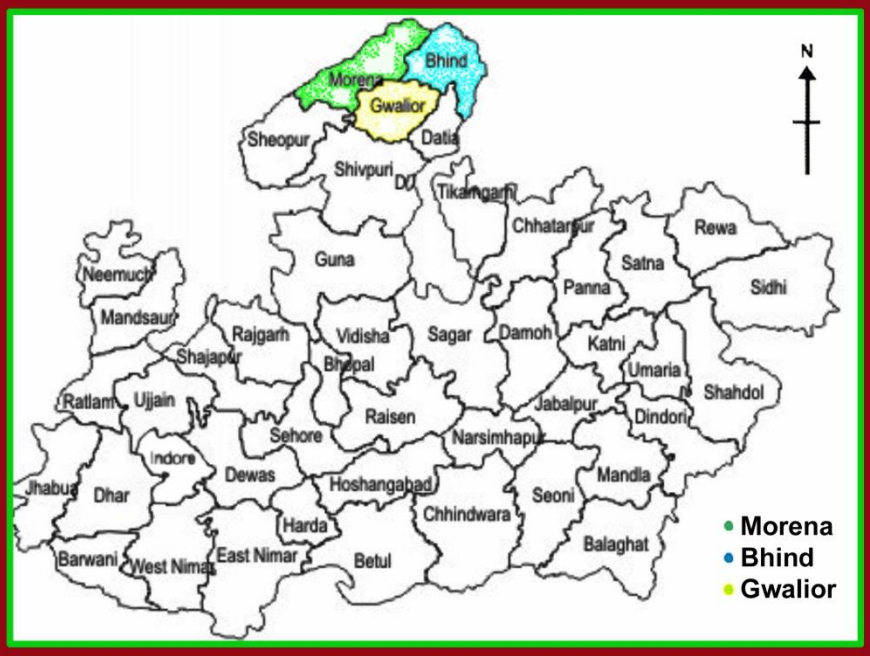

Plate 2- Madhya Pradesh Map: Major Pearl millet surveyed districts

Plate 3- Farmers field survey of pearl millet blast.

inoculating different liquid media in 100 ml Erlenmeyer flasks containing 25 ml broth medium, one disc of 5 mm diameter of fungal mycelium was allowed to float on the medium.

3.4.2 Incubation

The inoculated Petri – plates and flasks were incubated at 28±1ºC in B.O.D. incubator for required period.

3.4.3 Measurement of radial growth of colony

Radial growth of the regular colonies was measured in two directions at right angles with help of a linear scale. In case of irregular colonies, measurements were recorded at the broadest and narrowest diameter and average of two different directions were taken as growth. In all the cases radial growth was recorded after 168 hrs of incubation. In case of poisoned food techniques, it was recorded after 120 and 168 hrs of incubation.

3.5 Disease Survey

Pearl millet fields of Gwalior, Morena and Bhind were surveyed to find out the severity of blast in different blocks of the district. For such survey five villages from each district were randomly selected and from each village five fields were randomly selected. The blast severity was recorded at dough stage of the crop. In each selected field the disease severity was recorded on four randomly selected patches of 1 m^2. The mean disease severity of all the four patches of the field represents the severity of the field. The mean of all the five fields of the village will represent the severity of the village. The district severity was calculated by taking the mean of all the blocks of the district.Blast severity was recorded using 0 - 9 Scale of Mayee and Datar, 1986 as detailed below.

Score	Percent leaf area infected
0	No lesion
1	No lesion to small brown specks of pinhead size
2	No lesion to small brown specks of pinhead size
3	Small, roundish to slightly elongated, necrotic gray spots, about 1–2 mm in diameter with a brown margin
4	Typical blast lesions, elliptical, 1–2 cm long, usually confined to the area between main veins, covering <2% of the leaf area

5	Typical blast lesions covering <10% of the leaf area
6	Typical blast lesions covering 10–25% of the leaf area
7	Typical blast lesions covering 26–50% of the leaf area
8	Typical blast lesions covering 51–75% of the leaf area and many leaves dead
9	All leaves dead

Percent disease index (PDI) was worked out by using the formula given by Wheeler (1969).

$$\text{Percent Disease Index (PDI)} = \frac{\text{Sum of individual disease ratings}}{\text{No. of leaves assessed x Maximum grade}} \text{ x } 100$$

3.6 Effective medium for the growth of *Pyricularia grisea* under *in-vitro* condition

A total number of 7 traditionally used and self made media viz., Pearl millet dextrose agar, Oat meal agar, Pearl millet potato dextrose agar ,Richard's agar medium ,Czapeck's dox agar ,Yeast extract agar, Potato carrot agar evaluated against *Pyricularia grisea* and compared with check (Potato Dextrose Agar) to select out the suitable medium for the growth of fungus. All medium inoculated with 9 mm. disc of 72 hours old culture of *Pyricularia grisea*. After inoculation the plates incubated at 28+1^{0}C. The fungal growth of all inoculated medium measured regularly at 24 hrs. interval starting from 48 h of the inoculation.

Table 3.2: The culture media utilized for testing the efficacy of the growth of blast pathogen

S.No.	Name of the medium	Ingredients
1	**Potato Dextrose Agar (PDA)medium**	Peeled & sliced potato - 200 g ,Dextrose - 20 g, Agar-agar - 20 g ,Distilled water1000 ml
2	**Czapeks Dox Agar (CDA) medium**	Sodium nitrate - 2 g ,Di potassium hydrogen phosphate - 1 g, Magnesium sulphate - 0.5 g, Potassium chloride - 0.5 g, Ferrous sulphate - 0.01 g , Sucrose - 30 g, Agar-agar - 20 g
3	**Richards's Agar (RA) medium**	Potassium nitrate - 10 g ,Potassium monobasic phosphate - 5 g , Magnesium sulphate - 2.5 g, Ferric chloride - 0.02 g,

		Sucrose - 50 g , Agar- agar - 20 g
4	**Pearl millet dextrose agar**	Pearl millet grain 50 gm, Agar-agar 20 gm, Dextrose 20 gm , Distilled water 1lt
5	**Pearl millet potato dextrose agar**	Pearl millet grain 50 gm, sliced potato 100 gm, Agar-agar 20 gm, Dextrose 20 gm Distilled water 1lt.
6	**Potato carrot agar**	Sliced potato 100 gm, sliced carrot 100 gm, Agar-agar 20 gm, Distilled water 1lt.
7	**Oat meal agar**	Oat meal - 50 gm, Agar-agar 20 gm, Dextrose 20 gm, Distilled water 1lt
8	**Yeast extract agar**	Peptic digest of animal tissue- 5 gm, Yeast extract-3 gm, Agar- 15 gm ,Distilled water-1 lit

In the above mentioned media the ingredients were poured in the required quantity of water, boiled and there after media was prepare. The supernatant was filtered. The supernatant and melted agar was then mixed measured and the volume was restored to 1000 ml with hot water. It was again boiled for five minutes and then dextrose was added. The medium was poured into the flask and culture tubes. The flasks and culture tubes were plugged with non-absorbent cotton plug and sterilized in an autoclave. After this the medium was used under aseptic condition for bioassay of the test fungus.

3.7 *In-vitro* evaluation of botanicals and chemicals against *Pyricularia grisea*

A total number of 8 botanicals and 6 fungicides were evaluated against *Pyricularia grisea* under *in vitro* condition by adopting poisoned food technique.

Experimental details of Fungicides and Botanicals-

Design : CRD

Replications : 3

Treatments : 15 (Botanicals 8, Fungicides 6, +1 Check)

Technique : Poisoned food

(A)

(B)

Plate 4 (A and B) -Experimental field view at dough stage.

Treatments -

T1- *Azadirachta indica* (leaf)

T2– Bel patra (leaf)

T3- Periwinkle (leaf)

T4 - *Nicotiana tobacum* (leaf)

T5– *Datura stramonium* (leaf)

T6 - *Lantana camara* (Leaf)

T7– *Allium sativum* (Bulb)

T8- *Ocimum tenuiflorum* (Tulsi)

T9- Iprobenphos

T10- Tricyclazole

T11- Azoxystrobin

T12- Thiophenate methyl

T13- Propiconazole

T14- Trifloxystrobin+Tebuconazole

T15- Control

a) Preparation of botanicals

The fresh crude extracts were prepared by grinding the required quantity of plant parts viz., leaves, bulbs, and rhizomes. Before grinding the equal quantity of water was added in the respective plant parts (1:1 weight/volume basis). These crushed extract were used @ 20 percent adopting poisoned food techniques.

The growth of the fungus measured seven days after inoculation.

b) Fungicides

Following fungicides were evaluated against *Pyricularia grisea* under in-vitro condition by adopting poisoned food technique.

Table 3.3: List of fungicides tested against *Pyricularia grisea* under *in-vitro* condition.

S. No.	Common name	Trade name	Chemical name	Active ingredient
1	Iprobenphos	KITAZIN	S-benzyl o, o-diisopropyl phosphorothioate	48% EC
2	Tricyclazole	DhanTeam	5-methyl-1, 2, 4-triazolo[3, 4-	75%WP

			b]benzothiazole	
3	Azoxystrobin	AMISTAR	Methyl (2E)-2-(2-{[6-(2-cyanophenoxy pyrimin-4-yl]oxy}phenyl)-3 methoxyacrylate	23%SC
4	Thiophenate methy	ROKO	methyl N-[[2-(methoxycarbonylcarbamothioylamino)phenyl]carbamothioyl]carbamate	50%SC
5	Propiconazole	Dhanuka	1-[[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2 yl]methyl]-1,2,4-triazole	25%EC
6	Tebuconazole + Trifloxystrobin	Nativo	1-(4-chlorophenyl)-4,4-dimethyl-3-(1,2,4-triazol-1-ylmethyl)pentan-3-ol+Methyl (2E)-2-methoxyimino-2-[2-[[(E)-1-[3(trifluoromethyl)phenyl]ethylideneamino]oxymethyl]phenyl]acetate	50%WG, 25%WG

Percent inhibition over control was calculated by following formula suggested by Bliss (1934)

$$\text{Percent inhibition} = \frac{C-T}{C} x\ 100$$

where, C = Control, T = Fungal growth

3.8 Evaluation of hybrids varieties, locally collected and other pearl millet material against blast

Fifty hybrids varieties, locally collected and other pearl millet material were planted in 4 m single row length. The blast severity was recorded on the 5 randomly selected and tagged plants of each line. The details of the experiment were as follow:-

Entries	-	50
Design	-	RBD
Replication	-	2
Plot size	-	4 m single row
Distance	-	Row to Row - 50 cm
Plant to Plant	-	10 cm

Fertilizer	-	60 N, 40 P_2O_5, 20 K_2O kg/ha
Date of sowing	-	15.07.2017

3.9 Field evaluation of selected chemicals and botanicals for the management of Pearl millet blast

After in vitro evaluation the selective botanicals and chemicals further evaluated in field against the disease. The detail of experiments as follow :

Design	:	RBD
Replications	:	4
Treatments	:	8
Plot size	:	4 x 2 m
Spacing	:	50 x 10
Fertilizer application	:	60 : 40 : 20

Treatments-

T1	–	Iprobenfhos (Kitazin) 48 EC @ 0.1%
T2	–	Tricyclazole (Beam) @ 0.1%
T3	–	Azoxistrobin 25 EC @ 0.05%
T4	–	Propiconazole @ 0.05%
T5	-	Trifloxystrobin + Tebuconazole @ 0.05%
T6	-	*Allium sativum* (bulb) @ 20%
T7	-	*Lantana camera* @ 20%
T8	-	Control (Untreated)

Statistical Analysis:

For statistical analysis the data of blast scoring (1-9 scale) were transformed under angular transformation.

Design

The statistical model design in single environment could be defined as below (Chandel, 1999).

$$Yijk = + ti + bij + rijk$$

Where, k = the general

ti = the effect of ith treatment

bij = the effect of jth replication in ith treatment

eijk = the error associated with each observation.

The skeleton for analysis of variance for randomized complete block design is given below:

Source of variance	Degree of freedom	Observation mean sum of squares	Expected mean sum of squares
Replication	r-1	M.S1	2ei + 2tri
Treatment	t-1	M.S2	2ei + 2rri
Error	(r-1)(t-1)	M.S3	2ei
Total	**rt-1**		

Standard error of mean

It was calculated as formula given below :

$$\text{S.E. (m)} \pm = \sqrt{\frac{Ve}{r}}$$

Where,

Ve = Error mean sum of squares

r = Number of replication

Critical difference:

It was mean used as formula mentioned below :

CD = SE (m) ± × at 5 %

Where, C. D. = Critical difference

S.D. = Standard error of mean

t 5 % = Table value of ‘t’

Chapter – IV

RESULTS

4.1 To find out the status of Pearl millet blast in Northern Madhya Pradesh

A planned survey of Pearl millet blast was carried out in Morena, Bhind and Gwalior districts. The data presented in the table 4.1 reveals that blast is a commonly occurring foliar disease of Pearl millet and its severity across the villages was in the range of 9.8 % to 15.04 %. In Morena district the maximum severity of blast was recorded in Morena village 15.04% followed by Chhera (13.32%), Kharagpur (12.28%) and Tighra (11.8 %) while the minimum blast severity of 10.92 % was recorded in Jigani village (Fig.4.1).

Table 4.1: Village wise severity of Pearl millet blast in Morena, Bhind and Gwalior districts.

District	Village	Percent blast severity					Mean
		F1	F2	F3	F4	F5	
D1.Morena	V1.Kharagpur	12.2	17.4	7.6	13.4	10.8	12.28
	V2. Jigani	9.6	7.2	11.4	14	12.4	10.92
	V3.Tighra	15.4	8.8	14.8	12.4	7.6	11.80
	V4.Chhera	9.4	12	11.8	16.2	17.2	13.32
	V6.Morena	20.2	13.2	16.4	10.8	14.6	15.04
	D1. Mean %						**12.67**
D2.Bhind	V1.Badagaon	6.8	11.2	12.4	10.2	8.4	9.80
	V2.Barua	13.4	10.6	11.8	7.4	9.8	10.60
	V3.Mauch	12	14.6	10.2	8.2	10.6	11.12
	V4.Aatiri	9.2	8.6	7.4	11.2	14.6	10.20
	V5.Singarpura	13.8	14	12.8	15.4	13	13.80
	D2. Mean %						**11.10**
D3.Gwalior	V1.Ahroli ghat	7.6	13.4	17.8	7.8	10.8	11.48
	V2.Adokhar	14.2	11.6	10.2	13.6	14.8	12.88
	V3.Akoun	8.4	10	14.2	9.8	11.2	10.72
	V4.Amleda	9.8	13.8	11.2	7.6	13.4	11.16
	V5.Babedi	14.6	9.6	11.4	14.4	12.2	12.44
	D3. Mean %						**11.73**
	All District Mean %						**11.84**

D- District, V- Village, F-Field.

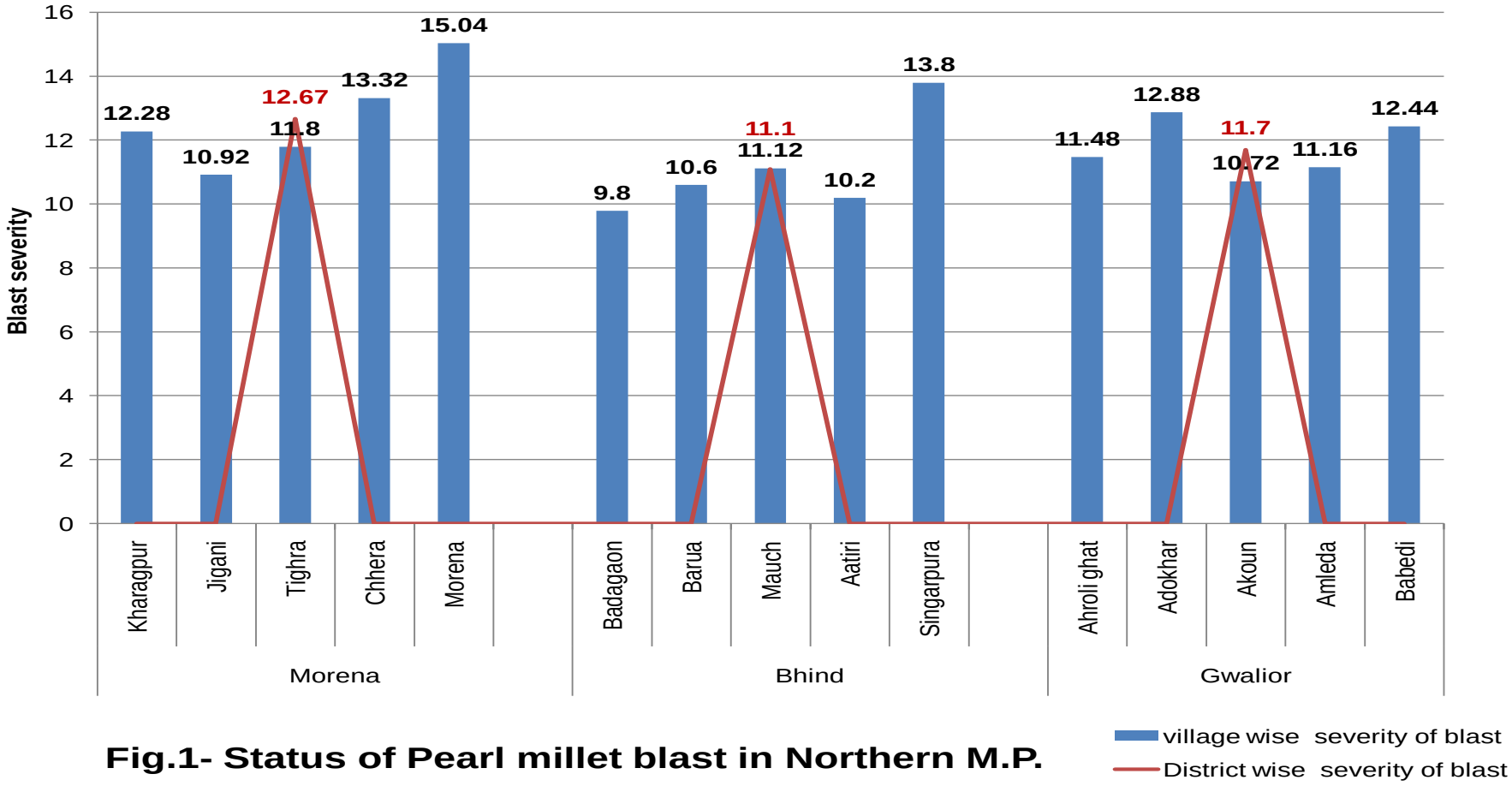

Fig.1- Status of Pearl millet blast in Northern M.P.

The maximum blast severity in Bhind district was recorded in Singarpur village (13.8%) followed by Mauch (11.12%), Barua (10.6%) and Aatiri(10.2%) while minimum blast severity of 9.8% was recorded in Badagaon village (Fig.4.1). Adokhar village of Gwalior district showed maximum severity (12.88%) of blast followed by Babedi (12.44%), Ahroli ghat (11.48%) and Amleda (11.16%) while minimum blast severity was recorded on the Akoun (10.72%) (Fig.4.1).

It is also obvious from the table 4.1 that the maximum blast severity (12.67%) was recorded in Morena district followed by Gwalior and Bhind where 11.73% and 11.10% blast severity was recorded respectively.

4.2 *In-vitro* evaluation of culture media to find out the most suitable medium for the growth of *Pyricularia grisea.*

Seven media were evaluated in the present study for the growth of *Pyricularia grisea* **under *in-vitro* condition and the data are summarized in the table 4.2.**

Table 4.2: *In-vitro* evaluation of culture media to find out the most suitable medium for the growth of *Pyricularia grisea*

Treatment No.	Name of the medium	Diameter growth of fungal mycelium (mm)
1	Pearl millet dextrose agar	48.5
2	Pearl millet potato dextrose agar	84.00
3	Czapeck's dox agar	81.00
4	Potato carrot agar	58.00
5	Oat meal agar	57.00
6	Richard's agar medium	51.00
7	Yeast extract agar	50.00
8	PDA (Control)	85.00

SE(m) 1.07
C.D (at 5%) 3.20

Data are the mean of three replications.

Plate 5-*In vitro* evaluation of culture media to find out the most suitable medium for the growth of *Pyricularia grisea*

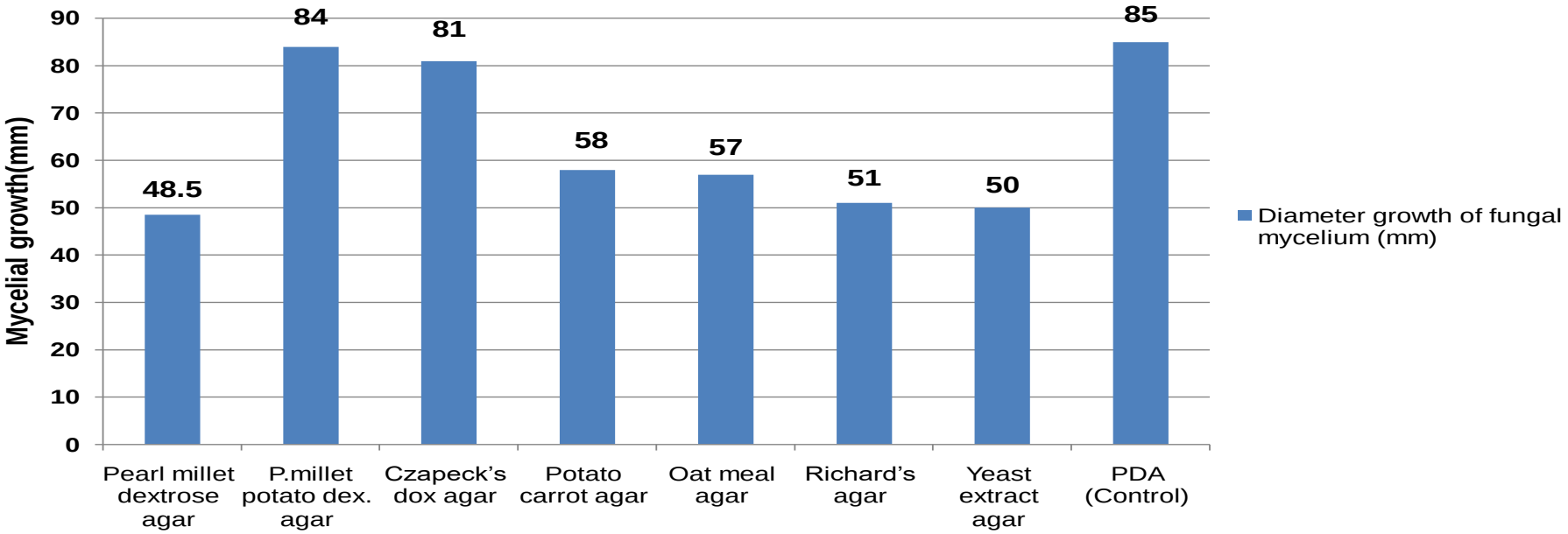

Fig.4.2-*In vitro* evaluation of culture media against *Pyricularia grisea.*

The maximum colony diameter (85.0 mm) was recorded on Potato dextrose agar medium followed by Pearl millet potato dextrose agar medium (84.0 mm), Czapek's dox agar medium (81.0 mm), Potato carrot agar medium (58 mm), Oat meal agar medium (57 mm), Richard's agar medium (51 mm) and Yeast extract agar medium (50 mm) while minimum **mycelial growth (48.5%) was recorded on** Pearl millet dextrose agar medium.

PDA was significantly superior over rest of the other tested medium except Pearl millet potato dextrose agar medium. Pearl millet potato dextrose agar medium significantly superior over Pearl millet dextrose agar medium, Yeast extract agar medium , Richard's agar medium , Oat meal agar medium and Potato carrot agar medium where as it was statistically at par with Czapek's dox agar medium.

4.3 *In-vitro* evaluation of botanicals and chemicals against *Pyricularia grisea*

A total number of eight botanicals (@ 20%) viz., *Azadirachta indica*(leaf); *Aegle marmelos* (leaf); *Catharanthus roseus* (leaf); *Nicotiana tobacum*(leaf); *Datura stramonium* (leaf); *Lantana camra*(leaf*); Allium sativum* (bulb); *Ocimum tenuiflorum*(Tulsi) and Six fungicides (@ 0.1%), viz**.,** Iprobenphos (Kitazin); Thiophenate methyl; Tricyclazole (Beam); Azoxystrobin; Trifloxystrobin+Tebuconazole; Propiconazole were evaluated against *Pyricularia grisea* under *in-vitro* condition. The data summerized in table 4.3 reveals that three fungicides viz., Tricyclazole**,** Propiconazole and Trifloxystrobin+Tebuconazole @ 0.1 per cent absolute inhibited fungal growth while maximum growth was recorded in control (87.33 mm). These three fungicides were significantly superior over all other tested fungicides and botanicals. Iprobenphos has also shown good inhibition of the fungus as only 5.3 mm growth was recorded in this treatment and it was significantly superior over all the tested botanicals and the remaining fungicides viz., Azoxystrobin 25 EC (21 mm) and Thiophenate methyl 70% WP (58.6 mm).

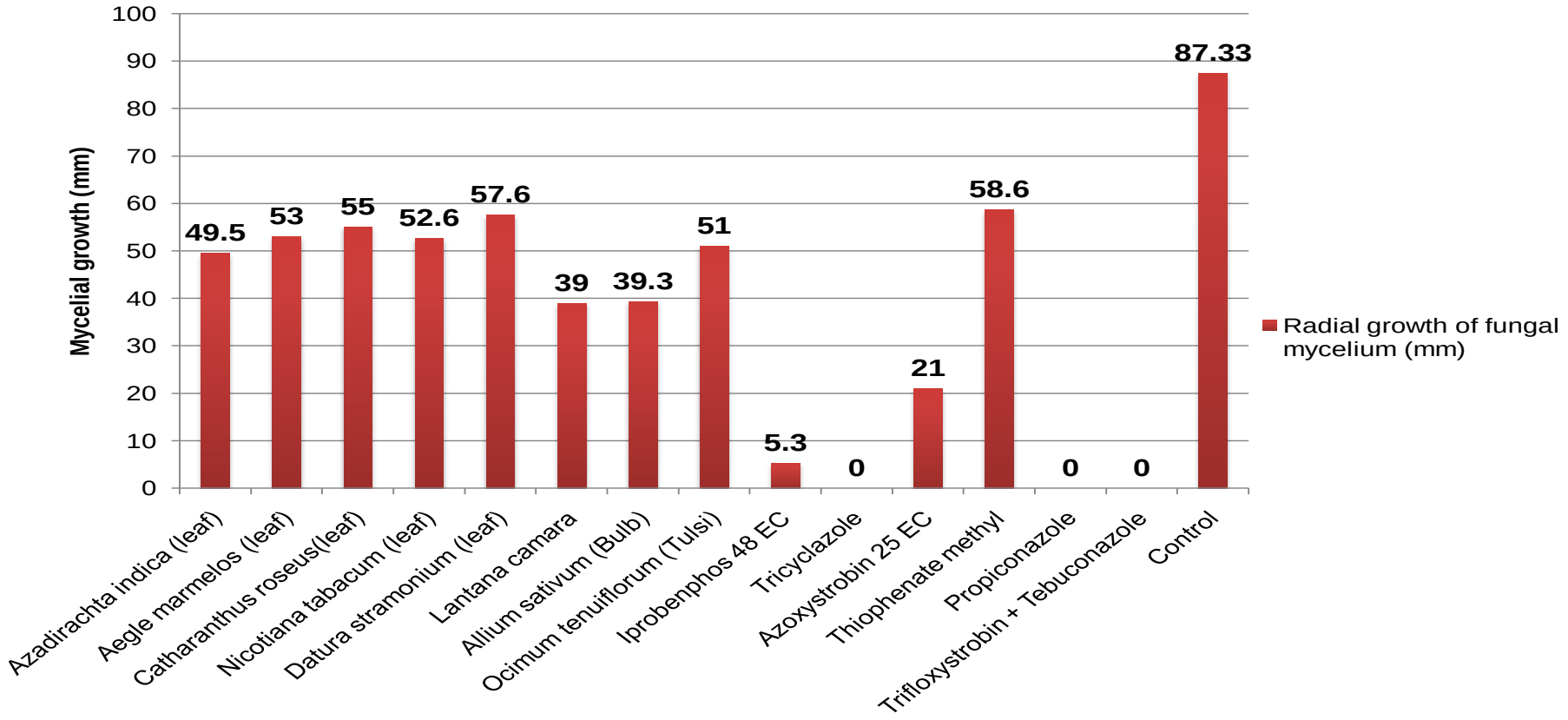

***Fig. 4.3 In vitro* evaluation of botanicals and chemicals against *Pyricularia grisea*.**

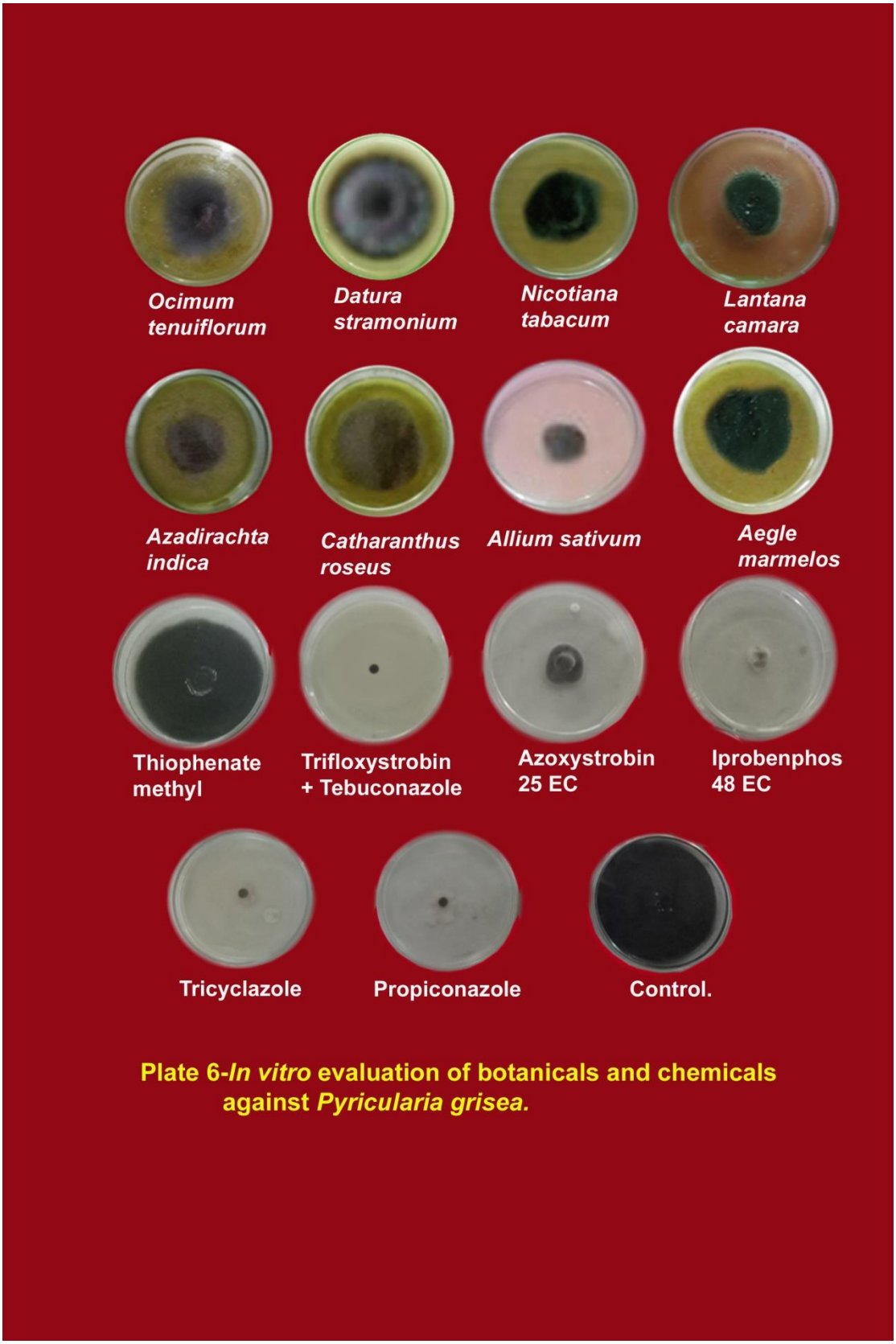

Plate 6-*In vitro* evaluation of botanicals and chemicals against *Pyricularia grisea.*

Among the botanicals *Lantana camra* leaf extract and *Allium sativum* bulb extract @ 20 per cent concentration were found effective against the pathogen as in these two treatments 39 mm and 39.3 mm growth showed respectively. These two botanicals were significantly superior over the rest of the tested botanicals and also these two were significantly superior over the fungicide Thiophenate methyl (58.6mm). In *A. indica* leaf extract 49.5 mm fungal growth was recorded which was significantly superior over *A. marmelos* leaf extract (53 mm), *C. roseus* leaf extract (55 mm), *D. stramonium* (57.6mm) and Thiophenate methyl (58.6mm) but statistically at par with *O. tenuiflorum* (51mm) and *N. tabacum*(52.6%)

Table 4.3: *In-vitro* evaluation of botanicals and chemicals against *Pyricularia grisea*

Treatment No.	Treatments	Radial growth of fungal mycelium (mm)
1.	*Azadirachta indica (leaf)*	49.5
2.	*Aegle marmelos (leaf)*	53.00
3.	*Catharanthus roseus (leaf)*	55.00
4.	*Nicotiana tabacum (leaf)*	52.60
5.	*Datura stramonium (leaf)*	57.60
6.	*Lantana camara*	39.00
7.	*Allium sativum (Bulb)*	39.30
8.	*Ocimum tenuiflorum (Tulsi)*	51.00
9.	Iprobenphos 48 EC	5.30
10.	Tricyclazole	0.00
11.	Azoxystrobin 25 EC	21.00
12.	Thiophenate methyl	58.60
13.	Propiconazole	0.00
14.	Trifloxystrobin + Tebuconazole	0.00
15.	Control	87.33
SE(m) 1.20		
C.D (at 5%) 3.48		

The data are the mean of three replications of the above botanicals and

fungicides which were used @ of 20 and 0.1 per cent, respectively.

4.4 Evaluation of hybrids, varieties, locally collected and other pearl millet material against blast

A total of fifty entries of Pearl millet consisting of hybrids , varieties, locally collected & other pearl millet material were screened against blast under favourable condition and the data are summarized in table 4.4. which reveals that the tested entries showed a great variation in response to blast as their blast PDI range from 0.0 to 99.99%

Table 4.4: Evaluation of hybrids varieties, locally collected and other pearl millet material against blast

S.No.	Lines	PDI (Mean)	S.No.	Lines	PDI(Mean)
1.	Krishna 9119	44.44(41.70)	**26.**	JBV-2	42.15(40.48)
2.	Kaveri super boss	35.50(36.46)	**27.**	JBV-3	59.95(50.84)
3.	krishna 7207	34.40(35.89)	**28.**	JBV-4	51.05(45.66)
4.	proagro 9444	59.95(50.73)	**29.**	Bhind local	22.15(27.93)
5.	Pioneer 86M88	33.30(35.05)	**30.**	Dhanshakti	83.25(65.93)
6.	Pioneer 86M86	48.87(44.29)	**31.**	ICMB01333	38.85(38.47)
7.	Pioneer 86M84	46.60(42.95)	**32.**	ICMB02444	77.70(62.59)
8.	Dhaanya 7888	54.35(47.61)	**33.**	ICMB07111	8.89(17.18)
9.	Dhaanya 7792	29.90(32.79)	**34.**	ICMB92777	73.25(38.12)
10.	HHB 272	31.05(33.83)	**35.**	ICMB93333	15.50(23.14)
11.	MPMH21	8.86(17.13)	**36.**	ICMB97222-P1	43.30(41.15)
12.	HHB226	25.50(30.29)	**37.**	ICMR02222	49.95(45.07)
13.	RHB177	13.31(21.33)	**38.**	ICMR06444	25.50(30.33)
14.	GHB538	0.00(0.00)	**39.**	ICMR11003	53.25(46.97)
15.	HHB67 Imp	25.52(30.33)	**40.**	ICMR07888	96.60(79.52)
16.	MPMH17	0.00(0.00)	**41.**	ICMR11333	62.15(52.07)
17.	RHB173	29.95(33.12)	**42.**	GHB905	15.51(22.98)
18.	HHB197	36.60(37.21)	**43.**	Pratap	61.05(51.40)
19.	GHB558	13.31(21.33)	**44.**	Nandi61	42.15(40.48)
20.	GHB744	24.40(29.58)	**45.**	Nandi65	52.15(46.34)
21.	HHB223	16.60(24.00)	**46.**	RAJ 171	47.70(43.78)
22.	KBH108	24.40(29.58)	**47.**	RAJ 179	81.05(64.28)
23.	GHB732	53.30(46.88)	**48.**	ICMV 221	52.15(46.34)
24.	NHB5767	25.50(30.28)	**49.**	Pusa Co. 383	75.50(60.32)
25.	NHB5061	0.00(0.00)	**50.**	ICMB95444(Check)	99.90(84.53)
SE(m)			**4.11**		
C.D (at 5%)			**11.69**		

Data in parenthesis indicates square root transformation.

Three entries viz., GHB538, MPMH17 and NHB5061 were absolutely free from the disease. In respect of blast reaction these three entries were significantly superior over rest of the 47 entries.

The maximum blast PDI (99.9 %) was recorded in the susceptible check ICMB95444 and due to severe blast this entry could not produce ear head and dried prematurely.

ICMB95444 showed significantly higher disease the rest of the other 49 tested entries.

4.5 Field evaluation of selected chemicals and botanicals for the management of Pearl millet blast.

An experiment was carried out to manage the Pearl millet blast by the foliar application of chemical and botanicals which were found effective under *in vitro* evaluation. The data summarized in table 4.5 and Fig. 4.4 revealed that all the five fungicides viz., Trifloxystrobin+Tebuconazole @ 0.05%, Propiconazole @ 0.05%, Azoxistrobin 25 EC @ 0.05%, Tricyclazole (Beam) @ 0.1%, Iprobenphos (Kitazin) 48 EC @ 0.1% and botanicals (@ 20 %) significantly checked the disease.

The minimum blast PDI (28.3%) was recorded in the treatment Trifloxystrobin + Tebuconazole followed by Propiconazole (33.3%), Azoxistrobin (35.5%), Tricyclazole (39.4%), Iprobenphos (Kitazin) (40.5%), *Allium sativum* bulb exract (42.1%) and *Lantana camera* leaf extract (51.05%), while maximum PDI (64.3%) was recorded in control.

The most effective treatment Trifloxystrobin + Tebuconazole was significantly superior over Tricyclazole, Iprobenphos, *Allium sativum* bulb extract and *Lantana camera* leaf extract where as it was statistically at par with Azoxistrobin and Propiconazole.

Trifloxystrobin + Tebuconazole gave 55.9 % disease control followed by 48.21 % in Propiconazole, 44.8% in Azoxistrobin ,38.72% in Tricyclazole, 37.01 % in Iprobenphos, 34.50 % in *Allium sativum* (bulb) whereas the least disease control (20.60%) was recorded in *Lantana camera* leaf extract.

Table 4.5: Field evaluation of selected chemicals and botanicals for the management of Pearl millet blast

S. No.	Treatments	PDI (Mean)	Disease percent control
1.	Iprobenphos 48 EC@ 0.1%	40.50(39.46)	37.01
2.	Tricyclazole @ 0.1%	39.40(38.76)	38.72
3.	Azoxistrobin 25 EC @ 0.05%	35.50(36.44)	44.80
4.	Propiconazole @ 0.05%	33.30(35.16)	48.21
5.	Trifloxystrobin+Tebuconazole @ 0.05%	28.30(32.01)	55.90
6.	*Allium sativum* @ 20%	42.10(40.46)	34.50
7.	*Lantana camara* @ 20%	51.05(45.59)	20.60
8.	Control (Untreated)	64.30(53.41)	0.00
SE(m) 2.29			
C.D (at 5%) 6.73			

The data given in parenthesis are angular transformed

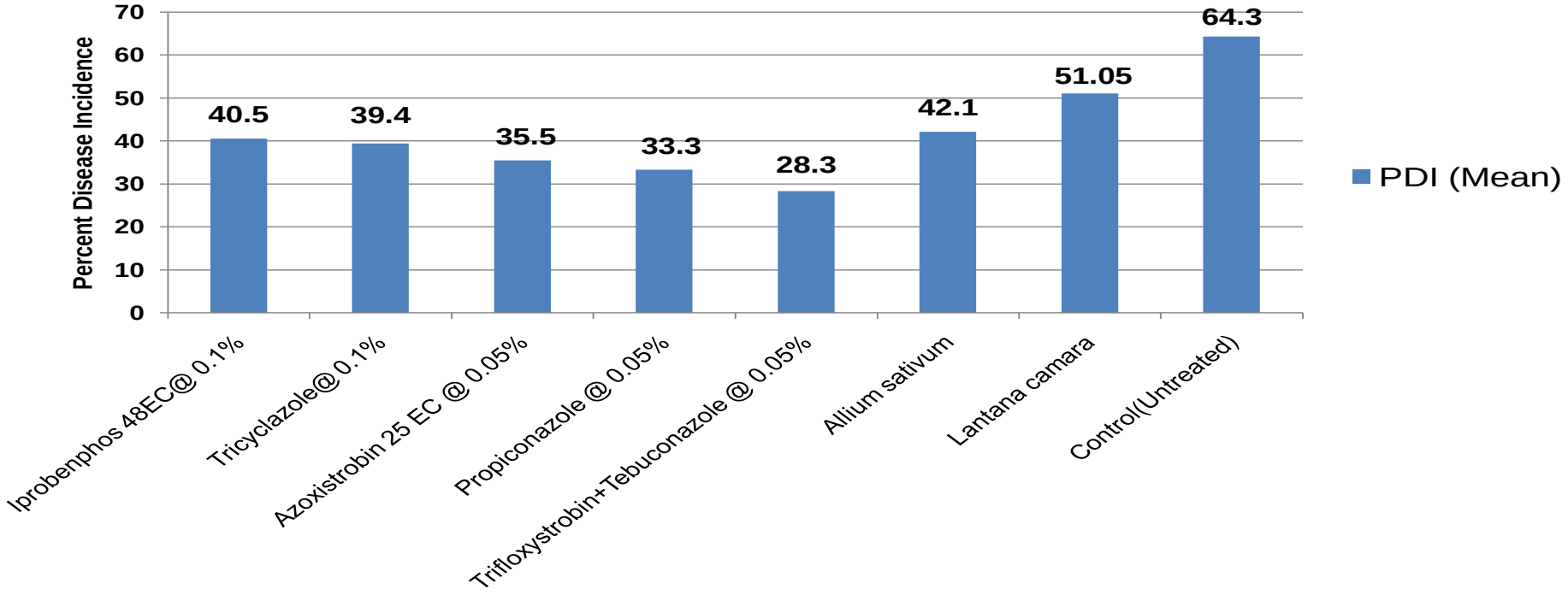

Fig.4.4 Field evaluation of selected chemicals and botanicals for the management of Pearl millet blast.

Chapter - V

DISCUSSION

An attempt has been made in this chapter to explain the experimental findings with possible reasons in the light of the literature available on different aspects under study.

Pearl millet [*Pennisetum glaucum* (L.) R.Br.] one of the common millet grown as a nutri cereal for human and fodder crop for livestock. Blast [*Pyricularia grisea (Cooke) Sacc.*] has been observed as a serious disease of Pearl millet in Northern Madhya Pradesh. During the present study it was observed that some of the highly susceptible genotypes were completely damaged by the pathogen and such genotypes were dried prematurely resulting in 100 % yield losses. The disease appears as grayish, water-soaked lesions on foliage that enlarge and become necrotic, resulting in extensive chlorosis and premature drying of young leaves. On the leaves, the lesions appear as small bluish green flecks, characteristic spindle shaped spots with grey centre and dark brown margin .Lesions are often surrounded by a chlorotic halo, which turns necrotic, giving the appearance of concentric rings. The lesions are usually confined to interveinal spaces on the foliage. Lesions grow and coalesce to cover large surface areas and cause necrosis of tissues. In case of a susceptible cultivar the entire foliage gives a burnt appearance.

Present finding is in conformity with the finding of Padmanabhan (1974) and Manibhushan Rao (1994) that the lesions or spots first appear as minute brown specks, then grow to become spindle shaped pointed at both ends, several cm long and about 0.5 – 1.0 cm wide. The centre is greenish grey often showing a brownish margin. The size, colour and shape of the lesions, however, vary with different climatic conditions and also varietal response. Under favourable conditions on a susceptible cultivar several greyish spots may appear, become larger and broader and coalesce, leading to withering of the whole leaf.

Survey

The results of the disease survey reveals that blast is one of the most important biotic constraints in the cultivation of pearl millet in Northern region of Madhya Pradesh. The farmers of the region usually normally grows dual purpose hybrids and varieties**.** The maximum blast severity was recorded in Morena district followed by Gwalior. While the severity of the disease was low in Bhind district. The higher severity of blast in Morena district. The possible reason of the higher severity of blast in Morena district might be due to soil fertility, monoculture, continous cultivation of hybrids in large areas.

The present finding is supported by the work of Devda (2009) who surveyed the pearl millet fields of Morena, Bhind and Gwalior and reported 5.5, 3.8 and 5.4 per cent severity of blast respectively. The severity of Pearl millet blast in Morena, Bhind and Gwalior was 11.53, 13.40 and 11.28 percent respectively. Yadav *et al.,* (2012).

Culture media

Eight culture media were evaluated against *Pyricularia grisea*. The findings reveals that the maximum fungal growth was recorded in Potato dextrose agar followed by Pearl millet potato dextrose agar medium while pearl millet dextrose agar medium was the least effective media. Potato dextrose agar was significantly superior over other tested media except Pearl millet potato dextrose agar medium. In the present study PDA was at par with Pearl millet potato dextrose agar medium. In pearl millet potato dextrose agar medium the quantity of potato has been reduced from 200 gram to 100 gram, even though the result are at par and result may be much better if we use recommended quantity of potato that is 200 gram per litre. Under such situation the pearl millet potato dextrose agar medium may be one of the more suitable media than PDA for the growth of *Pyricularia grisea* isolated from pearl millet leaves.

Present finding is supported by Malviya R. (2014) who tested four media namely Potato dextrose agar, Czapek's Dox Agar, Rechard Agar medium and Corn meal agar. However, PDA was found to be highly suitable

for the growth and sporulation of *P. grisea*. PDA medium could provide the best medium for *P. oryzae* vegetative growth. Mahdieh S (2013).

Mijan Hossain (2000) also documented that among the non synthetic media, potato dextrose agar supported maximum radial growth (85.00 mm), next was host extract + 2 per cent sucrose agar medium (80.33 mm) followed by oat meal agar (75.00 mm).

***In vitro* and *in vivo* evaluation of botanicals and chemicals**

A total number of eight botanicals and six fungicides were evaluated under *in vitro* condition. Out of them three fungicides viz. Tricyclazole**,** Propiconazole and Trifloxystrobin + Tebuconazole @ 0.1 percent absolute inhibited fungal growth and the two botanicals *Lantana camra* leaf extract and *Allium sativum* bulb extract @ 20 percent concentration were found effective against the pathogen. Further the present findings is in conformity with those of the effective chemicals and botanicals were evaluated in the field and the result reveals that foliar application of Trifloxystrobin+Tebuconazole @ 0.05% was found most effective control of Pearl millet blast followed by Propiconazole, Azoxistrobin, Tricyclazole, Iprobenphos, *Allium sativum* bulb extract and *Lantana camera* leaf extract while the maximum PDI was recorded in control.

Similarly finding Joshi and Gohel (2015) revealed that Propiconazole and Tricyclazole completely inhibited the growth of *Pyricularia grisea*.

Netam *et al.* (2014) also reported that complete inhibition of mycelial growth of *Pyricularia grisea* was given by Tricyclazole followed by Ediphenphos and Mancozeb 65 per cent WP+Carbendazim12 per cent WP under laboratory condition.

Pal (2014) who studied six fungicides like Kresoxim methyl, Azoxystrobin, Propiconiazole, Trifloxystrobin + Tebuconazole (Nativo), Difeconazole, and Tricyclazole to control the leaf blast of rice. Among them Trifloxystrobin + Tebuconazole (Nativo) was found to be a highly effective.

Similarly the effectiveness of *Allium sativum* was also reported by Hajano Jamul-U-Ddin *et al*., (2000) who evaluated five fungicides, three botanicals and six bio-agents. Among botanicals only higher dose of garlic

(Allium sativum L.) completely inhibited the mycelial growth of the test fungus.

Evaluation of hybrids varieties, locally collected and other pearl millet material

Out of fifty hybrids/varieties , three entries viz. GHB538, MPMH17 and NHB5061 were absolutely free from blast while the maximum PDI was recorded in ICMB95444 (99.9%).

Earlier finding Yadav *et al.* (2013) who evaluated twenty five promising pearl millet hybrids and varieties against blast. The minimum severity of 7.5% was recorded in PB 106, GHB-744, and GHB-732, while its maximum severity (32.50%) was recorded in B-2301, PB 106, GHB 744 and GHB-732. One thirty five pearl millet lines were evaluated against blast at Gwalior and reported blast severity in the range of 0 - 47.5%. The result reveals that only one entry MH 1541 remained completely free from blast while its maximum severity i.e. 47.5% was recorded in MH 1513. Devda (2009).

Chapter - VI

SUMMARY, CONCLUSION AND SUGGESTIONS FOR FURTHER WORK

Pearl millet (*Pennisetum glaucum*) is an important nutri-cereal for human as well as a forage/fodder crop for livestock. It is the only cereal crop that is capable for adopting harsh climate condition and marginal soil. Blast incited by *Pyricularia grisea* (Cooke) Sacc. has occupied a key position among the pearl millet diseases resulting severe losses in high yield potential hybrids/ varieties particular cultivated for fodder purpose. The results of the studied undertaken are summarized as under.

The disease survey was carried out in Morena, Bhind and Gwalior which jointly contribute more than 70% in area and production of the state. The survey result reveals that blast is a serious problem in the state as its severity in the above three district was 12.6%, 11.10% and 11.73%, respectively.

A total of eight culture media were evaluated against *Pyricularia grisea* isolated from blast infected pearl millet leaves. Out of them the maximum growth of fungus was recorded in Potato dextrose agar medium (85 mm). followed by Pearl millet potato dextrose agar medium (84.0 mm), Czapek's dox agar medium (81.0 mm) and Potato carrot agar medium (58 mm) while minimum mycelial growth (48.67%) was recorded **on** Pearl millet dextrose agar medium. PDA and Pearl millet potato dextrose agar medium were statistically at par with each other.

A total number of eight botanicals and six fungicides were evaluated under *in vitro* condition. Out of them three fungicides viz. Tricyclazole**,** Propiconazole and Trifloxystrobin+Tebuconazole @ 0.1 percent absolute inhibited fungal growth and two botanicals *Lantana camra* leaf extract and *Allium sativum* bulb extract @ 20 percent concentration were found effective against the pathogen as in these two treatments 39 mm and 39.3 mm growth showed respectively. After *in-vitro* evaluation five fungicides and two botanicals were further evaluated in the field condition. The findings reveals that all the tested fungicides and botanicals significantly check the blast but

the combination of Trifloxystrobin+Tebuconazole @ 0.05 was found most effective as in this treatment the minimum PDI (28.3%) was recorded followed by Propiconazole @ 0.05% (33.3%), Azoxistrobin 25EC@ 0.05% (35.5%) and Tricyclazole (Beam) @ 0.1% (39.4%). Among the botanicals *Lantana camra* @ 20% was also effective.

Out of fifty hybrids/varieties, three entries *viz*. GHB538, MPMH17 and NHB5061 were absolutely free from blast while the maximum disease incidence was recorded in ICMB95444 (99.9%).

CONCLUSION

The entire study carried out was successful in meeting its objectives as reflected in the following conclusions drawn there from;

1. Blast is one of the most serious diseases of Pearl millet in Northrn region of M.P.
2. Potato dextrose agar and Pearl millet potato dextrose agar medium are the more suitable media for the culture of *Pyricularia grisea* isolated from infected Pearl millet leaves.
3. Trifloxystrobin+Tebuconazole @ 0.05% and Propiconazole @ 0.05% are the more effective fungicide for the management of blast through foliar application.
4. Three entries viz. GHB538, MPMH17 and NHB5061 were identified as blast resistance under field condition.

SUGGESTIONS FOR FURTHER WORK

On the basis of above findings following measures are recommended for the study and management of Pearl millet blast.

1. More number of genotypes should also be screened to find out the resistance sources.
2. Work should also be carryout on cultural and genetic variability of the pathogen.
3. More number of botanicals and other non chemicals should be tested to find out and alternate to the chemical for the management of disease.

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APPENDIX-I

Meteorological Data (Average of the Week) Location Gwalior 2017

Std. Weather week	Date/Period	Temp°C Maxi.	Temp°C Mini.	R.H. Morning	R.H. Evening	Rain Fall mm	Evaporation (mm)
25	JUNE 18-24	39.4	26.6	67.9	40.3	037.2	7.0
26	June-July 25-1	37.3	27	70.9	55.7	064.0	5.4
27	July 2-8	35.5	27.8	77.0	60.0	030.4	6.4
28	July 9-15	34.6	26.6	79.4	58.7	030.0	4.0
29	July 16-22	34.5	26.2	90.0	67.3	043.2	4.5
30	July 23-29	32.0	25.7	87.1	74.6	004.2	3.2
31	July-Aug. 30-5	34.5	26.6	81.7	58.3	000.0	4.7
32	Aug 6-12	32.5	26.5	79.6	77.3	092.2	2.3
33	Aug 13-19	34.7	27.3	78.0	58.0	000.0	6.6
34	Aug 20-26	35.7	26.3	86.0	56.7	019.0	3.9
35	Aug-Sep. 27-2	31.1	24.9	91.6	74.7	196.2	2.1
36	Sep. 3-9	34.5	25.5	82.4	56.4	001.6	4.4
37	Sep. 10-16	36.3	26.2	80.3	52.7	000.0	4.7
38	Sep. 17-23	33.9	24.7	85.6	59.6	072.3	3.3
39	Sep. 24-30	35.5	23.4	81.3	50.6	000.0	4.9
40	Oct. 1-7	36.1	21.1	86.7	39.7	000.0	4.5
41	Oct 8-14	36.7	21.7	82.7	33.6	000.0	4.6
42	Oct 15-21	37.1	17.7	83.4	25.7	000.0	4.7
Total						590.3	

VITA

The author of this thesis Prerana Parihar D/o Dr. Pravin Singh Parihar was born on 4 February, 1993 at District- Kawardha, Chhattisgarh.

She completed Primary education at his native place, High School education with first division in the 2009 from S.S.M. School, District- Kawardha, Chhattisgarh and Intermediate education in the year 2011 with first division from ,Vivekanand Public School Khairagarh, District- Kawardha, (C.G).

Thereafter, she joined in Sant kabir College Of Agriculture And Research Station, Kawardha in 2012 and completed B.Sc. (Ag.) Degree in 2016 with 7.75 O.G.P.A. from Indira Gandhi Krishi Vishwavidyalaya Raipur.

After completing graduation she was selected for M.Sc. (Ag.) degree programme in Plant Pathology and was admitted in College of Agriculture, Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior (M.P.). She has completed all required courses in M.Sc. (Ag.), Plant Pathology; She is submitting thesis for M.Sc. (Ag), Plant Pathology. He had passed all required courses in M.Sc. by obtaining 82 per cent marks approx.

During all the period of education, from schooling to post graduation she was very sincere and honest towards studies and works.

PRERANA PARIHAR