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Assessment of maize grey leaf spot (*Cercospora zeae-maydis* Tehon and Daniels) in a main maize producing part of Western Oromia, Ethiopia

Debela Diro and Fikre Lemessa

Abstract

Grey leaf spot (GLS) is one of the fungal diseases caused by *Cercospora zeae-maydis*; it is a necrotrophic and polycyclic foliar disease of maize that poses a severe challenge in tropical maize production. This study was designed to determine the prevalence and intensity of maize grey leaf spot (GLS) and how maize varieties under production in the surveyed area reacted to GLS. A total of 81 farmer's fields in 9 districts from 3 zones were surveyed when the crop was in the flowering to grain filling stage. The global positioning system (GPS) was used to mark each sampling site using a GPS receiver for altitude and co-ordinates. Using meter tap, 9m² areas in each field were marked out in three randomly selected points in a diagonal form. Purposive multistage sampling method was used and data were analyzed using three-stage nested design. The survey's findings revealed that, although to various degrees, GLS was prevalent in each district. While the mean GLS incidence ranged from 15% in Ababo Guduru to 76.67% in Digga district, the mean percent severity index differed between 12.50% and 46.06% between these two districts. The findings suggested that it need to develop management strategies like crop rotation with non-host, balanced fertilizer utilization and use of tolerant varieties to minimize the negative impact of the disease on maize production

Keywords: *Cercospora zeae maydis*, Grey leaf spot, Incidence, Maize, Percentage severity index, Prevalence

Introduction

Maize (*Zea mays* L.) has been chosen as one of the primary national commodity crops to meet the food self-sufficiency program to feed the worlds, Africa's, and Ethiopia's rapidly growing populations. It is the most important crop in Ethiopia in terms of production and distribution. Thus, maize is second in area coverage to teff (*Eragrostis tef*) among all cereals, with 2.3 million hectares (17.7% of all cereals) land committed to the crop, but first in productivity (4.7 t ha⁻¹), with total annual production of 10.6 million tons (CSA, 2020) [4]. Despite maize's importance as a basic food security crop, Ethiopia's average yield (3.74 t ha⁻¹) remains low when compared to the global average (5.78t ha⁻¹) (FAO, 2020) [8]. As a result, the effects of abiotic and biotic factors, as well as insufficient deployment of varieties tolerant or resistant to these scenarios, account for a large amount of the yield gap.

Drought, heat, soil acidity, frost, and poor soil fertility, particularly in N and P, are some of the principal abiotic factors affecting maize productivity levels. And also, Ethiopian maize production is being hampered by biotic stresses like diseases such as Grey Leaf Spot (*Cercospora zeae-maydis*), Turicum Leaf Blight (*Exserohilum turcicum*), Common Leaf Rust (*Puccinia sorghi*), Maize Streak Virus (Mastre virus), Maize Lethal Necrosis, parasitic weeds (primarily *Striga hermonthica*), and insect pests (such as the maize stem borer, maize weevils (Keno *et al.*, 2018) [11].

Among fungal disease, GLS, caused by *C. zeae-maydis*, is a necrotrophic and polycyclic foliar disease of maize that poses a severe challenge in tropical maize production (Renfro and Ullstrup, 1976) [19]. This pathogen causes the plant to lose a lot of water, resulting in severe leaf blighting and impaired photosynthesis. Yield losses occur when a maize plant's ability to retain and produce carbohydrates (glucose) in the grain is damaged. When *C. zeae maydis* infects foliar tissue, the plant's capacity to photosynthesize and create byproducts of the process is reduced. This eventually results in undersized ears, reduced grain yield, and maize plant death (Stromberg, 2009) [24].

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Extreme blighting of the upper eight or nine leaves, which produce 75 to 90% of the photosynthates for grain fill, can cause stalk weakness or potentially infectious stalk rot diseases, which can result in premature stalk death and lodging (Ward *et al.*, 1999)^[28].

In 1997-1998, a study was undertaken in the western region of the nation on important maize foliar diseases (Wegary *et al.*, 2001)^[36]; then, possible threat of GLS to maize yield was first recognized through a survey conducted in 1997-1998 to determine the disease's distribution and importance in Ethiopia's maize-growing regions (Wegary *et al.*, 2001)^[36]. In the country, a big epidemic emerged in the early 2000s, causing significant maize grain yield losses. During the 2003/2004 cropping seasons in Bako, Wegary *et al.* (2004)^[31] found yield losses owing to GLS on resistant, moderately resistant, and susceptible varieties of 0-14.9%, 13.7-18.3%, and 20.8-36.9%, respectively. Likewise, study conducted in South Ethiopia from 2004 to 2006 revealed that GLS caused a 29.5% yield reduction (Tilahun *et al.*, 2012)^[27]. The disease has since been widely spread through severe outbreaks yearly, particularly in the country's warm and humid regions (Tilahun *et al.*, 2012; Negash, 2013)^[27, 16]. Western Oromia is one of Ethiopia's maize-producing provinces. Grey leaf spot was particularly severe in the country's warm, humid maize area. Furthermore, according to Nega *et al.*, (2016)^[15], the highest GLS prevalence, incidence, and severity were found in South and Southwest Ethiopia, with 74%, 71.2%, and 45.13%, respectively. In terms of reaction level to the disease, there is limited information about the varieties under production. As a result, the objectives of this work is to collect comprehensive and informative data on the level of grey leaf spot reaction in several commercial maize varieties now in cultivation, as well as disease epidemics in diverse agro ecologies and cropping practices. Thus, the present study was undertaken with the following objectives:

- To determine the prevalence and intensity of maize grey leaf spot in major growing area of western Oromia zones
- To determine how maize varieties in production in the surveyed area reacted to GLS.

Materials and Methods

Description of Survey Areas and Survey Method

During the 2019/20 growing season, maize grey leaf spot was observed in important maize-producing agro-ecologies in the Western section of Oromia, including certain districts in the Horro Guduru and West Shoa zones.

The studied area covers a Wollega, East Wollega, wide range of districts and agro-ecological zones and is located between 36°19'37" and 37°03'37" E East longitude and 8°28'7" N and 9°04'34" N latitude, with altitudes ranging from 1446 to 2418 meters above sea level. The prevalence, incidence, and severity of maize GLS were evaluated in these districts. Farmers fields were picked using a random sampling technique at 5-10 km intervals, whereas districts and kebeles were selected on purpose. A total of 81 farmer's fields were evaluated, with 9 districts out of the zones studied when the crop was in the flowering to grain filling period, which is when grey leaf spot development is at its peak (Latterell and Rossi, 1983)^[12]. Grey leaf spot status was recorded in respect to maize varieties in production. The global positioning system (GPS) was used to mark each sampling site using a GPS receiver for altitude and co-ordinates. Using meter tap, 9m² areas in each field were marked out in three randomly selected points in a diagonal form. Ten maize plant stands were randomly picked in the centre of each indicated area and evaluated for incidence and severity (Nwanosike *et al.*, 2015)^[17]. For disease assessment, each indicated area was regarded as a replicate.

Table 1: Description of the survey areas

Region	Zone	Districts	Coordinates		No. of PA Assessed/District	Total No. of Farmers Field Assessed/Zone	Altitude Range (m.a.s.l.)
			N	E			
		Digga	9°1'57"N-9°35'0"N	36°19'37"E-36°26'6"E	3	9	1446-2168
	East Wollega	Wayu Tuka	9°1'45"N-9°24'1"N	36°36'32"E-36°41'1"E	3	9	1886-1921
		Gudeya Billa	9°12'10"N-9°15'42"N	37°1'40"E-37°05'05"E	3	9	1800-2125
		Ababo Guduru	9°38'9"N-9°46'34"N	37°30'1"E-37°30'37"E	3	9	2289-2353
Oromia	Horro Guduru Wollega	Guduru	9°34'0"N-9°34'3"N	37°29'47"E-37°29'55"E	3	9	2300-2332
		Abay Choman	9°35'27"N-9°38'53"N	37°14'40"E-37°20'29"E	3	9	2356-2418
		Nono	8°28'7"N-8°32'5"N	37°24'58"E-37°26'13"E	3	9	1626-1871
	West Shoa	Jibat	8°39'12"N-8°42'15"N	37°26'48"E-37°27'17"E	3	9	2106-2361
		Liban Jawi	8°55'20"N-8°58'24"N	37°31'40"E-37°34'44"E	3	9	2267-2343
Total	3	9			27	81	

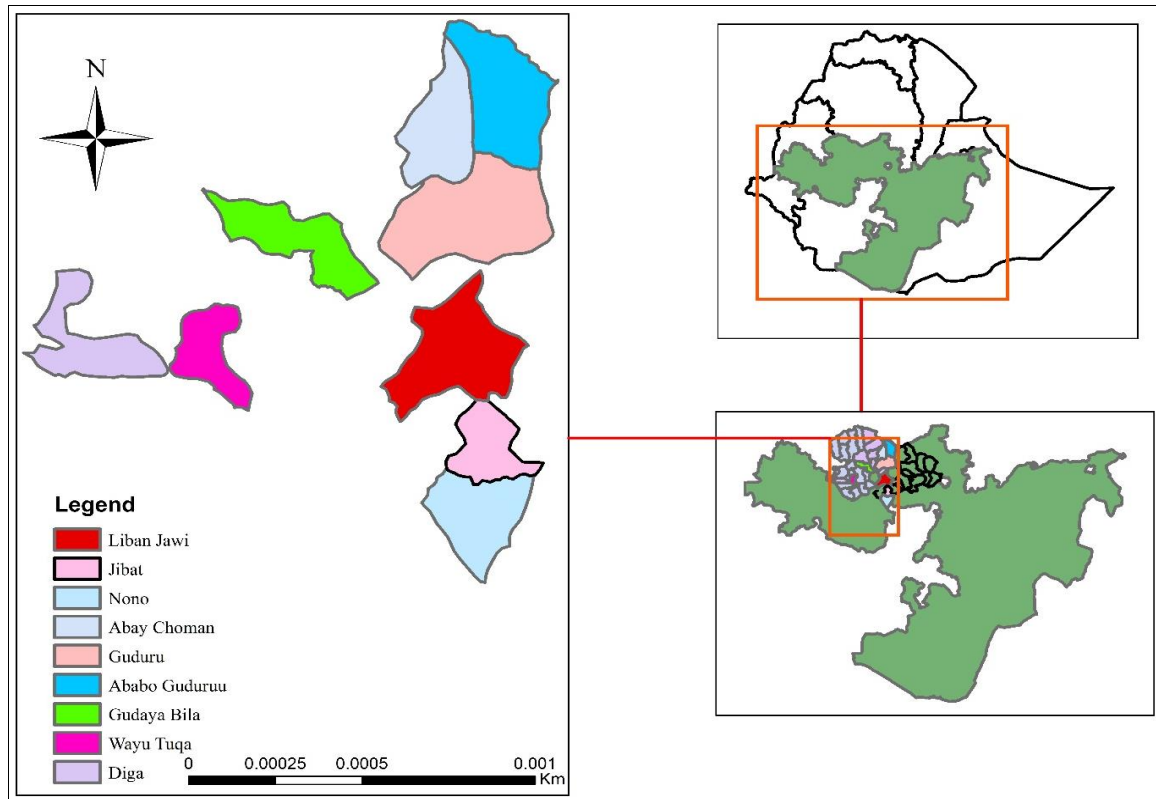


Fig 1: Map of the survey areas

Disease assessment

Disease incidence

Disease incidence was calculated as a percentage of infected maize plants out of total maize plants in the sample plot (Cooke *et al.*, 2006) [3] as follows:

$$\text{Incidence} = \frac{\text{No. of diseased plants}}{\text{Total no. of plants assessed}} \times 100$$

Disease severity

Data on disease severity was collected during the field survey by evaluating the field using 1-5 disease recording scale, where 1 = Very slight to slight infection, one or two to few Scattered lesions on lower leaves; 2 = moderate lesion development below the leaf subtending the ear; 3 = heavy lesion development on and below the leaf subtending the ear with a few leaves, 4=severe lesion development on all but the uppermost leaves, which may have a few lesions; and 5 = all leaves dead (Maroof *et al.*, 1993) [14].

Wheeler (1969) [33] suggested equation was used to convert the numerical rating to a percentage severity index (PSI).

$$\text{PSI}(\%) = \frac{\text{Sum of all numerical ratings}}{\text{Total No. of rated} \times \text{Max. disease score on scale}} \times 100$$

Disease prevalence

The disease's prevalence was calculated by dividing the number of fields infected by the total number of fields assessed and expressing the result as a percentage.

$$\text{Disease Prevalence} (\%) = \frac{\text{number of infected fields}}{\text{Total number of fields assessed}} \times 100$$

Data analysis

The survey data on incidence and severity index of GLS were arranged using three-stage nested design by nesting districts in Zone, peasant Association (PAs) within districts and farms within PAs with the following model to see their interaction.

$$Y_{ijk} = \mu + \beta_i + \tau_{j(i)} + \gamma_{k(ij)} + \epsilon_{l(ijk)}$$

Where: y_{ijk} is the GLS disease intensity where peasant association k is nested within district J nested within zone i , μ is the overall mean, τ_i is the effect of the i th zone, $\beta_j(i)$ is the effect of the j th district within the i th zone, and $\gamma_{k(ij)}$ is the effect of the k th peasant association within the j th district and i th zone, and $\epsilon_{l(ijk)}$ is the error term. Analysis of variance (ANOVA) was performed using SAS V 9.3 statistical (SAS, 2010) [20]. Means were separated using LSD t-test at significance levels of 0.05. Pearson correlation analysis was used to determine the relationships between GLS disease incidence and severity and altitude, cropping system, and growth stage.

Results and Discussion

Status of Maize Grey Leaf Spot in Western Oromia Prevalence and intensity of grey leaf spot at zone level

Studies have shown that grey leaf spots have been found in all assessed areas in western Oromia. Nonetheless, in the 2019/20 planting season, its prevalence varies among the assessed zones. Thus, the highest (70.37%) value was recorded in East Wollega zone; followed by West Shewa zone with 55.56% while the lowest was recorded in Horro Guduru Wollega zone with 29.63% (Figure 2).

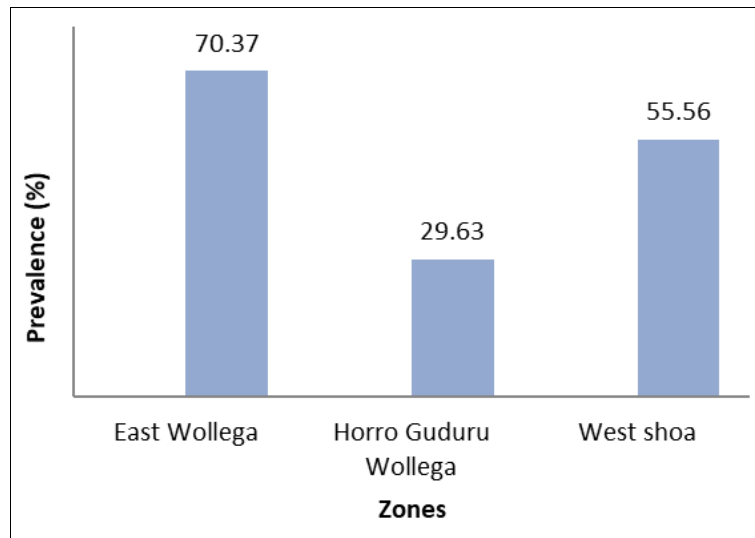
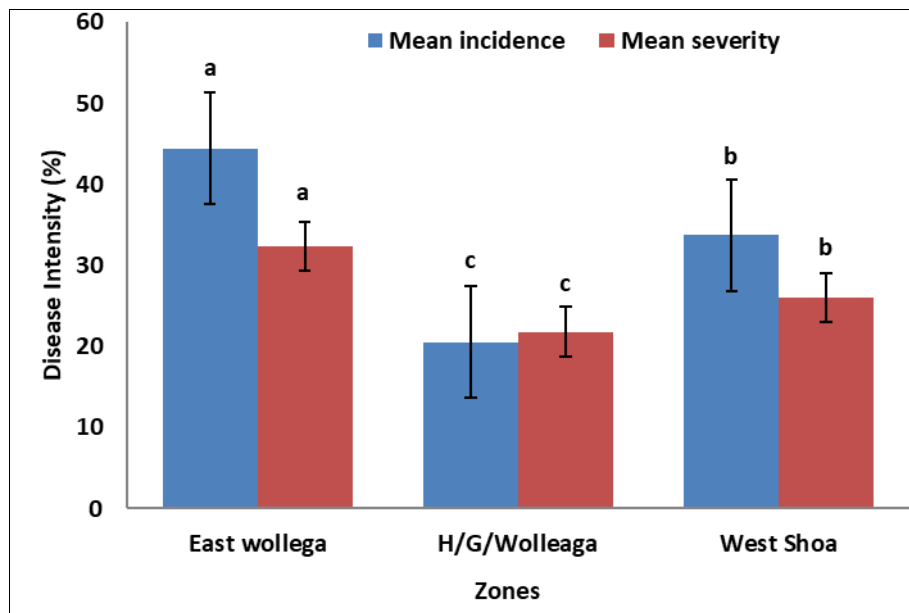


Fig 2: Prevalence of grey leaf spot across survey zone in 2019/20 main cropping season

Similarly, there was significant ($p < 0.05$) difference among the zones in intensity of maize grey leaf spot. GLS incidence and PSI score varied throughout the zones. East Wollega zone had the highest mean disease incidence of grey leaf spot of maize (44.44%), followed by West Shoa (33.705%), and Horro Guduru Wollega zone had the lowest (20.53%). Also, the zone of East Wollega had the greatest mean PSI of maize grey leaf spot with 32.32%, followed by West Shoa zone with 25.99%, and Horro Guduru Wollega zone had the lowest with 21.80% (Figure 3). Gray leaf spot was found in the major maize-producing

regions of Ethiopia's Western, Southern, and Northwestern regions, according to Wegari *et al.* (2001)^[36]. GLS has been Ethiopia's main maize disease since 1998, according to the report. GLS has also increased in prevalence and intensity in the major maize-producing regions of Ethiopia's southern and southwestern regions, according to Nega *et al.* (2016)^[15]. GLS disease intensity on maize producing areas varied depending on climatic conditions, cropping system, and altitude range, according to a survey done in northwestern part of Ethiopia (Sintayehu *et al.*, 2018)^[21].



Incidence (LSD=3.64); Severity (LSD=2.73)

Fig 3: Intensity of grey leaf spot across survey zone in 2019/20 main cropping season Bars with the same letter(s) are not significantly different at $p < 0.05$.

Prevalence and intensity of maize grey leaf spot at district level

The survey revealed that maize grey leaf spot was present in varied degrees in all maize-producing districts of western Oromia. Digga district has the greatest prevalence of GLS disease (100%), followed by Nono and Gudeya Billa districts with 77.77% and 66.66%, respectively. Also, at

55.55%, 55.55%, 44.44%, and 44.44%, respectively, the medium prevalence was found in Wayu Tuka, Guduru, Jibat, and Liban Jawi districts. GLS prevalence was found to be lower in Ababo Guduru and Abay Choman districts, with 33.33% each account (Table 2). The interaction between environmental factors and cultural practices and varieties in use could explain why the disease is so prevalent.

Table 2: Prevalence of GLS across districts in 2019/20 main cropping season

Zones	Districts	Altitude range	Number of assessed fields	Number of infected fields	Prevalence (%)
	Digga	1446-2168	9	9	100
East Wollega	Wayu Tuka	1883-1921	9	5	55.55
	Gudeya Bila	1800-2125	9	6	66.66
Horro Guduru Wollega	Ababo Guduru	2289-2353	9	3	33.33
	Guduru	2300-2332	9	5	55.55
	Abay Choman	2356-2418	9	3	33.33
	Nono	1626-1871	9	7	77.77
West Shoa	Jibat	2106-2361	9	4	44.44
	Liban Jawi	2267-2343	9	4	44.44

Dida *et al.* (2020) [7] reported that the highest altitude ranges from 2031–2422 m.a.s.l associated with high rainfall and also farming practices enhanced the disease. Similarly, the authors reported that, other things kept constant, the use of specific maize variety had its own effect as disease prevalence is minimum with Shone and Limu maize varieties as compared to others. The current result contradicts the authors' findings on the highest altitude; in fact, it indicated that there was a negative relationship between altitude and disease.

The mean grey leaf spot incidence in the studied districts ranged from 15.00% (Ababo Guduru district of Horro Guduru Wollega Zone) to 76.67% (Digga district of East Wollega Zone). Digga had the greatest disease incidence (76.67%) of all the districts surveyed, followed by Nono, Gudeya Billa, and Wayu Tuka, which had disease incidences of 53.33%, 30.11%, and 26.67%, respectively.

This implies that variation in existing weather condition, cultural practices, miss/less of crop rotation and difference in genetic background of maize varieties sown by farmers in the districts boost the development of the GLS disease. On the other hand, Ababo Guduru, Abay Choman, and Liban Jawi, districts had relatively lower GLS incidence of 15%, 18.33% and 23.33%, respectively. In similar manner, Jibat and Guduru districts recorded equal number of incidence with 24.44%. Like with that of incidence, the maximum PSI was recorded on Digga (46.06%) followed by Nono (36.72%). By way of addition, districts such as Gudeya Bila, Wayu Tuka, Guduru and Abay Choman had recorded with 29.44%, 24.47%, 24.44%, and 24.01% of PSI, respectively. In contrast, relatively the mean minimum PSI was scored in the Ababo Guduru district (12.50%) followed by Jibat (20.11%) and Liban Jawi (21.11%) (Table 3).

Variations in disease intensity and prevalence across districts may be related to differences in ecological parameters (altitude, temperature, relative humidity), as well as farming practices in the assessed area.

Planting susceptible maize varieties under high relative humidity was common in districts with a high disease incidence and severity index. Farmers in Digga and Nono,

for example, were mostly utilizing the local and BH540 maize varieties, both of which have a high disease intensity. Farmers in Ababo Guduru, Jibat, and Liban Jawi, on the other hand, predominantly used the BH661 and limu maize varieties. In comparison to the districts with the lowest disease incidence and severity index, those with the highest had higher relative humidity, less crop rotation, and used susceptible maize varieties. The finding of the current study was agreed with Sintayehu *et al.* (2018) [21] who stated that GLS was occurred higher incidence and severity during the main season in predominantly warm temperature and high humidity maize production areas of low and mid altitude of Metema and Takussa districts. According to the current survey, comparatively warm temperatures and high relative humidity could create a favorable environment for pathogen epidemics and increase disease intensity. According to Beckman and Payne (1983) [2], meteorological conditions that promote the development of GLS disease include warm temperatures, evenly distributed rain, and high relative humidity. De Nazareno *et al.* (1993) [5] also realized that a strong positive relationship between relative humidity and disease severity when the weather conditions were conducive for disease development.

According to Sisay *et al.* (2012) [22], the development of GLS disease is fostered by humid and warm conditions. According to Tadesse (2008) [25], the GLS disease is widespread and has a considerable influence on maize yield reduction in both local and improved susceptible varieties in Ethiopia's maize-growing regions. With yield losses as high as 29.10% and significant output losses in most maize-growing areas, the disease is basically one of Ethiopia's most fundamental problems to maize production (Tefferi, 1999; Wegary *et al.*, 2004; *et al.*, 2018) [26, 31, 11]. Regardless of environmental circumstances, low value of disease incidence and severity suggests that a lot of farmer-grown varieties are resistant or tolerable to GLS. Crop rotation (particularly previous crop) was done in the surveyed districts, which could be another justification for the observed data.

Table 3: Disease incidence and severity index of grey leaf spot across the study area in 2019/20 main cropping season at western Oromia

Zones	Districts	Mean incidence (%)	Incidence range (%)	Mean severity (%)	Severity range (%)
	Digga	76.67 ^a	40-100	46.06 ^a	30-64.5
E/wollega	Wayu Tuka	26.67 ^{cd}	10-60	24.47 ^d	18.7-51.3
	Gudeya Bila	30.00 ^c	10-50	29.44 ^c	20-40.5
	Ababo Guduru	15.00 ^f	0-40	12.50 ^e	20-30
H/G/wollega	Guduru	24.44 ^{cde}	0-50	24.44 ^d	20-30.5
	Abay Choman	18.33 ^{ef}	0-50	24.01 ^d	20-39.1
	Nono	53.33 ^b	10-100	36.72 ^b	20-60
W/shoa	Jibat	24.44 ^{cde}	0-50	20.11 ^d	16.2-39.8
	Liban Jawi	23.33 ^{de}	0-40	21.11 ^d	20-41.7

LSD		6.46		4.85	
CV		30.49		24.33	
Mean		32.47		26.54	

Mean values with the same letter within the column are not significantly different; LSD=least significant difference

Prevalence and intensity of grey leaf spot across peasant associations

The within district comparison of maize grey leaf spot distribution indicated that, highest prevalence (100%) of the disease were recorded in the peasant associations of Diga (Bikila, Gudisa and Firomsa), Nono (Chafe Oddo) districts. However, these peasant associations recorded different mean percent of incidences and severities of GLS. The disease was important in Bikila, Gudisa, Firomsa and Chafe Oddo localities with mean incidences of 80%, 80%, 73% and 73% in the order listed. Ula Qarre, Omo Tokofa and Achane recorded with the lowest disease incidence of 10% each account and Gudene Dado, Sirba Olma Hurumu Jibat and Qaso recorded with 13.33% of disease incidence (Table 4).

Similarly, the severity of grey leaf spot in the surveyed

peasant associations didn't show a similar trend to that of incidences. The highest mean disease severity of 58.17% was recorded in Bikila, followed by Chafe Oddo, Firomsa, Gongolcha, Karra, Gudisa and Imboro, with mean values of 46.83, 43.33 38.33%, 36.67, 36.67 and 36.37 in the same order. However, the lowest severity was recorded in Biftun Bate (10%) followed by Sirba Olma, Ula Qarre, Omo Tokofa, Achane a and Irense with mean values of (12.33%) each account. In the localities of Ababo Guduru district, the disease severity was very low as compared to others districts. The lower disease prevalence and intensity in this locality might be due to the fact that GLS is more important at higher temperature and at higher relative humidity in the area during the survey. Additionally, the farmers in the locality were practicing crop rotation and relatively the resistant variety (BH661).

Table 4: Prevalence and intensity of GLS across different peasant associations of the districts in 2019/20 main cropping season at Western Oromia

Zones	Districts	Peasant association	Field assessed	Prevalence (%)	Incidence (%)	Severity (%)
		Bikila	3	100	80.00 ^a	58.17 ^a
	Digga	Gudisa	3	100	80.00 ^a	36.67 ^{cd}
		Firomsa	3	100	73.33 ^a	43.33 ^{bc}
		G/ Badiya	3	66.66	23.33 ^{de}	27.17 ^{efg}
East Wollega	W/Tuka	W/B /Migna	3	33.33	20.00 ^{def}	19.57 ^{ghi}
		kara	3	66.66	36.67 ^{bc}	26.97 ^{efg}
		Gongolcha	3	66.66	40.00 ^{bc}	38.33 ^c
	G/Billa	Garado	3	66.66	30.00 ^{cd}	30.00 ^{de}
		Jere	3	66.66	20.00 ^{def}	12.33 ^{ij}
		B/Bate	3	33.33	20.00 ^{def}	10.00 ^j
	A/Guduru	G/Dado	3	33.33	13.33 ^{ef}	13.33 ^{ij}
		S/Olma	3	33.33	13.33 ^{ef}	12.33 ^{ij}
		Baro	3	66.66	40.00 ^{bc}	30.00 ^{de}
H/G/Wollega	Guduru	Guto	3	66.66	23.33 ^{de}	23.33 ^{efgh}
		Qarre	3	33.33	10.00 ^{fg}	12.33 ^{ij}
		O/Tokofa	3	33.33	10.00 ^{fg}	12.33 ^{ij}
	A/Choman	Irgi	3	33.33	36.67 ^{bc}	28.03 ^{ef}
		Achane	3	33.33	10 ^{fg}	12.33 ^{ij}
		C/Oddo	3	100	73.33 ^a	46.83 ^b
	Nono	Imboro	3	66.66	40.00 ^{bc}	36.37 ^{cd}
		Silkamba	3	66.66	46.67 ^b	26.97 ^{efg}
		M/Kombolcha	3	66.66	30.00 ^{cd}	21.00 ^{efghi}
West Shoa	Jibat	H/Jibat	3	33.33	13.33 ^{ef}	18.33 ^{hi}
		Qaso	3	33.33	13.33 ^{ef}	18.33 ^{hi}
		Irense	3	33.33	20.00 ^{def}	12.33 ^{ij}
	L/Jawi	D/Gudina	3	33.33	20.00 ^{def}	25.00 ^{efgh}
		S/Kombolcha	3	33	30.00 ^{cd}	18.33 ^{hi}
LSD					10.55	7.91
CV					30.49	24.33
Mean					32.10	24.82

Means with the same letter(s) within the column are not significantly different at $p < 0.05$.

Where:H/G/Wollega – Horro Guduru Wollega,W/Tuka-Wayu Tuka, G/Billa-Gudeya Billa, A/Guduru-Ababo Guduru, A/Choman-Abay Choman, L/Jawi-Liban Jawi, G/Badiya-Gute Badiya, W/B /Migna-Warra Babo Migna, B/Bate- Biftun Bate, G/Dado-Gudene Dado, S/Olma-Sirba Dado, M/Kombolcha-Marukombolcha, H/Jibat-Hurumu Jibat, D/Gudina-Dire Gudina and S/Kombolcha-Sadan Kombolcha

Status of grey leaf spot in relation to maize varieties under production

Approximately four hybrids (BH661, BH540, limu, and shone) and one local maize variety were grown over the studied area during the assessment. Thus, the BH661 variety had the largest coverage of 54.32%, followed by the limu variety, which had 19.75% assessed farms. Local and shone varieties had coverage with 11.11 and 8.64% respectively. Furthermore, BH540 had the lowest coverage (6.17%)

among maize varieties in the areas surveyed. There was a significant ($p < 0.05$) difference in disease incidence among the grown varieties. Survey result on incidence and percent severity index of GLS revealed that maize varieties under production varied in their reaction to GLS. The BH540 variety had the highest mean disease incidence (62.00%), while the limu variety had the lowest (20.71%). Local, shone, and BH661 exhibited disease incidences of 60.00%, 45.75%, and 27.37%, respectively. Similarly, there was a significant difference ($p < 0.05$) among varieties grown in disease severity index. The PSI for maize GLS ranged from 19.71% on the limu variety to 45.92% on the BH-540 variety. PSI was also greater in the local variety, at 40.67%. PSI values for shone and BH661 were 30.93% and 24.40%, respectively (Figure 4). The study revealed that GLS was more common and severely affected BH540 and local varieties. On the other hand, varieties such as limu, BH661 and shone performed better

than others in terms of the GLS reaction. This suggested that the nature of susceptibility/resistance of the varieties, as well as favorable climatic circumstances for disease development in the assessed area had a significant impact on the increase in maize GLS. This finding confirmed the finding of a survey conducted in western Oromia during the 2017 main cropping season, which found significant variation in maize varieties' responses to the disease, which could be attributed to genetic variation within varieties (Dida *et al.*, 2020)^[7].

Earlier results from different authors indicate that the increase in the incidence of GLS is related to the continued planting of maize and the use of susceptible maize varieties (Gevers *et al.*, 1994; De Nazareno *et al.*, 1993; Wegary *et al.*, 2008)^[35, 5, 32]. Likewise, Tefferi (1999)^[26] reported field assessment result; thus, among released hybrid maize, only BH660, and PHB30H83 were found relatively tolerant to maize GLS.

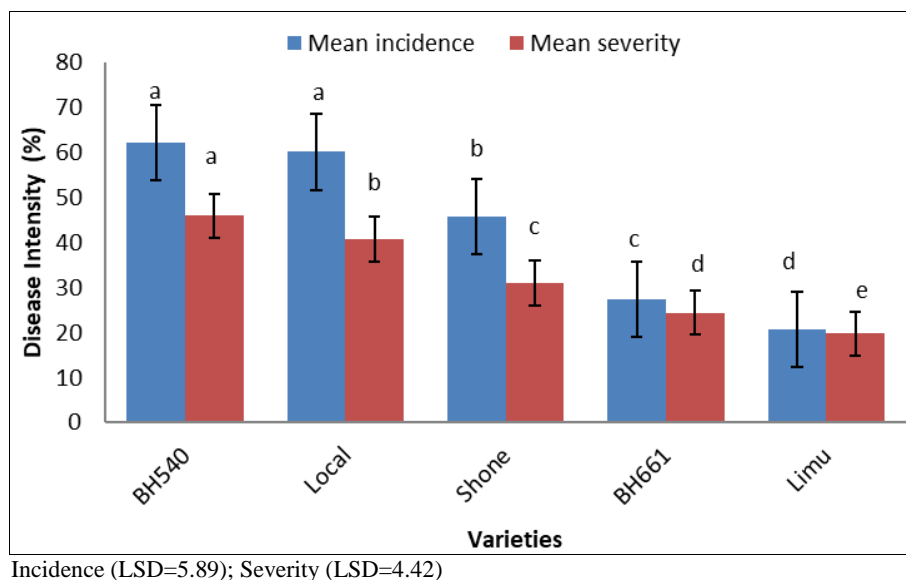


Fig 4: Incidences and percent severity index of GLS on maize varieties under production in surveyed districts Bars with the same letter(s) are not significantly different at $p < 0.05$.

Intensity of grey leaf spot at different growth stages of maize

Maize was grown in different agro-ecological zones with varying planting dates in the assessed area. During the study period, maize fields were at three different growth stages (tasseling, silking, and grain filling). For instance, with 38.27% at tasseling, 20.99% at silking and 40.74% at the grain filling stages in terms of area coverage. The highest (43.44%) mean disease incidence was scored at grain filling stage. Growth stages such as silking and tasseling (anthesis) had 29.33% and 25.77% of disease incidences respectively. Similarly, the highest (32.87%) mean PSI was observed at grain filling stage and the lowest (23.09%) at tasseling stage followed by silking stage with 24.22% (Table 5). Therefore, the present investigation revealed that the development of GLS is also influenced by the stage of maize growth. The disease is crucial, according to Manandhar *et al.* (2011)^[13], since it rapidly damages leaves when the plant is approaching grain maturity. Additionally, survey that was done in india, revealed that disease severity was highest during the grain filling stage (22.06 and 15.50%) and lowest at the vegetative stages (14.31 and 9.49%) (Kalappanavar, 2017)^[10]. The current study confirmed a greater distribution

of GLS in the assessed areas, which was connected to plant stages.

Table 5: Mean incidence and percent severity index of Grey leaf spot on maize at different growth stages of maize in surveyed areas during 2019/20 main cropping season

Growth stage	Farm%	Mean incidence	Mean severity
Grain filling	40.74	43.44 ^a	32.87 ^a
Silking	20.99	29.33 ^b	24.22 ^b
Tasseling	38.27	25.77 ^b	23.09 ^b
LSD		3.78	2.84

Mean values with the same letter within the column are not significantly different; LSD=least significant difference

Effects of preceding crop on maize grey leaf spot incidence and PSI

The results of the survey revealed that maize fields have diverse cultivation histories in the past. Thus, maize was grown 48.15%, teff was grown 17.28%, and wheat was grown 8.64%. There were an equal number of fields produced by Niger and fallow by coincidence; they accounted for 7.41%, while faba bean and field pea accounted for 6.17% and 4.94% of the field, respectively. The highest disease incidence and PSI were recorded on

maize field after maize cropping history with (47.90% and 35.26%) respectively. The lowest mean incidence of GLS (7.50%) was recorded from fields that stayed as fallow field. Maize fields that had wheat and teff cropping history were recorded with incidence of 30.00% and 24.17% respectively.

In the same way, fields with Niger, field pea, and faba bean as a previous crop history had 15%, 14.29%, and 10% disease incidence, respectively. The PSI of maize sown after wheat, Niger, and teff, respectively, was 24.08%, 20.00%, and 19.17%. Maize planted following a fallow system had the lowest PSI (13.98%), followed by faba bean and field pea, both with 17.50%. In the same way, fields with Niger, field pea, and faba bean preceding crop history had 15%, 14.29%, and 10% disease incidence, respectively. Maize grown after wheat, Niger and teff were recorded with 24.08%, 20.00% and 19.17% of PSI respectively. The lowest (13.98%) PSI was recorded on maize grown after fallow system; followed by faba bean and field pea with 17.50% each account (Table 6).

Rotating maize after maize produced the highest disease intensity, possibly because to an increase in the amount of inoculum in the field year after year; however, growing maize after non-host crops to GLS, such as fallow system, faba bean, field pea, and Noug, produced the lowest disease intensity.

Because GLS is a residue-borne disease, growing non-maize

crops in the same field can impair the availability of host residue in the field, reducing the pathogen's capacity to over season. Rotating non-maize crops minimizes inoculum levels (source) and the incidence of disease onset early in the season. To decompose maize residue successfully, it is better to make deep plough; otherwise, completely remove of maize residue is preferable. Rotation of maize with a non-host crop because maize is the only known host crop for GLS, soybean and potato can successfully reduce disease inoculums (Wolf, 2002; Dhami *et al.*, 2015) [35, 6]. According to Latterell and Rossi (1983) [12], disease intensity increased in monoculture approaches due to high inoculums established in the field year after year, however disease intensity decreased in rotation and fallow practices due to maize being the only host crop this fungus is known to attack. They also realized that grey leaf spot of maize is detrimental only to maize, and that rotating the non-host crop for two years can successfully minimize disease inoculums. Grey leaf spot has been related in Africa to cultural practices such as conservation tillage, continuous maize production, and the use of susceptible maize cultivars, according to Gevers *et al.* (1994) [35]. Similarly, a lack of the pathogen's necessary mineral nutrients could play a role in grey leaf spot epidemics (Smith, 1989; Ward, 1996) [23, 29].

Table 6: The effect of previous crop varieties on the mean of maize GLS disease intensity across assessed area during 2019/20 main cropping season at Western Oromia

Previous crop	Farm (%)	Mean incidence	Mean severity
Maize	48.15	47.90 ^a	35.26 ^a
Wheat	8.64	30.00 ^b	24.08 ^b
Teff	17.28	24.17 ^b	19.17 ^{bc}
Niger	7.41	15.00 ^c	20.00 ^{bcd}
Fallow	7.41	7.50 ^c	13.98 ^d
Faba bean	6.17	10.00 ^c	17.50 ^{cd}
Field pea	4.94	14.29 ^c	17.50 ^{cd}
LSD		7.98	5.99

Mean values with the same letter within the column are not significantly different; LSD=least significant difference

Effects of cropping system on maize grey leaf spot incidence and PSI

In the studied areas, cropping systems played a major impact in the development of maize grey leaf spot. According to the result, 83.95% of fields were sown only with maize, whereas 16.05% were sown with maize intercropped. In comparison, the sole cropping system had a higher disease incidence and severity index, with 34.84% and 28.08%, respectively, while the intercropping system

had 30.00% and 24.26% of disease incidence and severity index, respectively (Table 7). Cropping practices such as mono- or inter-cropping, as well as cultivar mixtures, have been demonstrated to have a favorable or negative impact on disease pressure (Agrios, 2005) [1]. Therefore, intercropping or mixed cropping inhibits air circulation inside the crop field, increasing relative humidity and boosting disease progression (Wolf, 2002; Dhami *et al.*, 2015) [35, 6].

Table 7: The effect of farming practices on mean incidence and severity of maize GLS disease during 2019/20 main cropping season across study area

Cropping System	Farm%	Mean Incidence	Mean severity
Sole cropping	83.95	34.84 ^a	28.08 ^a
Intercropping	16.05	30.00 ^b	24.26 ^b
LSD		4.46	3.35

Mean values with the same letter within the column are not significantly different; LSD=least significant difference

Incidence and percent disease index of grey leaf spot on maize in relation to fertilizer application

During the survey, the amount of GLS intensity in fertilized and unfertilized maize fields differed significantly. In terms

of area coverage, fertilized fields comprised 88.89% of the surveyed fields, while unfertilized fields composed 11.11%. GLS disease incidence and PSI were lower in fertilized maize fields compared with unfertilized maize fields.

Fertilized fields had the lowest mean GLS incidence (30.63%) and PSI (26.08%). Unfertilized maize fields, from the other hand, had the highest disease incidence (60.00%) and PSI (38.44%) (Table 8).

Okorai *et al.* (2004) [18] found that the GLS epidemic was much higher in unfertilized plots than fertilized plots, which is consistent with this finding. They also stated that the use of chemical fertilizers has a substantial impact on GLS progress. A single nitrogen application raised plant susceptibility to GLS, while a combined nitrogen and phosphorus application at a recommended level considerably reduced the predisposition effect of a high nitrogen dose (Okorai *et al.*, 2004) [18]. Unbalanced nutrient utilization resulted in nutrient deficit in the host, as well as a loss of resistance status, which predisposed the plants to GLS (Smith, 1989; Ward, 1996) [23, 29]. Plants with nutrient shortages are known to be weaker, slower to grow, and age more quickly. Pathogens are attracted to such plants (Agrios, 2005) [11].

Table 8: The effect of fertilizer application on mean incidence and severity of maize GLS disease during 2019/20 main cropping season across study area

Fertilizer application	Farm%	Mean incidence	Mean severity
Fertilized	88.89	30.63 ^b	26.08 ^b
Unfertilized	11.11	60.00 ^a	38.44 ^a
LSD		4.46	3.35

Mean values with the same letter within the column are not significantly different; LSD=least significant difference

Correlation between altitude, relative humidity, minimum temperature, maximum temperature and intensity of maize grey leaf spot: According to Pearson correlation analysis, there was a highly significant ($p < 0.001$) and altitude had negative correlation with maize grey leaf spot disease incidence ($r = -0.54$) and severity ($r = -0.56$). Similarly, there was highly significant ($p < 0.001$) and positive correlation between relative humidity and disease incidence ($r = 0.38$) and severity ($r = 0.36$). Grey leaf spot incidence and PSI negatively correlated with minimum and maximum temperature although not significantly differ (Table 11). Thus, with maximum temperatures ($r = -0.05$) and minimum temperatures ($r = -0.15$). Likewise, grey leaf spot PSI was negatively correlated with minimum ($r = -0.20$) and maximum ($r = -0.04$) temperatures. This finding suggests that disease intensity of maize GLS is influenced by altitude and weather parameter in general. Thus, it coincided with Sintayehu *et al.* (2018) [21] result states that GLS was occurred higher incidence and severity during the main season in predominantly warm temperature and high humidity maize production areas of low and mid altitude. Also, warm temperatures, evenly distributed rainfall, and high relative humidity, according to Wheeler (1969) [33], are weather conditions that favor disease progress. According to Sisay *et al.* (2012) [22], the development of GLS disease is facilitated by humid and warm conditions.

Because weather data was only obtained using portable devices during disease assessment dates in the current study, some differences between the current survey results and those of past findings may exist. It should be understood that weather conditions over longer time periods than just a few specific dates affect disease development in general. However, it is impracticable to maintain meteorological records throughout the growth season and beyond due to the absence of weather stations in the survey areas.

Table 9: Pearson's correlation coefficients of altitude and weather parameters with incidence and severity of maize GLS in 2019/20 main cropping season

Variable	Incidence	PSI	Altitude	RH	Min. temp	Max. temp
Incidence	1	0.83**	-0.54**	0.38**	-0.15NS	-0.05NS
PSI		1	-0.56**	0.36**	-0.20NS	-0.04NS
Altitude			1	-0.56**	0.10NS	-0.23*
RH				1	-0.58**	0.40**
Min. temp					1	-0.02NS
Max. temp						1

NS = non significant, *Significant level at $p < 0.05$ and **Significant level at $p < 0.01$

PSI= Percentage severity index, RH= Relative humidity

Conclusion

The findings of the survey revealed that Grey Leaf Spot was widespread throughout the majority of the studied Zones and districts, with a notable variation in incidence and severity index from district to district. As a result, East Wollega zone, followed by West Showa zone, recorded the highest disease incidence and severity index. Differences in ecological parameters (altitude, temperature, relative humidity), maize varieties under production, and farming practices in the studied area may all be contributing factors to variations in disease intensity and prevalence among districts. According to the survey's findings, farmers in areas with a high prevalence and intensity of disease should choose maize varieties that are less severe and practice crop rotation to reduce disease intensity. To determine the pattern of disease spread and the significance of maize grey leaf spot disease across seasons, further research would be needed. And also, races identification would be done.

Conflicts of Interests

The authors have not declared any conflict of interests

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