

YIELD STABILITY AND INTER-TRAITS RELATIONSHIPS OF MAIZE HYBRIDS UNDER LOW- AND OPTIMUM-NITROGEN CONDITIONS

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The menace of low soil nitrogen poses a significant challenge to maize production in sub-Saharan Africa, necessitating the development of high-yielding hybrids with increased nitrogen (N) tolerance. Two hundred and forty (240) maize hybrids were evaluated across eight environments at Zaria and Mokwa in Nigeria during the 2020 and 2021 growing seasons, encompassing both low- and optimum-N conditions. The primary objective of the study was to identify high-yielding, stable, and low-N-tolerant hybrids for potential adoption in Nigeria. The study results underscored the significant impact of nitrogen availability on various agronomic traits, causing a substantial 53% reduction in grain yield. Significant genetic variation was observed among hybrids for most measured traits. SMLW146 × IITA1878, SMLW147 × SAM50M and SMLW147 × IITA1878 were the highest-yielding hybrids across the test environments with a yield advantage of 48.2%, 46.6% and 43.1%, respectively over the best check. GGE biplot analysis identified Mokwa Low-N 2021 (MLN21) and Mokwa optimum-N 2021 (MOP21) as discriminating and representative environments for selecting superior hybrids. SMLW147 × SAM50M emerged as the most stable low-N tolerant hybrid with the highest yield performance. Conversely, hybrid SMLW147 × IITA1878 produced a high yield across environments but was unstable. The study concludes by recommending SMLW147 × SAM50M for cultivation across both low- and optimum-N conditions, while SMLW147 × IITA1878 is specifically recommended for cultivation in optimum-N environments. Anthesis-silking interval, plant aspect, ear aspect, and ear height were identified as the most suitable secondary traits for selecting hybrids for high grain yield under low-N conditions.

Key words: genotype × environment interaction, selection indices, correlation, biplot analysis, low-N tolerance, *Zea mays* L.

Maize (*Zea mays* L.) stands as a cornerstone in global agriculture, playing a pivotal role as a vital staple crop with far-reaching implications for food security, economic development and various industries (FAOSTAT, 2016). Beyond its importance in human consumption, maize is integral to the production of animal feed, supporting the livestock industry on which much of the world's meat and dairy production relies. Its industrial applications, including

the production of cornstarch, corn syrup and other derivatives, make maize a versatile commodity with impacts across various sectors (Bankole *et al.* 2019).

However, despite the economic importance of maize, challenges persist in optimizing its cultivation, particularly in regions with nitrogen-deficient soils. Maize, being a C4 plant with high photosynthetic efficiency, heavily depends on nitrogen (N) availability in the soil for substantial biomass sup-

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port. Unfortunately, tropical soils often face limitations in nitrogen availability due to factors such as high rates of leaching, runoff, ammonia volatilization, and microbial biomass immobilization (Alva *et al.* 2006; Amegbor *et al.* 2017). Additionally, dry periods exacerbate nitrogen availability issues by impeding nitrogen uptake due to decreased root activity and impaired nutrient transport in the soil. Drought-induced soil cracks can intensify the leaching of nitrate below the root zone, rendering it inaccessible to plants. Soil-N can at the same time volatilize as ammonia gas under dry and warm conditions, further reducing nitrogen availability for plants (Plett *et al.* 2020; Hoffmann *et al.* 2021).

While fertilisers offer a conventional solution to nitrogen deficiency, their indiscriminate use has raised environmental concerns, contributing to nitrogen pollution and ecosystem degradation (Tubiello *et al.* 2013; Giordano *et al.* 2021). Recent studies highlighted a positive correlation between nitrogen fertilisation and residual nitrogen losses, particularly in regions with intensive agricultural practices (Wang *et al.* 2018; Martínez-Dalmau *et al.* 2021; Hassan *et al.* 2022). Policies like the Nitrate Directive in the European Union emphasized the need to address nitrate pollution from agricultural activities (Musacchio *et al.* 2020), highlighting the consequences of excessive nitrogen application on water quality, eutrophication, and biodiversity loss (Billen *et al.* 2013; Jia *et al.* 2014; Martínez-Dalmau *et al.* 2021). In response to these challenges, the development of maize hybrids with low-N tolerance through genetic approaches emerges as a promising and environmentally friendly solution. These hybrids often exhibit more efficient mechanisms for nitrogen uptake, characterized by specialized root traits facilitating better access to nitrogen sources in the soil (Kant *et al.* 2011). Additionally, they demonstrate higher nitrogen use efficiency, strategically allocating nitrogen to critical processes like grain filling, photosynthesis and root development (Asibi *et al.* 2019; Li *et al.* 2019).

Furthermore, understanding the interaction between genotype and environment ($G \times E$) is crucial for developing adaptable maize varieties with stable yield performance across diverse conditions. When $G \times E$ interactions are insignificant, genotypic performance remains relatively consistent across environ-

ments, allowing for reliable predictions of performance regardless of specific conditions (Badu-Apraku *et al.* 2011; Mafouasson *et al.* 2018). However, significant $G \times E$ interactions can lead to variations in genotype performance across locations, requiring the selection of different varieties for different environments (Yan & Tinker 2006; Badu-Apraku *et al.* 2011).

Various statistical tools have been employed to explore the interactive effects of genotype and environment on yield performance and stability, including the genotype \times environment interaction (GGE) biplot, additive main effects and multiplicative interaction (AMMI) analysis, and regression analysis (Badu-Apraku *et al.* 2011; Olaoye *et al.* 2017; Mafouasson *et al.* 2018; Bankole *et al.* 2023). Among these tools, the GGE biplot stands out as a preferred choice due to its ability to provide a visual representation of complex trait interactions, simultaneous assessment of genotype and environmental effects, dimension reduction for easier interpretation, identification of mega-environments, intuitive interpretability, accommodation of non-linear interactions, and flexibility in data analysis (Yan & Tinker 2006; Badu-Apraku *et al.* 2011; Bankole *et al.* 2023). Given the significance of maize production in Nigeria and the prevalence of nitrogen-deficient soils in the region, it is imperative to identify maize hybrids with low-N tolerance for potential adoption. This study was therefore carried out to assess the agronomic performance, analyse genotype-environment interactions, and investigate the inter-traits relationships of 237 testcrosses and three hybrid checks under varying soil-N conditions.

MATERIAL AND METHODS

Germplasm

The genetic materials used in this study comprised 237 testcrosses and 3 hybrid checks (SAM-MAZ50, Oba Super 2 and SC619). Detailed information regarding the inbred lines, testers, and the development of the testcrosses has been provided in an earlier report. In summary, the 237 testcrosses were generated by crossing seventy-nine inbred lines (comprising 35 low-N tolerant and 44 susceptible to low-N) with three inbred testers, in a line \times

tester mating design at the Institute for Agricultural Research (IAR) experimental field in Zaria in 2019. SAMMAZ50 is a high-yielding hybrid released in Nigeria and adapted to the Southern and Northern Guinea Savanna agroecological zone. Oba Super 2 is also a high-yielding hybrid maize variety from Premier Seeds Nigeria adapted to the Forest and Savanna agroecological zone.

Experimental sites

The evaluation was conducted at the IAR experimental fields in Zaria, Kaduna State and Mokwa, Niger State. Zaria is in the Northern Guinea Savanna ecology of Nigeria with an Elevation of 640 m, a Longitude of 8°22' E, a Latitude of 12°N, and an annual rainfall of 1,200 mm. Mokwa is in the Southern Guinea savanna of Nigeria with an Elevation of 45 m, a Longitude of 5°N 4'E, a Latitude of 9°18'N and an annual rainfall of 1,100 mm. The study was conducted under low-N and optimum-N conditions over two successive years (2020 and 2021). The IAR low-N sites at Zaria and Mokwa established by soil depletion of available nitrogen through continuous planting of maize for several years without any fertiliser application were used as the low-N field for the research. The nitrogen content of the low-N fields at Mokwa and Zaria were 0.085% and 0.11% respectively, both of which were below the minimum threshold of 0.2% based on the interpretation by Landon (1991). The combination of years, locations and soil nitrogen level was therefore treated as an environment in the study. Environments 1, 2, 3, 4, 5, 6, 7 and 8 represent Zaria low-N 2020, Zaria low-N 2021, Zaria Optimum-N 2020, Zaria Optimum-N 2021, Mokwa Low-N 2020, Mokwa low-N 2021, Mokwa Optimum-N 2020, and Mokwa Optimum-N 2021, respectively.

Field evaluation and management

The 240 hybrids were evaluated using a 15 × 16 lattice design with two replications under low- and optimum-N conditions at each location during the 2020 and 2021 growing seasons. The plot layout comprised a single row measuring 4 m in length, with inter-row and intra-row spacings set at 0.75 m and 0.4 m, respectively. Two seeds were sown per hill to achieve a population density of 66,667 plants/ha. Nitrogen fertiliser was applied at two rates: 30 kg N/ha for low-N and 90 kg N/ha for optimum-N. For

the low-N treatment, urea (30 kg N/ha) was applied evenly in two split doses at two and five weeks after sowing (WAS), along with Muriate of Potash at a rate of 60 kg K/ha and Single Super Phosphate at a rate of 60 kg P/ha at 2WAS. Optimum-N treatment involved NPK 15:15:15 (60 kg N/ha, 60 kg P/ha and 60 kg K/ha) at 2WAS, followed by an additional top dressing of 30 kg N/ha of urea at 4WAS. At all locations, weeds were managed in the field through the application of herbicides (Primextra and Paraquat at 5 L/ha) during the early phases of maize growth. Subsequent manual weeding was performed when necessary to maintain a weed-free field throughout the growing season.

Data collection and analyses

Agronomic data were recorded for each hybrid either on a plot or sampled plant basis in both low- and optimum-N environments. Flowering traits were recorded in days while growth traits were quantified based on the average measurements of five randomly selected plants within a plot. Aspect ratings were assessed visually, utilizing a phenotypic scale ranging from 1 to 10, where 1 denoted excellent overall phenotypic appeal, and 10 indicated poor overall phenotypic appeal. Leaf senescence, characterized by the stay-green trait, was assessed by visually evaluating the condition of the leaves, including colour, overall appearance, and visible signs of ageing for each plot in the low-N field. Leaf senescence data were collected 70 days after planting to capture the progression of this trait over time. To standardize this assessment, a visual scale was designed to quantify leaf senescence. This involved assigning numerical values from 1 to 10 to represent various stages of senescence. A score of 1 indicated plants with healthy, green leaves, while a score of 10 represented the highest degree of senescence, characterized by leaves exhibiting prominent signs of wilting, yellowing, or necrosis. The shelled grain weights per plot of the genotype were used to estimate grain yield [kg/ha] in the low-N field, while the ear weight, adjusted to an 80% shelling coefficient, was utilized in the optimum field. Subsequently, both datasets were adjusted to a 15% moisture content.

The analysis of variance using SAS software was conducted on the agronomic data collected (SAS

Institute, version 9.2, 2008). Means of traits for which the maize inbred lines differed significantly were separated using the least significant difference at probability levels of 0.01 or 0.05 (Steel & Torrie 1980). The level of tolerance to low soil N of each hybrid was assessed using the low-N tolerance index (LNTI) as outlined by Oyekunle and Badu-Apraku (2013). The standardized value of each trait was used for the estimation of LNTI.

$$LNTI = (2 \times YLDLN) + YLDON + EPP - ASI - PA - EA - STGR$$

Percentage yield reduction (YRD) was also calculated using the formula:

$$YRD (\%) = \frac{(YLDON - YLDLN)}{YLDON} \times 100$$

Where: YLDLN – grain yield in low-N environment; YLDON – grain yield in the optimum-N environment; EPP – number of ears per plant in a low-N environment; ASI – anthesis silking interval in low-N environment; PA – plant aspect in low-N environment; EA – ear aspect in low-N environment; STGR – stay green characteristic in low-N environment.

GEA-R software was used to investigate the G×E and the stability of the low-N tolerant hybrids (best 15 and worst 10) using their mean grain yield (Pacheco *et al.* 2016). The software was also used to examine the relationships between grain yield and all the measured traits in the study.

Broad sense Heritability (H^2) was estimated for each trait under both low-N and optimum-N conditions using the formula:

$$H = \frac{\sigma^2_G}{\sigma^2_G + (\sigma^2_{GE} / e) + (\sigma^2_E / re)}$$

Where: σ^2_G is genotypic variance, σ^2_E is error variance, σ^2_{GE} is G×E Interaction variance, e and r are the numbers of environments and replications within an environment, respectively (Fehr 1991).

RESULTS

Hybrids performance under different nitrogen conditions

For low-N trials, the environmental main effect was significant ($\rho \leq 0.05$) for the anthesis silking interval (ASI) and highly significant ($\rho \leq 0.01$) for grain yield (GY) and all other measured traits. The

genotypes exhibited highly significant differences ($\rho \leq 0.01$) for GY, days to anthesis (DA), days to silking (DS), ASI, Plant height (PHT), ear height (EHT), stay green characteristics (STGR) and significant differences ($\rho \leq 0.05$) for ear aspect (EA). However, the G×E interaction was highly significant ($\rho \leq 0.01$) for GY, EHT, and STGR, and significant ($\rho \leq 0.05$) for ASI, PHT and EA. Stay-green characteristic had the highest broad sense heritability estimate ($H^2 = 0.92$), followed by DA ($H^2 = 0.88$), DS ($H^2 = 0.87$) and ASI ($H^2 = 0.87$), while plant aspect (PA) had the least ($H^2 = 0.05$). Under optimum-N condition, the environmental main effect was highly significant ($\rho \leq 0.01$) for GY and other traits, while the genotypes showed significant differences ($\rho \leq 0.05$) for GY, DA, DS, ASI, PA, and highly significant differences ($\rho \leq 0.01$) for PHT and EHT. However, the interactive effect of Genotype and environment was only significant ($\rho \leq 0.05$) for DA, DS, PHT and PA. Grain yield ($H^2 = 0.53$), PHT ($H^2 = 0.42$) and EHT ($H^2 = 0.43$) had moderate heritability while other traits had below 0.03 (Table 1).

The overall mean GY of the hybrids under low-N was 2,473 kg/ha, whereas, under optimum-N condition, it was 5,262 kg/ha, representing a yield reduction of 53% under low-N (Table 2). Grain yield varied from 547 kg/ha (SMLW143 × IITA1876) to 5,742 kg/ha (SMLW74 × SAM50M) under low-N, and it ranged from 2,351kg/ha (SMLW140 × IITA1878) to 8,155kg/ha (SMLW147 × IITA1878) under optimum-N condition. SMLW74 × SAM50M, SMLW146 × IITA1878, and SMLW147 × SAM50M were the top yielders, with yields of over 5,000 kg/ha in low-N environments. SMLW147 × IITA1878 and SMLW25 × SAM50M stood out with the highest yield of over 8,000 kg/ha in optimum-N conditions, with a yield advantage of over 25% compared to the best check (SAMMAZ50). For the other agronomic traits, the hybrids consistently demonstrated improved mean performance under optimum-N conditions compared to low-N conditions (Figure 1). Under optimum-N conditions, both days to anthesis and days to silking decreased significantly, from 64 to 57 (DA) and from 68 to 60 (DS). The ASI was significantly higher under low-N conditions at 3.76 compared to optimal conditions, where it reduced to 2.61. The hybrids' mean PHT increased from 135 cm under low-N to 146 cm, and EHT exhibit-

T a b l e 1

Mean squares from combined ANOVA of grain yield and agronomic traits of maize hybrids evaluated under low and optimum-N conditions at Zaria and Mokwa in 2020 and 2021

| Source of variation | df | Grain yield | Days to anthesis | Days to silking | Anthesis-silking interval | Plant Height | Ear height | Ear per plant | Plant aspect | Ear aspect | Stay green characteristic |
|---------------------|-----|-------------------------|-----------------------|-----------------------|---------------------------|-------------------------|------------------------|---------------------|----------------------|---------------------|---------------------------|
| Low-N conditions | | | | | | | | | | | |
| Environment (E) | 3 | 394104880 ⁺⁺ | 67.09 ⁺⁺ | 45.22 ⁺⁺ | 2.56 ⁺ | 179525.2 ⁺⁺ | 194001.5 ⁺⁺ | 0.666 ⁺⁺ | 172.26 ⁺⁺ | 20.24 ⁺⁺ | 237.12 ⁺⁺ |
| Rep (Environment) | 4 | 32148421 ⁺⁺ | 15.17 ⁺ | 23.51 ⁺ | 0.93 | 6530.77 ⁺⁺ | 1215.4 ⁺⁺ | 0.231 ⁺ | 6.76 ⁺⁺ | 1.07 | 175.07 ⁺⁺ |
| Block (Env*Rep) | 112 | 5040356 ⁺⁺ | 5.80 | 6.11 | 0.64 | 1290.57 ⁺⁺ | 445.1 ⁺⁺ | 0.085 ⁺⁺ | 1.08 ⁺⁺ | 2.04 ⁺⁺ | 1.30 |
| Genotype (G) | 239 | 5236504 ⁺⁺ | 54.36 ⁺⁺ | 49.61 ⁺⁺ | 5.19 ⁺⁺ | 1447.1 ⁺⁺ | 310.7 ⁺⁺ | 0.053 | 0.39 | 1.03 ⁺ | 21.09 ⁺⁺ |
| G × E | 717 | 3063943 ⁺⁺ | 6.61 | 6.69 | 0.67 ⁺ | 760.4 ⁺ | 252.2 ⁺⁺ | 0.057 | 0.37 | 0.94 ⁺ | 1.63 ⁺⁺ |
| Pooled Error | 844 | 2309967 | 6.36 | 6.48 | 0.58 | 642.2 | 189.6 | 0.051 | 0.33 | 0.79 | 1.05 |
| CV [%] | | 61.45 | 3.94 | 3.76 | 20.21 | 18.84 | 22.37 | 26.30 | 16.25 | 21.40 | 22.98 |
| H ² | | 0.41 | 0.88 | 0.87 | 0.87 | 0.47 | 0.19 | 0.002 | 0.05 | 0.09 | 0.92 |
| Optimum conditions | | | | | | | | | | | |
| Environment, E | 3 | 694838179 ⁺⁺ | 2560.24 ⁺⁺ | 2183.84 ⁺⁺ | 171.67 ⁺⁺ | 131920.59 ⁺⁺ | 79429.81 ⁺⁺ | 1.24 ⁺⁺ | 97.08 ⁺⁺ | 23.70 ⁺⁺ | – |
| Rep (E) | 4 | 18316147 ⁺ | 77.85 ⁺⁺ | 112.19 ⁺⁺ | 12.37 ⁺⁺ | 4455.88 ⁺⁺ | 1813.50 ⁺⁺ | 1.18 ⁺⁺ | 11.36 ⁺⁺ | 8.54 ⁺⁺ | – |
| Block (E × Rep) | 112 | 6531293 | 16.71 ⁺⁺ | 20.24 ⁺⁺ | 1.92 ⁺⁺ | 1108.96 ⁺ | 450.91 ⁺⁺ | 0.08 ⁺ | 1.34 ⁺⁺ | 0.69 ⁺⁺ | – |
| Genotype, G | 239 | 6905449 ⁺ | 8.40 ⁺ | 10.07 ⁺ | 1.25 ⁺ | 1431.73 ⁺⁺ | 486.08 ⁺⁺ | 0.07 | 0.29 ⁺ | 0.42 | – |
| G × E | 717 | 3236153 | 7.46 ⁺ | 8.67 ⁺ | 1.12 | 836.25 [*] | 275.02 | 0.06 | 0.28 ⁺ | 0.46 | – |
| Error | 844 | 3182939 | 6.47 | 7.59 | 1.03 | 743.17 | 256.06 | 0.06 | 0.23 | 0.41 | – |
| CV [%] | | 43.48 | 4.45 | 4.61 | 38.78 | 18.71 | 25.98 | 24.93 | 18.03 | 24.17 | – |
| H ² | | 0.53 | 0.11 | 0.14 | 0.10 | 0.42 | 0.43 | 0.14 | 0.03 | – | – |

⁺, ⁺⁺, Significant at 0.05 and 0.01 probability levels, respectively; H² – broad sense heritability; CV – coefficient of variation; df – degree of freedom.

T a b l e 2

Mean grain yield and agronomic traits of 237 maize test crosses (best 15 and worst 5 based on LNTI) and three hybrid checks evaluated under low- (LN) and optimum-N (OPT) conditions at Mokwa and Zaria in 2020 and 2021

| Hybrids | GY | | DA | | ASI | | PHT | | EPP | | PA | | EA | | STGR | LNTI | YRD (%) |
|--------------------|--------|--------|-------|-------|------|------|-------|-------|-----|------|------|------|------|------|------|-------|---------|
| | LN | OPT | LN | OPT | LN | OPT | LN | OPT | LN | OPT | LN | OPT | LN | OPT | | | |
| SMLW147 × SAM50M | 5126.2 | 7337.0 | 60.5 | 56.2 | 5.1 | 2.7 | 144.9 | 124.8 | 0.8 | 1.0 | 3.0 | 2.5 | 3.4 | 2.4 | 1.9 | 13.53 | 30.13 |
| SMLW74 × SAM50M | 5742.1 | 5815.2 | 62.9 | 56.2 | 4.1 | 2.4 | 123.9 | 141.6 | 0.9 | 1.0 | 3.4 | 2.8 | 3.4 | 3.0 | 2.2 | 12.98 | 1.26 |
| SMLW105 × IITA1876 | 3683.9 | 4847.1 | 66.8 | 57.6 | 3.0 | 2.9 | 153.1 | 147.1 | 1.0 | 0.9 | 2.9 | 2.6 | 3.1 | 2.3 | 4.0 | 9.98 | 24.00 |
| SMLW44 × IITA1876 | 3342.1 | 4288.2 | 62.8 | 57.1 | 4.0 | 2.9 | 156.7 | 132.7 | 0.9 | 0.9 | 2.9 | 2.9 | 2.6 | 2.5 | 2.0 | 9.78 | 22.06 |
| SMLW100 × IITA1876 | 4486.0 | 5023.3 | 66.5 | 55.7 | 1.9 | 2.8 | 133.8 | 133.2 | 0.9 | 1.0 | 3.5 | 2.6 | 4.0 | 2.6 | 2.7 | 9.34 | 10.70 |
| SMLW106 × SAM50M | 3891.9 | 6144.3 | 63.0 | 54.9 | 3.0 | 2.9 | 153.8 | 139.2 | 0.8 | 1.0 | 3.1 | 2.6 | 4.1 | 2.9 | 3.2 | 8.74 | 36.66 |
| SMLW7 × IITA1878 | 4551.4 | 6399.0 | 63.3 | 59.0 | 3.3 | 2.2 | 125.8 | 143.1 | 1.0 | 1.1 | 3.7 | 2.5 | 3.7 | 2.7 | 4.0 | 8.53 | 28.87 |
| SMLW57 × IITA1878 | 4125.4 | 7143.9 | 62.1 | 56.3 | 3.4 | 2.2 | 128.5 | 157.4 | 1.2 | 1.2 | 3.6 | 2.8 | 3.7 | 2.5 | 4.3 | 8.51 | 42.25 |
| SMLW70 × SAM50M | 3962.9 | 4569.2 | 63.6 | 56.8 | 3.8 | 3.0 | 135.3 | 149.8 | 0.9 | 0.9 | 2.9 | 2.4 | 3.8 | 2.5 | 2.3 | 8.50 | 13.27 |
| SMLW22 × SAM50M | 2957.1 | 4629.1 | 67.8 | 55.9 | 2.6 | 2.6 | 148.2 | 162.6 | 0.9 | 0.9 | 3.3 | 2.6 | 2.8 | 3.0 | 2.7 | 8.41 | 36.12 |
| SMLW48 × IITA1878 | 2952.6 | 6502.5 | 61.4 | 58.5 | 4.0 | 2.2 | 162.2 | 139.4 | 1.0 | 1.2 | 3.0 | 2.5 | 3.0 | 2.7 | 5.3 | 8.10 | 54.59 |
| SMLW120 × SAM50M | 4028.4 | 5821.6 | 63.9 | 59.5 | 2.9 | 3.0 | 144.0 | 158.6 | 0.9 | 1.1 | 3.4 | 2.7 | 4.0 | 2.9 | 3.5 | 7.84 | 30.80 |
| SMLW10 × SAM50M | 3466.1 | 5496.6 | 66.3 | 57.2 | 2.4 | 3.2 | 146.0 | 134.8 | 0.8 | 1.1 | 3.7 | 2.6 | 3.5 | 2.3 | 1.7 | 7.70 | 36.94 |
| SMLW147 × IITA1878 | 4019.0 | 8155.0 | 61.4 | 56.7 | 3.5 | 2.7 | 128.9 | 123.6 | 0.9 | 1.0 | 3.3 | 2.5 | 3.7 | 2.3 | 3.5 | 7.66 | 27.31 |
| SMLW121 × IITA1876 | 3314.4 | 6594.6 | 64.8 | 59.0 | 2.9 | 3.2 | 151.7 | 159.4 | 0.9 | 1.0 | 3.3 | 2.6 | 3.9 | 2.6 | 4.1 | 7.40 | 49.74 |
| SMLW155 × SAM50M | 1615.6 | 4257.2 | 60.0 | 55.9 | 4.9 | 2.8 | 100.2 | 117.3 | 0.8 | 0.9 | 3.8 | 3.0 | 4.7 | 2.7 | 3.9 | -5.87 | 62.05 |
| SMLW140 × IITA1878 | 1692.6 | 2351.2 | 61.1 | 57.9 | 3.9 | 3.4 | 115.4 | 134.9 | 0.9 | 0.9 | 3.6 | 3.1 | 4.3 | 3.3 | 6.6 | -6.08 | 28.01 |
| SMLW93 × IITA1878 | 2132.0 | 3078.2 | 62.1 | 56.9 | 4.4 | 2.3 | 126.7 | 157.7 | 0.9 | 0.9 | 3.6 | 2.8 | 4.4 | 2.3 | 8.5 | -6.28 | 30.74 |
| SMLW5 × IITA1876 | 1650.1 | 3090.4 | 60.7 | 58.7 | 4.0 | 3.3 | 131.6 | 129.8 | 0.7 | 1.0 | 4.0 | 2.7 | 4.9 | 3.2 | 3.0 | -6.94 | 46.61 |
| SMLW169 × IITA1878 | 1451.2 | 5338.4 | 65.0 | 56.3 | 4.9 | 1.9 | 112.7 | 160.1 | 0.7 | 1.1 | 3.9 | 2.6 | 4.7 | 2.4 | 7.0 | -7.78 | 72.82 |
| SAMMAZ 50 | 1729.5 | 6334.7 | 65.6 | 59.1 | 2.7 | 3.2 | 124.8 | 117.7 | 0.9 | 0.8 | 3.6 | 2.9 | 4.4 | 2.3 | 5.7 | -2.58 | 72.70 |
| Oba Super 2 | 979.8 | 6088.3 | 67.2 | 56.5 | 3.0 | 2.7 | 133.9 | 139.2 | 0.8 | 0.9 | 3.4 | 2.5 | 4.4 | 2.5 | 4.0 | -3.29 | 83.91 |
| SC 619 | 2184.5 | 6320.7 | 67.2 | 58.9 | 2.9 | 2.7 | 129.3 | 132.6 | 0.9 | 0.9 | 3.7 | 2.5 | 4.0 | 2.6 | 4.7 | -0.75 | 65.44 |
| MEAN | 2473.2 | 5262.7 | 64.03 | 57.14 | 3.76 | 2.61 | 134.5 | 145.7 | 0.9 | 0.97 | 3.53 | 2.67 | 4.14 | 2.65 | 4.46 | - | - |
| LSD (α.0.05) | 1491.6 | 2245.6 | 2.48 | 2.5 | 0.75 | 0.99 | 24.9 | 26.75 | 0.2 | 0.24 | 0.56 | 0.47 | 0.00 | 0.01 | 1.01 | - | - |

GY – grain yield [kg/ha]; DA – days to anthesis; ASI – anthesis silking interval; PHT – plant height; EPP – numbers of ears per plant; PA – plant aspect; EA – ear aspect; STGR – stay green characteristics; LNTI – low-N tolerance index; YRD – yield reduction [%].

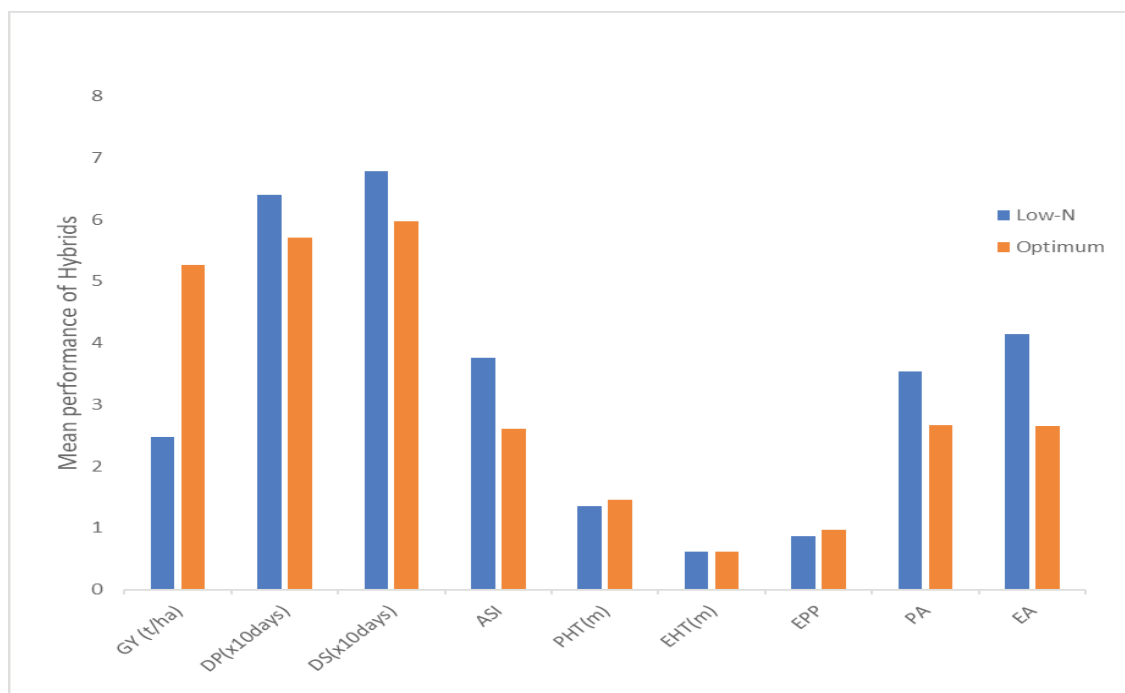


Figure 1. Mean grain yield and other agronomic traits of the 240 maize hybrids evaluated under low-N and optimum-N environments.

Note: GY – grain yield; PHT – plant height; EHT – ear height; EPP – number of ears per plant; PA – plant aspect; EA – ear aspect; DP – days to anthesis; DS – days to silking; ASI – anthesis silking interval; STGR – stay green characteristics.

ed a marginal increase from 60 cm under low-N to 62 cm under optimum-N. Plant aspect and ear aspect ratings improved under optimum-N (2.67 and 2.65, respectively) compared to under low-N (3.53 and 4.14, respectively). One hundred and eleven (111) hybrids had a positive low-N tolerance index (LNTI) greater than 1, twenty-two (22) had LNTI greater than 0 but less than 1, while the remaining one hundred and four (104) had a negative LNTI. Hybrids SMLW147 × SAM50M and SMLW74 × SAM50M ranked first and second, respectively, in terms of both grain yield and LNTI (Table 2).

Performance and stability of maize hybrids across environments

Figures 2 and 3 show the polygon view of the GGE biplot, depicting the grain yield performance of the top 15 and bottom 10 low-N tolerant hybrids, alongside three checks, across eight environments in Nigeria. The biplot explained 53.49% of the total variation for grain yield, with 20.09% attributed to principal component axis 1 (PC1) and 33.4% attributed to Principal component 2 (PC2). Figure 2 shows the “which-won-where/what” polygon view that enabled the identification of vertex hybrids, i.e.,

genotypes positioned at one of the vertices (corners) of the biplot polygon. SMLW147 × IITA1878 (14) was identified as the vertex hybrid for environments Mokwa optimum-N 2020 (MOP20), Zaria optimum-N 2020 (ZOP20), Mokwa optimum-N 2021 (MOP21) and Zaria optimum-N 2021 (ZOP21). Hybrid SMLW147 × SAM50M (1) exhibited the highest grain yield performance at environment Mokwa low-N 2020 (MLN20), Mokwa low-N 2021 (MLN21) and Zaria low-N 2021 (ZLN21) while SMLW74 × SAM50M (2) was the best for environment Mokwa optimum-N 2021 (MOP21). The vertex hybrids SMLW140 × IITA1878 (37), SMLW5 × IITA1876 (39) and SMLW23 × IITA1878(35), exhibited the lowest yields across all or some of the environments. The hybrids located within the polygon, especially closer to the biplot origin, demonstrated less responsiveness compared to those within the polygon vertices.

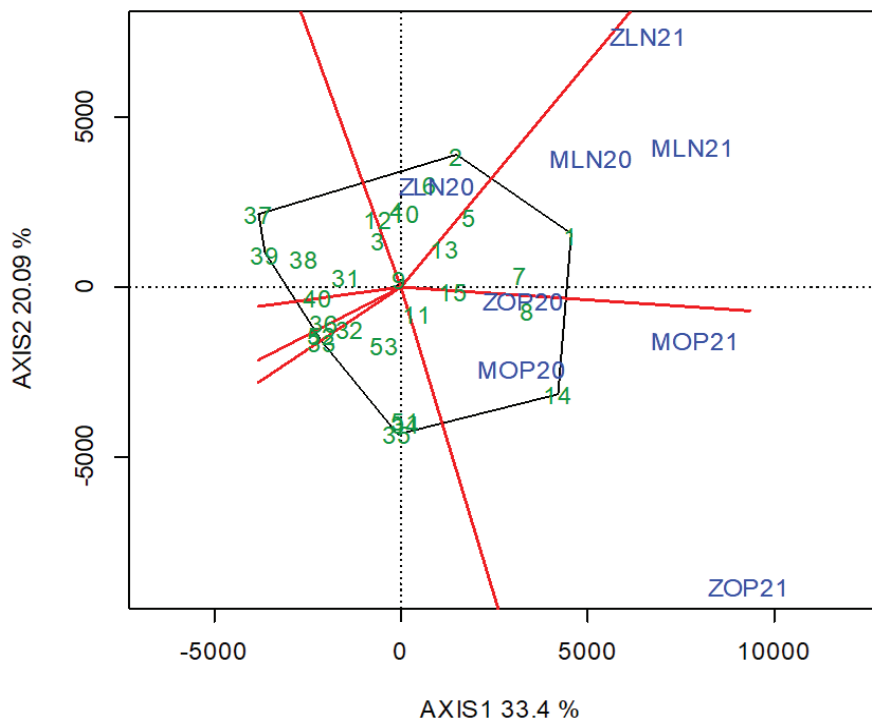
Figure 3 shows the “mean vs stability” view for the identification of low-N tolerant hybrids with high grain yield and stability across the eight (8) environments. The absolute length of each hybrid’s projection on the average tester axis represents its average yield performance, while its projection on

the ATC y-axis measures its stability. Based on these criteria, SMLW147 × SAM50M (1), SMLW147 × IITA1878 (14), SMLW57 × IITA1878 (8), SMLW7 × IITA1878 (7), and SMLW100 × IITA1876 (5) emerged as the highest yielding hybrids across the eight environments, while SMLW140 × IITA1878 (37) and SMLW5 × IITA1876 (39) were identified as the lowest yielders. Hybrids SMLW7 × IITA1878 (7), SMLW147 × SAM50M (1) and SMLW57 × IITA1878 (8) were the most stable hybrids with the highest yield performance. In contrast, hybrid

SMLW147 × IITA1878 (14) recorded the highest yield, but it was highly unstable across the environments.

Discriminating power and representativeness of the test environments

The test environments ZLN21, ZOP21, MLN21, MOP21, and MLN20 had long vector lengths, providing significant information for discriminating among hybrids (Figure 4). Conversely, environments ZOP20, MOP20, and ZLN20 had short vec-



| Code | Environment | Code | Hybrid | Code | Hybrid |
|-------|----------------------|------|---------------------|------|--------------------|
| ZLN20 | Zaria low-N 2020 | 1 | SMLW147 × SAM50M | 31 | SMLW104 × IITA1878 |
| ZLN21 | Zaria low-N 2021 | 2 | SMLW74 × SAM50M | 32 | SMLW52 × IITA1878 |
| ZOP20 | Zaria optimum-N 2020 | 3 | SMLW105 × IITA1876 | 33 | SMLW135 × SAM50M |
| ZOP21 | Zaria optimum-N 2021 | 4 | SMLW44 × IITA1876 | 34 | SMLW24 × IITA1878 |
| MLN20 | Mokwa low-N 2020 | 5 | SMLW100 × IITA1876 | 35 | SMLW23 × IITA1878 |
| MLN21 | Mokwa low-N 2021 | 6 | SMLW106 × SAM50M | 36 | SMLW155 × SAM50M |
| MOP20 | Mokwa optimum-N 2020 | 7 | SMLW7 × IITA1878 | 37 | SMLW140 × IITA1878 |
| MOP21 | Mokwa optimum-N 2021 | 8 | SMLW57 × IITA1878 | 38 | SMLW93 × IITA1878 |
| | | 9 | SMLW70 × SAM50M | 39 | SMLW5 × IITA1876 |
| | | 10 | SMLW22 × SAM50M | 40 | SMLW169 × IITA1878 |
| | | 11 | SMLW48 × IITA1878 | 51 | SAMMAZ50 |
| | | 12 | SMLW120 × SAM50M | 52 | Oba Super 2 |
| | | 13 | SMLW10 × SAM50M | 53 | SC 619 |
| | | 14 | SMLW147 × IITA 1878 | | |
| | | 15 | SMLW121 × IITA1876 | | |

Figure 2. A ‘which-won-where/what’ genotype and genotype × environment (GGE) biplot of 28 (best 15, worst 10, and 3 checks) low-N tolerant hybrids across low-N and optimum-N environments in Nigeria during 2020 and 2021 growing season.

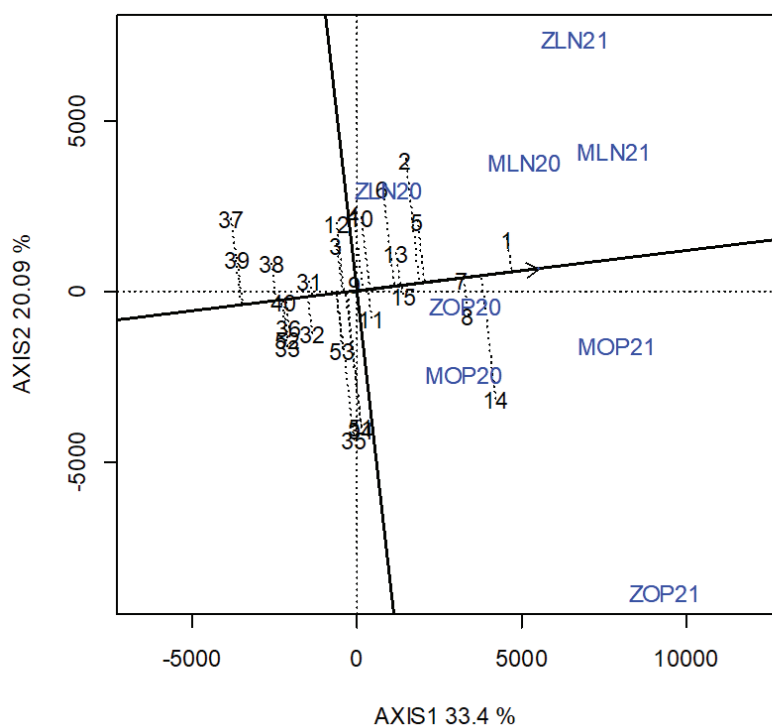


Figure 3. An entry/tester genotype and genotype \times environment (GGE) biplot of 28 (best 15, worst 10, and 3 checks) low-N tolerant hybrids across low-N and optimum-N environments in Nigeria during 2020 and 2021 growing season.

tor lengths, indicating they were less discriminating and offered limited information about the hybrids. Notably, ZLN21 and ZOP21 were characterized by long vectors and large angles with the AEC abscissa, while MLN21 and MOP21 had long vectors and small angles with the AEC abscissa. Two mega-environments, which showed strong correlations in hybrid rankings, were identified: the first comprised ZLN20, ZLN21, MLN20, and MLN21, while the second comprised MOP20, ZOP20, ZOP21, and MOP21.

Genotype – trait biplot analysis showing interrelationship among traits of maize hybrids

In the low-N conditions (Figure 5), acute angles were observed between GY and PHT, as well as between DA and DS. Additionally, ASI, EHT, and EPP trait vectors formed acute angles with the GY vector. STGR, EA and PA exhibited a near-perfect linear relationship with GY. Conversely, vectors for DS and DP were inclined at almost right angles to GY. Under optimum-N conditions (Figure 6), acute angles were observed between GY and EPP, between PHT and EHT, as well as between DA and DS. However, obtuse angles were noted between

GY and each of DA, DS, EA, PA, and ASI. Notably, the vectors for PHT and EHT traits exhibited a weak correlation (at an angle of 96°) with grain yield.

DISCUSSION

The significant genotypic factor across various traits, such as grain yield, flowering traits, growth traits, and leaf senescence, underscores the inherent variability among the studied genotypes. Some genotypes outperformed others, reflecting the genetic diversity within the maize population, providing a valuable resource for breeding programs seeking to develop improved and adaptable maize varieties. The greater contribution of environmental factors on yield and other traits, compared to genotype and genotype \times environment interaction highlights the high variability observed across the test environments. This finding aligns with earlier studies (Badu-Apraku *et al.* 2011; Mafouasson *et al.* 2018; Bankole *et al.* 2023), which consistently reported that the largest proportion of total variation in multi-environment trials was attributed to environmental factors or locations.

The observed significant $G \times E$ interaction for grain yield under low-N conditions, emphasizes the dynamic relationship between genotype and environment in influencing maize performance. This interaction led to varying hybrid yield rankings across different low-N environments, demonstrating the need for thorough testing to identify hybrids with both high yield potential and stability under N-stress conditions. Contrastingly, under optimum-N conditions, where the $G \times E$ interaction was less significant, genotypic performance remained more consistent across diverse environments. This consistency simplified the identification and selection of high-yielding hybrids, as genetic factors exhibited a more stable and predictable influence on the traits of interest.

Nitrogen, as a primary component of essential plant molecules such as amino acids, proteins, and chlorophyll, significantly influences the growth and development of maize plants (Havlin *et al.* 2014). Adequate nitrogen availability is crucial for optimal photosynthesis, energy transfer, and nitrogen metabolism, collectively contributing to overall maize crop performance (Havlin *et al.* 2014; Cassim *et al.* 2022). The mean performance of hybrids under both low-N and optimum-N conditions underscored the substantial impact of nitrogen availability on various agronomic traits, particularly grain yield. The

study's results highlighted the profound impact of nitrogen availability on grain yield, wherein under low-N conditions, the hybrids' mean grain yield was significantly lower than under optimum-N conditions. This aligns with established knowledge indicating that nitrogen plays a pivotal role in chlorophyll synthesis, essential for photosynthesis. Adequate nitrogen levels enhance the plant's ability to convert solar energy into carbohydrates, ultimately leading to increased grain production (Bänziger *et al.* 2000; Fageria & Baligar 2005).

Furthermore, nitrogen availability significantly influenced the timing of critical developmental stages in maize. Under optimum-N conditions, there was a notable acceleration in reproductive development, indicated by a reduction in both days to anthesis and days to silking. This acceleration can be attributed to nitrogen's role in promoting the synthesis of proteins and enzymes involved in flowering, ultimately enhancing pollination and fertilisation processes. The delayed development under low-N conditions, aligns with previous findings (Bänziger *et al.* 2000; Fageria & Baligar 2005; Havlin *et al.* 2014; Zuffo *et al.* 2021), underscoring the importance of nitrogen in promoting timely flowering and reproductive processes.

The Anthesis Silking Interval, a critical parameter influencing maize reproductive success, was

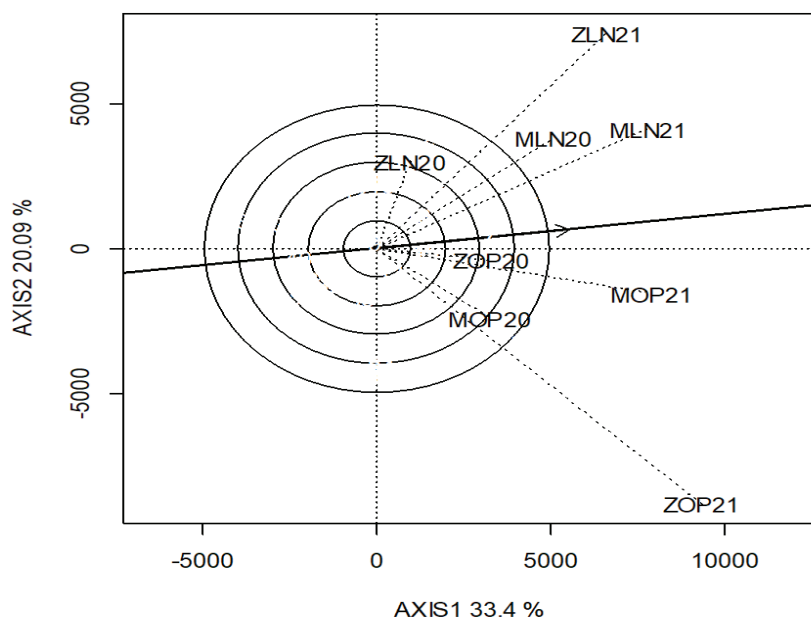


Figure 4. The ‘discriminating power and representativeness’ view of GGE biplot of 28 (best 15, worst 10, and 3 checks) low-N tolerant hybrids across low-N and optimum-N environments in Nigeria during 2020 and 2021 growing season.

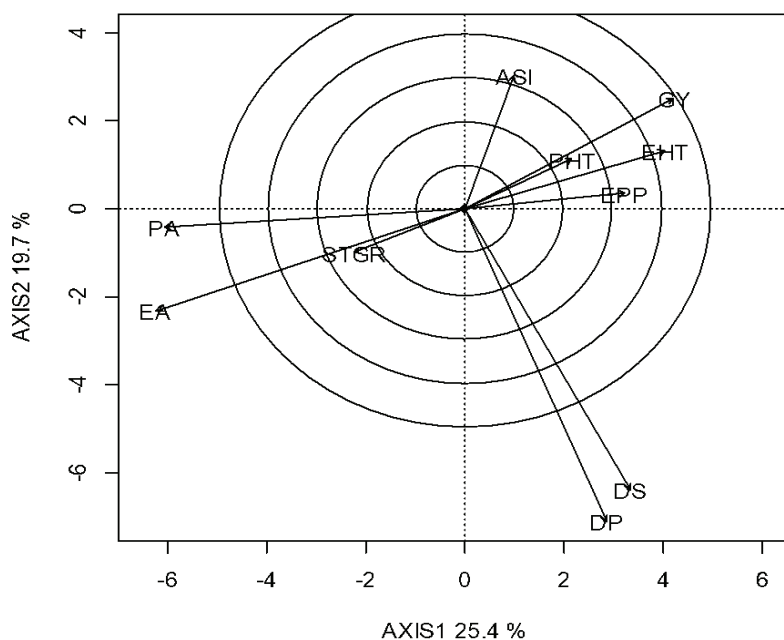


Figure 5. A vector view of the genotype-by-trait biplot showing interrelationships among traits of 60 selected maize hybrids evaluated under low-N conditions at Zaria and Mokwa in 2020 and 2021.
 Note: GY – grain yield; PHT – plant height; EHT – ear height; EPP – number of ears per plant; PA – plant aspect; EA – ear aspect; DP – days to anthesis; DS – days to silking; ASI – anthesis silking interval; STGR – stay green characteristics.

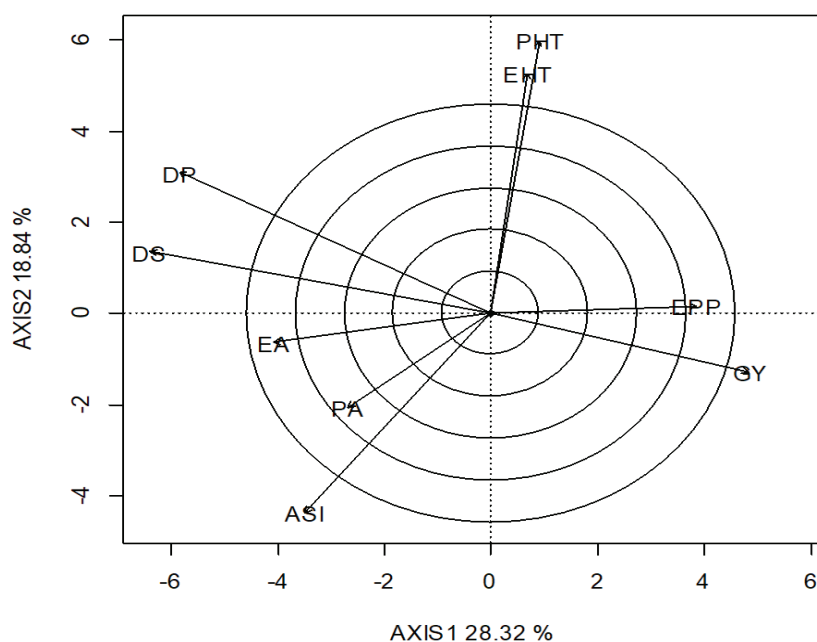


Figure 6. A vector view of the genotype-by-trait biplot showing interrelationships among traits of 60 selected maize hybrids evaluated under optimum-N condition at Zaria and Mokwa in 2020 and 2021.
 Note: GY – grain yield; PHT – plant height; EHT – ear height; EPP – number of ears per plant; PA – plant aspect; EA – ear aspect; DP – days to anthesis; DS – days to silking; ASI – anthesis silking interval.

significantly higher under low-N conditions compared to the optimal conditions. This suggests that nitrogen deficiency can extend the time gap between male and female flowering, potentially impacting pollination, and grain formation (Bänziger *et al.* 2000). Nitrogen availability also had a profound impact on the overall architecture of the maize hybrids. Plant height and ear height, crucial components of plant architecture influencing light interception and resource utilization, exhibited notable changes under optimum nitrogen conditions. The hybrids were taller under optimum-N conditions compared to low-N, highlighting the role of nitrogen in promoting vigorous vegetative and reproductive growth. Plant aspect and ear aspect, indicators of plant and ear morphology, respectively, exhibited significant improvements under optimum nitrogen conditions. These changes suggest that nitrogen availability contributed to more robust and visually appealing plant and ear structures, reflecting the overall health and vigour of the maize crop.

Furthermore, notable differences were observed in the performance of the maize hybrids under the different soil N conditions, with some hybrids standing out as top performers. Notably, SMLW74 × SAM50M and SMLW147 × SAM50M demonstrated exceptional yield performance and remarkable tolerance to low soil-N conditions, positioning them as the most tolerant hybrids in the study. Their high LNTI values indicate that these hybrids might have inherent genetic advantages or physiological traits that enable them to adapt well to low soil-N conditions. Under optimum-N conditions, SMLW147 × IITA1878, SMLW25 × SAM50M, and SMLW121 × IITA1878 emerged as the top-performing hybrids in terms of yield. These hybrids, with high yield performance only under optimum environments but with low yield performance under low-N conditions, could be considered suitable for cultivation under optimum-N condition.

The “which-won-where/what” biplot proved instrumental in identifying vertex hybrids in each environment. Vertex hybrids, positioned at the vertices of the biplot polygon, showcased distinct performance patterns and adaptability to specific environments within that sector (Yan & Tinker 2006). Meanwhile, the “mean vs. stability” biplot facilitated the identification of hybrids with high-yield per-

formance and stability across both low-N and optimum-N environments. Among the identified low-N tolerant hybrids, SMLW147 × SAM50M demonstrated excellent yields and moderate stability across test environments, making it a viable option for cultivation in both low-N and optimum-N conditions. Conversely, SMLW147 × IITA1878, despite its high yield, exhibited low stability across test environments, suggesting its suitability for location-specific plantings, particularly under optimum-N conditions at Mokwa and Zaria.

In addition, the purpose of test environment evaluation is to identify the ideal environment for distinguishing superior genotypes, considering discriminativeness and representativeness (Yan & Tinker 2006; Badu-Apraku *et al.* 2011). The length of the environment vector indicates its discriminating power in evaluating cultivars across test environments, i.e., the longer the vector, the higher the ability to discriminate among cultivars in that environment (Bankole *et al.* 2023). Thus, environments ZLN21 and ZOP21, with longer vector lengths, were the most discriminating and informative, followed by MOP21, MLN21 and MLN20. Conversely, test environments ZLN20, MOP20 and ZOP20, with shorter vector lengths, are considered less discriminating, providing little information about the low-N tolerant hybrids.

On the other hand, the representativeness of test environment reflects its ability to represent the mega-environment, determined by the angle between the test environment and the AEA. Smaller angles indicate greater representativeness, while larger angles suggest less representativeness. In this study, the test environments were divided into two mega-environments on the biplot: the four low-N environments (ZLN20, ZLN21, MLN20, and MLN21) constituting one mega-environment and the four optimum environments (ZOP20, MOP20, MOP21, and ZOP21) constituting the other mega-environment. MLN21 and MOP21, with longer vector lengths and the closest proximity to the AEA in their mega-environments, are considered the most discriminating and representative, ideal for selecting superior hybrids. In contrast, ZLN21 and ZOP21, with long vector lengths and a large angle with the AEA, are discriminating but non-representative test environments, making them unsuitable for selecting

superior hybrids but effective for culling unstable genotypes (Yan & Tinker 2006; Badu-Apraku *et al.* 2011). Additionally, the observed construction of mega-environments based on different nitrogen treatments, rather than different locations or growing seasons, implies that nitrogen availability emerges as a pivotal variable influencing the discriminative and representative qualities of the environments. While geographical and seasonal variations such as climate and soil types may affect other aspects of maize cultivation, the emphasis on nitrogen treatments in the mega-environment construction underscores the critical role of nitrogen in the performance of maize hybrids.

Genotype by trait biplots provided insights into associations between grain yield and various agronomic traits. Under low-N conditions, hybrids with increased plant height, greater ear height, shorter anthesis-silking intervals, and a higher number of ears per plant correlated positively with high grain yields. Conversely, traits related to plant and ear aspect ratings, as well as stay-green characteristics, displayed negative correlations with grain yield, suggesting that hybrids with less favourable phenotypic characteristics in terms of ear and plant aspects, accompanied by higher levels of leaf yellowing, are likely to experience reduced grain yields. Analysing the vector lengths of traits with significant effects on grain yield in the biplot, traits such as anthesis-silking interval, ear height, plant aspect, and ear aspect had long vectors, indicating their potential value in selecting hybrids for high grain yield under low-N conditions (Ribeiro *et al.* 2020).

Under optimum-N conditions, the plant aspect and ear aspect also displayed negative correlations with grain yield, suggesting that even under conditions with optimum nitrogen availability, the phenotypic characteristics of plant and ear aspects remain crucial factors influencing high grain yields. In contrast to low-N conditions, where the correlation between grain yield and both days to anthesis and silking was non-significant and weak, under optimum-N conditions, the correlation was significant and negative. Significantly, considering the long vectors for both days to anthesis and silking on the biplot under optimum-N conditions, it becomes evident that the timing of flowering events plays a pivotal role in achieving high grain. Additionally, most

high-yielding hybrids in the study exhibited a greater number of ears per plant compared to low-yielding hybrids. Surprisingly, these high-yielding hybrids achieved anthesis and silking earlier, while low-yielding hybrids took a longer time to complete these events. This observation contrasts with previous reports (Kamara *et al.* 2009; Hussain *et al.* 2011; Adu *et al.* 2021) that suggest late-maturing maize varieties are typically higher yielding compared to early-maturing ones. One possible explanation for these findings could be attributed to a combination of environmental conditions and specific genetic characteristics inherent in the maize hybrids studied.

CONCLUSIONS

This study unveiled the pivotal role of nitrogen availability influencing key agronomic traits of maize, from grain yield to developmental stages and overall plant architecture. Two test environments, MLN21 (Mokwa low-N 2021) and MOP21 (Mokwa optimum-N 2021), were identified as highly discriminating and representative, making them ideal for selecting hybrids with broad adaptability. Conversely, ZLN21 (Zaria low-N 2021) and ZOP21 (Zaria optimum-N 2021), though highly discriminating, were deemed non-representative and can be utilized for selecting hybrids specifically adapted to those environments.

Additionally, the study identified the hybrid SMLW147 × SAM50M as a standout performer, exhibiting high yield, tolerance to low soil-N, and stability across diverse test environments. This hybrid is recommended as the optimal choice for cultivation under both low-N and optimum-N conditions. Conversely, SMLW147 × IITA1878, the highest yielding hybrid in all four optimum-N environments, but with low stability across the test environments, is deemed more suitable for cultivation under optimum-N conditions. Further evaluations and on-farm assessments are recommended to confirm the consistency of these hybrids' performance across contrasting environments, potentially paving the way for commercialization in Nigeria. Importantly, this study justified the inclusion of anthesis-silking interval, plant aspect, and ear aspect in selection indices for identifying low-N tolerant genotypes.

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