Biplot analysis of multi-environment trial data: Principles and applications

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Yan, W. et Tinker, N. A. 2006. **Biplot analysis of multi-environment trial data: Principles and applications**. Can. J. Plant Sci. **86**: 623–645. Biplot analysis has evolved into an important statistical tool in plant breeding and agricultural research. Here we review the basic principles of biplot analysis and recent developments in its application in analyzing multi-environment trail (MET) data, with the aim of providing a working guide for breeders, agronomists, and other agricultural scientists on biplot analysis and interpretation. The review is divided into four sections. The first section is a complete but succinct description of the principles of biplot analysis. The second section is a detailed treatment of biplot analysis of genotype by environment data. It addresses environment and genotype evaluation from all perspectives. The third section deals with biplot analysis of various two-way tables that can be generated from a three-way MET dataset, which is an integral and essential part to a fuller understanding and exploration of MET data. The final section discusses questions that are frequently asked about biplot analysis. Methods described in this review are available in a user-friendly, interactive software package called "GGEbiplot".

Key words: biplot analysis; genotype by environment interaction; mega-environment; multi-environment trials

Yan, W. et Tinker, N. A. 2006. **Analyse par double projection des résultats des essais multi-environnementaux : principes et applications**. Can. J. Plant Sci. **86**: 623–645. L'analyse par double projection est devenue un outil statistique important pour la recherche en phytogénétique et en agronomie. Les auteurs passent en revue les principes de base de cette méthode et les progrès récents qu'a connus son application à l'analyse des données généalogiques multi-environnementales, l'intention étant de proposer un manuel pratique sur l'analyse par double projection et l'interprétation de ses résultats aux obtenteurs, aux agronomes et à d'autres scientifiques agricoles. L'article est divisé en quatre. La première partie donne une description complète mais succincte des principes de l'analyse par double projection. La deuxième explique en détail l'application de cette méthode aux données combinant génotype et environnement. Il y est question de l'évaluation du génotype et du milieu sous tous les angles. La troisième partie aborde l'analyse par double projection de tableaux à double entrée issus d'un jeu de données généalogiques multi-environnementales à triple entrée, et constitue un aspect intégral et essentiel à une solide compréhension et exploration de telles données. La dernière partie examine les questions que l'on se pose souvent sur l'analyse par double projection. Les méthodes décrites dans l'article sont disponibles sous la forme d'un logiciel interactif et convivial baptisé « GGEbiplot ».

Mots clés: Analyse par double projection, interaction entre le génotype et le milieu, méga-environnement, essais multi-environnementaux

Plant variety trials are routinely conducted to compare multiple genotypes in multiple environments (years and locations) for multiple traits, resulting in genotype by environment by trait three-way data. Variety trials provide essential information for selecting and recommending crop cultivars. However, variety trial data are rarely utilized to their full capacity. Although data may be collected for many traits, analysis may be limited to a single trait (usually yield) and information on other traits is often left unexplored. Furthermore, analysis of genotype by environment data is often limited to genotype evaluation based on genotype main effect (G) while genotype-by-environment interactions (GE) are treated as noise or a confounding factor.

GE has been a research focus among biometricians and quantitative geneticists since the early 1900s. With the notion that GE is undesirable and/or that it confounds genotype evaluation, much work has been devoted to developing stability indices to quantify and select against GE. Many stability indices have been proposed, as reviewed in Lin and Binns (1994) and more recently in Yan and Kang (2003). Several books and symposium proceedings have been published to document the advances in the study of GE (Byth and Montgomery1981; Kang, 1990, 2003; Gauch 1992; Imrie and Hacker 1993; Kang and Gauch 1996; Cooper and Hammer 1996) and most earlier research on GE can be traced from these publications. Although research on GE has contributed considerably to the understanding of this issue, there remains a gap in how GE is measured and addressed between biometricians and quantitative geneticists versus breeders and other practitioners.

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Abbreviations: **AEA**, average-environment axis; **AEC**, average-environment coordination; **GE**, genotype by environment interaction; **GGE**, G + GE; **MET**, multi-environment trials; **PC**, principal component; **PCA**, principal component analysis; **SVD**, singular value decomposition; **SVP**, singular value partitioning

The former groups concentrate primarily on quantification of GE, while the latter groups are often concerned primarily with matching genotypes with environments.

This gap may be partially bridged by the advent of biplot analysis methodology. A biplot is a scatter plot that approximates and graphically displays a two-way table by both its row and column factors such that relationships among the row factors, relationships among the column factors, and the underlying interactions between the row and column factors can be visualized simultaneously. Since its first report by Gabriel (1971), biplots have been used in visual data analysis by scientists of all disciplines, from economics, sociology, business, medicine, to ecology, genetics, and agronomy. Currently, most major statistical software packages include a procedure or macro for generating biplots. Numerous publications have been published in peer-reviewed journals documenting the use of biplot in understanding research data, and more than 50 000 web pages containing the word "biplot" are currently available on the Internet. A common misconception is that biplot analysis is equivalent to principle component analysis (PCA). While both biplot analysis and PCA use Singular Value Decomposition (SVD) (Pearson 1901) as a key mathematical technique, biplot analysis is a fuller use of SVD to allow two interacting factors to be visualized simultaneously.

The first application of biplots to agricultural data analysis was Bradu and Gabriel (1978), who used data from a cotton performance trial to illustrate the diagnostic role of biplots for model selection. Other early work of analyzing genotype by environment data using biplots includes Kempton (1984), Gauch (1992), and Cooper and DeLacy (1994). Kroonenberg (1995) distributed an excellent introduction to biplot analysis of genotype by environment tables on the Internet. More recently, the term "GGE biplot" was proposed and various biplot visualization methods developed to address specific questions relative to genotype by environment data (Yan et al. 2000). The term "GGE" emphasizes the understanding that G and GE are the two sources of variation that are relevant to genotype evaluation and must be considered simultaneously for appropriate genotype and test environment evaluation. GGE biplot analysis has evolved into a comprehensive analysis system whereby most questions that may be asked of a genotype by environment table can be graphically addressed (Yan et al. 2000; Yan 2001; Yan and Kang 2003; Yan and Tinker 2005a). The use of this system has been extended to visual analyses of other types of breeding related data, including genotype by trait tables (Yan and Rajcan 2002), host by pathogen tables (Yan and Falk 2002), diallel cross tables (Yan and Hunt 2002), and QTL by environment tables (Yan and Tinker 2005b).

Biplot analysis can be performed using many statistical packages either as a specialized feature or through customized programming or macros. A user-friendly software package for biplot analysis that is dedicated to simplifying the selection and construction of accurate biplot diagrams has also been developed (Yan 2001; Yan and Kang 2003). This software performs biplot analysis of genotype by environment tables (and other types of two-way tables), genotype by environment by trait three-way tables, and year by location by genotype by trait four-way tables. It creates an interactive analysis environment that is intended to be simple and informative, particularly for researchers with limited training in statistics and computer applications. The GGEbiplot software is continually enhanced and improved. All biplots presented in this review are direct outputs of this software.

This paper will review the basic concepts of biplot analysis and its applications in MET data analysis. The purpose is to provide a working guide for breeders, agronomists, and other agricultural scientists on biplot analysis and interpretation. The paper is divided into four sections. The first section is a complete but succinct description of the principles of biplot analysis. The second section is a detailed treatment of biplot analysis of genotype by environment data. It addresses environment and genotype evaluation from all possible perspectives. The third section deals with biplot analysis of various two-way tables that can be generated from a three-way MET dataset, which is an integral and essential part to a fuller understanding and exploration of MET data. The final section discusses questions that are frequently asked about biplot analysis. The order of layout is a conceptual reconstruction rather than a historical narration.

PRINCIPLES OF BIPLOT ANALYSIS

Biplot and its Inner-product Property

Mathematically, a biplot may be regarded as a graphical display of matrix multiplication. Given a matrix G with m rows and r columns, and a matrix E with r rows and n columns, they can be multiplied to give a third matrix P with m rows and n columns. If $r = 2$, then matrix G can be displayed as m points in a 2-D plot, with the 1st column as the abscissa (x-axis) and 2nd column the ordinate (y-axis). Similarly, matrix E can be displayed as n points in a 2-D plot, with the 1st row as the abscissa and 2nd row the ordinate. A 2-D biplot is formed if the two plots are superimposed, which would contain $m + n$ points. An interesting property of this biplot is that it not only displays matrices G and E, but also implicitly displays the m x n values of matrix P, because each element of P can be visualized as:

$$P_{ij} = x_i x'_j + y_i y'_j = \vec{g}_i \vec{e}_j = |g_i| \|e_j| \cos\theta_{ij} \quad (1)$$

Where (x_i, y_i) are the coordinates for row i and (x'_j, y'_j) are coordinates for column j; $\vec{g}_i$ is the vector for row i and $\vec{e}_i$ is the vector for column j; $|g_i|$ is the vector length for row i and $|e_j|$ is the vector length for column j. θ_{ij} is the angle between the vectors of row i and column j.

Equation 1 is referred to as the inner-product property of the biplot. It is the most important property of a biplot. It not only allows each element of matrix P to be estimated but also constitutes the basis for visualizing the patterns in matrix P, including ranking the rows relative to any column, ranking the columns relative to any row, comparing any two rows relative to individual columns, identifying the rows with largest (or smallest) values for each column, or vice versa (Yan and Kang 2003, chapter 3).

Singular Value Decomposition and Partitioning

The practical application of a biplot in data analysis was stated most clearly by the founder of biplot (Gabriel 1971): any two-way table can be graphically analyzed using a 2-D biplot as soon as it can be sufficiently approximated by a rank-2 (i.e., $r = 2$) matrix. Given a genotype by environment two-way table P of m genotypes and n environments, biplot analysis starts with its decomposition into three matrices G, L, and E, via SVD:

$$P_{m,n} = G_{m,r} L_{r,r} E^{T}_{n,r} \quad \left(r \le \min(m,n)\right) \tag{2}$$

Matrix G has m rows and r columns; it characterizes the m genotypes. Matrix E has r rows and n columns; it characterizes the n environments. Matrix L is a diagonal matrix containing r singular values. In summation notation, SVD decomposes P into r principal components (PC), each containing a genotype vector (ξ_i), an environment vector (η_j), and a singular value (λ):

$$P_{ij} = \sum_{l=1}^{r} \xi_{il} \lambda_l \eta_{lj} \quad \left(\lambda_l \ge \lambda_{l+1}\right) \tag{3}$$

where r is the rank of the two-way table, i.e., the number of PC required to fully represent P, with $r \le \min(m, n)$. If $r < m$ there are associations (linear relationships) among the genotypes. This is true whenever $m > n$. If $r < n$ there are associations among the environments. The environments are independent if and only if $r = n$. λ_l is the singular value for PCl, and λ_l^2 is the lth nonzero eigenvalue of P^TP or PP^T. The lth column of G is an eigenvector of PP^T (a row eigenvector) corresponding to λ_l^2, and the lth column of E is an eigenvector of P^TP (a column eigenvector) corresponding to λ_l^2. Another requirement of SVD is that $G^TG = I_{r,r} = E^TE$, where $I_{r,r}$ is the r by r identity matrix.

When $r = 2$, the two-way table P is said to be a rank-2 matrix and can be displayed in a 2-D biplot exactly. The goodness of fit of a 2-D biplot for the two-way table is measured by the ratio of $(\lambda_1^2 + \lambda_2^2)$/SS, where SS is the sum of squares of the two-way table. Because the PC are arranged such that $\lambda_l \ge \lambda_{l+1}$, a 2-D biplot of PC1 vs. PC2 always displays the most important patterns of P, even when the goodness of fit is poor. The goodness of fit reflects the strength of the associations (i.e., patterns) among the environments or among the genotypes. A poor fit implies that the dataset either has complicated patterns or has no discernible patterns at all. If all environments are independent of each other and all genotypes are independent of each other, each PC should explain 1/min(m, n) of the SS.

The singular values must be partitioned into the genotype and environment scores before a biplot can be constructed to approximate the two-way data:

$$P_{ij} = \sum_{l=1}^{r} \xi_{il}^{*} \eta_{ij}^{*} = \sum_{l=1}^{r} \left(\xi_{il} \lambda_l^{f}\right)\left(\lambda_l^{1-f} \eta_{lj}\right) \tag{4}$$

where f is the partitioning factor, which can be anything between 0 and 1, resulting in an unlimited number of ways of singular value partitioning. Among these, two methods are particularly useful: column-metric preserving and row-metric preserving (Gabriel 2002; Yan 2002). A third method is symmetrical partitioning, which has been the most used, but not necessarily the most useful, singular value partitioning method.

Column-metric Preserving and Associated Interpretations

When $f = 0$, the singular values are entirely partitioned into the column (here environment) eigenvectors, referred to as column-metric preserving. Since $E^* = EL$, we have $E^*(E^*)^T = P^TP$, which is the sum of squares and cross products matrix of P. If P is column-centred then P^TP is $(m - 1)$ times the covariance matrix. Therefore, this partitioning is appropriate for studying the relationships among column factors. Three important rules apply to this partitioning:

The dot product, which is $(m - 1)$ times of the covariance between two columns, can be visualized as:

$$\sum_{i=1}^{m} p_{ij} p_{ij'} = \left|e_j\right| \left\|e_{j'}\right| \cos\theta_{jj'}. \tag{5}$$

Since the correlation between two columns is estimated by

$$r_{ij'} = \frac{\sum_{i=1}^{m} \left(p_{ij} - \bar{p}_j\right)\left(p_{ij'} - \bar{p}_{j'}\right)}{\sqrt{\sum_{i=1}^{m} \left(p_{ij} - \bar{p}_j\right)^2 \sum_{i=1}^{m} \left(p_{ij'} - \bar{p}_{j'}\right)^2}}, \tag{6}$$

when the two-way data are column-centered (as all types of GGE biplots, see "data centering" later), i.e., when $\bar{p}_j = \bar{p}_j = 0$, we have:

$$r_{ij'} = \frac{\sum_{i=1}^{m} p_{ij} p_{ij'}}{\sqrt{\sum_{i=1}^{m} p_{ij}^2 \sum_{i=1}^{m} p_{ij'}^2}} = \frac{\sum_{i=1}^{m} p_{ij} p_{ij'}}{\left|e_j\right| \left\|e_{j'}\right|},$$

or

$$\sum_{i=1}^{m} p_{ij} p_{ij'} = \left|e_j\right| \left\|e_{j'}\right| r_{ij'}.$$

From Eq. 5, we have

$$r_{jj'} = \cos \alpha_{jj'} \tag{7}$$

This important relationship for column-centered data is called "equality between cosines and correlations." Verbally, the correlation between two columns is approximated by the cosine of the angle between their vectors *if* the

data are column-centered before subjecting to SVD (Kroonenberg 1995).

Another useful relationship is that the vector length of a column equals $\sqrt{m-1}$ times the standard deviation of the column factor across the rows because

$$s_j\sqrt{m-1} = \sqrt{\sum_{i=1}^{m}\left(p_{ij}-\bar{p}_j\right)^2} = \sqrt{\sum_{i=1}^{m}p_j^2} = \left|e_j\right|. \quad (8)$$

This relation can be used to visualize the relative discriminating ability of the column factors. That is, for a biplot based on column-centered data, the length of column vectors measures their ability to discriminate among the rows.

Row-metric Preserving and Associated Interpretations

When f =1, the singular values are entirely partitioned into the row eigenvectors, which is referred to as row-metric preserving. Since $G^* = GL$, we have $G^*(G^*)^T = PP^T$. Therefore, this partitioning recovers the Euclidean distances among row factors (here genotypes) and is, therefore, appropriate for visualizing the similarity/dissimilarity among row factors. More discussions on this partitioning method are presented in the "comparison among all genotypes" selection.

Data Centering Prior to Singular Value Decomposition

In a genotype by environment two-way table Y, the value of each cell can be regarded as mixed effect of the grand mean (μ) modified by the genotype (row) main effect (α_i), the environment (column) main effect (β_j), and the specific genotype (row) by environment (column) interaction (ϕ_{ij}), plus any random error (ε_{ij}):

$$y_{ij} = \mu + \alpha_i + \beta_j + \phi_{ij} + \varepsilon_{ij} \quad (9)$$

The matrix P that is subjected to SVD (Eq. 3) can be any part of Y, resulting in different models, ignoring random errors:

$$P_{ij} = y_{ij} = \mu + \alpha_i + \beta_j + \phi_{ij} \quad (10)$$

$$P_{ij} = y_{ij} - \mu = \alpha_i + \beta_j + \phi_{ij} \quad (11)$$

$$P_{ij} = y_{ij} - \mu - \alpha_i = \beta_j + \phi_{ij} \quad (12)$$

$$P_{ij} = y_{ij} - \mu - \beta_j = \alpha_i + \phi_{ij} \quad (13)$$

$$P_{ij} = y_{ij} - \mu - \alpha_i = \beta_j = \phi_{ij} \quad (14)$$

Obviously, biplots based on different models (Eqs. 10–14) have different interpretations. All models are useful, depending on the research purposes and the questions one wishes to address. If one is interested only in GE, Eq. 14 should be the choice. This model is also appropriate for visual study of microarray (gene expression) data (Pittelkow and Wilson 2003; Wouters et al. 2003), because it is the relative changes that are the focus. If one is interested in genotype evaluation, Eq. 13 is most appropriate, as it contains both G and GE, which must considered simultaneously. Biplots based on Eq. 13 are referred to as "GGE biplots" (Yan et al. 2000). If one is interested in visual study of the data per se, Eq. 10 should be the choice. This model is appropriate for constructing QQE biplots (Yan and Tinker 2005b) or genetic covariate by environment biplots (Yan and Tinker 2005a). These centering methods, plus various data scaling (see below) and transformation options, have been built into GGEbiplot.

Data Scaling Prior to Singular Value Decomposition

The GGE biplot model (Eq. 13) can be more generally presented as:

$$P_{ij} = (y_{ij} - \mu - \beta_j)/s_j = (\alpha_i + \phi_{ij})/s_j \quad (15)$$

where s_j is a scaling factor. Thus, there can be different GGE models, depending on the definition of s_j. Equation 13 is a special case of Eq. 15 with s_j = 1. When s_j is the standard deviation for column (environment or trait) j, the data is said to be "standardized" such that all columns are given the same weight (importance). When s_j is the standard error in environment j, any heterogeneity among the environments will (supposedly) be removed. Replicated data are essential for estimating standard errors in each environment. Data standardization is essential for biplot analysis of two-way tables in which the columns are of different units or scales. Genotype-by-trait tables belong to this category. GGEbiplot allows each of the models (Eqs. 10 to 14) to be scaled in various ways.

Four Questions to be Asked Before Trying to Interpret a Biplot

From the above discussions, four questions need to be asked to correctly interpret a biplot (Yan and Tinker 2005a).

First, what is the model on which the biplot is generated? That is, how are the data centered and scaled? This determines what kind of questions can be asked of the biplot. For example, it is not possible to visualize the mean yield of the genotypes in a biplot based on Eq. 14, which contains GE only. Similarly, it is not possible to visualize the main effects of environments in a GGE biplot, as this information is removed from the model.

Second, how are the singular values partitioned? This again determines if certain relationships can be properly visualized. For example, the relationships among environments cannot be accurately visualized in a GGE biplot that is based on genotype-metric preserving (row-metric preserving) or symmetrical partitioning.

Third, what is the goodness of fit of the biplot for the table that is to be visualized? That is, does the biplot adequately approximate the two-way table? If not, some patterns may not be displayed in the primary biplot (i.e., biplot of PC1 vs. PC2). More discussions on this are presented later in the "frequently asked questions on biplot analysis" section.

Finally, are the axes drawn to scale? All interpretations of a biplot are based on the assumption that both axes are drawn to scale. If not, the biplot may be misleading as the

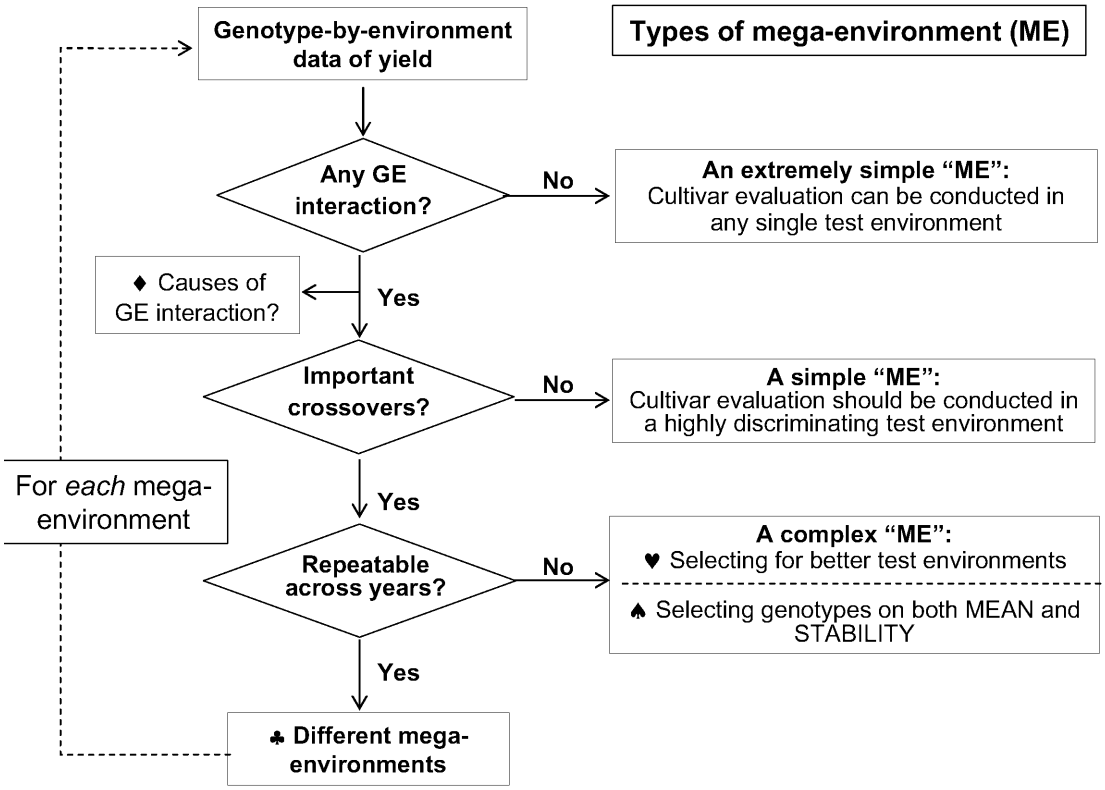

Fig. 1. A scheme of multi-environment trial data analysis.

relationships are distorted. Biplots generated using the GGEbiplot software explicitly address these concerns.

BIPLOT ANALYSIS OF GENOTYPE BY ENVIRONMENT DATA

Four Objectives of Genotype-by-environment Data Analysis

Performance trials have to be conducted in multiple environments because of the presence of GE. For the same reason, the analysis of genotype by environment data must start with the examination of the magnitude and nature of GE (Fig. 1). The first question to ask is whether there is significant GE in the data. If not, genotypes can be reliably evaluated in any single environment. Unfortunately, this situation rarely exists except perhaps for certain traits that are under simple genetic control. If GE exists, it is necessary to determine whether there are important crossovers, i.e., rank changes of the genotypes in different environments, such that different winners are picked up in different environments. If not, superior genotypes can be identified in any of the environments but there exists an ideal test environment in which the best genotypes can be most easily identified. If crossover interactions exist, it is necessary to determine whether the crossover GE patterns are repeatable across years. Data from multiple years are necessary to address this question. If there are repeatable interactions then the target environments should be divided into different mega-environments and genotype evaluation should be conducted separately for each mega-environment. Dividing target environments into meaningful mega-environments is the only way that GE can be exploited (Yan and Tinker 2005a). If there is no recognizable pattern of GE, then the target environment is a single mega-environment with unpredictable GE, and models addressing random sources of variation may be appropriate.

Within a single mega-environment, the objectives of data analysis are twofold: genotype evaluation to identify genotypes with both high performance and high stability, and test environment evaluation to identify test environments that are both informative (discriminating) and representative. In addition, whenever there is significant GE, potential causes of GE should be explored.

To summarize, genotype by environment data analysis should address the following four questions:

(1) Can the target environment be divided into meaningful mega-environments so that some of the GE can be exploited or avoided? Multi-year data are essential to address this question.
(2) What are the causes of GE? Data of genetic and environmental covariates are required to address this question.
(3) What are the best test environments (representative and discriminating)?
(4) What are the superior genotypes (both high and stable performance within a mega-environment)?

Given sufficient data, biplot analysis implemented by GGEbiplot can help address these questions effectively and conveniently.

Table 1. Mean yield (ton ha^{-1}) of 18 winter wheat varieties (G1 to G18) tested at nine Ontario locations (E1 to E9) in 1993

Genotypes	E1	E2	E3	E4	E5	E6	E7	E8	E9
G1	4.46	4.15	2.85	3.08	5.94	4.45	4.35	4.04	2.67
G2	4.42	4.77	2.91	3.51	5.70	5.15	4.96	4.39	2.94
G3	4.67	4.58	3.10	3.46	6.07	5.03	4.73	3.90	2.62
G4	4.73	4.75	3.38	3.90	6.22	5.34	4.23	4.89	3.45
G5	4.39	4.60	3.51	3.85	5.77	5.42	5.15	4.10	2.83
G6	5.18	4.48	2.99	3.77	6.58	5.05	3.99	4.27	2.78
G7	3.38	4.18	2.74	3.16	5.34	4.27	4.16	4.06	2.03
G8	4.85	4.66	4.43	3.95	5.54	5.83	4.17	5.06	3.57
G9	5.04	4.74	3.51	3.44	5.96	4.86	4.98	4.51	2.86
G10	5.20	4.66	3.60	3.76	5.94	5.35	3.90	4.45	3.30
G11	4.29	4.53	2.76	3.42	6.14	5.25	4.86	4.14	3.15
G12	3.15	3.04	2.39	2.35	4.23	4.26	3.38	4.07	2.10
G13	4.10	3.88	2.30	3.72	4.56	5.15	2.60	4.96	2.89
G14	3.34	3.85	2.42	2.78	4.63	5.09	3.28	3.92	2.56
G15	4.38	4.70	3.66	3.59	6.19	5.14	3.93	4.21	2.93
G16	4.94	4.70	2.95	3.90	6.06	5.33	4.30	4.30	3.03
G17	3.79	4.97	3.38	3.35	4.77	5.30	4.32	4.86	3.38
G18	4.24	4.65	3.61	3.91	6.64	4.83	5.01	4.36	3.11

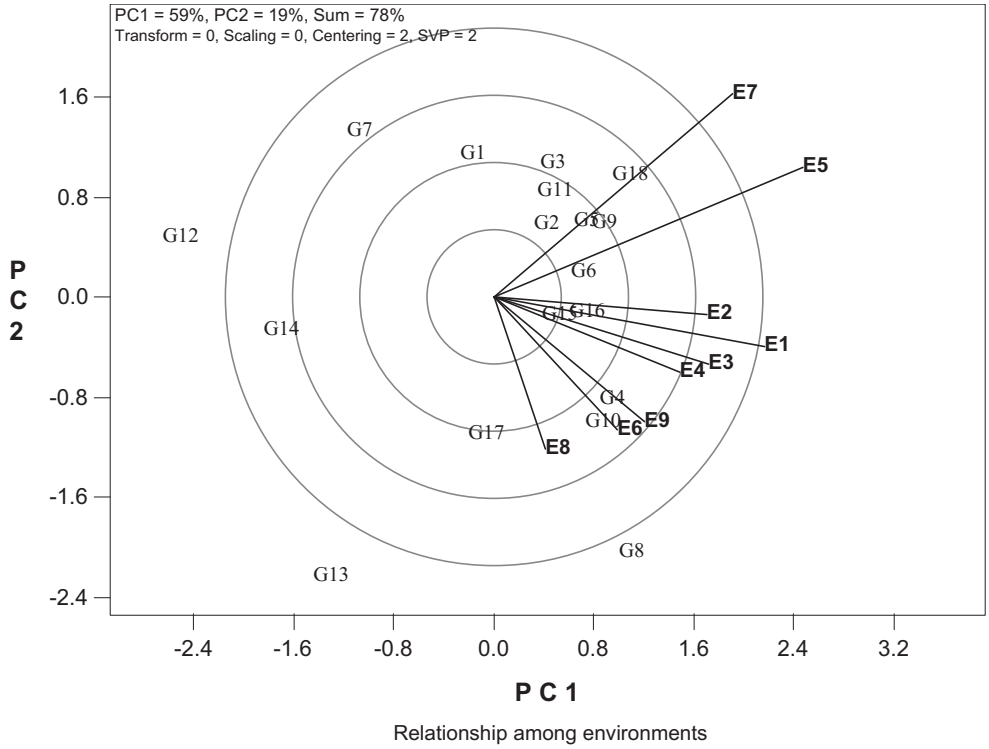

Fig. 2. The environment-vector view of the GGE biplot to show similarities among test environments in discriminating the genotypes.

Most of the discussions below are based on the yield data of 1993 Ontario winter wheat performance trials, in which 18 genotypes (G1 to G18) were tested at nine locations (E1 to E9). The data are presented in Table 1; interpretations based on the biplots can be checked against it for correctness and accuracy.

Environment Evaluation Based on GGE Biplots

Relationships Among Test Environments

Figure 2 is the environment-vector view of the GGE biplot for the data in Table 1. It is based on an environment-centered (centering = 2) G by E table without any scaling (scaling = 0), and it is environment-metric preserving (SVP = 2) and its axes are drawn to scale (default feature of GGEbiplot). This biplot explained 78% of total variation of the environment-centered G by E table.

Assuming that it adequately approximates the environment-centered two-way table (more discussion on this assumption in the "frequently asked questions on biplot analysis" section), Fig. 2 can be interpreted as follows.

(1) The lines that connect the test environments to the biplot origin are called environment vectors. According to Eq. 7, the cosine of the angle between the vectors of two environ-

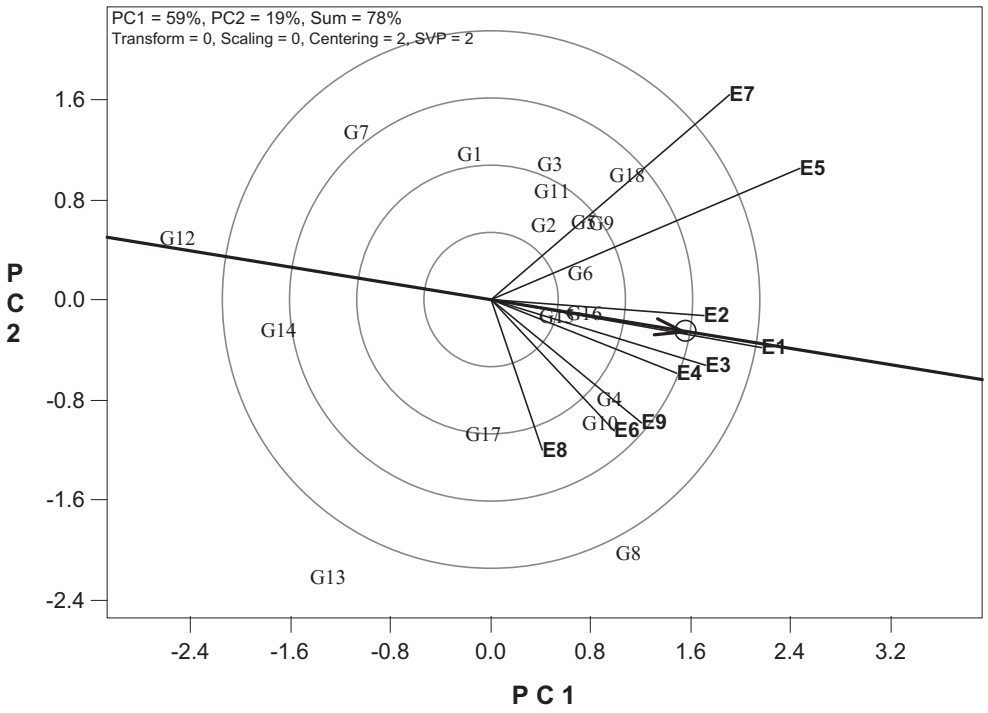

Fig. 3. The discrimination and representativeness view of the GGE biplot to show the discriminating ability and representativeness the test environments.

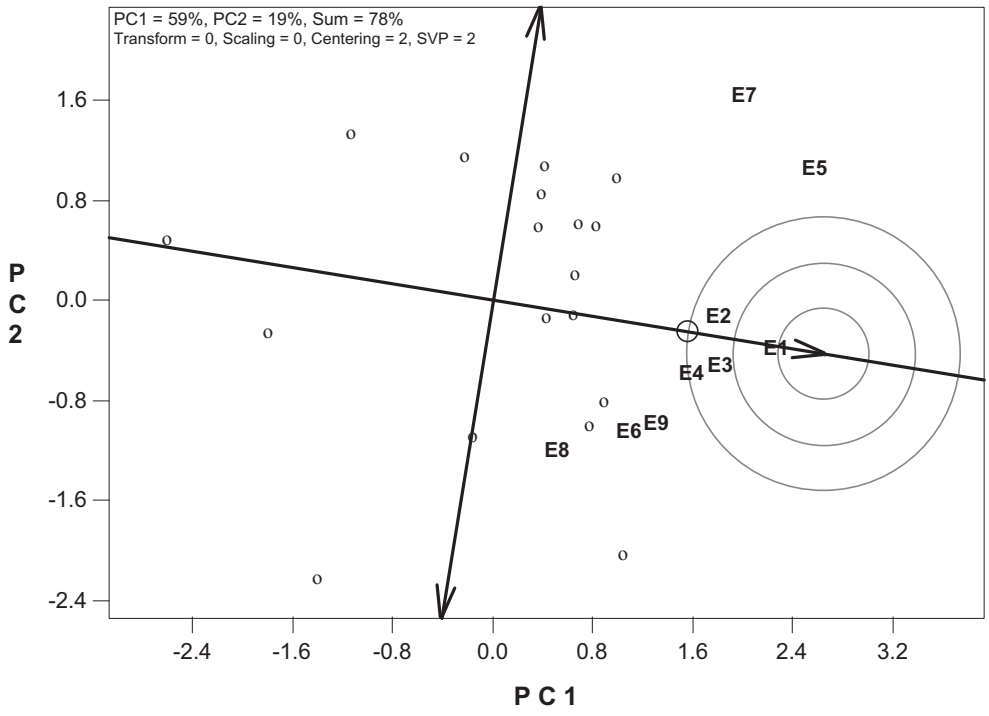

Fig. 4. The discrimination and representativeness view of the GGE biplot to rank test environments relative to an ideal test environment (represented by center of the concentric circles).

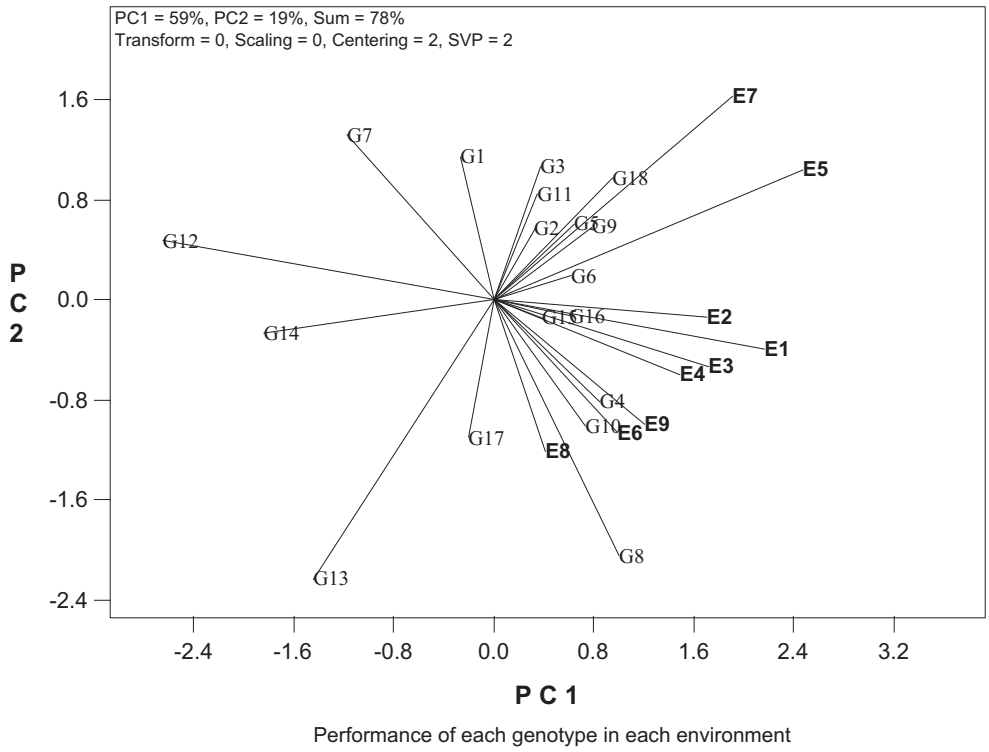

Fig. 5. The GGE biplot showing the performance of each genotype in each environment.

ments approximates the correlation between them. For example, E7 and E5 were positively correlated (an acute angle), E7 and E8 were slightly negatively correlated (an obtuse angle), and E5 and E8 were not correlated (a right angle).

(2) The presence of wide obtuse angles (i.e., strong negative correlations) among test environments is an indication of strong crossover GE. Here the largest angle is slightly larger than 90° (between E7 and E8), implying that the GE is moderately large.

(3) The *distance* between two environments measures their dissimilarity in discriminating the genotypes. Thus, the nine environments fell into two apparent groups: E7 and E5 formed one group, and the remaining environments formed another [according to Eq. 5, the similarity (covariance) between two environments is determined by both the length of their vectors and the cosine of the angle between them].

(4) The presence of close associations among test environments suggests that the same information about the genotypes could be obtained from fewer test environments, and hence the potential to reduce testing cost. If two test environments are closely correlated consistently across years, one of them can be dropped without loss of much information about the genotypes.

Discriminating Ability of Test Environments

(1) The concentric circles on the biplot help to visualize the length of the environment vectors, which is proportional to the standard deviation within the respective environments (Eq. 8) and is a measure of the discriminating ability of the environments. Therefore, among the nine environments, E7 and E5 were most discriminating (informative) and E8 least discriminating (Fig. 2).

(2) Test environments that are consistently non-discriminating (non-informative) provide little information on the genotypes and, therefore, should not be used as test environments.

Representativeness of Test Environments

Figure 3 presents the same biplot as Fig. 2 except that an "Average-Environment Axis" [AEA, or average-tester-axis, Yan (2001)] has been added. The average environment (represented by the small circle at the end of the arrow) has the average coordinates of all test environments, and AEA is the line that passes through the average environment and the biplot origin. Figure 3 can be interpreted as follows:

(1) A test environment that has a smaller angle with the AEA is more representative of other test environments. Thus, E1 is most representative whereas E7 and E8 least representative.

(2) Test environments that are both discriminating and representative (e.g., E1) are good test environments for selecting generally adapted genotypes.

(3) Discriminating but non-representative test environments (e.g., E7 and E8) are useful for selecting specifically adapted genotypes *if* the target environments can be divided into mega-environments.

(4) Discriminating but non-representative test environments (e.g., E7) are useful for culling unstable genotypes if the target environment is a single mega-environment.
(5) Non-discriminating test environments (those with very short vectors) are less useful because they provide little discriminating information about the genotypes.

Ideal Test Environments for Selecting Generally Adapted Genotypes

Within a single mega-environment, the ideal test environment should be most discriminating (informative) and also most representative of the target environment. Figure 4 defines an "ideal test environment", which is the center of the concentric circles. It is a point on the AEA in the positive direction ("most representative") with a distance to the biplot origin equal to the longest vector of all environments ("most informative"). E1 is closest to this point and is, therefore, best, whereas E7 and E8 were poorest for selecting cultivars adapted to the whole region. Note that additional years are required to confirm that a specific test location is "ideal".

Mega-environment Identification

The pattern of environments in the above biplots suggests the existence of two different mega-environments. Multi-year data are required to confirm this hypothesis, i.e., to see if this pattern is repeatable across years. In this example, E7 and E5 were from eastern Ontario whereas the others except E1 were from southern Ontario. This pattern did seem to be repeatable across years (Yan et al. 2000).

Yan and Tinker (2005a) presented another example of mega-environment investigation using biplots based on yield data of 145 barley double-haploids measured at multiple locations across northern North America in 1992 and 1993. Two spring barley (western vs. eastern) mega-environments were identified based on repeatable location by genotype interactions. This result implies that eastern and western Canada (plus the northwestern states of the United States of America) are different mega-environments that require different barley varieties for maximum yield.

Genotype Evaluation Based on GGE Biplots

Performance of the Genotypes in Specific Environments

Both the genotype vectors and the environment vectors are drawn in Fig. 5 so that the specific interactions between a genotype and an environment (i.e., the performance of each genotype in each environment) can be visualized. The interpretation rule is: the performance of a genotype in an environment is better than average if the angle between its vector and the environment's vector is <90°; it is poorer than average if the angle is >90°; and it is near average if the angle is about 90°. For example, G12 was below average in all environments (obtuse angles) whereas G8 was above average in all environments (acute angles) except E7. The angle determines the direction of the interaction, i.e., above or below average in the specific environment; the magnitude of the interaction is determined by both the cosine of the angle and the length of the vectors. The basis of the interpretation is the "inner-product" principle (Eq. 1), which is valid regardless of singular value partitioning method. Figure 5 can be used (1) to rank the genotypes based on performance in any environment, and (2) to rank environments on the relative performance of any genotype. Additional uses of this property are detailed below.

Ranking Genotypes Based on Performance in One Environment

To rank the genotypes based on their performance in an environment, a line is drawn that passes through the biplot origin and the environment. This line is called the axis for this environment, and along it is the ranking of the genotypes. Figure 6 ranks the genotypes based on performance in E5. Genotypes G12, G14, G13, G7, and G17 had lower than average yield, G1 and G8 had near average yield, and all others had higher than average yields. The highest yielder in E5 was G18 and the lowest yielder G12.

Ranking Environments Based on the Performance of a Genotype

To study the specific adaptation of a genotype, i.e., to rank the test environments on the relative performance of a genotype, a line is drawn that passes through the biplot origin and the genotype. This line is called the axis for this genotype, and along it is the ranking of the environments. For example, Fig. 7 ranks the test environments based on the relative performance of G8. It shows that G8 had lower than average yield in environment E7, near-average yield in E5, and higher than average yield in other environments.

Mean Performance and Stability of the Genotypes

Within a single mega-environment, genotypes should be evaluated on both mean performance and stability across environments. Figure 8 is the average-environment coordination (AEC) view of the GGE biplot. It is the same as Figs. 3 and 4 except that it is genotype-metric preserving (SVP = 1) and is, therefore, more appropriate for genotype evaluations with the following interpretations:
(1) The single-arrowed line is the AEC abscissa (or AEA); it points to higher mean yield across environments. Thus, G8 had the highest mean yield, followed by G4, G10, etc.; G17 had a mean yield similar to the grand mean; and G12 had the lowest mean yield.
(2) The double-arrowed line is the AEC ordinate; it points to greater variability (poorer stability) in either direction. Thus, G13 was highly unstable whereas G4 was highly stable. Note that if the biplot explained only a small portion of the total variation, some seemingly stable genotypes may not be truly stable as their variations may not be fully explained in this biplot.
(3) G13 was highly unstable because it had lower than expected yield in environments E7 and E5 but higher than expected yield in E6, E8, and E9. Its yield in E1 was just as expected from its average yield across environments.

Ranking Genotypes Relative to the Ideal Genotype

An ideal genotype should have both high mean performance and high stability across environments. Figure 9 defines an "ideal" genotype (the center of the concentric circles) to be a point on the AEA ("absolutely stable") in the positive

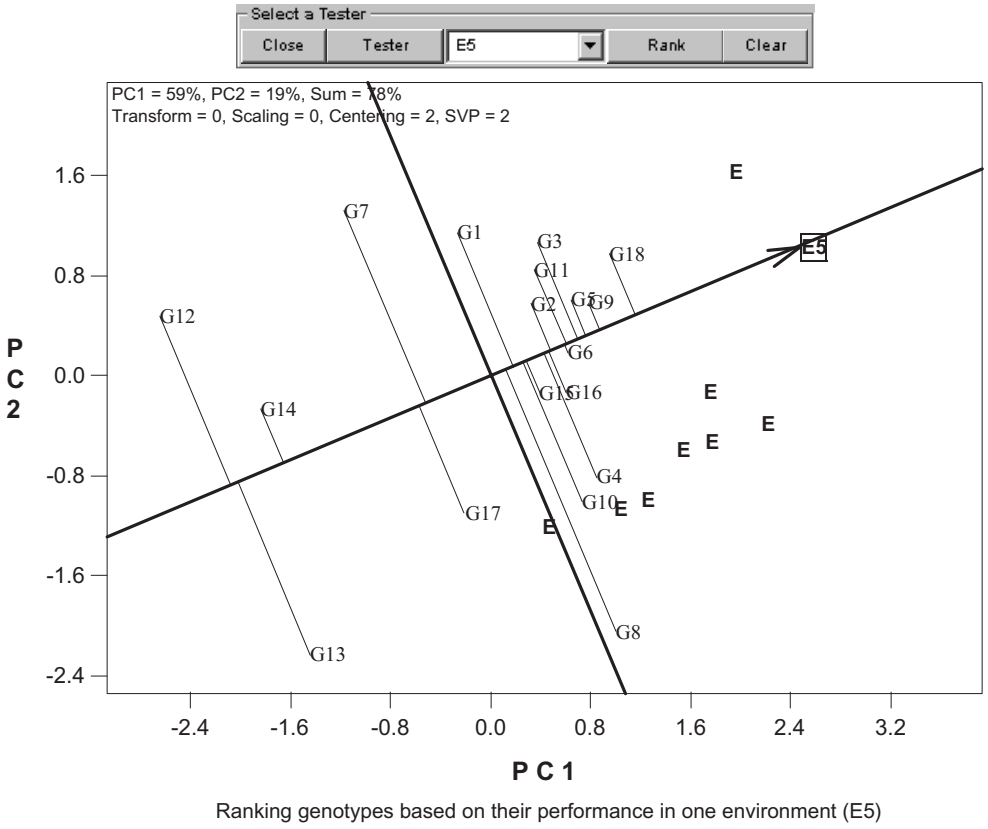

Fig. 6. Ranking genotypes based on performance in a specific environment (E5).

direction and has a vector length equal to the longest vectors of the genotypes on the positive side of AEA ("highest mean performance"). Therefore, genotypes located closer to the 'ideal genotype' are more desirable than others. Thus, G4 was more desirable than G8 even though G8 had higher average yield. G12 was, of course, the poorest genotype because it was consistently the poorest.

Figure 9 illustrates an important concept regarding "stability". The term "high stability" is meaningful only when associated with mean performance. According to Fig. 9, G12 is highly "stable". This does not mean G12 was any good; it only means that the relative performance of G12 was consistent. G12 was even poorer than the highly variable, least stable genotype G13, because G13 performed reasonably well in at least some environments. From this example, it should be easy to see how misleading it can be to search and select for "stability" genes. "Stable" genotypes are desirable only when they have high mean performances.

Comparison Among All Genotypes

Figure 10 is similar to the GGE biplot in Fig. 2 except that it is genotype-metric preserving (SVP = 1) and is, therefore, appropriate for comparing genotypes. The following interpretations can be made based on it.

(1) The distance between two genotypes approximates the Euclidean distance between them, which is a measure of the overall dissimilarity between them. For example, G8 and G12 are very different whereas G10 and G4 are quite similar. The dissimilarity can be due to difference in mean yield (G) and/or in interaction with the environments (GE).
(2) The biplot origin represents a "virtual" genotype that assumes an average value in each of the environments. This "average" genotype has zero contributions to both G and GE.
(3) Therefore, the length of the genotype vector, which is the distance between a genotype and the biplot origin, measures the difference of the genotype from the "average" genotype, i.e., its contribution to either G or GE or both. Therefore, genotypes located near the biplot origin have little contribution to both G and GE and genotypes with longer vectors have large contributions to either G or GE or both. Therefore, genotypes with the longest vectors are either the best (e.g., G8) or the poorest (e.g., G12) or most unstable (e.g., G13) genotypes.
(4) The angle between the vector of a genotype and the AEA partitions the vector length into components of G and GE. A right angle means that the contribution is to GE only; an obtuse angle means the contribution is mainly to G, which leads to lower-than-average mean performance; and an acute angle means the contribution is mainly to G, which leads to higher-than-average mean

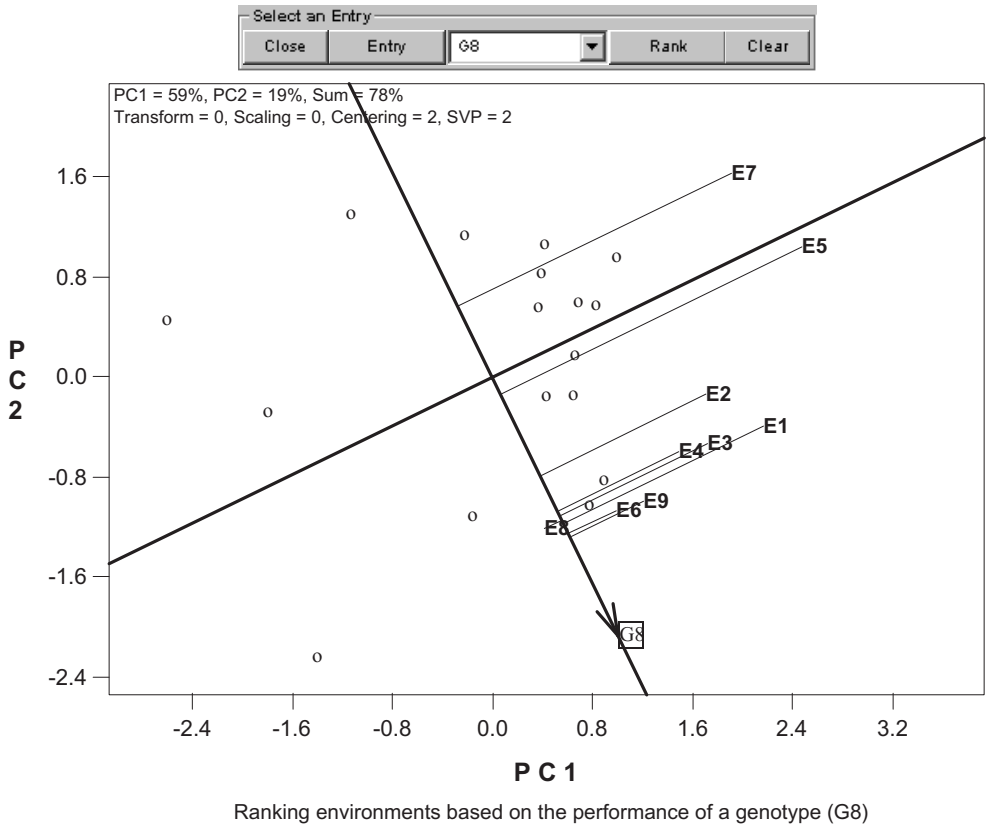

Fig. 7. Ranking test environments in terms of the relative performance of a genotype (G8).

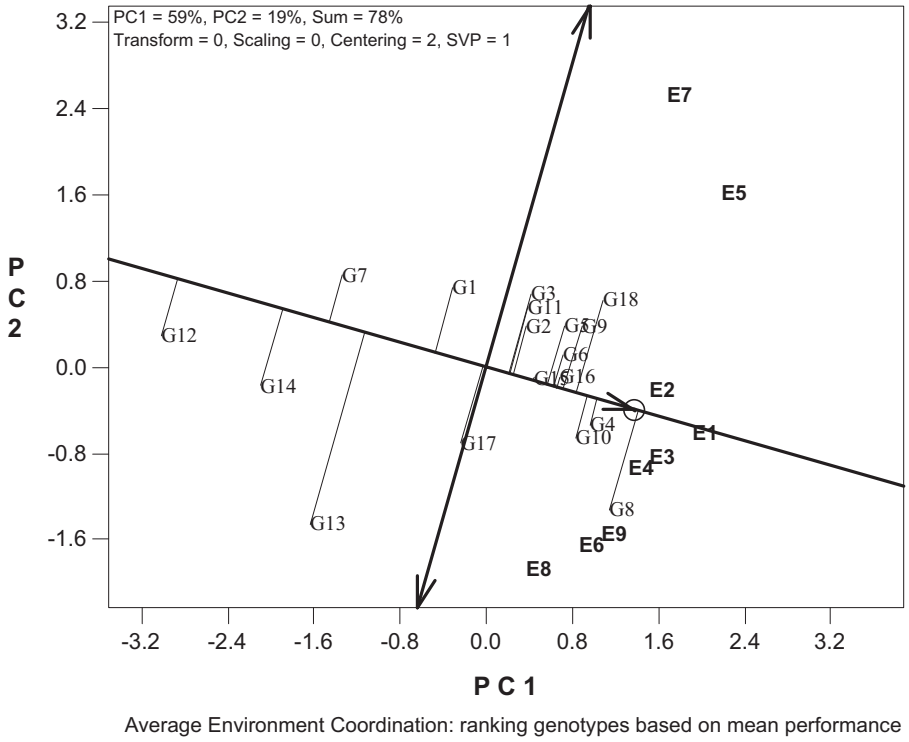

Fig. 8. The average-environment coordination (AEC) view to show the mean performance and stability of the genotypes.

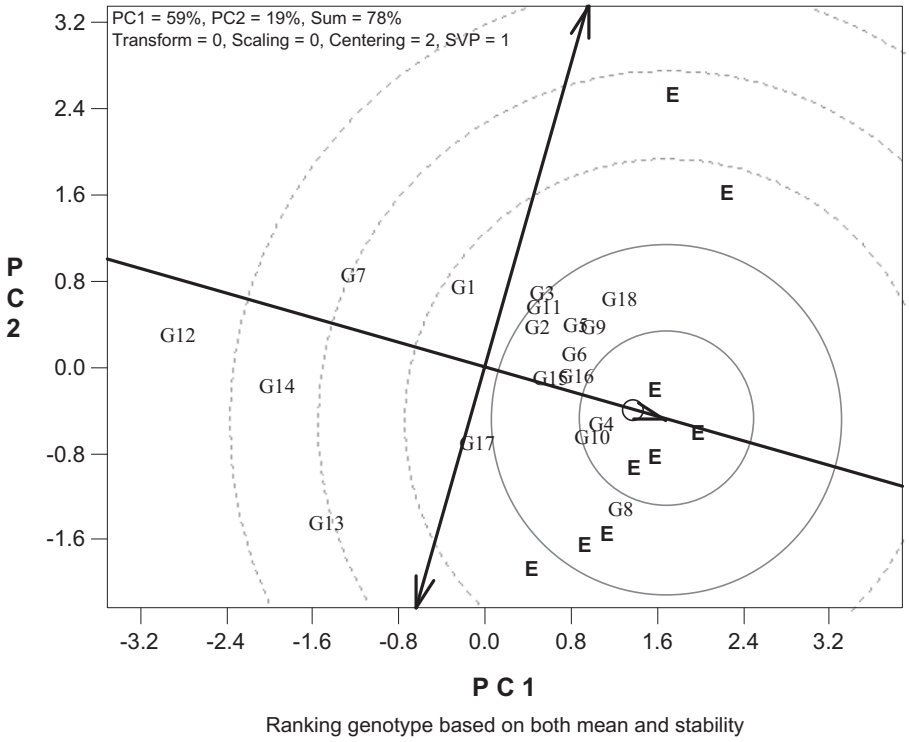

Fig. 9. The average-environment coordination (AEC) view to rank genotypes relative to an ideal genotype (the center of the concentric circles).

performance. Also see the "mean performance and stability of the genotypes" section.

(5) The angle between two genotypes indicates their similarity in response to the environments. An acute angle (e.g., G12 vs. G14) means that the two genotypes responded similarly and that the difference between them was proportional in all environments. An obtuse angle (e.g., G13 vs. G18) means that the two genotypes responded inversely and wherever the first genotype performed well the other genotype performed poorly. A right angle indicates that the two genotypes (e.g., G18 vs. G8) responded to the environments independently. In the first two cases the difference between the genotypes contributed more to G than to GE. In the third case, the difference contributed mostly to GE.

Comparison Between Any Two Genotypes

In a GGE biplot, two genotypes can be visually compared by connecting them with a straight line, followed by drawing a perpendicular line that passes through the biplot origin (Fig. 11). This perpendicular line is the "equality line" of the two genotypes. That is, the two genotypes to be compared should be equal in all environments that are located on this line. The following interpretations can be made based on Fig. 11:

(1) A genotype has higher values in environments that are located on its side of the equality line. Thus, G18 had higher yield in E5 and E7 whereas G8 had higher yield in other environments. This is a clear example of a "crossover" interaction.

(2) The difference between two genotypes varies by environment, being proportional to the distance of the environment to the equality line. Thus, the difference between G8 and G18 was relatively large in E7 and E8 but very small in E2.

(3) Since the biplot distance of the line that connects the two genotypes measures the Euclidian distance between them (when SVP = 1 is used), comparison using the Fig. 11 method is meaningful only if the connection line is long enough.

Note that SVP = 1 is required in Fig. 11 for point 3. Both SVP = 1 and SVP = 2 and any other singular value partitioning methods are equally valid for points (1) and (2) as both genotypes and environments are involved. Again, SVP = 1 is appropriate for comparing genotypes while treating environments as random samples; SVP = 2 is appropriate for studying relationships among environments while treating genotypes as random samples. Both are equally valid when studying specific genotype by environment relationships.

Which-won-where

One of the most attractive features of a GGE biplot is its ability to show the which-won-where pattern of a genotype by environment dataset (Fig. 12). Many researchers find this use of a biplot intriguing, as it graphically addresses important concepts such as crossover GE, mega-environment differentiation, specific adaptation, etc.

The "which-won-where" function of a GGE biplot is an extended use of the "pair-wise comparison" function described above. A polygon is first drawn on genotypes that

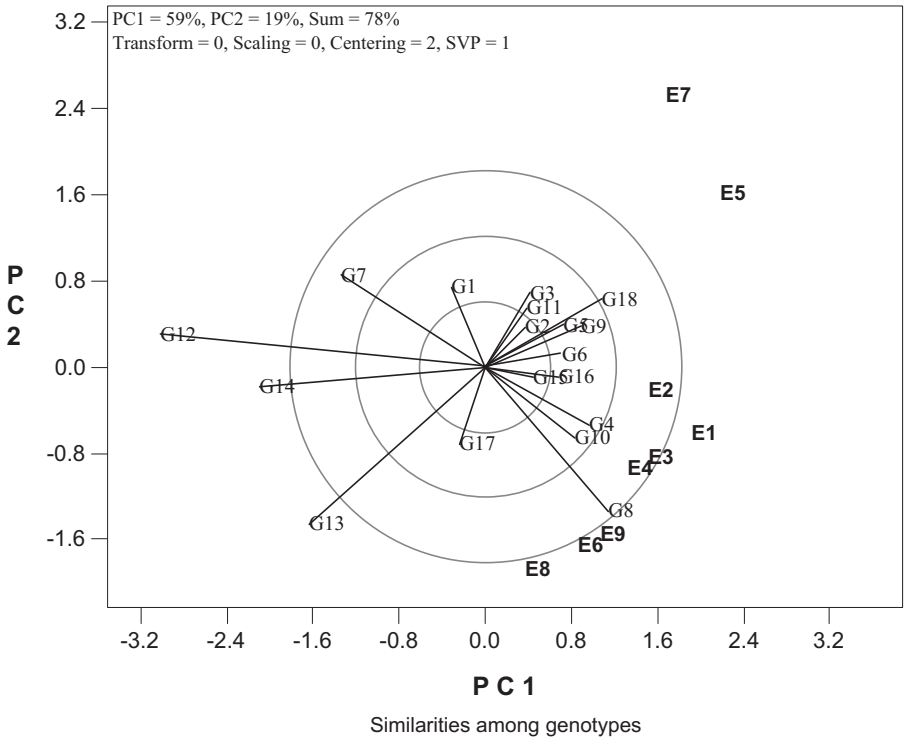

Fig. 10. The genotype-vector view to show similarities among genotypes in their performances in individual environments.

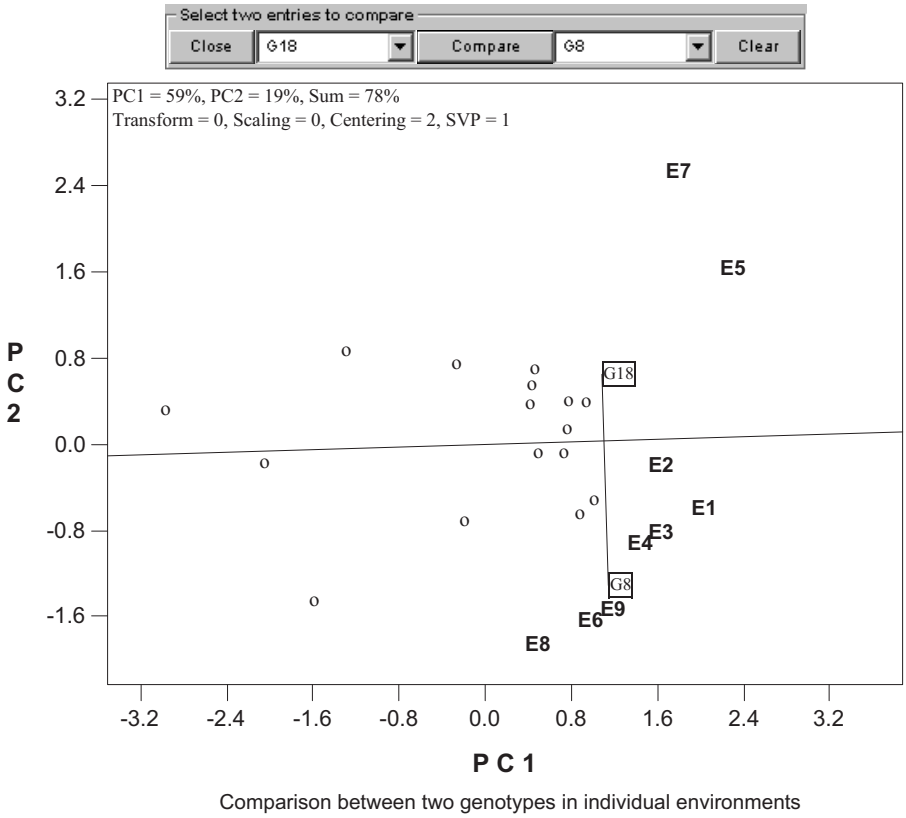

Fig. 11. Comparison between two genotypes in their performances in individual environments.

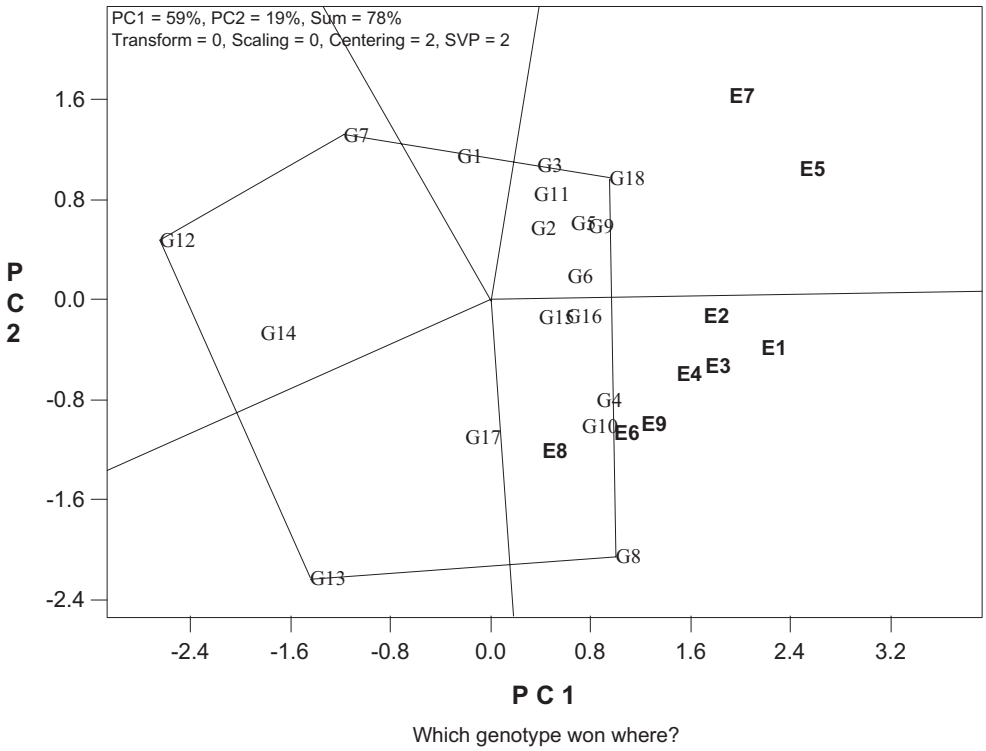

Fig. 12. The which-won-where view of the GGE biplot to show which genotypes performed bets in which environments.

are furthest from the biplot origin so that all other genotypes are contained within the polygon. Then perpendicular lines to each side of the polygon are drawn, starting from the biplot origin.

The interpretations are as follows:

(1) Genotypes located on the vertices of the polygon performed either the best or the poorest in one or more environments.

(2) The perpendicular lines are equality lines between adjacent genotypes on the polygon, which facilitate visual comparison of them. For example, the equality line between G18 and G8 indicates that G18 was better in E7 and E5, whereas G8 was better in the other environments. The equality line between G18 and G7 indicates that G18 was better than G7 in all environments. Note that G3 and G1 are located on the line that connects G18 and G7. This means that the rank G18 > G3 > G1 >G7 was true in all environments.

(3) The equality lines divide the biplot into sectors, and the winning genotype for each sector is the one located on the respective vertex. In this example, the nine environments fall into two sectors. G18 was the winner in environments E7 and E5, and G8 was the winner for the other environments. This pattern suggests that the target environment may consist of two different mega-environments and that different cultivars should be selected and deployed for each.

(4) As with Fig. 7, interpretation of Fig. 12 is based on the inner product property of the biplot and it is not altered by different singular value partitioning methods. However, environment-focused partitioning (SVP = 2) is preferred because it correctly shows the relationships among environments.

Comparison Among Three Genotypes

The which-won-where function can be very useful in comparing among three genotypes, because when the GGE biplot contains only three genotypes (i.e., when the other genotypes are deleted from the data), it will explain 100% of the variation due to G and GE (Fig. 13). For the same reason, the comparison is more accurate than the compare-two-genotype method (Fig. 11) in a biplot that does not explain 100% of the total G + GE. Therefore, it is recommended that this procedure be used even if the purpose is to compare two genotypes. GGEbiplot has a function that allows any subset of the full data to be easily selected for biplot visualization. In the example of Fig. 13, G18 was best in E5 and E7, G8 was best in E3, E4, E6, E8, and E9, whereas G10 was the best in E1. The performances of these three genotypes were about the same in E2, and G8 and G18 were very similar in E4. Note again SVP =1 was used in Fig. 13 for appropriate comparison among genotypes.

BIPLOT ANALYSIS SYSTEM FOR THREE-WAY MET DATA ANALYSIS

The basic structure of MET data is a genotype-environment-trait three-way table, which can be organized into various

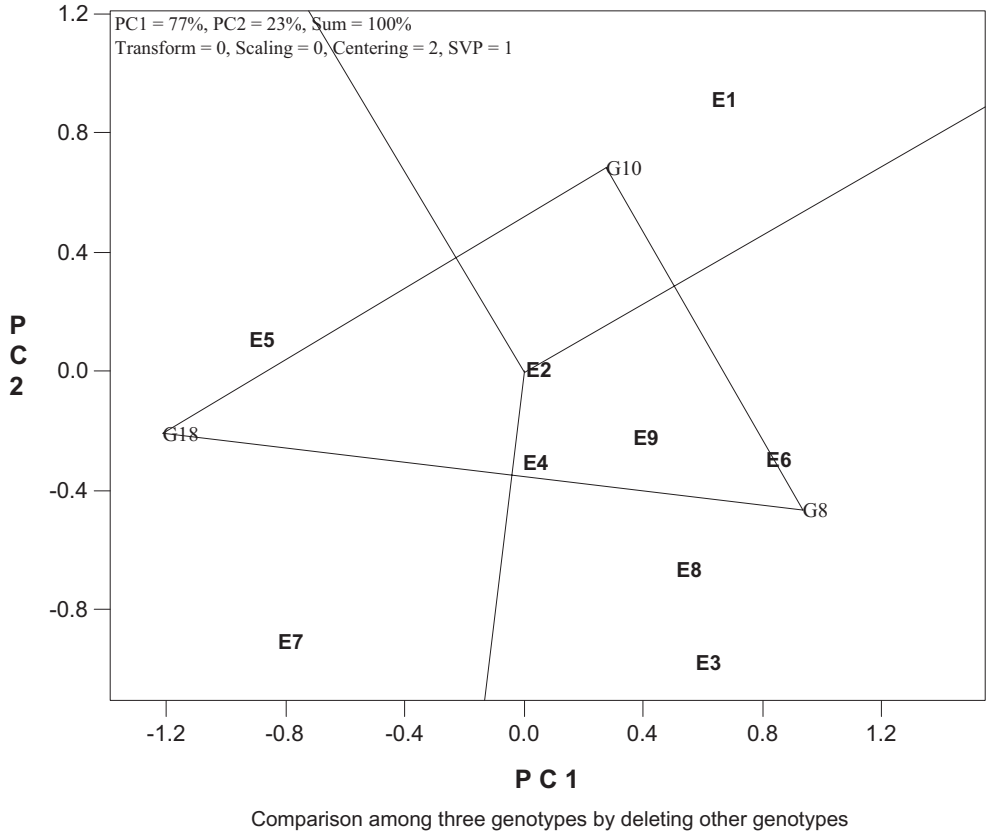

Fig. 13. Comparison among three genotypes.

two-way tables, which can then be studied in a biplot so that various questions can be graphically addressed. A full understanding of the three-way MET data involves understanding all of these two-way tables, although they are not equally important for a particular purpose. GGEbiplot reads location by genotype by trait three-way data or year by location by genotype by trait four-way data and provides options for biplot analysis of various two-way tables as discussed below.

Genotype-by-environment Tables

A three-way dataset can be dissected into genotype by environment tables for each trait, which can be studied using biplots as described in detail in the above sections. Although the genotype by environment two-way table of yield is most studied, it is possible and may be beneficial to study two-way tables of other traits as well.

Genotype-by-trait Tables

From a genotype by environment by trait three-way table, genotype-by-trait tables in any single environment, averaged across all environments, or averaged across a subset of the environments can be generated and investigated using biplots. Biplot analysis of genotype by trait tables is a typical example of biplot analysis of multivariate data. The model for biplot analysis of genotype by trait data is SVD of the trait-standardized two-way table, i.e., Eq. 15 with s_j being the standard deviation for trait j. A genotype by trait biplot can help understand the relationships among traits (breeding objectives) and can help identify traits that are positively or negatively associated, traits that are redundantly measured, and traits that can be used in indirect selection for another trait. It also helps to visualize the trait profiles (strength and weakness) of genotypes, which is important for parent as well as variety selection (Yan and Kang 2003). Most of the functions described in previous sections for biplot analysis of genotype by environment tables are applicable to genotype by trait data. To avoid unnecessary duplication, we will only present an example on how a genotype by trait biplot can assist in parent selection in breeding and genetics research.

The biplot in Fig. 14 presents data of 18 covered spring oat varieties determined for four traits in the 2004 Ontario oat performance trials: yield, groat percentage, oil, and protein concentration. It is trait-metric preserving (SVP = 2) and is, therefore, appropriate for visualizing the relationships among the traits. With the knowledge that higher yield, groat, and protein and lower oil are desirable for milling oat varieties, the purpose of this exercise is to formulate crosses for breeding better milling oat varieties as well as for studying the genetics of groat and oil content. The following can be seen from Fig. 14:

(1) Across the 18 tested genotypes, yield and groat were positively associated (an acute angle). These two traits

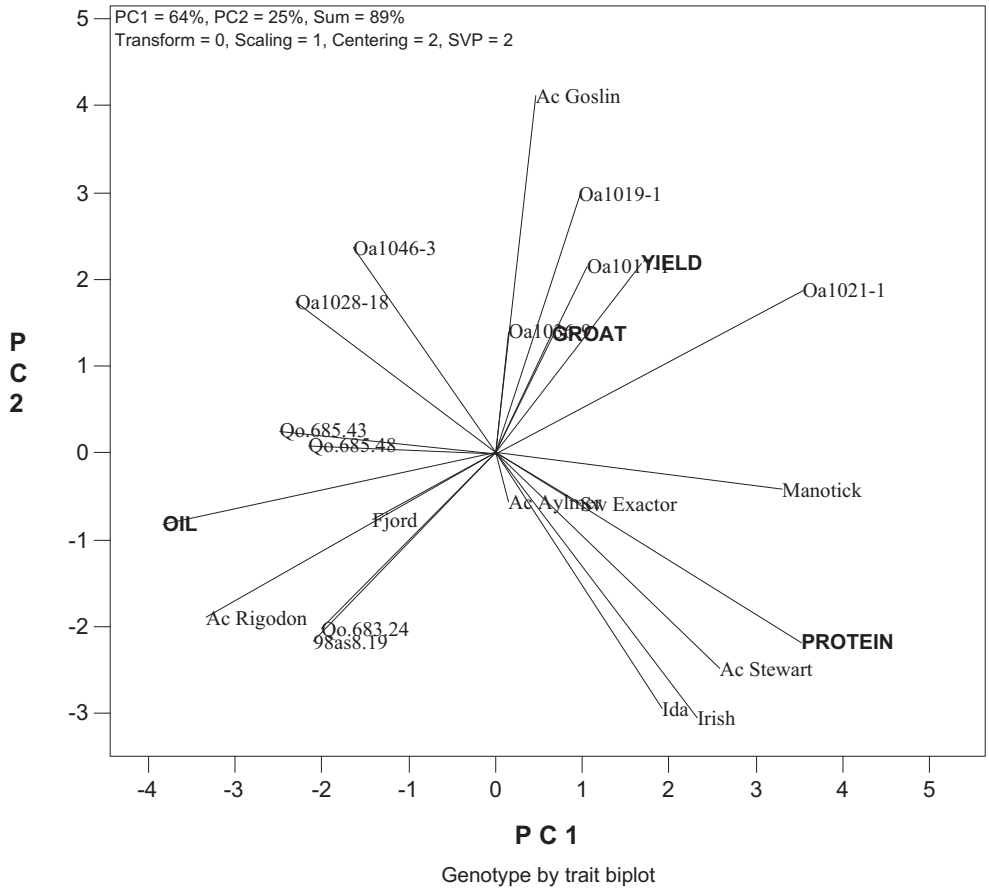

Fig. 14. A genotype by trait biplot representing 18 spring oat genotypes measured for four traits. Data from 2004 Ontario Oat Performance Trials, averaged across locations.

were negatively correlated with oil concentration (obtuse angles), and they were independent of protein concentration (near right angles). Oil and protein were negatively correlated (an obtuse angle). These relationships suggest that it is possible to combine higher yield, higher groat, higher protein, and lower oil in a single genotype.

(2) AC Goslin, a proven good milling variety, had the highest yield and groat, lower than average oil, and lower than average protein. It would be ideal if Goslin had higher protein content. Figure 14 indicates that "OA1021-1" combined all favorable attributes: it had similar yield and groat to Goslin but had higher protein and lower oil than Goslin.

(3) AC Rigodon was located opposite to OA1021-1 relative to the biplot origin because its trait profile was opposite to that of OA1021-1: it had the highest oil, the lowest yield and groat, and intermediate protein. It is, therefore, highly undesirable for milling. However, it might be a good parent for studying the genetic determination of oil and groat in oat. Therefore, OA1021-1 × AC Rigodon may make a useful cross for this purpose.

(4) AC Stewart had the highest protein content, intermediate groat and yield, and lower-than-average oil. If it is desirable to further improve the protein level of Goslin and OA1021-1, crosses of Stewart × Goslin and Stewart × OA1021-1 may be useful.

(5) In addition, many other relationships can be revealed from Fig. 14. For example, Cultivars AC Stewart, Ida, and Irish constitute a group of genotypes with similar trait profiles; QO685.43 and QO685.48 form another group with similar trait profiles, etc. It would be rational to guess that the genotypes within each group share similar origins/parentages.

Environment-by-trait Table

From a genotype-environment-trait three-way table, trait-by-environment tables for one genotype or averaged across genotypes can be generated and studied using biplots. Such biplots may be useful in studying trait by environment interactions and environmental correlations among traits. This type of analysis may be more relevant to production agronomists who are interested in knowing which environments are more favorable (or unfavorable) for production in terms of a particular trait.

Phenotype-by-trait Table

Treating each genotype-environment combination as a phenotype, a genotype-environment-trait three-way becomes a

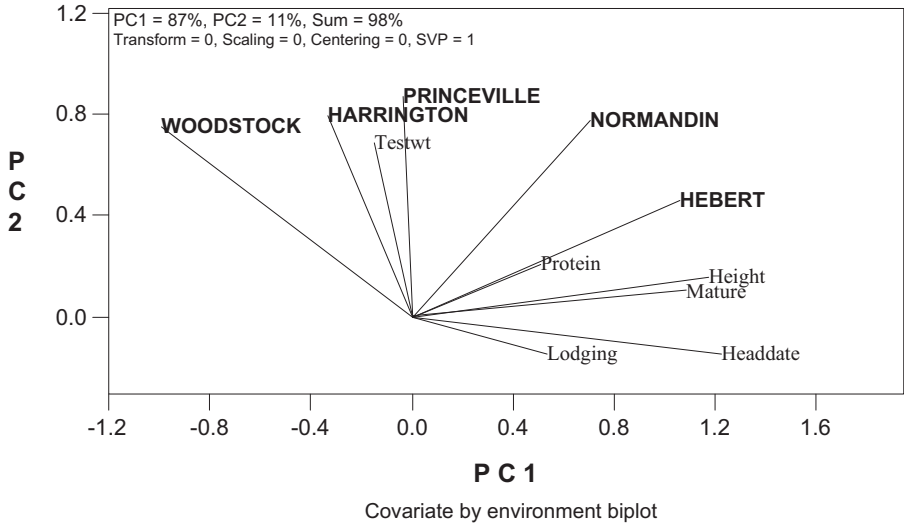

Fig. 15. A genetic covariate by environment biplot to interpret the genotype by location interaction of oat yield using genetic values of explanatory traits, based on 2003 Eastern Screening Trials data conducted by oat breeding program at ECORC, Ottawa.

phenotype by trait table. Biplot display of this table facilitates understanding the phenotypic correlations among traits (Lee et al. 2003).

Genotype by Trait × Environment Table

A genotype-environment-trait three-way table is a genotype by trait × environment two-way table if measurements of the same trait in different environments are treated as different traits. Such a two-way table can be analyzed in the same way as for an ordinary genotype by trait table but differences among genotypes can be more thoroughly studied.

Genetic Covariate by Environment Table

In MET data, one trait (usually yield) can be regarded as the target trait while other traits treated as explanatory traits or covariates of the target trait. Yan and Tinker (2005a) reported a "genetic covariate by environment biplot" whereby the G and GE of the target trait (yield) can be interpreted in terms of covariate by environment interactions. This analysis involves the following steps: (1) generating a genotype by environment table for yield, averaged across replicates; (2) generating a genotype by explanatory trait table, averaged across environments; (3) from these two tables, calculating the correlation coefficient between yield and each of the explanatory traits in each of the environments, resulting in an explanatory trait (covariate) by environment two-way table of correlation coefficients, and (4) displaying the covariate by environment table in a covariate by environment biplot by subjecting it to SVD without centering (Centering = 0) or scaling (Scaling = 0), corresponding to Eq. 10. The whole process is completed by a few mouse-clicks using GGEbiplot.

The covariate by environment biplot in Fig. 15 is based on the MET data of 2003 Eastern Oat Screening Trials conducted by the Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada at Ottawa. Thirty spring oat breeding lines and cultivars were tested at five locations representing eastern Canada: Woodstock in New Brunswick, Harrington in Prince Edward Island, and Hebertville, Normandin, and Princeville in Quebec. The entries were evaluated for agronomic traits including yield, plant height, days to heading, days to maturity, and lodging score, and for quality traits including kernel weight, test weight, groat percentage, oil content, and protein content.

The following interpretations can be drawn from Fig. 15:

(1) The five environments were quite different in terms of yield-trait relations, as indicated by the obtuse angle between Hebertville and Woodstock. This pattern is consistent with the GGE pattern based on the genotype by environment data of yield (biplot not shown).
(2) Days to heading, days to maturity, plant height, and test weight had stronger associations with yield (longer vectors) than lodging scores and protein concentration (shorter vectors). Days to heading, days to maturity, plant height, and test weight may, therefore, explain the observed G and/or GE of yield.
(3) Genotypes with greater test-weight tended to have yield in all environments, as indicated by its acute angles between test weight and the environments. Therefore, test weight explains some of the observed G for yield.
(4) Genotypes with greater values of days to heading tended to have higher yield in Herbertville and Normandin (acute angles) but lower yield in Woodstock and Harrington (obtuse angles). They tended to have average yield in Princeville (near-right angle). Therefore, days to heading explained some of the observed GE for yield.
(5) The associations of days to maturity and plant height with yield were similar to that of days to heading with yield.
(6) Figure 15 suggests possible indirect selection strategies when direct selection for yield is not possible. Later and taller cultivars should be selected for Herbertville and Normandin, whereas earlier and shorter varieties should be selected for Woodstock and Harrington to maximize

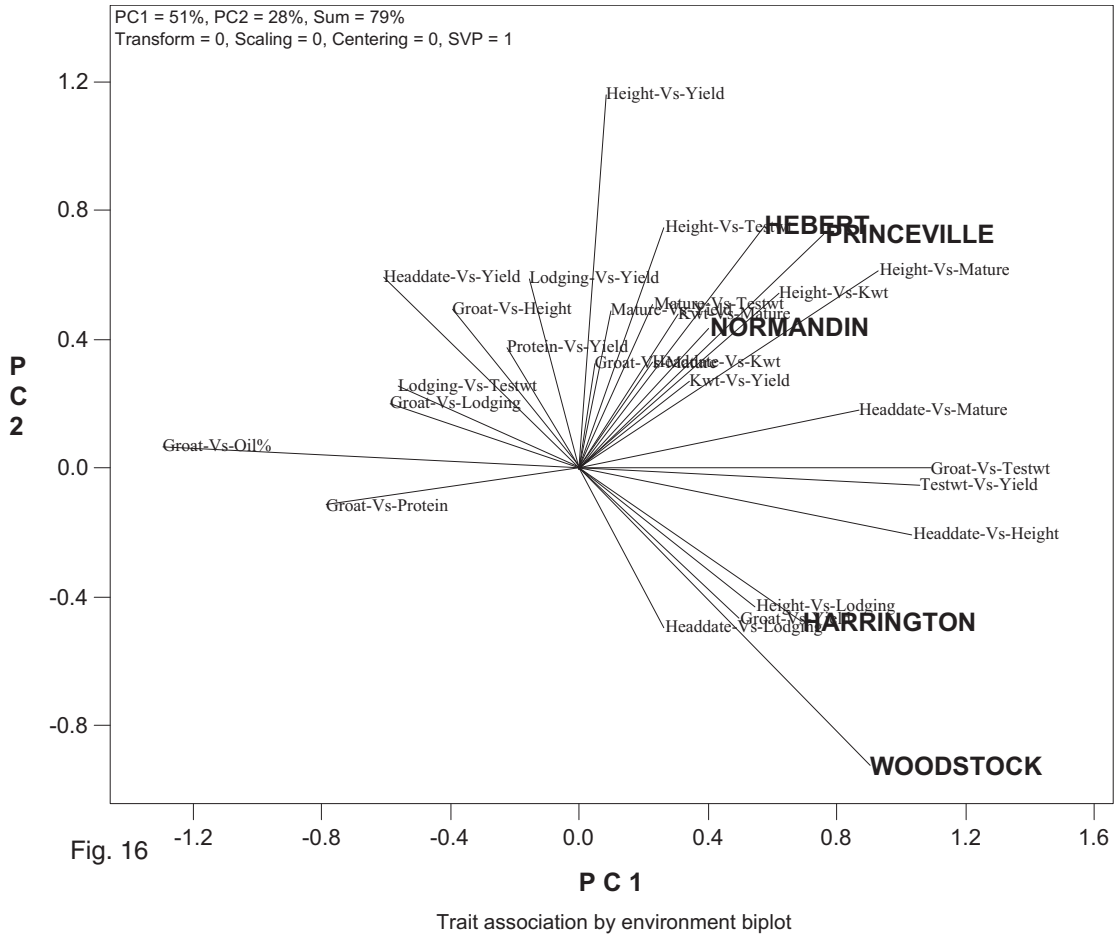

Fig. 16

Fig. 16. A trait association by environment biplot, based on the 2003 Eastern Screening Trials data conducted by oat breeding program at ECORC, Ottawa.

yield in different environments. Greater test weight, however, is desirable for all environments.

When interpreting a genetic covariate by environment biplot like Fig. 15, it should be kept in mind that the associations are between the genotypic component of the covariates (i.e., averaged over environments) and the yield in each environment, not the association between the covariate and yield within an environment (see next reference to Fig. 16). For example, it should be interpreted as "Genotypes with higher test weight tended to have higher yield in all environments" rather than "Test weight had positive associations with yield in all environments".

When genetic covariates are genetic markers, the covariate-effect biplot can be used to identify QTL based on phenotypic data from multiple environments (Yan et al. 2005). The genetic marker by environment biplot (or "QQE biplot", Yan and Tinker 2005b) can be used to identify genetic regions that are associated with yield (or other traits) in one or more environments (QTL identification), to visualize the effects of the QTL in individual environments, and to study QTL by environment interactions, which automatically lead to suggestions on strategies of marker-assisted selection specific to different (mega-) environments.

Trait Association by Environment Table

From a genotype-environment-trait three-way table, a trait association by environment (TAE) two-way table can be generated and analyzed in a biplot using GGEbiplot. When the TAE biplot procedure is activated, GGEbiplot generates a list of all traits present in the data and allows any number of the traits to be selected for analysis. If the number of selected traits is N, a list of $N(N-1)/2$ possible pair-wise associations will be formulated. Next, for each pair of traits, and within each environment, a correlation coefficient will be calculated across all genotypes tested in the respective environment, resulting in a trait-association by environment two-way table of correlation coefficients. This table will then be displayed in a TAE biplot.

Figure 16 is a TAE biplot based on the aforementioned 2003 Eastern Screening Trials data. The following can be seen from this biplot:

(1) The five environments fell into two distinct groups: one group consisted of three environments, all in Quebec,

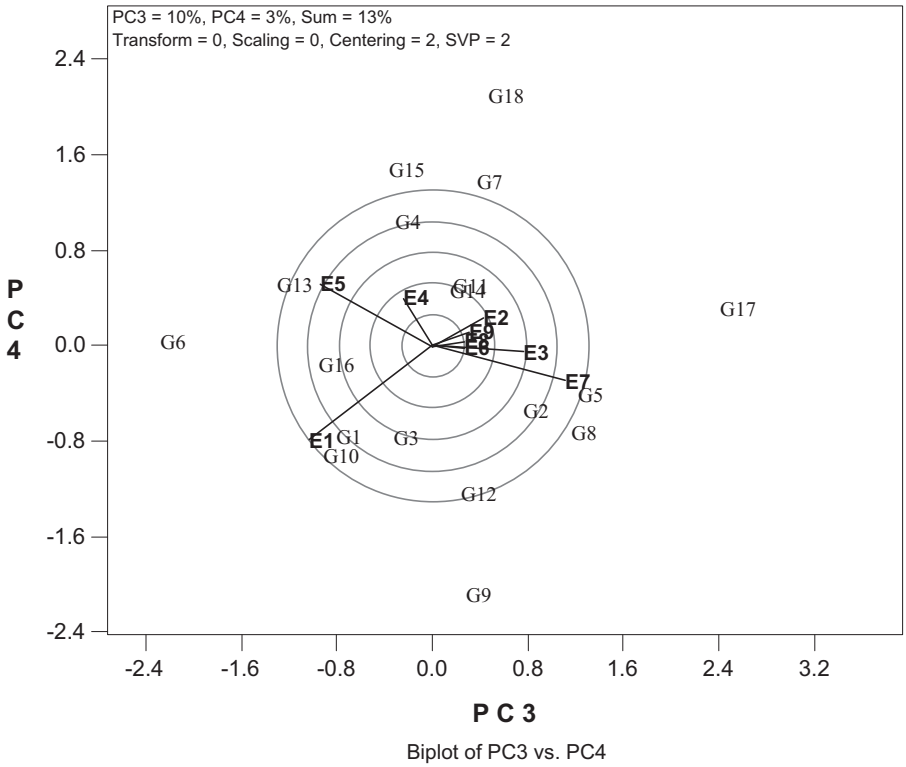

Fig. 17. A secondary biplot of PC3 vs. PC4.

and the other consists of the two maritime locations, Harrington (Prince Edward Island) and Woodstock (New Brunswick).

(2) The Quebec sites were characterized by strong positive associations of height vs. yield, height vs. test weight, height vs. kernel weight, and height vs. days to maturity. Therefore, taller genotypes had higher yield at these sites.

(3) The maritime sites were characterized by strong positive associations of groat vs. yield, days to heading vs. lodging, and height vs. lodging, and negative associations of height vs. yield, lodging vs. yield, etc. Thus, at these locations, taller and later genotypes tended to lodge more and yield less.

(4) The five locations were common in the positive associations of days to heading vs. days to maturity, days to heading vs. plant height, groat vs. test weight, and test weight vs. yield, and the negative association of groat vs. oil content.

Compared with Fig. 15, it is clear that Fig. 16 is more informative and represents a better use of the information contained in the MET data.

Figure 16 was based on MET data from the same year and the same set of genotypes. A TAE biplot can be generated across multi-year MET data in which the genotype sets are different from year to year (Yan et al., personal communication). When this is the case, the genotypes are treated as random samples and this factor should be taken into account in the biplot interpretation.

FREQUENTLY ASKED QUESTIONS ON BIPLOT ANALYSIS

How Many Principal Components are Needed to Represent the Data?

Usually biplot analysis uses only the first two PC, i.e., the primary biplot, to approximately display a two-way table. When the first two PC do not explain 100% of the two-way table that is to be investigated, which is usually the case, it is legitimate to ask if the primary biplot adequately displays the pattern of the two-way table and how many PC are needed to achieve this.

This can be answered by examining the size of each PC. Assume that the two-way table to be investigated has m rows and n columns. The maximum number of PC that are required to fully represent the two-way table is $K = \min(m, n)$. If there are no linear correlations either among the rows or among the columns, then the proportion of the total variation explained by each PC should be exactly $1/K$. When there are some linear correlations among the rows or among the columns, the proportion of variation explained by the first few PC would be more than $1/K$, while that for others would be less than $1/K$. Among the output of GGEbiplot is an "Information Ratio" (IR) for each PC, which is the proportion of variation explained by each PC divided by $1/K$. Therefore, any PC with an IR value substantially smaller than 1.0 carries little information, whereas a PC with an IR value substantially greater than 1.0 carries important patterns, i.e., summarizes information from different columns

and/or rows. Therefore, all PC that have an IR value not substantially smaller than 1.0 need to be considered for approximating the two-way table but for revealing the most important patterns, a PC should be retained only if has an IR substantially greater than 1.0. For the data of Table 1, the IR values of the first four PC were 5.31, 1.71, 0.90, and 0.27, suggesting that two PC are sufficient in revealing the patterns but three PC might be needed to approximate the data.

It should be pointed out that using IR as a criterion for retaining PC may lead to too few PC when there are dominating patterns, which can mask weaker patterns that may be more relevant. On the other hands, too many PC can be retained when there are no strong patterns. This problem can be largely avoided by selecting an appropriate model (Eqs. 10 to 14) for a particular research objective. For example, for genotype and test environment evaluation, Eq. 13 should be selected. For studying the GE pattern, Eq. 14 should be selected.

A graphical method to see if the primary biplot of PC1 vs. PC2 is sufficient is to examine whether there is additional pattern in the secondary biplot of PC3 vs. PC4 (Yan and Kang 2003; Yan and Tinker 2005b). For sample, Fig. 18 presents a biplot of the PC3 vs. PC4 that complements the primary biplot of PC1 vs. PC2 in Fig. 2. Environments E1, E5, and E7 have the longest vectors due to their relatively large variation in PC3, implying that Fig. 2 does not explain all the variation of these environments. Figure 17 suggests that the relation between E5 and E7 was not as close as Fig. 2 suggested. It also suggests that E1 was not as closely related to the other six environments as suggested in Fig. 2. Thus, the primary biplot (Fig. 2) and the secondary biplot (Fig. 17) collectively suggest that three PC are needed to fully present the data, consistent with the suggestion by the IR values of the PC. The primary biplot and various secondary biplots can be easily generated using GGEbiplot.

Many cross-validation methods have been used in determining the number of PC required to optimally approximate a two-way table. Recognizing that a genotype by environment two-way dataset is a mixture of patterns and noise (caused by random errors) and assuming that larger PC contain a greater pattern-over-noise ratio than smaller ones, Gauch (1988) proposed a "predictive success" criterion to determine the number of PC required to minimize the prediction error of the cell means of a genotype by environment table, using a drop-a-replicate procedure. Alternatively, Crossa and Cornelius (1993), Cornelius and Seyedsadr (1997), and dos S. Dias and Krzanowski (2003) adopted a drop-a-cell approach to achieve the same objective. More recently, Cornelius and colleagues developed a shrinkage factor approach [reviewed by Moreno-González et al. (2003)] to estimate the cell means and to discard principal components that carry little information.

When the experimental error mean square can be estimated, the heuristic approach proposed by Gauch and Zobel (1996) may be more useful for determining the number of required PC. For any dataset, an ANOVA table is generated first (Table 2). The expected noise SS for each variation source is estimated by its degrees of freedom (d.f.) multiplied by the error mean square, and the expected pattern SS is estimated by the total SS for the source minus its expected noise SS. The expected pattern SS vs. total SS ratio is then calculated for each variation source. For the 1993 Ontario winter wheat performance trial data (Table 1), the expected pattern percentage for G + GE is 84.5% (Table 2), meaning that a GGE biplot should explain about 84.5% of the total G + GE to be regarded as optimally approximated the G + GE pattern. Since the first three PC explained 59, 19, and 10% of the G + GE, respectively, the G + GE information contained in the data is slightly under fitted by a 2D GGE biplot but slightly over fitted by a 3D GGE biplot.

What if More than Two PC are Required?

If more than two PC are needed to adequately represent the two-way table, the primary biplot consisting of PC1 and PC2 still displays the most important pattern in the table. To achieve a fuller understanding of the data, however, the following proposals can be considered. The first proposal is to use a rotating 3D biplot that displays the first three PC (www.ggebiplot.com/3D-BiplotViewer.htm). For most cases, a rotating 3D biplot should suffice for revealing the most important patterns in the data. A static 3D biplot, however, is never more informative than a 2D biplot of the first two PC.

The second approach is to divide the data into subsets based on the pattern in the primary biplot of PC1 vs. PC2 and then use multiple biplots to examine each of them (Yan and Tinker 2005a, b). For example, Figs. 18 and 19 are constructed based on the pattern in Fig. 2. The difference between E1 and the other six environments is immediately revealed in Fig. 18, so is the difference between E5 and E7 in Fig. 19. Biplots based on any subset of the full data can be easily generated using GGEbiplot.

Can Biplots be Used for Hypothesis Testing?

Since there is no uncertainty measure in a biplot, the answer is "no". Biplots are an excellent tool for reducing the dimensionality of the data and allowing the researcher to visualize and explore relationships among rows, relationships among columns, and interactions between rows and columns of a two way table. Biplots complement but cannot replace tests of significance in MET data analysis particularly when an important decision is to be made. For example, while a biplot gives a convenient two dimensional picture approximating correlations among environments (or conversely genotypes), conclusions about specific correlations should be verified by checking the actual correlations and their significance; this information is also provided by GGEBiplot.

Can Biplots Reveal Non-linear Patterns?

Principal component analysis is useful only if linear correlations exist among the rows or among the columns. It is also useful for summarizing two-way tables in which the relationships among rows or columns can be easily transformed to linear ones, e.g., through logarithm or square root, or inverse transformation, which are options in GGEbiplot. Since most nonlinear but monotonic relationships can be easily transformed into linear relationships, principal component analysis and, therefore, biplot analysis, are widely applicable. However, some

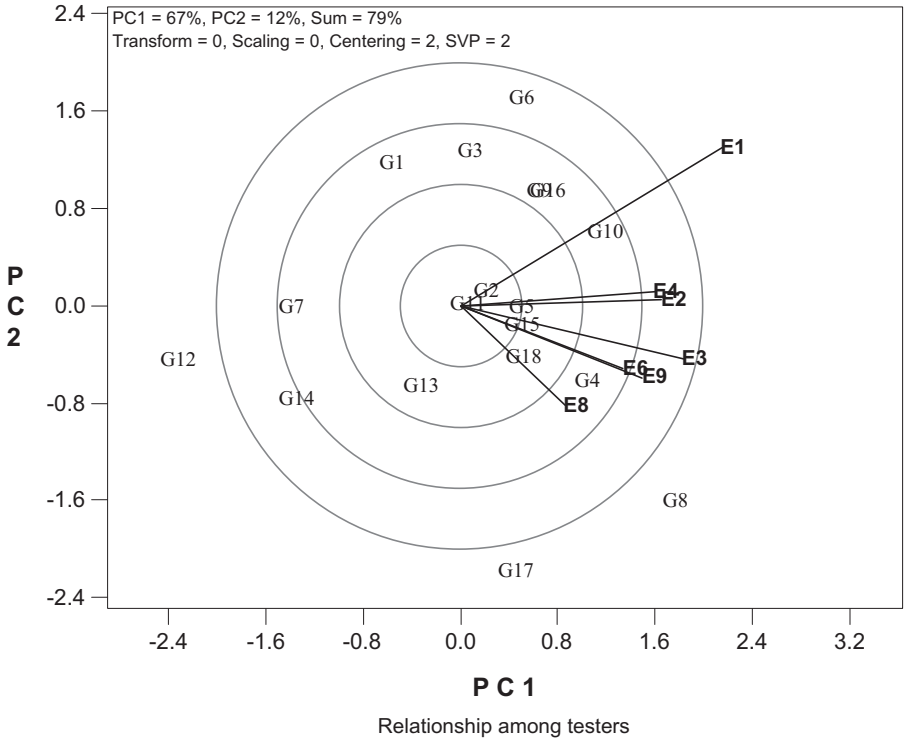

Fig. 18. A GGE biplot based on a subset of seven environments.

Table 2. ANOVA table, expected noise SS, expected pattern SS, and expected pattern ratio for each variation source or source combination for the yield data of the 1993 Ontario winter wheat performance trials

Source	DF	SS	MS	F	Expected error SS	Expected pattern SS	Expected pattern %
Total	646	740.1					
G	17	88.6	5.21	30.9	2.9	85.7	96.8
E	8	447.7	55.96	331.6	1.4	446.3	99.7
GE	136	78.5	0.58	3.4	23.0	55.5	70.7
Block (E)	27	48.0	1.78				
Error	458	77.3	0.17				
G + GE	153	167.1			25.9	141.2	84.5

non-linear relationships, for example quadratic relationships with maximum or minimum values near the mid-way, cannot be revealed in biplot analysis.

CONCLUSIONS

Biplot analysis has evolved into an important technique in crop improvement and agricultural research. GGE biplot analysis provides an easy and comprehensive solution to genotype by environment data analysis, which has been a challenge to plant breeders, geneticists, and agronomists. It not only allows effective evaluation of the genotypes but also allows a comprehensive understanding of the target environment and the test environments. Specifically, biplot analysis can help one understand the target environment as a whole, i.e., whether it consists of a single or multiple mega-environments, which determines whether GE can be exploited or avoided. Within a single mega-environment, biplot analysis can help one understand the test environments: whether they are informative, representative, and unique in terms of genotype discrimination. At the same time, biplot analysis can help one evaluate genotypes in terms of both mean performance and stability across environments. Thus, GGE biplot analysis of genotype by environment data not only addresses short-term, applied questions but also provides insights on long-term, basic problems.

Genotype by environment data analysis is but one aspect of MET data analysis. Genotype-environment-trait three-way MET data can be organized or dissected into various two-way tables, which can be effectively exploited using biplots. Biplot analysis of a genotype by trait table can help the researcher to understand the crop as an integrated system with interconnected components (breeding objectives), which is the basis for establishing realistic breeding objec-

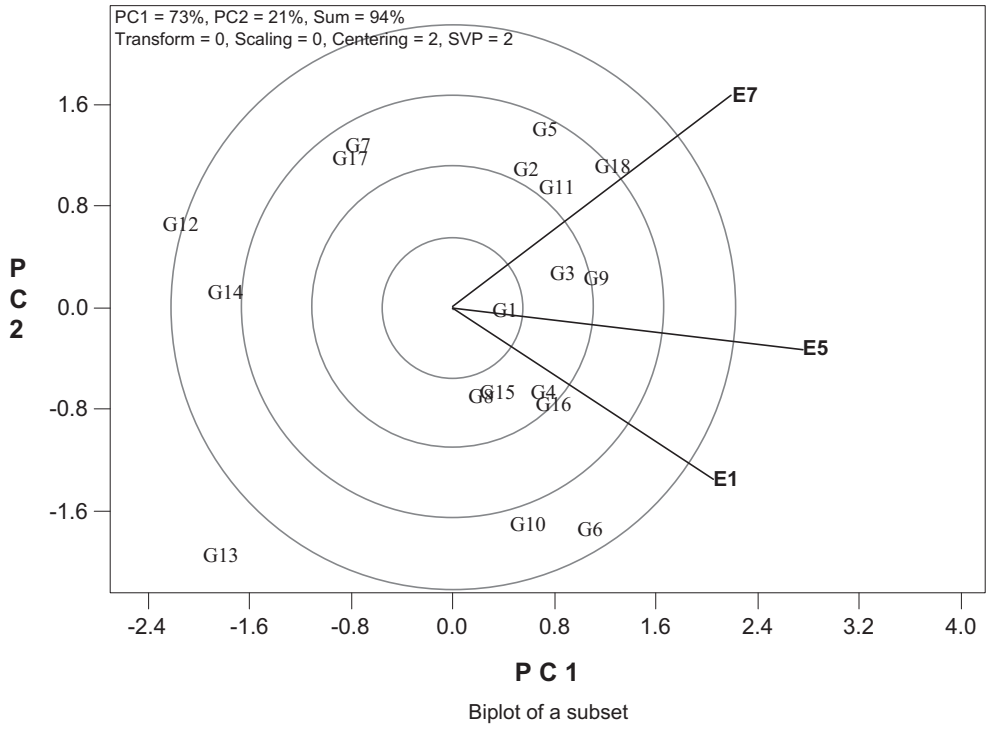

Fig. 19. A GGE biplot based on a subset of three environments.

tives and selection criteria. Simultaneously, it helps reveal the trait profiles (strength and weakness) of the genotypes, which are important for identifying superior cultivars and parents. The newly developed trait-association by environment biplots can help reveal, interpret, and explore genotype by environment interactions. Biplot analysis of other two-way tables extracted from MET data can be useful for addressing other research questions. User-friendly, feature-rich, interactive computer programs such as GGEbiplot can make biplot analysis of three-way MET data easy, informative, and interesting, and can greatly facilitate the use of biplot analysis by plant breeders and other agricultural scientists.

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