

Article

Adaptation and Grain Yield Stability Analysis of Winter Wheat Cultivars with and Without Fungicides Treatment from National Variety Trials in Sweden

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Abstract: The multi-environment evaluation of wheat genotypes for grain yield is an integral part of germplasm enhancement since it plays a pivotal role in sustainable production. A total of 178 winter wheat cultivars were evaluated across 20 environments in Sweden from 2016 to 2020, with 52 to 59 cultivars tested per year as part of the Swedish National Trials (Sverigeförsöken). The genotypes were evaluated for grain yield performance with and without fungicide treatments. Additive main-effects and multiplicative interaction (AMMI) and genotype plus genotype-by-environment interaction (GGE) biplot methods were explored to estimate the genotype-by-environment interaction (GEI) for grain yield performance. ANOVA revealed a significant variation between treatments, genotypes in all years, and GEI in all years except 2018. The majority of the explained variance came from the environment, with a range of 61–88% across the five-year trial. The 20 sites were grouped into two to four mega-environments in the yearly studies. From the fungicide-treated trials, G 0512LT3, Informer, SG SU1563-15, LG Imposanto, and Pondus were identified as the most stable and high-yielding cultivars each year. From the fungicide-untreated trials, Informer, Ancher Greece, RGT Saki, and Pondus were the best-performing cultivars and could be good candidates for organic wheat cultivation.

Keywords: wheat; yield; AMMI; GGE biplot; GEI; stability; organic agriculture



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1. Introduction

Wheat is one of the major cereal crops cultivated in Sweden, and, although with substantial fluctuation, its production has increased through the 1970–2019 period, ending at 3.48 million tonnes in 2019 [1]. Winter wheat is the primary wheat type grown in the country, and its cultivation has recently increased. For instance, in the autumn of 2020, 457.9 thousand hectares were planted with winter wheat in Sweden, which was 12, 19, and 29% larger compared to 2019, the average of the previous five and ten years, respectively [2]. The report indicated that this is by far the most extensive area coverage for winter wheat in Sweden, and particularly, the Västra Götaland and Västmanland counties made a remarkable increment with 23 and 42% more than the corresponding area in the previous autumn.

Phenotypic expression is a function of the genotype, the environment, and phenotypic response variation in genotypes to different environments, also known as the genotype-by-environment interaction (GEI) [3]. The environment includes soil fertility, temperature, the presence of biotic and abiotic influences, and other factors [4]. Breeding for optimum genetic gain will be possible if only a suitable test environment is utilized. However, the GEI makes the selection process difficult in identifying the best and most stable genotypes for cultivar recommendation [5]. The GEI is the most important but challenging issue for

plant breeders in developing high-yielding and stable cultivars [6]. In this case, multiple environment trials (MET) play a crucial role in quantifying the GEI effect and giving the relative performance of genotypes across environments [7,8]. Plant breeders conduct MET for several years to identify stable superior cultivars for particular environments and to delimit mega-environments. Due to the GEI, genotypes perform differently in various locations and years. Hence, specific adaptations and growing regions or mega-environments must be identified for a particular cultivar to optimize its productivity. Mega-environment is a crop species' growing region, not necessarily adjacent to a homogeneous environment that causes similar cultivars to perform best [9]. A cultivar is said to be stable when the GEI effect is non-significant across specific environments, and the genotype gives a similar yield (i.e., either low or high) across environments [10]. On the other hand, adaptability is when a cultivar adjusts to a particular environment to give a high yield but low in another environment [11].

The MET experiments include the following objectives: I) quantifying the magnitude of the GEI; II) evaluate the adaptability and stability of cultivars; III) delimit mega-environments to establish the relations between genotypes, environments, and the GEI simultaneously; and IV) predict the productivity of cultivars to particular environments that enables a precise selection for subsequent cropping cycles [12]. Consequently, appropriate methods are necessary to quantify the GEI since any inefficiency will create problems in selecting cultivars with the best performance in different environments [13].

Additive main-effects and multiplicative interaction (AMMI) and genotype main-effect plus genotype-by-environment interaction (GGE) biplot methods are the two most frequently used statistical approaches in GEI analysis. The AMMI model [14,15] combined the analysis of variance (ANOVA) with principal components for better exploitation of the GEI, its causes and consequences [16–18]. In AMMI, genotype and environment main effects are extracted using a two-way additive ANOVA model followed by the principal component analysis (PCA) that is applied to the ANOVA residuals, which includes the GEI. Yan et al. [19] proposed a biplot method, called GGE biplot, based on singular value decomposition (SVD) of environment-centered or within-environment standardized genotype by environment data, which displays based on the two sources of variation (i.e., the genotype and the GEI). The GGE biplot considers both the genotype main effects and GEI effects that are the basics during the GxE analysis in MET experiments [20,21]. The only difference between the AMMI and GGE biplot analysis comes at the initial stage, where the earlier method separates genotype from GEI while the latter analyzes both the genotype and GEI effects together, and finally, both methods build biplots for analysis interpretation [22]. The two analysis approaches depict their complicity despite cynicism among some authors on the effectiveness of AMMI and GGE biplots in depicting the adaptive responses of genotypes over environments [6,18,23,24]. However, such differences do not indicate the superiority of either method: the AMMI analysis method provides phenotypic stability, genetic divergence between genotypes, and environments with optimal performance, while the GGE biplot provides delineation of mega-environments along with genotypes with optimal performance [22,25].

The Swedish national trial (Sverigeförsöken, <https://sverigeforsoken.se/s> (accessed on 1 September 2023)) conducts multi-environment and year experiments of several crop cultivars, including winter wheat. The national trial works together with the Swedish University of Agricultural Sciences (SLU), Swedish Agricultural Research Foundation (SLF), crop and plant protection companies and advisers, farmers, and authorities aimed at creating an appropriate platform for plant breeders and other interested professionals. The national trial tests the cultivar's performance for several traits with agronomic importance, including yield, quality, and disease resistance traits. Genotype performance without any chemical treatment is one of the objectives of the MET, which is aimed at recommending cultivars appropriate for organic farming. This study investigated the GEI to determine the most adaptable and stable cultivars across environments based on the

AMMI and GGE biplots. To achieve this, 178 winter wheat cultivars treated and untreated with fungicides were tested in 20 experimental sites for five consecutive years.

2. Materials and Methods

2.1. Plant Materials and Experimental Design

The five-year data (2016–2020) of winter wheat from the Sweden national trials was used for this study. A total of 178 cultivars were tested within the five years in which 54–59 genotypes were included every year. The majority of these cultivars are released from Lantmännen Lantbruk and Scandinavian Seed AB but also encompassed from other sources, including Nordic Seed, RAGT, Syngenta, and Saaten-Union GmbH. Genotypes were tested in 20 different trial sites from the south, middle, and north growing zones of Sweden, in which only 10 were included every year (Table S1). These regions are labeled as the most important growing zones in Sweden [26]. The trial sites and their details, including soil type and weather conditions, can be found on the Swedish national trial webpage: <https://sverigeforsoken.se/s> (accessed on 1 September 2023). The appropriate experiments could be achieved by inserting the year followed by the crop type (winter wheat) and the particular field trial represented by the plant images shown on the map.

The trials were laid out in a split-plot design with two replicates in which the fungicide-treated and untreated plots were the main plot factor, and cultivars were then arranged in alpha lattice design in each treatment with two replications and eight sub-blocks. The plots had an area of 14 m², but only 12 m² were harvested.

2.2. Statistical Analysis

Given the variation in cultivars tested each year and the differing trial locations (Table S1), this study focuses solely on year-wise analyses, excluding across-year comparisons of cultivars.

Analysis of variance was estimated for grain yield using the *aoov* (Fit an analysis of variance model by a call to *lm*) package in the R environment [27] by including genotypes, treatments, environments, replications, and blocks. Additive main-effects and multiplicative interaction (AMMI) and genotype plus genotype-by-environment interaction (GGE) biplot methods were utilized to estimate the genotype-by-environment interaction, yield stability, and cultivars' performance in a particular environment. The AMMI model [14] utilized for specific environment adaptation and mean performance, while the GGE biplot [19] is used to estimate the which-won-where in mega-environments and yield stability analysis.

The AMMI analysis was exploited to estimate the overall performance and adaptation of genotypes across environments with the following model according to Pacheco et al. [28] and the result was displayed in biplots.

$$Y_{ij} = \mu + g_i + e_j + \sum_{n=1}^N \tau_n \gamma_{in} \delta_{jn} + \varepsilon_{ij}$$

where Y_{ij} is the grain yield from the i -th genotype ($I = 1, \dots, I$) in the j -th environment ($j = 1, \dots, J$); μ is the general mean while g_i and e_j represents the genotype and environment deviations from the general mean, respectively; τ_n is the eigen value of the interactive principal component analysis (IPCA) axis n while γ_{in} and δ_{jn} are the genotype and environment IPCA scores for axis n , respectively; N is the number of IPCs retained in the model, and ε_{ij} is the residual error. IPCA refers to the analytical process within AMMI that decomposes the interaction component of the data into principal components to capture patterns in the interaction effect between genotypes and environments. IPC is the actual component extracted from the interaction matrix in the AMMI model. Each IPC corresponds to a factor or dimension that captures a specific portion of the genotype-by-environment interaction.

The GGE biplots were applied to delineate mega-environments and estimate genotype stability based on the model given by Frutos et al. [29] with the following formula:

$$Y_{ij} - \mu - \beta_j = \sum_{k=1}^t \lambda_k \xi_{ik} \eta_{jk}$$

where Y_{ij} is the grain yield of the genotype i in the environment j ; μ is the overall mean and β_j is the environment (column) main effect; t is the number of SVD (singular value decomposition) axes retained in the model; λ_k is the singular value for the SVD axis k ; ξ_{ik} is the singular value of the i th genotype for the SVD axis k ; η_{jk} is the singular value of the j th environment for the SVD axis k .

The stability analysis of grain yield was estimated based on the environment and genotype interaction with a linear regression model using the formula:

$$Y_{ij} = \mu + d_i + (1 + \beta_i)e_j + \delta_{ij} + \varepsilon_{ij} \quad (1)$$

where Y_{ij} is the average phenotypic value of the i th genotype in the j th environment; μ is the general mean; d_i and e_j stand for the effect of the i th genotype and j th environment, respectively; $1 + \beta_i$ is the regression of Y_{ij} in e_j ; δ_{ij} is the deviation of the regression for the i th genotype in the j th environment, and ε_{ij} is the error.

Software packages, including genotype-by-environment interaction analysis in R (GEA-R) [28] and Multi-Environment Trial Analysis with R (META-R) [30] were used to exploit the GEI analysis. Various variance components were estimated using the lme4 package [31] applying the following model:

$$Y_{ijklm} = \mu + \text{Trt}_m + \text{Gen}_i + \text{Env}_j + \text{Gen}_i \times \text{Env}_j + \text{Gen}_i \times \text{Trt}_m + \text{Env}_j \times \text{Trt}_m + \text{Gen}_i \times \text{Env}_j \times \text{Trt}_m + \text{Rep}_k(\text{Env}_j) + \text{Block}_l((\text{Rep}_k(\text{Env}_j))) + \varepsilon_{ijklm}$$

where Y_{ijklm} is the phenotypic value (yield) for the i -th genotype, j -th environment, k -th replicate, l -th block, and m -th treatment. μ is the overall mean effect. Trt_m is the effect of the m -th treatment (fungicide-treated vs. untreated). Gen_i is the effect of the i -th genotype, and Env_j is the effect of the j -th environment. $\text{Gen}_i \times \text{Env}_j$ is the interaction between the i -th genotype and the j -th environment. $\text{Gen}_i \times \text{Trt}_m$ is the interaction effect between the i -th genotype and the m -th treatment. $\text{Env}_j \times \text{Trt}_m$ is the interaction effect between the j -th environment and the m -th treatment. $\text{Gen}_i \times \text{Env}_j \times \text{Trt}_m$ is the three-way interaction effect between the i -th genotype, j -th environment, and m -th treatment. $\text{Rep}_k(\text{Env}_j)$ is the effect of the k -th replication nested within the j -th environment. $\text{Block}_l((\text{Rep}_k(\text{Env}_j)))$ is the effect of the l -th block nested within the k -th replication and j -th environment. ε_{ijklm} is the residual error

The normality of residuals was assessed using frequency distribution plots and quantile-quantile (Q-Q) plots generated with the package ASReml v. 4 [32]. The analysis was conducted year-wise, including the fungicide-treated and untreated experiments, since new cultivars were added every year. To display on biplots, cultivar number IDs were used based on the data available in Sweden's national trial (<https://sverigeforsoken.se/s> (accessed on 1 September 2023)). For environments, only numbers were used excluding 0's; for example, 001 and 002 environments on the webpage (<https://sverigeforsoken.se/s> (accessed on 1 September 2023)) were labeled as 1 and 2 on the biplots, respectively. Trials can be found on the website after selecting the year (2016–2020), crop (autumn wheat), and the code L7-0101-field trials (001–010).

3. Results

3.1. Analysis of Variance and AMMI Biplots

The normality of the year-wise data entries was confirmed through normal distribution analysis and quantile-quantile (Q-Q) plots, which demonstrated a close alignment between the expected and observed residuals (Figure S1A–E). The analysis of variance revealed a

highly significant difference in grain yield between the cultivars and the two treatments (Table 1). The GE interaction was highly significant in all years except in 2018, which was non-significant. Environment dominantly explained the majority of grain yield variance with a range of 61–88% across the year’s trial, followed by the GEI, while genotypes contributed the least (Table 2). In AMMI analysis, the first two interactive principal components significantly explained most of the total variance in most year’s analyses, but up to AMMI9 was required to capture the total variance (Tables S2–S5).

Table 1. ANOVA of grain yield estimated from 178 winter wheat cultivars, including treatments and environments. E = environment; G = genotype; Trt = treatment; DF = degree of freedom; SS = sum square; MS = mean square; F = F test statistics; EV = explained variance. ** significant; *** highly significant; NS non-significant.

Year		DF	SS	MS	F	p-Value
2016	G	57	702	11.7	9.167	<0.001 ***
	E	9	22,252	2781.5	2177.877	<0.001 ***
	G×E	513	1424	3.0	3.303	<0.001 ***
	Trt	1	174	174.4	136.573	<0.001 ***
	Residuals	2092	2672	1.3	-	-
2017	G	53	815	15.1	6.383	<0.001 ***
	E	9	11,163	1240.3	524.842	<0.001 ***
	G×E	477	1760	3.6	1.451	<0.001 ***
	Trt	1	686	685.7	290.135	<0.001 ***
	Residuals	2018	4769	2.4	-	-
2018	G	58	706	12	8.262	<0.001 ***
	E	9	14,053	1561.4	1130.542	<0.001 ***
	G×E	522	758.4128	1.4529	0.92664	0.81417 NS
	Trt	1	11	11.3	7.810	<0.00524 **
	Residuals	2326	3369	1.4	-	-
2019	G	54	1283	23.8	14.543	<0.001 ***
	E	9	16,551	1839	1125.616	<0.001 ***
	G×E	486	1524	3.1	1.248	<0.001 ***
	Trt	1	2147	2147.4	1314.363	<0.001 ***
	Residuals	2130	3480	1.6	-	-
2020	G	57	1290	22.2	19.015	<0.001 ***
	E	9	27,453	3050.3	2607.869	<0.001 ***
	G×E	513	1008	1.9	1.696	<0.001 ***
	Trt	1	205.084	3.15514	12.67328	<0.001 ***
	Residuals	2284	2672	1.2	-	-

Table 2. Genotype, genotype by environment, and other sources of variations with their explained variation.

Source	Years				
	2016	2017	2018	2019	2020
Genotype	1.6358318	3.4125225	3.1980683	4.734364	3.494132
Env	87.7523246	60.9966980	76.5970001	74.63924	87.02654
Genotype:Env	4.1392496	6.6509106	1.6903638	5.596941	1.788598
Genotype:Trt	0.2888352	0.1388348	0.1114204	0.482911	0.076533
Env:Trt	0.4414176	10.2452944	1.9304268	6.031131	2.218385
Genotype:Env:Trt	0.8661147	0.0915487	0.0000000	0.509868	0.442382
Residual	3.6667502	8.3943257	6.6456719	4.157752	3.475491

On the provided biplots, genotypes are denoted in numbers (ID) written in blue, while environments with red on top of the green vector lines drawn from the center of biplots specified their interactive forces with cultivars. The farther the environments are located from the center, the higher the interactive force they have exerted on cultivars. The AMMI1

model was drawn with the IPCA1 (factor 1) and average grain yield scores of cultivars from each year (Figure 1).

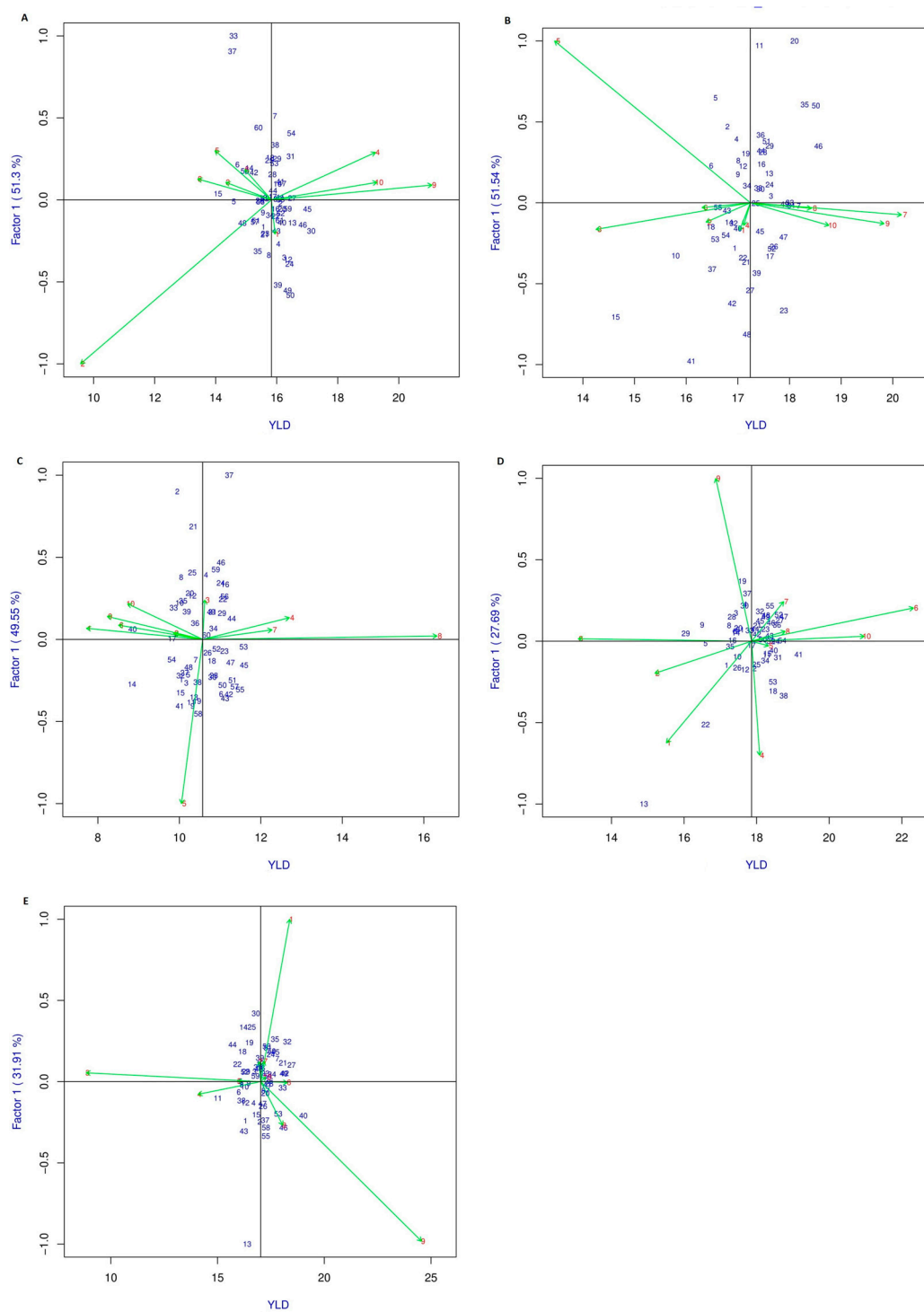


Figure 1. AMMI1 biplots with average grain yield vs. IPCA1 (factor 1) for fungicide-treated trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D), and 2020 (E). Yield scores are based on Kg/plot. Genotypes are denoted with numbers in blue, while environments with red are connected in green lines from the plot center.

Between 2016 and 2020, winter wheat cultivars were evaluated across diverse environments to identify high-yielding, stable genotypes and their specific adaptability.

The results highlight notable performances across genotypes, environments, and genotype-by-environment interactions. For instance, in 2016, genotypes 30 (G 0512LT3), 45 (Stinger), 46 (SJ 1378001), and 27 (Etana) were identified as the top performers in mean yield. Among the environments, high productivity was observed at sites characterized by optimal agronomic and climatic conditions, including those environments 7 (St. Deciduous farm), 9 (Russelbacka, Järpås), and 10 (Multorp, Sal) (Figure 1A).

In 2017, 46 (Informer), 50 (Jonas), and 35 (Ancher) were the top three high performing cultivars and environments; 7 (St. Deciduous farm), 9 (Russelbacka, Järpås), and 10 (Multorp, Sal) were the best productive trial sites than others (Figure 1B). In the year 2018, 45 (Jonas), 53 (SG SU1563-15), and 55 (Kron) were the top three cultivars with the best mean yield productivity, while 8 (Klostergården), 4 (Russelbacka, Järpås), and 7 (St. Deciduous farm) were the three top productive environments (Figure 1C). In 2019, 41 (RGT Saki), 38 (Terence), 27 (LG Imposanto), and 54 (LG Quadrant) were the top performers in mean yield, while 6 (Russelbacka, Järpås) and 10 (Brunnby farm) were the two best productive environments than others (Figure 1D). Cultivars such as 40 (Pondus), 27 (Kaldi), and 32 (RGT Koi) were the top three average yield performers in the year 2020, and 9 (Kiplingeberg 7) was the most productive environment (Figure 1E).

The angle between environments notifies their relationship in response to cultivar performance, in which the acute and obtuse angles indicated the positive and negative correlations, respectively, while environments with no correlation were represented by a right angle. For instance, in the year 2016, environment 2 (Sandbygårdsvägen 5) had a negative correlation with all other environments except with 1 (Russelbacka, Järpås) with a positive correlation. Environment 10 (Klostergården) had a positive correlation with 5 (Stora Uppåkravägen 117), no correlation with 6 (Skolgatan 37), and a negative correlation with other environments (Figure 1A). Trial sites found at the south zones of Sweden such as 1 (Stora Uppåkravägen 117), 2 (Sandbygårdsvägen 5), 3 (Skolgatan 37), and 4 (Ga Malmövägen 459) clustered together with positive correlations in 2017 (Figure 1B). The other trial sites from the middle and north zones, including 7 (St. Deciduous farm), 8 (Klostergården), 9 (Russelbacka), and 10 (Järpås Multorp, Sal), made another cluster with a positive correlation in the same year.

Cultivars were tested without fungicide treatment to select cultivars with stable and high-yield performance appropriate for organic farming. The AMMI1 model successfully identified highly performing genotypes in the five years. The result indicated that the cultivar Informer was the top-performing genotype in both years, 2016 and 2017, while 24 (Stinger), 41 (RGT Saki), and 40 (Pondus) were the best-performing cultivars in 2018, 2019, and 2020, respectively (Figure S2A–E).

The AMMI2 model was used for the specific adaptation of cultivars in a particular environment and the relationship between environments and cultivars' performance (Figure 2). In the year 2016, 12 (Ellen), 8 (Ceylon), and 15 (Norin) were best adapted to environment 1 (Russelbacka, Järpås). Cultivars such as 50 (Igloo), 39 (R 11333), and 49 (WPB Nigel) on environment 2 (Multorp, Sal); 33 (KWS Lili), 37 (RW41350), and 7 (Hereford) on environments 3 (Ga Malmövägen 459) and 6 (Skolgatan 37); 6 (Cubus), and 25 (RGT Hasseth) on environment 4 (Sandbygårdsvägen 5); 54 (RW41498), 31 (W 237) and 53 (R 11559) on environment 5 (Stora Uppåkravägen 117); 18 (Croft) and 41 (Hacksta) on environment 9 and 60 (LGWD 11-132070) and 45 (Stinger) on environment 10 (Klostergården) were the most productive cultivars along with their best adapted environments (Figure 2A). This year, genotype 11 (RGT Reform) was the most stable cultivar, followed by 15 (Norin), 47 (KWS Finn), and 22 (Festival) across the 10 tested trial sites. In the year 2017, genotypes 15 (Norin), 42 (Nord 11044/032) and 37 (KWS Ahoy) on environments 1 (Stora Uppåkravägen 117) and 4 (Ga Malmövägen 459); 20 (Gunilla), 11 (RGT Reform) and 5 (Olivine) on environment 5; 10 (Internship) and 14 (Nordh) on environments 2 (Sandbygårdsvägen 5) and 3 (Skolgatan 37); 23 (Rockefeller) on environments 7 (St. Deciduous farm) and 10 (Multorp, Sal); and 3 (Bronze) and 25 (Greece) were adapted well on environments 8 (Klostergården) and 6 (Brunnby farm) (Figure 2B). Genotypes, including 31 (KWS Ker-

rin) and 30 (WPB Nigel), were the most stable cultivars across tested environments in 2017. In 2018, genotypes such as 59 (RGT Blossom), 35 (KWS Talent), and 20 (Rockefeller) on environments 2 (Sandbygårdsvägen 5), 4 (Russelbacka, Järpås), 7 (St. Deciduous farm), 8 (Klostergården), and 9 (Gäsmesta 202); 24 (Stinger) on environment 10 (Brunnby farm); and 37 (Ancher), 2 (Ellvis), and 21 (Hacksta) were well adapted on environment 6 (Skepparslövsvägen 258) (Figure 2C). Genotype 60 (RGT Specialist) was a widely adapted cultivar across the 10 trials in 2018, followed by 7 (Frontal) and 47 (Bright). In 2019, the best-adapted cultivars in a particular environment were genotype 18 (Hacksta) on environment 1 (Ga Malmövägen 459); 13 (Norin) on environment 4 (Stora Uppåkravägen 117); 3 (Mariboss), 33 (Johanna), and 48 (SW 14527) on environments 6 (Russelbacka, Järpås), 7 (St. Deciduous farm) and 8 (Klostergården); and 29 (Axioma) and 4 (RGT Reform) on environments 3 (Isgrannatoprsvägen 134) and 10 (Brunnby farm). Cultivar 5 (Cubus) was the most stable genotype, while the effect of environment 5 (Astranna Gård 1, Flo) was close to average across tested cultivars (Figure 2D). Most tested cultivars exhibited average values for environments in 2020 and were crowded at the center of the plot (Figure 2E). However, some genotypes had a specific environmental adaptation such as 55 (SW 16100) on environment 9 (Kiplingeberg 7); 30 (SG SU1563-15) on environments 2 (Sandbygårdsvägen 5) and 7 (St. Deciduous farm); 8 (Ceylon) on environments 3 (Stora Uppåkravägen 117), 4 (Stureholmsvägen 177) and 10 (Brunnby farm); and 39 (LG Quadrant) on environment 1 (Gislöv Nobelvägen 79).

AMMI2 biplots were applied to fungicide untreated cultivars to highlight those suitable for organic farming adapted to a certain environment (Figure S3). In 2016, the cultivar 13 (Power) was the best-adapted genotype in environments 1 (Russelbacka, Järpås) and 2 (Multorp, Sal) while 59 (Informer) in 4 (Sandbygårdsvägen 5) and 10 (Klostergården); 54 (RW41498) in environments 3 (Skolgatan 37), 5 (Goal 6) and 9 (Russelbacka, Järpås) (Figure S3A). In 2017, the specific adaptation analysis indicated cultivars 42 (Nord 11044/032) and 52 (Bright) in environments 1 (Stora Uppåkravägen 117) and 4 (Ga Malmövägen 459); 35 (Ancher) in environment 5 (Goal 6); 27 (Stinger) and 28 (SJ 1378001) in environment 9 (Russelbacka, Järpås); and 50 (Jonas) in environments 6 (Brunnby farm), 8 (Klostergården) and 10 (Multorp, Sal) (Figure S3B). In the year 2018, only limited cultivars such as 25 (KWS Finn), 26 (Igloo), 37 (Ancher), and 46 (a cultivar from Lantmännen) showed a particular adaptation for all trial sites except for environments 5 (Stora Uppåkravägen 117) and 8 (Klostergården) (Figure S3C).

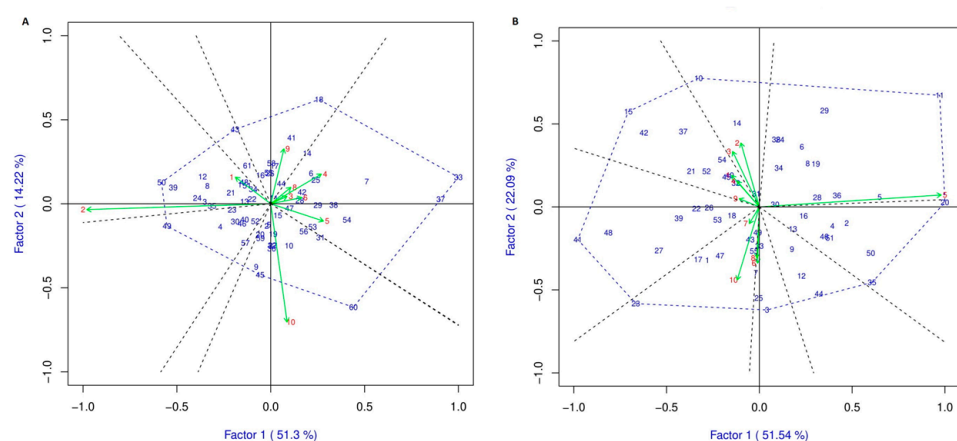


Figure 2. Cont.

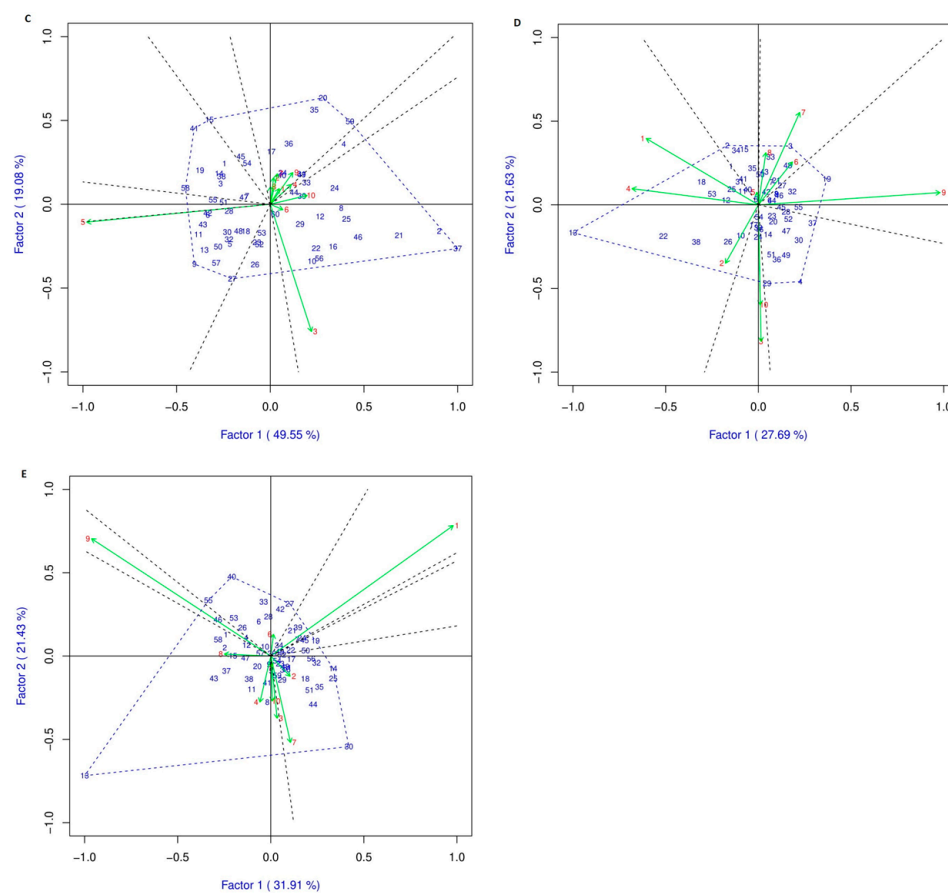


Figure 2. AMMI2 biplots of grain yield with the IPCA1 (factor 1) and IPCA2 (factor 2) components for trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D), and 2020 (E).

3.2. Mega-Environments and Which-Won-Where

The GGE biplot method was applied to estimate the mega-environments and well-performed genotypes on the delineated regions or mega-environments (Figure 3). The polygons were made with vertices using genotypes located farthest away from the origin of the biplot in all directions. In 2016, the 10 trial sites were grouped into four mega-environments (Figure 3A). Three mega-environments were established from the 10 tested environments in the year 2017 (Figure 3B). Three mega-environments were formed in the year 2018 (Figure 3C). Three mega-environments were formed in 2019 which the first included environments 1 (Ga Malmövägen 459), 2 (Sandbygårdsvägen 5), 4 (Stora Uppåkravägen 117), 5 (Astranna Gård 1, Flo), 6 (Russelbacka, Järpås), 7 (St. Deciduous farm), and 8 (Klostergården) with well-adapted cultivars such as 41 (RGT Saki), 27 (LG Imposanto), and 31 (Kaldi) (Figure 3D). In 2020, all trial sites except 1 (Gislöv Nobelvägen 79) were included in a mega-environment in which the two cultivars, 40 (Pondus) and 27 (Kaldi), from Nordic seed, were the best productive genotypes for the region (Figure 3E).

Mega-environments were also estimated from cultivar trials untreated with fungicides (Figure S4). The ten experimental sites were grouped into 2, 4, 3, 3, and 2 mega-environments during the years 2016, 2017, 2018, 2019, and 2020, respectively (Figure S4A–E).

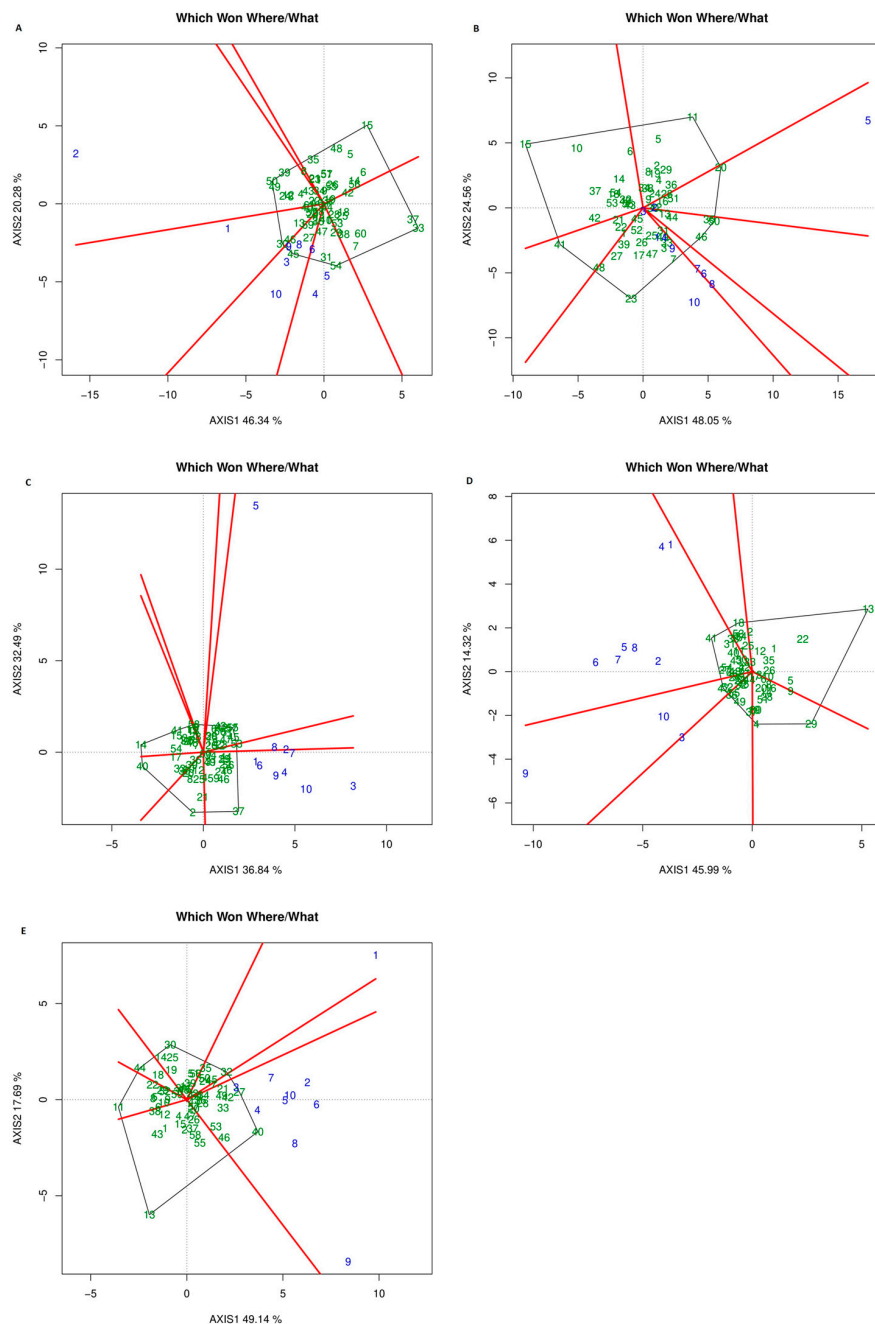


Figure 3. GGE biplot for delineation of mega-environments and the which-won-where analysis results for trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D), and 2020 (E).

3.3. Grain Yield Stability

The stability on the average yield performance of cultivars across trials was plotted with the average environment coordination (AEC) method, and the GGE biplots were built with environment-centered, non-scaled, and singular value partitioning (SVP) Figure 4). The analysis revealed that certain wheat cultivars maintained high yields and stability across diverse trials. For instance, in 2016, cultivars *G 0512LT3* and *Stinger* performed better across multiple environments, demonstrating both high yield and stability (Figure 4A). This resilience is essential for regions with unpredictable growing conditions, as cultivars that consistently perform well reduce the risk of yield loss, ensuring more reliable production. The current study also conducted a performance evaluation of cultivars under-treated and untreated conditions. In fungicide-free trials, cultivars such as *Informer* showed high yields and stability, as illustrated in Figure S5A for 2016, suggesting the suitability of the cultivar

for low-input and organic farming systems. Such cultivars are invaluable for regions aiming to reduce chemical inputs without compromising yields, supporting sustainable agricultural practices. In addition to year-specific stability, certain cultivars demonstrated broad adaptability across multiple locations and years. For example, in 2017, the cultivar Informer was again one of the most stable and high-performing genotypes, while Jonas led in yield but was slightly less stable (Figure 4B). By focusing on high-yielding cultivars that also display resilience, this study provides a valuable foundation for breeding programs globally. The findings emphasize the importance of cultivars that offer not only peak yield in favorable conditions but consistent performance across diverse and sometimes challenging environments.

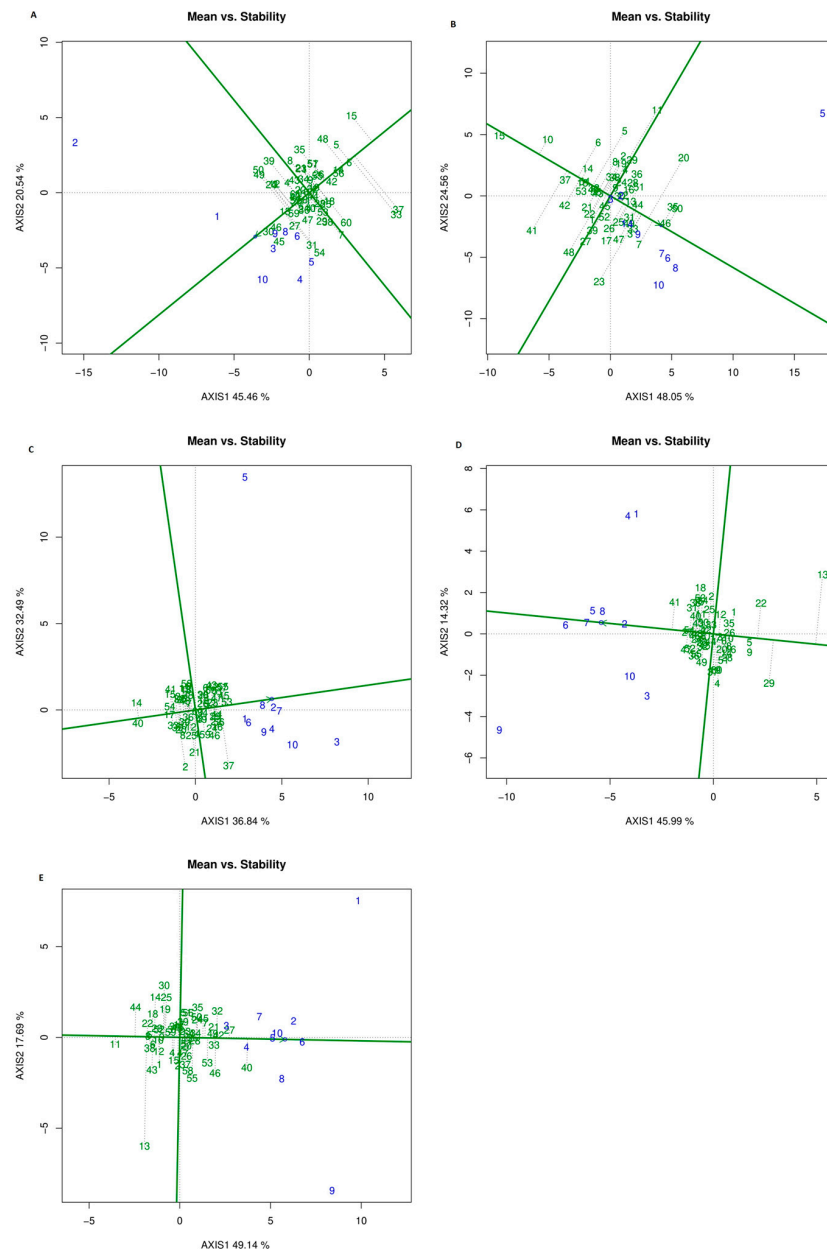


Figure 4. GGE biplot for mean vs. stability analysis results towards the ideal environment for trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D), and 2020 (E).

4. Discussion

This study specified a highly significant GEI existed in 178 winter wheat cultivars tested in 20 sites for five consecutive years from 2016 to 2020, except in the year 2018. This

leads us to the presence of significant performance variation in genotypes across tested environments and the immense importance of this study in quantifying the magnitude of the existing interaction, the adaptability of cultivars to a particular environment, and their stability. The year 2018 was the least productive, with non-significant differences across trial sites (Table 2). That year had a summer with prolonged drought that extremely affected the agriculture in Sweden [33] and up to 50% yield reduction was recorded in cereal crops [34].

Both the AMMI and GGE biplot analysis methods were successfully utilized in this study to quantify the existing GEI followed by detecting cultivar's adaptation to a particular environment, their stability across environments and delineation of mega-environments. The first two singular axes of the AMMI analysis usually capture the highest percentage of the variation/pattern of the data [13,16,35]. Likewise, the AMMI2 biplots captured the majority of the existing variation (50–74%) in the current year-wise study and illustrated the best yielding cultivars and environments in both fungicide-treated and untreated experiments. The AMMI1 biplots ranked cultivars and environments based on the grain yield performance across tested trial sites. For instance, Stinger, a cultivar from Scandinavian Seed AB, was 2nd best performing and stable genotype in 2016 (fungicide-treated) and the top yield performer in 2018 (fungicide-untreated). Informer, another cultivar from Scandinavian Seed AB, was the other top-performing genotype in 2017 from the fungicide-treated experiment and top-performing in both 2016 and 2017 from the fungicide-untreated experiment. One of the current challenging issues in organic farming is the average lower yield performance of cultivars [36,37] and increasing the yield in organic agriculture has been regarded as one of the foremost targets in Sweden [38]. Since approved crop protection products for organic agriculture are very limited [39], diseases and pests greatly affect the yield.

Consequently, organic farmers rely on plant cultivars/varieties suitable for organic agriculture [38]. The current study highlighted that the cultivar Informer in two years (i.e., 2016 and 2017) and Stinger, RGT Saki, and Pondus in the years 2018, 2019, and 2020, respectively, outperformed the other cultivars from the experiments with no fungicide treatment. This suggests that these cultivars could be highly suitable for farmers and other producers aimed at organic wheat cultivation.

The two principal components of the GGE biplots explained in a close range of variations to the AMMI2 in which the latter was slightly higher agreed with previous reports [22]. The GGE biplot is an effective method for the which-won-where analysis and demarcates the mega-environments [19,20,23]. Subdivision of growing regions into mega-environments is pivotal to exploiting the narrow adaptation of genotypes, resulting in a significant opportunity to increase yield [9,12]. The which-won-where analysis grouped the 20 national trial sites into three mega-environments in all years except in 2016, where environments were grouped into four mega-environments. Most of these trial sites were grouped into mega-environments based on their contiguity. For instance, in 2016, the two sites (Russelbacka, Järpås, and Multorp, Sal) were in one mega-environment, and these sites are found in close proximity to central Sweden while the other two trial sites (i.e., Sandbygårdsvägen 5 and Stora Uppåkravägen 117), found at the southern Sweden, included in the other mega-environment. The experimental site, Goal 6, found in central Sweden near Uppsala and far from the other trial sites, made its own mega-environment in the year 2017. Similarly, the 2019 trial grouped the two nearby trial sites (i.e., Brunnby farm and Hällby gårdsväg 13) in a mega-environment. The GGE biplot method was also utilized to determine the stability of cultivars across the studied environments. For a successful variety selection with wider adaptation, one should consider higher yield potential with lower GEI and if the variety gives high and stable performance across locations and for years, it is called a stable variety [40]. The stability and average yield performance of 178 winter wheat genotypes were plotted using the average environment coordination (AEC) method. A cultivar with the shortest length of projection from the AEC is generally the most stable genotype across tested environments [41]. Therefore, G 0512LT3, Jonas and

LG Imposanto from Lantmännen; Informer, SU1563-15, and Stinger from Scandinavian Seed AB; Pondus and Kaldi from Nordic Seed; and RGT Saki from RAGT were the most stable and high yielding cultivars screened in the five years study.

5. Conclusions

The study comprehensively evaluated the grain yield performance and stability of 178 winter wheat cultivars across 20 diverse environments in Sweden over five years. The results revealed the critical influence of genotype-by-environment interaction in determining yield variability, emphasizing the importance of tailored breeding programs. Notable high-performing and stable genotypes, such as Informer, RGT Saki, and Pondus, were identified under fungicide-free conditions, highlighting their potential for organic farming systems. The application of statistical models, such as AMMI and GGE biplots, facilitated the identification of mega-environments and genotype-specific adaptations, providing valuable insights for optimizing regional wheat cultivation strategies. These findings highlight the need for regionally adapted cultivars that offer resilience to environmental stresses while maintaining productivity.

The identification of cultivars suitable for organic farming aligns with the increasing global demand for environmentally friendly and low-input agricultural systems. Furthermore, the delineation of mega-environments serves as a valuable framework for breeding programs worldwide, enabling the efficient deployment of high-yielding, stable cultivars in diverse agro-ecological zones. By addressing critical challenges in crop production, such as the variability introduced by GEI and the need for disease-resistant genotypes, this study supports global food security initiatives and the transition toward sustainable crop management practices.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/agriculture14122229/s1>, Figure S1. Normality test based on the frequency distribution (top) and quantile-quantile (Q-Q) plots (bottom) of residuals for 178 winter wheat cultivars tested with and without fungicide treatment in 20 trial sites in 2016 (A), 2017 (B), 2018 (C), 2019 (D) and 2020 (E). Figure S2: AMMI1 biplots with average grain yield vs IPCA1 (factor 1) for fungicide untreated trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D) and 2020 (E). Yield scores is based on Kg/plot. Genotypes are denoted with numbers in green while environments with red connected in green lines from the plot center. Figure S3: AMMI2 biplots of grain yield with the IPCA1 (factor 1) and IPCA2 (factor 2) components for fungicide untreated trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D) and 2020 (E). Figure S4: GGE biplot for delineation of mega-environments and the “which-won-where” analysis results for fungicide untreated trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D) and 2020 (E). Figure S5: GGE biplot for mean vs stability analysis results towards the ideal environment for fungicide untreated trials in 2016 (A), 2017 (B), 2018 (C), 2019 (D) and 2020 (E). Table S1. List of varieties tested across different environments and years. Table S2. The Interactive principal components in the AMMI analysis for fungicide treated yield trial in 2016. Table S3. Interactive principal components in the AMMI analysis for fungicide treated yield trial in 2017. Table S4. Interactive principal components in the AMMI analysis for yield trial tested in 2018 with fungicide treatment. Table S5. The Interactive principal components in the AMMI analysis for yield trial with fungicide treatment tested in 2019. Table S6. The Interactive principal components in the AMMI analysis for yield trial with fungicide treatment tested in 2020.

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