ASSESSMENT OF ETHIOPIAN BARLEY (*HORDEUM VULGARE L.*) GENOTYPES RESISTANCE FOR SCALD (*RHYNCHOSPORIUM COMMUNE*) AND POWDERY MILDEW (*BLUMERIA GRAMINIS*)

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ABSTRACT: The existence of a high degree of variability in crop genetic resources for disease resistance can be taken as one of the strategies that allow crops to survive in equilibrium with the challenges posed by pathogenic organisms. Three hundred twenty barley (*Hordeum vulgare* L.) lines were evaluated for severity and area under disease progress curve (AUDPC) at two locations in Ethiopia. These disease resistance traits were for barley leaf scald (*Rhynchosporium commune*) and powdery mildew (*Blumeria graminis*). The lines were tested during the 2017/18 and 2018/19 cropping seasons for disease severity and in a single season for AUDPC, under natural infestation conditions. The REML analysis based on BLUP mean revealed that lines differ significantly ($p \leq 0.01$) for both disease severity and AUDPC in both diseases. Furthermore, there was a significant difference ($p \leq 0.01$) among the test environments for disease severity based on over years combined data. For leaf scald, the severity was found to be higher in the first year possibly due to a comparatively earlier planting date in the season which in turn facilitated repeated infection within the growing season. Hence, apart from the utilization of resistant lines, avoiding early planting can be taken as a good management strategy to reduce the damage caused by leaf scald. Lines HB-42 and Accn# 243209 were in the top ten for both leaf scald and powdery mildew resistance suggesting their potential for multiple disease resistance. These two lines can be used as parental lines in the attempt to develop high-yielding genotypes with multiple disease-resistance backgrounds.

Keywords: AUDPC, disease severity, genetic resources, farmers' varieties.

INTRODUCTION

In this era of climate change crop production can be hampered by various biotic and abiotic factors. In any breeding program to develop varieties of a crop of interest that can exist in equilibrium with challenges caused by pathogenic organisms, the availability of an important source of variability is imperative (Zhao et al., 2022). Reduction in the genetic base of crops, on the contrary, increases crop vulnerability to various pathogens (Bailey-Serres et al., 2019; Zhao et al., 2022).

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Concerning barley production in the world, diseases caused by fungal pathogens represent the key constraint, despite substantial efforts to manage the damage. Cultivated barley hosts more than 250 different pathogens with variable levels of importance (Walters et al., 2012; Singh et al., 2019). Repeated resistance breakdown in host plants as a result of increased pathogenicity is causing a threat to global barley production. Leaf scald, caused by *Rhynchosporium commune* (Avrova and Knogge, 2012), and powdery mildew, caused by *Blumeria graminis* f. *hordie*, are among the main and most widely distributed diseases in barley (Spies et al., 2012).

Previously, *Rhynchosporium secalis* was known to cause leaf blotch in rye, triticale and barley. However, with advances in pathogenicity studies, it was later discovered this pathogen has further host specialization over the mentioned crops. Hence, the pathogen causing scald in barley was reclassified to be *Rhynchosporium commune,* whereas the pathogen causing leaf blotch in rye and triticale remained *Rhynchosporium secalis* (Zaffarano et al., 2011; Avrova and Knogge, 2012; King et al., 2015; Zhang et al., 2020). Barley leaf scald is a haploid, polycyclic fungus with repeated pathogen generations in one growing season (Avrova and Knogge, 2012). The primary inoculum is coming either from crop debris or infected seed. Splash-dispersal from leaves infected by the pathogen are the source of secondary disease spread (Davis and Fitt, 1992; Zhan et al., 2008). *Rhynchosporium commu*ne a hemibiotrophic fungus (Oliver and Ipcho, 2004; Zhan et al., 2008) that can cause yield loss from 15 to 45% and reduce grain quality (Brown, 1985); 30 to 40% (Paulitz and Steffenson, 2010). In Ethiopia, it is the most widely distributed and destructive disease of barley. The disease is most prevalent in the highlands characterized by high rainfall and low temperature during the cropping season (Meles et al., 2004).

Powdery mildew, an ascomycetes fungi affects more than 10,000 plant species (Takamatsu, 2004; Kusch et al., 2023). *Blumeria graminis* f. sp. *Hordei (Bgh)* is a wind-borne pathogen that relies entirely on its host for its growth and reproduction as is an obligate biotroph (Both et al., 2005; Rsaliyev et al., 2017; Piechota et al., 2019). *Bgh* is the most widespread barley pathogen worldwide and causes yield reduction of 5-10%

and in severe cases yield loss may reach up to 40% (Chaure et al., 2000; Piechota et al., 2019). Owing to the potential to produce large numbers of sexual haploid spores, sexual recombination during the growing season and airborne dispersal over large distances, *Bgh* has a very rapid evolution. This in turn resulted in the development of stable resistance to be very difficult (Bouguennec et al., 2016).

Eighteen different genes conferring resistance to *R. commune* have been reported in barley, many of which have been mapped. On chromosome 3H more than 11 alleles were described (Bjørnstad et al., 2002), *Rsl5b* on chromosome 2H (Schweizer et al., 1995), *Rrsl4* on chromosome 1H (Garvin et al., 2000), *Rrsl6* on chromosome 4H (Pickering et al., 2006), *Rrsl3* on chromosome 6H (Abbott et al., 1995), *Rrs2,Rsl5a* on chromosome 7H (Genger et al., 2005; Hanemann et al., 2009). In addition, several QTL for scald resistance were identified on all chromosomes but 5H (Wang et al., 2014). Barley resistance genes known to confer resistance against powdery mildew could be race-specific (more than 85) (Jørgensen, 1994) or race-non-specific. The powdery mildew resistance genes mapped on the barley genome include *Mla*, the most thoroughly characterized race-specific locus conferring resistance to powdery mildew, *Mlat, MlGa, Mlk, Mlnn, and Mlra* on chromosome 1H; *MlLa* on 2H; *mlo, Mlg,* and *MlBo* on 4H; *Mlj* on 5H; *Mlh* on 6H; and *mlt* and *Mlf* on 7H (Jørgensen, 1994; Schönfeld et al., 1996).

Management of diseases following cultural methods is considered environmentally friendly, but is often less effective, especially under high disease incidence situations. Chemical control methods in Ethiopia are the last option because of the associated higher cost and their adverse effect on the environment. For this reason, genetic resistance is the best option for the sustainable management of pathogens. Deployment of resistant varieties is the most economically effective and environmentally friendly way to cope with the damage caused by pathogens. *mlo*, a recessive allele identified from an Ethiopian barley farmer variety, was found to be a very effective and durable source of resistance against barley powdery mildew. As a result, this resistance gene has been incorporated to a larger extent in European barley varieties (Piechota et al., 2019). Farmers' varieties and breeding materials resistant to scald were also identified (Daba et al., 2019).

However, the ability of the pathogens to evolve rapidly and generate new virulent pathotypes in a short period of time emphasizes the significance of hunting for new sources of resistance genes. To this end, farmers' variety collections represent valuable reservoirs of genetic diversity, which have not been fully employed, and could be successfully exploited in modern breeding programs for disease resistance (Jørgensen and Jensen, 1997). The study aimed to assess Ethiopian barley farmers' and improved varieties' resistance to leaf scald and powdery mildew diseases.

MATERIALS AND METHODS

Genetic materials

The current study comprises 320 barley lines where, 249 barley accessions originally obtained from the Ethiopian Biodiversity Institute were used to develop 501 lines through spike-to-row maintenance, of which 293 were included in this experiment. In addition to the developed lines, 27 varieties developed, released and maintained by national and regional agricultural centers were included, bringing the total number to 320. The experimental materials comprised 6-rowed, 2-rowed (both deficient and male fertile) and irregular (labile) barley variants.

Field experiment

The experiment was laid out in two replications in four rows of 2.5m length. The size of each plot was $2m^2$ and 17g of seed was sown to each of the plots. The experiment was conducted at two locations (Arsi Negelle and Holeta) for two years though the type of disease over the location and type of disease-specific traits over the years were variable. The plots were fertilized with DAP and Urea fertilizers as per the recommended rate of applications for the two sites and other agronomic management were uniformly applied to all the lines.

Data collection

The disease data were collected from lines in which the pathogens grew under natural infestation conditions. Scald disease severity was scored visually on plot bases (Amezrou et al., 2018) following a double-digit scale (D1D2, 00-99) in the experimental fields. The first digit (D1) indicates vertical disease progress on the plant and the second digit (D2) refers to severity measured as diseased leaf area (Saari and Prescott, 1975; Eyal et al., 1987). The disease severity percentage was computed using the following formula (Sharma et al., 2007).

$$\text{Disease severity\%} = \left(\frac{D1}{9}\right)\left(\frac{\mathrm{D2}}{9}\right) x100.$$

Powdery mildew severity was assessed visually as well, however, scored as a single digit (0-9) scoring scale (Saari and Prescott, 1975) and hence disease severity percentage was estimated as follows.

$$\text{Disease severity \%} = \left(\frac{D}{9}\right) x100$$

where D is the disease severity measured as a progress over the height of the plant and diseased leaf area. For both of the diseases scoring was done two times in the first year and four times in fifteen-day intervals in the second year and the final score was taken for percentage disease severity estimation. According to the severity percentage lines can be grouped as highly resistant (0–5%), resistant (5–10%), moderately resistant(10–20%), moderately susceptible (20–30%), susceptible (30–40%), and highly susceptible (> 40%) (Eyal et al., 1987). The disease severity percentage values were Arc-sine transformed.

The area under the disease progress curve (AUDPC) (percent days) gives a quantitative measure of epidemic development and disease intensity (Das et al., 1992). It was computed based on the transformed disease severity percentage corresponding to the four records using the following formula:

$$\mathrm{AUDPC} = \sum_{\mathrm{i=1}}^{\mathrm{n-1}} \left[\frac{(\mathrm{X_i} + \mathrm{X_{i+1}})}{2}\right] (\mathrm{T_{i+1}} - \mathrm{T_i})$$

where Xi=the disease severity on the $_i$th date, $T_{(i+1)}$-T_i= time or days between two disease scores, n=number of dates on which the disease was recorded. The disease data collected varied depending on the locations and year. Scald severity was estimated in years (2017/18 and 2018/19) at Holeta and scald AUDPC was estimated only in the second year (2018/19) at Holeta. Disease severity for powdery mildew was scored in both years and AUDPC was estimated only in 2018/19 at Arsi Negelle. Lower AUDPC values are associated with a better resistance of a particular line to a particular disease and a higher AUDPC score means higher susceptibility.

Statistical analysis

The restricted maximum likelihood (REML) algorithm was used to produce the best linear unbiased prediction (BLUP) mean, in which lines and environments were fitted as random effects. Both scald and powdery mildew severity BLUP mean was combined over two seasons. BLUP means for scald and powdery mildew AUDPC were estimated from a single environment. It was calculated using the META- R statistical software version 6.04 (Alvarado et al., 2019). The model used to generate the combined BLUP means was as follows:

$$Y_{ijk} = \mu + Env_i + Rep_j(Env_i) + Block_k(Env_iRep_j) + Gen_l + Env_i \text{ x } Gen_l + \varepsilon_{ijkl}$$

where Y_{ijk} is the trait of interest, μ is the general mean, Env_i is the effect of the $_i$th environment, $Rep_j(Env_i)$ is the effect of the $_j$th replicate within the $_i$th environment, $Block_k$ (Env_iRep_j) is the effect of the $_k$th incomplete block within the $_i$th environment and $_j$th replicate, Gen_l is the effect of the $_l$th genotype, Env_i x Gen_l is genotype by environment interaction and ε_{ijkl} is the error associated with the $_i$th environment, $_j$th replication, $_k$th incomplete block and the $_l$th genotype, which is assumed to be normally distributed. On the other hand, the model used to produce individual location BLUP mean was:

$$Y_{ijk} = \mu + Rep_i + Block_j(Rep_i) + Gen_k + \varepsilon_{ijk}$$

where Y_{ijk} is the trait of interest, μ is the mean effect, Rep_i is the effect of the $_i$th replicate, $Block_j(Rep_i)$ is the effect of the $_j$th incomplete block within the $_i$th replicate, Gen_k is the effect of the $_k$th genotype, ε_{ijk} is the error associated with the $_i$th replication, $_j$th incomplete block and the $_k$th genotype. The box plot and correlation among variables were performed using the ggplot 2 function of R statistical software version 3.6.1(R core team, 2019).

RESULTS

The panel of barley lines was evaluated for scald and powdery mildew depending on the prevalence of the diseases at Arsi Negelle and Holeta respectively under natural infestation. The lines showed highly significant ($p \leq 0.01$) variation for both disease severity and AUDPC. Treatment by environment (year) interaction effects for disease severity was also highly significant ($p \leq 0.01$) for both leaf scald and powdery mildew, suggesting a differential response of the genotypes over the test years (Table 1; Table 2).

Table 1. REML variance component analysis for barley leaf scald based on individual and combined environments for severity and AUDPC disease traits.

Source of variation	Year			
	2017/18	2018/19		Combined over years
	Scald severity	Scald severity	Scald AUDPC	Scald severity (%)
Year				199.68^{**}
Genotype	418.35^{**}	191.92^{**}	134960.3^{**}	490.09^{**}
Genotype x year				122.54^{**}
Residual	22.85	19.77	19850.66	21.62
Grand Mean	41.21	31.76	919.38	36.48
LSD	6.65	5.85	187.60	7.88
CV (%)	11.60	13.99	15.32	12.74

AUDPC= Area under disease progress curve, **=highly significant, *=significant

Table 2. REML variance component analysis for barley powdery mildew based on individual and combined environments for severity and AUDPC disease traits.

Source of variation	Year			
	2017/18	2018/19		Combined over years
	Powdery mildew severity (%)	Powdery mildew severity (%)	Powdery mildew AUDPC	Powdery mildew severity (%)
Year				38.82^{ns}
Genotype	204.86^{**}	260.85^{**}	323558^{**}	371.02^{**}
Genotype x year				92.86^{**}
Residual	53.25	58.56	42065.55	55.97
Grand Mean	53.58	52.52	1852.80	53.05
LSD	8.88	9.37	269.68	7.72
CV (%)	13.61	14.56	11.07	14.10

AUDPC= Area under disease progress curve, ns=non-significant, **=highly significant

Leaf scald

Although there was seasonal variation, it was possible to classify the barley lines from highly resistant to highly susceptible to leaf scald. In 2017/18, only variety HB-42 was found to be highly resistant (Figure 1; Table 3). Two lines, Acc# 24638-B, Acc# 16866 (0.6%), were resistant, 23 lines (7.1%) were moderately resistant, 39 lines (12.1%) turned out to be moderately susceptible and 93 lines (29%) were classified as susceptible; fifty percent of the lines fell in the highly susceptible category. In the second year (2018/19) on the other hand none of the lines appeared in the highly resistant groups. In this case, the majority of the lines were found to be susceptible but there was one variety (EH-1847) with resistant response and 45 lines (14%) with moderately resistant response. Combined over the two years, the majority of the lines were in the highly susceptible group for this disease. No lines were identified in the highly resistant or the resistant group, however, 23 lines (7.1%) were classified as moderately resistant.

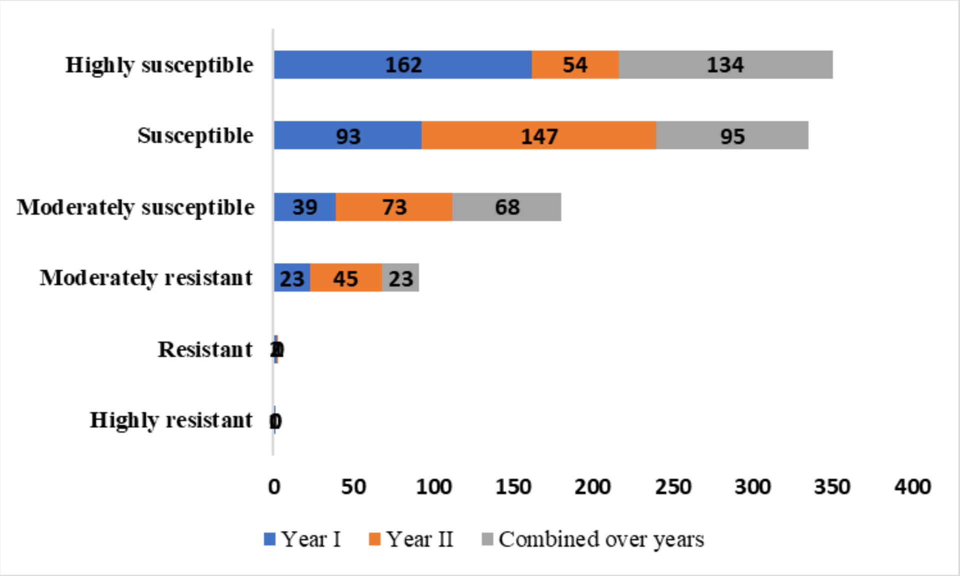

Figure 1. The distribution of the barley lines for leaf scald severity in each of the severity groups over the individual years and combined over the two years. The X-axis is the number of barley genotypes in each severity group and the y-axis severity groups for leaf scald.

Table 3. The response of the top 25 barley lines to severity and AUDPC disease traits of leaf scald on individual years and combined over the test years.

Serial no.	First year (2017-2018)		Second year (2018-2019)		Combined over years		Second year (2018-2019)	
	Genotype	Severity	Genotype	Severity	Genotype	Severity	Genotype	AUDPC
1	243209-A	20.42	HB-42	11.79	Bahati	14.02	Bahati	233.97
2	Bahati	21.21	Explorer	11.79	HB-42	14.29	HB-42	249.83
3	Traveller	21.29	Traveller	11.79	Traveller	14.31	Traveller	250.39
4	HB-42	21.58	Bahati	11.79	243209-A	17.64	Explorer	255.22
5	219026-B	21.95	64334	19.34	219026-B	18.48	243209-A	425.35
6	238360	25.90	219026-B	19.34	Explorer	18.66	219026-B	428.86
7	A hore 880/61	28.11	238360	19.34	238360	20.41	238360	439.97
8	208842-A	28.56	243209-A	19.34	208842-A	25.54	64334	448.70
9	Explorer	29.33	64333-B	22.70	64333-B	25.99	Derebie	676.83
10	64333-B	32.71	208842-A	26.89	A hore 880/61	26.10	64333-B	716.42
11	208841-A	37.87	A hore 880/61	26.89	64334	31.63	HB 1964	743.47
12	243307-A	38.56	HB 1964	26.89	HB 1964	31.86	A hore 880/61	771.20
13	Ibon174/03	39.39	Derebie	35.78	222969-A	38.06	HB-1965	821.44
14	HB 1964	39.57	HB-1965	35.78	Ibon174/03	40.82	208842-A	872.23
15	222969-A	39.73	17651	39.14	208841-B	41.86	Ibon174/03	1051.63
16	213594-A	42.42	18318-B	39.14	213594-A	42.76	17658	1296.15
17	219580-A	44.85	208841-B	39.14	Holker	42.82	219580-A	1318.89
18	17244-A	44.97	222969-A	39.14	208841-A	43.10	17663	1387.85
19	64334	45.68	242093-A	39.14	17244-A	43.47	HB-1963	1412.67
20	208841-B	46.15	Fanaka	39.14	243307-A	43.74	3514-C	1412.69
21	219612-A	47.25	Holker	39.14	18318-B	43.78	64336-A	1418.28
22	204802-B	47.27	17244-A	44.15	242093-A	43.82	213527-A	1424.50
23	Bekoji-1	47.46	17252-C	44.15	Derebie	43.91	4540-A	1429.07
24	Holker	47.50	17658	44.15	Fanaka	43.98	243307-A	1442.06
25	208816-A	47.53	17663	44.15	17651	44.36	222969-A	1499.58

The barley lines showed a considerably wider range for both severity and AUDPC for leaf scald (Table 4a). In 2017/18, the barley lines varied for leaf scald from 1.89 % (HB-42) to 62.24% (Accession# 237329). In 2018/19, it ranged from 9.55% (EH-1847) to 51.26 (Accession # 208923, 213527-A, 215217-A, 219026-B, 221325, 230814-A). Comparing the individual years, the range for scald severity was wider in the first year than in the second year (Figure 2). Besides, the overall mean performance of the lines for leaf scald severity was higher in the first year than in the second year. In accordance with this, the number of lines classified as susceptible (from moderately susceptible to highly susceptible category) was lower in the second year than the first, because of the higher disease pressure in the first year. Out of the 23 lines that were in the moderately resistant category combined over the test years, 13 (57%) were lines derived from farmer's varieties and 10 were improved varieties (Table 3). The variety HB-42 was found to be the most resistant one (11.62%) and the lines from farmers' varieties Accession# 208923, 213527-A and 221325 were the most susceptible with a severity percentage of 55%. Leaf scald AUDPC on the other hand varied between 342.46 (EH-1847) to 1584.95 percent days (Accession# 221325). In the best ten lines characterized by a slow scald development (AUDPC), 5 were improved varieties and among these two, HB-42 and EH-1847 were also in the top ten least scalding (lower scald severity). Likewise, five lines from farmers' varieties, i.e. Accession# 243209-A, 16866, 17148, 24639-A, 242093-A, with slow scald development were also in the top ten least scalding groups. In response to leaf scald, the lines of our panel showed a skewed distribution towards high susceptibility for both severity and AUDPC disease traits (Appendix 1). This pseudo-normal distribution may indicate the quantitative nature of both leaf scald resistance within the lines and varieties.

Table 4. The range of the mean performance of 320 barley lines for severity and AUDPC of leaf scald and powdery mildew based on individual years and combined over years.

a) Leaf scald

Traits	Year					
	2017/18		2018/19		Combined over years	
	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Severity	1.89	62.24	9.55	51.2	11.63	54.92
AUDPC			342.46	1584.95		

AUDPC= Area under disease progress curve

b) Powdery mildew

Traits	Year					
	2018		2019		Combined over years	
	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Severity	20.42	79.83	11.79	67.91	14.02	69.65
AUDPC			233.97	2353.34		

AUDPC= Area under disease progress curve

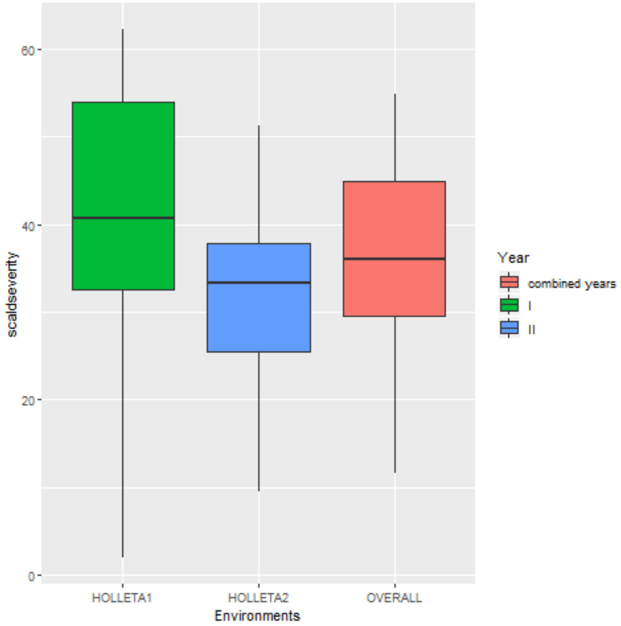

Figure 2. Leaf scald severity of the barley lines across the evaluation years. The x-axis shows the test environments (HOLETA1=Holeta first year, HOLETA2=Holeta second year and OVERALL=combined over the years) indicated as per the legend. The y-axis indicates the leaf scald severity level.

Powdery mildew

The evaluation of the performance of the barley lines to powdery mildew revealed that the majority were highly affected by the disease. In the first year, no single line was recoded to be either highly resistant or resistant, only one line was moderately resistant (243209-A) (Figure 3). In the second year, although no line was either highly resistant or resistant, eight lines (2.5%) were moderately resistant. Combined over the years seven lines (2.2%) were found to be moderately resistant whereas the rest were in the susceptibility category.

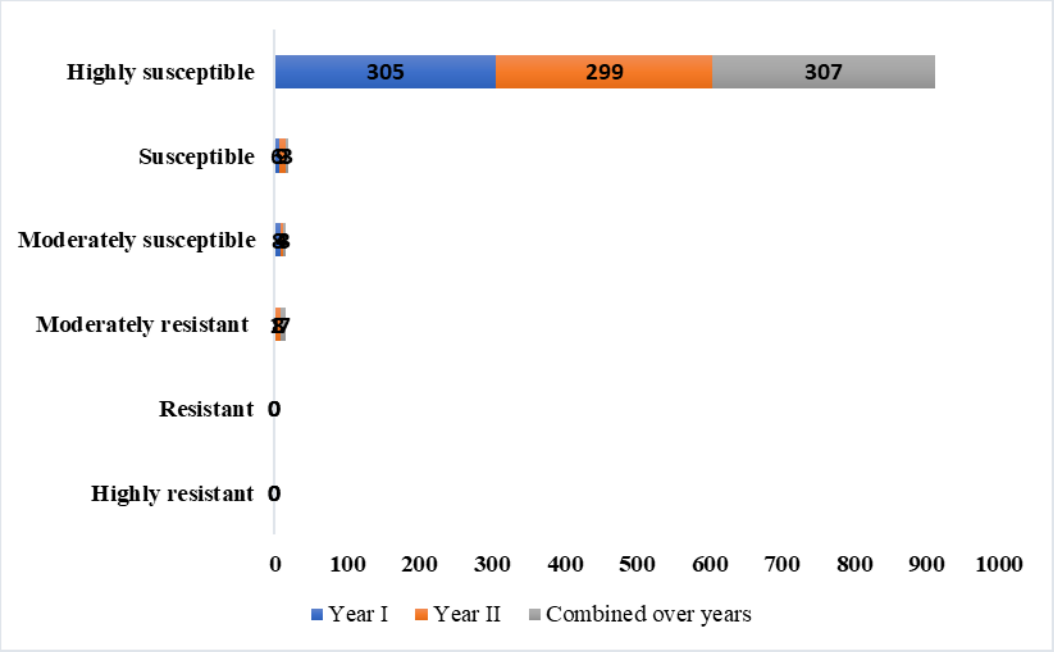

Figure 3. The distribution of the barley lines for powdery mildew severity to each of the severity groups over the individual years and combined over the two years. The X-axis is the number of barley genotypes in each severity group and the y-axis severity groups for leaf scald.

The barley lines also showed a wide range for both disease traits (severity and AUDPC) (Table 4b). For powdery mildew severity in the first year, the lines varied from 20.42% (Accession# 243209-A) to 79.83% (Accession#17244-B). In the second year, it spanned between 11.79% (HB-42) to 67.91% (Accession# 3545-C). The number of lines in the moderately resistant category increased from one in the first year to eight in the second year. Although there was no considerable variation in the range over the two years, the minimum and the maximum values in the second year were lower than it was in the first year (Figure 4) and had comparable mean performance over the two years. Seven barley lines (2.18%) based on over years combined data were found to be moderately resistant to barley powdery mildew and out of these lines four (57%) were improved varieties and three were lines from farmers' varieties. The line with the lowest severity percentage for powdery mildew was the improved variety Bahati (14.01%) (Table 5) and the most susceptible variety with a severity percentage of 69.65% was Accession# 17244-B. The same variety (Bahati) with 233.97 percent days showed the lowest AUDPC value and the line from farmer variety

Accession # 64233-C with 2353.34 percent days had the highest AUDPC. Five of the ten lines with low AUDPC were derived from farmers' varieties and were also in the top ten lines with low disease severity. Out of the five varieties with the lowest AUDPC all but one variety (Derebie) were in the top ten lines with low severity. For both severity and AUDPC disease traits the mean performance of the lines followed a skewed distribution towards higher susceptibility for both of the test years and combined over the years (Appendix 2).

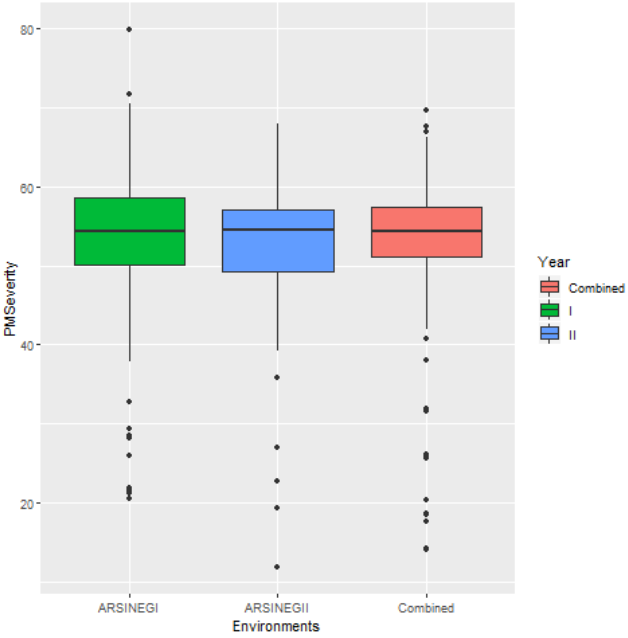

Figure 4. Leaf scald severity of the barley lines across the evaluation years. The x-axis shows the test environments (ARSINEGI= Arsi Negelle year I, ARSINEGII= Arsi Negelle year II and combined= combined over the two years) indicated as per the legend. The y-axis indicates barley powdery mildew severity level.

Table 5. The response of the top 25 barley lines to severity and AUDPC disease traits of powdery mildew on individual year and combined over the test year.

Serial no.	First year (2017-2018)		Second year (2018/19)		Combined over years		Second year (2018-2019)	
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20	208841-B	46.15	Fanaka	39.14	243307-A	43.74	3514-C	1412.69
21	219612-A	47.25	Holker	39.14	18318-B	43.78	64336-A	1418.28
22	204802-B	47.27	17244-A	44.15	242093-A	43.82	213527-A	1424.50
23	Bekoji-1	47.46	17252-C	44.15	Derebie	43.91	4540-A	1429.07
24	Holker	47.50	17658	44.15	Fanaka	43.98	243307-A	1442.06
25	208816-A	47.53	17663	44.15	17651	44.36	222969-A	1499.58

Correlation among leaf scald and powdery mildew disease traits

In order to determine the degree of association among the diseases, the disease traits and the test environments, the BLUP mean of the response of the lines for severity and AUDPC traits of both barley leaf scald and powdery mildew were analyzed for correlations (Figure 5). Correlation coefficients were calculated among disease traits within and between the diseases, among the test environments for both diseases. Significant, and positive correlations were observed between severity and AUDPC disease traits for both leaf scald and powdery mildew (r=0.83 between combined leaf scald severity and AUDPC, r=0.89

between combined powdery mildew severity and AUDPC). Likewise, correlations between the different test environments were also significantly high and positive (r=0.82 between severity in years I and II for scald and r=0.68 between severity in years I and II for powdery mildew). Differently, correlations of disease traits between the two diseases were found to be weak though positive (r=0.078 between leaf scald severity combined over years and powdery mildew severity combined over years).

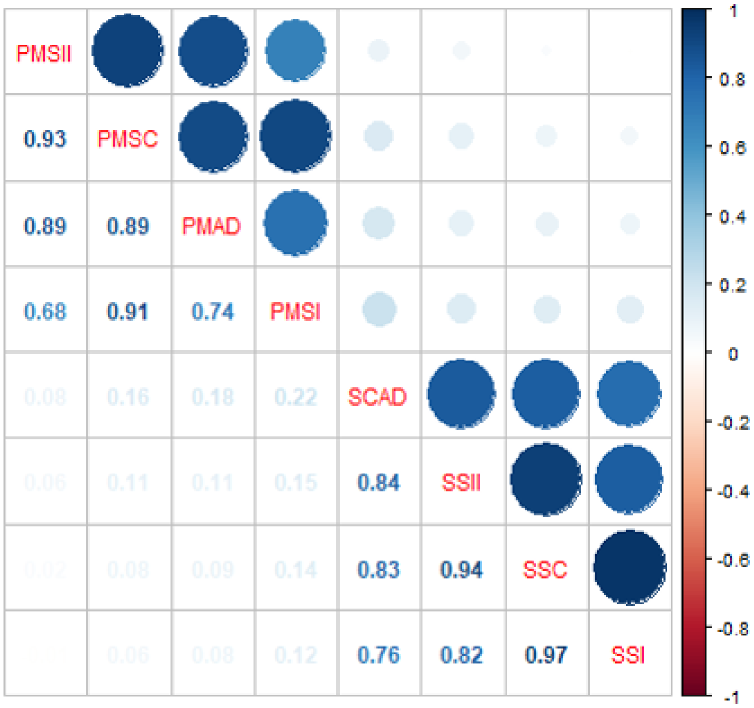

Figure 5. Correlation coefficients for leaf scald and powdery mildew disease traits SSI= leaf scald severity year one, SSII= leaf scald severity year II, SSC= leaf scald severity combined over years, SCAD= leaf scald area under disease progress curve, PMSI= powdery mildew severity year one, PMSII= powdery mildew severity year two, PMSC= powdery mildew severity combined over the test years, PMAD= powdery mildew area under disease progress curve. The strength and direction of the correlation are represented by the color and size of the circle in relation to the legend.

Correlation between the disease traits and test environments was highly significant *($P \leq 0.01$)* (Figure 6) for both leaf scald and powdery mildew. The high, positive and significant correlation between severity combined over years and AUDPC r=0.84 for leaf scald and r=0.89 for powdery mildew suggests the lower the severity percentage, the lower the AUDPC and hence the better the response of the line to the particular disease.

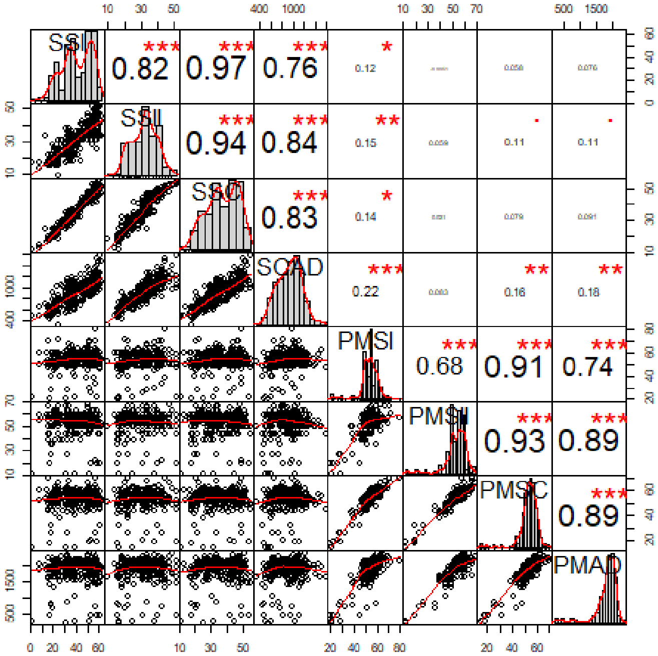

Figure 6. Correlation coefficients for leaf scald and powdery mildew disease traits SSI= leaf scald severity year one, SSII= leaf scald severity year II, SSC= leaf scald severity combined over years, SCAD= leaf scald area under disease progress curve, PMSI= powdery mildew severity year one, PMSII= powdery mildew severity year two, PMSC= powdery mildew severity combined over the test years, PMAD= powdery mildew area under disease progress curve. Above diagonal correlation coefficient values of the disease traits in both diseases. Below diagonal correlation plots between any two disease traits under study.

DISCUSSION

Developing varieties that can cope with virulent pathogens is considered as one of the most effective strategies for managing the damage caused by fungal diseases (Xu et al., 2022). Virulence of a pathogen frequently follows the gene-for-gene concept that underlines for every resistance gene in the host there is a corresponding gene conferring virulence in the pathogen (Gønneød et al., 2002; Gururani et al., 2012). Therefore, the development of resistant genotypes is a continuous process based on the identification of new sources of resistance.

In order for the spore to germinate, subsequent infection and disease development both leaf scald (Tekauz, 1991) and powdery mildew (Glawe, 2008; Scott and Punja, 2021) need enough moisture. Leaf scald specifically grows rapidly under cool and wet growing conditions. In our study, although there was a difference in disease prevalence, there was sufficient disease pressure for both diseases in the test years. The variance analysis based on BLUP mean combined over the test years for leaf scald was found to be significant for studied disease traits justifying the variation among the lines in response to leaf scald (Meles et al., 2004). In agreement with our result, significant variation for disease severity tested for two years and AUDPC among double haploid lines for leaf scald and net blotch in barley was reported (Cherif et al., 2007). The significant treatment-by-year interaction effect suggested the variability of environment and isolates that affect the response of the lines through different isolate line interactions (Yosef et al., 2017). Comparing the occurrence of the leaf scald over the test years it was more prevalent in the first year. As the date of planting in the first year was 10 days earlier than it was in the second year, earlier planting may also be considered as a probable factor explaining the higher prevalence of scald in the first year. Considering the polycyclic nature of the causal pathogen of leaf scald, an earlier planting date may have provided a temporal room for the pathogen to infect the host repeatedly during the growing season. Similar results were also described in barley (Xi et al., 2008; Zerihun et al., 2019). Hence, apart from the utilization of resistant varieties, avoiding early planting can be an alternative management strategy to reduce the damage caused by leaf scald. In addition, as leaf scald spores are dispersed by rain splash taller plants had a comparatively lower chance of being affected by the disease. Lines (Accession# 243209-A, 16866, 17148, 24639-A, 242093-A, HB-42 and EH-1847) that combine both lower scald severity and those that had slowly scalding response are capable of withstanding the reduction in yield caused by the leaf scald. Except for seven lines with moderate resistance, the rest of the lines in our experiment were in the susceptibility category for powdery mildew. However, among these moderately resistant lines for powdery mildew, two were also moderately resistant for leaf scald (HB-42 and Accn#243209) for both disease traits suggesting their potential for multiple disease resistance. Lines of this kind are valuable sources of resistance in crop improvement activities. These lines can be used as a parental line in breeding endeavors of crossing activities to develop genotypes with multiple disease resistance. Quantitative trait loci for multiple disease resistance in wild barley on leaf scald, powdery mildew and net blotch were reported by (Yun et al., 2005). In general, disease development was maximum at the last scoring time for both diseases as a result the final disease scoring can be considered ideal in discriminating lines for the mentioned diseases especially while handling a larger set of lines.

The association between the disease traits and test years for each of the diseases was found to be highly significant and positive. In this case, the correlation coefficient between scald severity in the first and second year was (r= 0.82). This correlation suggests resistance in the first year was resistant in the second year too despite the variation among the test years in the disease prevalence. Lines that showed low scald severity showed lower percent days for AUDPC and it is in agreement with (Paraschivu et al., 2013). Similarly genotypes with the least mildew severity exhibited the lowest AUDPC. A positive and significant correlation between severity and AUDPC disease traits for powdery mildew was in agreement with the findings of (Liatukas and Leistrumaite, 2007).

CONCLUSION AND RECOMMENDATION

In the current research, the barley genotypes were studied for their performance for two major barley diseases leaf scald and powdery mildew. The finding uncovered that adjusting planting time, particularly avoiding early planting can be considered as one of the mechanisms to minimize grain yield reduction that perhaps occurs as a result of repeated infection of leaf scald thereby reducing the cycle of disease infection within the season. The varieties identified to exhibit multiple disease resistance characteristics (HB-42 and Accn# 243209) are potential breeding materials as parental lines in future breeding activities to develop varieties that combine high-yielding characteristics under multiple disease resistance backgrounds. It will also be important to assess the response of the barley genotypes studied for the other major diseases of barley (net blotch) over different locations and years.

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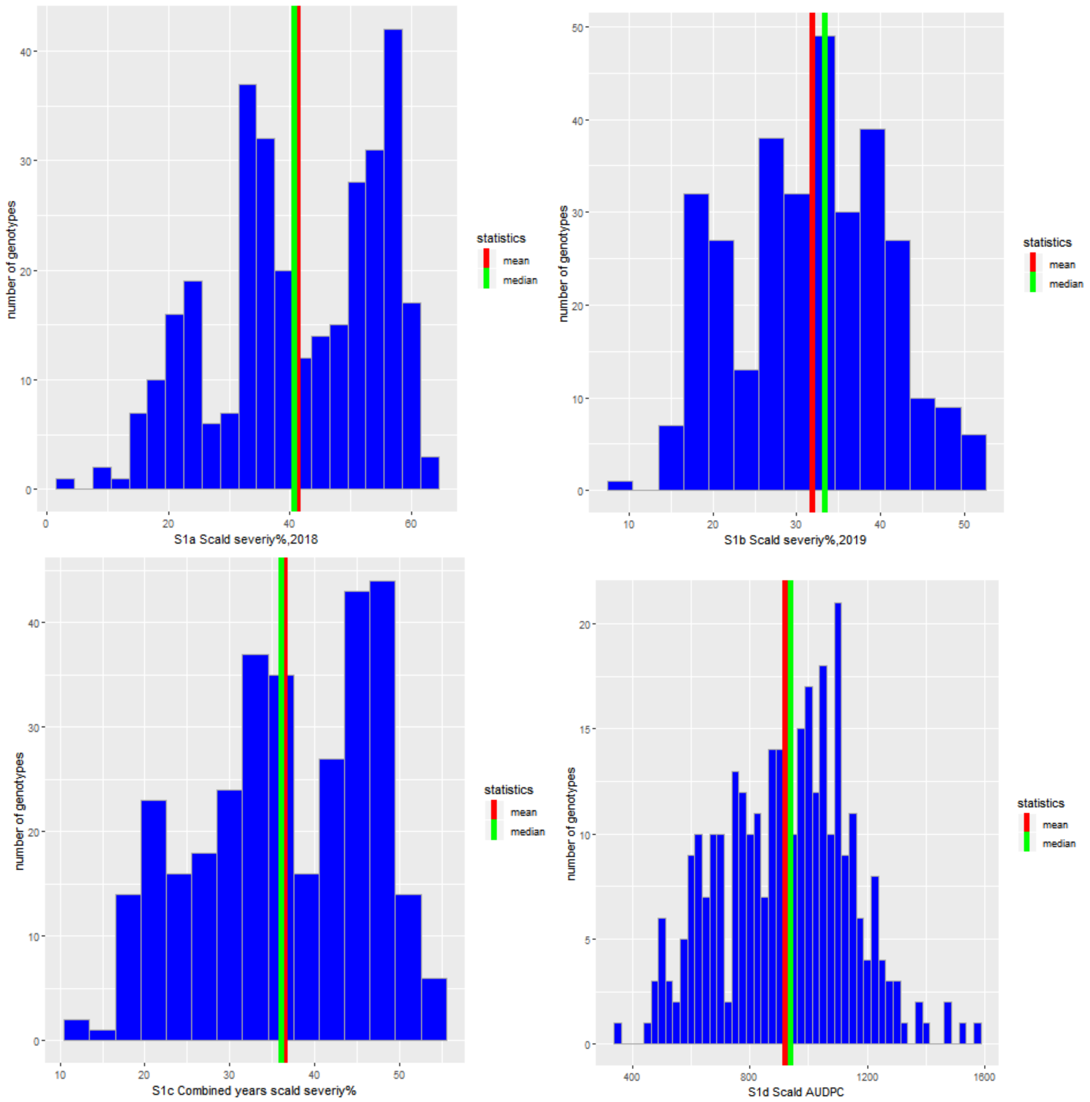
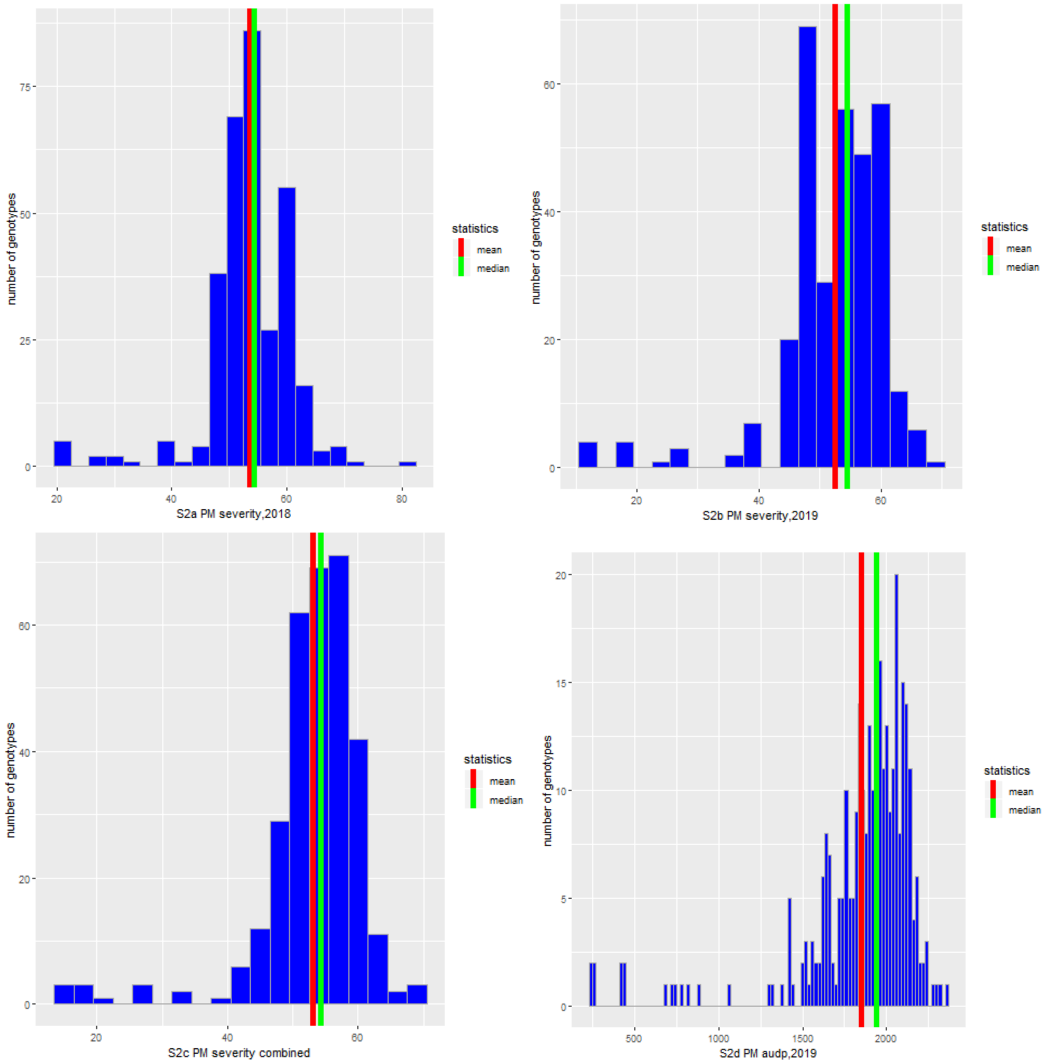


Appendix 1. The distribution of the mean performance 320 barley lines for Leaf scald severity and AUDPC traits. S1a=scald severity % for the first year (2018), S1b=Scald severity % for the second year, S1c scald severity % combined over the test years and S1d=scald AUDPC for 2019. The x-axis disease severity percentage and AUDPC (percent days) and the y-axis number of genotypes.



Appendix 2. The distribution of the mean performance 320 barley lines for Leaf scald severity and AUDPC traits. S2a=powdery mildew severity % for the first year (2018), S2b=powdery mildew severity % for the second year (2019), S2c=powdery mildew severity % combined over the test years and S1d=powdery mildew AUDPC for 2019. The x-axis disease severity percentage and AUDPC (percent days) and the y-axis number of genotypes.