# EFFECT OF LIGHT AND WHEAT VARIETY ON THE GROWTH AND SPORULATION OF WHEAT BLAST PATHOGEN *MAGNAPORTHE ORYZAE TRITICUM*

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# CERTIFICATE

*This is to certify that the thesis entitled,* **"EFFECT OF LIGHT AND WHEAT VARIETY ON THE GROWTH AND SPORULATION OF WHEAT BLAST PATHOGEN *MAGNAPORTHE ORYZAE TRITICUM*".** *submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of* ***MASTER OF SCIENCE in PLANT PATHOLOGY,*** *embodies the result of a piece of bona fide research work carried out by Registration No.* ***19-10236*** *under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

*I further certify that such help or source of information, as has been availed of during the course of this investigation has duly been acknowledged.*

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***The author***

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*Dhaka*

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## ABSTRACT

The effects of the visible light namely white, NUV, blue including dark condition was evaluated on seed germination, mycelial radial growth, and conidia production of wheat blast pathogen *Magnaporthe oryzae triticum* (MoT) on three wheat varieties namely BARI Gom-26, BARI Gom-28 and Prodip. The seed germination, number of conidia per seed, number of conidia per panicle of this plant pathogenic fungus was significantly differed among these light treatments. MoT produced significantly more conidia when the fungus grew under White, NUV and Blue lights. MoT produced more conidia on BARI Gom-26 and BARI Gom-28 compared to Prodip variety. Blue light produced a greater number of conidia per rachis and per panicle where white light enhanced to produce a greater number of conidia per seed. BARI Gom-28 variety produced a greater number of conidia on rachis, seed and panicle. Whether the number of conidia counted on rachis, seed and panicle are less produced on Prodip variety. Blue×BARI Gom-28 and Dark×BARI Gom-28 resulted highest seed germination, whether the number of conidia per rachis and per panicle was higher only in the interaction effect of blue light and BARI Gom-28 variety. No significant difference of conidia production per seed was obtained in the interaction effect of White×BARI Gom-26, White×BARI Gom-28 and Blue×BARI Gom-28. The number of conidia per rachis and per panicle was comparatively lower in dark light and Prodip variety, where the number of conidia per seed was lower for both interaction effect of NUV×Prodip and Dark×Prodip treatment combination. MoT was grown on oat meal agar medium (OMA) in the White, NUV, Blue light and Dark condition. Radial mycelial growth was significantly higher under Blue light and comparatively lower in White light condition.

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# CHAPTER 1

# INTRODUCTION

Wheat (*Triticum aestivum*) is one of the world's most important grains and a leading source of calories and plant protein in human foods (Curtis *et al.* 2002). Wheat is synonymous with food for a large part of the global food security and population of developing countries (ICAR). It is the primary staple food in North Africa and the Middle East and is gaining popularity in Asia. Wheat originated from the nearby Levant region of the east but is now cultivated worldwide. It grows in more than 70 countries on 5 continents (Dixon, 2007) and is the most widely grown crop in the world. In 2017, world production of wheat was 771.7 million tonnes, making it the third most produced cereal (FAOSTAT, 2017). Wheat is grown on more than 1.5 million hectares, which is larger than any other crop. The four largest producers of wheat in 2017 are China (134.3 million tonnes), India (98.5 million tonnes), Russia (85.9 million tonnes) and the United States (47.3 million tonnes) (FAOSTAT, 2017).

Wheat blast is basically a head disorder. Common Symptoms caused by *Magnaporthe oryzae triticum* (MoT) have also been described, from small elliptical lesions to complete bleaching and empty spikes (Igarashi *et al.* 1986; Igarashi 1990). Infected plants show signs of common wheat blast with partial or less obvious bleaching of the spike Showing of diseased trees as they are blackened, with a dark border on the leaves, on the rachis of diseased plants, with gray to tan necrotic lesions often present. Additionally, in some fields, blackening of lower nodes was observed. Grains from blast-infected heads were small, shriveled, deformed, and had low test-weight leading to serious yield losses (Malaker *et al.*, 2016).

In February 2016, Bangladesh was identified as the first Asian country to have the origin of the alarming wheat blast disease caused by a South American lineage of a hemibiotrophic filamentous fungus *Magnaporthe oryzae Triticum* (MoT) pathotype (Callaway, 2016; Islam *et al.,* 2016; Malaker *et al.,* 2016). With the first emergence of Bangladesh, wheat blast has entered Asia, which accounts for about 42% of the world's wheat production. The first occurrence of wheat blast during

2015-16 cropping season was restricted to eight districts of Bangladesh (Islam *et al.,* 2016). However, the outbreak spread to eight more neighboring districts: Magura, Faridpur and Rajshahi in 2016-17, 2017-18 cropping season. According to the data of the Bangladesh Department of Agriculture Extension, total area of wheat cultivation in the blast affected districts during the year 2015-16 and 2016-17 was 99,259 and 47,278 hectares, respectively. Infected wheat fields were burnt, which contributed to a 15% reduction in wheat production in nine infected districts (Islam *et al.,* 2016; Malaker *et al.,* 2016). Despite this reduction, the total wheat production in Bangladesh has increased slightly (35000 MT, 2.7%) in the total of 2016 compared to 2015. Importantly, 100% of the seed proportions of the state-owned Bangladesh Agricultural Development Corporation (BADC) in the affected districts (private 355 Ha) were completely cleared by the Ministry of Agriculture to destroy the pathogen inoculum. Farmer wheat fields that were severely damaged (up to 100%) were also burned (Islam *et al.,* 2016). It is now clear that wheat blast disease is well established in at least one country in Asia (Bangladesh). The incidence of the disease has certainly decreased in the amount, location, and size of the infected area. However, the new incidence of infection in new regions indicates gaps in the control of wheat blast and the spread. Since this wheat killer is in progress, the need for rapid development of effective management methods, including the development of blast-resistant varieties using modern biotechnological approaches, including genome editing is essential before it becomes catastrophizing (Islam *et al.,* 2019). Castruagudin *et al*. (2016) used four culture media such as CMA (Cornmeal agar), MEA (Malt-extract agar), OA and PDA for showing the growth of PoT. They showed that maximum growth was occurred on PDA media. But that work was not sufficient to evaluate the isolates. Now a days breeding for developing blast resistant wheat variety is utmost important and subsequent evaluation of resistant cultivars by inoculation with desired inoculum potential is very challenging. Sufficient sporulation of the MoT isolates is therefore important and challenging. In the present study different light treatments were applied and evaluated on seed germination, radial mycelial growth and sporulation of MoT isolates on seeds, wheat panicles and OMA.

Keeping the review in mind the present research work was conducted with the following objectives.

1. To determine the main effect of light and wheat variety on growth and sporulation of wheat blast pathogen *Magnaporthe oryzae triticum* (MoT).

2. To determine the interaction effect of light and wheat variety on the germination and sporulation of wheat blast pathogen *Magnaporthe oryzae triticum* on seeds and panicle.

# CHAPTER 2

# REVIEW OF LITERATURE

In 1985, a blast pathogen which was a major fungal foe of rice was identified in Parana state of Brazil on wheat. Wheat blast causes enormous loss to wheat production. Further it has also been reported from Uruguay, Paraguay, some parts of Argentina. Slowly it has spread to other South American countries. The biggest shock for wheat cultivation was occurrence of blast on wheat in Bangladesh during February 2016 outside of America. It is an alarming for wheat cultivation in tropical and sub-tropical areas in the world.

## 2.1. Survey on disease incidence and severity and collection of different isolates of *Magnaporthe oryzae triticum*

According to the report of the Department of Agricultural Extension (DAE), the wheat blast infected area was about 15,000 ha, which correspond to 3.5% of total wheat fields in Bangladesh. Wheat blast was observed in eight southwestern districts, viz., Pabna, Kushtia, Meherpur, Chuadanga, Jhenaidah, Jashore, Barisal and Bhola in 2016. The severity of wheat blast disease varied among districts and among wheat varieties. A disease surveillance program on wheat blast was organized in collaboration with CIMMYT and CU, USA in mid-February 2017. Out of 103 surveyed sites, 33 sites were found infected with wheat blast. Overall disease incidence in 2017 was comparatively lower than the previous season with low disease severity (5- 10%).

Malaker *et al.* (2016) reported that the highest percentage of infected wheat blast fields was observed in Meherpur (70 %) followed by Chuadanga (44 %), Jessore (37 %), Jhenaidah (8%), Bhola (5 %), Kushtia (2 %), Barisal (1 %) and Pabna (0.2 %). The infected wheat fields were burned, which causes 15% decrease in wheat production of the eight infected districts.

Islam *et al.* (2016) stated that yield losses in different affected districts varied. The highest yield loss was reported in Jhenaidah (51%) followed by Chuadanga (36 %), Meherpur (30%), Jessore (25 %), Barisal (21 %), Pabna (18 %), Kushtia (10 %) and Bhola (5 %).

## 2.2. Significance of blast disease of wheat

Zhang *et al.* (2016) stated that the wheat blast pathogen belongs to the *Magnaporthe oryzae* (syn. *Pyricularia oryzae*) species complex. Choi *et al.* (2013) also showed that the members of this species complex cause blast disease on more than one hundred of species in the Poaceae family including rice, wheat, barley and rye. Several phylogenetic species (e.g., pathotypes) are proposed by cladistic analyses based on the multi-gene sequence and the host specificity (Choi *et al.*, 2013; Hirata, 2007; Kato *et al,* 2000; Tosa *et al.*, 2004).

Fisher *et al. (*2012) showed that, the outbreaks caused by fungal diseases have increased in frequency and are a recurrent threat to global food security. Fisher *et al.* (2012); Pennisi (2010) and Liu *et al.* (2014) also showed that, one example is blast, a fungal disease of rice, wheat and other grasses, that can destroy enough food supply to sustain millions of people. Until the 1980s, the blast disease was not known to affect wheat, a main staple crop critical to ensuring global food security.

Urashima *et al.* (2009) also found that, these grains are often discarded during the post- harvest process of threshing or winnowing. Goulart and Paiva (1992, 2000) also reported that, the yield losses up to 100% due to the cultivation of susceptible cultivars. Urashima *et al.* (2009) experimented that, wheat blast is today considered a major disease affecting wheat production in Brazil.

Goulart *et al.* (2007) reported that, wheat blast is considered a major disease affecting wheat production. The economic importance of this disease derives from the fact that the fungus can reduce yield and grain quality. Grains from blast-infected spikes from highly susceptible cultivars are often small, shriveled and deformed, with low test weight. Highest yield losses occur when spike infections begin during flowering or early grain formation. Goulart (2005) also experimented the economic importance of this disease derives from the fact that the fungus can reduce yield and the quality of the wheat grain. Infected grains from highly susceptible cultivars are usually small, wrinkled, deformed, and have low-test weight. The highest yield losses occur when infections start during flowering or grain formation. Goulart and Paiva (1992, 2000) reported yield losses in Brazil on susceptible cultivars vary from 10.5 up to 100%.

### 2.3. *Magnaporthe oryzae triticum* (MoT)

Klaubauf *et al.* (2014) examined infected plant samples using a light microscope. A hallmark of blast fungi is the production of asexual spores that have a specific morphology consisting of three-celled pyriform conidia. Microscopic analyses revealed that gray colored lesions observed on both spikes and leaves which produce a large number of three- celled pyriform conidia from aerial conidiophores.

Valent *et al.* (2021) and Zeigler (1998) observed that, *M. oryzae triticum* is a filamentous, heterothallic ascomycete that has potential for sexual and asexual reproduction; however, there is evidence that sexual fertility has been lost in some populations.

### 2.4. Variation of fungal isolates on different lights

Costa *et al.* (2021) studied the effects of the visible light wavelengths on germination, mycelial radial growth, and conidial production of the plant pathogens *Colletotrichum acutatum* and *Fusarium fujikuroi* were investigated. Bot fungi were grown on PDA in the dark or on PDA under continuous white, blue, green or red light. In addition, the conidia from each treatment were exposed to UV radiation. The germination and growth of both plant pathogenic fungi were not affected by any of the treatments. *C. acutatum* produced more conidia when the fungus grew under white and red light. *F. fujikuroi* produced more conidia in the dark. Li *et al.* (2018) reported that under enhanced UV-B radiation, the infectivity of *Magnaporthe oryzae* was decreased, which could significantly inhibit its growth and sporulation. Following inoculation with Magnaporthe oryzae, levels of disease resistance related substances in the rice leaves were significantly increased. The production, survival, propagation, invasiveness and virulence of the fungal conidia is affected by UN-B radiation. Hui *et al.* (2018) reported that incubation period was shortened, and the infection efficiency, sporulation quantity and disease index increased when UV-B radiation was performed only pre-inoculation. When healthy seedlings were inoculated using urediospores collected from wheat leaves irradiated by UV-B only post-inoculation or both pre-and post-inoculation, infection efficiency, sporulation quantity and disease index were also reduced. However, in the latter, the disease incubation period did not differ under varying UV-B radiation intensities compared to that when wheat leaves were not treated with UV-radiation.

Cheng *et al.* (2014) looked at effects of UV-B radiation on epidemiological components of wheat stripe rust caused by three physiological races of Pst-(Cyr 31, Cyr 32 and Cyr 33). The results showed that UV-B radiation prolonged the incubation period, reduced the germination rate and infection efficiency of each physiological race, and caused the decreases in the lesion expansion rate, sporulation quantity and area under disease progress curve (AUDPC).

# CHAPTER 3

# MATERIALS AND METHODS

The present research work was carried out in the laboratory condition to evaluate different light and wheat variety on mycelial growth and sporulation of *Magnaporthe oryzae triticum* (MoT) on seeds and wheat panicle. The details of the materials and methodology adopted during this study are described here under the following points.

## 3.1. Survey area

The survey was made in the farmer's wheat field which are affected by wheat blast disease at Meherpur and Mujibnagar upazila in Meherpur districts and Chuadanga sadar upazila in Chuadanga districts in Bangladesh (Figure 1).

**Figure 1. Survey area and sampling sites (●) of wheat blast infected south western region of Bangladesh**

(https://bddirectories.com/khulna-division-of-bangladesh/khulna-division-map/)

### 3.2. Survey Period

Survey was made to the selected area for the collection of wheat blast infected sample during January 2020 to April 2020.

### 3.3. Experimental Site

The experiment was conducted in the Laboratory, Department of Plant Pathology, Sher-E- Bangla Agricultural University (SAU), Dhaka-1207, Bangladesh.

### 3.4. Collection of panicle blast sample

Bleached panicles of wheat were collected from farmers' fields. Infected ears were cut from the mother plant, field dried and placed in brown paper envelopes, which were labeled with all necessary information's including the name of the region, district, localities, cultivars and date of collection. Samples were kept in refrigerator at $4^0$C until the surveys in all the districts were finalized. Then samples were transported to Plant Pathology Laboratory, SAU for pathogen identification and characterization and further experimentation.

### 3.5. Isolation, purification and identification of wheat blast isolates

The water agar (Agar 20 g with 1000 ml distilled water), and potato dextrose agar media (200g of peeled potatoes, 20 g of dextrose, and 20 g of agar and 1000 ml of distilled water) were used for the isolation of blast pathogen and Oat Meal Agar (80 g oat, 15 g agar and 1000 ml water) was used for sporulation. Diseased spikes of wheat cultivars infected with MoT were cut into suitable size (3-4 cm in size) around the area showing the blast lesion and were surface sterilized with 1% sodium hypochlorite for 1 minute followed by 3 times washes with sterile distilled water. Then the cut pieces were placed in Petri dishes lined with moist filter papers and it was incubated at $26\pm1^0$C for 24 hours to encourage sporulation. After incubation, these infected spike pieces were examined under stereo- dissecting microscope. Abundant sporulation was observed from in and around the lesions with grey, dense and bushy appearance. A sterile moistened needle was used to pick out single conidia by the needle across the sporulating lesion. The conidia were placed on water agar. After 12 hours, mycelium was visible in petri dish and then hyphal tip was placed in potato dextrose agar media plates containing Streptomycin (40 mg/L) and the plates were incubated in $26\pm1^0$C. The marginal mycelial growth that

developed subsequently was picked-up aseptically for sub-culturing until pure culture of *M. oryzae triticum* was obtained. The pure culture was maintained by sub culturing at an interval every 15 days and preserved at low temperature (4ºC) in refrigerator.

After preparing the petriplates containing OMA the mycelial block of MoT was incubated in the centre of the petridishes were incubated at 26±1°C for about 10 to 15 days with alternate 12 hours.darkness and 12 hours light for sporulation. After conidia production in OMA plates, the conidia of *M. oryzae triticum* were checked under compound microscope (Figure 2). Identification of the pathogen was carried out according to the cultural and morphological characteristics as preciously described (Agrawal *et al.*, 1989; Mew and Gonzales, 2002).

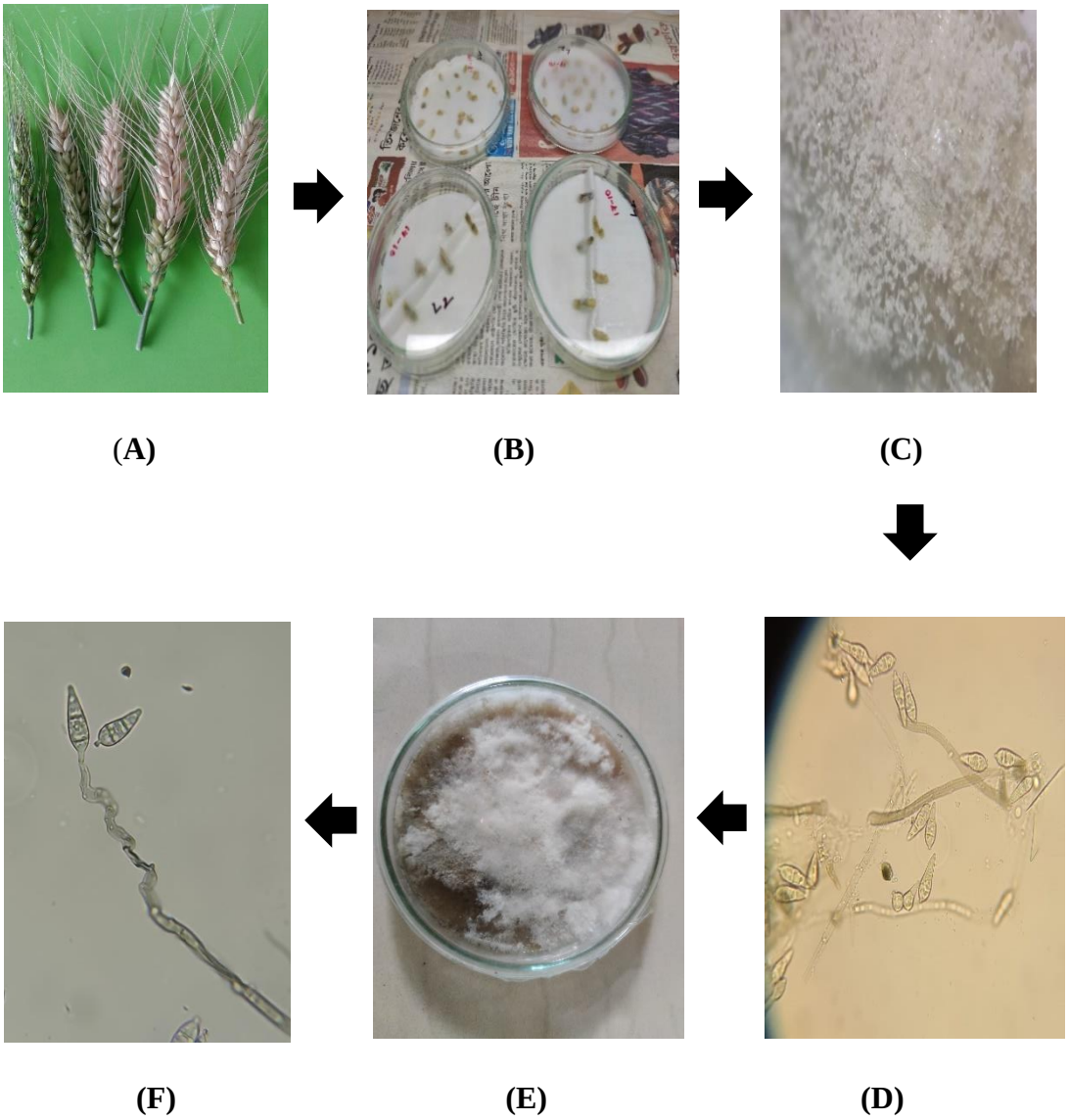



**Figure 2. Isolation and identification of *Magnaporthe oryzae triticum* (MoT). A. Bleached panicles; B. Seeds and rachis cut pieces placed on moist blotter paper; C. Sporulation of MoT on seeds (×40); D. Mycelia and conidia from seeds observed under compound microscope (×40); E. Pure culture of MoT and F. Conidia and conidiophore of MoT (×100)**

### 3.6. Treatments

The experiment was laid in two factor treatments

Factor A: Light

a. White
b. NUV
c. Blue
d. Dark

Factor B: Wheat variety

a. BARI Gom- 26
b. BARI Gom- 28
c. Prodip

### 3.7. Arrangement of light treatments

To investigate mycelial growth, sporulation, the *M. oryzae triticum* isolates were grown on OMA under different spectral light qualities at 26°C. Four different light regimes were established using 1) White light, 2) UV lights, 3) Blue light and (4) dark at 26°C as control. Different light sources were arranged in wooden rack provided different colored light. For the white light treatment, the petri dishes with cultures on OMA medium, seeds and rachis with lids in place, in a single layer (not stacked) were maintained under continuous light provided by two 15 W cool white Philips (TL-D 15 W/75–650) broad-spectrum fluorescent light bulbs suspended at a distance of 25 cm above the samples. For NUV and blue light treatments, the Petri dishes with cultures on OMA medium, seeds and rachis with lids in place, in a single layer (not stacked), were maintained under continuous NUV, blue and dark light provided within the wooden rack were adjusted to enable incubation of the cultures under different wavelengths of light. The distance of the incubator between the light source and the agar plates was 25 cm, the temperature of the incubators was adjusted to 26 °C, and no heating effect by the lights were detected. For dark treatments, all Petri dishes were maintained in the same incubator as the light treatment, but the Petri dishes were kept inside a perforated plastic box (to keep the cultures ventilated) and covered with a thick black cloth sleeve.

### 3.8. Seed germination and conidia production on seeds and rachis

Seed germination test was conducted following ISTA (1999). Seeds were threshed form panicle and 25 seeds were placed in each Petri dishes (90 × 15 mm) with moist blotter paper. The plates were kept under different kind of light treatments. In 7-8 days later, about maximum germination took place. Then all the data regarding germination were taken. After that, all the Petri dishes were checked under stereo-microscope. MoT infected seeds were marked with pencil and counted.

For taking the data regarding conidia production, about 16 Petri dishes were taken. 8 dishes were set with moist blotter paper and 25 seeds were placed in each dish, where the rest 8 dishes were set with moist blotter paper and single rachis cut for every dish. After 3-4 days later, all seeds and pieces of rachis were taken in test-tubes for making solution with 2 ml of distilled water in each test-tube for every respective Petri-dishes with marking the treatment. The seeds and rachises were vortexed for two minutes to separate conidia from seeds and rachises in water solution. From the solution, 5 µL was pipetted and taken in hemocytomter for spore counting. For each treatment, four Petri dish replicates were prepared on the same day.

### 3.9. Mycelial radial growth of MoT

From the colonies of *Magnaporthe oryzae triticum* on OMA medium in Petri dishes (90 × 15 mm), one disk of 5 mm diameter was removed with a cork borer and placed in the center of Petri dish containing 23 ml of OMA. The plates were kept at 26 °C for the treatments: dark (control), and white, NUV or blue light. The fungi *Magnaporthe oryzae triticum* was grown for 15 days. Colony diameter of mycelial growth was measured on the 15th day horizontally and vertically (at a perpendicular axis). For each treatment, six Petri dish replicates were prepared on the same day.

### 3.10. Measurement of conidial production

To measure conidial production under different treatment conditions, three agar plugs (per plate) were removed with a cork borer (5 mm dm) at different places on the medium surface with an even coverage of conidia, and the conidia were suspended in 2 ml sterile Tween 80 (0.1%) solution. After the conidial suspensions

were vigorously shaken on vortex shaker, the conidial concentrations were determined by hemocytometer counts. Each experiment was replicated three times and each experiment was repeated thrice.

### 3.11. Pathogenicity tests for *M. oryzae triticum* (MoT) isolates

The pathogenicity test of the isolates was done for further confirmation of pathogenic isolates of wheat blast pathogen. The pathogenicity test of *M. oryzae triticum* was confirmed by Koch's postulates using the method of Chevalier *et al.* (1991). The pot was prepared for this test using sterilized soil. The soil was collected from near field. Disinfected viable seeds of BARI Gom-24 (Pradip) susceptible to wheat blast variety were sown in pots with 6-7 seeds per pot. The plants were inoculated after germination, at the age of 3-4 leaves and the seedlings in each pot was sprayed with 40– 50 ml of spore suspension adjusted to $10^4$ spores/ml with the help of hemocytometer. The conidial suspensions were sprayed on to the wheat seedlings until runoff while water was used for spraying the control treatment. Inoculated pots were covered with polythene bags. The plants were placed inside the dew chamber at $26\pm1^0$C for 7 days. Periodical observations were made for the development of symptoms on the leaves starting 7 days after inoculation. Experiments were done with three replications. The fungus was re-isolated from the artificially inoculated wheat seedlings leaves showing typical blast symptom.

### 3.12. Statistical analysis

The effect of White, NUV, Blue and Dark light treatments on germination, sporulation and mycelial growth of *Magnaporthe oryzae triticum* was assessed with analysis of variance of a two-way factorial analysis. All analyses were carried out with the statistical program Staistix 10 (Trial version). The treatment means were compared by LSD.

# CHAPTER 4

# RESULTS AND DISCUSSION

### 4.1.1. Main effect of light and wheat variety on the germination (%) of wheat seed

The germination percentage (%) of three wheat varieties was recorded under different kind of light i.e. White, NUV and Blue (Figure 3 and Figure 4) etc. The germination percentage (%) varied significantly due to the application of different kind of light (Table 1). Statistically highly significant result was observed regarding germination percentage (%) by the Blue light treatment (Table 1). Germination percentage (%) under Blue light treatment gave the highest (90.22%) seed germination, while the White light treatment gave the lowest (66.67%) seed germination.

Seed germination of different wheat variety under different light treatments was significantly varied from one to another (Table 1). Statistically higher (88.33%) seed germination was observed by BARI Gom- 28. Prodip variety gave the lowest (68.33%) seed germination.

**Table 1.** Main effect of light and wheat variety on germination percentage (%) of wheat seed

| **Light** | **Germination %** |
|---|---|
| White | 66.67 c |
| NUV | 77.33 b |
| Blue | 90.22 a |
| Dark | 81.33 ab |
| **Variety** | **Germination %** |
| BARI Gom-26 | 80.00 a |
| BARI Gom-28 | 88.33 a |
| Prodip | 68.33 b |

**Figure 3. Experimental set up of different variety of wheat seed under different light treatment arranged in wooden rack.**

**Figure 4. Germination test of wheat seeds (ISTA, 1999)**

### 4.1.2. Interaction effect of light and wheat variety on the germination (%) of wheat seed

Interaction effect between light intensity and variety was found significant in respect of seed germination (Table 2). Maximum seed germination (98.67%) was obtained for Blue × BARI Gom-28 treatment, which was statistically similar with NUV × BARI Gom-26, NUV × BARI Gom-28, Blue × BARI Gom-26, Blue × Prodip and Dark × BARI Gom-28. Germination percentage (%) was significantly lower (48.00 %) for Prodip variety under White light treatment (White × Prodip).

**Table 2.** Interaction effect of light and wheat variety on germination percentage (%) of wheat seed

| Treatments (Light × Variety) | Germination % |
|---|---|
| White × BARI Gom-26 | 76.00 bc |
| White × BARI Gom-28 | 76.00 bc |
| White × Prodip | 48.00 d |
| NUV × BARI Gom-26 | 82.67 abc |
| NUV × BARI Gom-28 | 82.67 abc |
| NUV × Prodip | 66.67 c |
| Blue × BARI Gom-26 | 85.33 ab |
| Blue × BARI Gom-28 | 98.67 a |
| Blue × Prodip | 86.67 ab |
| Dark × BARI Gom-26 | 76.00 bc |
| Dark × BARI Gom-28 | 96.00 a |
| Dark × Prodip | 72.00 bc |

### 4.2.1. Main effect of light and wheat variety on the incidence (%) of *Magnaporthe oryzae triticum* on wheat seeds

Considering the main effect of light on the incidence of MoT (%) varied significantly (Table 3). Maximum *Magnaporthe oryzae triticum* incidence (5.51%) was obtained for Blue light treatment, which was statistically similar with White, NUV and Dark light treatment (Table 3).

Statistically highly significant result was observed regarding *Magnaporthe oryzae triticum* incidence (%) by BARI Gom-26 variety (Table 3). Mot incidence (%) of BARI Gom-26 gave the highest (7.77%) result, while BARI Gom-28 variety gave the lowest (1.87 %) MoT incidence.

**Table 3.** Main effect of light and wheat variety on incidence % of *Magnaporthe oryzae triticum*

| **Light** | **Mot incidence (%)** |
|---|---|
| White | 4.27 a |
| NUV | 4.27 a |
| Blue | 5.51 a |
| Dark | 3.20 a |
| **Variety** | **Mot incidence (%)** |
| BARI Gom-26 | 7.77 a |
| BARI Gom-28 | 1.87 b |
| Prodip | 3.30 b |

### 4.2.2. Interaction effect of light and wheat variety on the incidence (%) of *Magnaporthe oryzae triticum* on wheat seeds

Interaction effect between light intensity and variety was found significant in respect of MoT incidence (%) on wheat seeds.

Maximum % incidence (12.80) was obtained for White × BARI Gom-26 treatment, which was statistically similar with NUV × BARI Gom-26, Blue × BARI Gom-26, Blue × BARI Gom-28 and Dark × Prodip. Mot incidence (%) was significantly lower for White × BARI Gom-28, White × Prodip, NUV × BARI Gom-28, Blue × Prodip, Dark × BARI Gom-26 and Dark × BARI Gom-28 (Table 4).

**Table 4.** Interaction effect of light and wheat variety on incidence (%) of *Magnaporthe oryzae triticum*

| Treatments (Light × Variety) | Mot incidence (%) |
|---|---|
| White × BARI Gom-26 | 12.80 a |
| White × BARI Gom-28 | 0.00 c |
| White × Prodip | 0.00 c |
| NUV × BARI Gom-26 | 9.20 ab |
| NUV × BARI Gom-28 | 0.00 c |
| NUV × Prodip | 3.60 bc |
| Blue × BARI Gom-26 | 9.07 ab |
| Blue × BARI Gom-28 | 7.47 ab |
| Blue × Prodip | 0.00 c |
| Dark × BARI Gom-26 | 0.00 c |
| Dark × BARI Gom-28 | 0.00 c |
| Dark × Prodip | 9.60 ab |

### 4.3.1. Main effect of light and wheat variety on the number of conidia per rachis (×10⁴)

Main effect of light on the number of conidia of *Magnaporthe oryzae triticum* per rachis varied significantly (Table 5 and Figure 5). Statistically highly significant result was observed regarding number of conidia by the Blue light treatment. Number of conidia under Blue light treatment gave the highest (102.00 $\times 10^4$), while the White light treatment gave the lowest (40.33 $\times 10^4$) result, which was statistically different from all other light treatment (Table 5).

Statistically highly significant result was observed regarding number of conidia of *Magnaporthe oryzae triticum* per rachis ($\times 10^4$) of BARI Gom- 28 variety (Figure 5). Number of conidia per rachis of BARI Gom- 28 gave the highest ($130.00 \times 10^4$), while Prodip variety gave the lowest ($15.00 \times 10^4$) number of conidia (Table 5).

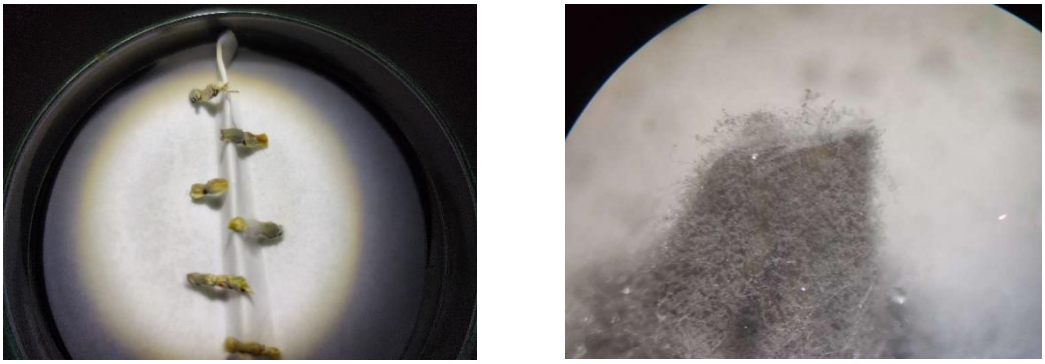
**Table 5.** Main effect of light intensity and wheat variety on number of conidia of *Magnaporthe oryzae triticum* per rachis

| **Light** | **Number of conidia per rachis (×10⁴)** |
|---|---|
| White | 40.33 b |
| NUV | 42.44 b |
| Blue | 102.00 a |
| Dark | 41.33 b |
| **Variety** | **Number of conidia per rachis (×10⁴)** |
| BARI Gom-26 | 24.83 b |
| BARI Gom-28 | 130.00 a |
| Prodip | 15.00 c |

### 4.3.2. Interaction effect of light and wheat variety on the number of conidia of *Magnaporthe oryzae triticum* per rachis (×$10^4$)

Interaction effect between light and variety was found significant in respect of number of conidia of *Magnaporthe oryzae triticum* per rachis (Table 6 and Figure 5).

Maximum number of conidia per rachis (270.00×$10^4$) was obtained for Blue × BARI Gom-28 treatment. Number of conidia per rachis (×$10^4$) was significantly lower (2.00×$10^4$) of Prodip variety kept under Dark treatment (Dark × Prodip), which was statistically similar with White × BARI Gom-26, NUV × BARI Gom-26, NUV × Prodip and Blue × BARI Gom-26 (Table 6).



**(A)** **(B)**

**Figure 5. Conidia production of *Magnaporthe oryzae triticum.* A. on rachis; B. Habit characters of MoT on rachis (×40).**

**Table 6.** Interaction effect of light and wheat variety on number of conidia of *Magnaporthe oryzae triticum* per rachis

| **Treatments (Light × Variety)** | **Number of conidia per rachis ($\times 10^4$)** |
|---|---|
| White × BARI Gom-26 | 6.00 g |
| White × BARI Gom-28 | 90.00 c |
| White × Prodip | 25.00 f |
| NUV × BARI Gom-26 | 9.33 g |
| NUV × BARI Gom-28 | 110.00 b |
| NUV × Prodip | 8.00 g |
| Blue × BARI Gom-26 | 12.00 fg |
| Blue × BARI Gom-28 | 270.00 a |
| Blue × Prodip | 25.00 f |
| Dark × BARI Gom-26 | 72.00 d |
| Dark × BARI Gom-28 | 50.00 e |
| Dark × Prodip | 2.00 g |

### 4.4.1. Main effect of light and wheat variety on the number of conidia per seed (×10⁴)

Main effect of different light on the number of conidia of *Magnaporthe oryzae triticum* per seed ($\times 10^4$) varied significantly (Table 7).

Statistically highly significant result was observed regarding number of conidia ($\times 10^4$) by the White light treatment (Table 7). Number of conidia ($\times 10^4$) at White light treatment gave the highest ($4.95 \times 10^4$), while the Dark treatment gave the lowest ($2.27 \times 10^4$) number of conidia.

Considering the main effect of wheat variety, statistically highly significant result was observed regarding number of conidia of *Magnaporthe oryzae triticum* per seed ($\times 10^4$) by BARI Gom-28 (Table 7). Number of conidia per seed of BARI Gom-28 was the highest ($5.96 \times 10^4$), while Prodip variety gave the lowest ($0.92 \times 10^4$) conidia per seed.

**Table 7.** Main effect of light and wheat variety on number of conidia of *Magnaporthe oryzae triticum* per seed

| **Light** | **Number of conidia per seed (×10⁴)** |
|---|---|
| White | 4.95 a |
| NUV | 3.85 b |
| Blue | 3.52 c |
| Dark | 2.27 d |
| **Variety** | **Number of conidia per seed (×10⁴)** |
| BARI Gom-26 | 4.06 b |
| BARI Gom-28 | 5.96 a |
| Prodip | 0.92 c |

### 4.4.2. Interaction effect of light and wheat variety on the number of conidia of *Magnaporthe oryzae triticum* per seed

Interaction effect between light intensity and variety was found significant in respect of number of conidia of *Magnaporthe oryzae triticum* per seed (Table 8 and Figure 6). Maximum number of conidia per seed ($6.72\times10^4$) was obtained for Blue × BARI Gom-28 treatment, which was statistically similar with White × BARI Gom-26 and White × BARI Gom-28. Number of conidia per seed ($\times10^4$) was significantly lower ($0.08\times10^4$) of Prodip variety under Dark treatment (Dark × Prodip), which was statistically similar with White × BARI Gom-26, NUV × BARI Gom-26, NUV × Prodip and Blue × BARI Gom-26 (Table 8).

**Table 8.** Interaction effect of light and wheat variety on number of spores of *Magnaporthe oryzae triticum* per seed

| Treatments (Light × Variety) | Number of conidia per seed ($\times10^4$) |
|---|---|
| White × BARI Gom-26 | 6.72 a |
| White × BARI Gom-28 | 6.72 a |
| White × Prodip | 1.40 e |
| NUV × BARI Gom-26 | 5.68 b |
| NUV × BARI Gom-28 | 5.68 b |
| NUV × Prodip | 0.20 f |
| Blue × BARI Gom-26 | 1.84 d |
| Blue × BARI Gom-28 | 6.72 a |
| Blue × Prodip | 2.00 d |
| Dark × BARI Gom-26 | 2.00 d |
| Dark × BARI Gom-28 | 4.72 c |
| Dark × Prodip | 0.08 f |

### 4.5.1. Main effect of light and wheat variety on the number of conidia per panicle (×10⁴)

The number of conidia of *Magnaporthe oryzae triticum* per panicle ($\times 10^4$) of three wheat varieties was recorded under different kind of light namely White, NUV and Blue etc. The number of conidia ($\times 10^4$) varied significantly due to the application of different kind of light. Statistically highly significant result was observed regarding number of conidia ($\times 10^4$) by the Blue light treatment (Table 9). Number of conidia ($\times 10^4$) at Blue light treatment gave the highest (190.00 $\times 10^4$), while the Dark treatment gave the lowest (98.00 $\times 10^4$) number of conidia (Table 9). The number of conidia of *Magnaporthe oryzae triticum* per panicle ($\times 10^4$) showed a general trend of gradual increase with the changing intensity of light (Figure 6).

**(A)** **(B)**

**Figure 6. Conidia production of *Magnaporthe oryzae triticum* seeds and panicle. (A. Set up of seeds and rachis cut under different kind of lights; B. Conidia production of *Magnaporthe oryzae triticum* on seed (×40).**

Considering the main effect of wheat variety, statistically significant result was observed regarding number of conidia of *Magnaporthe oryzae triticum* per panicle ($\times 10^4$) by BARI Gom-28 (Table 9). Number of conidia per panicle of BARI Gom-28 gave the highest ($279.00 \times 10^4$), while Prodip variety gave the lowest ($38.00 \times 10^4$) conidia per panicle.

### 4.5.2. Interaction effect of light and wheat variety on the number of conidia of *Magnaporthe oryzae triticum* per panicle

Interaction effect between light and variety was found significant in respect of number of conidia of *Magnaporthe oryzae triticum* per panicle (Table 10). Maximum number of conidia per panicle ($438.00 \times 10^4$) was obtained for Blue × BARI Gom-28 treatment. Number of conidia per panicle was significantly lower ($4.00 \times 10^4$) of Prodip variety under Dark treatment (Dark × Prodip), which was statistically similar with NUV × Prodip (13.00×104) (Table 10).

**Table 9.** Main effect of light and wheat variety on number of conidia of *Magnaporthe oryzae triticum* per panicle

| **Light** | **Number of conidia per panicle ($\times 10^4$)** |
|---|---|
| White | 164.00 b |
| NUV | 139.00 c |
| Blue | 190.00 a |
| Dark | 98.00 d |
| **Variety** | **Number of conidia per panicle ($\times 10^4$)** |
| BARI Gom-26 | 126.00 b |
| BARI Gom-28 | 279.00 a |
| Prodip | 38.00 c |

**Table 10.** Interaction effect of light and wheat variety on number of spores of *Magnaporthe oryzae triticum* per panicle

| **Treatments (Light × Variety)** | **Number of conidia per panicle ($\times 10^4$)** |
|---|---|
| White × BARI Gom-26 | 174.00 c |
| White × BARI Gom-28 | 258.00 b |
| White × Prodip | 60.00 f |
| NUV × BARI Gom-26 | 151.00 d |
| NUV × BARI Gom-28 | 252.00 b |
| NUV × Prodip | 13.00 g |
| Blue × BARI Gom-26 | 58.00 f |
| Blue × BARI Gom-28 | 438.00 a |
| Blue × Prodip | 75.00 f |
| Dark × BARI Gom-26 | 122.00 e |
| Dark × BARI Gom-28 | 168.00 cd |
| Dark × Prodip | 4.00 g |

### 4.6. Effect of light on radial mycelial growth (mm) of *Magnaporthe oryzae triticum*

The radial mycelial growth (mm) of MoT isolates was recorded under different kind of light i.e. White, NUV, Blue and Dark (Figure 7). Radial growth (mm) at Blue light treatment was the highest (69.67 mm), which was statistically similar with Dark treatment (65.00 mm) (Figure 7 and Figure 8). While the White light treatment gave the lowest (42.58 mm) mycelial growth (Figure 8). MoT radial growth showed a general trend of gradual increase with the decreasing intensity of light (Figure 8).

**(A)** **(B)**

**Figure 7. Radial mycelial growth of *Magnaporthe oryzae triticum* (A. Pure culture of Mot); (B. Some pure culture of MoT under light treatments)**

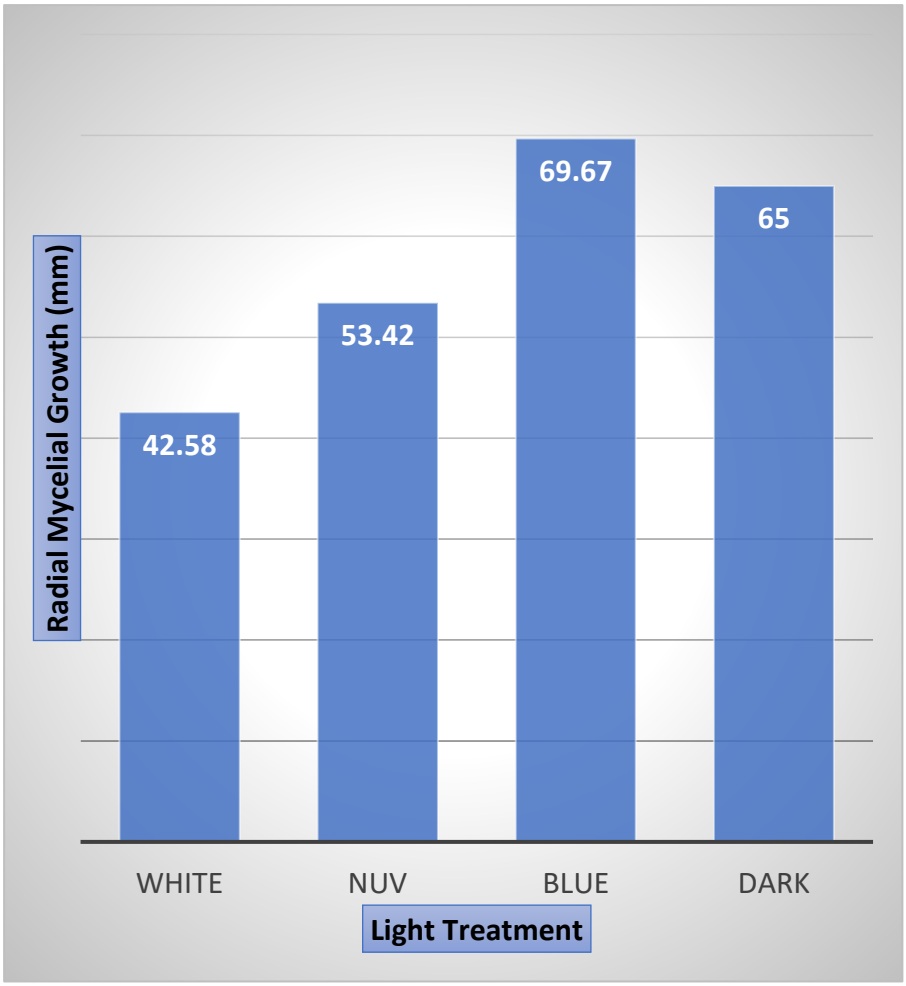



**Figure 8. Effect of different light sources on radial mycelial growth (mm) of *Magnaporthe oryzae triticum* on OMA**

### 4.7. Effect of different light on conidia formation

Different abiotic light of different color and wavelength had significant effect on mycelia growth and conidiation of MoT on OMA (Figure 9). Maximum number of conidia/cm$^2$ of OMA was produced under white light ($5.3\times10^3$) followed by UV-light and blue light. Minimum number of conidia ($2.0\times10^3$) was produced in dark condition without light.

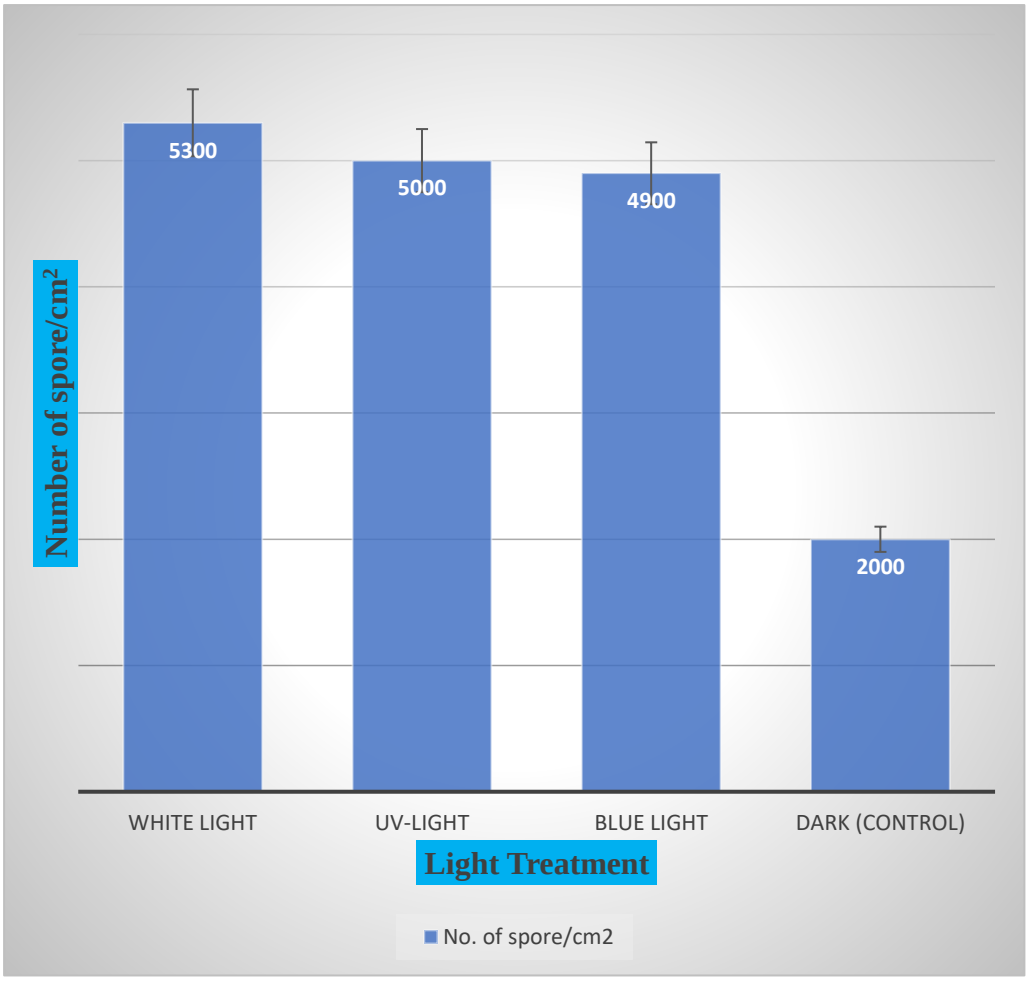



**Figure 9. Effect of different light sources on conidia production of *Magnaporthe oryzae triticum* on OMA.**

### 4.8. Pathogenicity test of *Magnaporthe oryzae triticum* (MoT)

MoT isolates were found pathogenic upon inoculation on wheat cv BARI Gom-24. The MoT isolates produced spindle shaped lesions on artificially inoculated wheat seedlings. MoT isolates produced two septate three celled pyriform conidia on inoculated leaf, when incubated on moist chamber.

Veloso *et al.* (2017) observed that *Copaifera oblongifolia* seeds sown under high light intensity had a lower germination percentage than seeds sown under low light intensity and darkness. In this study seeds treated with different light sources had significant effect on seed germination and MoT sporulation on seeds and panicles.

In another study the plant-pathogenic fungi *Colletotrichum acutatum* and *Fusarium fujikuroi* control several aspects of their physiology in response to light though light did not affect the germination and mycelial growth of these fungi. It has been reported that *Botrytis cinerea* grows less under light (Canessa *et al.,* 2013) emphasized that the effect of light on fungal genera is more important for reproduction in compare to vegetative growth (Gottlieb, 1950; Lilly and Barnett, 1951; Cochrane, 1958). Conversely, the conidial production as well as the conidial stress tolerance differs according to the different light treatments during mycelial growth.

*Colletotrichum acutatum* produced more conidia under white and red light than in darkness, and blue and green light generated the least conidia (de Menezes *et al.*, 2015) though in our study blue light generated more conidia. *Fusarium fujikuroi* produced more conidia in the dark however, *F. verticillioides* cultures grown under white, blue, yellow, green, and red wavelengths produce more conidia than the cultures grown in the dark (Fanelli *et al.,* 2012). Like our study conidiation is also stimulated by light in the wild type strain IMI58289 of *F. fujikuroi* (Avalos *et al.*, 1985; Avalos and Estrada, 2010), but in a different wild type strain of the same species mycelial growth in the dark produced more conidia than under light (Estrada and Avalos, 2008; Estrada and Avalos, 2009; Avalos and Estrada, 2010). Estrada and Avalos (2008, 2009) found that growth in the dark of the same *F. fujikuroi* isolate (FKMC 1995) generated more conidia than growth in the light. The results of our study didn't agree with Estrada and Avalos, in which our isolate produced less conidia in the dark. The reason behind this may be due the variation of response among fungal genera that have different physiological requirements of light intensity and light color.

In another study growth under white light also improved conidial UV radiation tolerance of C. *acutatum* and *F. fujikuroi* though *C. acutatum* conidia produced in the light that were similarly tolerant to UV radiationas conidia produced in the dark in the lowest UV irradiances, at the highest UV irradiances, conidia produced under light were more tolerant than conidia produced in the dark. Mycelial growth under blue, green or red light generated conidia less tolerant than conidia produced in the dark. Similarly, growth of *C. acutatum* colonies under low irradiance of white light increased conidia and mucilage production and conidia produced under the light were two- times more tolerant to UV radiation (Menezes *et al.,* 2015). *C. acutatum* is more pigmented under white and blue light, while the least pigmentation was observed in mycelia incubated

in the dark (Yu *et al*., 2013). Blue light also enhances melanin production, enhancing virulence of *C. acutatum* (Yu *et al*., 2013). Moreover, the effect of green and red light stimulates less melanin production than blue light, leading to reduced disease severity (Yu *et al.,* 2013) but in our study we didn't record melanin production of MoT isolates.

For *F. fujikuroi,* growth under white and blue light produces conidia two-fold more tolerant to UV radiation than conidia produced in the dark and under green and red light. Growth under illumination upregulates many stress genes that are important for producing conidia with increased stress tolerance (Wu *et al.,* 2014; Brancini *et al.,* 2019; Dias *et al.,* 2020). Light also promotes higher resistance of *Aspergillus fumigatus* against exogenous oxidative stress and enhances resistance to acute ultraviolet radiation (Fuller *et al.,* 2013). *M. robertsii* conidia produced under white light exhibit higher tolerance to osmotic stress (Dias *et al.,* 2020), heat (Rangel *et al.,* 2011; Rangel *et al.,* 2015), and UV radiation (Rangel *et al.,* 2011; Rangel *et al.,* 2015; Dias *et al.,* 2020).

In addition, exposure of fast-growing mycelia of *M. acridum* to white, blue or UV-A wavelengths induces tolerance to subsequent UV-B irradiation. However, red light induced lower mycelial tolerance to subsequent UV-B irradiation (Brancini *et al*., 2016). This observation may indicate that red light represses genes for tolerance to stress. Therefore, pathogenic fungi use environmental cues to prepare their conidial offspring against challenges in the environment (Rangel *et al*., 2008; Rangel, 2011; Rangel and Roberts, 2018; Medina *et al.,* 2020). Producing offspring more tolerant to the same or other stress conditions (Rangel *et al.,* 2004; Rangel *et al.,* 2011) as well as enhancing their virulence (Oliveira *et al.,* 2018; Oliveira and Rangel, 2018) will support greater dispersal distances under daytime conditions.

Many studies report effects of UV-B radiation on host plants or pathogens individually, but few studies look at the combined effects. In another study it has been found that the UV-B radiation both pre and post inoculation may affect the susceptibility of wheat and the pathogenicity of Pst. Pre inoculation UV-B radiation may increase the susceptibility of wheat, and then aggravate disease occurrence. Post-inoculation UV-B radiation may reduce the pathogenicity of Pst and decrease disease occurrence. Furthermore, wheat stripe rust was reduced when UV-B radiation was carried out both pre and post inoculation. The effect of UV-B radiation on pathogenicity of Pst was greater than on the susceptibility of wheat.

Studies on the effects of post-inoculation UV-B radiation on wheat stripe rust have also been conducted. Li *et al*. (2008) showed that wheat stripe rust severity can be reduced by post- inoculation UV-B radiation, and they suggested this finding may be related to direct damage of UV-B radiation to *Pst*.

In contrast, Hui *et al*. (2018) reported that post-inoculation UV- B radiation had no effect on wheat stripe rust. In another study, post-inoculation UV-B radiation prolonged the incubation period and reduced the infection efficiency, sporulation quantity and disease index, resulting in decreased wheat stripe rust occurrence. Although UV- B radiation affected both the host plant and the pathogen, the effect of UV-B radiation on *Pst* was greater than on the host plant. The results of other study demonstrate that pre- inoculation UV-B radiation on wheat seedlings could reduce damage of post-inoculation UV-B radiation to the pathogenicity of *Pst* to a certain extent. Pre-inoculation UV-B radiation may cause stress in wheat plants, leading to increased activity of oxygen scavenging systems which reduce the level of reactive oxygen species (ROS) in order to reduce damage from UV-B radiation (Cheng *et al.*, 1999).

Changes in epidemiological components of wheat stripe rust caused by urediospores collected from UV-B irradiated wheat leaves were also previously investigated. When healthy wheat seedlings were inoculated using urediospores collected from wheat leaves irradiated with UV-B only post inoculation, the incubation period was prolonged and the infection efficiency, sporulation quantity, and disease index decreased. Post- inoculation UV-B radiation reduced infectivity of *Pst*, which may be related to reduction of *Pst* vitality due to UV-B radiation (Cheng *et al*., 2014). When healthy wheat seedlings were inoculated using the urediospores collected from the wheat leaves irradiated both pre-and post-inoculation, *Pst* pathogenicity was influenced and wheat stripe rust was reduced. Moreover, under the same UV-B radiation distance, total sporulation quantity was greater on leaves inoculated with urediospores from pre- and post- inoculation UV-B radiation plants compared to those with the post-inoculation treatment only.

In another study, Jiang *et al*. (2013) performed stress treatments on *Pst*-inoculated wheat seedlings. They showed that UV-B radiation had no effect on wheat stripe rust, but also that the interaction of enhanced $CO_2$ concentration and UV-B

radiation had no effect on the wheat disease in terms of epidemiological components including infection efficiency, incubation period, lesion expansion rate and AUDPC. Enhanced UV-B radiation can influence the infectivity and sporulation of *Magnaporthe oryzae*, alleviate the disease and reduce the spread and diffusion of rice blast. UV-B radiation can stress the rice, induce the formation of a defence system and indirectly improve the resistance of rice. In other study, the enhanced UV-B radiation was within the tolerance range of the plants, which would not affect the normal physiological activities of the plants, but could improve the sensitivity of the plants' resistance to stress so that they could quickly respond to pathogen infections. The combination of pathogenic fungi and UV-B radiation changed the disease incidence and disease index of the traditional rice variety in a Yuanyang terrace. With an enhancement in UV-B radiation in the moderate range, the rice disease resistance increased because of the adjustment of physiological activities. Meanwhile, UV- B radiation inhibited the pathogenic fungi and alleviated the rice blast. When the irradiation exceeded the tolerance range of rice, the rice responded to the dual stresses of pathogen and UV-B radiation, and the growth energy distribution was unbalanced.

To date, most studies related to the effects of UV-B radiation on plant diseases were conducted in controlled environments. However, artificial light differs from natural light, and factors such as temperature and humidity may also differ from those in natural environments. Therefore, further studies on the effects of different field environmental factors on plant diseases should be conducted in the future. Moreover, mechanisms of the effects of different radiations on sporulation of MoT isolates on wheat diseased leaf and bleached panicle and the effect of different light intensity and light color on virulence and dissemination of spore and subsequent disease development need further investigation.

# CHAPTER 5

# SUMMARY AND CONCLUSION

The study aimed to determine seed germination, sporulation and radial mycelial growth of *Magnaporthe oryzae triticum* under different lights. The result from this research provided useful information for the development of strategies for the future control of wheat blast in field condition.

The response of the fungi expressed the effect of exposure to light varies with the variation in the light intensity employed. The growth of *Magnaporthe oryzae triticum* and conidia production was affected by changing the color of light exposure. In case of Blue light, germination was maximum while it was maximum for BARI Gom-28 wheat variety, which is statistically similar with BARI Gom-26 variety. Total interaction effect was maximum in case of Blue × BARI Gom-28, was the highest than the other light treatment. Maximum seed germination was obtained for Blue × BARI Gom-28 treatment, which was statistically similar with NUV × BARI Gom-26, NUV × BARI Gom-28, Blue × BARI Gom-26, Blue × Prodip and Dark × BARI Gom-28.

MoT incidence (%) has different reactions to light and crwheat variety. The incidence (%) was maximum for Blue light treatment, which was statistically similar with White, NUV and Dark light. In case of crop variety, BARI Gom- 26 gave the highest incidence (%) of MoT. The interaction effect was also varied due to light and crop variety, maximum % incidence was obtained for White × BARI Gom-26 treatment, which was statistically similar with NUV × BARI Gom-26, Blue × BARI Gom-26, Blue × BARI Gom-28 and Dark × Prodip.

Sporulation of *Magnaporthe oryzae triticum* was also varied due to effect of light and wheat variety. Sporulation was maximum under Blue light in rachis and panicle, while Sporulation was maximum under White light in seed. But, sporulation in rachis, seed and panicle was always maximum for BARI Gom-28 variety. Interaction effect was maximum for Blue × BARI Gom-28 in case of sporulation in rachis, seed and panicle.

Radial mycelial growth was also affected due to different kind of lights. *Magnaporthe oryzae triticum* gave the maximum radial growth under Blue light, which was statistically similar with Dark light.

In order to further investigate the differences among the growth and sporulation of *Magnaporthe oryzae triticum*, more isolates should be collected from other geographical locations from different variety. In addition, research should also be expanded to field conditions and associated with the study of wheat blast management.

# CHAPTER 6
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