

Identification of high-yielding barley genotypes with adequate foliar disease resistance in north delta of Egypt

ABSTRACT

In addressing the global impact of fungal foliar diseases on barley yield; namely powdery mildew, net blotch and leaf rust, this study undertakes a robust phenotypic and genotypic evaluation. A cohort of 132 barley genotypes, were examined under conditions of elevated pathogenic stress. Multivariate analyses revealed statistically significant inter-genotypic variances for all measured agronomic and pathological traits, concomitant with noticeable seasonal effects. In a nuanced observation, the genotype-by-season interaction elicited significant impacts across a plethora of traits, with the exception of spike length and thousand kernel weight. From a quantitative genetics perspective, broad-sense heritability estimates were remarkably high: 97.2% for powdery mildew, 89.3% for net blotch, and 97.5% for leaf rust. Concurrently, genetic advance metrics for biological yield, grain yield, powdery mildew, net blotch, and leaf rust registered in the high to very high categories. Correlational analyses substantiated a positive linear relationship between grain yield and maturity date, plant height and biological yield. Employing biplot analytic techniques, 12 genotypes were delineated that manifested superior yield parameters whilst demonstrating an amenable level of resistance to powdery mildew, net blotch, and leaf rust. Thus, investigation illuminates the possibility of deploying high-yielding, disease-resistant barley cultivars as an efficacious strategy for mitigating the deleterious effects of fungal foliar pathogens on barley production in Egypt.

Key words: *Fungal diseases, yield loss, resistance, heritability, correlation, biplot*

1. INTRODUCTION

In the northern regions of Egypt, barley crop is susceptible to three predominant mycotic afflictions: powdery mildew, net blotch, and leaf rust. Constituting significant impediments to both of yield and grain quality, these pathogenic agents exhibit considerable genomic plasticity, with multiple strains or pathotypes contributing to their virulence [1, 2, 3]. Notably, historical data has quantified yield debilitation, attributing up to a 20% loss to these fungal diseases [4, 5, 6]. Powdery mildew alone can account for a staggering 30% yield reduction [7]. In the case of leaf rust, yield diminutions can escalate to an alarming with mean losses generally hovering around 25% [8], in the epidemic conditions net blotch cause yield losses more than 70% [9].

The utilization of fungicides, though effective to some extent, raises concomitant issues concerning environmental degradation and public health [10, 11]. Consequently, the adoption of disease-resistant cultivars emerges as a more ecologically and biomedically benign countermeasure. Further impetus for this strategy arises from the significant grain yield diminutions engendered specifically by net blotch infection [12, 13]. As global food security remains a paramount concern, the breeding of high-yield, disease-resistant cereal crops becomes imperative, not merely to meet local needs but to mitigate worldwide alimentary demands.

2. MATERIAL AND METHODS

A comprehensive assemblage comprising 130 barley genotypes sourced from the International Center for Agricultural Research in the Dry Areas (ICARDA) was meticulously evaluated. This evaluation also included two check cultivars: one denoted as susceptible (Giza 123) and the other as

resistant (Rihane-03). Agronomic and yield performance were scrutinized via in situ field trials adhering to an 11 × 12 α-lattice experimental design, featuring dual replications. These trials were orchestrated at the Experimental Farm of the Sakha Agricultural Research Station, situated in the northern delta of Egypt, precisely at geographical coordinates 31° 05' N latitude and 30° 56' E longitude, during the biennial agronomic cycles of 2020-2021 and 2021-2022.

The experimental site on the average has annual precipitation of about 53.6 and 31.2 mm in the first and second seasons, respectively, with minimum and maximum temperature of 8 °C and 34 °C (Table 1).

Table 1: Maximum, minimum temperature, average and rainfall during the two growing seasons of barley crop at Sakha Agricultural Research Station, (ARC), Egypt.

Month	Temperature °C						Rain mm month ⁻¹	
	Maximum		Minimum		Average		2020/2021	2021/2022
	2020/2021	2021/2022	2020/2021	2021/2022	2020/2021	2021/2022		
December	24	20	17	12	21	17	20.7	9.1
January	22	18	14	8	19	14	5.4	10.6
February	21	21	12	9	17	16	13.5	6.4
March	21	22	11	8	17	16	13.3	2.6
April	23	33	14	14	19	25	0.6	2.5

In a meticulously designed agronomic setup, each barley genotype was sown within a tri-row plot, the dimensions of which spanned 0.6 m × 2 m long, total plot area 1.2 m². Seeding operations were executed on mid of December across both experimental seasons, adhering to a uniform seeding rate of 50 kg fed⁻¹, and were structured within an 11 (genotype) × 12 (BLK) α-lattice experimental framework that incorporated dual replications. Cultivation practices recommended by the Ministry of Agriculture were diligently employed at temporally optimal junctures, and cotton was utilized as the antecedent crop in both instances for soil health maintenance. The experiment was grown under natural infection in the field and the experiment was surrounded with highly sensitive cultivars for studied diseases as spread disease cultivars. Table 2 provides the names of the twelve selected genotypes.

Table 2. Name of the selected barley genotypes.

No	Genotype	Name
1	Line 12	PENCO/CHEVRON-BAR//PETUNIA 1
2	Line 22	CAPLUL/ZIGZIG
3	Line 23	CAPUL/PENTUNIA 1
4	Line 24	CAPUL//PENCO/CHEVRON-BAR
5	Line 29	LEGACY/CHAMICO//ADABELLA
6	Line 30	LA MOLINA 96/CIRU
7	Line 50	BLLU/CDC KENDALL
8	Line 67	P.STO/LBIRAN/UNA80//LIGNEE640/4/BLLU/5/PETUNIA1/6/LIMON/BICHY2000
9	Line 112	LIMON/BICHY2000//CANELA/3/MSEL
10	Line 114	MSEL/GUNTHER//BARKE
11	Line 116	PFC9202//CLI18/E.QUEBRACHO/3/CANELA
12	Line 117	ALEL/ESCOBA/3/ARUPO/K8755//MORA/4/CANELA/5/MSEL

Studied traits including, but not limited to, days to maturity (DM), plant height (cm), spike length (cm), biological yield (kg plot⁻¹), grain yield (kg plot⁻¹), 1000-grain weight (g). Also, powdery mildew

(%), net blotch (%), and leaf rust (%) infection severity were comprehensively cataloged for each genotype as percentage to facilitate multi-dimensional evaluation.

For the statistical dissection of phenotypic data, estimates of variance components were ascertained through the deployment of genstat statistical software. These variance metrics were subsequently harnessed to calculate the broad-sense heritability (H^2) via the equation:

$$H^2 = \left(\frac{V_g}{V_{ph}} \right) \times 100$$

where V_g and V_{ph} signify genetic and phenotypic variances, respectively.

The expected genetic advance (G a) expressed as a percentage of the mean value with an assumed 5% intensity of selection pressure was computed by the formula given by **Hallauer and Miranda [14]** as:

$$Ga = K. h. \sigma_{ph}$$

Where: $k = 2.06$, constant for 5% selection intensity (i.e. the highest- performing 5% are selected), $h =$ narrow-sense heritability, $\sigma_{ph} =$ Phenotypic standard deviation of the population.

The predicted genetic advance (Ga %) from selection as percentage of mean was calculated as: $Ga \% = (Ga / \bar{x}) \times 100$

The phenotypic and genotypic coefficients of variation are computed as follow **Burton [15]**.

$$PCV = \left(\frac{V_{ph}}{\bar{x}} \right) \times 100 \quad PGV = \left(\frac{V_g}{\bar{x}} \right) \times 100$$

Where: PCV, GCV are phenotypic and genotypic coefficients of variation, respectively; V_{ph} , V_g are corresponding variances.

Pearson correlation analysis was made using Minitab software to determine the relationship between all the studied traits. The Principal Component, biplot and factor analyses were performed using GenStat software.

The Relative Resistance Index (RRI) was calculated by using country Average Relative Percentage Attack (CARPA) which calculated on a 0 to 9 scale, where 0 represents the most susceptible and 9 shows highly resistant variety [16]. The standard for desirable index was maintained ≤ 7 whereas value for acceptable index was fixed as 5. The following formula was used for calculation of RRI [16, 17], $RRI = (100 - CARPA) / 100 \times 9$.

3. RESULTS

For analysis of variance, there were significant differences for all studied traits among the genotypes and the effects due to seasons. The genotypes and genotypes \times season interaction effects were significant for all traits except spike length and 1000 grain weight in genotypes \times season interaction (Table 3).

Table 3. Mean squares of the barley genotypes evaluated for all studied traits during 2020-2021 and 2021-2022 seasons.

SOV	d.f.	Days to Maturity	Plant height	Spike length	Biological yield	Grain yield	1000 grain weight	Powdery mildew	Net blotch	Leaf rust
Season	1	2698.26**	1699.9**	154.05**	12464.5**	5.85 ^{ns}	451.61**	1591.9**	30797.46**	2550.2**
Season x Rep	2	44.01**	1098.1**	0.51 ^{ns}	1405.0**	178.87**	61.82**	2280.8**	1012.65**	751.8**
Season x Rep x BLK	44	3.54*	48.19 ^{ns}	1.49*	178.68**	27.78**	43.27**	512.36**	129.24**	406.23**
Genotype	131	11.20**	138.88**	3.25**	319.79**	46.16**	119.07**	1612.20**	357.81**	938.0**
Season x Genotype	131	4.43**	85.82**	0.97 ^{ns}	370.85**	33.77**	7.84 ^{ns}	290.37**	155.68**	205.76**
Error	218	2.01	49.04	0.99	43.67	9.92	6.23	45.54	38.23	23.23

Results in Table 4 indicated that, grain yield ranged from 0.46 to 1.02 kg plot⁻¹ with a mean 0.67, while powdery mildew ranged from 0.1 to 77.5% and net blotch ranged from 2 to 60% and leaf

rust ranged from 0.1.6 to 85%. Although 73%, 58% and 96% of lines were resistance to powdery mildew, net blotch and leaf rust, respectively compared with local check (Giza 123). Meanwhile, 65%, 54% and 53% of lines were resistance to powdery mildew, net blotch and leaf rust, respectively compared with Introduced check (Rihane-03). 14% of genotypes were higher biological yield than Giza 123 and 11 genotypes were better than Giza 123 in 1000-grain weight. About 14% of lines including twelve genotypes produced superior grain yield than Giza123 (No. 12, 22, 23, 24, 29, 30, 50, 67, 112, 114, 116, and 117) and had adequate foliar diseases infection (Table 5).

Table 4: Mean and range for all studied traits of barley genotypes and checks evaluated.

Traits	Range		Mean	Local check (Giza 123)		Introduced check (Rihane-03)	
	Min	Max		Mean	Rank	Mean	Rank
Days to Maturity (day)	124.0	131.5	127.9	127.6	62	129.1	98
Plant height (cm)	78.8	106.3	90.9	98.2	15	91.7	61
Spike length (cm)	5.2	10.5	8.2	8.2	72	5.3	132
Biological yield (kg plot ⁻¹)	1.09	2.36	1.59	1.94	19	1.65	57
Grain yield (kg plot ⁻¹)	0.46	1.02	0.67	0.81	20	0.75	28
1000 grain weight (g)	36.9	62.6	47.9	55.9	12	52.1	34
Powdery mildew %	0.1	77.5	24.5	37.5	95	31.3	87
Net blotch %	2.0	60.0	24.9	26.2	78	18.8	73
Leaf rust %	1.6	85.0	19.5	62.5	128	15.0	71

Table 5: Superior grain yield genotypes with adequate values for powdery mildew, net blotch and leaf rust with comparisons to Giza123 (local check) and Rihane-03 (Introduced check) in descending order according to grain yield.

Genotype No.	Days to Maturity (day)	Plant height (cm)	Spike length (cm)	Biological yield (kg plot ⁻¹)	Grain yield (kg plot ⁻¹)	1000 grain weight (g)	Powdery mildew (%)	Net blotch (%)	Leaf rust (%)
Line 114	129.7	96.8	9.5	2.31	1.02	52.8	0.1	10	4
Line 67	129.2	106	8.8	1.91	0.96	52.7	26.3	16.3	8.7
Line 112	128.5	86.3	8.3	1.99	0.93	50.7	1.3	28.8	4.3
Line 117	129.5	95.3	9	2.02	0.92	53.6	10	25	4
Line 116	128.7	99	8	2.01	0.91	55.9	16.3	21.3	4
Line 29	126.2	97	7.5	1.62	0.88	50	26.2	35	3
Line 23	126.5	93	7.5	1.95	0.87	50.4	30	20	13.8
Line 24	128	94.8	8.3	1.86	0.85	52.3	8.8	8.8	6.5
Line 30	130.2	99.5	7.3	1.91	0.83	49.8	1.6	2	3
Line 22	128.8	89.8	7	1.81	0.82	49.8	13.8	16.3	13.8
Line 12	126.7	93	7.3	1.64	0.82	49.4	28.8	20	18.7
Line 50	125.7	100	8	1.79	0.81	44.6	10	11.5	10
Rihane-03	129.1	91.7	5.3	1.65	0.75	52.1	31.3	18.8	15
Giza 123	127.6	98.2	8.2	1.94	0.81	55.9	37.5	26.2	62.5
LSD 0.05	2.8	9.8	1.4	0.29	0.14	3.5	9.4	8.6	6.7

Heritability estimates in broad sense expected and predicted genetic advance from selection for all the studied traits are presented in Table (6). Broad sense heritability percentages ranged from 64.7% for plant height to 97.5% for leaf rust. PCV was slightly higher than all the traits. The heights estimate of phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) were observed for powdery mildew, net blotch and leaf rust traits. The expected genetic advance (Ga) ranged from 0.2 for grain yield to 40.2 for powdery mildew, while the estimates of predicted genetic advance (Ga%) ranged from 2.2% for days to maturity to 164.2% for powdery mildew.

Table 6: Estimates of broad sense heritability (H²), phenotypic and genotypic coefficients of variation, expected genetic advance (Ga) and predicted genetic advance (Ga %) from selection for all studied traits.

Traits	H ²	PCV	GCV	Ga	Ga %
Days to maturity	82.2	2.2	1.8	2.8	2.2
Plant height	64.7	38.2	24.7	7.9	8.6
Spike length	69.4	9.9	6.9	1.3	15.7
Biological yield	86.3	5.0	4.3	0.5	31.6
Grain yield	78.5	1.7	1.4	0.2	25.9
1000 grain weight	94.8	62.2	58.9	10.7	22.3
Powdery mildew	97.2	1646.5	1600.0	40.2	164.2
Net blotch	89.3	359.1	320.7	17.4	69.9
Leaf rust	97.5	1203.7	1173.9	30.8	157.8

The relative importance and contribution of each trait to multivariate polymorphism were tested using PCA (Table 7). PCA was performed, and the first four axes showed eigenvalues >1 and accounted for more than 71% of the observed phenotypic diversity for the barley genotypes examined (Table 7). The first PC1 explained 31% of the total variation and was positively correlated with grain yield (0.41) followed by biological yield and days to maturity (0.39), while the second component (PC2), explaining 17% variance, was positively correlated with biological yield (0.41). It was revealed that high yielding barley genotypes; 114, 67, 112, 117, 116, 29, 23, 24, 30, 22, 12 and 50. Table 5 showed high values for plant height, biological yield, spike length and 1000 grain weight, whereas have low values for powdery mildew, net blotch and leaf rust infections (Fig. 1).

Table 7. Eigenvalues, eigenvectors and percentage of variation explained by the first four principal components assessed for 9 traits in 132 barley genotypes.

Eigenvalues	2.81	1.48	1.16	0.94
Proportion	0.31	0.17	0.13	0.10
Cumulative Proportion	0.31	0.48	0.61	0.71
Days to heading	0.39	-0.33	0.03	0.23
Plant height	0.22	0.31	0.53	-0.20
Spike length	0.21	-0.40	0.54	0.20
Biological yield	0.39	0.41	0.10	0.24
Grain yield	0.41	0.39	0.03	0.10
1000 grain weight	0.35	-0.31	0.14	-0.47
Powdery mildew	-0.35	0.40	0.35	0.14
Net blotch	-0.30	-0.01	0.38	-0.54
Leaf rust	-0.32	-0.23	0.36	0.52

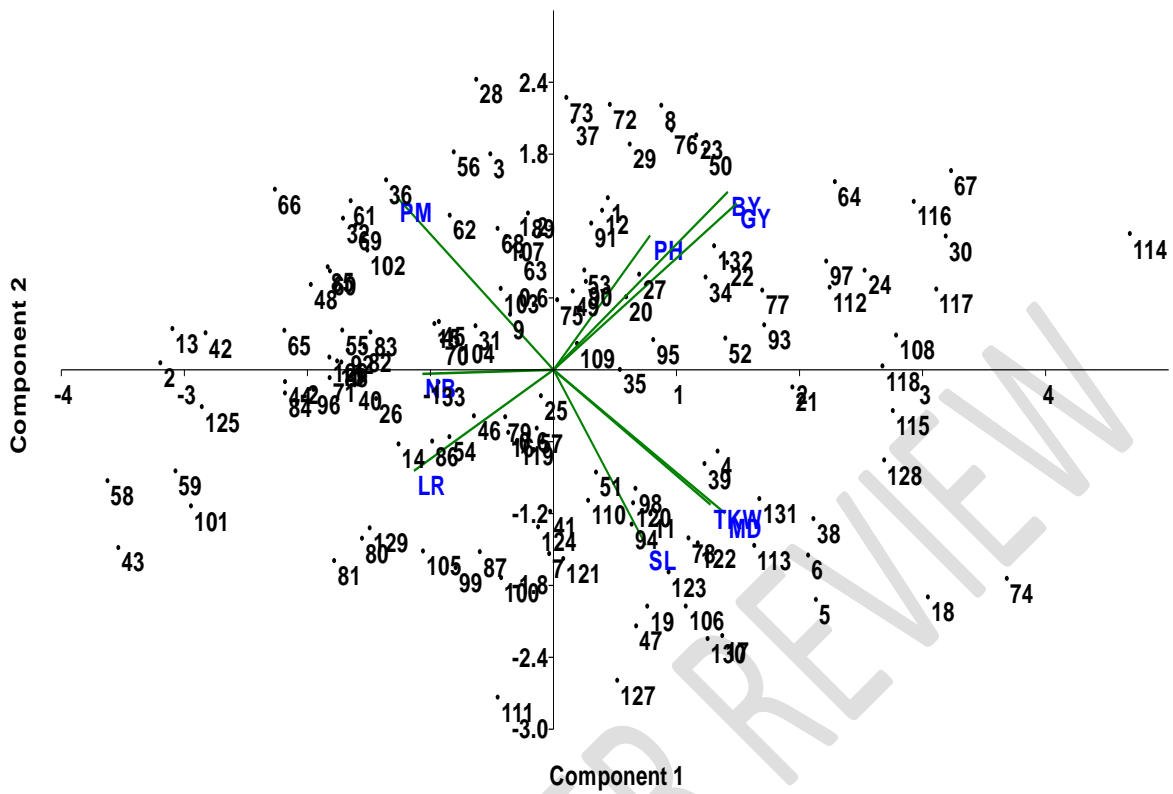


Fig. 1: Biplot of the 132 barley genotypes evaluated for grain yield, yield-related traits and foliar fungal diseases powdery mildew, net blotch, and leaf rust on the first two principal components.

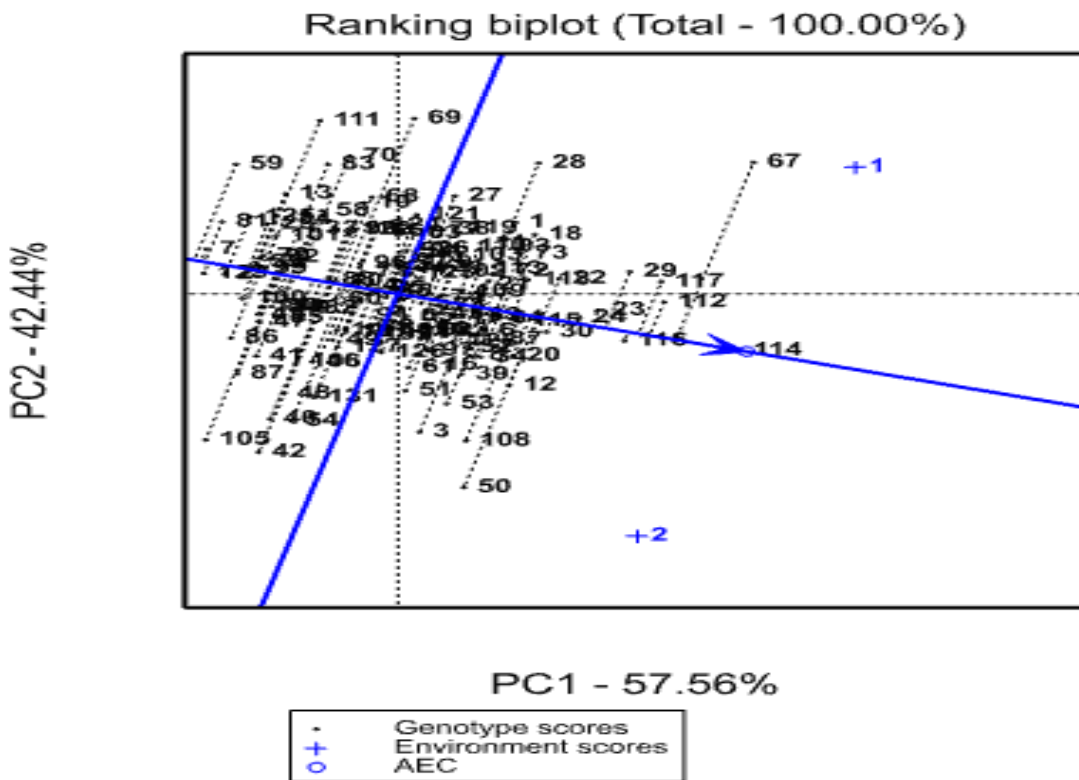


Fig 2. GGE biplot, for grain yield of 132 barley genotypes across 2 environments.

Factor Analysis of the yield- related traits on the first and second factors showed acute angles among grain yield, biological yield, and plant height, which depicted positive correlations among these parameters. Acute angles were also observed among days to maturity, spike length, and 1000-grain weight. Obtuse angle was also observed between grain yield and powdery mildew, net blotch, and leaf rust traits (Fig 3).

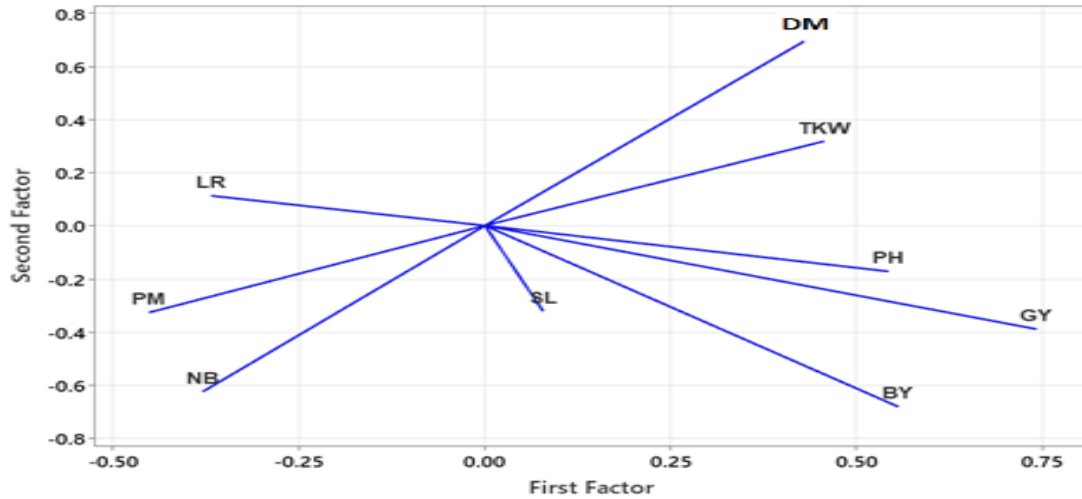


Fig 3: Factor Analysis of days to maturity, plant height, spike length, biological yield, grain yield, 1000-grain weight, powdery mildew, net blotch and leaf rust traits.

Positive and high significant correlation between grain yield and days to maturity ($r = 0.1$), plant height ($r = 0.356$), biological yield ($r = 0.643$) and 1000-grain weight ($r = 0.092$) as showed in Fig. 4, also positive and high significant correlation between biological yield and both of plant height ($r = 0.264$) and spike length ($r = 0.181$). A negative and high significant correlation between grain yield and powdery mildew (-0.092), net blotch (-0.092) and leaf rust (-0.147). Also, negative and high significant correlation between 1000-grain weight and powdery mildew (-0.318), net blotch (-0.091) and leaf rust (-0.216). Days to maturity had negative high significant correlation with powdery mildew ($r = -0.216$) and net blotch ($r = -0.497$).

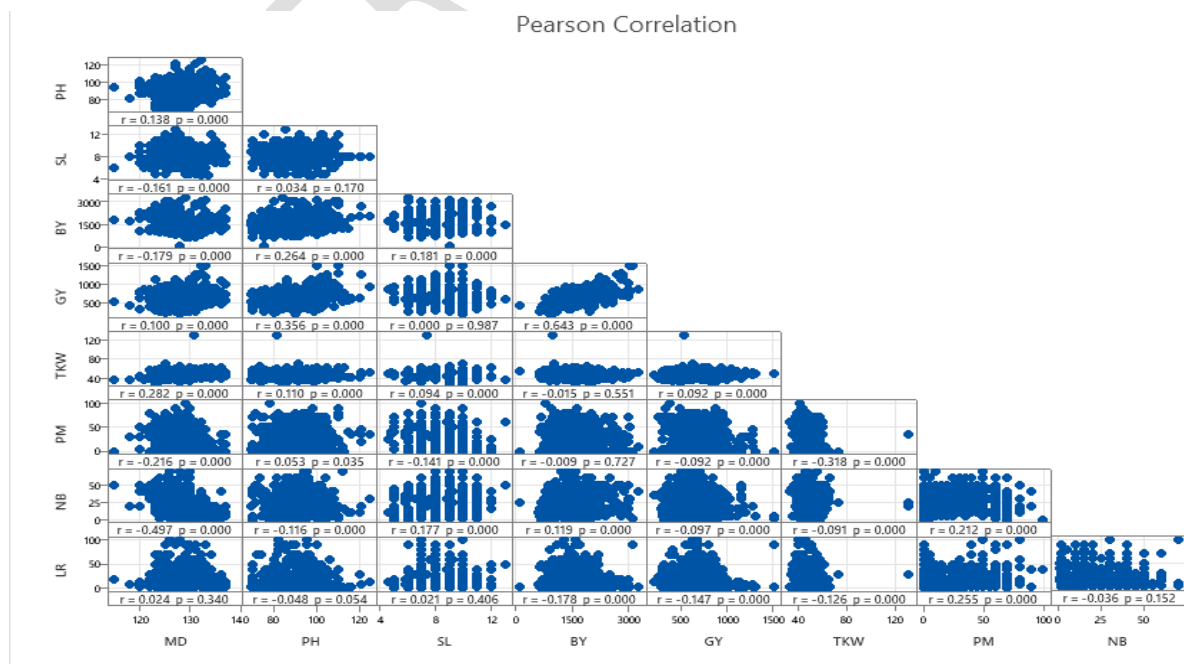


Fig. 4. Correlation coefficients between grain yield and all other studied traits overall the two growing seasons.

ACI values ranged from 0.1 to 37, 2.0 to 35.0 and 3.0 to 62.5 powdery mildew, net blotch and leaf rust, respectively (Table 8). Regarding powdery mildew disease, three tested barley genotypes i.e. Line 114, Line 112 and Line 30 showed low values of ACI were 0.1, 1.3 and 1.6 respectively. On the other hand, four barley genotypes i.e. Line 23, Rihane-03, and Giza 123, showed the highest values of ACI i.e. 30.0, 31.3, and 37.5, respectively (Table 8).

Table 8. Final severity (%) of 14 barley genotypes to powdery mildew (PM), net blotch (NB) and leaf rust (LR), along with the average coefficient of infection (ACI), country average relative percentage attack (CARPA) and relative resistance index (RRI).

Genotype No.	ACI			CARPA			RRI		
	PM	NB	LR	PM	NB	LR	PM	NB	LR
Line 114	0.1	10.0	4.0	0.27	38.12	6.4	8.98	5.56	8.42
Line 67	26.3	16.3	8.7	70.13	62.21	13.92	2.69	3.40	7.75
Line 112	1.3	28.8	4.3	3.47	109.92	6.88	8.69	0.89	8.38
Line 117	10.0	25.0	4.0	26.67	95.42	6.40	6.60	0.41	8.42
Line 116	16.3	21.3	4.0	43.47	81.30	6.40	5.09	1.68	8.42
Line 29	26.2	35.0	3.0	69.87	133.59	4.80	2.71	3.02	8.57
Line 23	30.0	20.0	13.8	80.00	76.34	22.08	1.80	2.13	7.01
Line 24	8.8	8.8	6.5	23.47	33.59	10.40	6.89	5.98	8.06
Line 30	1.6	2.0	3.0	4.27	7.64	4.80	8.62	8.31	8.57
Line 22	13.8	16.3	13.8	36.80	62.22	22.08	5.69	3.40	7.01
Line 12	28.8	20.0	18.7	76.8	76.34	29.92	2.09	2.13	6.31
Line 50	10.0	11.5	10.0	26.67	43.90	16.00	6.60	5.05	7.56
Rihane-03	31.3	18.8	15.0	83.47	71.76	24.00	1.49	2.54	6.84
Giza 123	37.5	26.2	62.5	100	100	100	0	0	0

Net blotch, three of the tested barley genotypes showed low values of ACI i.e. Line 30, Line 24, and Line 114 were recorded 2.0, 8.8., and 10.0, respectively. Leaf rust, the seven barley genotypes i.e. Line 29, and Line 30, displayed the lowest values of ACI i.e. 3.0, and 3.0, respectively (Table 8). As for, country Average Relative Percentage Attack (CARPA) of the tested barley genotypes showed the values, data in the same Table (36) run in a parallel line with those previously mentioned with ACI values. Concerning the Relative resistance index (RRI), data presented in Table 8 revealed the presence of one of the tested barley genotypes i.e. line 30 showed RRI acceptable and desirable (RRI) to three tested disease; powdery mildew, net blotch, leaf rust.

4. DISCUSSION

The present investigation endeavored to elucidate the genotypic diversity among 132 barley genotypes subjected to abiotic and biotic stress conditions at Sakha Agricultural Research Station. Exemplified by elevated humidity and increased precipitation, these conditions intensified the susceptibility to pathogenic invasion. Extensive genotypic variance was detected, particularly beneficial for the isolation of elite genotypes characterized by superior agronomic traits and heightened resilience to biotic stressors.

Analysis of variance (ANOVA) substantiated a non-trivial variance across the genotypes for all agronomic traits under investigation. A statistically significant genotype-environment interaction indicated the imperative for multi-seasonal evaluations to accurately assess yield stability. Noteworthy is the amplitude of phenotypic variance exhibited for foliar diseases such as powdery mildew, net blotch, and leaf rust, thereby signifying the feasibility of discriminative genotype selection for subsequent breeding programs.

Inherent genetic variance constituted the predominant contributor to the observed phenotypic dispersion among the cultivars for all evaluated traits. The genotypic coefficient of variation (GCV) demonstrated marginal divergence from the phenotypic coefficient of variation (PCV), thereby implying a minimal environmental perturbation on phenotypic manifestations, corroborated by **Katiyar [18]**.

Heritability estimates adhered to the categorization schema posited by **Johnson [19]**, with values exceeding 80% deemed exceptionally high. Genetic Advance as a Percentage of Mean (Ga%) delineated a stratification into low, moderate, and high categories, in alignment with **Johnson [19]**. Elevated Ga% estimates were observed particularly for biological yield, grain yield, powdery mildew, net blotch, and leaf rust, concordant with the findings of many researchers **[20, 21, 17, 11]**. Selection indices, as explicated by **Falconer and Mackay [22]**, indicated that both heritability and genetic advance estimates for biological yield, grain yield, powdery mildew, net blotch, and leaf rust were prodigiously high, implying a preponderance of additive gene action, corroborated by **Iannucci [23]**.

Correlational matrices revealed a direct association between days to maturity and several agronomic traits such as plant height, spike length, grain yield, biological yield, and 1000-grain weight. This affirms the assertion that late maturity is conducive to enhanced dry matter accumulation and, concomitantly, grain yield, as substantiated by **Msundi [24]**.

Principal component analysis (PCA) was deployed, revealing the first four principal axes with eigenvalues exceeding unity, accounting for 71% of the extant phenotypic diversity among the barley cultivars. The biplot analysis, mapping genotypes and traits onto the first two principal components, accounted for 48% of the cumulative variance and revealed acute and obtuse angles, indicative of positive and negative correlations among agronomic traits, respectively. High-yielding genotypes were explicitly identified, thereby informing future targeted breeding strategies for the amelioration of yield and disease resistance.

The results of ACI, RRI and CARPA analysis suggested that, the tested barley genotypes had a variety of genetic based on the varying disease reactions to three tested disease that were detected among them. One tested barley genotypes i.e. Line 30 it may be extrapolated, only exhibited a high level resistance of three tested disease resistance followed by Line 114.

5. CONCLUSION

In summary, this comprehensive study reveals significant genetic and phenotypic diversity among 132 barley genotypes assessed under specific environmental stressors. Statistical methods such as ANOVA and PCA validate the predominance of genetic variance as a key contributor to phenotypic variability. The research underscores the importance of heritability and genetic advance as robust indicators for successful selective breeding. High-yielding and disease-resistant genotypes have been identified, offering valuable insights for future agricultural strategies. The study thus constitutes a critical contribution to agronomic research, with implications for enhancing both yield and disease resistance in barley cultivation.

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