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Impact of sowing dates and different weather parameters on the occurrence of leaf spot diseases of groundnut

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Groundnut (*Arachis hypogaea* L.) is a vital legume crop whose productivity is significantly hampered by fungal diseases, particularly in tropical regions. This study aimed to identify, characterize and correlate the different weather parameters affecting groundnut, assessing diseases severity and incidence across four different sowing dates (D_1–D_4) during the 2023 *Kharif* season in Nagaland. The predominant fungal pathogens were isolated and morphologically identified as *Cercospora arachidicola* and *Cercosporidium personatum*. Disease incidence and severity varied significantly with sowing time, with late sowings (D_3, D_4) exhibiting increased prevalence of late leaf spot, likely due to favourable microclimatic conditions. Disease severity was monitored weekly, revealing that late-season sowings coincide with peak pathogen activity, exacerbating oxidative stress and downregulating host defence genes. The correlation matrix of disease severity (PDI) tends to increase with higher maximum relative humidity (RH_{max}) but decreases with higher maximum temperatures (T_{max}), higher minimum temperatures (T_{min}), higher minimum relative humidity (RH_{min}), total rainfall, and an increased number of rainy days and vice versa. Disease severity also fluctuates with the change in weather parameters. The findings emphasize the role of temporal sowing patterns in disease epidemiology and the importance of integrated management strategies. This research provides foundational data for selecting resistant varieties, optimizing sowing schedules, thereby enhancing groundnut productivity.

Keywords : Disease incidence, disease severity, groundnut, Tikka leaf spot

INTRODUCTION

Groundnut (*Arachis hypogaea* L.), is a vital leguminous crop cultivated worldwide for its edible seeds, which are rich in protein, oil and essential nutrients (Biswas *et al.* 2024). It has a major impact on food security, nutrition as well as economic sustainability, particularly in developing countries where it serves as a staple food and cash crop (Khan *et al.* 2021). Groundnut production is limited by a number of biotic and abiotic conditions, despite its significance in agriculture, with fungal pathogens being among the most devastating. Fungal diseases cause substantial yield losses, reduce seed quality and may lead to the accumulation of mycotoxins, posing serious health risks to humans and livestock (Pal *et al.* 2014).

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According to Joshi *et al.* (2020), groundnut is highly vulnerable to various plant pathogens such as bacteria, fungi, viruses, and nematodes, which can significantly reduce both yield and quality. Among the various diseases affecting groundnuts, fungal infections are especially damaging. These include early leaf spot (*Cercospora arachidicola*) and late leaf spot (*Cercosporidium personatum*). These pathogens reduce photosynthetic efficiency, weaken plant vigor and in severe cases, cause complete crop failure.

Understanding the distribution of fungal species associated with groundnut is crucial to develop effective disease management techniques. Cultural practices, resistant varieties, biological control agents, and chemical fungicides are commonly employed to mitigate fungal infections (Ons *et al.* 2020). However, the development of strains resistant to fungicides and environmental issues over chemical use necessitate sustainable alternatives, such as biocontrol agents and host plant resistance (Zade *et al.*2018).

MATERIALS AND METHODS

Field trial was performed at the experimental field, Department of Plant Pathology, School of Agricultural Sciences (SAS), Nagaland University, Medziphema, under Dimapur district, during Kharif season of 2023 and the laboratory works was carried out in the laboratory of the Department of Plant Pathology, SAS, N.U, Medziphema, located in the foothills of Nagaland at an elevation of 310 meters above sea level, at 25° 45' 43" North latitude and 93° 53' 04" East longitude. The region is characterized by a hyper-arid climate, sandy loam soil, and erratic rainfall. A randomized block design (RBD) was implemented in the experiment with four different dates of sowing *viz.* D_1 (27^{th} June), D_2 (20^{th}July), D_3 (12^{th} August), and D_4 (4^{th} September), each replicated four times. Standard agronomic practices were followed uniformly across treatments. Diseased plant parts showing typical symptoms were identified by referring to various literatures of previous researchers and collected during different crop growth stages. Samples were surface sterilized with 1% sodium hypochlorite, then rinsed in sterile distilled water. Small slices from lesion margins were deposited on Potato Dextrose Agar (PDA) medium in Petri dishes and incubated at 25 ± 2°C.

Emerging fungal colonies were sub-cultured to produce pure cultures. Colony morphology (colour, texture, growth rate) and microscopic features (shape, size, septation of conidia and conidiophores) were studied using compound microscopy. Identification was based on standard taxonomic keys and literature. Disease incidence (DI) and percent disease index (PDI) were calculated using formula according to Turaki *et al.* (2014) and Wheeler, (1969) respectively. Data were recorded at regular intervals and analyzed to determine the progression of diseases.

$$\text{Disease incidence (\%)} = \frac{\text{No. of infected plants}}{\text{Total number of plant assessed}} \times 100$$

$$\text{PDI (\%)} = \frac{\text{Sum of individual ratings}}{\text{Total number of plant observed} \times \text{maximum rating}} \times 100$$

Statistical Analysis

Analysis of variance (ANOVA) was performed with SPSS software to test the significance of differences among treatments as described by Gomez and Gomez (1984).

RESULTS AND DISCUSSION

Symptomatology and morphological characterization of leaf spot disease and the pathogens

In the field it was observed that all foliar parts of the plants were affected by early and late leaf spot diseases, with symptoms being most severe on the leaves. Initial symptoms appeared around one month after planting and persisted until plant maturity. Among the two leaf spot diseases, late leaf spot was more prevalent and caused greater damage.

Under *in vitro* conditions, the fungal pathogens were isolated and identified from infected groundnut (*Arachis hypogaea* L.) plants: early leaf spot (*Cercospora arachidicola*), late leaf spot (*Cercosporidium personatum*). The significant amount of differences among the different isolates was studied with respect to morphological characters like colony colour, mycelial characteristics and shape of spores per plate were recorded. Their symptomatology, morphological traits and pathogenicity confirmation as given below.

Early leaf spot (*C. arachidicola*)

Symptoms appeared 30–37 days after sowing (DAS) (Fig.1A, B). Lesions were sub-circular (1–10 mm diameter), brown to reddish-brown on the adaxial surface with a raised tuft, and light brown on the abaxial surface, surrounded by a bright yellow halo. Lesions coalesced, leading to extensive foliar damage.

Late leaf spot (C. personatum)

Symptoms emerged 58–74 DAS as smaller (1–6 mm), dark brown to black lesions with a rough, tufted abaxial surface and minimal halo. Defoliation was severe in advanced stages (Fig.1 C, D).

Table 1: Disease incidence of groundnut caused by tikka leaf spot at different date of sowing during July to December 2023

Date of sowing	*Disease incidence (%)*												*MEAN*
	30 DAS	*37 DAS*	*44 DAS*	*51 DAS*	*58 DAS*	*65 DAS*	*72 DAS*	*79 DAS*	*86 DAS*	*93 DAS*	*100 DAS*	*107 DAS*	
D_1: 27th June 2023	12.50	15.00	20.00	32.50	47.50	52.50	65.00	67.50	67.50	70.00	77.50	77.50	50.42
	(20.70)	(22.79)	(26.57)	(34.76)	(43.57)	(46.43)	(53.73)	(55.24)	(55.24)	(56.79)	(61.68)	(61.68)	
D_2:20th July 2023	12.50	17.50	25.00	27.50	40.00	65.00	85.00	100.00	100.00	100.00	100.00	100.00	64.38
	(20.70)	(24.73)	(30.00)	(31.63)	(39.23)	(53.73)	(67.21)	(90.00)	(90.00)	(90.00)	(90.00)	(90.00)	
D_3:12th August 2023	17.50	22.50	35.00	52.50	77.50	97.50	97.50	97.50	97.50	97.50	97.50	97.50	73.95
	(24.73)	(28.32)	(36.27)	(46.43)	(61.68)	(80.90)	(80.90)	(80.90)	(80.90)	(80.90)	(80.90)	(80.90)	
D_4:4th September 2023	0.00	10.00	30.00	70.00	87.50	100.00	100.00	100.00	100.00	100.00	100.00	100.00	74.79
	(0.00)	(18.43)	(33.21)	(56.79)	(69.30)	(90.00)	(90.00)	(90.00)	(90.00)	(90.00)	(90.00)	(90.00)	
SEm±	*2.92*	*2.50*	*2.04*	*2.67*	*3.36*	*5.07*	*4.10*	*3.00*	*5.83*	*6.68*	*4.49*	*4.49*	-
CD (P=0.05)	*9.33*	*8.00*	*6.53*	*8.54*	*10.75*	*16.22*	*13.13*	*9.61*	*18.66*	*21.37*	*14.36*	*14.36*	-

Note : The table shows mean values, with figures in parentheses representing angular transformed values

Table 2 : **P**ercent disease index (PDI) of groundnut caused by tikka leaf spot at different date of sowing during July to December 2023

Date of sowing	*Percentage Disease Index (%)*												*MEAN*
	30 DAS	*37 DAS*	*44 DAS*	*51 DAS*	*58 DAS*	*65 DAS*	*72 DAS*	*79 DAS*	*86 DAS*	*93 DAS*	*100 DAS*	*107 DAS*	
D_1:27th June 2023	1.39	1.66	4.43	6.94	8.60	16.93	22.22	28.05	33.05	41.66	46.39	52.50	21.99
	(6.76)	(7.40)	(12.15)	(15.27)	(17.05)	(24.30)	(28.12)	(31.98)	(35.09)	(40.20)	(42.93)	(46.43)	
D_2:20th July 2023	1.39	1.94	3.33	4.71	7.21	12.22	16.66	25.55	33.88	42.50	58.33	61.67	22.45
	(6.76)	(8.01)	(10.51)	(12.54)	(15.58)	(20.46)	(24.09)	(30.36)	(35.60)	(40.68)	(49.80)	(51.75)	
D_3:12th August 2023	1.94	2.50	3.89	5.83	9.72	16.39	25.27	33.61	49.72	69.72	90.27	97.50	33.86
	(8.01)	(9.09)	(11.37)	(13.97)	(18.16)	(23.88)	(30.18)	(35.43)	(44.84)	(56.61)	(71.83)	(80.90)	
D_4:4th September 2023	0.00	1.11	3.33	7.77	10.83	21.11	32.78	55.00	60.00	77.77	100.00	100.00	39.14
	(0.00)	(6.05)	(10.51)	(16.19)	(19.21)	(27.35)	(34.92)	(47.87)	(50.77)	(61.87)	(90.00)	(90.00)	
Sem ±	*0.21*	*0.24*	*0.29*	*0.47*	*0.46*	*1.11*	*1.37*	*2.30*	*1.82*	*1.88*	*3.90*	*2.49*	-
CD (P=0.05)	*0.68*	*0.78*	*NS*	*1.50*	*1.47*	*3.56*	*4.38*	*7.37*	*5.83*	*6.02*	*12.47*	*7.95*	-

Note: The table presents mean values,with values in parentheses as angular transformed data

Table 3 : Correlation coefficient (r) on percent disease index (PDI) of groundnut caused by tikka leaf spot at different dates of sowing with weather parameters during July to December 2023

	Temperature (^{0}C)		*Relative humidity (%)*			
Pearson's correlation coefficient	*Maximum*	*Minimum*	*Maximum*	*Minimum*	*Rainfall (mm)*	*No. of rainy days*
D_1: 27th June 2023	-0.41	-0.83**	0.12	-0.32	-0.65*	-0.54
D_2: 20th July 2023	-0.81**	-0.98**	0.41	-0.69*	-0.64*	-0.76**
D_3: 12th August 2023	-0.93**	-0.95**	0.74**	-0.74**	-0.43	-0.49
D_4: 4th September 2023	-0.97**	-0.89**	0.86**	-0.29	0.15	-0.04

* = Significant at the 5% significance level, ** = Significant at the 1% significance level

Table 4 : Multiple Regression Equation (MRE) between PDI (tikka leaf spot) and Weather Parameters

	Regression Equation	R^2
D_1: 27th June 2023	$Y = 288.46 + 4.89X_1 - 28.50X_2 + 3.53X_3 - 0.95X_4 - 0.36X_5 + 12.75X_6$	**0.96**
D_2: 20th July 2023	$Y = 578.50 – 4.45X_1 – 5.66X_2 - 2.97X_3 – 0.07X_4 – 0.17X_5 + 2.19X_6$	**0.91**
D_3: 12th August 2023	$Y = 651.21 - 16.08X_1 + 4.16X_2 + 0.53X_3 – 3.52X_4 + 0.19X_5 + 9.50X_6$	**0.99**
D_4: 4th September 2023	$Y = 885.76 – 8.46X_1 – 5.82X_2 – 5.58X_3 + 0.32X_4 + 0.59X_5 + 0.848X_6$	**0.98**

Note: Y = Disease severity (PDI); $X_1 = T_{max}$, $X_2 = T_{min}$, $X_3 = RH_{max}$, $X_4 = RH_{min}$, $X_5 = R_{total}$ & $X_6 = R_{no}$.

These findings align with reports by Dutta *et al.* (2021) and Kukadiya *et al.* (2024) who noted that late leaf spots were dark brown to black specks without a yellow halo but had a rough, tufted appearance, while early leaf spots were brown to reddish brown in colour and always had a yellow halo. Additionally, lesions may develop on petioles, stems, stipules. They also observed that early leaf spot as subcircular-irregular, 1-10 mm diameter, whereas late leaf spot is diagnosed as smaller, 1-6 mm diameter.

Study of morphological characteristics of *C. arachidicola* on PDA revealed that, colonies transitioned from white to dark brown with raised centers. Conidiophores (28.2–45 × 3–6 µm) were pale olivaceous, geniculate, and bore obclavate, 3–10-septate conidia (33.92–90.22 × 3.5–5.9 µm)(Fig. 2 A, B). Whereas, in case of *C. personatum,* colonies turned grayish-black with fluffy mycelia. Conidiophores (26.31–41.71 × 4.5–6 µm) produced hyaline to light brown, cylindrical conidia (14.1–70.12 × 5.1–7.0 µm) with 1–4 septa (Fig.2C, D).

The findings of the current investigation were in congruent with the descriptions provided by previous researchers (Surendra *et al.* 2015; Kumar *et al.* 2019; Thanh *et al.*2022; Udhayakumar *et al.* 2022) and thus, early and late leaf spot fungi were identified as *C. arachidicola* and *C. personatum* respectively.

The study demonstrates that traditional morphological characterization remains a reliable and effective approach for identifying major fungal pathogens in groundnut, as evidenced by the successful differentiation of *Cercospora arachidicola*, *Cercosporidium personatum,* based on distinct colony features, sporulation patterns, and microscopic structures.

Occurrence of Tikka leaf spot diseases of groundnut

For Tikka leaf spot, early leaf spot (ELS) appeared 30–37 days after sowing (DAS), while late leaf spot (LLS) occurred 58–74 DAS, with peak

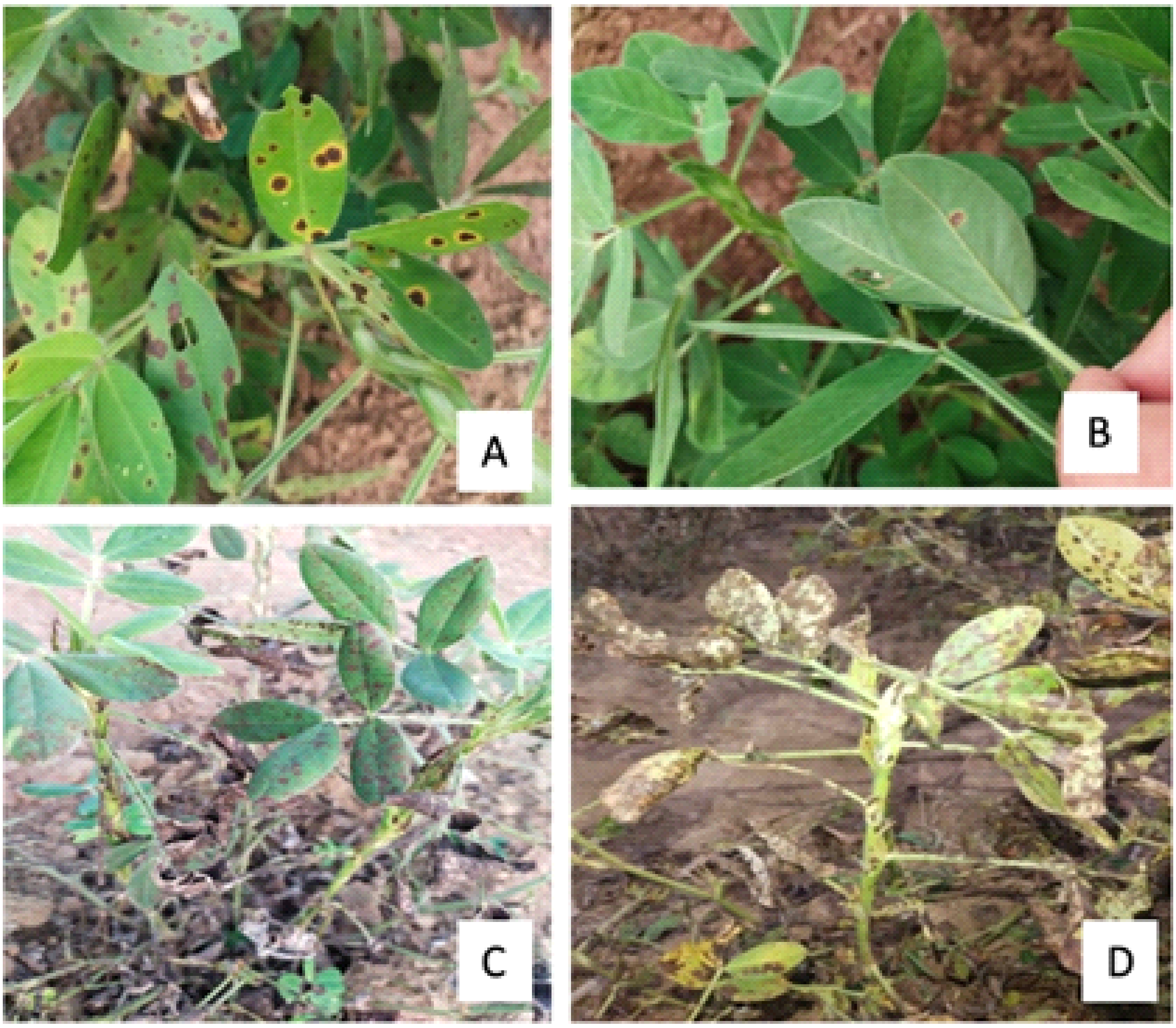

Fig.1: Symptoms associated with Early and Late Leaf spot of Groundnut.**A:** Early leaf spot on adaxial side; **B:** Early leaf spot on abaxial side **C.** Late leaf spot on adaxial side; **D.** Late leaf spot on abaxial side

disease incidence (100%) observed in D_2 (79 DAS) and D_4 (100 DAS) (Table 1). Disease severity was lowest in D_1 (52.50%) and D_2 (61.67%) at 107 DAS, while maximum severity (100%) occurred in D_4 at 100 DAS during the 2023 *Kharif* season (Table 2). These findings align with previous studies, including Aslam & Rafique (2018) who reported tikka leaf spot emergence at 30–35 DAS, primarily during August–October (pod formation stage), with disease incidence ranging from 9.35–22.48%. Similar trends were observed by Kumar *et al*. (2019), noting peak incidence (70%) and severity (48%) in Gandhinagar village, followed by Birgua (65%, 46%), while Tejpura recorded the lowest (22%, 12%). Additionally, Bwala *et al.* (2022) documented ELS and LLS occurrence at 44–51 DAS and 59–66 DAS, respectively, further supporting the observed disease progression patterns.

Epidemiology

The analysed correlation data as depicted in (Table 3), revealed that there was a highly significant negative correlation with T_{max} in D_2 (-0.81), D_3 (-0.93), D_4 (-0.97) and T_{min} D_1 (-0.83), D_2 (-0.98), D_3 (-0.95), D_4 (-0.89) were highly significant but negatively correlated. RH_{max} in D_3 (0.74), D_4 (0.89) was seen to be significant and positively correlated, while RH_{min} in D_2 (-0.69), D_3 (-0.74) showed significant and highly significant

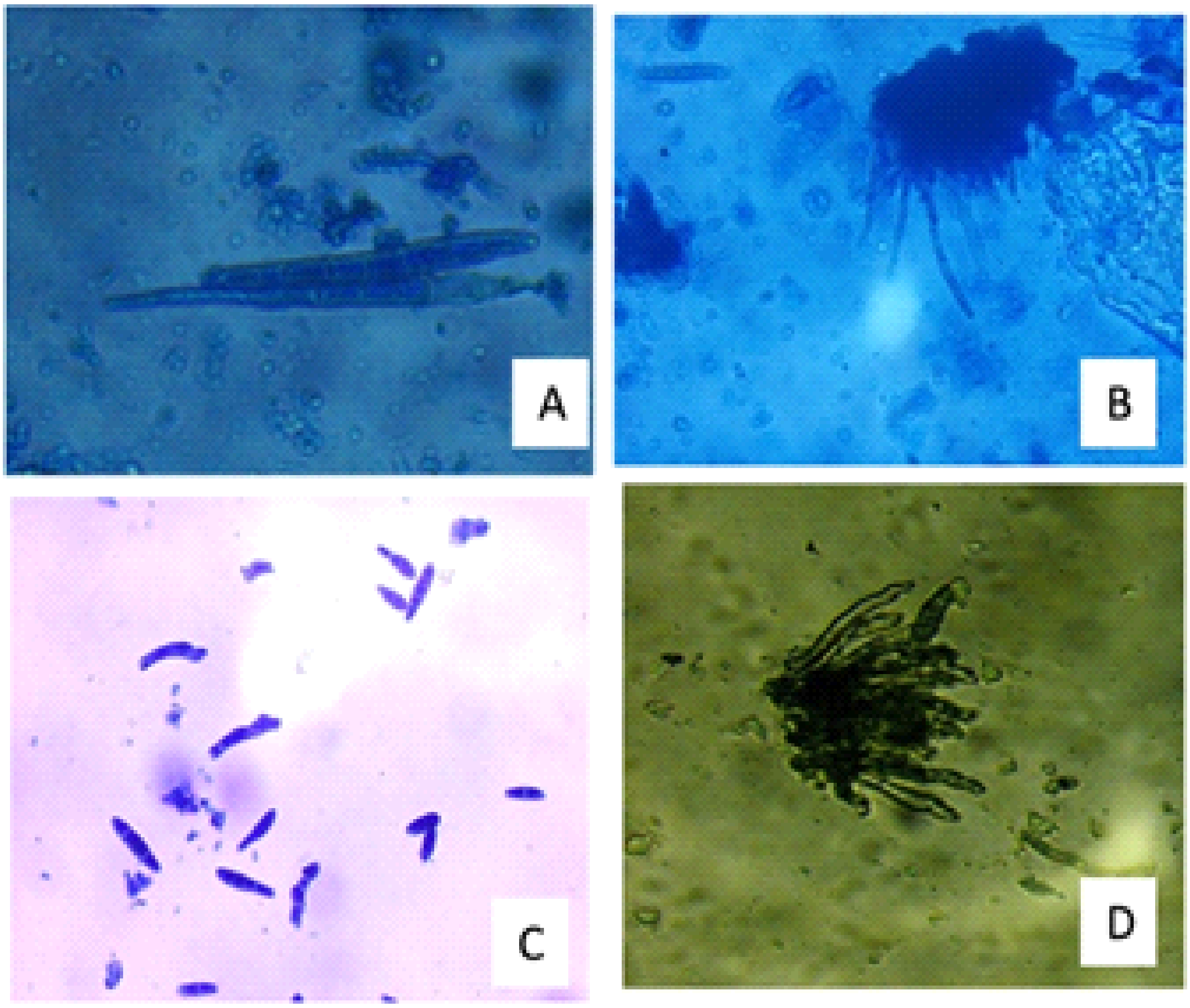

Fig 2: Morphological characterization of *C. arachidicola*and *C. personatum*. **A**. *C. arachidicola* Conidia (40X); **B.** *C arachidicola* conidiophore (40X); **C.** *C. personatum* conidia (40X); **D.** *C personatum* conidiophore (40X)

negative correlation. R_{total} in D_1 (-0.65), D_2 (-0.64) were seen significant and negatively correlated. R_{no} in D_2 (-0.76) was observed to be highly significant and negatively correlated. Thus, this indicated that the disease severity increases with increase in RH_{max} but increases with decrease in T_{max}, T_{min}, RH_{min}, R_{total}, and R_{no}.

While the coefficient of determination (R²) between disease severity and the six sets of independent variables as shown in (Table 4), was 0.96, indicating that 96% of the variation in disease severity was attributable to these six weather factors in D_1, 0.91 suggesting 91% change in disease severity was caused by the six weather parameters in D_2, similarly 99% in D_3 and 98% in D_4.

The above results were in conformity with the findings of earlier researchers like Dey *et al.* (2017) who reported that weather conditions such as average maximum temperatures of 30.92°C, minimum temperatures of 22.31°C, morning relative humidity of 89.67%, evening relative humidity of 64.25%, well-distributed average rainfall of 50.33 mm, average of 2.67 rainy days, and an average wind velocity of 4.24 km/hr were found to favor the onset, development, and spread of late leaf spot disease in susceptible groundnut. The current findings were also in agreement with Vyavhare *et al*. (2020) who reported that development of tikka leaf spot was influenced by the T_{max} ranged from 26.30°C to 32°C, mean temperature ranged from 25.10°C to 27.50°C, maximum relative humidity ranged from 94% to

98%, sunshine duration varied between 0.2 to 7 hours per day, evaporation ranged from 0.5 to 3.6 mm per day, and crop age ranged from 1 to 126 days. Conversely, minimum temperature, minimum relative humidity, mean relative humidity, wind speed, and rainfall were negatively correlated with tikka leaf spot.

The present study revealed that early and late leaf spots were significantly influenced by sowing dates, with disease severity progressively increasing from the vegetative stage to crop maturity. Late sowings (D_3, D_4) showed higher incidence of late leaf spot due to prolonged leaf wetness and conducive microclimatic conditions. Also, weather factors played a significant role on the onset of disease development and understanding the disease-weather association helps in providing vital insights for developing sustainable, climate-resilient strategies to protect groundnut crops and improve productivity and food safety in disease-prone regions.

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DECLARATION

Conflict of Interest. Authors declare no conflict of interest.

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