

# Combining ability and heterotic grouping of maize (*Zea mays* L.) inbred lines for tolerance to low soil nitrogen in Nigeria

## Habilidad combinatoria y agrupación heterótica de líneas de maíz (*Zea mays* L.) tolerantes al bajo nivel de nitrógeno del suelo en Nigeria

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### Abstract

Development of low soil – Nitrogen (N) tolerant maize hybrids is vital for the sustainability of maize production in the developing countries. This study investigated the potential of newly developed inbred lines for trait enhancement and population improvement by estimating their general combining ability effects (GCA). Employing a line x tester mating design involving 79 inbred lines and three testers, a total of 237 F1 hybrids were generated and evaluated alongside three checks at four low-N and four optimum-N environments in Nigeria in 2020 and 2021. Significant positive and negative GCA effects were observed across various traits, underscoring the presence of both superior and inferior combiners for these traits. Inbreds SMLW150 and SMLW155 exhibited significant ( $p \leq 0.01$ ) GCA effects for grain yield, recording 733.18 kg.ha<sup>-1</sup> and 776 kg.ha<sup>-1</sup> respectively under optimum-N conditions, thus emerging as the best combiners for grain yield and valuable genetic resources for grain yield improvement. Similarly, inbred lines SMLW7, SMLW9, SMLW57, SMLW43, and SMLW146 demonstrated noteworthy GCA effects for stay-green characteristics, positioning them as promising candidates for enhancing low-N tolerance. Inbred lines SMLW146 and SMLW147, along with tester IITA1876, emerged as best combiners for both grain yield and stay-green characteristics, making them attractive choices for parent selection in hybridization programs geared towards producing maize varieties with enhanced yield potential and nitrogen stress tolerance. Lines SMLW4, SMLW43, SMLW44, SMLW34, and SMLW37 were the best general combiner for days to anthesis and silking, making them potential parents for developing extra-early maturing maize varieties. Additionally, the inbred lines were classified into four heterotic groups under low-N and three groups under optimum-N conditions using the GCA effects of Multiple Traits (GCAMT) method. Each group exhibited distinct strengths and weaknesses. Notably, genotypes in Cluster 4 under low-N conditions and Cluster 3 under optimum-N conditions show promise as sources for developing hybrids with improved yield and nitrogen stress tolerance.

**Key words:** Additive gene action, cluster analysis, trait improvement, Per se performance, hybrid Maize.

### Resumen

El desarrollo de híbridos de maíz con baja tolerancia al nitrógeno (N) del suelo es vital para la sostenibilidad de la producción de maíz en los países en desarrollo. En este estudio se investigó el potencial de las líneas endocriadas de nuevo desarrollo para la mejora de rasgos y poblaciones mediante la estimación de sus efectos generales de capacidad combinatoria (GCA). Empleando un diseño de apareamiento línea x probador que incluía 79 líneas endocriadas y tres probadores, se generaron un total de 237 híbridos F1 y se evaluaron

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junto con tres controles en cuatro entornos de bajo N y cuatro de N óptimo en Nigeria en 2020 y 2021. Se observaron efectos GCA positivos y negativos significativos en varios rasgos, lo que subraya la presencia de combinadores superiores e inferiores para estos rasgos. Las líneas endocriadas SMLW150 y SMLW155 mostraron efectos GCA significativos ( $p \leq 0.01$ ) para el rendimiento de grano, registrando 733.18 kg.ha<sup>-1</sup> y 776 kg.ha<sup>-1</sup> respectivamente en condiciones óptimas de N, emergiendo así como los mejores combinadores para el rendimiento de grano y valiosos recursos genéticos para la mejora del rendimiento de grano. De forma similar, las líneas endocriadas SMLW7, SMLW9, SMLW57, SMLW43 y SMLW146 demostraron notables efectos GCA para las características de permanencia verde, posicionándolas como prometedoras candidatas para mejorar la tolerancia al N bajo. Las líneas endocriadas SMLW146 y SMLW147, junto con la probadora IITA1876, resultaron ser las mejores combinadoras tanto en rendimiento de grano como en características de permanencia en verde, lo que las convierte en opciones atractivas para la selección de parentales en programas de hibridación orientados a producir variedades de maíz con mayor potencial de rendimiento y tolerancia al estrés por nitrógeno. Las líneas SMLW4, SMLW43, SMLW44, SMLW34 y SMLW37 fueron las que mejor combinaron en general los días transcurridos hasta anthesis y ensilado, lo que las convierte en parentales potenciales para el desarrollo de variedades de maíz de maduración extratemprana. Además, las líneas endocriadas se clasificaron en cuatro grupos heteróticos en condiciones de bajo N y tres grupos en condiciones de N óptimo, utilizando el método de efectos GCA de Rasgos Múltiples (GCAMT). Cada grupo presentaba puntos fuertes y débiles distintos. En particular, los genotipos del grupo 4 en condiciones de bajo N y del grupo 3 en condiciones óptimas de N resultan prometedores como fuentes para el desarrollo de híbridos con mayor rendimiento y tolerancia al estrés por nitrógeno.

**Palabras clave:** *Acción génica aditiva, análisis de conglomerados, mejora de rasgos, rendimiento per se, Maíz híbrido.*

## Introduction

Maize (*Zea mays* L.) is a highly versatile crop known for its remarkable genetic plasticity and ecological adaptability, making it one of the world's most extensively cultivated food crops (Sangaré et al., 2018; Kamara et al., 2020). Sub-

Saharan Africa, in particular, demonstrates a strong dependence on maize, with an estimated daily consumption of 450 grams per person, emphasizing its pivotal role as a staple food (Ekpa et al., 2019). Beyond its significance in human nutrition, maize serves as a crucial energy feed in the livestock industry and a versatile raw material for various essential products, such as corn starch, corn syrup, corn flakes, corn oil, dextrose, lactic acid, and acetone (Fabunmi & Agbonlahor, 2012; Bankole et al., 2019). Nonetheless, despite its economic importance, maize production faces numerous challenges, including nutrient limitations, with a growing concern over low soil nitrogen further complicating these agricultural hurdles. As the demand for maize continues to rise, addressing these challenges becomes imperative. The development of maize genotypes capable of withstanding low-N stress and concurrently exhibiting high-yield potential is essential. This would not only ensure sustainable production and food security but also alleviate the financial burden on farmers associated with acquiring and applying fertilizers to enhance soil nitrogen content.

Combining ability, a fundamental concept in plant breeding, refers to the genetic potential of parental lines to positively or negatively contribute to the performance of their hybrids (Falconer & Mackay, 1996). This concept is important for identifying parents with superior genetic potentials for creating high-performing hybrids and accumulating desired alleles for targeted traits. Moreover, it serves as a tool to comprehend the genetic basis of trait expression, guiding plant breeders in selecting methods for genetic improvement and designing effective breeding strategies for future improvement programs (Oyekale et al., 2020). Additionally, information on combining abilities can be used to classify inbred lines into distinct heterotic groups. Heterotic groups comprise related or unrelated genotypes that exhibit similar combining ability effects when crossed with genotypes from other germplasm groups (Warburton et al., 2002). This classification is pivotal for effective hybrid breeding programs, as it enables breeders to strategically select parental lines from specific heterotic groups to maximize heterosis and develop hybrids with superior performance (Akinwale et al., 2021).

One widely used approach for estimating combining ability effect is the analysis of diallel mating systems, as proposed by Griffing (1956), which allows for the estimation of general combining ability and specific combining ability (SCA). Another commonly employed method is the factorial mating design, enabling the investigation of main effects and interactions among different genes or genetic factors. The line x tester mating design, introduced by Kempthorne (1957), involves crossing a set of selected lines with another set of different lines known as testers. This experimental design is extensively employed in plant breeding to estimate GCA and SCA effects. The GCA of a line is determined by evaluating its performance when crossed with various testers, and the average performance across these crosses reflects the line's GCA. SCA estimates identify line pairs that produce hybrids with either superior or inferior performance compared to what would be expected based on the GCA of the individual lines.

Furthermore, the Institute for Agricultural Research (IAR), Zaria, bears the mandate to continually develop and release maize varieties that address the challenges faced by maize growers and consumers in the country. This has led to the development of new maize inbred lines with varying genetic potentials. These newly introduced inbred lines have been evaluated under different stress conditions, with some identified as tolerant to low soil-N stress (Aboderin, et al., 2023). However, there remains a dearth of information on their combining abilities under low soil-N conditions. This study was therefore carried out to estimate the combining abilities of the inbred lines, identify the best combiner for each agronomic trait and classify the lines into heterotic groups based on their combining ability effects.

## Materials and Methods

### Source of Germplasm and Generation of Crosses

Eighty-two intermediate/late maturing inbred lines (79 lines and 3 testers) were used in the study. The seventy-nine inbred lines were selected from a pool of 110 intermediate/late

maturing white inbred lines developed by IAR maize scientist, Zaria. The selection criteria included both yield performance and tolerance to low soil nitrogen. Specifically, some lines were chosen for their good yield potential, while others were selected based on their tolerance to low soil nitrogen (Table 1). Subsequently, the seventy-nine selected lines were crossed with the three inbred testers in a line x tester mating design, resulting in 237 hybrids. This was done under irrigation during the dry season between January and April 2020 at the Institute for Agricultural Research field in Zaria. In the experimental set up, the inbred lines were planted in two rows per plot, while the testers were planted in 10 rows per plot. To ensure proper pollen synchronization, five rows of testers were planted on the same day as the inbred lines, while the remaining five rows of testers were planted seven days later. Prior to crossing, ears of both the lines and testers were bagged using ear shoot bags before the emergence of silk to prevent unnecessary cross-pollination. Tassel bags were used to collect pollen from the designated male plants (tester) and thoroughly mixed together. Pollination was performed by dusting the pollen collected in the pollen bags onto the silks of the specified ears. The pollen bags were used to cover the pollinated silks immediately after hand pollination to avoid pollen contamination. To prevent mix-ups and ensure proper identification, the pollen bags were properly labeled with permanent ink. At harvest, all ears from each testcross were bulk-shelled, dried, and packaged separately for subsequent evaluation.

### Experimental Sites Description

The evaluations were conducted at two distinct locations viz: Zaria (Elevation 640m, Longitude 8°22' E, Latitude 12°N) in the Northern Guinea savanna with an annual rainfall of 1200 mm, and Mokwa (Elevation 457m, Longitude 5°N 4'E, Latitude 9°18'N) in the Southern Guinea savanna of Nigeria with annual rainfall of 1100 mm. The IAR experimental fields in Zaria and Mokwa, established through the depletion of available soil nitrogen resulting from continuous maize cultivation without fertilizer application over several years and dedicated exclusively to

**Table 1: Description of the genetic materials used for the study**

| LINES   | PEDIGREE                            | Origin | Main Characteristics                |
|---------|-------------------------------------|--------|-------------------------------------|
| SMLW3   | SAM 18 S6-4-5-1-2-3-2-1-2-1-1-4-4   | IAR    | Susceptible to Low N                |
| SMLW4   | SAM 16 S6-3-6-1-2-2-3-1-2-1-1-1-2   | IAR    | High yielding and Tolerant to low N |
| SMLW5   | SAM 18 S6-4-5-1-2-1-2-1-2-1-1-3-3   | IAR    | Tolerant to low N                   |
| SMLW6   | SAM 18 S6-2-3-2-2-1-1-1-2-1-1-1-1   | IAR    | Susceptible to Low N                |
| SMLW7   | SAM 15 S6-1-6-1-2-1-1-2-2-1-2-1-1   | IAR    | High yielding and Tolerant to low N |
| SMLW9   | SAM 18 S6-4-5-1-2-1-3-1-2-1-1-2-3   | IAR    | Susceptible to Low N                |
| SMLW10  | TZEE-W S6-8-10-1-2-1-2-3-2-1-1-1-3  | IAR    | High yielding and Tolerant to low N |
| SMLW11  | TZEE-W S6-8-10-1-2-1-2-3-2-1-1-3-3  | IAR    | Susceptible to Low N                |
| SMLW14  | SAM 18 S6-4-3-1-2-3-2-1-2-1-1-4-5   | IAR    | Tolerant to low N                   |
| SMLW16  | SAM 18 S6-4-3-1-2-3-2-1-2-1-1-1-5   | IAR    | High Yielding                       |
| SMLW17  | SAM 16 S6-5-5-1-2-2-2-2-1-2-1-4     | IAR    | Susceptible to Low N                |
| SMLW19  | SAM 16 S6-5-5-1-2-2-2-2-1-2-3-4     | IAR    | Tolerant to low N                   |
| SMLW20  | SAM 16 S6-5-5-1-2-2-2-2-1-2-4-4     | IAR    | High Yielding                       |
| SMLW21  | SAM 16 S6-3-16-3-3-2-2-1-1-1-1-1-3  | IAR    | High yielding and Tolerant to low N |
| SMLW22  | SAM 26 S6-3-16-3-3-2-2-1-1-1-1-3-3  | IAR    | High Yielding                       |
| SMLW23  | SAM 26 S6-3-16-3-3-2-2-1-1-1-1-2-3  | IAR    | High Yielding                       |
| SMLW24  | SAM 26 S6-3-16-3-3-2-2-1-1-1-1-1-3  | IAR    | High yielding and Tolerant to low N |
| SMLW25  | SAM 16 S6-3-6-2-2-1-3-3-3-1-1-2-3   | IAR    | Susceptible to Low N                |
| SMLW26  | SAM 16 S6-3-6-2-2-1-3-3-3-1-1-3-3   | IAR    | High yielding and Tolerant to low N |
| SMLW27  | SAM 16 S6-3-6-2-2-2-3-1-2-1-1-3-3   | IAR    | High Yielding                       |
| SMLW33  | TZEE-W S6-10-10-1-2-1-1-1-2-1-1-1-3 | IAR    | Susceptible to Low N                |
| SMLW34  | TZEE-W S6-10-10-1-2-1-1-1-2-1-1-2-3 | IAR    | Susceptible to Low N                |
| SMLW37  | SAM 15 S6-3-6-2-2-1-1-2-2-1-1-2-3   | IAR    | High Yielding                       |
| SMLW43  | SAM 16 S6-4-6-2-2-2-2-2-1-1-1-3     | IAR    | Susceptible to Low N                |
| SMLW44  | SAM 16 S6-4-6-2-2-2-2-2-1-1-2-3     | IAR    | Tolerant to low N                   |
| SMLW48  | SAM 20 S6-4-5-1-2-1-1-1-2-1-1-1-3   | IAR    | Susceptible to Low N                |
| SMLW50  | SAM 20 S6-4-5-1-2-1-1-1-2-1-1-3-3   | IAR    | High Yielding                       |
| SMLW51  | TZE-W S6-9-10-1-2-2-2-1-3-1-1-1-3   | IAR    | Tolerant to low N                   |
| SMLW52  | TZE-W S6-9-10-1-2-2-2-1-3-1-1-2-3   | IAR    | High Yielding                       |
| SMLW53  | SAM 18 S6-4-5-1-2-1-2-2-2-1-1-2-2   | IAR    | Susceptible to Low N                |
| SMLW57  | TZE-W S6-3-3-2-2-1-1-1-2-1-1-2-2    | IAR    | Susceptible to Low N                |
| SMLW58  | SAM 16 S6-3-6-2-2-1-3-1-3-1-1-1-2   | IAR    | High Yielding                       |
| SMLW64  | SAM 16 S6-3-6-2-2-1-1-2-2-1-1-2-2   | IAR    | Susceptible to Low N                |
| SMLW69  | TZEE-W S6-2-3-1-2-1-2-1-2-1-1-1-2   | IAR    | Susceptible to Low N                |
| SMLW70  | TZEE-W S6-2-3-1-2-1-2-1-2-1-1-2-2   | IAR    | Susceptible to Low N                |
| SMLW74  | TZE-W S6-2-11-1-2-2-2-2-2-1-1-2-2   | IAR    | High Yielding                       |
| SMLW75  | SAM 16 S6-2-1-1-2-3-3-2-2-2-2-1-2   | IAR    | Susceptible to Low N                |
| SMLW77  | TZE-W S6-1-3-1-2-1-2-1-2-1-1-2-2    | IAR    | Tolerant to low N                   |
| SMLW78  | TZE-W S6-1-3-1-2-1-2-1-2-1-1-1-2    | IAR    | Tolerant to low N                   |
| SMLW84  | SAM 16 S6-2-6-1-2-2-3-2-2-1-1-1-2   | IAR    | Susceptible to Low N                |
| SMLW86  | SAM 15 S6-2-6-2-2-1-2-1-2-1-1-2-2   | IAR    | High yielding and Tolerant to low N |
| SMLW91  | SAM 16 S6-3-6-1-2-3-3-1-3-1-1-1-1   | IAR    | Susceptible to Low N                |
| SMLW93  | SAM 18 S6-3-5-2-2-1-1-1-2-1-1-1-2   | IAR    | Susceptible to Low N                |
| SMLW96  | SAM 15 S6-4-6-1-2-1-1-1-2-1-1-1-1   | IAR    | Tolerant to low N                   |
| SMLW99  | SAM 16 S6-2-6-1-2-3-3-1-2-1-1-1-1   | IAR    | High yielding and Tolerant to low N |
| SMLW100 | TZEE-W S6-2-3-1-2-1-2-1-2-1-1-1-1   | IAR    | Tolerant to low N                   |
| SMLW101 | TZE-W S6-2-3-1-2-1-2-2-2-1-2-1-1    | IAR    | Tolerant to low N                   |
| SMLW102 | SAM 16 S6-1-1-1-2-1-2-1-1-1-1-1-1   | IAR    | Tolerant to low N                   |
| SMLW104 | TZE-W S6-6-10-1-2-2-2-2-2-1-1-1-1   | IAR    | Susceptible to Low N                |
| SMLW105 | SAM 18 S6-1-5-2-2-1-2-2-2-2-2-1-1   | IAR    | Susceptible to Low N                |
| SMLW106 | SAM 15 S6-3-6-1-1-1-1-2-2-1-1-1-1   | IAR    | Susceptible to Low N                |
| SMLW107 | SAM 16 S6-3-6-1-2-3-2-2-2-1-1-1-1   | IAR    | Tolerant to low N                   |
| SMLW108 | SAM 15 S6-2-6-1-2-1-1-2-2-2-2-1-1   | IAR    | Susceptible to Low N                |
| SMLW119 | TZE-W S6-2-3-2-2-1-2-2-2-1-1-2-2    | IAR    | Susceptible to Low N                |
| SMLW120 | TZE-W S6-2-3-2-2-1-2-2-2-1-1-1-2    | IAR    | Susceptible to Low N                |

Continuación de Tabla 1

| LINES       | PEDIGREE                              | Origin       | Main Characteristics                |
|-------------|---------------------------------------|--------------|-------------------------------------|
| SMLW121     | SAM 16 S6-2-6-1-2-1-3-1-2-1-1-1-2     | IAR          | Susceptible to Low N                |
| SMLW122     | SAM 16 S6-2-6-1-2-1-3-1-2-1-1-2-2     | IAR          | Susceptible to Low N                |
| LINES       | PEDIGREE                              | Origin       | Main Characteristics                |
| SMLW127     | SAM 20 S6-5-5-1-2-2-2-1-2-1-3-1-1     | IAR          | Susceptible to Low N                |
| SMLW134     | TZE-W S5-1-12-1-2-1-2-1-1-2-2         | IAR          | Susceptible to Low N                |
| SMLW135     | TZE-W S5-3-3-1-2-2-2-1-2-1-1          | IAR          | Tolerant to low N                   |
| SMLW140     | 2009 DTE S5-8-10-1-2-1-2-1-1-2-2      | IAR          | Susceptible to Low N                |
| SMLW143     | TZE-W S5-3-13-2-2-1-2-1-2-1-2         | IAR          | Tolerant to low N                   |
| SMLW144     | TZE-W S5-5-13-3-6-1-2-1-3-2-2         | IAR          | Tolerant to low N                   |
| SMLW145     | TZE-W S5-5-13-3-3-1-2-1-3-1-2         | IAR          | High yielding and Tolerant to low N |
| SMLW146     | TZE-W S5-3-13-2-2-1-2-2-2-1-2         | IAR          | Tolerant to low N                   |
| SMLW147     | TZE-W S5-1-13-2-2-1-2-1-1-2-2         | IAR          | Tolerant to low N                   |
| SMLW150     | TZE-W S5-3-13-1-2-2-2-2-2-3-3         | IAR          | Susceptible to Low N                |
| SMLW155     | SAM 26 S5-4-6-2-2-2-2-1-1-3-3         | IAR          | Susceptible to Low N                |
| SMLW156     | SAM 26 S5-4-6-2-2-2-2-1-1-2-3         | IAR          | High yielding and Tolerant to low N |
| SMLW157     | SAM 26 S5-4-6-2-2-2-2-1-1-1-3         | IAR          | Tolerant to low N                   |
| SMLW158     | SAM 26 S5-1-12-2-2-2-2-1-1-2-2        | IAR          | High Yielding                       |
| SMLW159     | SAM 26 S5-1-12-2-2-2-2-1-1-1-2        | IAR          | High yielding and Tolerant to low N |
| SMLW160     | SAM 27 S5-2-11-2-3-3-3-3-3-3-3        | IAR          | Susceptible to Low N                |
| SMLW162     | SAM 27 S5-2-11-2-3-3-3-3-3-1-3        | IAR          | Tolerant to low N                   |
| SMLW163     | TZE-W S5-5-13-1-3-1-1-2-2-2-2         | IAR          | Tolerant to low N                   |
| SMLW165     | TZE-W S5-2-13-3-3-2-2-1-1-1-1         | IAR          | Tolerant to low N                   |
| SMLW167     | SAM 27 S5-2-11-2-3-3-3-1-3-1-1        | IAR          | High yielding and Tolerant to low N |
| SMLW169     | TZE-W S5-3-8-2-3-1-2-2-2-1-1          | IAR          | High yielding and Tolerant to low N |
| SMLW183     | SAM 27 S5-7-11-1-2-1-2-1-1-1-1        | IAR          | Tolerant to low N                   |
| Testers     |                                       |              |                                     |
| IITA 1878   | HIx4269-1x1368-7-2-B-B-B-B            | IITA         | High yielding and Tolerant to low N |
| IITA 1876   | P43SRC9FS100-1-1-8-1-B1-13-B1-B-B-B-B | IITA         | Tolerant to low N                   |
| SAM 50M     | IWD-SYN-STR-C3-46-2-B                 | IAR          | High Yielding                       |
| Checks      |                                       |              |                                     |
| SAMMAZ 50   |                                       | IAR          |                                     |
| Oba Super 2 |                                       | Premier Seed |                                     |
| SC 619      |                                       | Seed Co      |                                     |

\*, \*\*, Significant at 0.05 and 0.01 probability levels respectively  
 GY (Grain yield); DP (Days to anthesis); DS (Days to silking); ASI (Anthesis silking interval); PHT (plant height); EHT (Ear height); EA (Ear aspect); STGR (Stay green characteristics).

low N trials, were used as the low-N fields for the study. The nitrogen content of the soils at Mokwa and Zaria were 0.085 % and 0.11 % respectively, both of which were considered to be below the minimum threshold of 0.2 % based on the interpretation by Landon (1991). As a standard practice, every year, complete plant removal was carried out on the low N fields during harvest to prevent the decomposition of organic matter, which could otherwise result in the release of nitrogen into the soil.

### Experimental Design and Field Management

The 237 testcrosses together with three hybrid checks, were evaluated under two different soil

nitrogen conditions (low and optimum) during the 2020 and 2021 planting seasons at Mokwa and Zaria. The trial was laid out in each location using a 15 x 16 alpha ( $\alpha$ ) lattice design in single-row plots of 4 m long. The inter and intra-row spacing were set at 0.75 m and 0.4 m, respectively. Two (2) seeds were planted per hole, supplying was carried out seven days after planting to achieve an optimum population of 66 667 plants for hectare, and the trials were replicated twice in all the test environments. Nitrogen fertilizer was applied at two rates: 30 kg.ha<sup>-1</sup> for low N and 90 kg.ha<sup>-1</sup> for optimum N. For the low N treatment, urea (30 kg.ha<sup>-1</sup>) was applied evenly in two split doses at two and five weeks after sowing (WAS), along with muriate of potash (MOP) and

single superphosphate (SSP), both at a rate of 60 kg.ha<sup>-1</sup> at 2 WAS. Optimum N treatment involved application of compound fertilizer NPK 15:15:15 at the rate of nitrogen (60 kg.ha<sup>-1</sup>), phosphorus (60 kg.ha<sup>-1</sup>) and potassium (60 kg.ha<sup>-1</sup>) at 2 WAS, followed by an additional top dressing of 30 kg ha<sup>-1</sup> of urea at 4 WAS. Weed management was carried out using appropriate herbicides and supplemented with hoe weeding when necessary.

### Data Collection and Analysis

Agronomic data were recorded on each line on plot basis under both low and optimum-N condition. Flowering traits were recorded in days and growth traits were measured based on the average measurements of five randomly selected plants in a plot. Aspect ratings were assessed visually on a phenotypic scale ranging from 1 to 10, with higher values indicating poor phenotypic appeal. Leaf senescence, characterized by the stay green trait, was assessed by visually evaluating the condition of the leaves, including color, overall appearance, and visible signs of aging 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 different stages of senescence, with 1 indicating a healthy, green leaf and higher numbers representing decreasing degrees of senescence. For low-N trial, the harvested ears of each inbred line per plot were shelled, weighed, and recorded in grams before converting them to kilograms. Moisture content was also determined from the shelled grain. The values were then used to estimate the grain yield of the line in kilograms per hectare, adjusted to 15% moisture content. Under optimum-N conditions, the harvested ears per plot of each inbred line were weighed and recorded in kilograms. A sample size was taken among the harvested ears, which were then shelled to determine their moisture content. The ear weight was later converted to kilograms per hectare using a shelling coefficient of 80 % and adjusted to 15 % moisture content.

The estimates of the general combining ability (GCA) of the line and tester, as well as the

specific combining ability (SCA) between the line and tester, were derived using the AGD-R software version 4.1 (Rodríguez et al., 2015).

The statistical model used for line x tester analysis in each nitrogen (N) environment is as follows:

$$Y_{ijke} = \mu + a_e + R_{ke} + B_{mke} + g_i + g_j + S_{ij} + (ag)_{ie} + (ag)_{je} + (as)_{ije} + \varepsilon_{eijkm}$$

where,  $Y_{ijke}$  = the response of the of the  $i \times j^{\text{th}}$  progeny of the  $K^{\text{th}}$  block of the  $m^{\text{th}}$  replication of the  $e^{\text{th}}$  environment;  $\mu$  = the experimental mean;  $a_e$  = the main effect of Environment;  $R_{kl}$  = the effect of replication within the environment;  $B_{mke}$  = the block effect within each replication within each environment;  $g_i$  = general combining ability for the  $i^{\text{th}}$  parental line;  $g_j$  = general combining ability effect of  $j^{\text{th}}$  tester;  $S_{ij}$  = the specific combining ability (SCA) for the  $ij^{\text{th}}$   $F_1$  hybrid;  $(ag)_{ie}$  = the interaction effect of  $i^{\text{th}}$  line and  $e^{\text{th}}$  environment;  $(ag)_{je}$  = the interaction effect of  $j^{\text{th}}$  tester and  $e^{\text{th}}$  environment;  $(as)_{ije}$  = the interaction effect of the  $i^{\text{th}}$  hybrid (SCA) and the  $e^{\text{th}}$  environment;  $\varepsilon_{eijkm}$  = the residual effect.

### Heterotic Grouping of the Inbred Lines

Heterotic grouping of the inbred lines under each nitrogen environment was determined using the GCA of multiple traits approach (HGCAMT) (Badu-Apraku et al., 2013). This approach involved utilizing the standardized values of the GCA effects for all traits with significant mean squares under the respective N conditions. Eight traits were considered under both N conditions, including grain yield, days to anthesis, days to silking, plant height, ear height, and ear aspect. Stay green characteristics were additionally considered under low-N, while plant aspect was considered under optimum-N conditions. For each nitrogen condition, the standardized GCA values of the selected traits were converted to Euclidean distance estimates using PROC DISTANCE in SAS version 9.2. This distance metric quantified the genetic similarity between pairs of inbred lines based on their performance across all considered traits. Subsequently, these genetic similarity coefficients were subjected

to Ward's minimum variance cluster analysis. This algorithm organized the inbred lines into a hierarchical tree structure, known as a dendrogram, providing a visual representation of the relationships between the inbred lines based on their genetic similarity.

## Results

### *Estimation of Line and Tester GCA effects for Grain Yield and other Traits under low -N conditions*

The GCA effects of the inbred lines for grain yield and other agronomic traits under low N conditions are presented in [Table 2](#). Out of the 79 inbred lines, only two (SMLW74 and SMLW146) exhibited positive and significant GCA effects for grain yield, while two others (SMLW140 and SMLW143) exhibited negative and significant GCA effects. Notably, inbred line SMLW146 displayed the maximum GCA effect for grain yield ( $321.57 \text{ kg}\cdot\text{ha}^{-1}$ ), whereas SMLW143 exhibited the lowest ( $-226.15 \text{ kg}\cdot\text{ha}^{-1}$ ). Among the three testers, only IITA1876 demonstrated a positive and significant GCA effect for grain yield, while tester SAM50M exhibited a negative and significant GCA effect. Regarding days to anthesis, twenty-five lines showed significant negative GCA effects, while twenty-three lines displayed positive significant GCA effects. Inbred lines SMLW135, SMLW134, and SMLW122 exhibited the lowest significant negative GCA effects, whereas SMLW25 and SMLW3 had the highest positive significant GCA effects. None of the testers displayed significant GCA effects for days to anthesis. A similar trend was observed for days to silking, with twenty-five lines displaying significant negative GCA effects and twenty-two lines showing positive GCA effects. SMLW134, SMLW122, and SMLW135 recorded the lowest significant negative GCA effects for days to silking. Again, none of the testers exhibited significant GCA effects for this trait.

Regarding plant height, eight lines displayed significant positive GCA effects, while ten lines showed significant negative GCA effects. SMLW104 and SMLW48 exhibited the highest positive significant GCA effects, while SMLW155 and SMLW156 had the

lowest negative significant GCA effects. Tester IITA1876 exhibited a significant positive GCA effect for plant height, whereas SAM50M had a significant negative GCA effect for this trait. No significant differences were observed among the lines and testers for ear height. However, for ear aspect, only two inbred lines (SMLW48 and SMLW44) exhibited significant negative GCA effects, while tester SAM50M also displayed significant negative GCA effects. Regarding the stay green characteristic, five lines (SMLW7, SMLW9, SMLW57, SMLW43, and SMLW146) and two testers (IITA1876 and SAM50M) displayed significant negative GCA effects

### *Estimation of Line and Tester GCA effects for Grain Yield and other Traits under optimum-N conditions*

Four inbred lines (SMLW146, SMLW147, SMLW150, and SMLW155) and one tester (IITA1876) exhibited significant and highly positive GCA effects for grain yield, while only one line (SMLW17) and tester SAM50M had significant negative GCA effects ([Table 3](#)). Regarding days to anthesis, twenty-one lines and one tester (IITA1878) exhibited significant negative GCA effects, with the highest negative GCA effect observed in SMLW3, followed by SMLW25 and SMLW19. For days to silking, sixteen inbred lines exhibited significant negative GCA effects, while nine lines had significant positive GCA effects. Notably, these sixteen inbred lines also displayed significant negative GCA effects for days to anthesis. Line SMLW120 had the highest GCA effect for days to silking, while SMLW3 had the lowest. Only one tester (IITA1878) exhibited significant negative GCA effects for days to silking, while the others had significant positive effects for the trait.

Regarding plant height, six lines (SMLW4, SMLW25, SMLW22, SMLW167, SMLW165, and SMLW20) and one tester (IITA1876) exhibited significant positive GCA effects, whereas inbred lines SMLW160 and SMLW163, along with tester IITA1878, had significant negative GCA effects for the trait. Similarly, for ear height, nine lines and one tester (IITA1876) showed significant positive GCA effects, while five inbred lines (SMLW5, SMLW43, SMLW44,

**Table 2: Estimates of Line and Tester General Combining Ability effects for Grain Yield and other Traits under Low-N Condition (Mokwa and Zaria) in 2020 and 2021**

| Lines      | GY<br>(kg ha <sup>-1</sup> ) | DP<br>(days) | DS<br>(days) | ASI<br>(days) | PHT<br>(cm) | EHT<br>(cm) | EA<br>(1-9) | STGR    |
|------------|------------------------------|--------------|--------------|---------------|-------------|-------------|-------------|---------|
| SMLW - 3   | 17.21                        | 4.58**       | 4.74**       | 0.21          | 3.30        | 0.92        | 0.35        | -0.15   |
| SMLW - 4   | 7.05                         | -2.98**      | -2.73**      | 0.22          | 13.24*      | -0.67       | 0.44        | 0.14    |
| SMLW - 5   | -94.56                       | -1.88**      | -1.66**      | 0.21          | 3.00        | -0.31       | 0.42        | -0.57   |
| SMLW - 6   | -138.71                      | 0.26         | -0.33        | -0.59*        | -8.54       | -0.21       | 0.28        | -0.39   |
| SMLW - 7   | 143.11                       | -0.59        | -0.71        | -0.12         | -4.46       | -2.84       | 0.35        | -1.09** |
| SMLW - 9   | 41.87                        | -0.04        | 0.03         | 0.06          | -7.61       | -2.77       | 0.32        | -1.02*  |
| SMLW - 10  | 80.20                        | 2.36**       | 0.87         | -1.47**       | 3.38        | -3.37       | -0.12       | -0.44   |
| SMLW - 11  | -45.13                       | 1.35*        | 0.60         | -0.74**       | -0.77       | 0.39        | -0.19       | -0.13   |
| SMLW - 14  | -164.88                      | -3.17**      | -2.92**      | 0.22          | 5.23        | 0.74        | -0.40       | 0.01    |
| SMLW - 16  | 23.24                        | 1.73**       | 2.85**       | 1.16**        | -13.98*     | 0.91        | -0.34       | -0.08   |
| SMLW - 17  | -118.45                      | 2.02*        | 3.15**       | 1.16**        | 0.17        | 2.25        | -0.31       | 0.43    |
| SMLW - 19  | 10.49                        | 1.77*        | 0.59         | -1.16**       | 8.94        | 1.75        | -0.39       | 0.01    |
| SMLW - 20  | 40.98                        | 2.24**       | 0.42         | -1.82**       | 3.47        | -0.17       | -0.60       | -0.02   |
| SMLW - 21  | -58.10                       | 4.39**       | 2.73**       | -1.64**       | 5.37        | 0.09        | -0.49       | 0.09    |
| SMLW - 22  | 66.07                        | 4.40**       | 3.25**       | -1.12**       | 8.00        | 2.81        | -0.52       | -0.03   |
| SMLW - 23  | 57.03                        | 2.60**       | 1.84*        | -0.73**       | -0.59       | 1.07        | -0.60       | -0.22   |
| SMLW - 24  | -146.73                      | 3.77**       | 3.93**       | 0.21          | -5.59       | 0.80        | -0.50       | 0.44    |
| SMLW - 25  | -40.00                       | 5.12**       | 5.29**       | 0.22          | 8.79        | 0.45        | -0.60       | 0.63    |
| SMLW - 26  | 154.38                       | 4.11**       | 4.27**       | 0.21          | 12.73*      | 1.40        | -0.60       | -0.34   |
| SMLW - 27  | -32.10                       | 3.91**       | 4.09**       | 0.21          | 1.85        | 2.64        | -0.60       | -0.33   |
| SMLW - 33  | -42.73                       | 0.31         | 0.53         | 0.22          | 6.84        | 0.44        | -0.58       | -0.07   |
| SMLW - 34  | -34.51                       | -2.29**      | -2.04*       | 0.22          | 5.00        | 1.62        | -0.55       | -0.34   |
| SMLW - 37  | 13.43                        | -2.35**      | -2.13*       | 0.20          | 1.61        | 0.78        | -0.54       | -0.45   |
| SMLW - 43  | -78.87                       | -3.42**      | -3.16**      | 0.21          | 13.47*      | 1.70        | -0.66       | -0.78*  |
| SMLW - 44  | -25.26                       | -1.77*       | -1.54*       | 0.21          | 15.10**     | 2.34        | -0.76       | -0.31   |
| SMLW - 48  | 67.73                        | -1.94*       | -1.71*       | 0.21          | 16.81**     | 3.23        | -0.76       | -0.30   |
| SMLW - 50  | -24.66                       | 0.20         | 0.42         | 0.22          | 15.69**     | 2.06        | -0.45       | -0.67   |
| SMLW - 51  | 4.12                         | 0.56         | 0.76         | 0.20          | 5.37        | 1.27        | -0.32       | -0.49   |
| SMLW - 52  | -2.41                        | 0.92         | 1.11         | 0.20          | 10.16       | 1.82        | 0.36        | -0.67   |
| SMLW - 53  | -75.81                       | 0.79         | 0.61         | -0.17         | 4.81        | 2.38        | 0.42        | -0.39   |
| SMLW - 57  | -59.62                       | 0.39         | 0.03         | -0.35         | -7.36       | 1.95        | 0.40        | -0.86*  |
| SMLW - 58  | -169.26                      | 0.50         | -0.11        | -0.62*        | -5.05       | 3.26        | 0.45        | 0.02    |
| SMLW - 64  | -171.42                      | 0.90         | 0.49         | -0.41*        | -8.41       | -1.10       | 0.34        | 0.44    |
| SMLW - 69  | 184.51                       | 0.44         | -0.15        | -0.59*        | -2.10       | -2.31       | 0.38        | -0.48   |
| SMLW - 70  | 171.25                       | -0.42        | -0.26        | 0.16          | -7.61       | -2.41       | 0.31        | -0.66   |
| SMLW - 74  | 238.07*                      | 0.35         | 0.56         | 0.20          | -10.47*     | -2.40       | 0.33        | -0.50   |
| SMLW - 75  | 85.54                        | -0.13**      | -0.14        | -0.02         | -6.12       | -3.17       | 0.29        | -0.11   |
| SMLW - 77  | 94.45                        | 2.74         | 1.76*        | -0.96*        | 8.13        | -1.96       | 0.27        | -0.31   |
| SMLW - 78  | -19.02                       | 0.14         | 0.06         | -0.07         | 5.73        | -1.27       | 0.33        | 0.77*   |
| SMLW - 84  | -8.44                        | -0.51        | -0.26        | 0.25          | -3.43       | -1.43       | 0.40        | 0.09    |
| SMLW - 86  | -64.04                       | -0.28        | 0.05         | 0.32          | -3.11       | 0.06        | 0.29        | 0.53    |
| SMLW - 91  | -61.28                       | -0.53        | -0.25        | 0.27          | -8.02       | -1.75       | 0.55        | 0.90*   |
| SMLW - 93  | -3.53                        | -0.09        | -0.07        | 0.02          | -5.93       | -1.91       | 0.88*       | 0.44    |
| SMLW - 96  | -30.40                       | -0.40        | -0.39        | 0.01          | -9.83       | -3.58       | 0.90*       | 0.45    |
| SMLW - 99  | -27.40                       | -0.75        | -0.84        | -0.10         | -12.48*     | -2.77       | 0.86        | 0.45    |
| SMLW - 100 | 136.56                       | 2.64**       | 1.50*        | -1.13**       | 4.23        | -0.34       | 0.99*       | -0.03   |
| SMLW - 101 | 78.73                        | 2.11**       | 0.86         | -1.24**       | 6.57        | -1.78       | 0.96*       | 0.54    |
| SMLW - 102 | 39.03                        | 2.21**       | 1.48*        | -0.72**       | 8.86        | 0.17        | 0.90*       | 0.62    |
| SMLW - 104 | 89.51                        | 2.68**       | 1.92**       | -0.74**       | 17.18**     | -0.72       | 0.64        | 0.71*   |
| SMLW - 105 | 87.39                        | 1.64*        | 0.91         | -0.73**       | 8.24        | -1.26       | 0.48        | 0.32    |
| SMLW - 106 | 169.15                       | -1.25*       | -1.95**      | -0.72**       | 13.66*      | 0.39        | 0.52        | -0.04   |
| SMLW - 107 | 10.89                        | -1.39*       | -2.09**      | -0.73**       | 6.41        | -1.32       | 0.59        | 0.95*   |
| SMLW - 108 | 3.31                         | -1.90*       | -2.60**      | -0.73**       | 1.76        | -1.46       | 0.60        | 0.59    |



Continuación de Tabla 2

| Lines             | GY<br>(kg ha <sup>-1</sup> ) | DP<br>(days) | DS<br>(days) | ASI<br>(days) | PHT<br>(cm) | EHT<br>(cm) | EA<br>(1-9) | STGR        |
|-------------------|------------------------------|--------------|--------------|---------------|-------------|-------------|-------------|-------------|
| SMLW - 119        | 6.25                         | 0.64         | -0.07        | -0.72**       | -1.70       | -1.04       | 0.76        | 0.43        |
| SMLW - 120        | 20.65                        | 0.26         | -0.47        | -0.73**       | 8.61        | 1.15        | 0.62        | 0.06        |
| SMLW - 121        | 80.37                        | -1.76*       | -2.46**      | -0.73**       | 11.23*      | 0.44        | 0.44        | 0.19        |
| SMLW - 122        | -84.59                       | -4.15**      | -4.82**      | -0.72**       | 4.30        | -0.93       | 0.65        | 0.07        |
| SMLW - 127        | -130.14                      | -2.87**      | -3.56**      | -0.72**       | 7.59        | -1.99       | -0.21       | 0.08        |
| SMLW - 134        | -170.35                      | -4.22**      | -4.88**      | -0.73**       | 2.71        | -0.33       | -0.40       | 0.14        |
| SMLW - 135        | -126.80                      | -4.25**      | -3.99**      | 0.22          | 4.77        | -1.40       | -0.36       | 0.42        |
| SMLW - 140        | -210.88*                     | -2.96**      | -2.72**      | 0.21          | -1.55       | -0.71       | -0.31       | 0.41        |
| SMLW - 143        | -226.15*                     | -2.77**      | -2.53**      | 0.21          | -14.73**    | 0.03        | -0.31       | 0.60        |
| SMLW - 144        | 123.11                       | -3.06**      | -2.82**      | 0.22          | -5.39       | -0.05       | -0.41       | -0.26       |
| SMLW - 145        | 164.52                       | -3.49**      | -2.52**      | 0.95**        | -7.36       | 0.04        | -0.30       | -0.20       |
| SMLW - 146        | 321.57**                     | -3.48**      | -2.31**      | 1.15**        | -7.01       | -0.78       | -0.38       | -0.78*      |
| SMLW - 147        | 209.48                       | -3.37**      | -2.18**      | 1.16**        | -1.18       | 1.27        | -0.40       | -0.59       |
| SMLW - 150        | -29.75                       | -2.74**      | -1.56*       | 1.16**        | -14.88**    | 1.41        | -0.38       | 0.64        |
| SMLW - 155        | -114.43                      | -3.55**      | -2.38**      | 1.15**        | -19.75**    | -2.07       | -0.35       | 0.04        |
| SMLW - 156        | 6.11                         | -2.26**      | -1.10        | 1.15**        | -18.00**    | -2.09       | -0.27       | -0.07       |
| SMLW - 157        | -15.70                       | -3.54**      | -2.36**      | 1.16**        | -13.64*     | 0.40        | -0.43       | 0.04        |
| SMLW - 158        | -12.46                       | -1.51*       | -0.36        | 1.14**        | -6.39       | 0.52        | -0.34       | 0.13        |
| SMLW - 160        | -15.38                       | 1.12         | 2.22**       | 1.14**        | -15.82**    | 2.21        | -0.30       | -0.08       |
| SMLW - 162        | 65.88                        | 2.07**       | 3.19**       | 1.15**        | 0.61        | 2.63        | -0.34       | 0.04        |
| SMLW - 163        | -10.47                       | 1.30*        | 2.41**       | 1.14**        | -8.17       | 2.16        | -0.29       | 0.03        |
| SMLW - 165        | 42.40                        | 0.32         | 1.47*        | 1.16**        | -2.80       | 2.09        | -0.38       | 0.03        |
| SMLW - 167        | -97.35                       | 2.04         | 3.15**       | 1.14**        | -6.36       | 3.45        | -0.36       | 0.50        |
| SMLW - 169        | -173.86                      | 1.42*        | 2.54**       | 1.15**        | -15.47**    | 0.43        | -0.24       | 0.14        |
| SMLW - 183        | -42.74                       | 2.11**       | 2.00**       | -0.08         | -0.36       | -1.90       | -0.25       | 0.59        |
| SMLW - 159        | 42.75                        | 2.69**       | 2.51**       | -0.15         | -0.92       | -1.33       | -0.22       | 0.09        |
| <b>Testers</b>    |                              |              |              |               |             |             |             |             |
| IITA 1876         | 401.78**                     | 0.05         | 0.06         | 0.00          | 27.61**     | -1.01       | 0.84**      | -0.93**     |
| IITA 1878         | -84.70                       | -0.01        | 0.01         | 0.00          | 3.32        | 0.51        | 0.06        | 1.99**      |
| SAM 50M           | -317.1**                     | -0.04        | -0.06        | 0.00          | -30.92**    | 0.50        | -0.90**     | -1.06**     |
| <b>SE Testers</b> | <b>46.57</b>                 | <b>0.07</b>  | <b>0.08</b>  | <b>0.01</b>   | <b>2.05</b> | <b>0.77</b> | <b>0.11</b> | <b>0.10</b> |
| <b>SE Lines</b>   | <b>111.93</b>                | <b>0.61</b>  | <b>0.62</b>  | <b>0.19</b>   | <b>5.18</b> | <b>3.03</b> | <b>0.44</b> | <b>0.36</b> |

\*, \*\*, Significant at 0.05 and 0.01 probability levels respectively  
 GY (Grain yield); DP (Days to anthesis); DS (Days to silking); ASI (Anthesis silking interval); PHT (plant height); EHT (Ear height); PA (Plant aspect); EA (Ear aspect)

SMLW50, SMLW69) and tester IITA1878 displayed significant negative GCA effects for the trait. No significant differences were observed among the lines for plant aspect and ear aspect. However, tester IITA1878 exhibited significant negative GCA effects for both plant and ear aspect, while IITA1876 showed significant positive GCA effects for these traits.

***Heterotic grouping of the inbred lines under low- and optimum-N condition***

The 79 inbred lines were classified into four primary clusters (1, 2, 3, and 4) upon truncating the dendrogram at a 38 % genetic similarity level in low-N environments (Figure 1). Cluster 1 consisted of fifteen inbred lines with a 66 %

genetic similarity coefficient, while Cluster 2 encompassed twenty inbred lines with a 44 % genetic similarity coefficient. Cluster 3 was comprised of twenty-six inbred lines with a 69 % genetic similarity coefficient, and Cluster 4 contained eighteen inbred lines with a 38 % genetic similarity coefficient. Based on the *per se* performance of inbred lines within each cluster under low-N conditions (Table 4), lines in Cluster 4 exhibited the highest mean grain yield, followed by inbred lines in Cluster 1, whereas Cluster 3 inbred lines displayed the lowest mean yield. Concerning flowering traits, lines in Clusters 1 and 4 exhibited similar flowering times, with Cluster 2 lines showing delayed anthesis and silking. Inbred lines in Cluster 4 had the shortest

**Table 3: Estimates of Line and Tester General Combining Ability effects for Grain Yield and other Traits under Optimum-N Condition (Mokwa and Zaria) in 2020 and 2021**

| Lines      | GY<br>(kg ha <sup>-1</sup> ) | DP<br>(days) | DS<br>(days) | ASI<br>(days) | PHT<br>(cm) | EHT<br>(cm) | PA<br>(1-9) | EA<br>(1-9) |
|------------|------------------------------|--------------|--------------|---------------|-------------|-------------|-------------|-------------|
| SMLW - 3   | 68.17                        | -3.71**      | -2.81**      | 0.37          | 4.94        | -2.02       | -0.01       | 0.01        |
| SMLW - 4   | -32.70                       | -2.13**      | -1.63*       | 0.24          | 10.96*      | 7.58*       | -0.01       | -0.01       |
| SMLW - 5   | -91.28                       | -1.51*       | -0.93        | 0.31          | 1.96        | -9.07**     | -0.01       | 0.02        |
| SMLW - 6   | -108.32                      | -0.28        | -0.07        | 0.17          | -3.78       | -5.20       | 0.00        | -0.01       |
| SMLW - 7   | -60.58                       | -0.02        | 0.30         | 0.34          | -4.47       | 0.16        | -0.01       | 0.00        |
| SMLW - 9   | -112.68                      | 1.26*        | 1.67*        | 0.67*         | 6.39        | 4.20        | -0.02       | -0.01       |
| SMLW - 10  | 109.94                       | 1.15         | 0.46         | -0.52         | -6.51       | 3.51        | 0.00        | -0.01       |
| SMLW - 11  | -147.23                      | 0.23         | -0.39        | -0.58*        | 5.63        | 6.69*       | 0.00        | -0.01       |
| SMLW - 14  | 1.36                         | 1.52*        | 0.96         | -0.35         | -3.71       | -5.99       | 0.01        | 0.01        |
| SMLW - 16  | -52.56                       | 1.88**       | 1.12         | -0.48         | -2.50       | 5.82        | 0.01        | -0.01       |
| SMLW - 17  | -378.69*                     | -0.56        | -0.04        | 0.44          | 7.50        | -3.02       | -0.01       | 0.01        |
| SMLW - 19  | -74.30                       | -3.09**      | -2.18**      | 0.47          | 7.32        | 0.14        | 0.01        | 0.00        |
| SMLW - 20  | -235.80                      | -1.18        | -0.41        | 0.60*         | 12.93*      | 8.98**      | 0.01        | -0.01       |
| SMLW - 21  | -77.90                       | -2.81**      | -1.80**      | 0.34          | 6.53        | -2.00       | 0.00        | 0.01        |
| SMLW - 22  | -232.04                      | -2.78**      | -2.26**      | 0.17          | 11.87*      | -1.81       | -0.01       | 0.02        |
| SMLW - 23  | -26.42                       | -2.34**      | -1.52*       | 0.47          | 8.78        | -3.89       | 0.02        | 0.01        |
| SMLW - 24  | -89.59                       | -2.41**      | -1.83**      | 0.27          | 9.36        | -3.70       | 0.01        | 0.01        |
| SMLW - 25  | -43.45                       | -3.21**      | -2.16**      | 0.57          | 11.58       | -0.72       | 0.01        | 0.01        |
| SMLW - 26  | -204.13                      | -1.74**      | -1.23        | 0.31          | 9.10        | -1.06       | -0.01       | 0.00        |
| SMLW - 27  | 85.98                        | -2.45**      | -1.38*       | 0.70*         | 9.38        | -0.08       | 0.00        | 0.00        |
| SMLW - 33  | -125.95                      | -2.41**      | -1.34*       | 0.70*         | -1.24       | -3.61       | 0.02        | -0.01       |
| SMLW - 34  | -59.35                       | -2.96**      | -1.79**      | 0.77*         | -5.00       | -3.62       | 0.02        | 0.00        |
| SMLW - 37  | -36.68                       | -1.74**      | -1.30*       | 0.24          | 1.98        | -0.96       | 0.00        | -0.01       |
| SMLW - 43  | -196.46                      | -2.93**      | -1.86**      | 0.70*         | 4.18        | -7.11*      | 0.02        | 0.01        |
| SMLW - 44  | -267.29                      | -2.18**      | -1.33*       | 0.54          | -4.80       | -9.40**     | 0.00        | 0.01        |
| SMLW - 48  | -59.12                       | -1.59*       | -1.01        | 0.41          | 2.72        | -5.94       | 0.00        | 0.01        |
| SMLW - 50  | -153.39                      | -2.02**      | -0.86        | 0.90**        | -0.15       | -7.15*      | -0.01       | 0.01        |
| SMLW - 51  | -102.92                      | -1.92**      | -1.01        | 0.67*         | -4.36       | -5.53       | 0.02        | 0.00        |
| SMLW - 52  | -206.95                      | -2.03**      | -1.34*       | 0.41          | 2.61        | -6.08       | -0.01       | 0.00        |
| SMLW - 53  | -18.28                       | -1.17        | -0.67        | 0.31          | -2.29       | -5.82       | -0.01       | 0.01        |
| SMLW - 57  | 45.26                        | -0.46        | -0.21        | 0.21          | -1.06       | -5.75       | 0.00        | -0.01       |
| SMLW - 58  | -56.16                       | -0.53        | -0.23        | 0.24          | -3.20       | -5.62       | 0.01        | -0.01       |
| SMLW - 64  | -92.17                       | 0.45         | 0.62         | 0.24          | 0.94        | -0.57       | -0.01       | 0.01        |
| SMLW - 69  | 74.99                        | -0.45        | -0.07        | 0.31          | -7.62       | -6.97*      | 0.01        | 0.00        |
| SMLW - 70  | -177.94                      | 0.56         | 0.84         | 0.37          | -1.75       | 1.95        | 0.00        | 0.01        |
| SMLW - 74  | -158.33                      | -0.27        | 0.42         | 0.70*         | -2.48       | 1.41        | 0.01        | 0.02        |
| SMLW - 75  | -8.93                        | -0.31        | 0.48         | 0.77*         | -0.51       | 4.56        | -0.01       | 0.00        |
| SMLW - 77  | -112.08                      | 2.06**       | 1.19         | -0.58*        | -1.07       | 3.95        | 0.00        | 0.01        |
| SMLW - 78  | -97.88                       | 0.56         | 1.08         | 0.57          | 7.44        | 6.49        | -0.01       | 0.00        |
| SMLW - 84  | 267.86                       | -0.50        | 0.05         | 0.50          | 5.20        | 2.26        | -0.02       | -0.02       |
| SMLW - 86  | 143.28                       | -0.71        | -0.04        | 0.64*         | -3.52       | 2.42        | 0.02        | 0.00        |
| SMLW - 91  | -77.51                       | -0.75        | -0.38        | 0.27          | -3.35       | 3.83        | 0.01        | 0.00        |
| SMLW - 93  | -167.18                      | -0.54        | 0.20         | 0.64*         | -0.58       | -2.88       | 0.01        | -0.03       |
| SMLW - 96  | -58.90                       | -0.59        | 0.21         | 0.77*         | -3.48       | 0.35        | 0.02        | 0.00        |
| SMLW - 99  | 155.55                       | 0.09         | 0.69         | 0.67*         | -1.21       | -5.35       | -0.01       | -0.01       |
| SMLW - 100 | -10.15                       | 1.48*        | 0.83         | -0.48         | -6.76       | -0.53       | -0.01       | -0.01       |
| SMLW - 101 | -137.02                      | 1.04         | 0.38         | -0.48         | -2.40       | 8.07*       | 0.00        | 0.01        |
| SMLW - 102 | -209.60                      | 2.12**       | 1.52*        | -0.29         | -3.74       | 8.03*       | 0.01        | 0.04        |
| SMLW - 104 | -54.95                       | 1.82**       | 1.29*        | -0.29         | -4.62       | 7.50*       | 0.01        | 0.00        |
| SMLW - 105 | 175.30                       | 1.81**       | 0.87         | -0.71*        | -1.94       | 11.12**     | 0.00        | -0.02       |
| SMLW - 106 | -45.47                       | 1.08         | 0.58         | -0.42         | -2.58       | 7.12*       | 0.00        | 0.00        |
| SMLW - 107 | -171.98                      | 1.35*        | 0.59         | -0.61*        | 3.03        | 3.35        | 0.01        | 0.01        |
| SMLW - 108 | -7.38                        | 2.32**       | 1.63*        | -0.38         | 2.38        | 7.28*       | 0.00        | 0.01        |
| SMLW - 119 | -72.23                       | 0.85         | 0.28         | -0.45         | -3.24       | -2.05       | 0.00        | 0.00        |

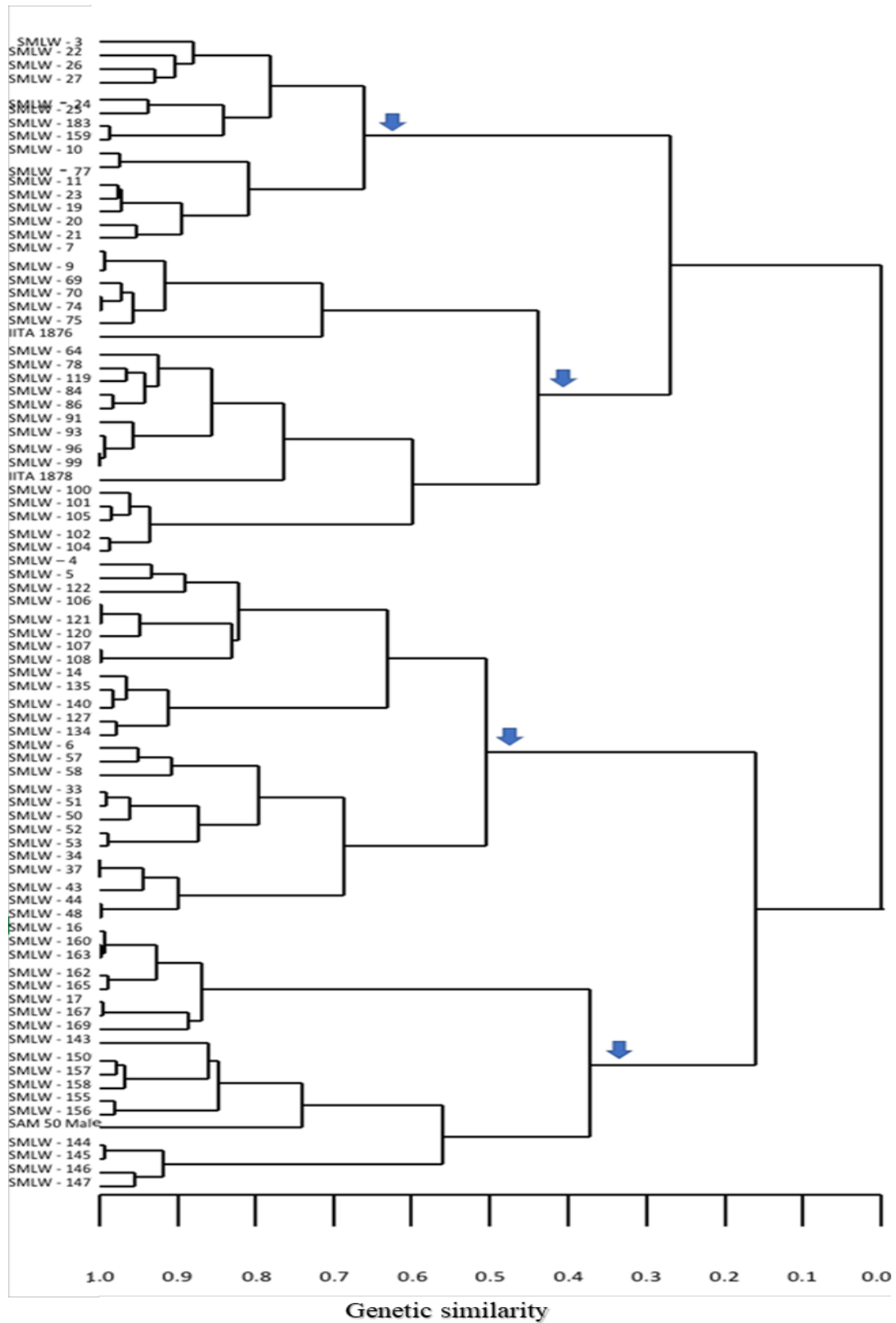
Continuación de Tabla 3

| Lines             | GY<br>(kg ha <sup>-1</sup> ) | DP<br>(days) | DS<br>(days) | ASI<br>(days) | PHT<br>(cm) | EHT<br>(cm) | PA<br>(1-9) | EA<br>(1-9) |
|-------------------|------------------------------|--------------|--------------|---------------|-------------|-------------|-------------|-------------|
| SMLW - 120        | 122.32                       | 2.41**       | 1.71*        | -0.35         | 1.74        | 0.17        | 0.01        | 0.00        |
| SMLW - 121        | 110.58                       | 2.10**       | 1.35*        | -0.52         | 2.27        | -2.51       | 0.01        | -0.01       |
| SMLW - 122        | -234.55                      | 2.21**       | 1.20         | -0.71*        | 0.31        | -0.14       | 0.01        | 0.01        |
| SMLW - 127        | 174.46                       | 1.71*        | 1.03         | -0.38         | -7.58       | -0.64       | 0.01        | -0.01       |
| SMLW - 134        | 97.32                        | 0.96         | 0.66         | -0.19         | -6.37       | 0.57        | -0.01       | 0.00        |
| SMLW - 135        | -130.03                      | 1.89**       | 1.37*        | -0.25         | -4.93       | -2.57       | 0.01        | 0.00        |
| SMLW - 140        | -258.69                      | 1.77**       | 0.98         | -0.55         | 0.45        | -1.52       | 0.01        | 0.03        |
| SMLW - 143        | -0.10                        | 2.23**       | 1.61*        | -0.35         | -3.18       | -5.81       | 0.01        | 0.01        |
| SMLW - 144        | 128.02                       | 2.33**       | 1.23         | -0.71*        | 0.30        | 1.69        | -0.01       | -0.01       |
| SMLW - 145        | -69.18                       | 1.39*        | 0.26         | -0.94**       | -6.82       | 0.91        | -0.01       | 0.00        |
| SMLW - 146        | 759.71**                     | 0.87         | 0.34         | -0.38         | -2.39       | 3.97        | -0.01       | 0.00        |
| SMLW - 147        | 879.75**                     | 0.95         | 0.40         | -0.45         | -6.52       | -0.36       | 0.00        | -0.02       |
| SMLW - 150        | 733.18**                     | 1.46*        | 0.93         | -0.32         | -5.66       | 3.90        | 0.00        | 0.00        |
| SMLW - 155        | 776.47**                     | 1.78**       | 1.38*        | -0.12         | -10.85      | -2.51       | 0.00        | -0.01       |
| SMLW - 156        | 194.00                       | 1.15         | 0.25         | -0.75*        | -5.02       | -2.44       | -0.01       | -0.01       |
| SMLW - 157        | -58.96                       | 1.68*        | 1.05         | -0.38         | -6.52       | 3.73        | -0.01       | -0.01       |
| SMLW - 158        | -68.18                       | 1.26*        | 0.43         | -0.65*        | -6.20       | 3.57        | -0.01       | -0.03       |
| SMLW - 160        | 32.82                        | 1.77**       | 0.66         | -0.91**       | -11.54*     | -3.90       | 0.00        | 0.00        |
| SMLW - 162        | 143.06                       | 1.14         | 0.20         | -0.81**       | -7.47       | 0.13        | -0.01       | -0.01       |
| SMLW - 163        | -6.60                        | 1.12         | 0.13         | -0.81**       | -12.11*     | -1.89       | 0.01        | 0.03        |
| SMLW - 165        | 360.34                       | 1.03         | -0.01        | -0.94**       | 12.46*      | 2.12        | -0.01       | 0.00        |
| SMLW - 167        | -25.80                       | 0.94         | 0.13         | -0.68*        | 12.11*      | -0.93       | 0.00        | -0.02       |
| SMLW - 169        | 168.22                       | -1.31*       | -1.40*       | -0.25         | 7.73        | -0.81       | 0.00        | -0.01       |
| SMLW - 183        | 2.49                         | -0.86        | -0.03        | 0.70*         | 9.65        | 1.23        | -0.01       | 0.00        |
| SMLW - 159        | -48.39                       | 0.65         | -0.01        | -0.61*        | -4.62       | 5.88        | 0.00        | -0.02       |
| <b>TESTERS</b>    |                              |              |              |               |             |             |             |             |
| IITA 1876         | 856.63**                     | 1.40**       | 1.07**       | -0.31*        | 51.87**     | 21.84**     | 0.49**      | 0.47**      |
| IITA 1878         | -239.22                      | -1.59**      | -1.66**      | -0.08         | -43.5**     | -18.63*     | -0.49**     | -0.5**      |
| SAM 50M           | -617.41*                     | 0.18         | 0.58*        | 0.39*         | -8.41       | -3.21       | 0.01        | 0.04        |
| <b>SE Testers</b> | <b>123.91</b>                | <b>0.25</b>  | <b>0.21</b>  | <b>0.09</b>   | <b>5.56</b> | <b>3.64</b> | <b>0.03</b> | <b>0.04</b> |
| <b>SE Lines</b>   | <b>196.53</b>                | <b>0.61</b>  | <b>0.62</b>  | <b>0.29</b>   | <b>5.72</b> | <b>3.29</b> | <b>0.05</b> | <b>0.04</b> |

Anthesis-Silking Interval (ASI), while Cluster 1 had a longer ASI. Regarding growth traits, Cluster 4 lines were characterized by the tallest plants and ears, followed by Cluster 1, whereas inbred lines in Cluster 3 had the shortest plants and ears height. Aspect ratings and Stay Green Ratings were relatively consistent across clusters, with only minor variations observed (Table 4).

Under optimum-N conditions (Figure 2), the inbred lines were classified into three primary clusters (1, 2, and 3) upon truncating the dendrogram at a 70 % genetic similarity level (Table 3). Cluster 1 contained the same lines as under low-N conditions, with the addition of a few lines from Cluster 3. Cluster 2 contained almost the same lines as under low-N, at an 82 % genetic similarity level. Cluster 3 contained the highest number of inbred lines, with thirty-

seven inbred lines at a 70 % genetic similarity coefficient. It included all the lines from heterotic group 4 in low-N conditions, with the exception of SMLW168 and SMLW17, along with a few lines from heterotic group 3 under low-N conditions. Based on the *per se* performance of the inbred lines under optimum-N conditions (Table 4), the mean grain yield of the inbred lines in Cluster 1 was the highest, followed by Cluster 3, while the inbred lines in Cluster 2 had the lowest mean grain yield. Concerning flowering traits, lines in Clusters 1 and 3 displayed similar flowering behavior. In contrast, Cluster 2 exhibited delayed flowering. With respect to growth traits, inbred lines in Cluster 1 had the highest plants and ear heights, with an average of 125.83 cm and 59.46 cm, respectively. Inbred lines in Cluster 3 were also relatively tall with mean plants and ear heights of 121.77 cm and



**Figure 1: Dendrogram of 79 maize inbred lines (low-N) constructed from GCA effects of grain yield and other traits (HGCAMT) using Ward’s minimum variance cluster analysis**

**Table 4: Mean *Per se* Performance of Maize Inbred Lines across different Heterotic Group**

| Character             | LOW – N         |       |       |       | OPTIMUM – N     |        |        |
|-----------------------|-----------------|-------|-------|-------|-----------------|--------|--------|
|                       | Heterotic Group |       |       |       | Heterotic Group |        |        |
|                       | 1               | 2     | 3     | 4     | 1               | 2      | 3      |
| Grain yield (kg/ha)   | 377.1           | 327.4 | 300.6 | 398.4 | 2003.3          | 1528.2 | 1763.5 |
| Days to anthesis      | 66.3            | 67.6  | 67.0  | 66.2  | 62.1            | 64.4   | 63.5   |
| Days to silking       | 70.3            | 71.1  | 70.5  | 69.3  | 64.5            | 66.7   | 66.0   |
| ASI                   | 3.9             | 3.4   | 3.5   | 3.2   | 2.4             | 2.3    | 2.5    |
| Plant height (cm)     | 92.6            | 83.1  | 83.1  | 89.9  | 125.8           | 109.8  | 121.8  |
| Ear height (cm)       | 39.1            | 36.0  | 34.9  | 37.0  | 59.5            | 49.2   | 56.1   |
| Ears per plant        | 0.9             | 0.9   | 0.9   | 1.0   | 0.9             | 0.9    | 0.9    |
| Plant aspect (1 – 10) | 4.4             | 4.6   | 4.5   | 4.5   | 3.1             | 3.4    | 3.3    |
| Ear aspect (1 – 10)   | 4.2             | 4.5   | 4.5   | 4.3   | 2.9             | 3.3    | 3.2    |
| Stay green (1 – 10)   | 5.9             | 5.8   | 5.7   | 5.7   |                 |        |        |

Source: Aboderin et al., (2023)

56.07 cm, respectively. In contrast, inbred lines in Cluster 2 had significantly shorter plants and ear heights, with an average of 109.85 cm and 49.25 cm. Regarding Aspect ratings, inbred lines in Cluster 2 displayed the highest aspect ratings for both plant (3.41) and ear (3.27). Cluster 3 closely followed with aspect ratings of 3.31 for plants and 3.21 for ears. Cluster 1 had the lowest aspect ratings for both plant (3.10) and ear (2.93) (Table 3).

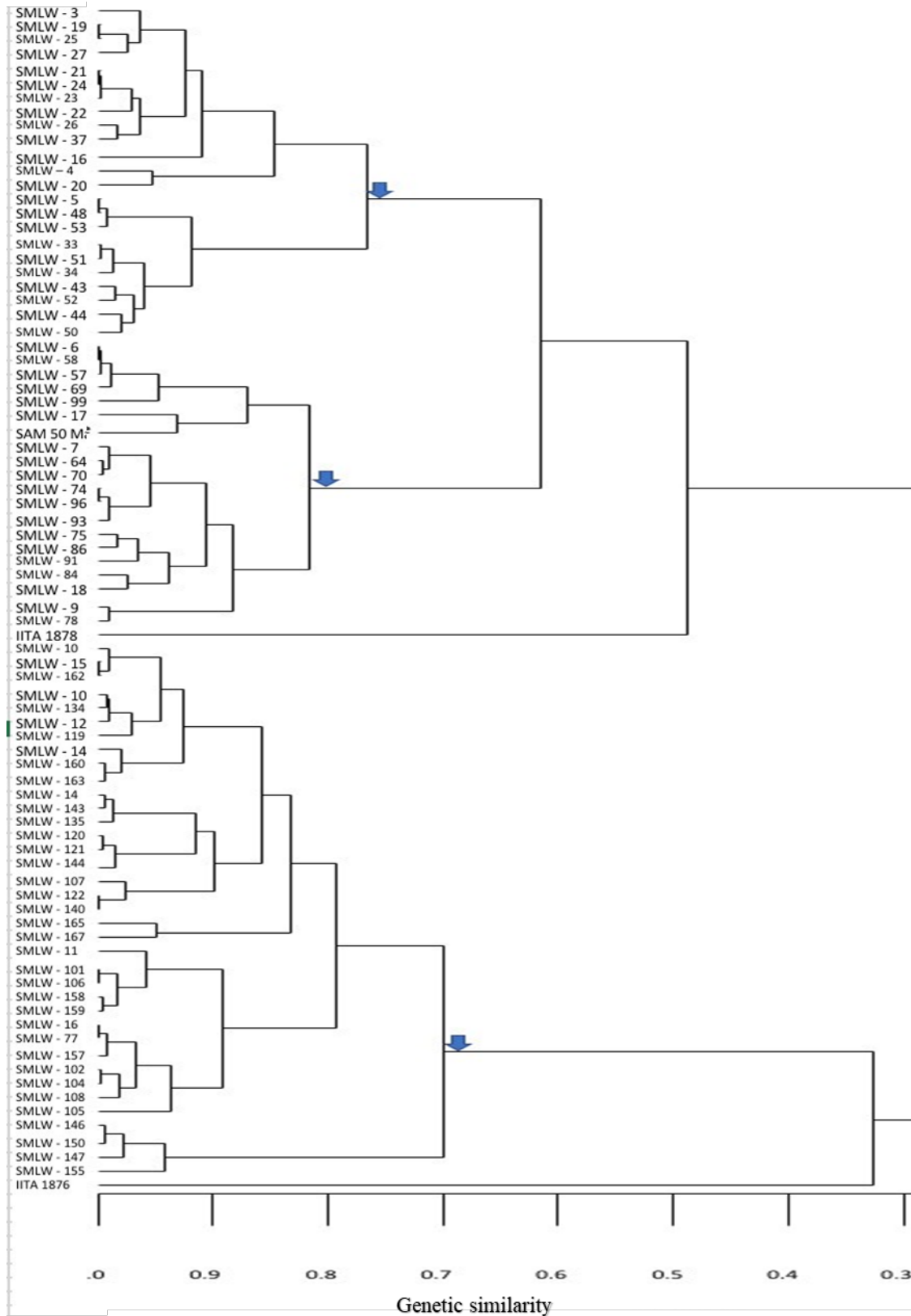
## Discussion

Estimating the general combining ability effects of parents in crosses is an important tool for breeders to assess the overall genetic potential of individual parents in contributing desirable traits to their offspring and for population improvement (Ertiro et al., 2017; Oluwaseun et al., 2022). GCA helps identify parents with superior additive genetic effects for specific traits, enabling breeders to efficiently develop superior hybrid varieties through controlled crosses (Amejbor et al., 2017). In this study, the presence of significant positive and negative GCA effects among the inbred lines for grain yield indicates the existence of both best and poorest combiners for this trait. Specifically, lines SMLW74 and SMLW146 (under low-N conditions), as well as lines SMLW146, SMLW147, SMLW150, and SMLW155 in the optimum-N environment, along with tester IITA 1876 under both nitrogen conditions, exhibited significant positive GCA

effects for grain yield, indicating their potential as excellent general combiners for this trait. As GCA is associated with additive gene action, traits under such influence tend to be highly heritable and less influenced by environmental factors (Ertiro et al., 2017; Abu et al., 2021). This underscores the significant additive genetic factors possessed by these lines and tester, which can consistently be passed on to their progeny. Thus, they represent valuable genetic resources for breeding high-yielding varieties.

Moreover, parental lines SMLW7, SMLW9, SMLW57, SMLW43, and SMLW146, exhibiting significant negative GCA effects for stay-green characteristics, emerged as the best combiners for these traits, positioning them as promising candidates for developing low-N tolerant maize varieties. The importance of stay-green traits in low-N conditions lies in their association with the plant's ability to retain greenness and sustain photosynthetic activity during grain filling, indicating improved stress tolerance and better performance in nitrogen-deficient environments. However, the exceptional performance of inbred lines SMLW146 and SMLW147, alongside tester IITA1876, as best combiners for both grain yield and stay-green characteristics, underscores their inherent genetic factors significantly contributing to higher grain production and stress tolerance. Their dual capability to excel in both traits makes them promising candidates for parent selection in hybridization programs aimed at producing maize varieties with improved yield potential

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**Figure 2: Dendrogram of 79 maize inbred lines (Optimum-N) constructed from GCA effects of grain yield and other traits (HGCAMT) using Ward's minimum variance cluster analysis**

and nitrogen stress tolerance (Olayiwola et al., 2021).

Furthermore, the significant negative GCA for days to anthesis and silking observed in lines SMLW 4, SMLW 43, SMLW 44, SMLW 34, and SMLW 37 under low- and optimum-N condition indicates that these lines tend to contribute to early flowering in their offspring when crossed with other parents. This characteristic could be advantageous in breeding programs aiming to develop maize varieties with shorter maturity periods or those adapted to specific growing conditions where early flowering is desirable, such as in savannas characterized by short rainfall durations or periodic terminal drought.

Similarly, the significant negative GCA effects for plant height observed in parental lines SMLW155, SMLW156 and tester SAM50M (under low-N) as well as lines SMLW160, SMLW163 and IITA1878 in optimum-N conditions, indicate that these lines and testers possess inherent genetic factors associated with shorter stature or reduced vertical growth, which can be inherited by their progeny. Likewise, the significant negative GCA effects for ear height observed in parental lines SMLW5, SMLW43, SMLW44, SMLW50, SMLW69 and tester IITA1878 under optimum-N condition, suggest that these lines and testers contribute genetic factors associated with reduced ear height in their offspring when crossed with other parents. These characteristics could be advantageous in breeding programs aiming to develop maize varieties with compact plant architecture or improved lodging resistance, as shorter plants or those with reduced ear height are often more stable and less prone to lodging, especially under adverse environmental conditions such as high winds or heavy rainfall, thereby contributing to reduced yield loss (Olakojo and Olaoye, 2005; Salami et al., 2007).

The GCA effects of multiple traits method (HGCAMT) was employed for the heterotic grouping of the inbred lines (Badu-Apraku et al., 2013). The HGCAMT method successfully identified four distinct clusters of inbred lines with varying characteristics under low-N conditions, while under optimum-N conditions,

these lines were classified into three primary clusters. Surprisingly, the composition of these clusters did not show significant differences between low- and optimum-N conditions; the majority of lines remained in the same groups in both growing conditions, with the exception of Cluster 3 in the optimum-N condition. Lines in heterotic group 1 under both low- and optimum-N conditions were characterized by GCA values close to the average for most traits, indicating a balanced genetic contribution. Interestingly, this group, based on *per se* performance evaluation, exhibited moderate grain yield, medium plant height, an average number of days to maturity, and a mixed response to low-N stress (Aboderin et al., 2023). The balanced GCA effects aligned well with their moderate performance, making lines in this heterotic group versatile options suitable for various growing conditions and valuable for hybrid breeding programs aiming for balanced performance.

Lines in Cluster 2 under both low- and optimum-N conditions were characterized by a mixture of positive and negative GCA values for different traits, potentially indicating their trait-specific strengths and weaknesses. In terms of *per se* performance, lines in this heterotic group exhibited lower grain yield and a higher susceptibility to low-N stress (Aboderin et al., 2023). Similarly, lines in Cluster 3 under low-N conditions displayed strong negative GCA effects for grain yield and susceptibility to low-N stress, which aligned with their observed low *per se* performance. This suggests that heterotic Group 2 and 3 lines may have specific traits of interest, but their overall lower *per se* performance suggests they may not be ideal for stand-alone cultivation. However, they can still be valuable for trait improvement when used in hybrid breeding programs with complementary lines (Akinwale et al., 2014; Obeng-Bio et al., 2019). Cluster 4 under low-N conditions and Cluster 3 under optimum-N conditions represent the desired heterotic groups containing lines with commendable *per se* performance. The high positive GCA values for grain yield of most lines in these clusters reflect their positive attributes, making them a promising source for developing hybrids anticipated to exhibit both

high grain yield and traits conducive to low-N stress tolerance.

Finally, having identified the strengths, weaknesses and potential use of lines within each heterotic groups in low- and optimum-N conditions, these inbred lines can be strategically recombined to create different base populations, which can then be improved over time using recurrent selection methods to develop better-performing varieties (Osuman et al., 2022).

## Conclusions

In conclusion, the significant positive and negative GCA effects observed across various traits in the study highlight the presence of both superior and inferior combiners for these traits. This enables the identification of best general combiners for each trait. For enhancing grain yield in optimum-N condition, inbred lines SMLW146, SMLW147, SMLW150, SMLW155, and tester IITA1876 emerged as valuable genetic resources. For low-N tolerance improvement, inbred lines SMLW7, SMLW9, SMLW57, SMLW43, and SMLW146 stand out as promising candidates. Notably, for simultaneous improvements in both traits, lines SMLW146 and SMLW147, along with tester IITA1876, are recommended as parents. Moreover, lines SMLW4, SMLW43, SMLW44, SMLW34, and SMLW37 are suggested as potential parents for developing extra early maturing maize varieties, while SMLW155, SMLW156, SMLW160, and SMLW163 could be incorporated as parents for breeding short-statured hybrids.

Furthermore, our analysis led to the classification of inbred lines into four heterotic groups under low-N and three groups under optimum-N conditions. Each group demonstrates distinct strengths and weaknesses. Notably, genotypes in Cluster 4 under low-N conditions and Cluster 3 under optimum-N conditions show promise as sources for developing hybrids with improved yield and nitrogen stress tolerance.

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## Conflicts of Interest

The authors declare there are no conflicts of interest.

## Authors Contribution

MO: developed the inbred lines used in the study; OSA and MO: conducted the experiments; OSA: and ZM: collected data; MO: and OSA: analyzed and interpreted data; OSA: drafted the manuscript. GO, FAB and MO: supervised the research and critically revised the manuscript. All authors reviewed the manuscript.

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