# Imposition of *Pyricularia oryzae* Stress on Growth and Yield of BRRI dhan29 and IR 64



**A THESIS**

**BY**

**FATIMA CHOWDHURY**

**STUDENT ID: MS-230822**

**A Thesis (Course no:081108 PLP 6118) Submitted to the Agrotechnology Discipline in Partial Fulfillment of the Requirements for the Degree of Master of Science in Plant Pathology**

**AGROTECHNOLOGY DISCIPLINE**

**LIFE SCIENCE SCHOOL**

**KHULNA UNIVERSITY**

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DEDICATED

TO

MY BELOVED FAMILY

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JANUARY 2025 The author

# Table of Contents

# ABSTRACT

Rice blast, caused by *Pyricularia oryzae*, is one of the most significant fungal diseases of rice, affecting growth and yield worldwide. The impact of *P. oryzae* on two rice genotypes, BRRI dhan29 and IR 64 was selected based on their resistance level against *Pyricularia oryzae.* The experiment was carried out under controlled conditions during the Boro season in order to assess morphological, physiological, and yield-related parameters in both genotypes artificially inoculated and non-inoculated *Pyricularia oryzae* stress. The results of this experiment show that *Pyricularia oryzae* exhibits severe stress in both genotypes with regard to plant height, tiller number, fresh and dry biomass, and leaf area. BRRI dhan29 showed greater susceptibility compared to IR64 in all studied morphological and physiological parameters. Under inoculated conditions, IR 64 maintained greater effective tiller number $plant^{-1}$ (23) than BRRI dhan29 (11). In case of dry weight of biomass IR 64 showed a reduction of 13.42%, while BRRI dhan29 displayed a higher reduction of 19.95%. IR 64 had fewer unfilled grains (391) compared to BRRI dhan29 (600) under *P. oryzae* inoculated conditions. Grain filling drastically affected by *Pyricularia oryzae* stress in BRRI dhan29 (44.73%) compared to IR 64 experienced a smaller reduction (11.45%). Only 12.5% reduction was found in IR 64 considering thousand grain weight, whereas BRRI dhan29 exhibited a more significant reduction (37.60%). Yield-contributing traits, including panicle length, filled grain number, and thousand-grain weight, were also adversely affected which lead to a higher yield loss in BRRI dhan29 (32%) compared to IR64 (10.96%). Leaf Area and leaf greenness by SPAD showed significant difference ($P<0.05$) in both rice genotypes at 15, 30, 45 and 60 days after inoculation. After 15 days of inoculation, IR 64 showed reductions in leaf area and leaf greenness of 8.21% and 4.13%, respectively, but BRRI dhan29 showed reductions in leaf area of 30.67% and leaf greenness of 34.25%. BRRI dhan29 had shown 50.94 % and 80.44% Percent Disease Index (PDI) respectively at 10 and 20 days after inoculation, which is noticeably greater than IR 64.Under *Pyricularia oryzae* stress, IR64 maintains better physiological performance and yield stability.

Keywords: BRRI dhan29, IR 64, *Pyricularia oryzae,* Stress, Growth, Yield

# List of Figures

# List of Tables

# Chapter I

# Introduction

Rice (*Oryza sativa* L.) belonging to the family gramineae, is a basic crop grown in 113 countries, providing income for around 100 million households in Asia and Africa (Nepune and Bhusan,2020). It is a key staple food and one of the main sources of revenue and jobs. According to reports, Asia, where a sizeable portion of the world's population lives, accounts for around 90% of worldwide rice production and consumption (www.fao.org; accessed on 20 January 2022). Rice is the most widely grown cereal grain in Bangladesh, occupying 28754 acres of cultivable land and yielding 39095 metric tons (Yearbook of Agricultural Statistics, 2023). Bangladesh's economy is centered on rice, which contributes roughly 20% of the country's GDP and one-sixth of its total national revenue (Rahman and Uddin, 2017).

Bangladesh's rice production is divided into *Aus*, *Aman*, and *Boro* seasons, with *Boro* growing exclusively in the irrigated dry season. Modern varieties account for 99% of *Boro*'s 5.00 million hectares, with higher yields during winter due to lower insect and disease populations, increased solar radiation, and improved water management (About BRRI, 2023). The Bangladesh Rice Research Institute (BRRI) has released 113 high-yielding rice varieties, including BRRI dhan28 and BRRI dhan29, which are highly susceptible to blast disease (Hossain et al., 2017). The IRRI-developed indica cultivar IR 64, introduced in the Philippines in 1985, has been successful in its early maturity, disease resistance, and exceptional cooking quality. It has been rapidly spread and cultivated across over 10 million hectares in two decades. Rice production is influenced by both biotic and abiotic factors, with fungal infections reducing yearly production by 14% globally (Mackill and Khush, 2018). Rice blast disease is a major biotic stress affecting rice yield and is being intensively studied due to its economic threat to world rice production (Onyango, 2014).
Rice blast disease is caused by the fungus *Pyricularia oryzae* [Telemorph: *Magnaporthe oryzae* (Hebert) Barr.,]. This disease has the potential to reduce rice yield by 70% to 80% (Nasruddin and Amin, 2013; Miah et al., 2013). This disease can destroy entire rice plants in 15 to 20 days and cause production losses of up to 100% under favorable conditions (Dhruw and Sahu, 2023). As a result, it is regarded as one of the most significant barriers to expanded rice production (Nasruddin and Amin, 2013). The *Pyricularia oryzae* might attack at any of the growth stages of

the rice plant from seedling to the pre-maturity stage of the crop as well as all aerial parts of the plant and causes lesions on them, including the leaf blade, leaf sheath, ligule, collar region, stem, panicle (rachilla and neck) and grain (hull), and restricts yield potential under environments favoring the disease (Rahman and Uddin, 2017). The *Pyricularia oryzae* is most common on leaves, causing leaf blast during the vegetative stage of growth, or on neck nodes and panicle branches during the reproductive stage, causing neck blast (Asibi et al., 2019).

The incidence of rice blast was recorded on *Boro* season and Transplanted *Aman* in all over Bangladesh (Wang et al., 2014). In recent years, the frequency of explosion occurrence in Bangladesh has increased due to the country's expansion into new territories (the north and northwest). The most popular and massive varieties reported as highly susceptible to blast illness are BRRI dhan29 and BRRI dhan28 (Anonymous, 2011). Furthermore, all local and enhanced aromatic rice types produced in the rainy season are susceptible to neck blast (Rahman and Uddin, 2017). This disease is found in 85 nations across all continents where rice is grown, in both paddy and upland environments. The damage is heavily influenced by environmental factors; climatic changes, particularly water scarcity, encourage researchers to consider developing production technologies for rice cultivation under lower water conditions, which may increase the incidence of many rice diseases, particularly those caused by *Pyricularia oryzae* (Rahman et al., 2017).

*Pyricularia oryzae* infection is a significant threat to rice plants during critical growth stages, including seedling, early tillering, and heading phases. During the early growth phase, the Pyricularia oryzae primarily affects leaves, leading to symptoms known as leaf blast (Rahman and Uddin, 2017). Later stages, neck and node blast manifest as necrotic lesions, causing severe damage. Neck blast is more devastating, directly disrupting grain filling and leading to complete yield loss in extreme cases.Blast disease outbreaks often result in catastrophic losses, including total seedling destruction in nurseries and large-scale epidemics in fields (Khan et al., 2014). Cultural management practices, such as planting resistant cultivars, applying fungicides, and adjusting planting schedules, fertilizers, and irrigation, are commonly employed to control rice blast. However, breeding disease-resistant or tolerant rice varieties has proven to be the most effective and sustainable strategy (Nurgoho et al., 2021).Significant progress has been made in developing disease-resistant varieties, contributing to increased global rice yields. Over 100 rice blast resistance genes have been identified, derived predominantly from japonica (45%) and indica (51%) cultivars, with a smaller proportion originating from wild rice species (Sharma et al., 2012). However, the adaptability of *Pyricularia oryzae* often limits the durability of resistance, as single-gene resistance tends to break down within three to five years of release. This study aims to evaluate and compare the morphological, physiological, and yield performance of BRRI dhan29 and IR 64 under rice blast stress. The findings will support the development of sustainable strategies to manage rice blast and enhance rice production in Bangladesh and other blast-prone regions.

Though most field research on rice blast disease has been undertaken in tropical and subtropical conditions, specific information on the effect of blast disease on growth and grain production of rice in Bangladesh is scarce. As a result, the current study was done to explore the influence of blast disease on growth and yield-contributing characteristics of two rice variety, BRRI dhan29 and IR 64 in Bangladesh.

So the objective of this study was to investigate the effect of blast disease caused by *P. oryzae* on morphological, physiological, and yield performance of BRRI dhan29 and IR64.

# Chapter II

# Review of literature

## 2.1 Rice Blast Occurrence and Distribution

Rice blast disease has been identified in 85 rice-growing nations (Wang et al., 2014). Blast is the most damaging rice disease due to favorable environmental circumstances for its occurrence and spread. Rice blast disease has been documented in rice-growing countries worldwide, including China, Japan, America, India, and Australia (Shafaullah et al., 2011). It has been identified as a damaging disease in West Africa, Iran (Mousanejad et al., 2010), Malaysia (Rahim, 2010), and South America's Savanas (Karamian et al., 2015). Rice blast was first documented in Africa in 1930 (Neupune and Bhusal, 2020). Global warming's impact on climate may spread globally (Kohli et al., 2011). Blast-resistant rice varieties often lose their resistance within a few years due to fungal strain changes (Huang, 2011).

In Myanmar, the occurrence of rice blast disease in Ayeyarwady (Central Agricultural Research Institute [CARI], 2000), leaf blast epidemic and neck blast around Yezin area on the variety IR50 in 2002-2003 cold and dry seasons, the occurrence of blast disease in 2013 early summer and 2014 rainy season in Nay Pyi Taw Union Territory (Aye et al., 2015), the leaf blast and neck blast occurrence in Aungban, Pindaya, Taunggyi, Kyaukme, Yezin, Lapputa and Bago during 2015 to 2018 rice growing season (Khaing et al., 2018) and leaf blast disease incidence in Aungban research farm every year (Department of Agricultural Research [DAR], 2018) has been reported.

## 2.2 Economic importance

*Pyricularia oryzae*, a filamentous fungus, causes the disease, which has been recorded in over 85 countries worldwide. Rice blast outbreaks have caused 50-90 percent grain yield losses globally (Agrios, 2005). According to Thu (2019), neck blast is more damaging than leaf blast in farmers' fields as it leads to higher output losses. Rice blast disease has caused significant crop losses in several rice-growing countries. Panicle infection leads to full yield loss.

The first known epidemic of blast in India in 1918 resulted in a 69% loss of rice crop. Blast epidemics in Malaysia and the Philippines resulted in yield decreases of 50-70%. In India, sensitive cultivars lost 75% of their grain while in the Philippines, thousands of hectares were affected, resulting in more than 50% yield loss and 40% grain loss in Nigeria (Thu,2019).

Neck blast infection reduces yield by twice as much as leaf blast. Rice blast can lead to crop loss in favorable weather conditions, including warmth and humidity (Thu, 2019). In Nepal, the disease reduces yield by 10-20% in sensitive types and up to 80% in severe cases. In rice-growing areas, a blast outbreak can result in a loss of 35-50% of production, and in severe cases, up to 100% (Thu, 2019).

Blast disease is the most devastating fungal disease of rice encountered by farmers in Nigeria. In rice-growing areas, a blast outbreak could cause the loss of about 35–50 % of rice yield, and in a serious outbreak of the disease, up to 100 % of yield could be lost (Thu, 2019). Rice blast is the most harmful fungal disease causing rice yield losses up to 70 to 80 % (Miah et al., 2013; Nasruddin and Amin, 2013).

### 2.3 Impact of *P. orayzae* on growth and yield of rice

Neupane and Bhusal (2020) conducted an experiment in Nepal. According to them, blast disease causes symptoms at all stages, from seedling in the nursery to heading in the field. However, the seedling, tillering, and panicle initiation stages are the most destructive. Blast symptoms impact leaves, nodes, necks, collars, panicles, rachis, and glumes. Every year, rice blast causes 10-30% of global yield losses. The disease reduces yields by 10-20% in susceptible types, but can reach up to 80% in severe conditions, as shown in Nepal. Blast fungus thrives in cloudy conditions, high relative humidity (93-99%), low night temperatures (15-20°C), and prolonged dew periods.

Asibi et al. (2019) stated that, Rice blast, a devastating fungal disease affecting rice (*Oryza sativa* L.), poses a danger to world food security. Rice is widely studied due to its importance in production and consumption, as well as its global distribution and destructive potential. Rice blast, produced by *Pyricularia oryzae* Cavara 1892 (A), can infect aboveground tissues of rice plants at any growth stage, resulting in crop failure. The Pyricularia oryzae causes lesions on leaves (leaf blast), collars (collar blast), culms (culm nodes), neck nodes (neck rot), and panicles (panicle blast). The color and shape of the lesions vary based on varietal resistance, climatic circumstances, and age. Understanding the impact of environmental circumstances on rice blast at the cellular and genetic levels is crucial for making informed decisions and managing the illness in the future.

*Magnaporthe grisea* (anamorph: *Pyricularia grisea*), a fungal Pyricularia oryzae , limits rice yield in major rice-growing regions worldwide. *Magnaporthe oryzae*, the most serious rice blast fungus, poses a continuing threat to global food security. Nonetheless, the fungus has developed as a model experimental organism for studying plant infection mechanisms caused by Pyricularia oryzae ic fungus. This hemibiotrophic Pyricularia oryzae infects epidermal cells, creating lesions on leaves, leaf collars, culms, nodes, and panicles, leading to seed filling failure. After successful penetration, the invasive hyphae grow rapidly in the host cells and caused blast lesions. in 5 to 7 days, the Pyricularia oryzae produces numerous conidia from the lesions and initiates a new infection cycle (Karamian et al., 2015).

In some lowland rice cultivars, neck blast disease causes a decrease in grain yield and quality. Neck blast disease in rice reduces yields both in number and quality. The study conducted by Subiadi et al. (2019) examined how neck blast disease affects grain attributes and yields in three high producing varieties of lowland rice. The types used included Ciherang, Inpari 4, Inpari 7, Inpari 8, and Inpari 9. The characteristics studied included the frequency and severity of neck blast illness, the seed size ratio between healthy and sick panicles, and predicted production. Neck blast disease affected all planted kinds, however Ciherang and Inpari 8 were more sensitive, Inpari 7 and 9 were moderate, and Inpari 4 was resistant. Neck blast illness affected panicle grain quality, resulting in lower grain width and substantial differences compared to healthy panicles across all kinds. The Inpari 4 variety had the highest projected productivity of 10,378 kg/ha but saw a 35.86% decline in production. The Ciherang variety had the lowest estimated productivity of 8,367 kg $ha^{-1}$ but experienced a 48.37% decrease in production after being infected with neck blast disease.

An experiment was done by Khan et al. (2014) to determine the impact of neck blast disease on yield-contributing features and seed quality traits in aromatic rice in Bangladesh. The Plant Pathology Division of the BRRI in Gazipur, Bangladesh, collected healthy and neck-blast-infected panicles from three aromatic rice cultivars, including high-yielding and local varieties. All evaluated types were particularly sensitive to neck blast disease in natural settings, but no leaf blast signs were observed on the leaves. Neck blast disease caused increased grain sterility, decreased grain size, yield, and seed quality. Disease severity and variety genetics influenced yield and seed quality reductions. Unfilled grains were the primary source of seed-borne

Pyricularia oryzae s, particularly blast in the seed lot. Blast Pyricularia oryzae transmission from the neck (panicle base) to seed was minimal. These findings are significant, particularly for seed certification programs that rely on field inspections. Controlled experiments are necessary to reach more accurate conclusions.

Rice blast disease is a serious constraint on rice production and threatens world food security. Disease-related rice production losses result in economic losses for farmers and an increase in global rice prices due to insufficient supply compared to consumer demand. The annual illness losses were anticipated to feed more than 60 million people. Researchers have extensively investigated the disease due to its significant impact on rice production and global spread (Agbowuro et al., 2020).

Rice (*Oryza sativa* L.) is the primary grain crop consumed by more than 50% of the global population. It accounts for 23% and 50% of total calories consumed by the global and Nepalese population, respectively. Rice blast is the most damaging abiotic agent impacting rice, resulting in 70-80% yield loss. This disease developed in China approximately 7000 years ago. In Nepal, it was first documented in Thimi, Bhaktapur in 1966. *Magnaporthe oryzae*, also known as *Pyricularia oryzae*, is the filamentous ascomycete fungus responsible for this condition. The infection affects all stages of plant development, causing symptoms on leaves, collars, necks, panicles, and glumes. It decreases rice production by enough to feed 60 million people annually. Cloudy weather, high relative humidity (93-99%), low night temperature (15- 20°C), longer duration of dew is the most favorable condition for the outbreak of disease (Simkhada and Thapa 2021).

In 2012 and 2013, Koutroubas et.al. (2015) looked at how *Pyricularia oryzae*, the cause of rice blast disease, affected plant development, grain output, and grain quality in a Mediterranean environment. By early blast fungal inoculation and natural infection, four rice types were grown under high and low disease pressure, respectively. The results demonstrated that inoculation had an impact on plant growth and yield in general and led to a decrease in plant height, the quantity of productive tillers per plant, grain weight, and grain yield. The severity of the decline varied according to how susceptible each variety was to the illness. Grain yield and both forms of the illness (leaf blast and neck blast) exhibited a negative correlation. Each unit increase in the neck blast and leaf blast ratings was predicted to reduce production by 5.97 and 0.23 g plant$^{-1}$, respectively. In contrast to agronomic variables, traits related to grain quality were less impacted by disease treatments. The disease's most noticeable impact was on total milling yield, which in some cases was lowered by immunization by up to 11%.

According to Zewdu (2021), rice blast (*Magnaporthe grisea*) was initially discovered in China and later in Africa in 1922. The disease has become the most pervasive and destructive rice disease globally. The disease can result in modest production decline or entire crop loss, depending on the variety and severity degree. The rice blast isolate is named *Magnaporthe grisea* and is closely linked to other blast-like fungus isolates. Rice blast fungus initiates infection when a three-celled conidium settles on the rice leaf surface. A single lesion can yield thousands of spores within 15 days of infection. Infected leaves have little brown necrotic lesions that grow to be elliptical or spindle-shaped, pale to gray, with darker margins. Infected seeds show brown patches, possibly caused by infection of the florets as they mature into seeds. It requires a minimum of 12 hours of moderate temperatures (25-30°C), high relative humidity (90-92%), and high wetness for development. To manage the illness, use resistant types, combine disease management methods, and apply necessary nitrogen and silicon fertilizers. The rice plant has two responses to disease: either the fungus cannot cause sporulating lesions on the plant, or the plant develops residual resistance that persists even after the *Pyricularia oryzae* has been defeated.

# Chapter III

# Materials and Methods

This chapter presents a brief description of the experimental period, experimental site, climatic conditions, soil, experimental design and layout, treatments, crop growing procedure, harvesting and post harvesting operation, data collection, experimental measurement and statistical analysis which are described below under different headings and sub- headings.

## 3.1 Experimental site and duration

The present experiment was conducted at Plant Pathology Laboratory and net house of Plant Breeding and Biotechnology Laboratory, Agrotechnology Discipline, Khulna University, Khulna. The geographical elements of the trial site are latitude at 22°84' N and longitude of 89°54' E and 9 m elevation.

## 3.2 Duration of the experiment

The experiment extended for three months in *Boro* season. It was conducted from December 2023 to April 2024.

## 3.3 Climatic condition

Khulna has a tropical monsoon climate which is characterized by a distinct rainy and dry period. Most of the rain falls from May till September. However, precipitation figures are much lower than in areas with a monsoon climate. Temperatures are warm all year round. During the rainy period, temperatures may rise a few degrees. Its annual average rainfall is 1,878.4 mm (73.95 in), with about 87 percent falling between May and October. Khulna also receives heavy rain from cyclones that form in the Bay of Bengal. The region has an annual average temperature of 26.3°C (79.3°F), with monthly averages ranging from 11.4°C (52.5°F) on January mornings to 34.6°C (94.3°F) during April afternoons (Khatun et al., 2016). The climate at the experimental site is categorized as AEZ 11, High Ganges River Floodplain (Bappy and Haque, 2019).

### 3.4 Collection of Seed

Seeds of BRRI dhan29 and IR 64 where the first one is blast disease susceptible and the other one is blast disease resistant variety. Seeds were collected from BRRI, Gazipur

### 3.5 Collection of Fungal Isolate

A virulent *P. oryzae* strain (RL-17) was used in this experiment. It was preserved in silica gel in a refrigerator at -80° c in the Plant Protection Laboratory of Agrotechnology Discipline, Khulna University.

### 3.6 Seed treatment

At first the seeds were taken in a clean and dry petridish. Then the seeds were treated with Vitavax-200@ 0.3% (w/w). Seeds were mixed with Vitavax-200 in petridishes for one hour (Shahrin et al., 2016). The treated seeds were then plated on water soaked filter papers in petridish for germination.

### 3.7 Soil and Pot Preparation

The soil filled in the pot was collected from the non-saline zone of the Khulna division. The physical and chemical properties of collected soil were clay loam textured, pH- 6.6, EC- 1.02, Soil organic matter- 1.87%, available N- 1140.5 ppm, available P- 15.04 $\mu g\ g^{-1}$, available S- 176.8 $\mu g\ g^{-1}$ with an inherent exchangeable K, Ca, and Mg content at 0.65 Cmol $kg^{-1}$, 16.5 Cmol $kg^{-1}$, and 6.5 Cmol $kg^{-1}$ soil, respectively. After 7 days of sun drying, the soil was gently crushed with a hammer to break up large particles. After removing undesired elements such as dried roots, grass, and stones, the soil was properly ground.15 kg of soil were placed in each plastic pot with compost and sand in a ratio of 2:1:1 and the pot size was (30.5×30.5×74) cm. Before adding soil, plastic pots were rinsed with tap water and dried in the sun. The experiment was designed using the factorial CRD with five replications. Each pot was considered as one replication. Each pot received equal amounts of fertilizer (urea, TSP, MoP, and gypsum) at rates of 1.3, 0.5, 0.6, and 0.4 g $pot^{-1}$ for nitrogen, phosphorus, potassium, and sulfur (Rahman et al.,2007). Before 2 days of transplantation, the first split dose of urea and full doses of all other fertilizers were manually mixed into the soil. The second split dose of urea was used after 35 days (at maximum tillering stage).

### 3.8 Germination and transplanting of seeds

The treated seeds were placed on soaked filter paper on petridish for germination and later incubated at 28°C in the dark for 5 days until they germinate. Afterwards, two seedlings were sown into each plastic pot. Before transplanting the soil was watered thoroughly before to ensure it is evenly moist but not waterlogged. The pots were watered gently using a sprinkler to avoid displacing the seeds.

### 3.9 Culture of *P.oryzae*

Initially, the *P. oryzae* strain was grown for 7 days at 23°C on a Potato Dextrose Agar (PDA) culture medium. A mycelial lawn with a diameter of 2 mm was cut with a borer, transferred to an oat meal medium, and cultivated for 14 days in the dark at 23°C. After 14 days on oat agar media, mycelial development was scraped using sterilized toothbrushes. Scrapped petridishes were covered with transperant plastic and properly ventilated for air flow, and they were kept under continuous light at 24°C for 72 hours. *P.oryzae* sporulation was detected using a microscope. After confirming sporulation, the plates were gently scraped with a painting brush. The spore suspensions were filtered using double layered gauze cloth. Spore suspensions were used as inoculants at a concentration of $3\times10^5$ m $L^{-1}$, as measured by a cell counting chamber. Few drops of Tween20 and distilled water were added to the spore suspension to make it easier to spray spores onto leaves.

### 3.10 Inoculation using *P.oryzae*

Plants were infected with *P.oryzae* at 35 days of age (emergence of the eighth leaf from the main tiller), with the exception of the control plants. The seedlings were sprayed with a fine mist from a spray bottle. Following that, the treated trays were transported to a growth chamber at 24°C with a 24-hour dark period. After 24 hours, the trays were moved to the net house.

### 3.11 Experimental Design

The experiment was laid out following by using factorial completely randomized design with two factors and five replications. Factor A consisted of two rice genotypes (IR 64 and BRRI dhan29) and factor B was two status of inoculation (non-inoculated and inoculated).

### 3.12 Statistical analysis

Results were expressed as means ± standard deviations (SDs). Tukey's range tests at a 0.05 probability level were conducted to detect differences between treatments by using Statistix 10 computer program.

### 3.13 Data collection

The following data were collected during the study period:

- Plant Height (cm)
- Total Tillers Plant$^{-1}$ (No.)
- Effective Tillers Plant$^{-1}$ (No.)
- Leaf Number
- Leaf Area ($m^2$)
- Fresh Weight (gm)
- Dry Weight (gm)
- Root Length (cm)
- Leaf Greenness by SPAD
- Percentage of Disease Index
- Panicle length (cm)
- Panicle number
- Sterile grain panicle$^{-1}$ (No.)
- Filled grain panicle$^{-1}$ (No.)
- Grain length (mm)
- Grain width (mm)
- 1000 grain weight (g)
- Grain yield (t ha$^{-1}$)

#### 3.13.1 Plant Height

One plant was taken from each pot and data of plant height was taken in centimeters (cm) from the base part of the plant to the tip of the tallest panicle of each plant and the average of the 5 plants were taken.

### 3.13.2 Tiller Number Plant$^{-1}$

The number of tillers plant$^{-1}$ was recorded from 5 plants and the average was calculated. It includes both effective and non-effective tillers.

### 3.13.3 Effective Tiller Number Plant$^{-1}$

The number of panicle-bearing tillers plant$^{-1}$ was counted from 5 plants, and the average was calculated. The panicle, which had at least one grain, was considered an effective tiller.

### 3.13.4 Leaf Number Tiller$^{-1}$

Total leaf number of the plant was counted in four stages- 15 days after inoculation, 30 days after inoculation and 45 days after inoculation and finally 60days after inoculation.

### 3.13.5 Panicle Length

Panicle length was recorded in centimeters (cm) from the base node of the rachis to the apex of each panicle. In addition, the length of 5 panicles plant$^{-1}$ was recorded and the average was calculated.

### 3.13.6 Unfilled Grains Panicle$^{-1}$

Grain was considered to be sterile if any kernel was not present therein. The total unfilled grain number of previously selected panicle plant$^{-1}$ was counted from 5 plants, and the average was calculated.

### 3.13.7 Filled Grains Panicle$^{-1}$

Grain was considered to be filled if any kernel was present therein. The total filled grain number was counted from 5 plants, and the average was calculated.

### 3.13.8 Grain Length

The grain length was taken with the help of a digital slide caliper. The length of the grain was recorded in millimeters (mm).

### 3.13.9 Grain Width

The width of the grain was taken with the help of a digital slide caliper. The width of the grain was recorded in millimeters (mm).

### 3.13.10 Thousand Grain Weight

One thousand cleaned grains were counted randomly from each sample and weighed using a digital electric balance. The grain retained about 14% moisture at that stage, and the mean weight was expressed in grams (g).

### 3.13.11 Total Yield

The grains were cleaned, dried, and then recorded weight with the help of an electric balance. Finally, grain yield was determined and expressed as t $ha^{-1}$.

### 3.13.12 Fresh Weight of Biomass

One plant was uprooted from the soil and cleaned thoroughly. Then after removing excess water, the plant was weighted with digital balance. The data was recorded in gram.

### 3.13.13 Dry Weight of Biomass

The uprooted plant was dried in sun for 72 hours and then the weighted of dried plant was taken with digital balance in gram unit.

### 3.13.14 Root Length

The plant was gently uprooted from the soil, ensuring safe removal with as much of the root system as possible without breaking it. Then the roots were soaked in a bucket of water for 5–10 minutes to loosen any adhering soil particles and washed carefully. The roots were spread on a flat surface and the primary root or the longest root was identified. Then a measuring tape was placed alongside the root. Data was measured from the base of the plant (crown region) to the tip of the longest root centimeters (cm).

### 3.13.15 Leaf Area

A fresh leaf was taken from plant. Then it was laid on a smooth surface. The length of the leaf was measured with a measuring tape in cm unit and recorded. Then from the widest part of the leaf, the width was measured in the same process. After that to measure leaf area, the length and width was put in a formula. The formula is given below:

$$\text{Leaf area} = \text{Leaf length} \times \text{Leaf width} \times K$$

On the formula. K is a correction factor for the shape of the leaf (commonly K= 0.75 for rice leaves, as they are not perfect rectangles). The leaf area was measured in $cm^2$ unit. Leaf area of the plant was counted in four stages- 15 days after inoculation, 30 days after inoculation, 45 days after inoculation and finally 60 days after inoculation.

### 3.13.16 Leaf Greenness by SPAD

For measuring chlorophyll content the topmost fully expanded leaf (usually the flag leaf) was chosen. This is critical because it is the most photosynthetically active part of the plant. After ensuring the leaf is clean and dry to avoid measurement errors, the SPAD meter clamp was placed on the middle section of the leaf blade, avoiding the midrib. The midrib can affect light transmittance and lead to inaccurate readings. The sensor was aligned so it captures the leaf surface properly. Then the clamp was closed gently to avoid damaging the leaf. The SPAD reading displayed on the device was recorded. This process was repeated multiple readings (3–5) per leaf and the average was calculated. This ensures the data is representative of the actual chlorophyll content. For measuring 5 plants, the same process was repeated for each plant. Chlorophyll Content of the plant was counted in four stages- 15 days after inoculation, 30 days after inoculation, 45 days after inoculation and finally 60 days after inoculation.

### 3.13.17 Percentage of Disease Index (PDI)

The leaves were observed for typical blast symptoms (elliptical lesions with gray centers and brown margins) and a score was assigned to each leaf based on a standard disease severity scale. PDI of the plant was counted in two stages- 10 days after inoculation and 20 days after inoculation. The Standard Evaluation System (SES) for rice developed by the International Rice Research Institute (IRRI) is commonly used. It provides a 0–9 scoring scale based on the percentage of infected leaf area.

Table 1. Standard scoring system for leaf blast based on the dominant type of infection

| Lesion type (score) | Description | Reaction |
|---|---|---|
| 0 | No lesion observed | Resistant (R) |
| 1 | Small brown specks of pinpoint size or larger brown specks without sporulating center | Resistant (R) |
| 3 | Small, roundish to slightly elongated necrotic sporulating spots, about 1-2 mm in diameter with a distinct brown margin or yellow hallow | Resistant (R) |
| 5 | A narrow or slightly elliptical lesion, 1–2 mm in breadth, more than 3 mm long with a brown margin | Moderately Resistant (MR) |
| 7 | A broad spindle-shaped lesion with a yellow, brown, or purple margin | Susceptible (S) |
| 9 | Rapid coalescing small, whitish, grayish, or bluish lesions without distinct margins | Susceptible (S) |

The PDI can be calculated using the formula (James,2003):

$$\text{PDI}=\frac{\text{Sum of all numerical ratings}}{\text{Total number of leaves or plants observed}\times\text{Maximum disease score}}\times 100$$

### 3.13.18 Relative Performance

The relative performance between the control and inoculated data were measured by following formula (Saha et al.,2010):

$$RP = \frac{\text{Variables measured under inoculated condition} - \text{Variables measured under control condition}}{\text{Variables measured under control condition}} \times 100$$

# Chapter IV

# Results

This chapter compares the morphological, physiological and yield responses of two rice genotypes, IR 64 and BRRI dhan29 which were evaluated under non-inoculated (control) and *Pyricularia oryzae* inoculated conditions. The measured parameters include plant height, tiller number, effective tiller number, fresh weight, dry weight, root length include panicle length, panicle number, sterile and filled grains, thousand grain weight, grain dimensions, and total yield under control and inoculated conditions.

## 4.1 Plant Height

Plant height was significantly reduced ($P<0.05$) in both rice genotypes under *Pyricularia oryzae* inoculated conditions (Table 2). The genotype IR 64 exhibited a 5.19% reduction in plant height compared to the control, whereas BRRI dhan29 showed a more pronounced reduction was 13.38%. IR 64 showed a smaller reduction (90.4 cm to 85.7 cm) compared to BRRI dhan29 (86.82 cm to 75.2 cm). These results suggest that BRRI dhan29 is more susceptible to *Pyricularia oryzae* induced stress in terms of plant height reduction compared to IR 64.

## 4.2 Tiller Number Plant$^{-1}$

*Pyricularia oryzae* inoculation significantly ($P<0.05$) reduced the tiller number in both genotypes (Table 2). BRRI dhan29 exhibited a substantial reduction of 25%. IR 64 maintained a higher number of tillers (24) compared to BRRI dhan29 (12) under *Pyricularia oryzae* inoculated conditions. The considerable decline in tiller number in BRRI dhan29 showed its reduced tillering ability under *Pyricularia oryzae* stress compared to IR 64.

## 4.3 Effective Tiller Number Plant$^{-1}$

A similar trend was observed for effective tiller numbers. Effect of *Pyricularia oryzae* inoculation significantly ($P<0.05$) observed in case of effective tiller number in both genotypes

(Table 2). BRRI dhan29 exhibited a substantial reduction (26.67%). IR 64 maintained a higher number of effective tillers (23) compared to BRRI dhan29 (11) under *P. oryae* stress conditions.

### 4.4 Fresh Weight of Biomass

Fresh weight decreased significantly (*P*<0.05) in both rice genotypes under *P. oryzae* inoculated conditions (Table 2). IR 64 showed a smaller reduction (99.95 g to 92.72 g), while BRRI dhan29 experienced a larger decrease (73.18 g to 62.48 g). IR 64 demonstrated a relatively smaller reduction of 7.23%, whereas BRRI dhan29 experienced a greater decrease of 14.62%.

### 4.5 Dry Weight of Biomass

Dry weight was also significantly reduced (*P*<0.05) in both genotypes under *P. oryzae* inoculated conditions (Table 2). IR 64 showed a smaller reduction (42.24 g to 36.57 g), while BRRI dhan29 experienced a larger decrease (37.4 g to 28.94 g). The genotype IR 64 exhibited a 13.42% reduction in dry weight, while BRRI dhan29 displayed a higher reduction of 19.95%.

### 4.6 Root Length

The root length of both control and *P. oryzae* inoculated plants of IR 64 showed no statistically significant difference. It was same in case of BRRI dhan29. (Table 2). IR 64 showed a smaller reduction (33.38 cm to 29.64 cm) compared to BRRI dhan29 (30.18 cm to 23.02 cm). BRRI dhan29 experienced reduction of 30.62%. The highest root length was recorded in the control condition of IR 64 (33.38 cm).

Table 2. Impact of *Pyricularia oryzae* on morphological and physiological parameters of IR 64 and BRRI dhan29

| Genotype | Treatment | Plant Height (cm) | RP % | Tiller No | RP % | Effective Tiller No | RP % | Fresh weight (gm) | RP % | Dry weight (gm) | RP % | Root length (cm) | RP % |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| IR 64 | Control | 90.4 a | | 25 a | | 25 a | | 99.95 a | | 42.24 a | | 33.38 a | |
| | Inoculated | 85.7 b | 5.19 | 24 b | 4 | 23 a | 8 | 92.72 b | 7.23 | 36,57 b | 13.42 | 29.64 a | 11.20 |
| BRRI dhan29 | Control | 86.82 b | | 16 b | | 15 b | | 73.18 c | | 37.4 b | | 30.18 a | |
| | Inoculated | 75.2 c | 13.38 | 12 c | 25 | 11 c | 26.67 | 62.48 d | 14.62 | 28.94 c | 19.95 | 23.02 b | 30.62 |
| CV (%) | | 1.10 | | 4.74 | | 7.24 | | 0.15 | | 1.23 | | 11.05 | |

RP= Relative Performance

Values with different letters are significantly differed at 5% level of probability according to Tukey's HSD All-Pairwise Comparisons Test

### 4.7 Panicle Length

There was no statistically significant difference was observed between panicle lengths of the control and inoculated plant by *P. oryzae* of IR 64. BRRI dhan29 also showed the same trend in case of panicle length reduction (Table 3). IR 64 showed a smaller reduction (34.56 cm to 30.2 cm) compared to BRRI dhan29 (30.4 cm to 21.62 cm). BRRI dhan29 had experienced reduction of 28.88%. The highest root length was recorded in the control condition of IR 64 (34.56 cm).

### 4.8 Panicle Number

The control and inoculated plants by *Pyricularia oryzae* of two rice genotype- BRRI dhan29 and IR 64 showed no significant differences in case of panicle number (Table 3). The highest panicle number of IR 64 was 35 in control condition whereas it was 23 in case of BRRI dhan29 also in control condition.

### 4.9 Unfilled Grain

Unfilled grain was also significantly reduced ($P<0.05$) in both genotypes under inoculated conditions (Table 3). IR 64 had fewer sterile grains (391) compared to BRRI dhan29 (600) under *P. oryzae* inoculated conditions.IR 64 exhibited a smaller increase (34.62%) in sterile grain numbers compared to BRRI dhan29 (51.89%).

### 4.10 Filled Grain

Filled grain decreased significantly ($P<0.05$) in both rice genotypes under inoculated conditions (Table 3). IR 64 maintained a higher number of filled grains (1,160) compared to BRRI dhan29 (420). IR 64 experienced a smaller reduction (11.45%) compared to BRRI dhan29, which showed decrease of 44.73%.

### 4.11 Thousand Grain Weight

The weight of thousand grains decreased significantly ($P<0.05$) under *Pyricularia oryzae* inoculated conditions in both genotypes (Table 3). IR 64 showed a higher grain weight (2.8 g) than BRRI dhan29 (1.46 g) under inoculation. IR 64 showed a higher thousand grain weight, showing a reduction of 12.5%, whereas BRRI dhan29 exhibited a more significant decrease (37.60%). This indicates better grain weight stability in IR 64 under stress conditions.

### 4.12 Grain Length and Width

Both grain length and width were reduced significantly ($P<0.05$) under *Pyricularia oryzae* inoculated conditions (Table 3). The highest grain length of IR 64 was 5.7 in control condition whereas it was 4.02 in case of BRRI dhan29 also in control condition. The highest grain width of IR 64 was 2.3 in control condition whereas it was 1.4 in case of BRRI dhan29 also in control condition.

### 4.13 Total Yield

Total yield decreased significantly for both genotypes under *Pyricularia oryzae* inoculated conditions (Table 3). IR 64 showed a smaller reduction (8.76 t $ha^{-1}$ to 7.8 t $ha^{-1}$) compared to BRRI dhan29 (7.5 t/ha to 5.1 t/ha). IR 64 demonstrated a smaller reduction (10.96%) in total yield compared to BRRI dhan29, which showed a significant reduction of 32%.

Table 3. Impact of *Pyricularia oryzae* on yield contributing parameters of IR 64 and BRRI dhan29

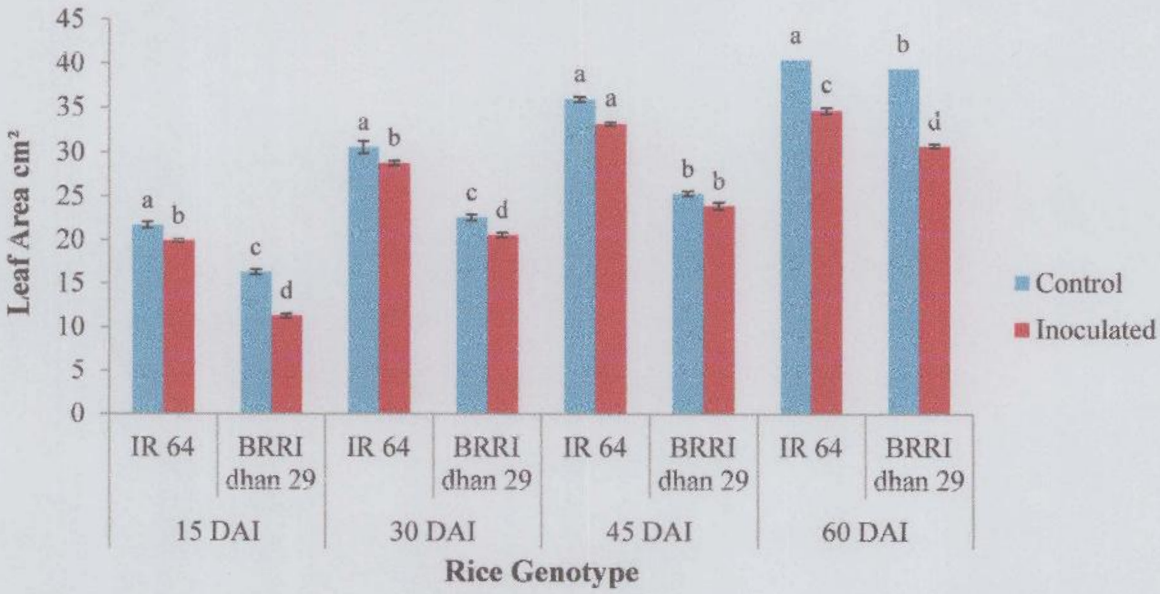
| Genotype | Treatment | Panicle Length (cm) | RP % | Panicle No | RP % | Unfilled Grain | RP % | Filled Grain | RP % | Thousand Grain Weight (gm) | RP % | Grain Length (mm) | RP % | Grain Width (mm) | RP % | Total Yield (t $ha^{-1}$) | RP % |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| IR 64 | Control | 34.56 | | 35 | | 290 c | | 1310 a | | 3.2 a | | 5.7 | | 2.3 | | 8.76 a | |
| | Inoculated | 30.2 | 12.61 | 32 | 8.57 | 391 b | 34.82 | 1160 b | 11.45 | 2.8 ab | 12.5 | 5.08 | 10.87 | 2 | 13.04 | 7.8 b | 10.96 |
| BRRI dhan29 | Control | 30.4 a | | 23 b | | 395 b | | 760 c | | 2.34 b | | 4.02 b | | 1.4 b | | 7.5 b | |
| | Inoculated | 21.62 b | 28.88 | 19 b | 17.39 | 600 a | 51.89 | 420 d | 44.73 | 1.46 c | 37.6 | 3.7 b | 7.96 | 1.17 b | 16.42 | 5.1 c | 32 |
| CV (%) | | 10.28 | | 8.77 | | 1.13 | | .29 | | 13.33 | | 10.28 | | 14.12 | | 6.15 | |

RP= Relative Performance

Values with different letters are significantly differed at 5% level of probability according to Tukey's HSD All-Pairwise Comparisons Test

### 4.14 Leaf Area

The leaf area of two rice genotypes, IR 64 and BRRI dhan29, has been shown in Figure. 1 under the control and inoculation treatment conditions. Four time points 15, 30, 45, and 60 days after inoculation (DAI) were used to display the data.




In case of 15, 30 and 60 days after inoculation leaf area were decreased significantly ($P<0.05$) in both rice genotypes under inoculated conditions except 45 days after inoculation (Figure. 1). At 15, 30 and 60 days after inoculation IR64 had experienced various degree of reduction, which was 8.21%, 5.88% and 13.86% respectively (Figure. 2) compared to non inoculated IR64. In addition, BRRI dhan29 had exhibited a significant reduction in leaf area of 30.67%, 8.99%, and 21.95%, respectively after 15, 30 and 60 days after inoculation in comparison to BRRI dhan29 that was not infected (Figure. 2). This finding had implied that under *Pyricularia oryzae* stress, IR 64 was better able to sustain a greater leaf area. At 45 days after inoculation (45 DAI), no significant differences were observed between the control and inoculated conditions of any genotype (Figure. 1). The highest leaf area of IR64 was 35.98 $cm^2$ and 25.3 $cm^2$ in case of BRRI dhan29 in non inoculated condition.

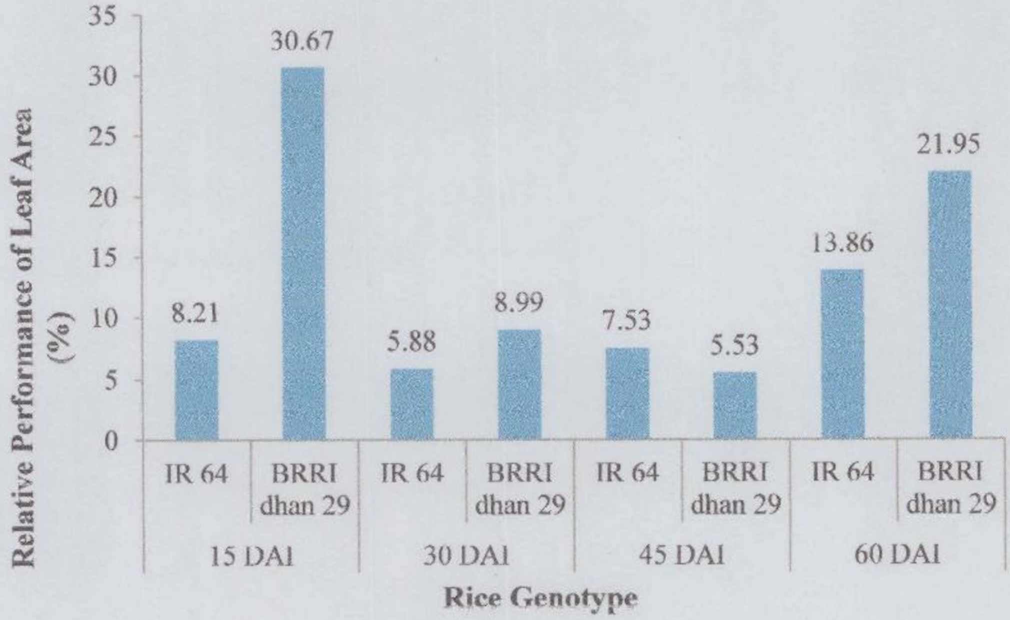



Figure. 2. Relative Performance of Impact of *Pyricularia oryzae* on leaf area of IR 64 and BRRI dhan29

(DAI= Days after Inoculation)

### 4.15 Leaf Number:

The leaf number of two rice genotypes, IR 64 and BRRI dhan has been exhibited in Figure. 3 under control and inoculation circumstances. The data were displayed at four time points: 15, 30, 45, and 60 days after inoculation (DAI).

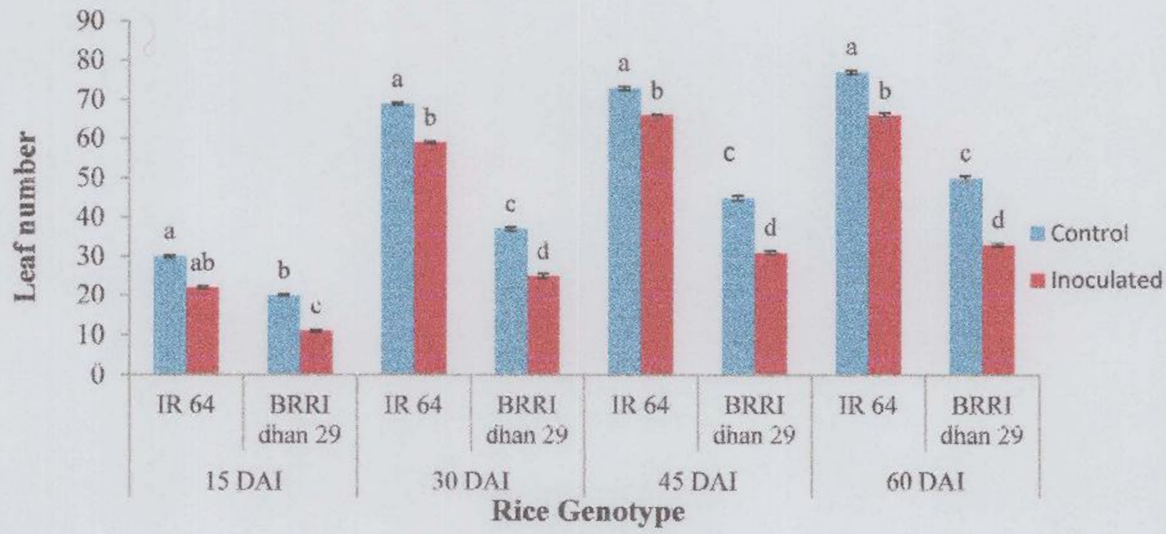



Figure. 3. Impact of *Pyricularia oryzae* on leaf number of IR 64 and BRRI dhan29

(DAI= Days after Inoculation.

All values represent the mean ± standard error (n= 3) at $P$ <0.05 probability according to Tukey HSD All-Pairwise Comparisons Test)

Leaf numbers were reduced significantly ($P$<0.05) in both rice genotypes at 15, 30, 45, and 60 days after inoculation (Figure 3). At 15, 30, 45, and 60 days following inoculation, IR64 had exhibited varying degrees of decrease, which were 26.67%, 14.49%, 9.59, and 14.28%, respectively (Figure 4), compared to non-inoculated IR64. Furthermore, BRRI dhan29 had showed a substantial drop in leaf number of 45%, 32.43%, 31.11%, and 34%, respectively, after 15, 30, 45, and 60 days after inoculation when compared to BRRI dhan29 that was not infected (Figure 3). This finding had suggested that IR 64 was better able to maintain a higher leaf number in the face of *Pyricularia oryzae* stress.

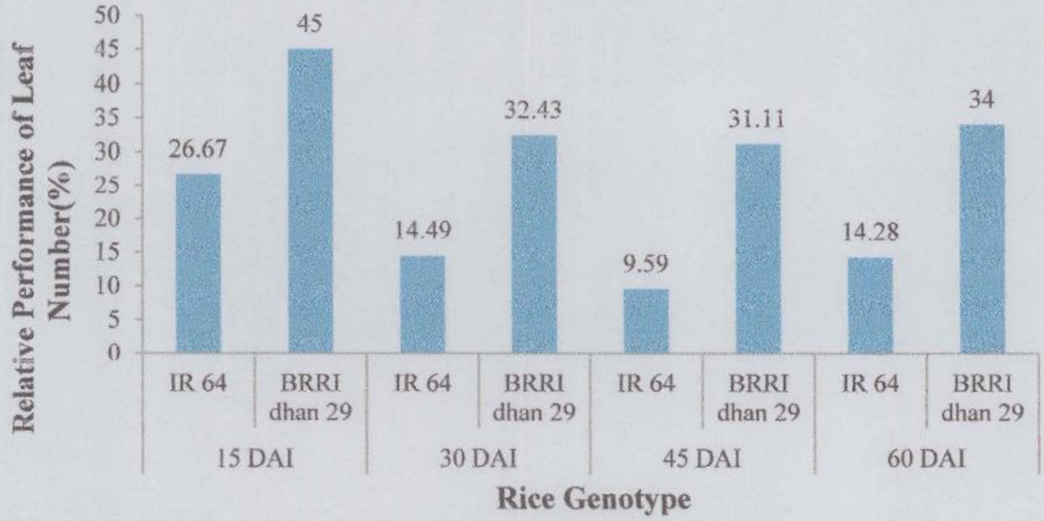



Figure. 4. Relative Performance of Impact of *Pyricularia oryzae* on leaf number of IR 64 and BRRI dhan29

(DAI= Days after Inoculation.)

### 4.16 Leaf greenness by SPAD

This bar graph (Figure. 5) compares the leaf greenness of two rice genotypes, IR 64 and BRRI dhan29, under control and inoculation conditions. The data were exhibited at four time points: 15, 30, 45, and 60 days after inoculation (DAI).

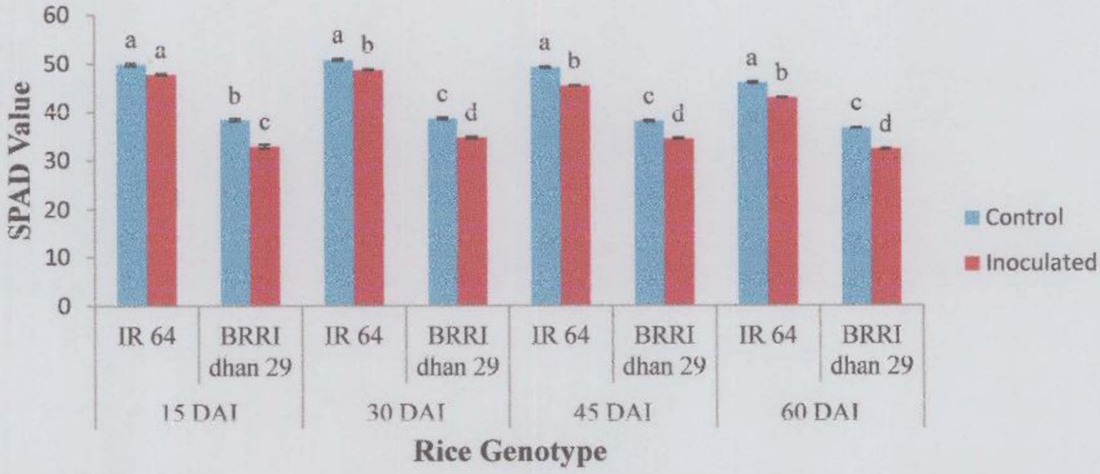



Figure. 5. Impact of *Pyricularia oryzae* on leaf greenness of IR 64 and BRRI dhan29

DAI= Days after Inoculation

All values represent the mean ± standard error (n= 3) at $P$ <0.05 probability according to Tukey HSD All-Pairwise Comparisons Test

Leaf greenness was declined significantly ($P$<0.05) in both rice genotypes 30,45 and 60 days after inoculation, with the exception of 15 days (Figure 5). At 30, 45, and 60 days after inoculation, IR64 had showed varying degrees of decrease, which were 4.13%, 7.64%, and 6.73%, respectively (Figure 6), compared to non-inoculated IR64. Furthermore, BRRI dhan29 had shown a substantial drop in leaf greenness of 34.25%, 10.36%, 9.76%, and 12.11% after 15, 30, 45, and 60 days after inoculation, compared to BRRI dhan29 that was not infected (Figure. 6). This study had suggested that IR 64 can maintain a higher level of leaf greenness under *Pyricularia oryzae* stress. At 15 days after inoculation (15 DAI), no significant differences were observed between the control and inoculated conditions of IR 64 genotype (Figure. 5).

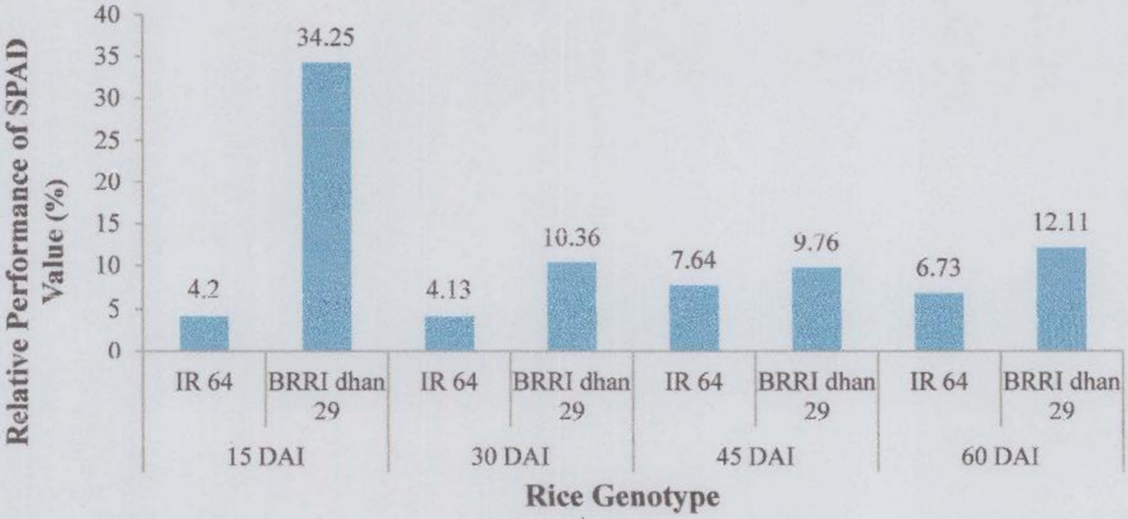



Figure 6. Relative Performance of Impact of *Pyricularia oryzae* on leaf greenness of IR 64 and BRRI dhan29

(DAI= Days after Inoculation.)

### 4.17 Percentage of Disease Index

The percentage of disease index of two rice genotypes, IR 64 and BRRI dhan29 has been displayed in Figure. 7 under the control and inoculation treatment conditions. Two time points 10 and 20 days after inoculation (DAI) were used to display the data.

The results indicated that IR 64 had exhibited significantly ($P$<0.05) lower PDI values compared to BRRI dhan29 at both time points, demonstrating greater resistance to the *Pyricularia oryzae* . In contrast, BRRI dhan29 shows higher PDI values which indicate greater susceptibility to the disease. This graph had demonstrated the varying levels of disease susceptibility between the two genotypes. BRRI dhan29 had shown a significantly higher PDI at both time points compared to IR 64 which suggest that BRRI dhan29 is more vulnerable to the disease. Furthermore, the significant increase in PDI values from 10 DAI to 20 DAI for both genotypes showed the progressive nature of the disease over time.



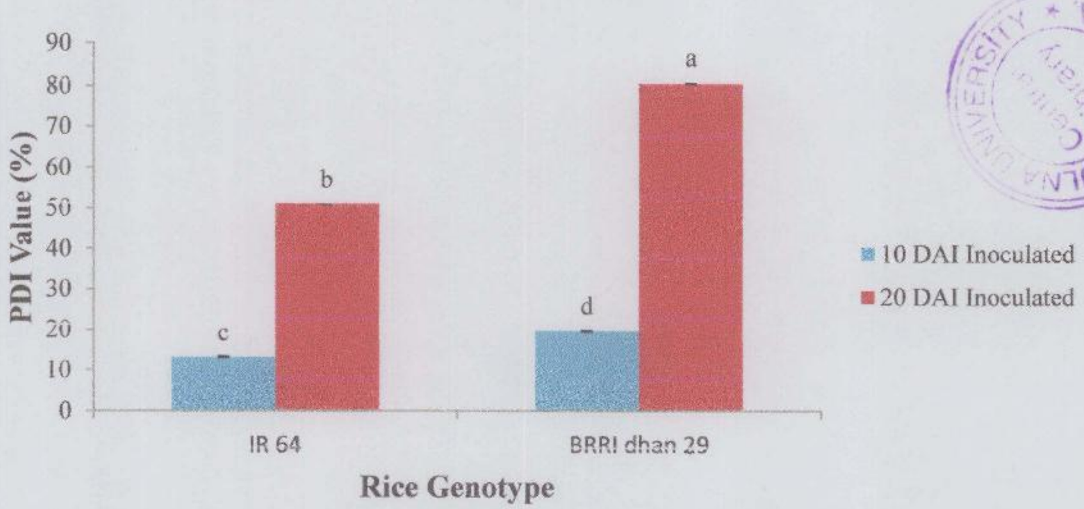



Figure. 7. Impact of *Pyricularia oryzae* on percentage of plant disease index of IR 64 and BRRI dhan29

(DAI= Days after Inoculation.

All values represent the mean ± standard error (n= 3) at $P$ <0.05 probability according to Tukey HSD All-Pairwise Comparisons Test)

# Chapter V

# Discussion

Rice blast disease (*Pyricularia oryzae*) had significantly affects the growth and physiological parameters of rice genotypes IR 64 and BRRI dhan29 (Table. 2). Plant height and tiller numbers were reduced under blast infection due to fungal colonization and damage to growing points, which stunts growth and limits tillering (Mousanejad et al.., 2010; Khan et al., 2014). Similarly, the number of effective tillers, essential for grain production, decreased which further reduce yield potential. Both fresh and dry weight of plants had decline significantly in inoculated plants, indicating a negative impact on biomass accumulation. Additionally, root length was reduced, which may hinder water and nutrient uptake which further compromising plant health and productivity. These results highlight the broader physiological disruptions caused by the disease.IR 64 had exhibited a smaller reduction in these parameters compared to BRRI dhan29, suggesting partial resistance or tolerance which is potentially linked to genetic traits such as enhanced defense mechanisms. These findings showed the importance of breeding resistant cultivars like IR 64 and adopting strategies such as fungicide application and integrated pest management to minimize yield losses caused by blast (Zewdu, 2021).

Rice blast disease (*Pyricularia oryzae*) had significantly reduced key yield-contributing parameters in rice genotypes IR 64 and BRRI dhan29 (Table. 3). Panicle length and number are adversely affected due to *Pyricularia oryzae* colonization and damage to reproductive tissues, resulting in reduced fertility and shorter panicles (Li et al., 2022). Sterile grains increase significantly, while filled grains decrease, highlighting the *Pyricularia oryzae* disruption of fertilization and grain development processes (Khan et al., 2014).Thousand-grain weight is also reduced, attributed to poor grain filling and interrupted maturation, consistent with Nalley et al. (2016). Additionally, grain length, width, and total yield are negatively impacted, with BRRI dhan29 showing greater susceptibility than IR 64. Yield losses, which can reach up to 50% in susceptible varieties, emphasize the need for resistance breeding (Neupane and Bushal, 2021; Khan et al., 2014).The partial tolerance observed in IR 64 suggests the presence of resistance traits, such as enhanced defense mechanisms, as noted by Khatun et al. (2016). To minimize yield losses, strategies such as cultivating resistant genotypes, applying fungicides, and adopting integrated pest management practices are essential for sustainable rice production.

Figure 1 illustrates changes in leaf area for two rice genotypes, IR 64 and BRRI dhan29, under control and inoculated conditions at different time points (15, 30, 45, and 60 days after inoculation). The results reveal significant reductions in leaf area under blast infection (*P. oryzae*), with BRRI dhan29 showing greater sensitivity compared to IR 64.In control conditions, both genotypes exhibited larger leaf areas, reflecting optimal growth. However, inoculated plants showed a consistent decline in leaf area, particularly in BRRI dhan29, indicating its higher susceptibility. This aligns with findings by Chuwa et al. (2015), who reported reduced leaf development in sensitive cultivars due to necrosis and stunting. The gap between control and inoculated conditions widened over time, especially at 45 and 60 DAI, highlighting the progressive impact of the disease. Shahriar et al. (2020) attributed these effects to disrupted cellular processes, leading to reduced leaf expansion and tissue damage. In contrast, IR 64 exhibited a smaller reduction in leaf area, suggesting partial resistance through delayed lesion development or enhanced physiological tolerance. The reduction in leaf area directly affects photosynthetic capacity, ultimately impacting yield. Prior studies (Devanna et al., 2022) emphasize the role of resistant genotypes in mitigating such losses by maintaining physiological performance under *Pyricularia oryzae* stress. These findings showed the importance of developing and utilizing resistant genotypes like IR 64 in combination with integrated disease management strategies to minimize the negative impact of rice blast on plant growth and yield.

The graph in Figure. 3 shows how rice blast disease affects leaf number in two rice genotypes, IR 64 and BRRI dhan29, at different growth stages (15, 30, 45, and 60 days after inoculation). Under control conditions, both genotypes showed an increase in leaf number over time, which is consistent with normal plant growth. However, IR 64 consistently had more leaves than BRRI dhan29, indicating possible genotypic variations in leaf growth and development. At all growth stages, both genotypes saw a considerable reduction in leaf number after being inoculated with the blast *Pyricularia oryzae*. Several factors contribute to this reduction:the fungus causes localized lesions on leaves, resulting in necrotic tissue and decreased leaf extension (Miah et al., 2017).

Blast infection can slow plant growth and development, resulting in fewer leaves and a smaller leaf area. While both genotypes were vulnerable to blast, IR 64 appeared to be more resistant than BRRI dhan29, as shown by a lower drop in leaf number in inoculated plants. This shows that IR 64 may have genetic features that offer some resistance to blast infection. The considerable fall in leaf number induced by blast infection demonstrates the possibility of significant yield losses in rice cultivation. Strategies to reduce leaf number decrease, such as using resistant cultivars or applying fungicides, can help to attenuate the disease's impact (Bastiaans,1993).

Figure.5 highlights the SPAD values, an indicator of leaf chlorophyll concentration and photosynthetic efficiency, in two rice genotypes, IR 64 and BRRI dhan29, measured at 15, 30, 45, and 60 days after inoculation (DAI). The study reveals a progressive decline in SPAD values over time, with inoculated plants exhibiting a more pronounced reduction compared to control plants. This aligns with previous findings by Acharya et al., (2019) which reported significant reductions in chlorophyll content and photosynthetic activity in blast-infected rice plants. The reduction in SPAD values in inoculated plants can be attributed to several factors: the blast *Pyricularia oryzae* produces enzymes that degrade chlorophyll, damages chloroplast membranes and organelles critical for photosynthesis, and competes with the host plant for nutrients such as nitrogen and magnesium essential for chlorophyll synthesis. Although both genotypes displayed reduced SPAD values following inoculation, the extent of reduction varied, indicating potential genotypic differences in their response to rice blast infection. This suggests that certain genotypes may have greater tolerance to the disease, preserving chlorophyll levels and photosynthetic function to a greater extent (Acharya et al., 2019).

The graph in Figure. 7 depicts the PDI (Percent Disease Index) values of two rice genotypes, IR 64 and BRRI dhan29, under two different inoculation times: 10 DAI (Days After Inoculation) and 20 DAI. Significant variations in disease severity are observed among the genotypes and inoculation periods (Nurgoho, 2021).

BRRI dhan29 exhibited the highest PDI value at 20 DAI (approximately 80), followed by IR 64 at the same time point. This indicates that BRRI dhan29 is more susceptible to blast disease at later stages of inoculation. The statistical grouping denoted by letters (a, b, c, and d) confirms that the differences in PDI values are significant. IR 64 displayed comparatively lower susceptibility, with the lowest PDI value observed at 10 DAI (Nalley,2016).

The results align with findings from Neupane and Bushal (2021), who highlighted the critical role of inoculation timing in determining disease severity in rice blast studies conducted in Nepal. Similarly, Aye et al. (2015) also emphasized that delayed infection often exacerbates yield losses due to the longer *Pyricularia oryzae* development cycle. The higher PDI values at 20 DAI can be attributed to increased *Pyricularia oryzae* establishment and spread, corroborating these studies.

# Chapter VI

# Conclusion

This study highlights the contrasting responses of two rice genotypes, IR 64 and BRRI dhan29, to stress induced by *Pyricularia oryzae*, the causative agent of rice blast disease. IR 64 exhibited greater tolerance to the *Pyricularia oryzae*, maintaining higher values in morphological, physiological, and yield-contributing traits compared to BRRI dhan29 based on their relative performance. Plant height, tiller number, biomass, leaf area, and yield-contributing characteristics are all markedly reduced by *Pyricularia oryzae*, indicating BRRI dhan29's increased vulnerability.

Plant height, tiller number, fresh and dry biomass, and leaf area of both genotypes exhibited significant reductions under inoculated conditions. These declines indicate the broad physiological disruptions caused by *P. oryzae* infection, which impairs photosynthetic capacity and biomass accumulation.

Significant decreases were also seen in both genotypes for two crucial indicators of photosynthetic capacity: leaf area and SPAD readings. The more significant decreases observed in BRRI dhan29 correlate with the disease's more severe expression and increased physiological suffering.

Yield-contributing traits were also adversely affected in both genotypes under inoculated conditions. *Pyricularia oryzae* stress reduced panicle length, filled grain number, thousand grain weight, and overall grain yield. Yield losses were notably higher in BRRI dhan29 (32%) than in IR 64 (10.96%) which showed the vulnerability of the former and the partial resistance of the latter.

Percent Disease Index (PDI) was used to measure disease severity which was consistently higher in BRRI dhan29, which displayed higher susceptibility to *P. oryzae*. By contrast, the ability of IR 64 to limit disease progression suggests the presence of genetic traits that contribute to its resilience.

The study demonstrates the severe impact of rice blast disease on growth and productivity, emphasizing the importance of resistant varieties like IR 64 for sustainable rice cultivation. The findings underline the need for effective disease management strategies, particularly in regions where rice blast is a recurring threat.

# Chapter VII

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