Assessment of black rot (*Xanthomonas campestris*) of cabbage (*Brassica oleracea*) in East Khasi Hills, Meghalaya

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ABSTRACT

Cabbage (*Brassica oleracea* L. var. capitata) holds significant economic importance in Meghalaya, thriving in its high altitude and cool climate. East Khasi Hills (EKH) district, where cabbage cultivation remains viable throughout the year, contributes 70% of the state's total cabbage production. A severe outbreak of cabbage black rot, caused by *Xanthomonas campestris* p.v. *campestris*, has raised concerns in this region. To comprehensively understand the outbreak and assess its economic impact, an extensive disease survey was conducted across 6 districts of Meghalaya, with a particular focus in the EKH during 2021–22 and 2022–23. The survey was on different cruciferous vegetables such as cabbage, broccoli (*Brassica oleracea* L. var. italica), cauliflower (*Brassica oleracea* L. var. botrytis), radish (*Raphanus sativus* L.), mustard (*Brassica juncea* L.), and knol khol (*Brassica oleracea* L. var. gongylodes). It was found that on an average, Meghalaya experiences a disease incidence (DI) of a 52.7% and per cent disease index (PDI) of 30.9% for black rot in cabbage, that surpassed knol khol and cauliflower. Highest DI and severity were observed during monsoon season. Further analysis revealed significant correlations (r) between black rot incidence and severity in cabbage with temperature (0.91), humidity (0.87) and rainfall (0.88). Additionally, multiple regression predictive model performed using weather parameters and DI led to the prediction of PDI with accuracy of 95% in EKH.

Keywords: Black rot, Cabbage, Disease incidence, Per cent disease index, Xanthomonas campestris

India is the second largest producer of cabbage (*Brassica oleracea* L. var. capitata) in the world boasting a substantial cultivation area of 0.4 million hectares, coupled with an impressive output of 9.59 million tonnes. Meghalaya features a land area of 1,947 hectares and yields a production output of 42,783 metric tonnes with an average yield of 21,964 kg/ha (Directorate of Economics and Statistics, Meghalaya 2020). Notably, the East Khasi Hills (EKH) district, situated at an average elevation of 940 m and temperature of 25–26°C, annual rainfall of 191.9 inches with approximately 109 rainy days/year (India Meteorological Department, Meteorological Centre, Shillong 2023), creates an ideal environment for the year-round cultivation of cole crops.

Black rot, caused by gram negative bacterium Xanthomonas campestris pv. campestris (Xcc), affects

¹College of Post Graduate Studies in Agricultural Sciences (Central Agriculture University, Imphal), Umiam, Meghalaya; ²Sher-e-Kashmir University of Agricultural Sciences and Technology, Jammu, Jammu and Kashmir; ³ICAR-Research Complex for North Eastern Hill Region, Umiam, Meghalaya. *Corresponding author email: sudharshankr1996@gmail.com cruciferous crops and is a leading cause of substantial agricultural losses on a global scale (Sharma et al. 2017). The disease, originating from seeds and soil has the capacity to invade plants through roots, wounds, hydathodes, and leaf stomata. Bacterial cells combine with xanthan, obstruct xylem vessels, impeding water transport and giving rise to distinct v-shaped lesions that initiate at leaf edges. These lesions are accompanied by darkened veins, tissue chlorosis, discolouration, leaf detachment and ultimately leads to plant death. Significant reduction in yield through early defoliation and a decline in the quality of cabbage heads have been observed. In cabbage and cauliflower, losses range from 50-70% (Gupta et al. 2013). It affects cruciferous vegetables including broccoli (Brassica oleracea L. var. italica), brussels sprouts, cabbage, cauliflower, radish (Raphanus sativus L.), mustard (Brassica juncea L.), kale (Brassica oleracea L. sabellica), rutabaga (Brassica napus rapifera) and turnip (Brassica rapa), as well as cruciferous weeds such as shepherd's purse and wild mustard.

The primary objectives of the present study were to conduct a comprehensive surveillance of the incidence and severity of black rot in cruciferous crops, with a particular focus on cabbage, across all four seasons in the EKH of Meghalaya and to identify and analyse the various factors that contribute to the proliferation of the disease.

MATERIALS AND METHODS

Survey area: In the present study-survey on prevalence of black rot in cabbage and other cruciferous vegetables was made during 2021–22 and 2022–23 in three different ways for different areas of the states. During the survey, fields were sampled randomly at an interval of 5–10 km along roads and distances between fields depending on the topography and the relative importance of crop and cultivation within each district. The districts of Meghalaya for the present study were selected based on production and productivity of cabbage as well as accessibility by road. For the disease assessment in cabbage, 27 villages each with three fields were surveyed and for other crucifers, 10 villages with three fields were surveyed. The surveys were carried out at the time of crop harvest. Surveyed areas represent different agro-ecological zones and constitute 85–90% of the total cropped area in Meghalaya. Notably around 70% of the total cabbage cropped area is from the East Khasi Hills (EKH) district.

Survey 1, part 1: The prevalence of black rot disease in cabbage in 6 prominent cultivation districts of Meghalaya, viz. Ribhoi, East Khasi Hills (EKH), East Garo Hills (EGH), West Garo Hills (WGH), East Jaintia Hills (EJH) and West Jaintia Hills (WJH) was recorded during 2021–22 crop season (Table 1).

Survey 1, part 2: A comprehensive survey on black rot in cabbage was conducted in the EKH district in all the 4 growing seasons, viz. spring, monsoon, autumn, and winter to assess the levels of disease incidence (DI) and severity in 2022–23.

Survey 2: An encompassing survey was carried out in the EKH district to evaluate DI and severity of black rot in 5 different cruciferous vegetables, viz. broccoli, cauliflower,

Table 1 Estimation of disease incidence and per cent disease index of black rot of cabbage in major cabbage growing areas during 2021–22

istrict Village Agro-clima		Agro-climatic zone	Previous crop	Disease incidence (DI)	Per cent disease index (PDI)	
East Khasi Hills (EKH)	Laitkor	Cold moist	No crop	49.06±7.8 efg	19.85±2.1 ^{abcd}	
	Myllium	Cold moist	Cabbage	61.61±3.0 ^{ijk}	43.31 ± 3.2^{ij}	
	Mawlot	Cold moist	No crop	48.45±3.2 ef	29.71 ± 6.5^{fg}	
	Smith	Cold moist	Cabbage	85.33±3.5 op	$52.66{\pm}1.5^{k}$	
	Ladmawreng	Cold moist	Cabbage	66.92±1.7 ^{jkl}	$30.00{\pm}3f^g$	
	Nangkrem	Cold moist	Cabbage	67.91±2.5 kl	39.38±3.9 ^{hi}	
	Mawlong	Cold moist	Cauliflower	73.45±3.0 ^{lm}	$44.0{\pm}2.6^{ij}$	
	Mawngap	Cold moist	Cauliflower	78.68±3.6 mno	49.76 ± 4.7^{jk}	
	Pepbah	Cold moist	Cabbage	91.00±1.0 ^p	$80.33{\pm}1.5^{1}$	
	Umphyrani	Cold moist	Cabbage	77.31±2.8 mn	$44.73{\pm}\ 2.7^{ij}$	
Ri Bhoi	Umiam	Mild moist	Potato	52.33±1.5 fgh	32.24 ± 5.8^{gh}	
	Nongpoh	Mild moist	Potato	47.53±5.1 ef	21.55±6.2abcde	
	Bhoirymbong	Mild moist	Potato	50.64 ± 5.4 fg	$25.52\pm6.3d^{efg}$	
	Umsning	Mild moist	Cabbage	53.74±5.7 fgh	13.27±4.9 ^a	
East Jaintia Hills (EJH)	Khliehriat	Mild moist	Cauliflower	56.84±6.1 ghi	18.50±4.6 ^{abcd}	
	Moosyiem	Mild moist	Cauliflower	59.95±6.4 hij	26.57 ± 5.4^{defg}	
	Pammanik	Mild moist	No crop	28.12±1.7 ab	15.88±3.6 ^{abc}	
	Mukhle	Mild moist	No crop	20.96±2.1 a	22.00 ± 0.9^{bcdef}	
West Jaintia Hills (WJH)	Jowai	Cold wet	Cabbage	82.33±1.5 no	29.09±5.9efg	
	Amlarem	Cold wet	Potato	31.00±1.7 bc	$41.94{\pm}2.7^{ij}$	
	Ummulong	Cold wet	Potato	28.92±7.5 b	14.30±4.1 ^{ab}	
East Garo Hills (EGH)	Songsak	Mild moist	Potato	25.66±2.1 ab	$39.28{\pm}4.7^{hi}$	
	Rongrenggre	Hot moist	Cauliflower	41.49±7.1 de	18.48±6.3 ^{abcd}	
West Garo Hills (WKH)	Garobadha	Host moist	No crop	21.0±2.2 a	23.37±4.1 ^{cdef}	
	Tura	Hot moist	Potato	37.40±1.9 cd	21.30±4.0 abcde	
	Sangsanggre	Hot moist	Potato	53.94±8.3 fgh	16.32±4.1 ^{abc}	
	Rongram	Host moist	No crop	29.93±1.7 b	20.86±3.4abcd	
Mean				52.70	30.90	
CD (P=0.05)				6.55	6.50	
SEm				2.30	2.29	

Disease assessment: For disease assessment, in each surveyed field, a quadrat of 1 m^2 was used to sample plants along an X-shaped transect. The quadrat was thrown three times on sampling points that were at least 10 m apart, with the three throws representing three replications/field. Following each throw, the number of healthy and black rot-infected leaves of 10 plants within the area was recorded. Black rot incidence was rated as the mean percentage of diseased plant leaves within the quadrat.

Severity was rated on leaves from the same 10 representative plants in each quadrat, disease severity was recorded according to a scale of 0–9 given by Williams (1985): 0=No visible attack; 1=Minute necrotic zones, 1-3 mm in diameter, at the hydathodes, or light brown leaf margins; 2=Marginal chlorotic, necrotic lesions, 0.5-3.0 cm in diameter, often at hydathodes; 3=Small to medium V-shaped lesions with distinct marginal chlorosis and blackened veins within the lesion, 1-5 cm in diameter; 4=Medium V-shaped lesions extending to the midrib with distinct marginal chlorosis and blackened veins within the lesion, more than 5 cm in diameter; 5=Large V-shaped lesions coalescing and expanding beyond the midrib, leaves appear scorched with coalescing lesions; 6=Many, 50-75%, of wrapper leaves exhibit symptoms, and a few are necrotic; 7=Almost all, 75-100%, of wrapper leaves exhibit symptoms, many wrapper leaves are necrotic; 8=All wrapper leaves exhibit symptoms, many are necrotic; 9=All wrapper leaves are necrotic. Disease severity scores were converted into percentage severity index (PSI) for the analysis.

$$DI (\%) = \frac{\text{Number of infected plants}}{\text{Total number of plants assessed}} \times 100$$
$$PDI (\%) = \frac{\text{Total sum of numerical rating}}{\text{Total no. of leaves observed } \times 100}$$
$$\text{Maximum category value}$$

where DI, Disease incidence; PDI, Per cent disease incidence.

Diverse meteorological variables such as rainfall, temperature, and relative humidity, were obtained from Meteorological Centre, Shillong (India Meteorological Department) and ICAR Research Complex for North Eastern Hill Region, Barapani, Meghalaya. Data were also collected on, evaluation of agronomic factors including the prior crop history, field dimensions, cropping patterns, cultivation systems, altitude, planting date, weeding practices, fertilizer application, soil composition, and the developmental stage of the plants, which were later used to determine their relationship and establish their correlation among these factors.

Statistical analysis: Aggregation of independent variables was used to analyse the survey of black rot of crucifers in 2021–22 with numbers of corresponding fields for each class (Table 1). The collected data were subjected to analysis using one-way analysis of variance (ANOVA),

followed by the implementation of Duncan's multiple range test (DMRT) at a significance level of 0.05 using SPSS software. This was performed to facilitate pairwise comparisons between the means of DI and PDI across 6 districts within the geographical expanse of Meghalaya, as well as for all 6 cruciferous crops within the EKH district. Two-way repeated measure ANOVA was performed to study the PDI in different villages in EKH across the season.

The correlation and multiple regression analysis was performed to assess the relationships between PDI of cabbage and DI and prevailing weather parameters in EKH using R software version 4.3.

RESULTS AND DISCUSSION

Assessment of disease occurrences: During the survey, black rot was observed to be pervasive in all cruciferous crops across the surveyed area of Meghalaya, with notable variations in disease incidence (DI) and severity. The disease was found to be prevalent at different levels in all surveyed cabbage fields in the 6 districts of the state (Table 1), consistent with the findings of Singh et al. (2016). The disease incidence of black rot of cabbage ranged from 21-91%, with the lowest incidence in WGH and EGH districts and the highest in EKH district. Disease severity observed during the present study ranged from 13.3–80.3% and closely followed the pattern of DI. The highest severity of 80.3% was observed in Pepbah village of EKH district, while the lowest (13.3%) was in Umsning village of Ri Bhoi district. Interestingly, among the six districts, the highest mean disease severity was recorded in EKH.

Influence of cropping pattern: Cropping pattern significantly influenced disease incidence (Table 1). Notably, cabbage fields with crucifers as a preceding crop exhibited the highest mean DI and severity. Conversely, fields with either fresh planting (no preceding crop) or only non-crucifers in the previous crop cycle suffered lesser incidence of black rot. The higher DI and severity observed in cabbage fields with crucifers as preceding crops may be attributed to the accumulation of pathogen inoculum over time due to continuous cultivation of susceptible host crops, leading to higher DI and disease severity (Laala *et al.* 2021).

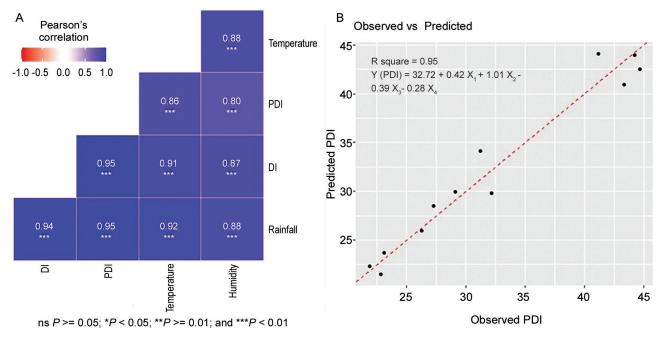
Seasonal assessment of black rot of cabbage: Seasonal assessment of black rot of cabbage revealed varying weather conditions and disease metrics across different seasons. In the spring season, villages within the EKH district experienced average rainfall of 5.2 mm, temperature of 17.9°C, and humidity level of 74.5%, corresponding to DI and PDI of 45.51 and 26.37%, respectively. During the monsoon season, EKH experienced increased rainfall (12.86 mm), higher temperatures (21.24°C), and elevated relative humidity (89.2%), resulting in notably higher DI and PDI of 69.97% and 43.37%, respectively. The autumn season saw lower rainfall, temperature, and humidity, leading to lower DI and PDI of 59.91% and 31.05%, respectively. The winter season had the lowest rainfall, temperature, and humidity, resulting in the lowest disease metrics of DI at 39.24% and PDI at 22.85%. These variations highlight the

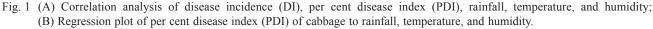
monsoon season's conducive nature for cabbage crop growth and development, as well as for the pathogen responsible for causing black rot of crucifers (Meena *et al.* 2021). Bacterial pathogens thrive in warm and humid climates (Vicente and Holub 2013) and consequently, black rot tends to be most serious in regions characterized by tropical, subtropical, and humid continental climates. Bila *et al.* (2009) reported that under wet and rainy weather conditions the occurrence of black rot can escalate significantly to 69.0%.

Association of disease and weather parameters: The present study observed significant positive correlations between DI and PDI with each of the analysed weather parameters in EKH (Fig. 1A). DI was significantly correlated with temperature (0.91), humidity (0.87), and rainfall (0.88). Multiple regression based on DI and weather parameters showed a prediction accuracy of PDI of 95% (Fig. 1B). These findings underscore the pivotal role of temperature, humidity, and rainfall in cabbage disease development in Meghalaya. Considering all weather variables together, the PDI showed a multiple regression predictive accuracy of 95%. A previous study by Jarial and Shyam (2004) highlighted the substantial impact of meteorological factors on disease dynamics, supporting the utility of these findings for disease forecast and effective management strategies. The results also support the hypothesis of the polycyclic nature of the pathogen and its inoculum-dependent development, as the progression rate of epidemics and spatial spread positively correlated with the strength of the source (Kocks et al. 1999). High rainfall and humidity during specific times of the year were found to contribute to longer Xcc survival periods in the phyllosphere of some species (Silva et al. 2017).

Interaction effect of DI and PDI in villages: The interaction effect between disease incidence and per cent

disease index (PDI) across various villages and seasons elucidates the intricate dynamics of occurrence of black rot in cabbage (Fig. 2) (Jensen et al. 2005). During the monsoon season, villages like Pepbah (DI, 91%; PDI, 80.33%) and Smith (DI, 85.33%; PDI, 52.67%) exhibit notably high disease incidence rates, recording percentages as high as 91% and 85.33%, respectively (Table 2). This heightened disease pressure is further corroborated by the PDI data, where these villages also showcase elevated mean values. Conversely, villages such as Laitkor (DI, 49.06%; PDI, 19.85%) and Mawlot (DI, 48.45%; PDI, 29.72%) demonstrate relatively lower disease incidence rates and PDI during the monsoon period, indicating a comparatively lower susceptibility to diseases under these climatic conditions. As the season transitions to autumn, there are discernible fluctuations in disease incidence and PDI across different villages. While certain villages experience a decline in both disease incidence and PDI, others witness an increase. For instance, villages like Nangkrem (DI, 48.23%; PDI, 22.12%) and Ladmawreng (DI, 63%; PDI, 25.13%) display a reduction in disease severity metrics during autumn compared to the monsoon season, suggesting a potential alleviation of disease pressure. In contrast, villages such as Mawlong (DI, 61.67%; PDI, 46.40%) and Umphyrani (DI, 58.33%; PDI, 39.83%) show an uptick in disease incidence and PDI during autumn, indicating heightened disease susceptibility under changing environmental conditions. Throughout spring and winter, similar patterns persist, with some villages maintaining consistent disease levels while others exhibit fluctuations. Villages like Mawngap (DI, 78.87%; PDI, 49.76%) and Myllium (DI, 61.62%; PDI, 43.31%) demonstrate relatively stable disease incidence and PDI across seasons, indicative of a consistent disease burden. Conversely, villages like Smith (DI, 29.33%; PDI, 15.42%) and Pepbah (DI, 44.33%; PDI,





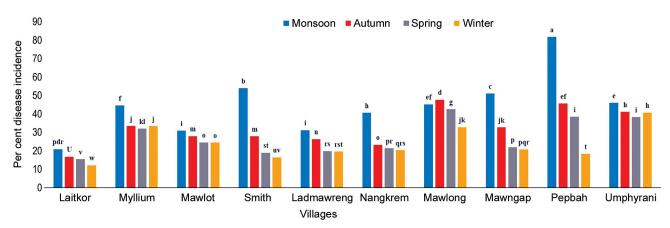


Fig. 2 Interaction effect of per cent disease index of black rot of cabbage in East Khasi Hills across different seasons and villages. Note: Different letters above bars indicate statistically significant difference at *P*>0.05 in Two-way ANOVA.

 Table 2
 Survey and estimation of disease incidence and per cent disease index of black rot of cabbage in different season in East Khasi

 Hills during 2022–23

Village	Monsoon		Spring		Autumn		Winter	
	Disease incidence (DI)%	Per cent disease index (PDI)						
Laitkor	49.10±7.8 ^a	19.85±2.0 ^a	32.00±2.6 ^a	14.56±3.0 ^a	36.31±3.2 ^a	15.70±4.0 ^a	30.66±1.2 ^a	11.14±0.1 ^a
Myllium	$61.60{\pm}3.0^{b}$	43.31±3.2°	$39.83{\pm}1.6^{a}$	30.96 ± 2.1^{b}	41.34 ± 2.3^{b}	32.21 ± 3.7^{b}	$38.0{\pm}3.0^{b}$	32.31 ± 5.5^{d}
Mawlot	48.50±3.2ª	29.71±6.5 ^b	31.46±3.1 ^a	23.55 ± 1.2^{b}	$34.68{\pm}2.4^{a}$	26.71 ± 2.2^{b}	29.46±0.7 ^a	23.44±4.3°
Smith	85.30±3.5 ^e	52.66±1.5 ^e	33.37±3.0 ^a	18.02±1.2 ^{ab}	$93.66{\pm}2.1^{f}$	26.66±1.5 ^{bc}	29.33±1.2 ^a	15.42±1.1 ^{ab}
Ladmawreng	66.90±1.7 ^b	$30.00{\pm}3.0^{b}$	31.65±2.9 ^a	18.92±7.5 ^{ab}	$63.0{\pm}1.0^{d}$	25.13±3.5 ^{bc}	27.00±2.6 ^a	18.55±6.5 ^{bc}
Nangkrem	67.90±2.5 ^{bc}	39.38±3.9°	41.66±1.5 ^b	$20.45{\pm}0.5^{ab}$	48.23±2.8°	22.12±2.5°	36.86 ± 2.8^{b}	19.25±2.6bc
Mawlong	73.50±3.0 ^{cd}	44.00±2.6 ^{cd}	56.16±2.9°	41.49 ± 7.1^{d}	61.66 ± 1.5^{d}	46.40±2.3°	$49.33{\pm}1.5^d$	31.71 ± 1.7^{d}
Mawngap	$78.70{\pm}3.6^{d}$	49.76±4.7 ^{de}	$64.46{\pm}3.8^{d}$	21.02±2.2 ^{ab}	78.86±4.3 ^e	$31.42{\pm}1.3^{d}$	62.46±2.3e	19.74±2.1bc
Pepbah	$91.00{\pm}1.0^{f}$	$80.33{\pm}1.5^{f}$	71.33±6.7 ^e	37.40±1.9 ^{cd}	82.99±1.7 ^e	44.29±3.6 ^{de}	44.33±3.5°	17.34±1 ^{abc}
Umphyrani	77.30±2.8 ^d	44.73±2.8 ^{cd}	53.13±5.9°	37.27±2.5 ^{cd}	$58.33{\pm}5.9^{d}$	39.82±6.5 ^e	44.95±4.9 ^{cd}	39.59±4.3e
Mean	70.00	43.40	45.5	26.4	59.9	31.1	39.2	22.9
CD (P=0.05)	6.41	6.22	6.68	4.25	5.59	4.65	4.63	6.35
SEm	2.158	2.0	2.24	1.43	1.88	1.56	1.56	2.10

17.34%) continue to grapple with elevated disease levels even during these seasons, highlighting ongoing challenges in disease management efforts (Burlakoti *et al.* 2018).

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Disease assessment in different crucifers: The assessment of disease incidence (DI) and disease severity across various cruciferous crops revealed notable variations in susceptibility to black rot. Among the surveyed crops, cabbage exhibited the highest DI and severity, with values of 69.98% and 43.38%, respectively (Fig. 3). Following cabbage, cauliflower demonstrated a significant DI of 51.02% and a severity of 26.93%. Knol khol also exhibited considerable susceptibility, with a DI of 62.59% and a severity of 15.23%. Radish displayed a DI of 36.85% and a severity of 15.61%, while broccoli showed a DI of 34.34% and a severity of 14.02% (Fig. 3). Conversely, mustard exhibited the lowest DI and severity among the surveyed crops, with values of 26.81% and 13.72%, respectively. These findings underscore the varying levels

of vulnerability among cruciferous crops to black rot, with cabbage being the most susceptible and mustard being the least affected. These findings are consistent with those of Dhar and Singh (2014).

In our comprehensive survey in Meghalaya and specifically the EKH district, we encountered the strong presence of black rot across various cruciferous crops. While the incidence and severity of this disease may fluctuate across the seasons and villages, it is evident that all cruciferous crops are facing a significant challenge due to the prevalence of black rot. The continuous cultivation of cabbage has led to the inoculum build up which is the major factor driving DI. However, our analysis indicates that disease severity is primarily dependant on weather parameters. As climate change reshapes regional climate patterns and disrupts traditional cropping practices, it also has a profound influence on disease epidemics. Our study unequivocally demonstrates that temperature, rainfall, and

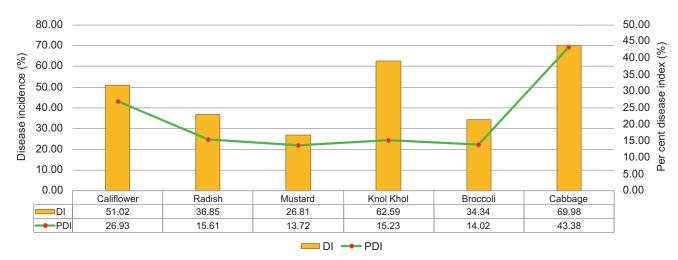


Fig. 3 Per cent disease index (PDI) and disease incidence (DI) of black rot of cruciferous vegetables in the East Khasi Hills district of Meghalaya during 2022–23.

relative humidity exert a robust and consequential impact on disease severity, underlining the pressing need for climateinformed disease management strategies in cruciferous crop cultivation.

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