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RESEARCH PAPER

Visual observation and image analysis method of blight disease severity for resistance assessment of two rice varieties

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ABSTRACT

Bacterial Leaf Blight (BLB), caused by *Xanthomonas oryzae* pv. *oryzae*, is a major threat to global rice production, causing yield losses of up to 80%. Accurate assessment of disease severity is essential for developing resistant rice varieties and implementing effective management strategies. However, traditional visual observation methods, while widely used, are prone to subjectivity and reduced accuracy. This study evaluates the accuracy of image analysis for assessing rice plant resistance to BLB. Disease severity was assessed using both visual observation and image analysis, with results quantified through the Area Under the Disease Progress Curve (AUDPC) and infection rate calculations. Image analysis outperformed visual observation, achieving an accuracy rate above 96%, compared to less than 90% for the latter. The Ciherang variety demonstrated greater resistance to BLB, with lower AUDPC and infection rates when assessed using image analysis. Conversely, visual observation produced contradictory results, highlighting its limitations. This study concludes that image analysis provides a more objective, reproducible, and accurate approach to assessing disease severity, with implications for breeding programs and integrated disease management systems. Further research is recommended to validate these methods across a broader range of rice genotypes and environmental conditions.

Key words: accuracy, AUDPC, bacterial leaf blight, *Ciherang*, *Inpari 32*

INTRODUCTION

Bacterial leaf blight (BLB), caused by the bacterium *Xanthomonas oryzae* pv. *oryzae* (Xoo), is one of the major diseases affecting rice crop production worldwide, particularly in Asian countries. This disease can lead to significant yield losses, with estimates ranging from 15% to as high as 80% under severe conditions (My, 2024; Lestari, 2023; Bae et al., 2018). The impact of BLB on rice production is profound, as it not only reduces the quantity of the harvest but also affects the quality of the grains, thereby threatening food security for millions of people who depend on rice as a staple food (Jiang et al., 2020; Long et al., 2023). The disease primarily manifests through leaf wilting and necrosis, which can severely impair photosynthesis and overall plant health. The pathogen enters the rice plant through hydathodes at the leaf margins or through wounds, proliferating within the xylem and leading to systemic infection (Jiang et al., 2022).

Various studies have focused on developing strategies to control BLB, including the development of inhibitors against Xoo (Jiang et al., 2019), the application of organic fertilizers combined with Xoo-controlling microbes (Rifki et al., 2018), and the long-term effects of niclosamide use to inhibit BLB (Kim et al., 2016). Other efforts include the identification of resistant genes in rice cultivars using SSR markers (Acharya et al., 2018), and the screening of BLB-resistant rice genotypes (Nihad et al., 2021). According to Kim & Reinke (2019), evaluating plant resistance to BLB in the early breeding stages is vital for developing resistant varieties.

An essential component of BLB disease management in the field is monitoring and assessing disease severity. This step is crucial to determine whether the implemented control measures are effective in reducing disease intensity. Monitoring disease severity enables farmers and agronomists to make informed decisions regarding the continuation or modification of management strategies (My, 2024; Lestari, 2023). Regular assessments also allow early detection of outbreaks, facilitating timely interventions that prevent further spread and minimize yield losses. This proactive approach is particularly

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important, as BLB can escalate rapidly under favorable environmental conditions (Bae et al., 2018; Jiang et al., 2020). Systematic monitoring generates valuable data for research purposes and supports the development of more effective control measures and resistant varieties. It also informs future breeding programs aimed at enhancing resistance to BLB (Long et al., 2023; Jiang et al., 2022).

Several methodologies are employed to monitor and assess BLB severity in rice fields. One of the most widely used methods is visual observation or scoring, in which disease severity is determined based on the percentage of leaf area affected by lesions. This approach often relies on standardized scales such as the Standard Evaluation System (SES) for rice, providing a uniform framework for quantifying disease impact (Haidary et al., 2018; Acharya et al., 2018). The conventional approach, involving field-based visual observation and calculation of the Area Under the Disease Progress Curve (AUDPC), is commonly used to assess crop resistance to BLB (Jirankali et al., 2023). However, subjectivity and low accuracy —often influenced by the observer's skill and environmental conditions—remain major limitations (Hassan et al., 2023; Vettori & Rice, 2020). Despite these drawbacks, visual observation remains the primary method for many practitioners due to its ease of use (Bock et al., 2021).

To overcome these limitations, researchers increasingly adopt "quantitative measurements" using advanced techniques such as image analysis and remote sensing. These approaches allow more precise assessments by generating spatial data on disease distribution, offering deeper insights into disease progression and dynamics (Shi et al., 2021; Nanayakkara et al., 2020).

The novelty of this research lies in the application of digital image analysis technology to enhance the accuracy of assessing plant resistance to BLB. Unlike traditional visual observation, digital image analysis provides a more objective and precise approach, offering several key advantages: 1) Objectivity and precision: Digital image analysis eliminates the subjectivity of human observation by providing objective measurements of disease symptoms. It quantifies severity using specific parameters such as lesion area or color changes, which are more reliable than subjective ratings (Long et al., 2023; Jiang et al., 2022; Haidary et al., 2018); 2) High throughput: Digital imaging enables the rapid evaluation of large plant populations, making high-throughput phenotyping feasible—particularly beneficial for breeding programs that assess numerous genotypes (Acharya et al., 2018); 3) Non-destructive assessment: This method allows non-invasive monitoring of living plants, ideal for studies that require continuous observation of plant development (Zhang et al., 2018).

Despite these advancements, Barbedo (2013) notes that the effectiveness of disease detection through digital images can be limited when symptoms are subtle or overlap with other physiological changes in the plant, potentially leading to misdiagnosis or oversight.

Digital image analysis has been successfully applied in various studies to evaluate resistance to BLB and other diseases. For example, image analysis methods have been used to estimate disease severity in rice varieties affected by blight and spot diseases (Bande & Hasan, 2024). Automated Disease Severity Assessment systems that utilize digital imaging have been developed to quantify lesion size and color intensity, offering detailed insights into plant health (Goriewa-Duba et al., 2018). Furthermore, integrating image analysis with machine learning has demonstrated potential for enhancing disease assessment accuracy. Machine learning algorithms can detect patterns in images, enabling more sophisticated analyses of plant resistance (Askey et al., 2019).

However, there is currently no study that explicitly compares the accuracy of visual observation and image analysis methods in assessing rice plant resistance to BLB in field conditions and their implications. Therefore, this research aims to validate the accuracy of visual observation compared to image analysis and to evaluate its implications for BLB disease control strategies.

This research is expected to make a significant contribution to the field of crop protection by providing a more accurate method for resistance assessment. It can be applied in breeding programs to develop BLB-resistance assessment. It can be applied in breeding programs to develop BLB-resistant rice varieties, thereby contributing to global food security through improved crop protection and higher yields. Furthermore, the developed method may be integrated into agricultural insurance systems to provide more precise and fair crop damage assessments (Hongo et al., 2022).

MATERIALS AND METHODS

Research Site. Surveys and image recording were conducted on rice varieties *Inpari 32* (3°46'56.9 "S 122°02'18.2 "E) and *Ciherang* (3°47'10.8 "S

122°01’40.7 “E) as test plants (105 days after planting). Both varieties were cultivated by farmers in adjacent areas in Puundombi Village, North Tongauna Subdistrict, Konawe District, Southeast Sulawesi Province, Indonesia. Confirmation of the causal agent of the disease in rice plants was carried out in the Laboratory of the Department of Plant Protection, Faculty of Agriculture, Halu Oleo University. All stages of the research were conducted from April to June 2024.

Determination of Plant Leaf Samples. Sampling location were determined using the purposive sampling method, with the criterion that visible symptoms of BLB were present on the plant leaves. Subsequently, five observation plots were determined using the diagonal sampling method. In each plot, four leaves exhibiting BLB symptoms were selected, each from a different rice clump, totaling 20 symptomatic leaves per variety. Efforts were made to ensure that sampled leaves showed no disease symptoms other than BLB (confirmed through laboratory testing) to avoid bias during image analysis.

Confirmation of the Cause of Blight. This stage began with the preparation of infected rice leaf slices (from the border area between diseased and healthy tissue), each measuring 0.5 cm × 0.5 cm. The slices were surface-sterilized using 70% ethanol for 5 min and rinsed twice with sterile distilled water. The sterilized slices were then chopped with a sterile knife, and two drops of sterile distilled water were added before incubating for 5 min. The supernatant, containing bacterial cells, was spread on PSA medium (potato 30 g, sucrose 20 g, and agar 20 g per L) and incubated for 3–4 days. Bacterial colonies with round, mucoid, yellow characteristics (typical of Xoo) (Khaeruni & Wijayanto, 2013), were then Gram-stained (Rachmawati et al., 2017) and observed under a Leica DM500 microscope (Leica Microsystems, Germany) at 400× magnification.

Plant Leaf Image Recording and Environmental Relative Humidity Measurement. Image recording of BLB-symptomatic leaves was carried out starting at 08:00 AM UTC+8 under clear sky conditions. Leaves were exposed to direct sunlight to clearly capture BLB symptoms. A smartphone camera (Realme C35) mounted on a tripod, approximately 15 cm away from the leaf, was used for image capture. A white background was placed behind the leaf to aid image analysis, along with a 30 cm scale bar for calibration. Image recording was conducted every other day for a week (days 1, 3, 5, and 7) on the same leaf samples. Environmental relative humidity was measured before each image capture using an HTC-1 digital hygrometer (Aptechdeals, China).

Image Analysis of BLB-Symptomatic Leaves. The analysis consisted of three main steps: (1) Calibration – RGB images of symptomatic leaves were calibrated (from pixels to centimeters) to enable measurement of diseased and healthy areas. The RGB image was then converted to the CIELAB color space (Lab*), resulting in three separate images: L*, a*, and b*; (2) Segmentation – The a* image was used for segmentation, as it best distinguished between diseased and healthy leaf areas; (3) Estimation of disease severity – A binary image (black and white) resulting from segmentation was used to estimate disease severity. All image analysis steps were conducted using the Fiji-ImageJ software (ImageJ 1.54g) (Schindelin et al., 2012).

Assessment of BLB Disease Severity, Disease Infection Rate, AUDPC, and Plant Resistance. Two methods were used to assess disease severity: visual observation and image analysis. In visual observation, BLB symptoms were scored and the disease severity percentage was calculated using the following formula (Soesanto et al., 2024):

$$DS = \left[\frac{\Sigma (v_i \times n_i)}{Z \times N}\right] \times 100\%$$

DS = Disease severity (%);
v_i = Leaf damage score;
n_i = Number of leaves with the same damage score;
Z = Highest damage score;
N = Total number of leaves observed.

Scoring was based on Seshu (1989), with slight modifications: 1–5% damage (score 1), 6–12% (score 3), 13–25% (score 5), 26–50% (score 7), and 51–100% (score 9).

Assessment of disease severity through image analysis is carried out on binary images as the final result of the final stage of image analysis using the following formula (modified) (Rodrigues et al., 2019).

$$DS = \frac{\text{Leaf area with blight (cm}^2\text{)}}{\text{Total leaf area (cm}^2\text{)}} \times 100\%$$

In image-based assessment, binary images were used to calculate disease severity (modified from Rodrigues et al., 2019). White areas in the binary image represented diseased tissue, while black areas represented healthy tissue (Figures 4C1 and 4C2). The total leaf area was the sum of diseased and healthy areas, calculated using the Analyze Particle plugin in Fiji-ImageJ.

BLB disease infection rate was calculated using Van der Plank (1963) formula:

$$r = \frac{e}{t}\left(\log\frac{X_t}{1 - X_t} - \log\frac{X_o}{1 - X_o}\right)$$

r = Infection rate;
e = Conversion number (2.718);
t = Observation time interval (days);
X_t = Proportion of diseased leaves at time t (%);
X_0 = Proportion of diseased leaves at the beginning of observation (%).

Plant resistance to BLB was further assessed by calculating the AUDPC (Area Under the Disease Progress Curve) using the following formula (Madden et al., 2017):

$$AUDPC = \sum_{i=1}^{n_i-1}\left[\frac{(y_i + y_{i+1})}{2}\right] \times (t_{i+1} - t_i)$$

n = Number of observations;
y_i = Disease severity at the initial observation;
y_{i+1} = Disease severity at the next observation;
ti = initial observation time;
t_{i+1} = Next observation time.

A lower AUDPC value indicates greater plant resistance to disease (Madden et al., 2017).

Accuracy of Disease Severity Assessment Method. The accuracy of the disease severity assessment was evaluated using the coefficient of determination (R^2) (Hasan et al., 2021).

Data analysis. All research data were tabulated and analyzed using descriptive statistics and correlation analysis with the Microsoft Excel (Microsoft Office Home and Student 2021).

RESULTS AND DISCUSSION

Bacterial leaf blight (BLB) in rice, caused by *Xanthomonas oryzae* pv. *oryzae* (Xoo), is a major disease affecting rice production worldwide. In this study, we successfully confirmed the presence of Xoo as the causative agent of blight in the rice varieties *Inpari 32* and *Ciherang*, based on laboratory analyses (Figure 1). The identification of Xoo was consistent with the findings of Rachmawati et al. (2017), who described the bacteria as forming round, shiny, slimy, pale-yellow colonies. Additionally, Gram staining and microscopic observations confirmed that Xoo is a Gram-negative, rod-shaped (bacillus) bacterium. These characteristics were crucial in verifying the pathogen responsible for the observed symptoms in the sampled rice plants.

Monitoring disease progression caused by the Xoo pathogen is crucial for developing effective control strategies. Two common approaches for evaluating the severity of plant diseases include visual observation and image analysis, both of which serve as a basis for assessing plant resistance levels. Visual observation relies on the direct assessment of disease symptoms, typically based on visible changes in the leaves. In the case of bacterial blight, initial symptoms appear as grayish-green stripes along the leaf edges, which progressively extend toward the base. As the disease advances, the leaves turn yellow, develop a straw-like appearance, and dry at the tips. These visible symptoms form the basis for determining disease severity in the field (Figure 2).

In contrast, image analysis employs digital technology to objectively and consistently detect and quantify disease symptoms. This method offers significant advantages by minimizing observer bias. The results of digital image analysis are highly reproducible, as illustrated in Figure 3. For example, visual assessments of blight-symptomatic leaves from *Inpari 32* and *Ciherang* yielded severity values of 50% and 49%, respectively, whereas image analysis indicated a uniform severity of 100%.

The percentage of disease severity based on visual observation often differs from that derived from image analysis (Figure 4). This discrepancy may be due to the inherent difficulty observers face in precisely distinguishing between healthy and diseased leaf areas. Visual evaluations are subjective and influenced by factors such as lighting, observer experience, and leaf texture. In contrast, image analysis provides a more objective and consistent assessment, clearly distinguishing between diseased (white areas in Figure 4C1) and healthy (black areas in Figure 4C2) tissues. However, some leaf regions appear grayish or nearly white (Figure 4B), indicating transitional zones that may become diseased over time. Until these areas fully progress and the color shifts to yellow-straw, image analysis may still classify them as healthy.

Both visual observation and image analysis in this study showed that BLB severity increased over time in both rice varieties, as reflected in the rising average severity at each observation point (Figures 5 and 6). However, data analysis confirmed that image-based assessments were significantly more accurate than visual observations (Figure 6). This is evident

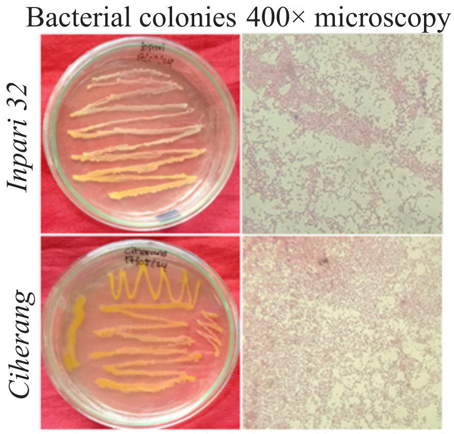

Figure 1. Visualization of bacterial colonies on artificial media isolated from blight-symptomatic *Inpari 32* and *Ciherang* rice leaves, along with microscopic observation (400× magnification) following Gram staining.

Figure 2. RGB images of rice leaf samples showing the progression of blight symptoms, observed every two days over the course of one week.

Figure 3. Images of blight-symptomatic rice leaf samples from the *Inpari 32* and *Ciherang* varieties. Examples of disease severity (DS) assessment results are shown based on visual observation (visualized with RGB images) and image analysis (visualized by grayscale images).

from the higher coefficient of determination (R^2 > 96%) achieved through image analysis, compared to lower R^2 values (< 90%) from visual assessments (Figure 5). These findings suggest that image analysis is a more reliable method for estimating disease progression, offering a closer approximation of actual severity than traditional visual observation.

Accurate assessment of disease severity is crucial for evaluating plant resistance. However, visual observations often result in inaccuracies that can compromise this evaluation. Such errors may lead to the exclusion of genuinely resistant plants and the selection of susceptible ones, undermining breeding programs aimed at improving disease resistance

(Méline et al., 2023). The subjectivity of human scorers introduces variability that hampers the identification of resistance genes and complicates the development of resistant cultivars (Bock et al., 2010). Additionally,

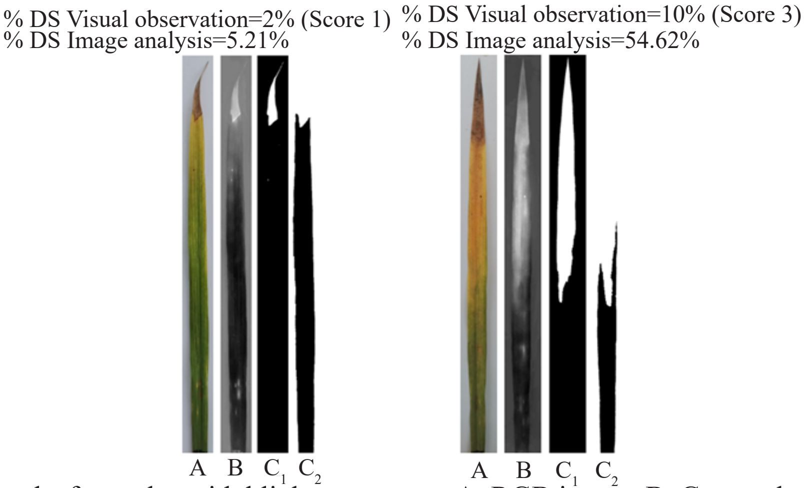

Figure 4. Images of rice leaf samples with blight symptoms. A. RGB image; B. Grayscale image; C1–C2. Binary (black-and-white) image, where C1 = represents the diseased leaf area and C2 represents the healthy leaf area; DS = disease severity.

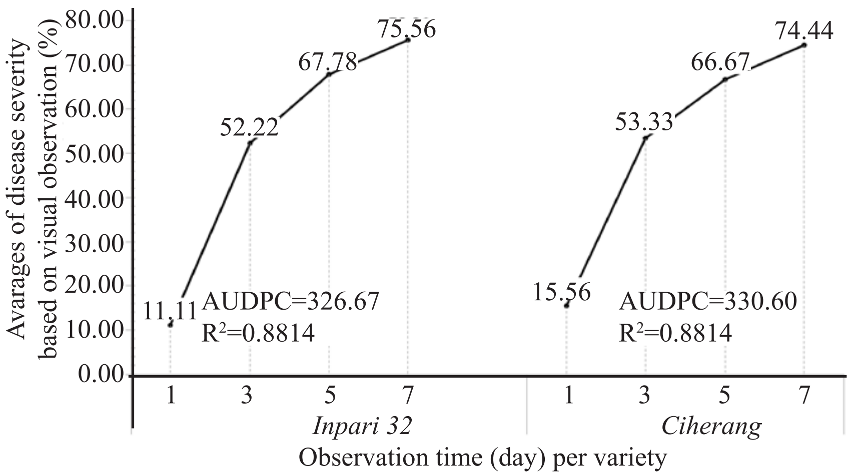

Figure 5. AUDPC value, coefficient of determination, and average percentage of BLB disease severity based on visual observation in the *Inpari 32* and *Ciherang* varieties.

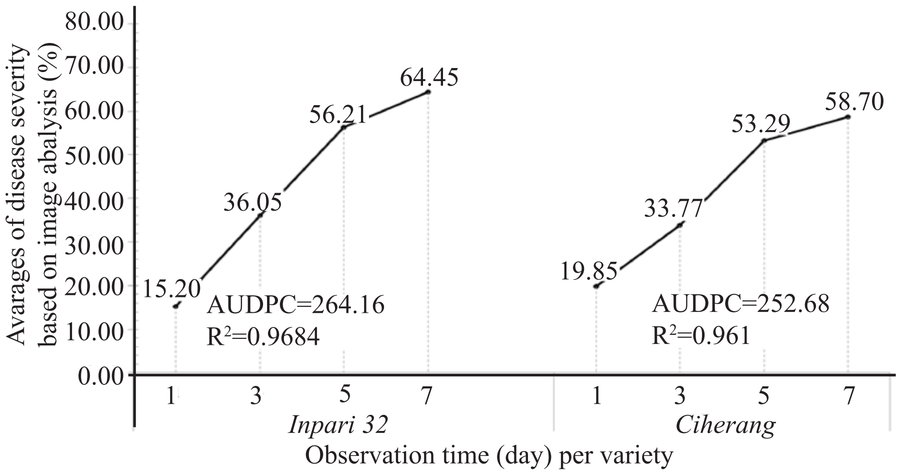

Figure 6. AUDPC value, coefficient of determination, and average percentage of BLB disease severity based on image analysis in *Inpari 32* and *Ciherang* varieties.

flawed assessments can result in inappropriate disease management practices, potentially exacerbating disease spread and causing significant crop losses. These inaccuracies may also have serious economic consequences, as crops incorrectly classified as resistant may fail to meet yield expectations (Bock et al., 2021). Furthermore, misallocated resources—such as time, labor, and capital—may be wasted on ineffective control strategies. Conversely, the adoption of disease-resistant varieties can reduce the need for chemical inputs, promoting more sustainable agriculture (Zander et al., 2023; Mooney et al., 2022). Therefore, improving resistance assessment accuracy is vital not only for yield optimization but also for efficient resource use and environmental sustainability.

Based on disease severity measurements, *Inpari 32* exhibited a consistently higher rate of BLB infection than *Ciherang*. Using image analysis, the infection rate for *Inpari 32* reached 1.089 units per day, while visual observation recorded 0.818 units per day (Table 1). This difference suggests that *Inpari 32* may be more susceptible to BLB, possibly due to genetic or physiological differences in response to the pathogen. Agronomically, the lower resistance of *Inpari 32* raises concerns about its field resilience. Such varieties may require enhanced management strategies, including chemical treatments or more robust protective measures, to mitigate yield losses.

The observed differences between measurement techniques also underscore the importance of method selection. Image analysis, which detected a higher infection rate in *Inpari 32*, can identify subtle or early-stage infections that might be overlooked during visual inspections. In the case of *Ciherang*, however, visual observation recorded a slightly higher infection rate (0.512 units/day) compared to image analysis (0.446 units/day). This variation may stem from human error or subjective interpretation, which can lead to over- or underestimation. These findings highlight the critical role of method selection in disease measurement. While image analysis offers objectivity and precision, especially for subtle symptoms, visual methods remain practical for field use, though they carry potential biases.

Subsequently, AUDPC (Area Under the Disease Progress Curve) values derived from image analysis showed a clear difference in resistance between the two rice varieties. *Ciherang* exhibited greater resistance to BLB, as indicated by its lower AUDPC value (252.68%-day) compared to Inpari 32 (264.16%-day) (Figure 6). This supports findings by Suryaningsih et al. (2023), who reported that Ciherang is highly resistant to BLB, while Yuliani & Sudir (2022) classified *Inpari 32* as resistant. Interestingly, visual observations showed a slightly different result, suggesting that *Inpari 32* appeared more resistant than *Ciherang* based on AUDPC values (326.67%-day and 330.00%-day, respectively) (Figure 5). This further reinforces the idea that inaccurate assessments can negatively affect resistance classification. As explained by Febriyanto et al. (2022), AUDPC is widely used in plant pathology to measure disease development over time and assess plant resistance. Lower AUDPC values indicate higher resistance, while higher values suggest susceptibility. This is because more resistant plants slow disease development, resulting in a flatter disease curve and lower AUDPC. In contrast, susceptible plants show rapid progression and steeper disease curves, leading to higher AUDPC values (Madden et al., 2017).

Several factors may contribute to differences in plant resistance to BLB infection. The presence of specific resistance genes is a critical defense mechanism. Research has shown that both quantitative trait loci (QTLs) and resistance (R) genes play important roles in disease resistance. Notable resistance genes such as Xa7, Xa10, and Xa23 enhance rice defenses by recognizing pathogen-associated molecular patterns and triggering immune responses. Identification of QTLs across multiple chromosomes also emphasizes the importance of genetic diversity in breeding programs (Liu et al., 2024). Additionally, pathogen virulence factors significantly influence the interaction between pathogen and host. Highly virulent pathogens may overcome plant defenses through enhanced infection efficiency, shorter latency periods, and increased sporulation (Fontyn et al., 2023). Some pathogens even neutralize antimicrobial peptides produced by plants through specialized virulence mechanisms (Farvardin et al., 2024). Understanding these factors is crucial for developing more resilient

Table 1. Infection rates of bacterial leaf blight in the *Inpari 32* and *Ciherang* rice varieties based on image analysis and visual observation

Measurement methods	BLB disease infection rate (r) in rice variety (units per day)	
	Inpari 32	*Ciherang*
Image analysis	1.089	0.446
Visual observation	0.818	0.512

crop varieties.

Environmental conditions also play a vital role in disease development and spread. Recent studies have highlighted the influence of humidity, temperature, and soil factors on BLB severity. For instance, spatial analysis in Pakistani rice fields showed strong correlations between environmental variables and BLB outbreaks (Ahmad et al., 2023). Similarly, studies have linked high relative humidity to increased severity of sheath blight, suggesting similar effects on BLB (Naveenkumar et al., 2022). High humidity combined with elevated temperatures during critical growth stages has also been linked to increased disease incidence (Iqbal et al., 2022).

The strong correlation between environmental humidity and disease severity in the *Inpari 32* and *Ciherang* rice varieties—regardless of the measurement method—reinforces findings that humidity is one of the key factors influencing plant disease progression, particularly during critical growth stages (Table 2). In this study, *Inpari 32* demonstrated a slightly higher correlation, suggesting that this variety is more susceptible to fluctuations in environmental humidity. Elevated humidity levels create optimal conditions for pathogen proliferation, as pathogens generally thrive in moist environments. Consequently, in humidity-sensitive varieties like *Inpari 32*, the risk of disease intensifies when plants are exposed to high humidity during essential growth phases, as noted by Iqbal et al. (2022).

In contrast, although *Ciherang* also exhibited a strong correlation between humidity and disease severity, its slightly lower correlation value may indicate greater resilience to humidity variations or reduced sensitivity to environmental fluctuations. Despite this relative tolerance, *Ciherang* remains vulnerable to disease outbreaks under high-humidity conditions.

These findings underscore the critical importance of integrated disease management systems that incorporate real-time monitoring of environmental variables to mitigate the risk of BLB and other humidity-related diseases. By adopting proactive management strategies that account for environmental dynamics, rice cultivation can significantly improve its disease control measures. Advanced technologies—such as image analysis and/or remote sensing—should be integrated into these strategies to enhance early detection and intervention, thereby reducing the overall impact of disease outbreaks on crop yield and food security.

Scientifically, this study contributes to developing more accurate and efficient methods for monitoring crop disease resistance. Image analysis can enhance data quality, allowing for deeper analysis and better decision-making in disease management. Practically, this technology can be adopted by farmers and agronomists to monitor disease development and reduce losses. For instance, smartphone-based applications can help measure disease symptoms in the field.

Despite its contributions, this study has some limitations. First, only two rice varieties were evaluated, limiting the generalization of findings to a broader genetic background. Second, varying environmental conditions can affect the results, indicating the need for further research across diverse field conditions.

CONCLUSION

Accurate assessment of bacterial leaf blight (BLB) severity is essential for effective disease management. This study shows that image analysis is more reliable and objective than visual observation, with over 96% accuracy. It eliminates subjective bias and improves reproducibility. Based on image analysis, *Ciherang* demonstrated greater resistance to BLB than *Inpari 32*, with lower infection rates and AUDPC values. In contrast, visual assessments produced inconsistent results, highlighting their limitations. Image analysis offers valuable support for breeding programs and crop insurance systems. However, further research involving more rice varieties and diverse field conditions is needed to broaden these findings.

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Tabel 2. Correlation values (*) between environmental relative humidity (%) and disease severity (%) in the *Inpari 32* and *Ciherang* rice varieties, measured using image analysis and visual observation methods

	Inpari 32	*Ciherang*
Image analysis	0.958	0.948
Visual observation	0.983	0.981

* = Pearson correlation

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AUTHORS' CONTRIBUTIONS

GHS, AH, and R planned the research. R recorded plant images in the field and confirmed the disease-causing pathogens in the laboratory. AH and R analyzed the plant image. AH conducted the statistical analysis. GHS, AH, and R interpreted the research results and prepared the manuscript. GHS, AH, LOSB, AK, MT, and VNS reviewed the manuscript.

COMPETING INTEREST

The authors declare no competing interests.

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