

## GENOTYPE × ENVIRONMENT INTERACTION EFFECT ON SOYBEAN GENOTYPES UNDER RHIZOBIA INOCULATION IN THE SAVANNA REGION OF NIGERIA

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### ABSTRACT

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A study aimed at exploiting soybean genotypes differences, assessing genotype by environment effect on seed yield and rhizobia inoculation as a means of improving nitrogen fixation and productivity of soybean across three locations in the savanna was conducted in 2015 rainy season. The experiments were arranged in a split plot design and replicated three times. Treatments were twenty four soybean genotypes and three levels of rhizobial inoculation (uninoculated and inoculated with *Legumefix* and *Nodumax*). Results indicated that with the exception of percentage emergence, number of days to 50% flowering and 100-seed weight, there were significant differences between the inoculated and uninoculated plants in all the parameters measured. The effects of genotype (G), environment (E) and G×E interactions on height, number of leaves and, number of branches per plant, above ground biomass and seed yield were significant ( $p=0.05$ ). Two soybean genotypes (TGx 1989-45F and TGx 1989-40F) were identified as the most promising in relation to yield stability. Of the three locations, Abuja produced the least interaction effects followed by Igabi and may be most appropriate environments for soybean production and evaluation. Selection in these environments will be effective as the relative performance of these genotypes would be fairly stable.

**Keywords:** Genotype by Environment, Interaction, Soybean, Seed yield and Breeding.

### INTRODUCTION

Soybean (*Glycine max* L.) is a globally important food legume of great nutritional value. The crop has the highest protein content (40%) of all food crops and is equivalent to proteins of animal products (Dugje *et al.*, 2009). It is second only to groundnut in oil content (20%) among food legumes. The current competitive prices for soybean in global markets have made the cultivation of the crop to be potentially rewarding to farmers (Ene-Obong and Carnovale, 1992). Therefore, interest in soybean production in Africa has increased considerably over the past few decades. This is attributable to a number of factors such as increased utilization of most commercially grown pulses as supplements in livestock feed (Uguru, 1996), usefulness as a source of cheap quality plant protein and the increasing prohibitive cost of animal protein (Ene-Obong and Carnovale, 1992).

In Nigeria, the cultivation of soybean is increasing in the savannas because it's a major cash crop widely used in food and feed. The crop provides opportunity to diversify the cereal cropping systems in the savannas. However there is a gap between the soybean yields on farmer's fields (1.2 t ha<sup>-1</sup>) and yields obtained in research stations (2.3 t ha<sup>-1</sup>). The theoretical limit of soybean productivity was suggested to be 8 t ha<sup>-1</sup> based on the amount of light energy available in the field (Specht, *et al.*, 1999). However, world productivity during 2016 was 3.72 t ha<sup>-1</sup>. Even this has not been achieved in the country due mainly to poor soil fertility, poor agronomic practices, limited varietal stability and narrow genetic base of soybean cultivars. Soybean obtains significant portion of its nitrogen requirement from symbiotic N<sub>2</sub> rhizobium strains. However, soils may not contain these strains to establish an effective association and hence inoculation is essential to ensure presence of effective rhizobium population in the rhizosphere of the crop (Kyei-Baohen *et al.*, 2002).

Soybean yields on farmers' field can also be substantially increased through the use of high yielding varieties. These improved varieties should also have end user preferred traits (e.g. high protein content and, big seed size) to stimulate high levels of adoption by farmers. In Nigeria, the commonest soybean varieties cultivated are the TGx varieties developed to fix nitrogen with indigenous rhizobium populations (Kneneman *et al.*, 1982; Sanginga *et al.*, 2003). Nonetheless, these varieties have been reported to benefit from rhizobial inoculation in the Nigeria savanna (Osunde *et al.*, 2003). Although a number of TGx varieties have been released for use by farmers, a substantial number of lines are yet to be evaluated for genetic stability across the savanna agroecology. Therefore, this study was aimed at exploiting soybean genotype differences; assessing genotype by environment effects on seed yield and evaluating rhizobia inoculation as a means of improving nitrogen fixation and productivity of soybean.

## MATERIALS AND METHODS

The study was conducted during the 2015 cropping season at three experimental sites- Abuja in the Southern Guinea Savannah (9° 16'N and 7° 20'E), Igabi in the Northern Guinea Savanna (112° 12'N and 7° 20'E) and Gwarzo in the Sudan Savanna (11° 19'N and 8° 51'E) in Nigeria. Twenty four soybean genotypes including five commercial checks were evaluated at each location. The inoculants (Legumefix and Nodumax) were used in the study. To achieve seed inoculation, the inoculants were mixed with the soybean seeds as seed treatment at the rate of 200g/50kg of seeds (N2 Africa Nigeria, 2015). The experiments were laid in a 3×24 factorial combination made up of 3 inoculation types (Without Inoculation, Legumefix and Nodumax) and 24 Soybean genotypes fitted into a Split-plot design with three replications. The main plots consisted of the soybean lines and the sub-plots were the inoculant applications. Plots were five rows of 2m length spaced at 0.75m × 0.2m between rows and plants respectively. Planting was done using three seeds per hole and the seedlings were thinned to one plant per stand at two weeks after planting. Standard cultural practices (land preparation, seed inoculation, planting, weeding and, fertilizer application, insect control and harvesting) were applied at each location. Data were taken on 5 plants per plot for height, number of leaves, chlorophyll content was determined using chlorophyll meter SPAD – 502Plus with optical density difference at 2 wavelengths at 10 weeks after sowing, number of branches per plant, number of pods per plant. Days to 50% flowering was taken when half of each plot had flowered. One-hundred seed weight was determined by counting 100 seeds each from each plot after threshing and weighed. Above ground biomass per plot was taken after harvesting the whole plant from the net plots and weighed. Grain weight per plot was taken after threshing the harvested pods from the net plots and weighed. Harvesting index was computed as a ratio of seed yield to total biomass yield. Analysis of variance procedure was adopted to test the effects of inoculation, location, genotype and interactions assuming the location effects as random and genotype effect as fixed.

The Additive Main Effects and Multiplicative Interaction (AMMI) model (Gauch, 2006) was used to evaluate soybean genotypes × environment interaction using the following relationship:  $Y_{ij} = \mu + g_i + e_j + \lambda_k Y_{ik} \delta_{jk} + \epsilon_{ij}$

Where  $Y_{ij}$  is the grain yield of the  $i$ -th genotype in the  $j$ -th environment,  $\mu$  is the grand mean,  $g_i$  and  $e_j$  are the genotype and environment deviation from the grand mean, respectively,  $\lambda_k$  is the eigen value of the principal component analysis (PCA) axis  $k$ ,  $Y_{ik}$  and  $\delta_{jk}$  are the genotype and environment principal component scores for axis  $k$ ,  $N$  is the number of principal components retained in the model, and  $\epsilon_{ij}$  is the residual term.

GGE – biplot methodology, which is composed of 2 concepts, the biplot concept (Gabriel, 1971) and the GGE concept (Yan *et al.*; 2000) was used to analyze multi-environment trials data. This methodology uses a biplot to show the factors (G and GE) that are important in genotype evaluation. In this study, genotype-focused scaling was used in visualizing for genotype comparison, with environment-focused scaling for environment comparison. The statistical analysis was conducted using the Integrated Breeding Platform Breeding Management System Version 3.09.

## RESULTS AND DISCUSSION

Significant differences were observed for all traits except percentage emergence, number of leaves and days to 50% flowering. Soybean grain yields for the 24 genotypes ranged from 1245.38 kg ha<sup>-1</sup> – 1610.80 kg ha<sup>-1</sup> (Table 1). Eleven genotypes (TGx 1989 – 11F, TGx 1990 – 110FN, TGx 1989-42F, TGx 1989 – 45F, TGx 1990 – 114FN, TGx 1993 – 4FN, TGx 1993 – 4FN, TGx 1989 – 75FN, TGx 148-2E check, TGx 1990-52F, TGx 1989-48FN, TGx 1990 – 57F) gave higher grain yields than the grand mean yield (1399.69 kg ha<sup>-1</sup>), grain yield of environments ranged from 39.93 kg ha<sup>-1</sup> in Gwarzo to 2148.44 kg ha<sup>-1</sup> in Abuja and was significant in all the three locations except Gwarzo (Table 1) Also, Abuja location also had the highest mean performance than other locations. This is explained by the box plot (Fig. 1). The boxplot encloses observations between the 25<sup>th</sup> (lower quartiles) and 75<sup>th</sup> (upper quartiles) with the lines extending to the minimum and maximum of observed values. The large yield variations explained by environments indicate that the environments were diverse, with large differences between environmental means contributing most of the variation to grain yield.

Inoculation significantly influenced grain yield of soybean except in TGx 1989-75FN, TGx 1990-78F, TGx 1990-40F and TGx 1485 –ID – check genotype (Table 2). According to Khalili *et al.* (2011) an ideal cultivar would have both a high average performance over a wide range of environments plus stability. Although genotypic main effect was not significant, environment effect was highly significant (Table 3) which shows differences in genotypic performances across environments resulting in genotype × environment interaction. The existence of genotype × environment interaction (GEI) raised the need to identify stable and high yielding genotypes. The mean values for yield and regression coefficient for 20 best genotypes of soybean over three environments are presented in Table 4. Slope ( $b$  value) is the genotypic sensitivity to changes in the environmental quality; where values for  $b > 1$  means genotypes with a higher than average sensitivity, and less stable while  $b < 1$  means genotypes that are less sensitive and more stable. The regression showed that TGx 1989-48FN had mean grain yield (1433.50 kg ha<sup>-1</sup>) greater than average mean 1399.69 (kg ha<sup>-1</sup>) and showed average genotypic sensitivity

based on the regression coefficient ( $b = 1$ ) hence averagely stable. One genotype, TGx 1990-57F, had more than average mean performance and above average sensitivity (i.e below average stability,  $b > 1$ , less stable). Among the genotypes, TGx 1989-45F, TGx 1993-4FN, TGx 1990-110FN and TGx 1989-42F had more than average mean performance and below average sensitivity (i.e above average stability,  $b < 1$  more stable). Although TGx 1989 – 49FN did better than TGx 1883-4FN in the average performance, TGx 1993-4FN has the potential to respond to increase in environmental quality in a predictable way (genotype superiority). Most of the high yielding genotypes had similar stability potentials. High yielding genotypes like TGx 1989-45F, TGx 1990-110FN, TGx 1990-46F and TGx 1990-52F also had good static stability (i.e ability to give same performance across environments).

The AMMI analysis of variance revealed that variances due to environments, genotypes and  $G \times E$  interactions were significant (Table 5). The result showed that the environment main effect (E) was the most important source of variation, due to its large contribution to the total sum of squares for yield. The large sum of squares for environment indicated that the environments were diverse. This might be due to differences in growing season rainfall which has been known to have positive impacts on soybean yield (Cucolotto *et al.*, 2007). Moreover, the significant ( $P < 0.05$ )  $G \times E$  interaction for soybean yield is an indication of differential performance of genotypes across environments and this necessitates the investigation of the nature of differential response of genotypes to environments. In spite of this significance, the magnitude of the GEI sum of squares was smaller than that of genotypes, indicating the presence of moderate variation among the genotypes over environments. Similar observations were obtained by Admassu *et al.* (2008). The presence of GEI was demonstrated by the AMMI model when the interaction was partitioned among the first two interaction principal component Axis (IPCA) as they were significant. The IPCA1 explained 70.1% of the interaction while IPCA2 explained 29.9% (Figure 1); they cumulatively captured 100% GEI. This implies that the interaction of the 24 genotypes of soybean with three environments was predicted by the first two principal components of genotypes and environments, which is in agreement with Guach and Zobel (1996). The differences among genotypes in direction and magnitude along the X-axis (yield) and Y-axis (IPCA 1 scores) are provided by AMMI biplot using the main effect and the first principal component scores of interactions (IPCA1) of both genotypes and environment (Figure 1). In the bi-plot, genotypes or environments that appear almost on a perpendicular line of the graph had similar mean grain yields and those that fall almost on a horizontal line had similar interaction (Crossa *et al.*, 1990) than that due to genotype differences. Genotypes or environments on the right side of the midpoint of the perpendicular line had higher yields than those on the left side. The genotype TGx 1989-49FN, TGx 1990-46F, TGx 1990-110FN, TGx 1993-44FN and TGx 1989-45F were high yielding. In contrast, TGx 1990-40F, TGx 1989-53FN, TGx 1835-10E Check and TGx 1987-62F check were low yielding. Genotypes or environments with large negative or positive IPCA1 scores had high interactions, while those with IPCA1 scores near zero (close to the horizontal line) had little interaction across environments (Egesi and Asiedu, 2002) and are considered more stable than those further away from the line. In the biplot, the genotypes TGx 1989-11F, TGx 1989-45F, TGx 1990-110FN and TGx 1993-4FN fell almost on a horizontal line near the zero point on IPCA1. This implies that these genotypes showed high and stable yield. The genotypes TGx 1990-46F, TGx 1987-10F check and TGx 1989-68FN were a little far away from the horizontal and implies that the genotypes are high yielding but relatively unstable. In terms of environment, Abuja, Igabi and Gwazo in that order were unstable producing highest interaction scores. The biplot of the best genotypes in each of the environments for grain yield is presented in Figure 2. The polygon view of the GGE-biplot explicitly displays best genotype in each environment and it is a summary of the GEI pattern of a multi-environment yield data. The polygon was formed by connecting the genotypes that are further away from the biplot origin such that all other genotypes are contained within the polygon. To each side of the polygon, a perpendicular line, starting from the origin was drawn and extended beyond the polygon so that the biplot was divided into several sectors, and the different environment were separated into different sectors. The genotype at the vertices of each sector is the best performer at environments included in that sector, provided that GGE is sufficiently approximated by PC1 and PC2. Hence, though there were five sectors in all, two mega environments were identified. Abuja and Gwazo was one mega environment with TGx 1989-42FN, TGx 1989-45F, TGx 1990-114FN, TGx 1989-75FN and TGx 1990-46F as the best genotypes in this environment. The best genotypes for the second mega environment, Igabi, were TGx 1989-11F, TGx 1993-4FN and TGx 1989-49FN. The remaining sectors had no environment within them and contained the following genotypes in their vertices TGx 1990-52F, TGx 1989-48FN, TGx 1990-57F, TGx 1990-95F, TGx 1989-40F, TGx 1990-55F, TGx 1987-10F-CHECK, TGx 1835-10E-CHECK, TGx 1485-1D- CHECK, TGx 1990-78F, TGx 1967-62F-CHECK, TGx 1989-68FN, TGx 1989-53FN and TGx 1990-40F. These genotypes without environment in the sectors were not the highest yielding genotypes at any environment. However, genotypes within the polygon, particularly those located near the plot origin, were less responsive than the vertex genotypes.

Table 1: Grain yield (kg ha<sup>-1</sup>) of soybean genotypes across the three locations

Genotypes	Abuja	Igabi	Gwarzo	Mean
TGx 1989-11F	1881.43	1981.03	1394.37	1418.94
TGx 1990-110FN	2086.43	1995.80	1480.06	1520.76
TGx 1989-42F	2068.40	1955.11	1435.05	1486.18
TGx 1990-95F	1692.02	2005.81	1404.11	1367.31
TGx 1989-45F	2023.13	1910.06	1435.96	1456.38
TGx 1990-114FN	1958.77	1885.64	1432.32	1425.57
TGx 1989-53FN	1768.58	1725.90	1467.85	1320.77
TGx 1993-4FN	2148.44	2087.01	1510.40	1581.95
TGx 1989-75FN	1995.28	1811.06	1509.15	1438.49
TGx 1990-78F	1563.63	1763.36	1447.40	1258.13
TGx 1987-62F CHECK	1678.30	1821.61	1431.26	1310.39
TGx 1448-2E CHECK	2058.71	1967.68	1393.93	1473.44
TGx 1989-40F	1672.20	2011.29	1425.07	1369.52
TGx 1990-52F	1829.54	2086.56	1406.93	1441.01
TGx 1989-48FN	1786.20	2096.23	1418.08	1433.50
TGx 1990-40F	1710.18	1743.98	1464.79	1306.31
TGx 1989-49FN	2140.88	2188.35	1503.19	1610.80
TGx 1990-57F	1740.74	2112.58	1494.51	1449.27
TGx 1989-68FN	1518.91	1794.45	1422.79	1245.38
TGx 1990-46F	2215.53	1824.43	1463.93	1501.29
TGx 1990-55F	1792.85	1856.93	1449.51	1366.43
TGx 1987-10F CHECK	1430.71	1867.98	1397.86	1232.18
TGx 1835-10E CHECK	1699.30	1799.13	1456.08	1318.17
TGx 1485-ID CHECK	1697.47	1642.89	1441.34	1260.56
Mean	1839.90	1913.95	1445.23	1399.69
Mean SED	217.96	242.39	153.22	217.96
Mean LSD	429.91	478.08	104.98	429.91
Heritability	0.49	0.04	0.27	0.49
p-value	0.01	0.02	0.59	0.01

Table 2: Grain yield (kg ha<sup>-1</sup>) of soybean genotype as affected by inoculation across the 3 locations

Genotypes	Without inoculation	Nodumax	Legumefix	SE±
TGx 1989-11F	1717.0c	1886.6b	2359.5a	70.0
TGx 1990-110FN	1541.0b	2126.1a	2320.3a	68.7
TGx 1989-42FN	1729.7b	1943.5a	2192.0a	65.7
TGx 1990-95F	1596.2b	2190.2a	2231.1a	71.2
TGx 1989-45F	1521.4c	2430.1a	1778.7b	59.3
TGx 1990-114FN	1492.2b	2182.4a	1982.3a	59.9
TGx 1989-53FN	1643.4b	1718.3a	1816.0a	48.4
TGx 1993-4FN	1691.1b	2242.7a	2327.2a	65.1
TGx 1989-75FN	1629.1	1985.2	1818.9	59.2
TGx 1990-78F	1516.6	1944.9	1828.6	60.7
TGx 1967-62F-CHECK	1266.3b	2126.9a	2071.6a	69.8
TGx 1448-2E-CHECK	1617.4b	1582.9b	2702.7a	72.1
TGx 1989-40F	1832.8b	2219.7a	1981.3b	64.2
TGx 1990-52F	1489.6b	2604.7a	2165.4a	75.5
TGx 1989-48FN	1795.9b	2013.5a	2479.2a	82.9
TGx 1990-40F	1701.7	1639.4	1890.8	70.6
TGx 1989-49FN	1761.2b	2563.7a	2240.2a	95.2
TGx 1990-57F	1765.8b	2143.8a	2428.2a	69.5
TGx 1989-68FN	1642.4b	2632.5a	2441.7a	57.6
TGx 1990-46F	1405.2b	1593.3b	2474.5a	92.8
TGx 1990-55F	1408.1b	1794.1b	2368.6a	77.3
TGx 1987-10F-CHECK	1142.7b	2153.7a	2307.6a	69.4
TGx 1835-10E-CHECK	1245.4c	2479.3a	1672.7b	71.2
TGx 1485-ID-CHECK	1648.6	1633.5	1646.6	59.3

Means followed by the same letter in a row are not significantly different at  $P \leq 0.05$  using DMRT. SE± = Standard error

Table 3: Analysis of variance using Finlay and Wilkinson regression analysis

Source	d.f.	s.s.	m.s.	v.r.	F pr.
Genotypes	23	751977.4547	32694.6719	1.50	0.169
Environments	2	32861072.3962	16430536.1981	753.99	<0.001
Sensitivities	23	300194.3423	13051.9279	0.60	0.887
Residual	23	501201.6828	21791.3775		
Total	71	34414445.8759	484710.5053		

Table 4: Mean values for yield and stability coefficient (b) for genotypes of soybean over three environment

Genotype	Mean Yield	Sensitivity (b value)	Genotype Superiority	Static stability	Ecovalence Stability	Mean square Deviation
TGx 1989-45F	1456.38	0.8528	937	502935	2163	10098
TGx 1993-4FN	1581.95	0.8554	2462	504092	7701	6339
TGx 1990-110FN	1520.76	0.8671	9111	526480	10909	23546
TGx 1989-42F	1486.18	0.8809	13621	531412	13089	335
TGx 1448-2E CHECK	1473.44	0.8833	14476	546069	13130	71356
TGx 1989-49FN	1610.80	0.8909	20001	559884	19938	5315
TGx 1990-46F	1501.29	0.9040	22433	569875	22275	747
TGx 1990-114FN	1425.57	0.9229	27277	584785	24037	3271
TGx 1989-11F	1418.94	0.9605	28012	631585	24718	372
TGx 1990-52F	1441.01	0.9673	28342	656248	26248	31234
TGx 1989-75FN	1438.49	0.9961	31811	697729	28852	36957
TGx 1989-48FN	1433.50	1.0077	33556	718252	29745	46165
TGx 1990-57F	1441.06	1.0151	38570	720440	31570	30076
TGx 1990-55F	1366.43	1.0369	48701	741249	33419	10291
TGx 1990-95F	1367.31	1.0644	53114	784145	34298	17169
TGx 1989-40F	1369.52	1.0696	55641	789790	35273	28275
TGx 1989-53FN	1320.77	1.0739	69240	797343	35542	362
TGx 1835-10E CHECK	1318.17	1.0752	70157	814351	35703	107782
TGx 1987-62F CHECK	1310.39	1.0861	71563	818505	37083	13499
TGx 1990-40F	1306.31	1.0878	75821	831873	38883	16837

Table 5: Additive main effect and multiplicative interactions (AMMI) analysis of variance for soybean genotype grain yield ( $\text{kg ha}^{-1}$ ) across environments

Source	d.f.	s.s.	m.s.	F pr
Genotypes	23	751977	32695	0.0215
Environments	2	32861072	16430536	<0.001
Interactions	46	801396	17422	0.0312
IPCA 1	24	561569	23399	<0.001
IPCA 2	22	239827	10901	0.0305
Residuals	0	0		

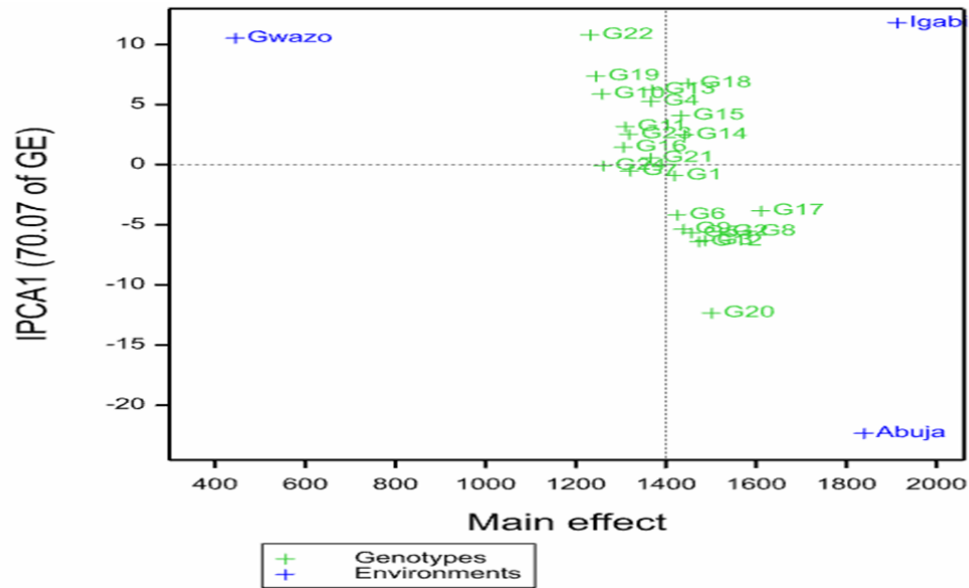


Fig. 1: Yield AMMI Plot for 24 soybean genotypes grown across the savanna  
 1= TGx 1989-11F; 2= TGx 1990-110FN; 3= TGx 1989-42FN; 4= TGx 1990-95F; 5= TGx 1989-45F; 6= TGx 1990-114FN; 7= TGx 1989-53FN; 8= TGx 1993-4FN; 9= TGx 1989-75FN; 10= TGx 1990-78F; 11= TGx 1967-62F-CHECK; 12= TGx 1448-2E-CHECK; 13= TGx 1989-40F; 14= TGx 1990-52F; 15= TGx 1989-48FN; 16= TGx 1990-40F; 17= TGx 1989-49FN; 18= TGx 1990-57F; 19= TGx 1989-68FN; 20= TGx 1990-46F; 21= TGx 1990-55F; 22= TGx 1987-10F-CHECK; 23= TGx 1835-10E-CHECK; 24= TGx 1485-1D- CHECK

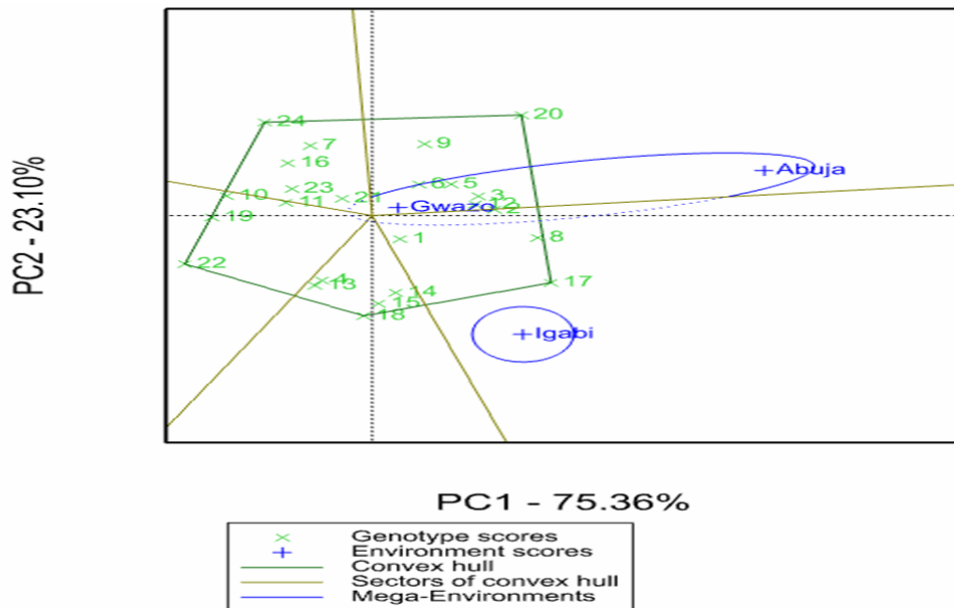


Fig. 2: Yield GGE biplot (symmetric scaling) for 24 soybean genotypes grown across the savanna  
 11= TGx 1989-11F; 2= TGx 1990-110FN; 3= TGx 1989-42FN; 4= TGx 1990-95F; 5= TGx 1989-45F; 6= TGx 1990-114FN; 7= TGx 1989-53FN; 8= TGx 1993-4FN; 9= TGx 1989-75FN; 10= TGx 1990-78F; 11= TGx 1967-62F-CHECK; 12= TGx 1448-2E-CHECK; 13= TGx 1989-40F; 14= TGx 1990-52F; 15= TGx 1989-48FN; 16= TGx 1990-40F; 17= TGx 1989-49FN; 18= TGx 1990-57F; 19= TGx 1989-68FN; 20= TGx 1990-46F; 21= TGx 1990-55F; 22= TGx 1987-10F-CHECK; 23= TGx 1835-10E-CHECK; 24= TGx 1485-1D- CHECK

**CONCLUSION**

The results from this study indicated that the GGE and AMMI biplots are useful techniques that were able to effectively detect the existence of a significant amount of GE interaction between 24 soybean genotypes across three environments. Two soybean genotypes (TGx 1989-45F and TGx 1989-40F) were identified as the overall best in performance in relation to yield and stability. These suggest that for reliability and optimum result it is better to combine the result of two or three analytical tools for yield and stability in the recommendation of

genotypes to farmers. Also, inoculation of soybean proved positive as it enhanced yield across the environments. Genotypes with large interaction with the environment are unpredictable in performance; Abuja produced the least interaction effect and may be most appropriate environment for soybean production and evaluation. Selection in this environment would be effective as the relative performance of these genotypes would be fairly stable.

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