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Stratification of SADC regional pearl millet testing sites based on grain yield performance of lines

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Abstract

Sequential retrospective (SeqRet) pattern analysis was used to stratify pearl millet testing sites according to their similarity of line-yield differentiation using grain yield data from 90 multi-environment trials (METs) conducted in the Southern African Development Community (SADC) region. The METs, conducted across 25 sites over 9 years were split into two sets: Set 1 (1989/1990–1992/1993) included introductory genetic materials, and Set 2 (1994/1995–1998/1999) included advanced genetic materials. Site stratification analysis from Set 1 and Set 2 partitioned the testing sites into six and five groups with R^2-values of 76 and 79%, respectively. Analysis of the cumulative dataset (1989/1990–1998/1999) clustered the 25 sites into six groups with $R^2 = 76\%$ and captured the major patterns of site similarities in Set 1 and Set 2. Based on our experience from running multi-year METs in the SADC region, the cumulative dataset was more informative in judging the relevance of site-stratification results. SeqRet pattern analysis, exploiting available highly imbalanced historical MET data, provided an objective basis for stratifying the test-sites to facilitate selection of a few representative test-sites for future testing of lines. The results from cumulative dataset suggest that future line testing could be restricted to a few sites picked from within each of the identified site-groups.

Keywords: Pearl millet; Site stratification; Sequential retrospective pattern analysis

1. Introduction

Maximization of crop productivity requires accurate selection and targeting of genotypes for appropriate production areas. The number and location of testing sites are critical factors that affect the efficiency of and potential gains from breeding. The selected testing sites must be representative of the conditions of target production areas. Within a large region, such as the Southern African Development Community (SADC), knowledge of underlying production zones within the region could help not only in choosing appropriate testing sites, but also in objective targeting of lines for maximizing production (Peterson, 1992). The availability of long-term yield data from regional trials conducted in SADC region over the past one decade provided a unique opportunity to identify intra-regional production zones based on grouping of testing sites by the line response to varying production conditions.

Plant breeders, over the years, often change both the lines and the locations in regional trials. Unlike the well-designed balanced genotype (G) × location (L) × year (Y) investigations, where both the genotypes and locations remain the same over years, the

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analysis of such regional trials is statistically more difficult due to highly imbalanced GLY data. Statistical techniques, developed over the last decade to stratify the testing sites according to their similarities in line response, attempt to account for this imbalance in GLY data basically through averaging of location proximity matrices across years. This approach tends to minimize the influence of missing data and short-term weather events or rare disease epidemics on relative relationships among the testing sites (Peterson, 1992). Based on this basic approach, Peterson (1992) and Peterson and Pfeiffer (1989) applied factor analysis on the average correlation matrix to stratify international winter wheat testing sites using 17 years of trial data. The average correlation matrix was derived from the correlation matrices from individual trial years, the correlations within a year being computed between line yields for pairs of locations. DeLacy et al. (1990) used pattern analysis technique (Williams, 1976) to stratify Australian cotton testing sites based on 6 years' data. They computed squared Euclidean distance (SED) between locations for each year and averaged the SEDs across years to produce a single average dissimilarity matrix for site classification. The individual years' dissimilarity matrices were either simply averaged or weighted by the number of lines grown in different years to obtain the single average dissimilarity matrix.

The objective of the research reported here is to stratify the pearl millet testing sites in the SADC region based on available historical grain yield data from regional trials to facilitate identification of key benchmark testing sites representative of the underlying production zones in the SADC region. The site-stratification will help to effectively use and target exchanges of germplasm and information.

2. Materials and methods

2.1. Structure and history of field trials

Initiated in 1983, and since then, implemented by ICRISAT, the Sorghum and Millets Improvement Program (SMIP) is a major crop breeding project for the SADC region. The program's major objective is to develop improved sorghum and millet lines for the drought-prone SADC region currently consisting of 14 Southern African countries. This study included grain yield data from 90 pearl millet METs conducted over a period of 9 years (1989/1990–1992/1993 and 1994/1995–1998/1999) covering 25 different locations. In the process of identifying suitable lines for recommendation to farmers, the number and composition of lines being evaluated, as well as test-sites, changed over successive years. Elite lines were often tested for 2–3 years across a range of locations in regional trials before the best lines were selected for subsequent stages of evaluation.

The major criterion in selecting the testing sites was their proximity to major pearl millet growing areas. The number of testing sites in a country was determined by the importance of the crop and the strength of the national research program. For example, there were five sites in Namibia where pearl millet is the major staple food, and four sites in Zimbabwe where the regional headquarter of SMIP is located. Zambia and Tanzania each had three sites whereas the other countries each had two sites. Pearl millet growing areas in the SADC region fall under four broad agro-ecosystems: (1) short season and often drought stricken environments with <3 months growing season, (2) warm humid environments with <4 months growing season, (3) environments characterized by cool nights as a result of high altitude, and (4) environments characterized by long season (>4 months) growing season with sporadic and unreliable rainfall regime. Within each agro-ecosystem, there are ranges of soil types, texture, and soil water holding properties. The major biophysical characteristics of testing sites are presented in Table 1.

Genetic materials consisted of elite material derived from pearl millet populations (lines, composite populations, synthetics) developed to suit the Southern African region. These can be broadly classified into two major categories. During the early years of the program (1989/1990–1992/1993) most lines were mainly derived from introductions. During the subsequent 5 years (1994/1995–1998/1999), the lines were developed in the region based on selection from local and regionally adapted germplasm. The lines were arranged in randomized complete block designs (a few in simple lattice) with number of lines ranging from 8 to 36 with three to four replications. Individual plots were four rows each 5 m long and spaced 75 cm apart with plant-to-plant spacing of 40 cm within the

Table 1
Biophysical characteristics of SADC[a] pearl millet test sites

Site	Country	Soil type[b]	SWHC[c]	pH	Drainage[d]	Longitude	Latitude	Altitude (m)	Annual rainfall (mm)	First month[e]	Minimum temperature (°C)	Maximum temperature (°C)	LGP (months)[f]	Site group (Fig. 3)
Sebele (seb)	Botswana	M/F	H	6.4	MWD	26.0	−24.6	976	495	11	12	28	2	Grp5
Maun (mau)	Botwana	C	M	6.6	WD	23.4	−20.0	898	445	12	15	31	3	Grp6
Kasinthula (kas)	Malawi	F	M	6.6	PD	34.8	−16.1	122	793	12	19	32	4	Grp5
Ngabu (nga)	Malawi	F	H	7.3	ID	34.9	−16.5	115	760	11	19	32	4	Grp5
Umbeluzi (umb)	Mozambique	M	H	6.4	WD	32.3	−26.0	64	667	12	17	29	5	Grp5
Bagani (bag)	Namibia	C	VL	6.4	ID	20.7	−18.1	1049	551	12	14	30	4	Grp6
Katima (kat)	Namibia	C	VL	6.4	ED	24.3	−17.6	966	682	12	13	30	4	Grp6
Mahanene (mah)	Namibia	C/M	M	6.4	WD	15.2	−17.5	1110	505	11	13	29	3	Grp3
Mashare (mash)	Namibia	M	M	6.6	ID	20.2	−17.9	1061	568	1	14	31	4	Grp1
Ogongo (ogo)	Namibia	C	VL	6.4	SED	14.6	−17.9	1225	403	1	12	26	3	Grp6
Okashana (oka)	Namibia	C	VL	8.5	WD	16.5	−18.3	1097	446	1	15	31	3	Grp6
Hombolo (hom)	Tanzania	C/M	M	5.4	MD	35.9	−6.0	1019	562	12	16	30	4	Grp4
Ilonga (ilo)	Tanzania	F	M	5.7	WD	37.0	−6.8	914	978	11	16	28	6	Grp1
Ukiriguru (uki)	Tanzania	C/M	M	5.4	ID	33.0	−2.7	1239	952	11	17	28	7	Grp4
Kaoma (kao)	Zambia	C	VL	6.4	ED	24.4	−14.4	1041	967	11	14	29	5	Grp6
Longe (lon)	Zambia	C	L	4.8	WD	24.9	−14.9	1124	930	11	13	29	5	Grp4
Simulumbe (sim)	Zambia	C	L	4.3	ED	23.8	−14.6	1017	968	11	15	29	5	Grp5
Panmure (pan)	Zimbabwe	M	M	6.4	MWD	31.6	−17.3	1037	817	11	13	27	5	Grp1
Kadoma (kad)	Zimbabwe	F	H	6.3	MWD	29.9	−18.3	1107	735	12	14	28	5	Grp1
Lucydale (luc)	Zimbabwe	C	M	6.4	MD	28.5	−20.4	1416	591	11	12	25	4	Grp4
Makoholi (mak)	Zimbabwe	M	M	6.4	MWD	30.8	−19.8	1111	628	12	13	26	5	Grp1
Matopos (mat)	Zimbabwe	F	H	6.4	MD	28.5	−20.4	1416	591	11	12	25	4	Grp6
Muzarabani (muz)	Zimbabwe	M	H	6.4	MWD	31.0	−16.4	427	665	12	17	32	3	Grp6

[a] Southern African Development Community.
[b] M: medium; F: fine; C: coarse.
[c] Soil water holding capacity; H: high; M: medium; L: low; VL: very low.
[d] WD: well drained; ID: imperfectly drained; MD: moderately drained; MWD: moderately well drained; PD: poorly drained; ED: excessively drained; SED: somewhat excessively drained.
[e] Officially declared month for beginning of planting.
[f] Length of growing period.

rows. The two center rows were harvested for yield determination.

2.2. Statistical analysis

Sequential retrospective (SeqRet) pattern analysis (Mirzawan et al., 1994; DeLacy et al., 1996a,b) was used to stratify the testing sites according to their similarity of line-yield-differentiation patterns. The methodology was implemented using the SEQRET package Version 1.1 (DeLacy et al., 1998). The SEQRET package and its manual are available at the website http://pig.ag.uq.edu.au/qgpb.

The 90 METs were individually analyzed using the applied experimental design to obtain least squares estimates of line performance. At seven sites, two (2) trials with individual experimental designs were conducted so that extra lines could be tested at those sites. Since the estimates of the EMS for the two trials at those sites were not significantly different (EMS were homogeneous), the data was amalgamated and a single analysis used. This gave 67 unique site–year combinations.

SeqRet pattern analysis was applied on mean data y_{ijk} derived as above from the 67 unique site–year environments for line $k = 1, \ldots, \delta_{ij}$ at site $i = 1, \ldots, n_l$ in year $j = 1, \ldots, \gamma_i$, where δ_{ij} is the number of lines tested in (i, j)th site–year environment, n_l the number of sites, and γ_i the number of years in which site i was present. The set of δ_{ij} lines grown in the (i, j)th site–year environment was assumed as a random (representative) sample of all test-lines. This assumption is an appropriate model to investigate the similarity among testing sites for selection purposes (DeLacy et al., 1996a). For each (i, j)th site–year environment, the y_{ijk} value was transformed to an environment-standardized (ES) value $w_{ijk} = (y_{ijk} - m_{ij})/v_{ij}$, where m_{ij} is the average yield and v_{ij}^2 the phenotypic variance of δ_{ij} line mean yields in (i, j)th site–year environment. The ES transformation was adopted because ES-data-based pattern analysis relates the sites by their similarity of discrimination among lines (Fox and Rosielle, 1982; DeLacy et al., 1994). The sites that cluster together in classification or occur together in ordination are expected to be similar with respect to discrimination among lines.

For each individual year j $(j = 1, \ldots, n_y)$, where n_y is the number of trial years, the SED $D_{ii'j}$ between ES values w_{ijk} and $w_{i'jk}$ of sites (i, i') $(i \neq i' = 1, \ldots, n_l)$ present in year j was computed as per equation (12.29) in DeLacy et al. (1996a). The weighted (average) ES-SED between sites (i, i') over m_y years was sequentially computed as $D_{ii'} = (1/\eta_{ii'})\Sigma_j \eta_{ii'j} D_{ii'j}, j = 1, \ldots, m_y, m_y = 1, \ldots, n_y$, where $\eta_{ii'} = \Sigma_j \eta_{ii'j}$. The $(n_l \times n_l)$ ES-SED matrix $D = \{D_{ii'}\}$was sequentially constructed up to the last year adding 1 year at a time. The corresponding $(n_l \times n_l)$ (complementary) similarity matrix $A = \{a_{ii'}\}$ was computed as $a_{ii'} = 1 - (\frac{1}{2})D_{ii'}$ using the relationship $D_{ii'} = 2(1 - a_{ii'})$ (Gower, 1966). Whenever, of the unique n_1 sites, no comparison among sites (i, i') existed, the corresponding cells in the full $(n_l \times n_l)$ matrices D and A were empty. With these empty cells, the matrices D and A cannot be subjected to a classification and ordination analyses, respectively. Rule 2 (DeLacy et al., 1996a) was used to eliminate empty cells (and the corresponding sites) to calculate *reduced* D and A matrices without empty cells.

The reduced D matrix was used to classify the sites represented in it using the incremental-sum-of-squares (ISS) clustering algorithm (Ward, 1963). Site-proximity plots were constructed from a principal coordinate analysis (PCoA) of the corresponding reduced similarity matrix A. The first two principal coordinate axes were used to graphically depict the sequential change in, and convergence of, site relationships as more years' data were sequentially added to the analysis. After classification of sites represented in reduced D matrix, each eliminated site, not present in the reduced D matrix because of empty cells, was allocated to one of the available site-groups based on its SED from the centroid of the site-groups (DeLacy et al., 1996a, p. 262). Each eliminated site was then assigned to the site-group with the nearest centroid. The adequacy of the chosen site-stratification model was determined from the R^2-statistic as suggested in DeLacy et al. (1996a). A site-stratification model with $R^2 \geq 70\%$ was considered as adequate.

The nine trial years' data were split into two sets: Set 1 (1989/1990–1992/1993) spanning 4 years in which the trials included introductory genetic materials, and Set 2 (1994/1995–1998/1999) spanning 5 years in which advanced genetic materials were tested in the trials. This splitting was done to meet the assumption of representativeness of the genetic materials. The above analytical approach was used separately on each data

and on the combined data covering the nine trial years. Another reason to split the nine trial years in two sets was to assess the site relationships based on different genetic materials. The combined analysis over 9 years, though not satisfying the assumption of representativeness of both introductory and advanced materials, was conducted to assess the repeatability of certain patterns, if any, in site similarity in Set 1, Set 2, and the combined data.

3. Results

The dendrograms in Figs. 1–3 summarize the results of site classification for Set 1, Set 2, and the combined set (Set 3), respectively. The set-wise results are given below.

3.1. Set 1 (1989/1990–1992/1993)

Out of a total of 18 sites present in this set, the cumulative analysis retained nine sites, the remaining nine sites eliminated due to lack of comparisons as explained in material and methods. Each of the nine eliminated sites was assigned to an appropriate site-group (derived from the retained nine sites in the cumulative analysis) with the nearest centroid. A clustering of the 18 sites into six groups, with assignment of eliminated sites to known site-groups, retained 76% of the variance. Of the six groups, two groups (Ogongo) and (Lucydale1) are individual-site groups. The other four groups are (Mahanene, Lucydale, Hombolo, Longe, Matopos, Ukiriguru), (Makoholi, Ilonga, Kaoma, Panmure), (Sebele, Kasinthula, Simulumbe, Umbeluzi), and (Ngabu, Muzarabani). Lucydale and Lucydale1 (i.e. the same location) grouped separately. This could be explained by a difference in planting date which even in the same environment can completely change the way the site discriminates amongst line ranking.

3.2. Set 2 (1994/1995–1998/1999)

This set included 13 sites. The cumulative analysis retained nine sites, four sites were eliminated due to lack of comparisons. A classification of these 13 sites into five groups, with assignment of the four eliminated sites to known site groups, yielded $R^2 = 79\%$. The sites Lucydale and Katima formed separate groups. The other three groups were (Ogongo,

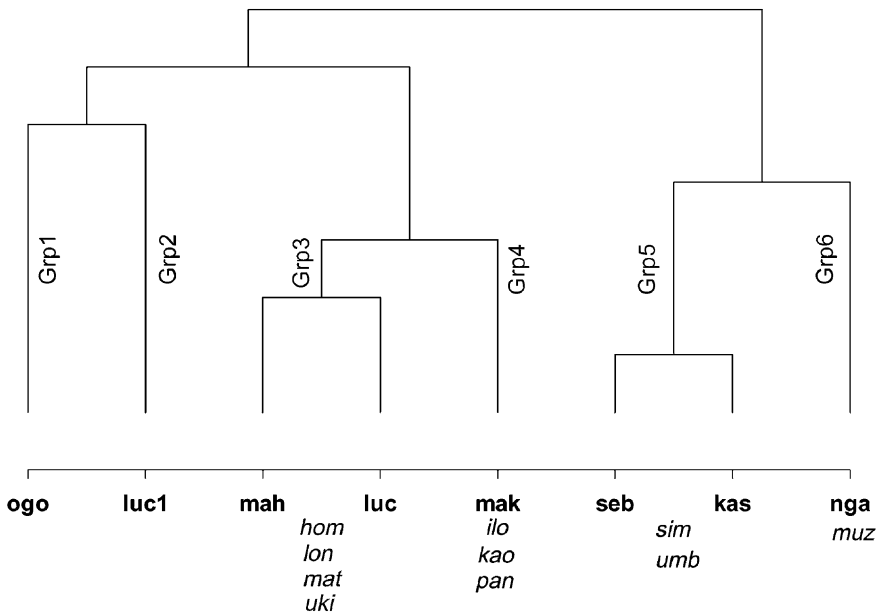

Fig. 1. Dendrogram of cumulative classification of sites (1989/1990–1982/1993) based on grain yield using ES-SED as dissimilarity measure and ISS as clustering strategy (site codes in Table 1); italics indicate the sites added to site-groups based on nearest centroid criterion.

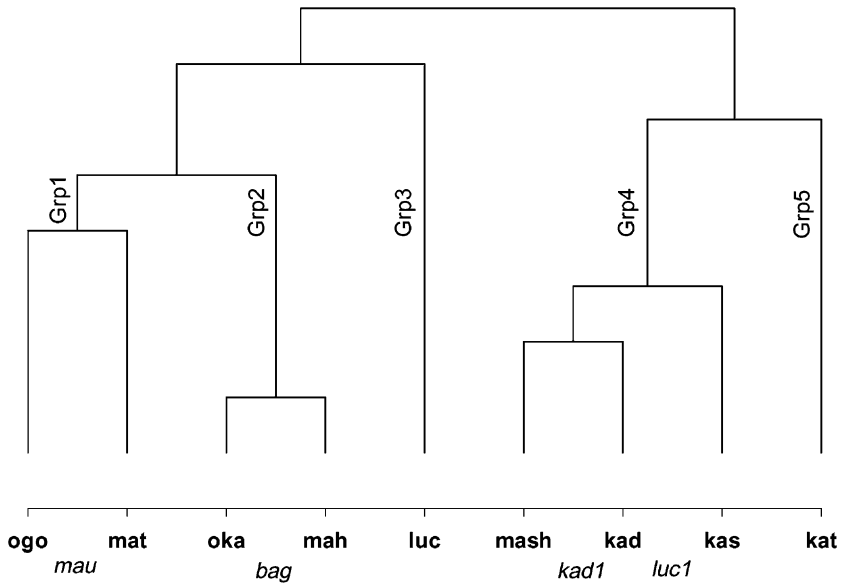

Fig. 2. Dendrogram of cumulative classification of sites (1994/1995–1998/1999) based on grain yield using ES-SED as dissimilarity measure and ISS as clustering strategy (site codes in Table 1); italics indicate the sites added to site-groups based on nearest centroid criterion.

Matopos, Maun), (Okashana, Mahanene, Bagani), and (Mashare, Kadoma, Kasinthula, Kadoma1, Lucydale1). As in Set 1, Lucydale and Lucydale1 again separated into different groups.

Across the two sets, there were six common sites Kasinthula, Lucydale1, Lucydale, Mahanene, Matopos, and Ogongo. The two sites Kasinthula and Lucydale1, belonging to separate groups in Set 1, joined

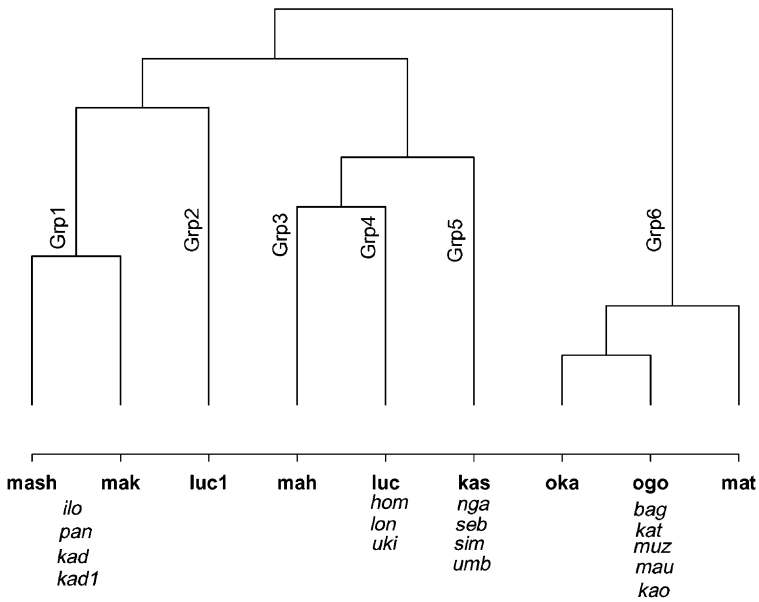

Fig. 3. Dendrogram of cumulative classification of sites (1989/1990–1998/1999) based on grain yield using ES-SED as dissimilarity measure and ISS as clustering strategy (site codes in Table 1); italics indicate the sites added to site-groups based on nearest centroid criterion.

together in Set 2. The sites Lucydale and Mahanene, together in Set 1, separated themselves in Set 2. The two sites Matopos and Ogongo, separate in Set 1, joined together in Set 2. In general there was little congruence in grouping of common sites in the two sets. This is a reflection of the difference in the composition of the genetic materials tested in the two sets.

3.3. Set 3 (1989/1990–1998/1999)

This set included 25 sites across the 9 years (Lucydale and Lucydale1, Kadoma and Kadoma1 treated as different sites). The cumulative analysis retained nine sites, eliminating 16 sites due to lack of comparisons. A clustering of these 25 sites into six groups, with the 16 eliminated sites assigned to known site groups, retained 76% of the variance. The sites Lucydale1 and Mahanene formed separate groups. These two sites also maintained their separate identity in Set 1 and Set 2. The other four groups were (Mashare, Makoholi, Ilonga, Kadoma, Kadoma1, Panmure), (Lucydale, Hombolo, Longe, Ukiriguru), (Kasinthula, Ngabu, Sebele, Simulumbe, Umbeluzi), and (Okashana, Ogongo, Matopos, Bagani, Kaoma, Katima, Maun, Muzarabani). The site Lucydale1 and Lucydale, like in Set 1 and Set 2, remain in separate groups. The sites (Hombolo, Longe, Ukiriguru), (Makoholi, Ilonga, Panmure), and (Sebele, Kasinthula, Simulumbe, Umbeluzi) that remained together in Set 1 also showed similar grouping and hence similar line discrimination in the combined set. Also, the sites ((Ogongo, Matopos, Maun), (Okashana, Bagani)), and (Mashare, Kadoma, Kadoma1) that were together in Set 2, also retained their similarity in the combined set. Thus the major site groupings in the separate sets were maintained in the combined set.

The site proximity plot (Fig. 4) of nine retained sites in the combined cumulative ordination analysis shows that the relative positions of these nine sites converged to fixed positions in the Euclidean space as the 9 years' data were sequentially added to the analysis. This

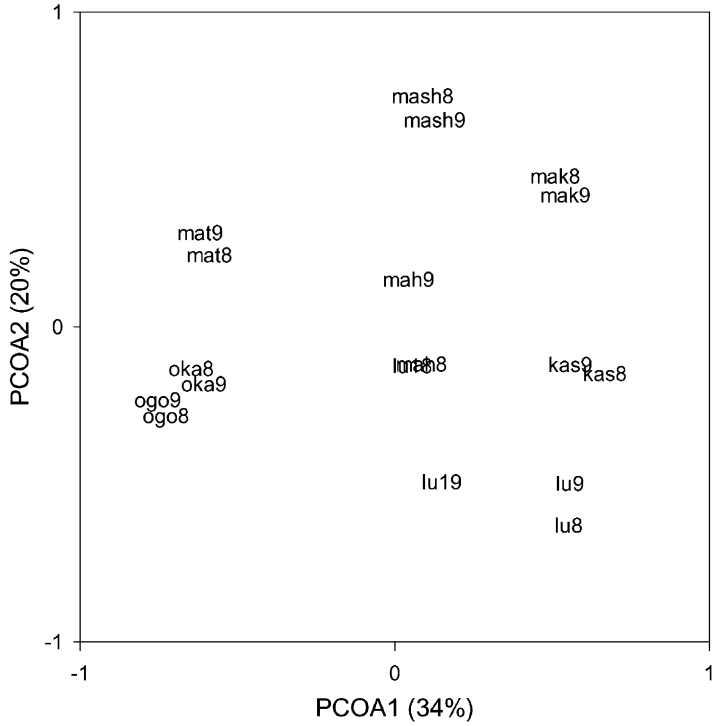

Fig. 4. Proximity plot of first two vectors from cumulative PCoA (1989/1990–1998/1999) based on grain yield using weighted ES-SED as dissimilarity measure; numerals 8 and 9 indicate cumulative analysis up to eight and nine trial years (site codes in Table 1).

convergence did not take place so well in Set 1 and Set 2 due to the smaller sample of years.

4. Discussion

Site stratification analysis utilizing introduced (Set 1) and locally developed germplasm (Set 2) partitioned the testing sites into six and five groups with R^2-values of 76 and 79%, respectively. The separation of Lucydale from Mahanene in Set 2 (Fig. 2), and not in Set 1 (Fig. 1) was expected. In the early years of the program, the main focus was on early maturity accessions to escape drought. As such, these materials could not differentiate location of a different length of growing period (LGP). The use of more diversified lines (Set 2) allowed for more differentiation among the sites based on LGP. This became even clearer in Set 3 where the over 4 months LGP Lucydale, Hombolo, Longe, and Ukiriguru clustered together, leaving Mahanene a site with below 3 months LGP, in a separate group.

The environments generally classified as warm and humid formed their own clusters—Muzarabani, Ngabu in one group, Kasinthula, Simulumbe and Umbeluzi in another group. These sites are generally in low-lying areas (60–427 m above sea level (a.s.l.)) except Simulumbe which is at higher altitude (1017 m a.s.l.) but situated in the humid Zambezi valley. Similarly, sites from environments generally characterized by long growing cycles clustered together in two groups. In one of the groups, three of the four sites were long growing cycle (>4 months LGP)—(Ilonga, Kaoma, Panmure), while the second group had the remaining five sites—(Lucydale, Matopos, Hombolo, Longe, Ukiriguru).

The elite germplasm (Set 2) developed for the SADC region, provided more useful stratification of the sites. The sites Ogongo, Maun, Okashana and Mahenene, all of which are very short duration environments (<3 months LGP) stratified together (Fig. 2). The warm humid environment site of Kasinthula also grouped together with other warm humid sites—Kadoma and Mashare, which were not included in the introductory years. The Set 2 lines were also generally higher yielding than the introductions Set 1 as a result of genetic improvement for local constraints (downy mildew and drought). The overall adaptation of the improved lines also contributed to the better discrimination of sites.

The combined analysis (1989/1990–1998/1999) gave a clearer picture of site stratification (Fig. 3). The differences in site groupings between Sets 1, 2 and 3, indicated that different relationships among sites relative to the longer-term perspective could result from analysis when a small MET data set is used. Similar observations were also made by DeLacy et al. (1996a,b) in spring bread wheat. Two of the three extra short duration sites, Ogongo and Okashana, stratified together. Using the centroid method, the site Maun is added to this cluster, which also included Matopos, the major ICRISAT testing site in Zimbabwe. It seems that the cool nights of Matopos (high altitude 1416 m a.s.l.) could have similar effects on relative development and performance of lines as the drought induced stress of the short season environments (<3 months LGP). Despite the geographic proximity of Matopos and Lucydale, the two sites were not grouped together. This coincides with a known difference in soil type; Lucydale has a generally sandy loam soil whereas Matopos is characterized by clay loam soils. The differentiation of sites of close geographic location but of different soil types, has also been reported by DeLacy et al. (1996a,b).

The long growth cycle sites (>4 months LGP) also stratified together into two groups encompassing Ukiriguru, Longe, Umbeluzi and Simulumbe. Four out of the five warm humid sites—Kasinthula, Ngabu, Umbeluzi, Simulumbe—clustered together. The site grouping ranged from one site per group to a maximum of eight sites (Fig. 3). With the exception of Lucydale1 (later than normal planting at the Lucydale site), the remaining five groups could be regarded as distinct production zones within SADC region.

5. Conclusions

The SeqRet pattern analysis, using the long-term historical data for pearl millet line testing in the SADC region, enabled an objective assessment of similarities among the sites for the way they discriminated among lines, and thus provided a basis to facilitate selection of few representative sites for future testing of lines. Future pearl millet line testing in the SADC region could be restricted to few sites picked from within

each of the five site-groups (Grp1, Grp3, Grp4, Grp5, Grp6) shown in Fig. 3.

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