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RESEARCH ARTICLE

Resistance sources of rice cultivars against *Pantoea* blight: A new threat to rice cultivation in India

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Abstract

The climatic variation in recent times has added a new problem to the rice crop, where leaves and panicles are blighted and the grains remain unfilled. The *Pantoea* genus is linked to a novel rice disease that induces leaf and panicle blight, adversely affecting rice output and quality. *Pantoea dispersa* is a Gram-negative bacterium recognized for its extensive environmental adaptation and potential as a plant pathogen and an opportunistic human disease. This study aimed to identify the bacterial pathogen by the 16S rDNA gene, the growth requirement of the bacteria and the resistant cultivar source against the panicle blight disease of rice. The bacterial pathogen was isolated from the Bargarh district of Odisha, India and the isolate was named ODBP1 and identified based on morphological and biochemical characterization. The isolate was amplified in molecular Identification using the universal primer pair 27F and 1492R. Optimization of growth parameters for *Pantoea dispersa* strain ODBP1 was conducted on M9 minimal medium with various carbon and nitrogen sources added and maintained at different pH and storage conditions. Screening of rice varieties for resistance against *Pantoea dispersa,* causing panicle blight disease, was scored in 65 paddy cultivars with different durations. None of the varieties showed an immune reaction to *Pantoea* blight; only resistant, moderately resistant, moderately susceptible, susceptible and highly susceptible disease reactions were recorded. The yield attributes of highly susceptible late maturing varieties were observed, with disease and disease incidence percentages of 88.72 % and 87.77 %, respectively. By investigating the specific growth requirements of *P. dispersa* and evaluating the resistance profiles of different rice cultivars, this study aims to bridge gaps in understanding pathogen behaviour and plant defence, contributing to effective integrated disease management strategies that support crop productivity and agricultural resilience.

Keywords: 16s rRNA; integrated disease management; *Pantoea* blight; *Pantoea dispersa*

Introduction

Rice is the principal sustenance for over 3.5 billion people worldwide, with *Oryza sativa* L. being the most prevalent variation among the several varieties. India ranks as the second-largest producer of rice globally and has the most extensive area dedicated to rice farming, around 43 million hectares (1, 2). Rice accounts for almost 40 % of India's overall food grain output. From 2019 to 2020, the rice cultivated area included 43.7 mha, with a total output of 118.4 mt and an average productivity of around 2705 kg/ha (1).

There has been an increase in a new rice disease associated with the *Pantoea* genus, which has been documented in several countries such as China, Malaysia, Germany, Turkey, Togo, Korea, India, Thailand, Brazil and Venezuela. The etiology of this disease has been attributed to *P. ananatis*, *P. stewartii*, *P. agglomerans* and *P. dispersa* (3, 4). The pathogenic *Pantoea* may infiltrate rice hosts via blooms, injuries inflicted by feeding insects, mechanical damage and plant contact during high winds, leading to severe instances that result in production losses ranging from 20 % to 100 % in rice (5). *Pantoea dispersa* is a Gram-negative bacterium recognized for its broad environmental adaptability and potential as a plant and opportunistic human pathogen (6, 7). In recent years, *P. dispersa* has emerged as a significant pathogen in rice (*Oryza sativa*), causing leaf and panicle blight, a disease that negatively impacts rice yield and quality (8-10). The bacterium *Pantoea dispersa* is increasingly recognized for its role in Pantoea panicle blight, a devastating disease affecting rice crops worldwide (11). Known for its versatility in adapting to various environmental conditions, *P. dispersa* can infect rice plants, leading to yield loss and compromised crop quality (12, 13). In early infection by *Pantoea dispersa*, the flag leaf becomes blighted, symptoms progress gradually and the panicles appear with brownish spots and gradually become completely chaffy. Some grains of the panicle remain green (Fig. 1). Recent studies have highlighted the importance of molecular characterization in understanding the pathogenicity and genetic diversity of

Fig. 1. Infected leaf and panicle showing blighted symptoms.

P. dispersa, which can lead to more effective disease management strategies (14). Molecular techniques, including 16S rRNA sequencing and multilocus sequence typing (MLST), have proven valuable in elucidating the genetic structure of *P. dispersa* populations and their evolutionary relationships (15).

In recent years, many species of *Pantoea* have been identified as tightly associated with rice-growing environments, functioning as epiphytes, endophytes, or pathogens. *Pantoea* spp. were often seen to promote plant development when identified as epiphytes or endophytes of rice and other crop too. The Identification of deceased *C. fimbriata* cells using Evans blue staining indicated that these *P. dispersa* strains exhibit fungicidal, rather than fungistatic, properties. Four *P.* dispersa strains significantly hampered the mycelial development and spore germination of *C. fimbriata,* while also altering the shape of the fungal hyphae (16). The enhancement of plant development by diverse *Pantoea* species may primarily be ascribed to many processes, including the manufacture of phytohormones such as indole-3-acetic acid (IAA), auxins, cytokinins, abscisic acid and gibberellic acid (17).

This pathogenic bacteria has the capacity to colonize and multiply in many habitats, with growth factors such as carbon and nitrogen sources, temperature, pH, inoculum concentration and incubation duration significantly influencing its development and virulence. Research demonstrates that growth characteristics affect bacterial virulence, as nutrition availability and environmental circumstances might influence bacterial survival and pathogenicity (18). The pathogenicity of *Pantoea* in rice plants may result from the synthesis of signal molecules linked to quorum sensing, which has been shown to influence bacterial pathogenicity, biofilm development and the production of exopolysaccharides and hydrolytic enzymes (19). The pathogenicity of *Pantoea* may also be ascribed to phytohormones, the hypersensitive response and pathogenicity (hrp) system and several secretion systems, including the T6SS in *P. ananatis* and the type III secretion system (T3SS) in *P. agglomerans* and *P. stewartii* (20). The increase in *Pantoea* panicle blight occurrence has necessitated the evaluation of rice cultivars for resistance to *P. dispersa* (21). Disease-resistant cultivars are fundamental to sustainable crop management, since they reduce reliance on chemical treatments and provide a long-term strategy for reducing crop losses (22). *Pantoea dispersa* has been sporadically documented about plant diseases; nonetheless, its position as a causal agent of panicle blight in rice is still inadequately investigated, particularly within Indian conditions. Knowledge of its epidemiology, host-pathogen interactions and survival mechanisms across many environmental conditions is lacking. Moreover, no extensive earlier research has discovered rice genotypes that are resistant or tolerant to this emerging disease, hence limiting the formulation of an effective disease management approach. This research intends to bridge the gaps in understanding pathogen behaviour and plant defence by examining *P. dispersa's* particular development needs and the tolerance levels of rice cultivars.

Materials and Methods

Isolation of pathogen

In November and December of 2023, seed samples from rice (*Oryza sativa*) exhibiting panicle blight symptoms were collected from paddy fields in Bargarh district, Odisha, India, with about 70 % disease incidence. Symptoms were observed in the panicles in which rusty, brownish water-soaked lesions appeared on the lemma or palea of the grains. Five strains of bacteria were isolated from the diseased samples from the field. Bacterial colonies were isolated from surface-sterilized infected seeds using 70 % ethanol and 1 % sodium hypochlorite, rinsed three times in sterilized water, placed on King's B agar medium and incubated for 48 hr at 30 °C. For preservation, single colonies were placed to King's B broth medium and shaken at 200 rpm overnight. All isolated bacteria were gram-negative facultative anaerobes, motile, positive for catalase and citrate utilization test and incapable of producing indole & hydrosulfuric acid. Colonies were round, smooth, slimy with irregular edges and produced yellow pigment on King's B agar medium.

Molecular identification and characterization

DNA isolation was done in the virology laboratory in the Department Plant Pathology by using 3 days old bacterium inoculated nutrient broth culture and using DNeasy Ultraclean Microbial Kit (Cat. No. 12224-250, Qiagen). DNA amplification was performed using ProFlex PCR system (ThermoFischer Scientific). The genomic DNA from sample was used at a concentration of 25 ng per µl with the reaction mixture containing 10X HiBuffer A (With 15mM $MgCl_2$) 2.5 µL, dNTPs solution (10mM) 2.5 µL, Primers (Forward + Reverse) 1 +1 µL, Taq polymerase (5U/µL) 0.5 µl, dH_2O 11 µL and the template DNA 5µL. For PCR amplification of 16S nuclear small subunit rRNA, universal primers were used. 27F- (5' GAGTTTGATCATGGCTCAG 3'), 1429R (5' GGTTACCTTGTTSCGACTT 3'). The PCR program included pre-denaturation at 95 °C for 4 min followed by denaturation at 94 °C for 30 sec, annealing temperature at 57 °C for 30 sec, extension at 72 °C for 1 min and final

extension at 72 °C for 10 min. The resulting amplicons were analyzed by resolving them in a 1 % agarose gel using 1X Tris-acetate-EDTA (TAE) buffer.

To further purify the DNA fragments from agarose gel QIAquick® Gel Extraction Kit (Qiagen, Germany) was used for gel extraction. Sanger sequencing of the purified PCR product was carried out at Eurofins Genomics India Pvt. Ltd., Bengaluru, Karnataka. The instrument used for sequencing was ABI 3730XL.

Study of growth parameters of the bacteria

The carbon, nitrogen sources, optimum temperature, pH, inoculum concentration and incubation period for bacterial biomass production were optimized using M9 minimal medium and the biomass was measured by its optical density at 600nm using multiscan nanodrop cum spectrophotometer cum ELISA reader (Multiskan skyhigh, Thermofischer Scientifics, Massachusetts, United States) after 24 hr of incubation. The M9 medium containing 95 mM Na_2HPO_4, 44 mM KH_2PO_4, 17 mM NaCl, 37 mM NH_4Cl, 0.1 mM $CaCl_2$, 2 mM $MgSO_4$ and 50 mM glucose (23). All experiments were performed with three replications to evaluate the best conditions for highest biomass production of the test pathogen.

The carbon source for bacterial biomass production was identified using M9 minimal medium supplemented with various carbon sources, including glucose, fructose, maltose, lactose, sucrose and galactose. Each 100 mL of medium was inoculated with 10 mL of 24 hr fresh culture, incubated at room temperature for 24 hr and biomass was measured at OD_{600} using a UV-Vis spectrophotometer. The nitrogen source was optimized using M9 minimal medium with glucose as the carbon source and varied nitrogen sources such as ammonium dihydrogen phosphate, ammonium molybdate, peptone, tryptone, yeast extract and beef extract. Each medium was inoculated and incubated as before and OD_{600} measurements were taken. The optimum temperature for biomass production was determined by incubating the optimized medium (M9 with glucose and tryptone) at different temperatures (20 °C, 25 °C, 30 °C, 35 °C and 40 °C) and OD_{600} was measured after 24 hr. The optimum pH was evaluated by adjusting the medium's pH between 5 and 8 using 1N HCl and 1N NaOH, followed by incubation at the optimal temperature, with OD600 readings determining the optimum pH. Inoculum concentration was optimized using varying inoculum sizes (1 %, 5 %, 10 %, 15 % and 20 %) and the resulting biomass production was measured. Finally, the incubation period was optimized by incubating the culture for 24, 48, 72, 96 and 120 hr, with OD_{600} measurements taken at each time point to determine the optimum incubation time. All experiments were performed with three replications.

Screening for *Pantoea* panicle blight resistance

For screening of the rice germplasm against bacterial leaf and panicle blight resistance, artificial inoculation was preferred over natural infection as it ensures the rice plants are properly exposed to right amount of inoculum for cause of the disease. Screening for *Pantoea* blight was carried using the method described at OUAT Instructional farm situated at university agriculture farm road, Bhubaneswar, Odisha (24). All the 65 genotypes with different durations were planted in a line of three rows of 2 m length. Three replications were maintained, adopting a spacing of 20 × 15 cm for screening against resistance to *Pantoea* panicle blight. The inoculum was prepared for field inoculation by suspending two days old *Pantoea* cultures in sterile distilled water. The concentration was adjusted to approximately 108 CFU mL^{-1} (OD at 600= 0.5) using spectrophotometer. The inoculum was immediately used within 2 hr. Plants were inoculated artificially by infiltrated method at 80 days after sowing. In this method, bacterial suspension in filtered at about 2-5 cm below the tip of the leaves. The data was recorded three weeks after inoculation. Observation on reaction to bacterial blight was recorded by physical measurement of lesion length to that of the leaf length and per cent diseased leaf area was worked out, categorized based on 0-9. Further disease index was computed based on the scores using the formula presented in Table 1.

Table 1. Disease scoring scale

Percentage disease severity (%)	Disease score	Disease reaction
0 %	0	Immune
1-10 %	1	Resistant
10-30 %	3	Moderately Resistant
30-50 %	5	Moderately Susceptible
50-70 %	7	Susceptible
70-100 %	9	Highly susceptible

Observations recorded

Plant height was measured in centimetres from the soil surface to the tip of the tallest panicle at harvest. Productive tillers per plant were determined by counting the number of ear-bearing tillers at maturity. Panicle length was recorded as the distance from the neck node to the tip of the last spikelet. Test weight was measured by weighing 1000 well-filled grains, randomly selected at 12 % moisture level, using a grain counter and expressed in grams. Grain yield per plant was calculated by harvesting, threshing, cleaning and drying five randomly selected plants to 12 % moisture, then weighing the grains and expressing the yield in grams.

Per cent (%) disease incidence =

$$\frac{\text{Number of plants infected}}{\text{Total plant population}} \times 100 \quad \text{(Eq. 1)}$$

Per cent (%) disease severity =

$$\frac{\text{Leision length (cm)}}{\text{Total height of the same plant}} \times 100 \quad \text{(Eq. 2)}$$

Per cent (%) disease index =

$$\frac{\text{Sum of all disease ratings}}{\text{Total number of rating} \times \text{Max. disease grade}} \times 100 \quad \text{(Eq. 3)}$$

Results

Molecular identification of bacterial pathogen by 16S rDNA gene

The bacterial colony are straw to slight yellowish in colour with mucoid surface, raised elevation, rounded shape with entire margin and no pigmentation and sporulation after 48 hr of incubation period in King's B culture medium (Fig. 2). The bacterial DNA was isolated and the 16S rDNA region of

Fig. 2. Colonies of *Pantoea dispersa* isolate ODBP1 on King's B agar medium.

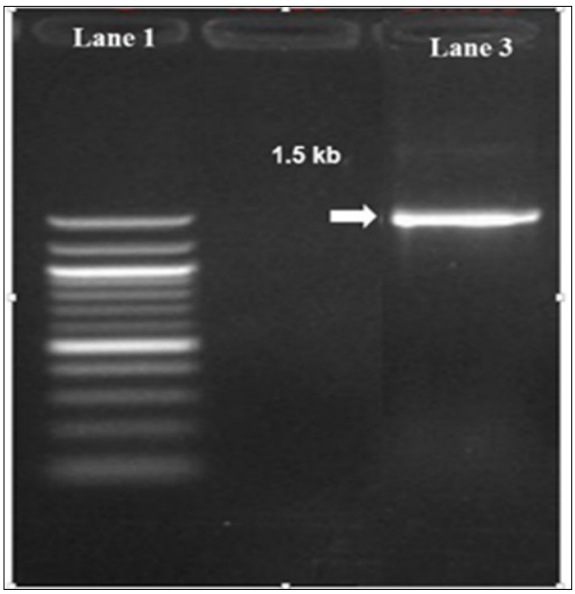

Fig. 3. Lane 1 100bp ladder; Lane 3- isolate ODBP1 Amplicon size of 1.5kb on 1 % agarose gel.

the pathogen isolates was amplified with the universal primer pair 27F and 1492R. The three isolates namely ODBP1 produced a band of ~1500 bp or 1.5kb upon PCR amplification (Fig. 3). These amplicon were subjected to Sanger sequencing and the obtained nucleotide sequences aligned with the help of MEGA-XI alignment software. Finally, the FASTA sequences of the isolate was analyzed through NCBI mega BLAST and submitted in NCBI GenBank with under 16S rRNA of isolate ODBP1 with accession numbers (OR672917.1). The isolate ODBP1 showed 100 % sequence similarity with *Pantoea dispersa* (Fig. 4).

Optimization of growth parameters for *Pantoea dispersa*

The *Pantoea dispersa* strain ODBP1 was grown on M9 minimal medium with various carbon and nitrogen sources added and maintained at different pH and stored at various temperature for different periods to see how they affect the bacterial growth (Table 2). Six carbon sources were tested and the isolate grew positively. However, the strain produced much more biomass in the presence of galactose. Other carbon sources, such as sucrose, fructose, glucose, maltose and lactose supplied with M9 minimal medium, likewise encouraged growth, but the biomass production was substantially lower than galactose. Sucrose exhibited slightly higher biomass production than fructose, followed by maltose, glucose and lactose. Compared to other carbon sources, maltose, glucose and lactose were at par with each other in M9 minimal medium produced lower biomass. The

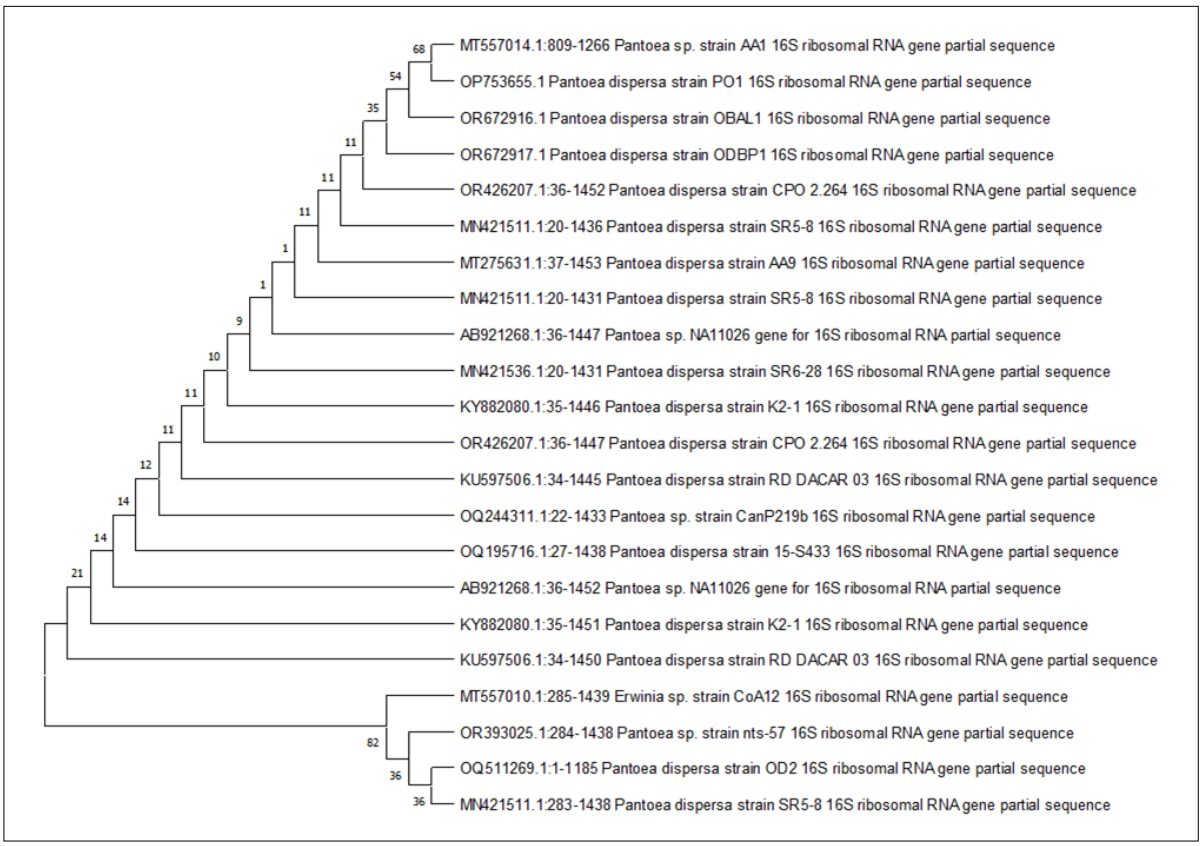

Fig. 4. Phylogenetic tree of the isolate ODBP1.

Table 2. Growth requirement of *Pantoea dispersa* isolate ODBP1

Sl. No.	Carbon source		Nitrogen source		Temperature		Ph		Inoculation concentration		Inoculation periods	
1.	T_1 (Glucose)	2.362^{d} (0.171)	T_1 (Ammonium dihydrogen phosphate)	3.838^{e} (0.449)	T_1 (20°C)	1.462^{d} (0.065)	T_1 (5)	1.564^{f} (0.075)	T_1 (1 %)	2.592^{e} (0.205)	T_1 (24 hrs)	4.325^{c} (0.569)
2	T_2 (Fructose)	3.27^{c} (0.326)	T_2 (Ammonium molybdate)	1.919^{f} (0.113)	T_2 (25°C)	1.855^{d} (0.105)	T_2 (5.5)	2.026^{f} (0.125)	T_2 (5 %)	5.355^{a} (0.872)	T_2 (48 hrs)	5.516^{a} (0.925)
3.	T_3 (Maltose)	2.705^{d} (0.223)	T_3 (Peptone)	4.612^{d} (0.647)	T_3 (30°C)	3.866^{b} (0.455)	T_3 (6)	3.265^{e} (0.325)	T_3 (10 %)	4.608^{b} (0.646)	T_3 (72 hrs)	5.040^{b} (0.772)
4.	T_4 (Lactose)	1.966^{d} (0.118)	T_4 (Tryptone)	7.463^{a} (1.688)	T_4 (35°C)	4.372^{a} (0.582)	T_4 (6.5)	4.084^{d} (0.508)	T_4 (15 %)	3.939^{c} (0.472)	T_4 (96 hrs)	3.733^{d} (0.424)
5.	T_5 (Sucrose)	4.171^{b} (0.530)	T_5 (Yeast extract)	6.079^{c} (1.123)	T_5 (40°C)	3.462^{c} (0.365)	T_5 (7)	4.679^{b} (0.666)	T_5 (20 %)	3.124^{d} (0.297)	T_5 (120 hrs)	2.600^{e} (0.206)
6.	T_6 (Galactose)	5.285^{a} (0.850)	T_6 (Beef extract)	6.631^{b} (1.335)	-	-	T_6 (7.5)	5.305^{a} (0.856)	-	-	-	-
7.	-	-	-	-	-	-	T_7 (8)	4.426^{c} (0.596)	-	-	-	-

Values in the parentheses indicate corresponding arcsine transformed values. Mean value in a column followed by same superscript letter are not Significantly different according to Duncan's multiple range test (DMRT) at P≤0.05

optimal nitrogen source for biomass production was determined to be inorganic nitrogen source tryptone. Other nitrogen sources, such as Beef extract and Yeast extract, favoured the growth of the pathogen, but biomass production was significantly lower than the tryptone. Next best in the order of merit was Beef extract which was at par with Yeast extract followed by peptone and Ammonium dihydrogen phosphate. Less or negligible growth was observed in Ammonium molybdate. Based on result obtained, galactose as carbon source and tryptone as nitrogen source were utilized in subsequent investigations. The most favourable temperature for the growth was determined to be 35 °C with remarkable bacterial biomass, but when the temperature decreased to 25 °C or less than it, bacterial biomass production was decreased. The maximum level of biomass production was obtained at pH 7.5, however the isolate could grow in wide range of pH 6 to 8, but biomass production was less when the pH was 5.5 or lower. Growth of the *Pantoea dispersa* isolate BP1 was studied using five different levels of inoculum concentration viz., 1 %, 5 %, 10 %, 15 %, 20 % concentration of isolate produced a substantial amount biomass in the M9 minimal medium which had Galactose as carbon source, Tryptone as nitrogen source and maintained at 35 °C temperature and pH at 7.5. Subsequent increase in inoculum concentration to 10 %, 15 % and 20 % witnessed a decrease in growth of the bacterium. The least growth was observed with inoculum concentration of 1 % compared to the other inoculum concentrations. The highest growth was recorded on 2nd day of incubation followed by 3 day of incubation and 1 day. Then on 4th day afterwards the bacterial growth was significantly reduced. Thus, the significant growth was observed in M9 minimal medium had Galactose as carbon source and Tryptone as nitrogen source, with inoculum size 5 %, pH 7.5 and temperature 35 °C and incubation period of 48 hr.

Screening of rice varieties for resistance against *Pantoea dispersa* causing panicle blight disease

Rice varieties were screened to check the resistance sources against the *Pantoea dispersa* causing panicle blight disease (Table 3). The reaction of varieties for resistance and susceptibility against *Pantoea dispersa* was scored in 65 paddy cultivars with different durations. Rice cultivars were categorized in order of their maturation period viz. late maturing (130-150 days) mid maturing (120-130 days) and early maturing (90-120 days). Different varieties were inoculated on varying dates coinciding with their booting stage. Panicle emerged 20 days after inoculation in almost all the varieties and exhibited symptoms in the panicle Since this is a newly identified disease in rice, a novel disease scoring scale was developed and presented in Table 1. None of the variety was found to show immune reaction to *Pantoea* blight and only resistant, moderately resistant, moderately susceptible, susceptible and highly susceptible disease reaction, disease incidence % and per centage disease index was recorded. As this disease mainly affects the panicles, grain yield per plant and test weight was recorded. In early maturing varieties (90-120 days) only Ghanteswari cultivar exhibited resistance towards the pathogen while BRRI Dhan 75, Sidhanta exhibited moderately resistant reaction towards the pathogen. In case of early maturing varieties, Mandakini cultivar exhibited moderately susceptible reaction towards the *Pantoea* blight. Rice cultivar Jyortirmayee, Khandagiri,

Table 3. Varietal screening of different rice genotypes against the bacterial pathogen *Pantoea dispersa*

Sl. No.	Rice cultivar	Duration	% Disease incidence	% Disease index	Disease score	Reaction	Grain yield/ plant (g)	Test wt (g)
1	Samanta	140	53.33	48.88	5	MS	27.54	23.84
2	Gajapati	130	56.66	52.22	7	S	19.30	18.25
3	Bhoi	125	64.88	58.88	7	S	23.27	20.73
4	Meher	140	33.33	32.22	5	MS	28.63	24.45
5	Jajati	135	9.09	8.88	1	R	40.67	31.49
6	Indravati	150	42.86	44.44	5	MS	28.57	23.96
7	Ashutosh	150	15.95	15.55	3	MR	33.30	27.21
8	Surendra	135	72.05	72.22	9	HS	13.40	13.31
9	Kanchan	160	16.53	16.66	3	MR	37.43	28.78
10	Prachi	155	25.27	26.66	3	MR	37.30	28.26
11	Prativa	125	65.66	67.77	7	S	21.25	18.73
12	Kalingadhan 1021	135	82.50	82.22	9	HS	12.93	12.03
13	Rambha	160	26.66	27.77	3	MR	32.97	26.61
14	Sarathi	120	35.70	36.66	5	MS	25.60	21.86
15	Tejaswini	135	77.19	78.88	9	HS	14.40	13.31
16	Kharavela	125	56.66	56.66	7	S	21.33	18.35
17	Gouri	135	29.97	31.11	5	MS	24.73	21.47
18	Manaswini	132	10.23	11.11	3	MR	33.10	26.62
19	MTU 1053	125	83.92	85.55	9	HS	15.43	12.84
20	Bhanja	140	51.58	49.99	5	MS	26.90	22.08
21	ARIZE 644 Gold	110	45.02	44.44	5	MS	27.73	23.69
22	Geeta	135	65.66	66.66	7	S	20.87	19.55
23	Khandagiri	95	35.08	36.66	5	MS	26.27	23.19
24	Daya	125	33.33	33.33	5	MS	26.70	23.47
25	Anjali	150	76.47	78.88	9	HS	12.87	12.55
26	MTU1159 Mahamaya	130	44.11	45.55	5	MS	22.30	20.28
27	Salivahan	135	26.04	25.55	3	MR	33.10	27.19
28	Chinikamini	135	91.56	91.11	9	HS	9.82	9.97
29	Ghanteswari	95	7.29	7.77	1	R	41.03	32.86
30	Rajeswari	135	63.17	63.33	7	S	16.63	16.94
31	Urvashi	145	53.33	54.44	7	S	18.33	17.41
32	Jagannath	150	33.33	32.22	5	MS	27.53	22.89
33	Pratikshya	142	23.33	23.33	3	MR	32.63	26.44
34	Hema	135	51.61	52.22	7	S	19.33	18.41
35	Hasanta	145	46.30	47.77	5	MS	25.67	22.51
36	Jyotirmayee	96	31.25	32.22	5	MS	23.37	21.74
37	Pratap	135	32.55	33.33	5	MS	22.33	20.27
38	Sarala	125	17.23	19.99	3	MR	33.83	27.48
39	Kalingadhan 1052	135	83.33	84.44	9	HS	17.13	15.63
40	Mahalaxmi	155	22.22	21.11	3	MR	35.97	28.04
41	Savitri	110	14.30	14.44	3	MR	32.27	26.71
42	Manika	155	54.07	54.44	7	S	15.50	15.46
43	Ranidhan	115	18.02	18.88	3	MR	34.87	27.06
44	Kalingadhan 1501	135	88.46	87.77	9	HS	12.47	12.35
45	Uphar	162	18.38	18.88	3	MR	33.20	26.66
46	Swarna sub 1	145	80.95	82.22	9	HS	13.53	12.39
47	Sidhanta	96	19.59	17.77	3	MR	31.87	25.99
48	Jagabandhu	150	61.05	61.11	7	S	20.13	18.30
49	Mahanadi	150	47.97	48.88	5	MS	24.83	21.48
50	Santepheep	155	6.74	6.66	1	R	39.73	31.49
51	Tanmayee	146	26.57	26.66	3	MR	33.43	26.78
52	Mrunalini	146	78.56	79.99	9	HS	13.67	12.92
53	Hiranmayee	135	10.29	11.11	3	MR	34.13	26.69
54	Birupa	135	39.34	39.99	5	MS	25.87	22.00
55	Ramachandi	155	66.31	66.33	7	S	19.87	18.17
56	Mandakini	100	54.99	55.55	7	S	21.41	18.54
57	Savaitri	135	36.46	37.77	5	MS	27.53	22.84
58	Pooja	150	32.35	32.22	5	MS	29.30	23.34
59	CR 1009	160	51.52	51.11	7	S	17.57	16.63
60	Tezz Gold	115	72.22	73.33	9	HS	18.17	14.72
61	Ajit	135	42.31	42.22	5	MS	26.17	21.26
62	Brri Dhan 75	115	22.10	23.33	3	MR	34.73	27.44
63	Swarna	150	86.72	87.77	9	HS	14.17	12.93
64	Bhuban	140	9.09	9.99	1	R	41.93	33.21
65	Nua Aacharmati	140	90.66	92.22	9	HS	8.96	9.87

Arize 644 Gold exhibited susceptible reaction towards the pathogen. In case of mid maturing varieties (120-130 days) no variety showed resistant reaction against the pathogen while Ranidhan, Savitri, Sarala, Salivahan exhibited moderately resistant reaction towards the pathogen. The mid maturing cultivars Gajapaati, Bhoi, Prativa, Kharavela and Geeta exhibited moderately susceptible against the *Pantoea* blight. In contrast, cultivars Ajit, Savitri, Pooja, Daya, Sarathi, MTU 1159 and Mahamaya exhibited susceptible reaction against the pathogen. The cultivars Nua Acharmati, Kalingadhan 1052, Chinikamini, Anjali, MTU 1053 and Kalingadhan 1021 exhibited highly susceptible disease reaction against the pathogen. In the late maturing varieties (130-150 days), Bhubana, Santeheep, Jajati exhibited resistant reaction towards the pathogen. In contrast, Hiranmayee, Uphar, Mahalaxmi, Pratikshya, Manaswini, Rambha, Kanchan, Prachi, Ashutosh and Tanmayee exhibited moderately resistance and Urvashi, Rajeswari, Manika, Jagabandhu, Ramachandi, CR1009 showed moderately susceptible reaction towards the pathogen (Table 4). Among the cultivars Birupa, Mahanadi, Hasanta, Pratap, Jagannath, Bhanja, Gouri, Indravati, Meher, Samanta were susceptible to the pathogen. In contrast, Swarna, TEZZ Gold, Mrunalini, Swarna sub-1, Tejaswini, Surendra exhibited highly susceptible reaction against the pathogen. The yield attributes were observed where in case of some highly susceptible late maturing varieties like Chinikamini, the disease incidence was found to be 91.56 % and grain yield per plant was only 9.82 g and test weight was 9.97 g. In the popular cultivar Swarna, the disease incidence % and PDI % was found to be 88.72 % and 87.77 % respectively. Similar trends were observed in highly susceptible cultivars against the *Pantoea* blight pathogen. In case of resistant cultivars like in Bhuban the disease incidence was observed to be 9.09 % and % disease index was 9.99 % and the grain yield per plant and test weight were found to be 41.93 g and 33.21 g, respectively.

Discussion

The findings from the growth study of *Pantoea dispersa* strain ODBP1 provide insights into its optimal conditions for growth in M9 minimal medium, highlighting the effects of various carbon and nitrogen sources, pH, temperature and inoculum concentration. The study indicates that tryptone is the optimal nitrogen source for the growth of *P. dispersa* strain ODBP1, followed by beef extract, yeast extract, peptone and ammonium dihydrogen phosphate. Tryptone, a rich source of peptides and amino acids, has been previously identified as a preferred nitrogen source for various bacteria due to its ability to support both fast growth and high biomass production (25). The other nitrogen sources tested, such as beef and yeast extract, also provided substantial growth, suggesting that *P. dispersa* has versatile nitrogen utilization capabilities. However, ammonium dihydrogen phosphate, a simpler, inorganic nitrogen source, did not support growth as efficiently, likely due to its limited ability to provide the necessary organic compounds for optimal bacterial metabolism (26).

Regarding carbon sources, the study found that galactose was the best carbon source for promoting the growth of *P. dispersa*, which aligns with other studies indicating that monosaccharides like galactose can be readily metabolized by many bacteria (27). While *P. dispersa* and related species often utilize other sugars such as glucose and fructose, galactose may be preferred in this strain because its metabolic pathways are more directly linked to the strain's enzymatic activities (28). This suggests that *P. dispersa* strain ODBP1 may harbour specialised enzymes that enable more efficient breakdown and utilization of galactose compared to other sugars. The optimal growth temperature range for *P. dispersa* was between 30 °C and 35 °C, with maximum biomass production occurring at pH 7.5. These results are consistent with the typical growth conditions for mesophilic bacteria, which generally thrive at moderate temperatures (29, 30). The near-neutral pH of 7.5 further supports the idea that *P. dispersa* is adapted to environments where pH conditions are close to neutral, which is common for many environmental and gut-associated bacteria (31) . The effect of inoculum size was also significant, with the highest growth observed on the second day of incubation, which is typical for many bacteria that require an initial lag phase to adapt to the growth medium before exponential growth ensues. The 5 % inoculum concentration was optimal, suggesting that *P. dispersa* strains generally require a moderate starting cell density to reach

Table 4. Category of rice varieties screened against *Pantoea dispersa*

Sl. No.	Days to maturation	Immune	Resistant	MR	S	MS	HS
1	**Early maturing (90-120 days)**	-	Ghanteswari	Brri Dhan 75 Sidhanta	Jyotirmayee Khandagiri Arize 644 Gold	Mandakini	
2	**Mid maturing (120-130 days)**	-		Ranidhan Savitri Sarala Salivahan	Ajit Savitri Pooja Daya Sarathi MTU1159 Mahamaya	Gajapati Bhoi Prativa Kharavela Geeta	Nua Acharmati Kalingadhan 1052 Chinikamini Anjali MTU 1053 Kalingadhan 1021
3	**Late maturing (130-150 days)**	-	Bhubana Santeheep Jajati	Hiranmayee Upahar Mahalaxmi Pratikshya Manaswini Rambha Kanchan Prachi Ashutosh Tanmayee	Birupa Mahanadi Hasanta Pratap Jagannath Bhanja Gouri Indravati Meher Samanta	Urvashi Rajeswari Manika Jagabnadhu Ramachandi CR 1009	Swarna TEZZ Gold Mrunalini Swarna sub 1 Tejaswini Surendra

optimal growth. Too low of an inoculum may result in slower colonization and growth due to limited cell-to-cell interactions and nutrient availability. Overall, the growth conditions identified in this study-M9 minimal medium with galactose as the carbon source, tryptone as the nitrogen source, a pH of 7.5, a temperature range of 30-35 °C and a 5 % inoculum size-provide an efficient environment for the cultivation of *P. dispersa* ODBP1. These conditions could be utilized for further studies aimed at understanding the metabolic pathways of this strain and its potential applications in biotechnology, particularly in processes requiring the conversion of galactose or other sugars into valuable products (32). The bacteria were shown to flourish at a wide spectrum of temperatures, from 4 to 41 °C and pH values between 2 and 8 (33). However, their most favourable conditions for growth of the bacteria occur at a temperature of 28-30 °C and a pH of 7, as indicated by previous research (34). Different researchers were observed the growth of *B. glumae is* temperature dependent. The growth was preferably more in the semi-selective than in the non-selective media at 30 °C. Similar growth patterns were observed over different temperatures among virulent and avirulent strains of *B. glumae*, with varying rates of growth expressed at 30 °C and 35 °C (35).

The study examined the resistance and susceptibility of 65 paddy cultivars *against Pantoea dispersa.* The cultivars were classified based on their maturation period, with no immune reactions observed. However, only resistant, moderately resistant, moderately susceptible, susceptible and highly susceptible disease reactions were recorded. Early maturing varieties showed only Ghanteswari resistance, while mid-maturing cultivars showed moderate resistance. Late maturing cultivars showed moderate resistance, while highly susceptible cultivars had high disease incidence and PDI %.

The research indicated that disease responses varied among the three categories of paddy varieties-early, mid and late developers-ranging from resistant to susceptible. Most early and mid-maturing varieties showed moderate resistance to the disease. The shorter incubation period observed in these varieties can be linked to inoculation during the booting stage, which coincides with the panicle emergence at the heading stage. This brevity in incubation may hinder the pathogen's ability to reach the population density required for high disease severity (36, 37). Thus, the maturation duration of paddy varieties appears to influence the expression of panicle blight symptoms.

Additionally, genetic factors of specific varieties may play a role, as some early and mid-maturing types exhibited a moderate susceptibility to the disease (38). Notably, early maturing varieties seem to have minimized the impact of environmental conditions due to their shorter pathogen incubation period following artificial inoculation. Their genetic traits might also contribute to a resistant response, minimizing susceptibility to *Pantoea* blight.

No varietal screening was done against the *Pantoea* spp. previously. Screening was carried out for other rice diseases like BLB, bakane, etc. Screening was performed on cultivated rice cultivars to assess their resistance to bacterial grain rot (39). The results indicated that none of the cultivars had a resistant reaction against BPB (bacterial grain rot pathogen), with the majority exhibiting a moderate resistance to susceptibility reaction.

Conclusion

The study on *Pantoea dispersa* reveals significant insights into the pathogen's growth requirements, molecular Identification and the resistance profiles of various rice cultivars. The optimal growth conditions for *P. dispersa* included galactose as a carbon source, tryptone as a nitrogen source, a temperature range of 30-35 °C, a pH of 7.5 and an inoculum concentration of 5 %. These findings provide a deeper understanding of the pathogen's biology and offer essential information for its management. Screening of 65 rice cultivars revealed a range of responses to the pathogen, with early and mid-maturing varieties exhibiting moderate resistance, while late-maturing cultivars showed varying degrees of susceptibility. Importantly, no rice cultivars displayed complete immunity to *P. dispersa*, underscoring the need for continued efforts in breeding and management practices aimed at enhancing resistance. The findings contribute to developing integrated disease management strategies, emphasizing the importance of optimizing environmental conditions and selecting resistant cultivars for sustainable rice production.

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Authors' contributions

BJ, AKS, SKB, JP, PN, SSP and HCK contributed equally to the discussion of the results and the preparation of the final manuscript. All the authors approved and read the final manuscript.

Compliance with ethical standards

This study did not involve human or animal subjects and thus did not require ethical approval.

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