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# **REGULAR ARTICLE**

# Pattern analysis of multi-environment trials in bread wheat

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## ABSTRACT

To detect genotype-by-environment interactions (GEIs), pattern analysis (PA) was performed on yield data of 20 bread wheat (Triticum aestivum L.) genotypes tested across nine environments during the 2001-2002 growing season in the Central Anatolian Region of Turkey. Nine clusters of genotypes with similar patterns in performance, mostly reflecting their origin and pedigree, were identified. Most of the genotypes from the National Bread Wheat Improvement Program (NBWIP), Turkey, fell into one of the two broad genotypic clusters, whereas most of those from the International Winter Wheat Improvement Program (IWWIP), a collaborative breeding program among Turkey, CIMMYT and ICARDA, were clustered together in another cluster. Six clusters of the environments with similar trends in discriminating genotypes were identified; discrimination of the environments tested indicated that there existed two distinct main types of environments: rain-fed and irrigated. However, the rain-fed environment E2 (Eskisehir) joined the irrigated environments, whereas the irrigated environment E9 (Haymana) was clustered with the rain-fed environments. Irrigated environments E6 (Konya) and E7 (Cumra) contributed trivially to discrimination of the genotypes, as they exhibited a pattern that was almost identical to that of the irrigated environments. The first two principal components explained 53.3% of the total variation in GEI data. This study revealed that the genotypes with a common parent in their pedigree or with the same origin tended to be clustered together. All the genotypes from the NBWIP and one-third of genotypes from the IWWIP were stable; therefore, the NBWIP could be strengthened by introductions from the IWWIP. Environmental cluster analysis effectively identified trials that received rainfall and supplementary irrigation. It might be possible to reduce the number of test environments by eliminating one or more of those that differentiate among genotypes in a similar manner (e.g., irrigated environments E6 in Konya and E7 in Cumra).

**Key Words**: bread wheat (T. aestivum L.); cluster analysis; multi-environment trials; genotype-byenvironment interaction; pattern analysis; principal component analysis.

**Abbreviations:** CA—cluster analysis; GEI—genotype-by-environment interaction; IWWIP—International Winter Wheat Improvement Program; NBWIP—National Bread Wheat Improvement Program; METs—multi-environment trials; PA—pattern analysis; PCA—principal component analysis.

#### INTRODUCTION

Genotype-by-environment interaction (GEI) is the differential response of genotypes evaluated under different environmental conditions. It is a complex phenomenon as it involves environmental (agro-ecological, climatic and agronomic) conditions and all physiological and genetic factors that determine the plant growth and development.

There are many statistical methods for assessing, studying and interpreting GEIs (Flores et al., 1998; Hussein et al., 2000; Sabaghnia et al., 2006). Some methods are based on linear regression of a genotype means on environmental index, e.g., Finlay and Wilkonson (1963) and Eberhart and Russell (1966). Nonparametric stability statistics, requiring no statistical assumptions, have been proposed by Hühn (1990) and Kang and Pham (1991). Many of the nonparametric methods have recently been compared by Sabaghnia et al. (2006). Three newer methods, which help identify important characteristics of GEI, are worth mentioning: the Additive Main effects and Multiplicative Interactions (AMMI), which was popularized by Gauch and Zobel (1988), Pattern Analysis (PA), which was developed and updated by Watson et al. (1996), and GGE Biplot Analysis, which was developed by Yan (2001) and thoroughly documented by Yan and Kang (2003).

Genotype-by-environment interaction data obtained from multi-environment trials (METs) across a wide range of environments can be investigated by PA to identify genotypes with similar responses across environments, and to identify those environments that discriminate among genotypes in a similar manner (Cooper and Delacy, 1994; Alagarswamy and Chandra, 1998; Delacy et al., 2000). Pattern Analysis is based on the joint complementary use of cluster (CA) and principal component analysis (PCA) to study different aspects of response patterns of genotypes. Since there is an exponential increase in a number of pair-wise comparisons with an increase in a number of environments, inspection of individual comparisons becomes impractical. To overcome this problem, the use of PA has been proposed (Cooper and Delacy, 1994). Inspection of two-way response plots from environmental and genotypic clusters or the biplots from PCA provides an alternative and complementary way of examining the relationships among genotypes and environments (Cooper and Delacy 1994). In particular, a biplot represents a versatile graphical approach for analyzing METs (Yan, 2001; Yan and Kang, 2003).

The objectives of this study were to (i) interpret magnitude and causes of GEI via PA of yield performances of 20 bread wheat genotypes tested across nine environments, (ii) identify high yielding genotypes on the basis of differential genotypic responses to environments, and (iii) identify similar or redundant environments to help streamline performance trials.

#### **MATERIALS AND METHODS**

Twenty bread wheat genotypes were grown in nine environments, including five rainfed environments: Cumra (E1), Eskisehir (E2), Konya (E3), Obruk (E4) and Haymana (E5), and four irrigated environments: Konya (E6), Cumra (E7), Eskisehir (E8) and Haymana (E9), during the 2001-2002 growing season at the Central Anatolian Region of Turkey. Of the 20 advanced lines used, eight were from the NBWIP and 12 from the IWWIP (Table 1). An experimental layout was a randomized complete block design with four replications. Sowing was done with an experimental drill in 1.2 m × 7 m plots, consisting of 6 rows spaced 20 cm apart. The seeding rate was 450 seeds m-2 for the irrigated and 550 seeds m-2 for the rain-fed environments. Fertilizer application was 27 and 36 kg of N ha<sup>-1</sup> and 69 and 92 kg of P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> at the planting, and 50 and 80 kg of N ha<sup>-1</sup> at stem elongation stage for rain-fed and irrigated environments, respectively. Experiments E6 and E7 (Table 2) were irrigated twice, i.e., before and after the heading stage, 50 mm each time, whereas those in E8 and E9 were irrigated once, 80 mm and 50 mm, respectively, following the heading stage. Harvesting was done in 1.2 m × 5 m plots with an experimental combine. Grain yield was obtained by expressing plot grain yields on ha basis (t ha-1). Details of 20 genotypes and nine environments of study are given in Tables 1 and 2, respectively.

Table 1. Code, origin, pedigree, selection history, yield mean (t ha-1) and cluster of genotypes.

Code	Pedigree and selection history	Origin	Mean (t ha-1)	Cluster
1	TX71A1039-VI*3/AMI(TX81V6603)//MVR16-85	NBWIPa	4.66 g*	IV
	BDKE 920008 -2F5 BD-0BD			
2	PLK70/LIRA"S//30-KZ-1	NBWIP	5.15 bd	V
	BDKE 920012 -2F5 BD-OBD			
3	ES 14/FLAMURA 85	NBWIP	5.09 ce	111
	YE 7907 6F5 BD OBD			
4	SDY/ALD/3/NAI60/HN7//BUC/4/KEA/TOW/5/YAN7578.128 CMWW91M00067T 1F5 BD OBD	IWWIP <sup>b</sup>	4.76 g	VI
5	SDY/ALD/3/NAI60/HN7//BUC/4/KEA/TOW/5/YAN7578.128	IWWIP	5.26 b	Ι
	CMWW91M00067T 3F5 BD OBD			
6	83(1).16.1//KA/NAC	IWWIP	4.73 g	II
	CMSW91M 00321S 6F5 BD OBD			
7	TAST/SPRW//BEZ2B/CGN/4/INIA66(R)//HBGN/DRC/3/BEZ ICWH 900759-0AP-0YC-0YC-0YC-1YC-0YC	IWWIP	4.70 g	VI
8	TAM200/KAUZ	IWWIP	5.11 be	Ι
	960686 CMSW91M 00414S-OSE-OYC-1YC-OYC			
9	LFN/VOGAF//LIRA/5/K134(60)/4/TOB/BMAN//BB/3/CAL/6/F339P1.2	IWWIP	4.74 g	VII
	TCI 935039-OSE-OYC-3YE-OYC		0	
10	ATAY/3/MII/GLEN//TRT	IWWIP	5.42 a	V
	MX-TCI CMSW90M407-0YC-0YC-0YC-3YC-0YC			
11	BILINMIYEN967	IWWIP	5.08 ef	VI
	F2 96 7-0SE-4YA-4YC-0YC			
12	ATLAS 66//HYS/7C	NBWIP	4.92 f	V
	BDKE 900096 -255 BD OBD	112111	1.7 - 1	•
13	BOLAL 2973/THUNDERBIRD	NBWIP	4.96 ef	Ш
10	BDKE 900003 -1E5 BD OBD	112111	100 01	
14	I ND/SW0791095A/4/YMH/TOB//MCD/3/I IRA	IWWIP	4 75 σ	VII
	ICWH90-0217 7F5 BD OBD	100001	1	• 11
15	KS2142/4/KRC66/3/TT-50-18/P101//11-50-18/VGDWVF	NBWIP	4 75 σ	IV
10	BDKF 910010 1F5 BD OBD	1 D M H	1.70 5	1.
16	BILINIEVEN	NBWIP	5.24 bc	V
10	YXX	1 D WII	5.24 00	v
17	63 122 66 2/NO / /I OV 2 E1 /2/E1 KVZ /HVS / $h$ /TIB 016 46 /	ПАЛАЛІР	4 02 f	V
17	$(0)^{-122-00-2} (100) / 100 (2.111) (5/11.10) (4/110) (4/110) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) (6/10) ($	100 0011	4.921	v
10	VEE /TSL//CDV70/2/NISE5.02/5/C124.15/COENT/2/NI10B/D14//D101/4/VDC44	ПАЛАЛІР	5 07 df	п
10	VEE/13///GRN79/3/100000/3/CL20.13/COFIN/3/1000/114//1101/4/RRC00	100 0011	5.07 ui	11
10	VV7/AU//CDV70	NIDIAIID	106 h	IV
19	NNL/AU//GKN/9	INDVVIP	4.06 n	IA
20		TT & 7T & 7T T	E 40	\$ 7111
20	TCI 922142 -OSE-OYC-3YC-OYC	100 00112	э.48 a	V 111

<sup>a</sup> National Bread Wheat Improvement Program-Turkey.

<sup>b</sup> International Winter Wheat Improvement Program-Turkey/CIMMYT/ICARDA.

\* A minor letter indicates a membership to the corresponding homogeneous group as revealed via Duncan range test.

Environment	Code	Soil properties	Rainfall + Irriga- tion (mm)	Mean (t ha <sup>-1</sup> )	Cluster
Cumra	E1ª	pH = 7.8, clayey, loam, hyromorphic alluvial	303	2.64 g*	IV
Eskisehir	E2 <sup>a</sup>	pH = 7.8, red brown	431	6.01 d	III
Konya	E3 <sup>a</sup>	pH = 8.2, clayey, alluvial	383	5.02 e	V
Obruk	E4 <sup>a</sup>	pH = 8.0 clayey, loam, brown	315	1.58 h	Ι
Haymana	E5 <sup>a</sup>	pH = 7.8, silty, loam	503	3.44 f	Ι
Konya	E6 <sup>b</sup>	pH = 8.2, clayey, alluvial	383 + 100	6.01 d	II
Cumra	Е <b>7</b> ь	pH = 7.8, clayey, loam, hyromorphic alluvial	303 + 100	6.47 a	III
Eskisehir	E8 <sup>b</sup>	pH = 7.8, red brown	431 + 80	6.35 b	II
Haymana	Е9ь	pH = 7.8, silty, loam	503 + 50	6.09 c	VI

Table 2. Code, soil properties, status of rainfall + irrigation, yield mean (t ha-1) and cluster for each environment.

<sup>a, b</sup> rain-fed and irrigated, respectively.

\* A minor letter indicates a membership to the corresponding homogeneous group as revealed via Duncan range test.

Analysis of variance of mean yield data for the 20 genotypes and nine environments was used to determine the relative magnitude of sums of squares attributable to G, E, and GEI. Before cluster analysis, the yield matrix was transformed within environments, whereby the main-effects of environment and grand mean were removed, and the remainder was divided by the within-environment standard deviation (Fox and Rosielle, 1982; Cooper and Delacy, 1994). From the transformed yield matrix, the squared Euclidean distance matrix (i.e., a dissimilarity matrix) was computed for genotypes and environments. Hierarchical agglomerative clustering (Williams 1976) was applied via Ward's clustering procedure (Ward 1963), in which incremental sums of squares were as the fusion criterion; in this method, in any part of the dendrogram, the members or clusters are joined to minimize the new within-cluster sums of squares. Dendrograms were constructed on the basis of fusion level to examine similarities in pattern of performance among genotypes (in reaction to environments) and environments (in discriminating among genotypes).

A biplot was constructed in the dimension of first two principal components, using a singular-value decomposition procedure (Gabriel, 1971; Kempton, 1984). The genotypes were represented on the biplots as the points derived from their scores for the first two components, and the environments as the vectors from the biplot origin to their points. The co-sine of angle between a pair of environment vectors approximates correlation between them (Yan and Kang, 2003). An acute angle (<900) indicates a strong positive correlation; an angle close to 900 indicates the environments are not correlated, whereas an obtuse angle close to 1800 represents a strong negative relationship (for more details, see Kroonenberg, 1995).

### **RESULTS AND DISCUSSION**

Pattern analysis showed that variation among the nine genotypic clusters and six environmental clusters accounted for only 60.1% of the GEI sum of squares, which was approximately 5.7 times larger than the within-clusters mean square. However, the reduced matrix retained only 65.2% of the total sum of squares (tables of ANOVA of PA are not given).

The dendrogram from a hierarchical clustering of 20 genotypes based on transformed yield data set across nine environments was truncated at the 9-cluster level wherein the similarity and fusion level between clusters changed noticeably, i.e., considerably more than a gradual change in the amalgamation steps (Figure 1). Cluster analysis showed that varia-

tion among the identified genotypic clusters accounted for 82.9% of the genotype sum of squares and their mean square was approximately 6.7 times the within-group mean square.

Clusters contained from one to five genotypes (Figure 1 and Table 1). As might be expected, genotypes with a common parent in their pedigree or from the same origin tended to be clustered together. Except for genotypes 2, 12 and 16, all the genotypes from the IWWIP amalgamated together at one of the two broad clusters, revealing that their response pattern was more similar than that of the remainder of the genotypes. Genotypes 1, 3, 13, 15 and 19 from the NBWIP were clustered at the other broad cluster, indicating that their selection histories and origins were similar. The NBWIP-derived genotypes were likely tolerant to drought conditions, due to the fact that most of them were crossed with the drought-tolerant genotypes, such as Kirac-66, Gerek-79 and Bolal-2973, and the fact that most of them were selected in the same target environments.

Pattern for the IWWIP-derived genotypes varied, indicating that the percentage contribution for their pedigree was different. For example, genotypes 4 and 5 had identical pedigree; however, response pattern for these genotypes was substantially different, as shown by the dendrogram. On the other hand, the IWWIP-derived genotypes tended to gather in the same part of the broad cluster, even if they originated from a drought-tolerant parent, or the genotypes well adapted to irrigated conditions tended to deviate from unity at the base of the sub-clusters (Figure 1). The dendrogram from a hierarchical clustering of nine environments based on transformed yield data set for 20 genotypes was truncated at the 6cluster level, wherein the similarity and fusion level between clusters changed by a noticeably different amount from a gradual change in the amalgamation steps (Figure 2). The among-environment clusters source of variation accounted for 96.4% of the environment sum of squares, with the mean square approximately equaling the within-cluster mean square.

The final cluster of environments comprised two broad clusters, one referring to the irrigated and the second to rain-fed environments (Figure 2). One cluster included the irrigated environments E6, E7, and E8, and the rain-fed environment E2. The other cluster included the rain-fed environments E1, E3, E4, and E5 as well as irrigated environment E9. As expected, the mean yield, 6.21 t ha<sup>-1</sup>, of the irrigated environments, including the rain-fed environment E2, was relatively higher than that for rain-fed environments together with the irrigated environment E9 (3.75 t ha<sup>-1</sup>; Table 2).

The response plot of the transformed data set of nine genotypic clusters across six environmental clusters indicated certain patterns (Figure 3). Genotypic clusters III, IV, V and VI expressed nearly no interaction with environmental clusters and therefore may be considered to be stable in performance (for yield) across all environmental clusters. All genotypes from the NBWIP and one-third of genotypes from the IWWIP were stable. This might be due to the improved genotypes being tolerant to zinc deficiency and boron toxicity as well as to drought conditions in Central Anatolia, Turkey (Cakmak et al., 1999)

The differences in magnitude and orientation of the specific effects for particular environmental clusters can be used to identify basic distinctions in adaptation of genotypic clusters. Genotypic cluster-pairs I-IV and III-VI had the greatest contrast in performance across environmental clusters. These contrasts confirmed that the IWWIP-derived genotypes exhibited reasonable differential responses across the environmental clusters. Environmental cluster I was characterized by relatively small interaction effects, whereas environmental cluster VI by large interaction effects for most of the genotypic clusters (Figure 3).

This reflected the degree of the differences in mean yield for the environments included within these clusters. Genotypes of clusters I and VIII showed the greatest adaptation to environments of the clusters I, II, III and IV, that is, these genotypes, belonging to clusters I and VIII, from the IWWIP tended to adapt to seven out of nine environments. Therefore, these genotypes had high general adaptability. Similarly, genotypes of the clusters IV and V showed good adaptive responses to the environments of the clusters V and VI, indicating that the corresponding genotypes had high specific adaptability for the these environments. On the other hand, genotypes of the clusters II, VI, VII and IX demonstrated poor adaptation to most of the environment clusters.



Figure 1. Dendrogram presenting hierarchical clustering of 20 genotypes (details of genotypes are given in Table 1).



Figure 2. Dendrogram presenting hierarchical clustering of nine environments (details of environments are given in Table 2).

The results of PCA of GEI are presented in Figure 4 as per Cooper and Delacy (1994). The first two principal components in the biplot explained 53.3% of the total variation in GEI. The environment vectors covered a wide range of the Euclidean space, indicating that the nine environments of study represented a wide range of environments (Table 2).

The maximum angle among the vectors of the irrigated environments was below 900, corresponding from irrigated environment E7 to rain-fed environment E2 (Figure 4). This suggests that these environments tend to discriminate among genotypes in a similar manner. Genotype 5 was the top-yielding in three out of four irrigated environments, whereas genotype 19 was the lowest-yielding in these environments. The rain-fed environments also showed the same degree of closeness as the irrigated environments, giving the angle of slightly greater than 900 between E3 and E9. Genotype 5 was the lowest-yielding in environments and E9.



ronment 9, whereas genotype 20 was the highest-yielding in E4 and E5 of the rain-fed environments (Table 2).

Figure 3. Response plots of nine genotype clusters over six environment clusters based on transformed yield data (details of genotypes and environments are given in Tables 1 and 2)

The environmental vector for E3 from the rain-fed environments made the angle of nearly 1800 with the irrigated environments. The genotypic discrimination of this environment was therefore expected to be almost opposite in direction to that of the irrigated environments. For example, in environment E3, genotypes 15 and 19 were high-yielding, whereas the rankings of the corresponding genotypes at almost all of the irrigated environments were below the environmental means (Table 2 and Figure 4).

High-yielding genotypes should have a large PC1-score and a small (absolute) PC2-score (high stability) (Yan and Rajcan, 2002). Genotypes 8, 16 and 17, being closer to the biplot origin, were average in their performances across the environments. Genotype 17 could be considered to be a widely adapted genotype across the environments, due to the fact that this genotype had both a larger score for PC1 and was closer to favorable environments E6 and E7 than genotypes 8 and 16 (Figure 4).

The position and perpendicular projection of genotypic points onto an environmental vector can be used to identify a genotype or genotypes having specific adaptation in that environment(s) (Yan et al., 2000). The genotypes that are farther along the positive direction of the vector tend to give higher yields, and are better adapted to those environments. Genotypes 6, 11, and 18 were adapted to the irrigated environments, but poorly adapted to the other environments. On the other hand, genotypes 3, 13, 15, 19 and 20 were well adapted to the rain-fed environments, but poorly adapted to the rest of the environments. Incidentally, genotypes 1, 4, 7 and 12 did not respond to almost all of the environments, that is, nearly all

the environments studied failed to discriminate among the corresponding genotypes (Figure 4).



Figure 4. Biplot for PC1 vs. PC2 scores obtained from yield data of 20 genotypes across nine environments. The nine environments are indicated as vectors drawn from origin. Genotypes are denoted by dots. Circles and rectangles refer to irrigated and rain-fed environments, respectively. (Details of genotypes and environments are given in Tables 1 and 2).

## CONCLUSIONS

Pattern analysis has assisted in analyzing the bread wheat testing environments leading to the identification of the existence of two mega-environment clusters (irrigated and rainfed). Within the mega-environment clusters, several sub-environment clusters were identified. The irrigated environments tended to be closer in the biplot, indicating that they similarly discriminated among these bread wheat genotypes. This reveals that it may be possible to reduce a number of bread wheat irrigated test environments and thereby economizing and optimizing the conduct of METs.

With repeatable GEI, it is possible to structure economically the METs. The results of the present study suggest the existence of two-mega environments in this MET. The results presented here, obtained via pattern analysis, are preliminary in nature. The repeatability of the pattern revealed in this MET needs to be established across a number of years, however, before this information can be used with confidence to structure the bread wheat METs.

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