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Responses of elite sorghum (*Sorghum bicolor* [L.] Moench) lines developed via gamma-radiation for grain yield, component traits and drought tolerance

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ABSTRACT

Induced plant mutagenesis is a powerful technique to create genetic variation for agronomic traits and drought tolerance selection programs. The objective of this study was to determine the response of elite sorghum (*Sorghum bicolor* [L.] Moench) lines developed via gamma-radiation for grain yield, component traits, and drought tolerance to select best performing lines for cultivation in water-stressed environments. Ten newly developed mutant lines and four check varieties were evaluated in two growing seasons under drought-stressed (DS) and non-stressed (NS) conditions in Namibia. Mutant lines were evaluated using a factorial experiment laid out in a randomized complete block design with three replications in custom-made rainout-shelter facility. Data on grain yield and yield-related traits were collected and drought tolerance selection indices were computed using mean genotype yield under non-stressed condition and drought-stressed condition. Data were subjected to standard analysis of variance, correlation and principal component analyses. The interaction effect of genotype × drought stress × season was non-significant for most assessed traits suggesting the relatively stable performance of the test lines for selection. Grain yield response of test genotypes varied from 0.55 to 2.27 t/ha under DS and 1.84 to 4.05 t/ha in NS conditions. Grain yield positively and significantly ($P < 0.05$) correlated with harvest index ($r = 0.79$), panicle weight ($r = 0.75$) and panicle length ($r = 0.37$), and negatively correlated with days to flowering ($r = -0.35$) under DS condition. Principal component (PC) analysis identified two PCs accounting for 96.35 % of total genotypic variation based on drought tolerance selection indices. Biplot analysis using a combination assessed traits allowed selection of drought tolerant mutant lines designated as ML4, ML10, ML6, and ML5 with mean grain yield of 2.27, 2.05, 1.89 and 1.67 t/ha under DS conditions, in that order. The selected lines are recommended for further multi-environment evaluations for release and large-scale production in Namibia or other related agro-ecologies.

1. Introduction

Sorghum [*Sorghum bicolor* (L.) Moench, $2n = 2x = 20$] is the 5th most widely produced cereal crop after maize, wheat, rice and barley globally. It is annually cultivated across the globe at an estimated area of 42 million hectares with a total grain production of 59 million tons ([1]; FAOSTAT, 2020). Sub-Saharan Africa (SSA) contributes to 70 % of the total area and 50 % of sorghum production worldwide. Sorghum yield is the lowest in Africa (≈1 t/ha) compared with potential yield of the crop reaching up to 10.5 t/ha [2,3]. The major constraints to sorghum production and productivity include biotic stresses notably pests (e.g., bird damage, parasitic weeds, pre- and post-harvest insect pests) and diseases (e.g., anthracnose, mildew and head smut), and abiotic stresses particularly drought, extreme temperatures and poor soil fertility [4].

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Sorghum is relatively drought tolerant and thrives under harsh growing conditions where other dominant cereal crops fail [5,6]. However, severe and recurrent drought due to climate change is the leading cause of sorghum's yield gap and food insecurity in the continental Africa particularly in water-limited countries such as Namibia and South Africa [7]. Drought stress during the reproductive and grain filling stages reduces grain yield by up to 80 % [8]. A combination of moderate drought stress during pre- and post-flowering stages led to grain yield loss of 96 % [9]. Severe drought stress, use of low yielding, poorly adapted and drought susceptible sorghum genotypes reportedly accounted for a total yield loss [10]. Various options are recommended to mitigate drought stress in crop production including use of irrigation water, cultural practices (e.g. mulching and cover crops) and adoption of drought-tolerant varieties [11]. Breeding and deployment of high yielding and drought adapted cultivars is the most economic and sustainable approach under water scare with smallholder sorghum production systems where high temperatures and low and erratic rainfall are prevalent.

Sorghum is the second most widely cultivated cereal crop behind pearl millet (*Pennisetum glaucum* [L.] R. Br.) in Namibia. It is widely cultivated in the northern regions of the country such as in Zambezi, Kavango East and West, Otjozondjupa, Oshikoto, Oshana, Ohangwena, Omusati and Kunene under rainfed production condition ([7]; FAOSTAT, 2020). It is mainly cultivated by smallholder farmers to prepare various food and local beverages contributing to household food security, enhanced livelihoods and cash income [7]. To date there are only two sorghum varieties that are officially released and grown in Namibia. The mean grain yield of sorghum in the country is considerably low (<300 kg/ha) than potential yields of 4.05 and 4.78 t/ha reported in Zimbabwe and South Africa, respectively ([12,13]; FAOSTAT, 2020). Low sorghum productivity in Namibia is attributable to continued use of a few low yielding varieties which are susceptible to drought and heat stress. According to the Namibian Statistic Agency (https://nsa.org.na/) close to 91.14 % of sorghum farmers use seeds of low yielding landraces due to unavailability of improved varieties. Landrace varieties are the main sources of seed for sorghum production in most rural communities in Namibia. Prominent landraces widely grown in the country include 'Nyova' and 'Nswe' for their high stem sugar content and 'Kotovava', 'Kakumbama', 'Kankota', 'Omusamane iteka ondaku' which have gooseneck type panicle grown for grain production. Other distinguished landraces grown include the white grain 'Ekoko' and 'Okambete' mainly used to prepare porridge locally referred to as 'pap', "isima" and "oshifima", while the red grain landrace variety 'Okatombo' is widely grown to make local beverages such as "sikundu" and "marovhu" [14]. Landraces are highly valued for possessing diverse farmer-preferred attributes such as unique taste, eating and brewing quality, adaptation to grow under low input farming systems and marginal agricultural lands. The genetic variability present in the traditional varieties is not well studied to select ideotypes with novel traits for sorghum breeding programmes and conservation.

A collaborative sorghum improvement program was initiated between the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) and the Namibian Government in 1991. The goal was to select best performing ICRISAT-bred and introduced lines and landraces targeting high grain yield and drought-adaptive traits. This led to development and release of two sorghum varieties Macia (ICRISAT-bred and introduced pure line) and red sorghum (local line developed through mass selection) in 1999 and 2004, respectively. Reportedly variety Macia showed lower emergence, poor crop establishment and less grain yield under reduced watering, erratic rainfall distribution and late planting [13,15,16]. Hence, there is a need to develop locally adapted and best performing new generation sorghum genotypes combining farmer-preferred traits, high yield and drought tolerance.

Genetic variation is the foundation for breeding programs to select distinct and complementary parents with economic traits. Conventional plant breeding programmes rely on genetic diversity present among the available genetic pool to develop superior progenies [17]. Trait-based breeding has been adopted in the design, selection and deployment of cultivars by ICRISAT and various national sorghum improvement programs [18]. The use of the natural genetic variation present among landraces and genetic resources maintained at gene banks and broadening the gene pool in presently cultivated and obsolete varieties would aid selection of superior parents for drought tolerance breeding.

The low genetic base of sorghum in Namibia hindered selection efforts and breeding progress in the development and release of high-yielding and drought adapted tolerant genotypes. Thus, there is a need for genetic enhancement of sorghum for breeding. Induced plant mutation creates genetic variation in a relatively shorter period than variation achievable through natural mutation (varying from 10^{-5} to 10^{-8} per loci in higher plants) and controlled crosses [19]. Induced mutagenesis is a powerful tool to create genetic variation for key traits including early maturity, grain yield, yield-improving traits and drought tolerance in sorghum [19]. Various reports are available that reported the value of induced mutagenesis in variety selection, design and release globally. According to the FAO/IAEA Mutant Variety Database [20] a total of 3365 mutant crops varieties in 228 crop species across 73 countries have been released for cultivation globally. For sorghum, 18 mutant varieties with desirable traits such as high grain yield, short plant height, early maturity and grain quality (high contents of protein, and starch) were developed between 1955 and 2014 using mutation breeding [20].

Gamma irradiation is the most preferred mutagenic agent due to its relatively higher degree of plant tissue penetration, reproducibility and greater mutation frequency [21]. Kenga et al. [22] selected eight sorghum mutant lines with high yield and drought tolerance through gamma irradiation which were recommended for production in Nigeria. Human et al. [23] developed and released three high-yielding and drought tolerant sorghum mutant varieties namely Pahat, Samurai I and Samurai II using gamma radiation in Indonesia. In Mali, mutant varieties including Djeman, Djemanin, Sandje and Tiedjan were developed using gamma radiation for traits such as early and late maturity, short plant height, longer panicle, white grain colour, larger grain size and high yield [20]. In China, mutant varieties Jinfu 1, Jinza 1 and Longfuliang 1 were developed using gamma radiation ideal for machine harvesting, higher yield potential, wide adaptability, early maturity and short plant height suitable for high density planting.

A collaboration between the Government of Namibia and the International Atomic Energy Agency (IAEA) was initiated in 2009 with the aim to develop superior sorghum varieties with novel traits including high grain yield, yield-improving and drought-adaptive traits. Gamma irradiation technique was recommended by the Namibian Radiation Regulatory Authority on the bases that irradiated seed have no negative impact on the environment. Thus, seed of sorghum variety Macia and Red sorghum were irradiated at the Joint FAO/IAEA laboratories in Seibersdorf, Austria for genetic enhancement. Subsequently, 45 mutant populations were selected and advanced from the M_2 through M_5 generations of variety Macia. The elite advanced mutant lines should be screened to identify the best performing lines for high yield and yield components and drought tolerance. This will enable selection of phenotypically stable mutant lines with drought tolerance characteristics, distinct agronomic traits including reduced days to flowering, shorter plant height, higher panicle length and better panicle weight for further recommendation. Understanding agronomic traits association [24] and use of drought selection indices [25] enhances selection efficiency and can aid development of superior lines suited for water-limited conditions. In light of the above background the objective of this study was to determine the response of elite sorghum lines developed via gamma-radiation for grain yield and component traits, and drought tolerance to select best performing lines for cultivation in water-stressed environments.

2. Materials and methods

2.1. Plant material

The study used 14 sorghum genotypes consisting of 10 newly bred elite mutant lines (M_6 generations) and four check varieties (Table 1). The check variety Macia (SDS 3220) is the most widely grown in Namibia, while variety NAM 738/2 was sourced from the National Botanical Research Institute (NBRI)/Namibia. Genotypes ICSR 55 and ICSR 59 were sourced from ICRISAT/India and used as comparative controls. The test genotypes were selected for their short plant height, early-maturity and high yield performance based on preliminary yield tests. The advanced 10 mutant lines were developed from gamma irradiated (350 Gy) seed of variety Macia which was followed by progeny selection with a pedigree selection method.

2.2. Description of study site and environment

The study was conducted using a custom-made rainout-shelter facility in two seasons (SN1-in-2019 and SN2-in-2020) at Mannheim Crop Research Station, Tsumeb, Namibia (19°10′07.3″S 17°45′52.2″E). The first season trial was conducted from 7 August to December 13, 2019, whereas the second season trial was from 17 February to 26 June 2020. Monthly mean, maximum and minimum temperatures and relative humidity during the experiments are shown in Fig. 1. In the first season, the maximum, average and minimum monthly temperatures gradually increased from 31.24 °C, 20.8 °C and 10.36 °C–34.54 °C, 26.73 °C and 19.63 °C, respectively, and the corresponding relative humidity values increased from 37.54 %, 25.19 % and 10.42 %–79.80 %, 53.28 % and 26.75 %. The maximum, average and minimum monthly temperatures in the second season decreased from 32.43 °C, 25.41 °C and 18.4 °C to 27.29 °C, 16.98 °C and 6.67 °C, respectively, while the relative humidity gradually decreased from 82.29 %, 26.9 % and 31.5 %–60.75 %, 38.17 % and 15.58 %, respectively.

3. Experimental design and trial establishment

The newly improved 10 advanced mutant lines and the four check (constituting 14 genotypes) were evaluated under drought-stressed (DS) and non-stressed (NS) conditions (representing two water regimes) in two seasons (SN1 and SN2) in Namibia. Test lines were evaluated using a 14 genotypes x 2 water regimes x 2 season's factorial experiment laid out in a randomized complete block design with three replications. Three to five seed were planted per hill on 2-m long single-row plots with the spacing of 60 cm between rows and 15 cm within rows. Fourteen days after emergence thinning was carried out by keeping two seedling plants per hill, and 21 days after emergence to one plant per hill. This provided a plant density of 11.11 plants m^{-2}. The NS condition involved maintaining soil moisture at field capacity by continuous irrigation until physiological maturity. DS condition was imposed by withholding water six weeks after planting and survival irrigation was supplied to avoid a permanent wilting and to promote seed set. Compound synthetic fertilizers composed of Nitrogen [N], Phosphorus [P] and Potassium [K] with a ratio of 2:3:2 by 30 kg N/ha, 45 kg Phosphorus pentoxide (P_2O_5)/ha and 30 kg Potassium oxide (K_2O)/ha as basal dressing. Urea (30 kg/ha) was applied at 35 days after sowing. All standard agronomic practices such as weeding and pest control were adopted during the entire cropping period.

Table 1
Name and origin of sorghum genotypes used in the study.

Sr. No	Genotype name/designation	Code	Source	Seed colour
1	L3P15-16	ML1	MAWLR	White
2	L7P9-9	ML2	MAWLR	White
3	L3P15-9	ML3	MAWLR	White
4	L7P9-4	ML4	MAWLR	White
5	L7P9-2	ML5	MAWLR	White
6	L7P9-14	ML6	MAWLR	Red
7	L3P15-13	ML7	MAWLR	White
8	L3P15-40	ML8	MAWLR	White
9	L7P9-13	ML9	MAWLR	Yellow
10	L7P7-3	ML10	MAWLR	White
11	MACIA	SDS 3220	ICRISAT	White
12	NAM 738/2	738/2	NBRI	Red
13	ICSR 55	55	ICRISAT	White
14	ICSR 59	59	ICRISAT	White

Sr. No, serial number; entries 1 to 10 are mutant lines derived from variety Macia using gamma radiation with a dose of 350Gy; MAWRL, Ministry of Agriculture, Water and Land Reform/Namibia; NBRI, National Botanical Research Institute/Namibia; ICRISAT, International Crop Research Institute for the Semi-Arid Tropics/India.

4. Data collection

Data were collected on agronomic traits following the methods described by the International Board for Plant Genetic Resources (IBPGR) and ICRISAT [26]. Five plants per plot were randomly selected and tagged for individual plant data collection. The days taken to 50 % flowering (DF) was recorded as number of days from planting to the date when 50 % of the plants showed anthesis. At maturity, plant height (PHT) in cm was measured from the ground to the tip of the panicle. Panicle length (PLT) in cm was measured from the lower panicle branch to the tip of the panicle. Panicle weight (PWT) in grams was weighed and recorded before threshing. Grain yield per plant (GYP) in grams was measured by weighing grains per panicle recorded after threshing and converted to t/ha (GYH). Thousand-grain weight (TGW) in grams was measured by weighing 1000-grains at 12 % moisture content. Harvest index (HI) was computed as the ratio of GYP and DMW and expressed as a percentage. Above ground biomass was harvsted and a dry mater weight (DMW) measured in grams as the sum of total dry matter of leaves, stems and panicles. Fresh above ground biomass were first sun-dried for 3–5 days, then oven-dried at 70 °C temperature for 72 h before weighing. Chlorophyll content (SPAD value) of flag leaves at physiological maturity was measured using a portable meter (SPAD-502, Konica-Minolta, Plainfield, IL, USA). Leaf canopy temperature (CT) in ºC was recorded from flag leaves at physiological maturity between 10 a.m. and 12 p.m. using a non-contact laser infrared digital thermometer (EC-Technology, N10IT001–5, USA).

Yield-based drought tolerance indices including mean productivity (MP), harmonic mean (HM), stress tolerance (TOL), geometric mean production (GMP), stress susceptible index (SSI), yield index (YI), yield stability index (YSI) and stress tolerance index (STI) were calculated (Table 2). Drought indices were computed using grain yield of each test line under NS and DS conditions.

5. Data analysis

Collected data were subjected to combined analysis of variance (ANOVA) after homogeneity test of variance using Genstat 18th edition [33]. Data were analysed to determine the effects of genotypes, water regimes, seasons and their interaction on the studied agronomic traits. The least significant difference (LSD) test procedure was used for mean separation at 5 % level of significance. Pearson's correlation coefficients were computed to determine the level of trait association using IBM SPSS Statistics version 27.0. Principal component analysis (PCA) based on the correlation matrix was performed to determine influential traits and drought tolerance indices. PCA bi-plots analysis was constructed using R version 4.0 (R Core Team, 2020) to display relationship between test genotypes, drought tolerance indices and agronomic traits to aid selection.

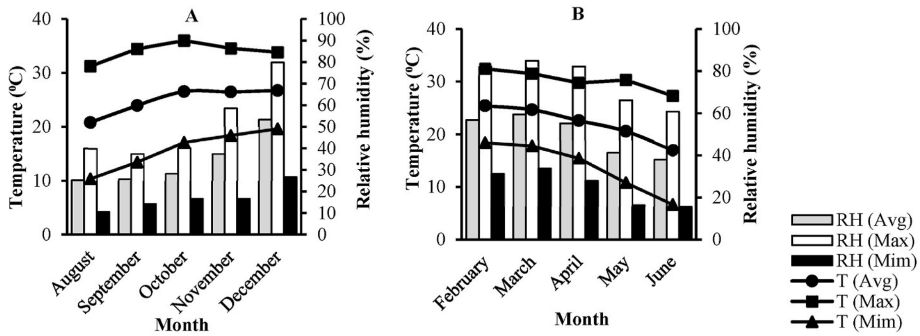

Fig. 1. The combined charts of histogram and lines showing monthly mean (Avg), maximum (Max) and minimum (Min) temperatures (T) and relative humidity (RH) during SN1-in-2019 (A) and SN2-in-2020 (B). Source: Namibia Meteorological Service (http://www.meteona.com/).

Table 2
Drought tolerance indices and computational formulas used to evaluate drought response of 14 sorghum genotypes.

Indices	Computation	Reference (s)
Mean productivity (MP)	$MP = \frac{(Y_p + Y_s)}{2}$	Rosielle and Hamblin [27]
Harmonic mean (HM)	$HM = \frac{2(Y_p \times Y_s)}{Y_p + Y_s}$	Farshadfar and Sutka [25]
Geometric mean production (GMP)	$GMP = (Y_p \times Y_s)^{0.5}$	Sio-Se Mardeh et al. [28]
Tolerance index (TOL)	$TOL = (Y_p - Y_s)$	Rosielle and Hamblin [27]
Stress susceptible index (SSI)	$SSI = \frac{1 - \left(\frac{Y_s}{Y_p}\right)}{SI}$ Where: $SI = 1 - \left(\frac{\hat{Y}_s}{\hat{Y}_p}\right)$	Mahalakshmi et al. [29]
Yield index (YI)	$YI = \frac{Y_s}{\hat{Y}_s}$	Gavuzzi et al. [30]
Yield stability index (YSI)	$YSI = \frac{Y_s}{Y_p}$	Bouslama and Schapaugh [31]
Stress tolerance index (STI)	$STI = \frac{(Y_s \times Y_p)}{(\hat{Y}_p)^2}$	Fernandez [32]

Note: Y_p, mean genotype yield under non-stressed condition; Y_s, mean genotype yield under drought-stressed condition; $\hat{Y}_p$ mean of all genotypes yield under non-stressed condition; $\hat{Y}_s$, mean of all genotypes yield under drought-stressed condition.

6. Results

6.1. Effects of genotype, drought, season and their interactions on agronomic traits

Combined analysis of variance showing the effects of genotype, water regime and season and their interactions on the assessed agronomic traits is presented in Tables 3 and 4. Genotype had a significant ($p < 0.05$) effect on most assessed traits except for TGW, CT and SPAD. Water regime had significant ($p < 0.05$) effect on most assessed traits, whereas season had significant effect on most traits except DF, PLT and CT. Genotype × water regime interaction effect was significant ($p < 0.05$) for DMW only. Genotype × season interaction effect was significant for DMW, PWT, TGW and SPAD. Water regime × season interaction effect was significant for PHT, DMW, PWT, TGW, GYH and SPAD. Genotype × water regime × season interaction effect was significant ($p < 0.05$) for PLT only.

6.2. Mean performance of sorghum mutant lines under non-stressed and drought-stressed conditions

The mean values for the assessed agronomic traits among the mutant lines and checks under NS and DS conditions are summarized in Figs. 2 and 3, and Supplemental Table 1. The mean DF in SN1 and SN2were 80 and 81 days, respectively. Lines ML1 and ML10 were early flowering types at 69.5 days under NS condition, whereas lines ML8 and ML7 were late flowering at 89.5 and 93.5 days, respectively. Under DS condition, line ML6 was the earliest to flower (74.5 days) followed by ML1 and ML4 (75.5 days to flowering) compared to the late flowering lines ML2 at 95 days and ML7 at 94 days. The mean PHT under NS condition was 151.89 cm compared to 123.52 cm under DS condition. The check variety ICSR 59 had the shortest plant height (90.9 cm) followed by lines ML7 and ML8 (98.7 cm), whereas lines ML4, ML10 and check variety NAM 738/2 were the tallest with PHT of 152.8, 148 and 47.6 cm under DS condition, respectively. Under NS condition, ICSR 55, ML7 and Macia were the shortest with PHT of 117.8, 119.6 and 123.9 cm, respectively, compared to the tallest entries such as the check variety NAM 738/2, lines ML3 and ML10 with PHT of 196.8, 184.8 and 181.2 cm under NS condition, respectively.

The mean PLT under NS and DS conditions were 22.71 and 19.29 cm, respectively. Under NS condition, lines ML3, ML1 and check variety NAM 738/2 recorded the longest panicle of 28.2, 27.5 and 26.3 cm,

Table 3
Mean-squares values and significant tests for agronomic traits of 14 sorghum lines evaluated in two seasons (SN1 and SN2) under drought-stressed and non-stressed conditions.

Sources of variation	d.f.	DF (days)		PHT (cm)		PLT (cm)		PWT (g/panicle)		TGW (g/1000 seed)	
Replication	2	3.79		1009.2		4.98		79.9		63.8	
Genotype (Gen)	13	512.62	**	7286.2	**	78.35	**	717.5	*	18.8	ns
Water regime (WR)	1	1382.90	**	33811.9	**	491.00	**	12388.0	**	148.0	*
Season	1	21.43	ns	7501.2	**	10.51	ns	4500.0	**	4359.0	**
Gen x WR	13	90.03	ns	385.8	ns	10.99	ns	275.7	ns	26.6	ns
Gen x Season	13	162.45	ns	315.0	ns	5.90	ns	396.8	*	70.5	**
WR x Season	1	110.10	ns	1881.5	*	5.63	ns	1328.0	*	90.7	*
Gen x Season x WR	13	90.56	ns	523.1	ns	22.95	**	228.0	ns	17.9	ns
Residual	56	91.88		285.0		6.58		199.2		21.6	

Note: d.f., degrees of freedom; DF, days to 50 % flowering; PHT, plant height; PLT, panicle length; PWT, panicle weight; TGW, 1000-grain weight; * significant at $p \leq 0.05$; ** significant at $p \leq 0.001$; ns, non-significant difference.

Table 4
Mean-squares values and significant tests for agronomic traits of 14 sorghum lines evaluated in two seasons (SN1 and SN2) under drought-stressed and non-stressed conditions.

Sources of variation	d.f.	GYH (t/ha)		HI (%)		DMW (g/plant)		SPAD value		CT (°C)	
Replication	2	1.45		84.1		3874.3		174.2		114.0	
Genotype (Gen)	13	3.79	*	315.81	**	4671.6	**	116.3	ns	12.0	ns
Water regime (WR)	1	107.2	**	2650.66	**	54762.6	**	1302.0	**	1663.0	**
Season	1	31.39	**	3159.97	**	5982	*	4248.0	**	1.5	ns
Gen x WR	13	1.23	ns	67.01	ns	1722.4	*	19.2	ns	7.0	ns
Gen x Season	13	1.29	ns	87.28	ns	6190.8	**	78.3	**	12.2	ns
WR x Season	1	16.74	**	215.25	ns	5802.3	*	3225.0	**	70.5	ns
Gen x Season x WR	13	1.04	ns	68.82	ns	1350.9	ns	34.1	ns	3.3	ns
Residual	56	1.09		91.60		885.6		28.4		24.4	

Note: d.f., degrees of freedom; GYH, grain yield (t/ha); HI, harvest index; DMW, above ground dry mater weight; CT, canopy temperature; SPAD, SPAD value; * significant at $p \leq 0.05$; ** significant at $p \leq 0.001$; ns, non-significant difference.

compared to the relatively shortest panicle recorded at 17.1, 18.4 and 21 cm for ML2, ML5 and ICSR 55, respectively. Under DS condition, lines ML1, ML7 and ML3 recorded the longest PLT of 24.2, 21.9 and 21.7 cm, respectively, compared to the shortest PLT exhibited by ML2, ML5 and ML9 at 16.0, 16.8 and 17.3 cm, respectively. The mean PWT under NS and DS conditions were 36 and 18.8 g/panicle, respectively. Lines ML8, ML6 and ML9 recorded the highest PWT of 56.5, 47.6 and 45 g/panicle, in that order, compared to the lowest PWT of 19.4, 20.3 and 25.9 g/panicle for NAM 738/2, ML2 and ML1 under NS condition. Under DS condition, lines ML4, ML6 and ML10 recorded the highest PWT of 30, 29.8 and 28.1 g/panicle, respectively, compared to the lowest PWT recorded for NAM 738/2, ML2 and ML3 at 7.9, 12.5 and 13.2 g/panicle, respectively. The mean TGW under NS and DS conditions were 21.4 and 19.53 g/1000-seed, respectively. ICSR 59, ML1 and ICSR 55 recorded the highest TGW of 25.2, 23.6 and 22.3 g under NS condition, respectively, whereas lines ML6, ML2 and ML7 had the lowest TGW of 18.3, 18.8 and 19 g under similar conditions. Under DS condition, line ML5 and check varieties ICSR 55 and NAM 738/2 recorded the highest TGW of 23.1, 21.8 and 21.3 g, compared to the lowest TGW recorded for ML8, ICSR 59 and ML6 of 16, 17 and 17.5 g, in that order. The mean GYH of 2.55 and 1.69 t/ha were recorded under NS and DS conditions, respectively. Lines ML8, ML7 and check variety Macia recorded the highest GYH of 4.05, 3.66 and 3.66 t/ha under NS condition, whereas ML1, NAM 738/2 and ML3 recorded the lowest GYH of 1.84, 1.99 and 2.18 t/ha, respectively. Under DS condition, lines ML4, ML10 and ML6 recorded the highest GYH of 2.27, 2.05 and 1.89 t/ha, in that order, compared to the lowest GYH recorded for ML2, NAM 738/2 and ML3 of 0.55, 0.59 and 0.72 t/ha.

The mean HI values under NS and DS condition were 24.7 % and 16.8 %, respectively. Macia, ML4 and ICSR 55 recorded higher HI of 32.9 %, 32.7 % and 31.9 % under NS condition, whereas check variety NAM 738/2, lines ML1 and ML3 recorded the lowest HI of 17.9 %, 18.3 % and 18.3 %, in that order under similar conditions. Under DS condition, Macia, ML4 and ML10 recorded the highest HI of 24.5 %, 22.4 % and 22.2 %, compared to the lowest HI recorded for ML2, NAM 738/2 and ML3 at 4.1 %, 9.5 % and 10.3 %, in that order. The mean DMW under NS and DS conditions were 113.5 and 77.41 g/plant, respectively. ML7, NAM 738/2 and ML8 recorded the highest DMW of 179.6, 136.5 and 136.1 g/plant under NS condition compared to the lowest DMW of 69.1, 81.8 and 97.1 g/plant for check varieties ICSR 55, ICSR 59 and Macia, respectively. Under DS condition, ML2, NAM 738/2 and ML7 recorded DMW values of 109.5, 105.6 and 93.9 g/plant, respectively, whereas the lowest DMW at 59, 54.2, 56 and 59.2 g/plant were recorded for ML9, Macia, ICSR 59 and ICSR 59, respectively. The mean SPAD values under NS and DS conditions were 30.9 and 25.3, respectively. NAM 738/2, ML10 and ICSR 59 respectively recorded the highest SPAD values of 39.5, 34.3 and 33 under NS condition, whereas lines ML8, ML1 and ML3 recorded the lowest SPAD values of 25.8, 27.3 and 28.5, respectively under NS condition. Under DS condition, NAM 738/2, ML7 and ICSR 59 recorded the highest SPAD values of 31.7, 29.1 and 28.7, compared to the lowest values of 20, 21.1 and 22.6 recorded for ML9, ML8 and ML3, respectively. The mean CT under DS and NS conditions were 32.1 °C and 25.8 °C, respectively. The check varieties NAM 738/2 and ICSR 59 recorded the lowest CT of 25 °C under NS condition, whereas mutant lines ML7, ML4 and ML8 recorded the highest CT of 26.1 °C, 26.7 °C and 26.8 °C, respectively. Under DS condition, ML7, NAM 738/2 and ML6 recorded the lowest CT at 29.7 °C, 29.9 °C and 30.1 °C, respectively, compared to the highest CT at 34.6 °C, 34.1 °C and 34 °C recorded for ML1, ML9 and Macia, respectively.

6.3. *Drought tolerance indices*

Yield-based drought tolerance selection indices calculated using grain yield under DS and NS conditions is summarized in Figs. 4 and 5. Under DS condition the mean grain yield (Y_s) was 1.33 t/ha which was lower by 54.8 % compared with the potential yield (Y_p) under NS condition (2.92 t/ha). Line ML4 recorded the highest MP (2.95), HM (2.63), GMP (2.78), YI (1.00) and STI (0.89) averaged across seasons. The local check NAM 738/2 had the lowest MP (1.29), whereas line ML2 recorded the lowest HM (0.66), GMP (0.87), YI (0.44) and STI (0.14) across seasons. ML1 displayed the lowest TOL (0.65), SSI (0.16) and highest YSI (1.00), while ML9 showed the lowest YSI (0.28) across seasons.

6.4. *Correlations among assessed agronomic traits*

The correlation coefficients (r) of agronomic traits assessed under DS and NS conditions among the test lines is summarized in Table 5. PHT significantly and positively correlated with DMW ($r = 0.40$), PLT ($r = 0.52$) and PWT ($r = 0.26$) under DS condition. DMW exhibited significant and positive correlations with PWT ($r = 0.32$; $p \leq 0.01$) and SPAD ($r = 0.43$; $p \leq 0.01$), and negatively associated with HI ($r = -0.32$; $p \leq 0.01$) and CT ($r = -0.40$; $p \leq 0.01$) under DS condition. PLT was significantly and positively correlated with PWT ($r = 0.41$; $p \leq 0.01$), GYP ($r = 0.37$; $p \leq 0.01$) and HI ($r = 0.22$; $p \leq 0.01$) under DS condition. PWT exhibited a significant and positive correlations with GYP ($r = 0.75$; $p \leq 0.01$) and HI ($r = 0.45$; $p \leq 0.01$) under DS condition. Relatively low and positive correlation was exhibited between TGW with HI ($r = 0.23$; $p \leq 0.05$) under DS condition. GYP positively and significantly correlated with HI ($r = 0.79$; $p \leq 0.01$), whereas SPAD had negative and significant correlation with CT ($r = -0.40$; $p \leq 0.01$) under DS condition.

Under NS condition, low and negative correlations were recorded between DF with PHT ($r = -0.38$), PLT ($r = -0.34$) and TGW ($r = -0.22$). PHT moderately and positively correlated with PLT ($r = 0.47$; $p \leq 0.01$) under NS condition. Also, DMW exhibited highly moderate and positive correlations with PWT ($r = 0.53$; $p \leq 0.01$) and GYP ($r = 0.39$; $p \leq 0.01$), and a low and negative correlation with HI ($r = -0.30$; $p \leq 0.01$) under NS condition. PWT exhibited a low and positive correlation with TGW ($r = 0.29$; $p \leq 0.01$), high correlation with GYP ($r = 0.78$; $p \leq 0.01$) and low correlation with HI ($r = 0.30$; $p \leq 0.01$), and negative

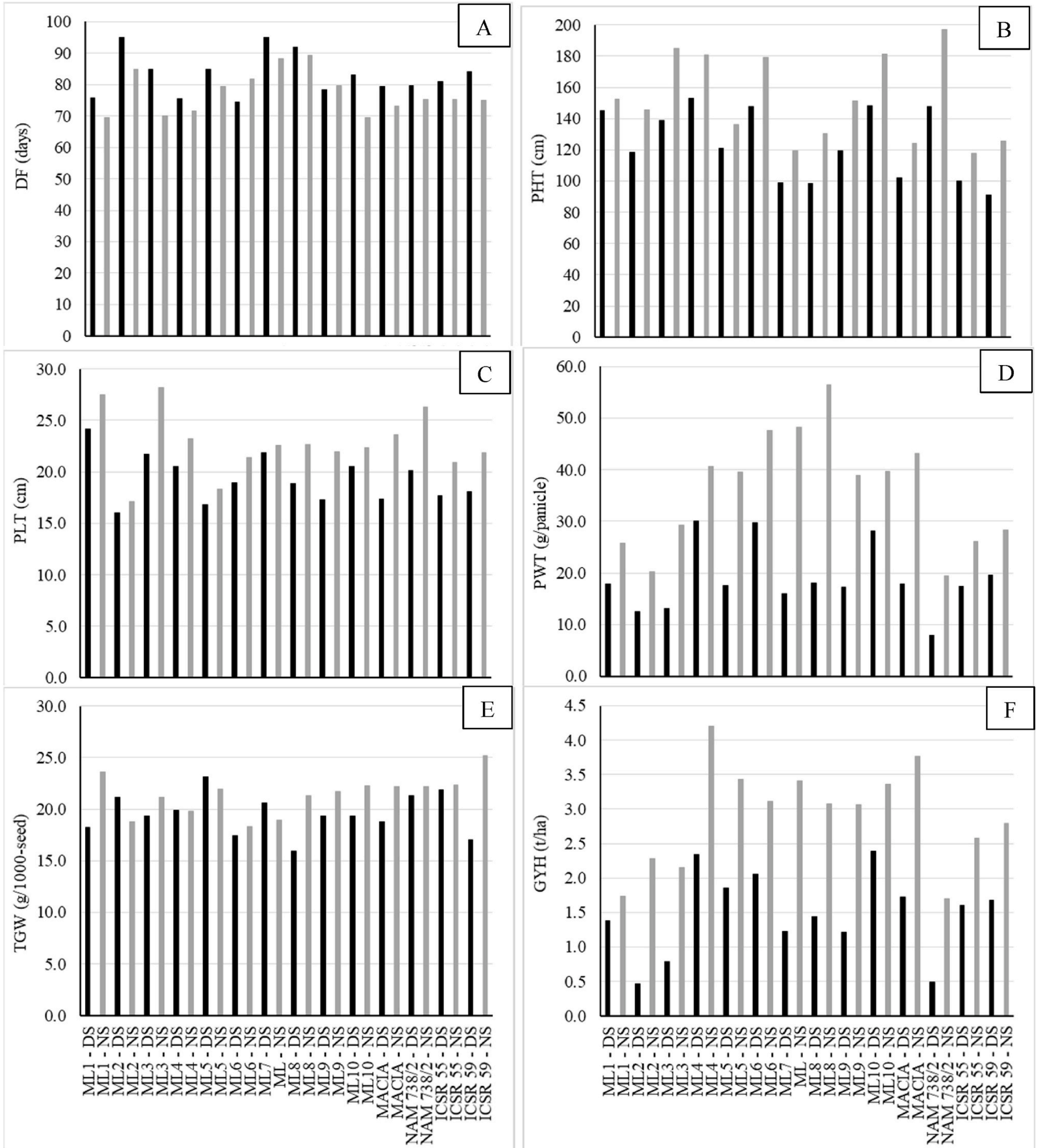

Fig. 2. Mean values for agronomic traits of 14 sorghum lines evaluated across two seasons (SN1 and SN2) under drought-stressed (DS) and non-stressed (NS) conditions. DF, days to 50 % flowering (A); PHT, plant height (B); PLT, panicle length (C); PWT, panicle weight (D); TGW, 1000-grain weight (E); GYH, grain yield (t/ha) (F).

association with SPAD (r = -0.35; $p \leq 0.01$) under NS condition. TGW exhibited a low and positive correlation with GYP ($r = 0.29$; $p \leq 0.01$) and HI ($r = 0.38$; $p \leq 0.01$), and negative correlation with SPAD ($r = -0.47$; $p \leq 0.01$) under NS condition. GYP positively and significantly correlated with HI ($r = 0.66$; $p \leq 0.01$) and negatively correlated with SPAD ($r = -0.38$; $p \leq 0.01$) under NS condition. HI displayed a low and negative correlation with SPAD ($r = -0.33$; $p \leq 0.01$) under NS condition.

6.5. *Correlations of yield and drought tolerance selection indices*

The correlations of Y_p, Y_s and drought tolerance indices are summarized in Table 6. Y_P showed relatively low correlation with Y_s (r = 0.27) and YI (r = 0.27), but moderate correlation with SSI (r = 0.49) and high correlations with MP (r = 0.92), HM (r = 0.60), GMP (r = 0.75), TOL (r = 0.87), and STI (r = 0.75), and negative correlation with YSI (r = −0.49). Ys significantly and positively correlated with MP (r = 0.63), HM (r = 0.87), GMP (r = 0.80), YI (r = 1.00), YSI (r = 0.52 and STI (r =

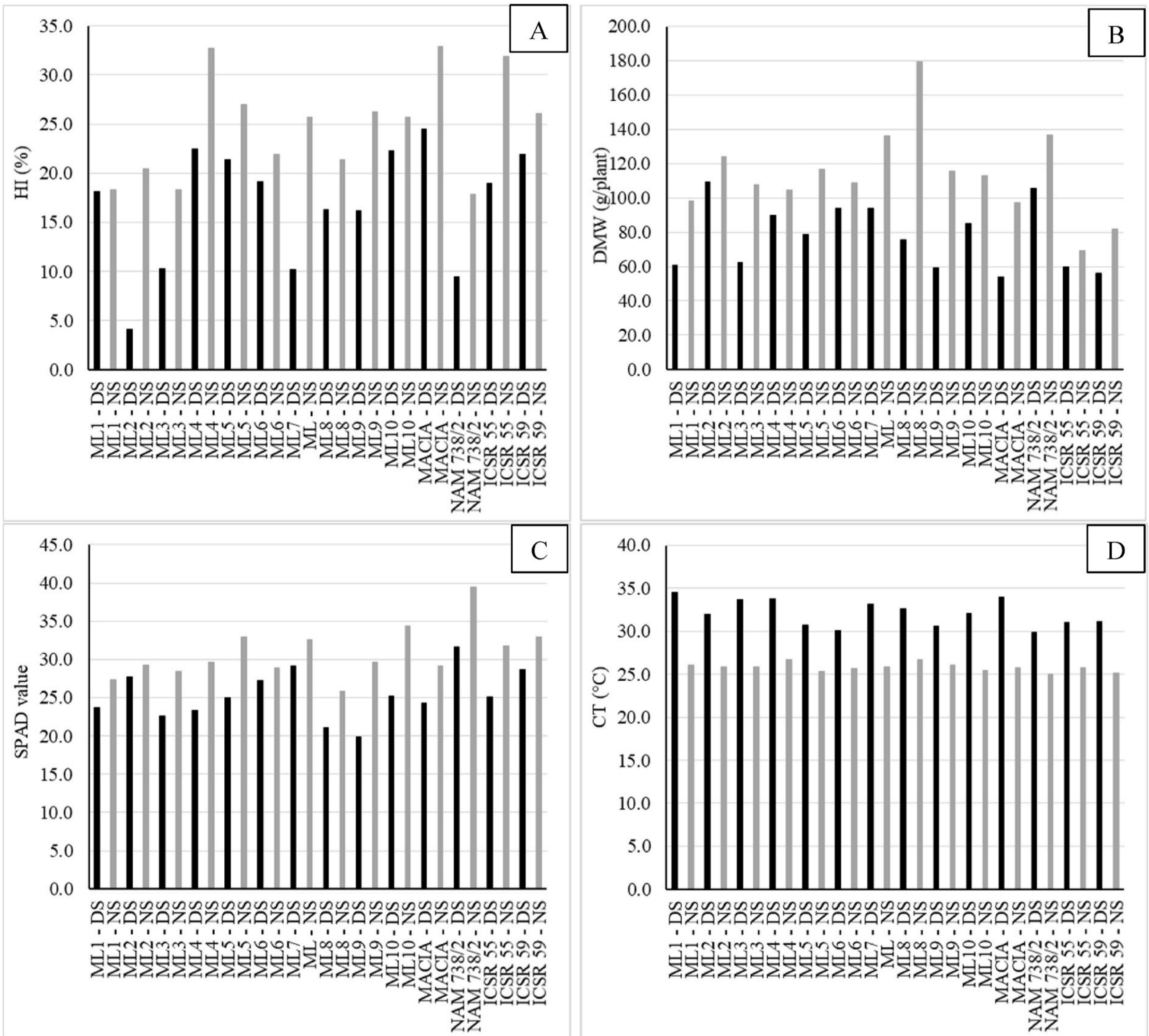

Fig. 3. Mean values for agronomic traits of 14 sorghum lines evaluated across two seasons (SN1 and SN2) under drought-stressed (DS) and non-stressed (NS) conditions. HI, harvest index (A); DMW, above ground dry mater weight (B); CT, canopy temperature (C); SPAD, SPAD value (D).

0.76), and negatively correlated with TOL (r = −0.25) and SSI (r = −0.52). MP displayed significant positive correlations with HM (r = 0.85), GMP (r = 0.94), YI (r = 0.63) and STI (r = 0.91). HM significantly and positively correlated with GMP (r = 0.98), YI (r = 0.87) and STI (r = 0.95). GMP exhibited a significant and positive correlations with TOL (r = 0.34), YI (r = 0.80) and STI (r = 0.97). TOL positively and significantly correlated with SSI (r = 0.76) and STI (r = 0.36), and negatively correlated with YSI (r = −0.76). SSI displayed significant negative correlations with YI (r = −0.52) and YSI (r = −1.00), while YI significantly and positively correlated with YSI (r = 0.52) and STI (r = 0.76).

6.6. Principal component analysis (PCA)

Principal component analysis showing loading scores, explained and cumulative variances for agronomic traits under DS and NS conditions, and for drought tolerance indices is summarized in Tables 7 and 8. The PCA identified three principal components (PCs) accounting for 73.3 % of total genotype variation contributed by agronomic traits under DS condition. Principal components 1 (PC1), PC2 and PC3 explained 34.02, 22.53 and 16.75 % of the total variation, respectively. High and positive loadings were observed for HI, GYP and PWT with PC1, while DMW, SPAD and PWT recorded high and positive loadings with PC2. PHT and PLT recorded high and positive loadings with PC3. High and negative loadings were recorded for DF and SPAD with PC1, whereas CT recorded high and negative loadings with PC2 under DS condition. Under NS condition, four PCs contributing 83.46 % of total variation were identified for agronomic traits. PC1, PC2, PC3 and PC4 explained 36.21, 20.48, 15.85 and 10.91 % of total variation, respectively. High and positive loadings were observed for GYP, PWT, DF, and CT with PC1, whereas DMW and PHT were associated with PC2, and PLT and CT associated with PC3. SPAD recorded high and positive loadings with PC4. High and negative loadings were computed for TGW and SPAD with PC1 and HI with PC2. DF recorded high and negative loading with PC3. Two PC's accounting for 96.35 % of total variation for drought tolerance indices were noted. PC1 explained to 65.02 % and PC2 to 31.33 % of total variation. High and positive loadings were observed for GMP, HM, YI, Y_s, STI, MP and Y_p with PC1, and TOL, SSI and Y_p with PC2. High and negative loading was observed for YSI with PC2.

6.7. Principal component biplot analysis

The biplots of PC1 vs PC2 showing groupings of test lines for agronomic traits under drought stressed and non-stressed conditions are shown in Fig. 6. Also, relationships between drought tolerance indices

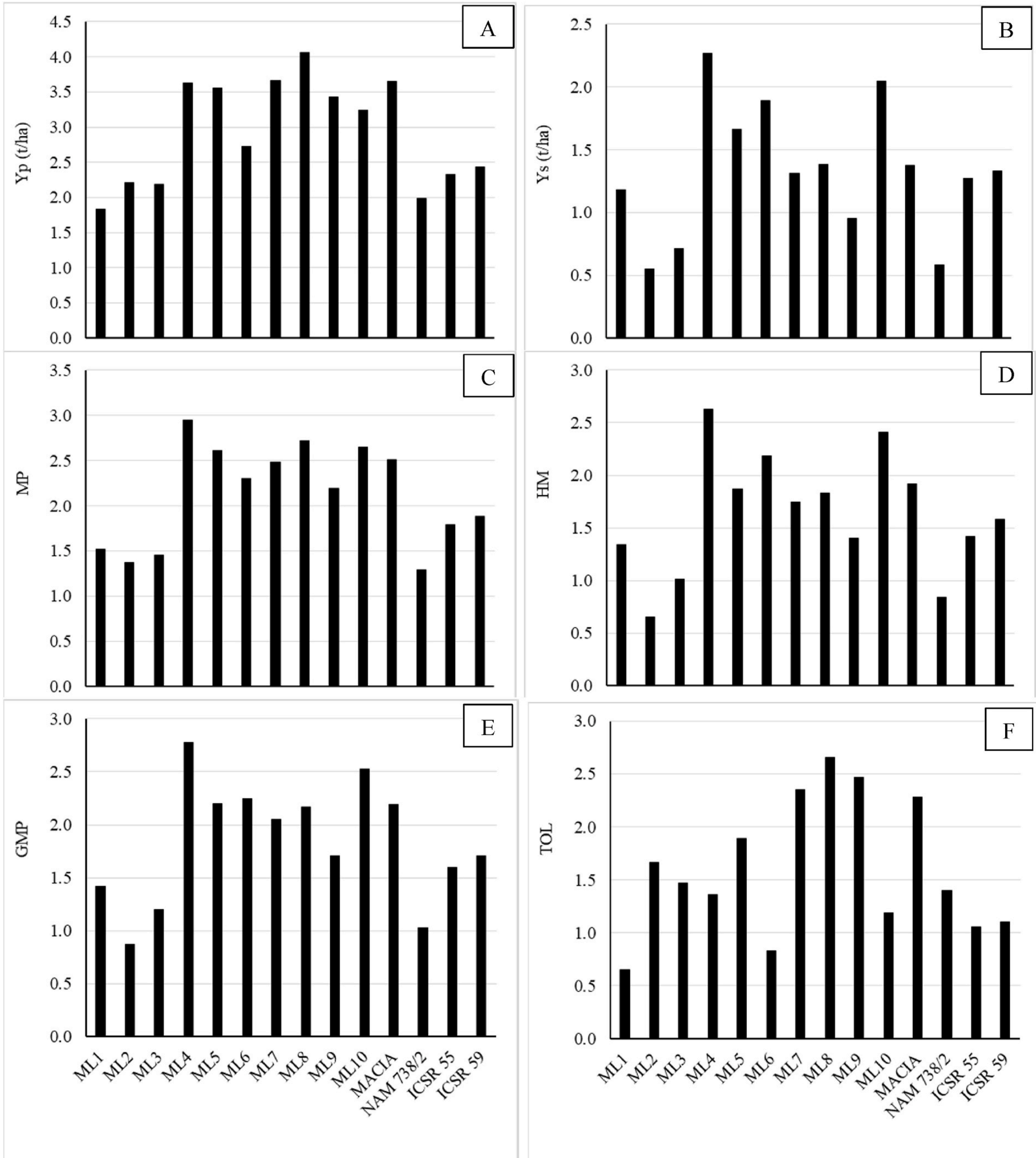

Fig. 4. Mean values for drought tolerance indices based on assessment of 14 sorghum lines evaluated across two seasons (SN1 and SN2) under drought-stressed (DS) and non-stressed (NS) conditions. Yp, mean genotype yield under non-stressed condition (A); Ys, mean genotype yield under drought-stressed condition (B); MP = mean productivity (C); HM = harmonic mean (D); GMP = geometric mean productivity (E); TOL = tolerance index (F).

with test lines are presented in Fig. 7. Average performing genotypes are plotted closer to the centre of the PC biplot, whereas most influential agronomic traits and drought tolerance indices were plotted furthest shown by the longest vector line. The smallest angles dimension in the same direction indicating strong and positive correlation was shown for GYP, PWT and PLT (Fig. 6). The largest obtuse angle indicating negative correlation was observed between GYP and DF under DS condition. Under NS condition, the smallest angles dimension in the same direction indicating strong and positive correlation was shown for DF and CT, and between GYP and PWT (Fig. 6). The largest obtuse angle indicating negative correlation were observed for GYP with PHT, PLT and SPAD were observed under NS condition. The smallest angles dimension in the same direction indicating strong and positive correlation was shown by GMP, HM, YI, Y_S, STI, and MP (Fig. 7). Vectors in opposite direction indicating high negative correlations was observed between SSI and YSI. Lines ML4, ML10 and ML5 were plotted in closer proximity of vectors for Y_s and YI indicating high grain yield performance under DS condition. Also, these mutants exhibited relationships with HM, GMP, STI and MP. Lines ML8, ML7 and check variety Macia were plotted in closer proximity of the vectors for Y_p and TOL indicating their high grain yield performance under NS condition. Mutant line ML6 was plotted in closer proximity of vectors for YSI which was negatively correlated with SSI and TOL.

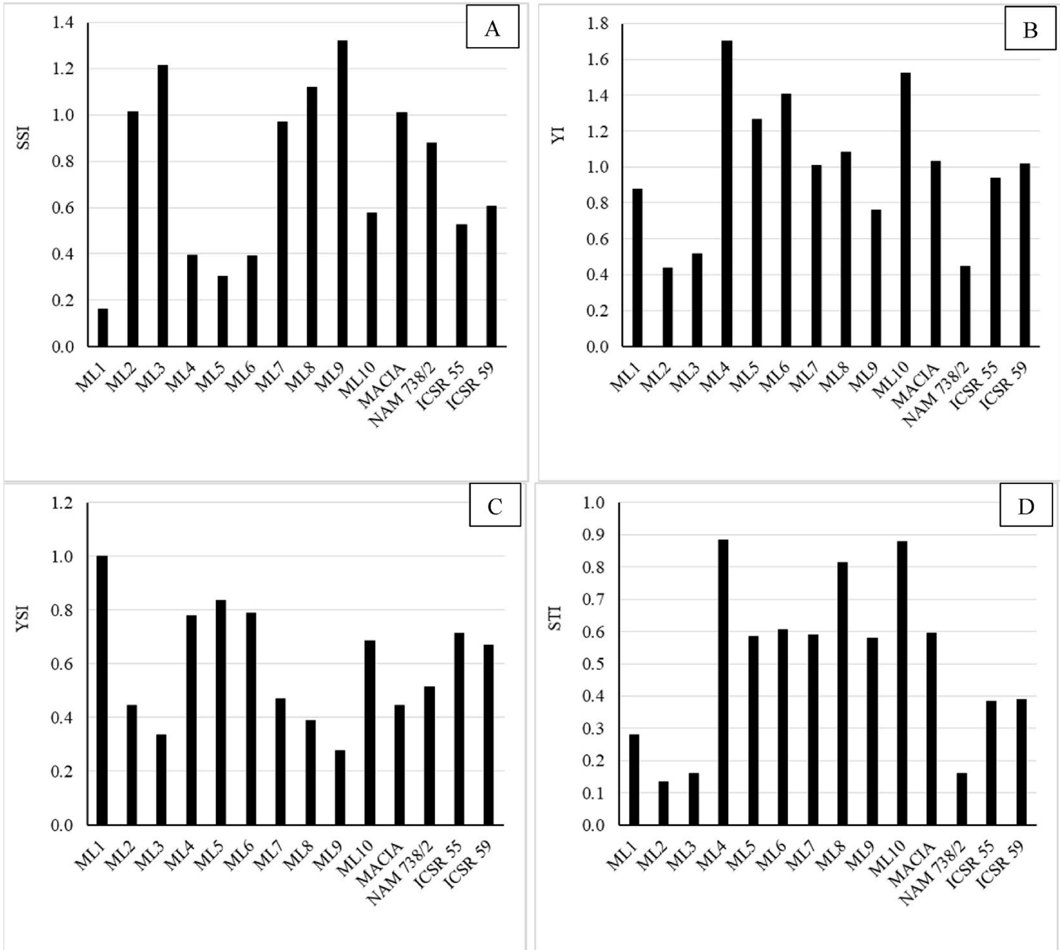

Fig. 5. Mean values for drought tolerance indices based on assessment of 14 sorghum lines evaluated across two seasons (SN1 and SN2) under drought-stressed (DS) and non-stressed (NS) conditions. SSI = stress susceptibility index (A); YI = yield index (B); YSI = yield stability index (C); STI = stress tolerance index (D).

7. Discussion

In Namibia sorghum exhibits a narrow genetic diversity which limited development of high-performing and drought-adapted genotypes possessing desirable agronomic traits such as grain yield and drought-adaptive traits for cultivation in water-stressed environments. Mutation breeding provides opportunities to induce genetic variation to aid selection of mutant lines with economic traits such as tolerance to drought and heat stress. The present study employed mutation breeding using gamma radiation to induce genetic variation for yield, yield-related traits and drought tolerance to identify and select novel mutant lines that combine high grain yield potential and drought tolerance for selection and subsequent release.

The study found considerable genotypic variation among the assessed lines for key agronomic traits including grain yield and drought tolerance (Table 3). Grain yield response of test genotypes varied from 0.55 to 2.27 t/ha under DS and 1.84 to 4.05 t/ha in NS conditions (Figs. 2 and 3). The following new lines: ML4, ML10, ML6 and ML5 were high yielding (>1.64 t/ha) compared to the parental variety Macia (1.38 t/ha) under drought stress condition (Figs. 4 and 5). Further, the lines were drought tolerant exhibiting low values of tolerance and susceptibility indices suggesting their higher yield potential under drought stress condition (Figs. 4 and 5). Thus, the four mutant lines (i.e., ML4, ML10, ML6 and ML5) are recommended for further multiple environment evaluation under drought-stress condition for variety registration and release. Contrastingly, mutant lines ML2 and ML3 were poor yield performers (≤0.72 t/ha) and drought sensitive (Figs. 4 and 5). The yield levels of the two lines were comparatively lower than that of Macia, ICSR 55 and ICSR 59. Therefore, these mutant lines were not ideal for further selection.

Trait-based selection targeting secondary yield-improving traits is useful to develop superior sorghum genotypes suited for cultivation in water-limited conditions. Early flowering is an important trait associated with drought escape where terminal drought is prevalent such as in many parts of SSA including Namibia [7]. Lines ML4 and ML6 were identified to be early flowering and high yielders. These lines could serve as sources of useful alleles for improving drought tolerance in sorghum. Conversely, lines ML1 and ML9 were identified as early flowering types albeit poor grain yield under water-limited conditions (Figs. 2 and 3).

In the present study, days to flowering significantly and negatively correlated with key agronomic traits including plant height, panicle length, panicle weight, harvest index and grain yield (Figs. 2 and 3). Reportedly, earliness has yield penalty due to reduced photo-assimilate production and hence affecting yield and yield-related traits [34]. Abdallah et al. [35] reported negative correlation between days to flowering with grain yield ($r = -0.67$; $p \leq 0.001$) agreeing with the present findings (r = -0.35). The authors also reported negative correlations between days to flowering with plant height (−0.33), number of grains per panicle (−0.52) and harvest index (−0.85). The present findings suggest that selection of early flowering lines have not compromised the expression of yield and yield-related traits, indicating the usefulness of the selected lines for water-restricted environments.

Panicle length is a key agronomic attribute that influence grain yield

Table 5
Pearson's correlation coefficients showing pair-wise traits association among 14 sorghum lines evaluated in two seasons (SN1 and SN2) under drought-stressed (below diagonal) and non-stressed (above diagonal) conditions.

Traits	DF (days)	PHT (cm)	PLT (cm)	PWT (g/panicle)	TGW (g/1000-seed)	GYH (t/ha)	HI (%)	DMW (g/plant)	SPAD value	CT (°C)
DF	–	−0.384 **	−0.342 **	0.017 ns	−0.218 *	0.003 ns	−0.131 ns	0.203 ns	0.050 ns	−0.005 ns
PHT	−0.548 **	–	0.467 **	−0.025 ns	−0.139 ns	−0.092 ns	−0.123 ns	0.115 ns	0.129 ns	0.114 ns
PLT	−0.412 **	0.522 **	–	0.174 ns	0.041 ns	0.130 ns	−0.005 ns	0.196 ns	0.029 ns	−0.019 ns
PWT	−0.326 **	0.258 *	0.407 **	–	0.294 **	0.782 **	0.302 **	0.528 **	−0.345 **	0.027 ns
TGW	−0.014 ns	−0.190 ns	0.028 ns	0.170 ns	–	0.293 **	0.378 **	−0.083 ns	−0.465 **	−0.052 ns
GYP	−0.349 **	0.213 ns	0.365 **	0.749 **	0.125 ns	–	0.659 **	0.39 **	−0.379 **	−0.108 ns
HI	−0.258 *	−0.095 ns	0.219 *	0.448 **	0.226 *	0.788 **	–	−0.302 **	−0.333 **	−0.028 ns
DMW	−0.062 ns	0.400 **	0.182 ns	0.320 **	−0.183 ns	0.163 ns	−0.318 **	–	0.051 ns	−0.082 ns
SPAD	0.032 ns	0.053 ns	0.043 ns	0.082 ns	−0.105 ns	0.092 ns	−0.032 ns	0.434 **	–	0.189 ns
CT	−0.073 ns	0.002 ns	−0.076 ns	−0.117 ns	0.113 ns	−0.124 ns	0.029 ns	−0.404 **	−0.401 **	–

Note: DF, days to 50 % flowering; PHT, plant height; CT, canopy temperature; SPAD, SPAD value; PLT, panicle length; PWT, panicle weight; TGW, 1000 grain weight; GYH, grain yield (t/ha); HI, harvest index; DMW, above ground dry mater weight; * significant at p ≤ 0.05; ** significant at p ≤ 0.001; ns, non-significant difference.

in sorghum [24,36]. In the present study, panicle length had a low and positive correlation with panicle weight and grain yield. This suggested that increased panicle length could directly improve panicle weight and grain yield under water-limited conditions. Tovignan et al. [37] found that larger panicle size would contribute to high carbohydrates remobilization from the stem to panicle to attain maximum grain filing. In the present study lines ML4, ML10 and ML6 exhibited longer and heavier panicle and higher grain yield than the parental variety Macia under drought-stressed condition. The panicle weight consists of a main rachis on which many primary branches develop bearing secondary and tertiary branches at which many spikelets are produced for ultimate grain production. In the present study, grain yield was highly and positively correlated with panicle weight and harvest index under drought-stressed and non-stressed conditions (Table 4). Panicle weight was highly correlated with harvest index under drought stress condition, implying that increased panicle weight and harvest index can be selected simultaneously to improve grain yield for water limited conditions. Bordoloi et al. [38] report similar correlation between grain yields with harvest index, panicle weight and panicle length agreeing with the present study. High yielding lines such as ML4, ML6 and ML10 had high panicle weight (>28 g/panicle) under drought stress condition (Figs. 2 and 3). Panicle length was poorly correlated with harvest index that may render poor selection response based on the two traits (Figs. 4 and 5). Thus, future selection and genetic advancement of lines after mutagenesis using gamma-radiation need to improve panicle length and grain weight to achieve improved grain yield. Targeting long panicle with grain weight gains during early generation of selections and line development stages could improve harvest index under water-limited conditions. Lines ML4 and ML10 which recorded higher harvest index under drought stress condition (Figs. 2 and 3) could be useful germplasm for further selection and genetic advancement to improve this trait.

Biomass production is a useful attribute in enhancing maximum photosynthesis and other key physiological and biochemical processes and for attaining grain yield potential in sorghum. In the present study, dry matter weight had a low and positive correlation with panicle weight and a low negative correlation with harvest index (Table 5). This suggested that increased harvest index in the mutant lines could directly improve grain yield under water-limited conditions. Biomass production showed non-significant relationship with grain weight under water limited condition compared to positive correlation recorded under non-stressed condition (Table 5). This implied that drought stress affected the translocation of photo-assimilates accumulated in the shoot biomass to promote seed set and grain filling in test lines. Van Oosterom and Hammer [39] reported that the proportion of shoot biomass allocated to panicle weight is genotype-dependent. This was observed in the present study where lines ML9, ML1 and ML3 with low biomass production were among poor grain yielders under drought-stressed condition (Figs. 2 and 3). Also, sorghum line ML2 with high biomass production was among poor yielder under drought-stressed condition. Therefore, targeting enhanced shoot biomass production combined with higher harvest index among the test lines could enhance grain yield and drought tolerance. Lines such as ML4, ML6 and ML10 had high biomass production and grain yield than the parental genotype Macia under water-limited condition (Figs. 4 and 5). Thus, these lines are recommended for breeding purposes targeting high shoot biomass production and high grain yield potential for water-limited environments.

Thousand-grain weight is an important yield attribute for attaining maximum yield potential in sorghum. In the present study, 1000-grain weight had positive correlation with panicle weight and grain yield (Table 5). This suggested that increased 1000-grain weight could directly improve grain yield under water-limited conditions. PC biplot revealed strong and positive correlation for 1000-grain weight with SPAD value and biomass weight. This suggests that simultaneous selection of these traits could improve yield under water-limited condition (Fig. 2). Girma et al. [40] reported strong and positive correlation of 1000-grain weight with grain filling rate and panicle weight attributed

Table 6
Pearson's correlation coefficients showing pair-wise association of drought tolerance indices based on 14 sorghum lines evaluated in two seasons under drought-stressed and non-stressed conditions.

Index	Y_p	Y_s	MP	HM	GMP	TOL	SSI	YI	YSI	STI
Yp	–									
Ys	0.267 *	–								
MP	0.917 **	0.629 **	–							
HM	0.602 **	0.873 **	0.847 **	–						
GMP	0.753 **	0.804 **	0.941 **	0.976 **	–					
TOL	0.867 **	−0.249 *	0.596 **	0.154	0.342 **	–				
SSI	0.489 **	−0.523 **	0.178	−0.089	0.014	0.761 **	–			
YI	0.267 *	1.000 **	0.629 **	0.873 **	0.804 **	−0.249	−0.523 **	–		
YSI	−0.489 **	0.523 **	−0.178	0.089	−0.014	−0.761 **	−1.000 **	0.523 **	–	
STI	0.745 **	0.756 **	0.914 **	0.945 **	0.966 **	0.358 **	0.046	0.756 **	−0.046	–

Note: Y_p, mean genotype yield under non-stressed condition; Y_s, mean genotype yield under drought-stressed condition; MP = mean productivity; HM = harmonic mean; GMP = geometric mean productivity; TOL = tolerance index; SSI = stress susceptibility index; YI = yield index; YSI = yield stability index; STI = stress tolerance index; * significant at $p \le 0.05$; ** significant at $p \le 0.001$.

Table 7
Rotated principal components showing loading scores, explained and cumulative variations for 10 agronomic traits computed from 14 sorghum lines evaluated across two seasons (SN1 and SN2) under drought-stressed and non-stressed conditions.

Trait	Drought-stressed			Non-stressed			
	PC1	PC2	PC3	PC1	PC2	PC3	PC4
DF	**−0.654**	−0.169	−0.347	**0.705**	0.224	**−0.575**	−0.227
PHT	0.284	0.492	**0.764**	−0.407	**0.604**	0.198	0.480
PLT	0.290	0.175	**0.624**	−0.414	0.422	**0.637**	0.132
PWT	**0.750**	**0.509**	−0.232	**0.814**	−0.067	0.247	0.379
TGW	−0.478	0.283	0.028	**−0.547**	−0.421	0.222	0.030
GYP	**0.778**	0.470	−0.333	**0.824**	−0.261	0.078	0.462
HI	**0.813**	0.120	−0.448	0.261	**−0.864**	0.156	0.229
DMW	−0.473	**0.731**	0.172	**0.562**	**0.648**	−0.204	0.221
SPAD	**−0.538**	**0.641**	−0.119	**−0.528**	−0.075	**−0.586**	**0.521**
CT	0.488	**−0.657**	0.415	**0.695**	0.178	0.532	−0.246
Eigen value	3.40	2.25	1.68	3.62	2.05	1.59	1.09
Explained variance (%)	34.02	22.53	16.75	36.21	20.48	15.85	10.91
Cumulative variance (%)	34.02	56.55	73.30	36.21	56.70	72.54	83.46

DF, days to flowering; PHT, plant height; CT, canopy temperature; SPAD, SPAD value; DMW, above ground dry mater weight; PLT, panicle length; PWT, panicle weight; TGW, 1000 grain weight; GYP, grain weight/plant; HI, harvest index; PC, principal component; Bold font values within a column denote significant scores correlated with PC.

Table 8
Rotated principal components showing loading scores, explained and cumulative variations for drought tolerance indices computed from 14 sorghum lines evaluated across two seasons (SN1 and SN2) under drought-stressed and non-stressed conditions.

Trait	Drought tolerance indices		
	Index	PC1	PC2
DF	Yp	**0.741**	**0.646**
PHT	Ys	**0.965**	−0.238
PLT	MP	**0.944**	0.320
PWT	HM	**0.985**	−0.050
TGW	GMP	**0.992**	0.083
GYP	TOL	0.084	**0.951**
HI	SSI	−0.424	**0.871**
DMW	YI	**0.974**	−0.202
SPAD	YSI	0.384	**−0.882**
CT	STI	**0.946**	0.255
Eigen value	Eigen value	6.50	3.13
Explained variance (%)	Explained variance (%)	65.02	31.33
Cumulative variance (%)	Cumulative variance (%)	65.02	96.35

Y_p, mean genotype yield under non-stressed condition; Y_s, mean genotype yield under drought-stressed condition; MP = mean productivity; HM = harmonic mean; GMP = geometric mean productivity; TOL = tolerance index; SSI = stress susceptibility index; YI = yield index; YSI = yield stability index; STI = stress tolerance index; PC, principal component; Bold font values within a column denote significant scores correlated with PC.

to stay-green traits observed in drought tolerant genotypes. In the present study, mutant lines ML5 and ML6 were identified as sources of useful alleles for high 1000-grain weight combined with high biomass and high grain yielders for breeding targeting. Harvest index is also an important determinant of the net drought stress effects on key physiological and biochemical processes. Phenotypic expression of other traits including plant height and panicle weight are direct related with HI [41]. In the present study, HI had strong and positive correlation with grain yield. This HI aid selection of drought tolerant sorghum mutant lines for moisture stressed conditions. Hadebe et al. [13] reported varying harvest index responses ranging between 0.46 and 0.63 to rainfall amount received ranging between 226- and 500-mm. In the present study, lines ML4 and ML10 were identified as sources of useful alleles for high harvest index (>22.23 %) combined with high grain yield (1.89 t/ha) for breeding targeting water-limited conditions.

High chlorophyll content associated with green leaves production under water-limited conditions would allow plants to have a prolonged and active photosynthesis for grain filling and yield development. High SPAD value is correlated with chlorophyll content and stay-green trait in sorghum [42]. In the present study, chlorophyll content showed a positive correlation with shoot biomass signalling that biomass affects photosynthetic capacity under water-limited condition (Table 6). Lines ML6 and ML10 are useful selections for green biomass production and high grain yield potential for breeding targeting moisture stress areas. Lower leaf canopy temperature in relation to ambient air temperature is

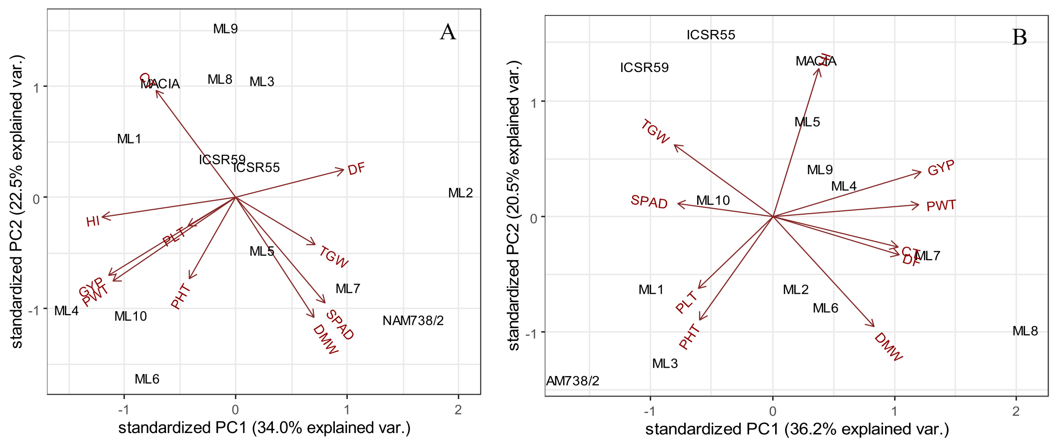

Fig. 6. Biplot showing grouping of 14 sorghum lines under drought-stressed (A) and non-stressed (B) conditions. PC, principal component; DF, days to flowering; PHT, plant height; DMW, above ground dry mater weight; PLT, panicle length; PWT, panicle weight; TGW, 1000 grain weight; GYP, grain weight/plant; HI, harvest index; SPAD, SPAD value; CT, leaf canopy temperature; red line denote vectors; red fonts denote agronomic traits.

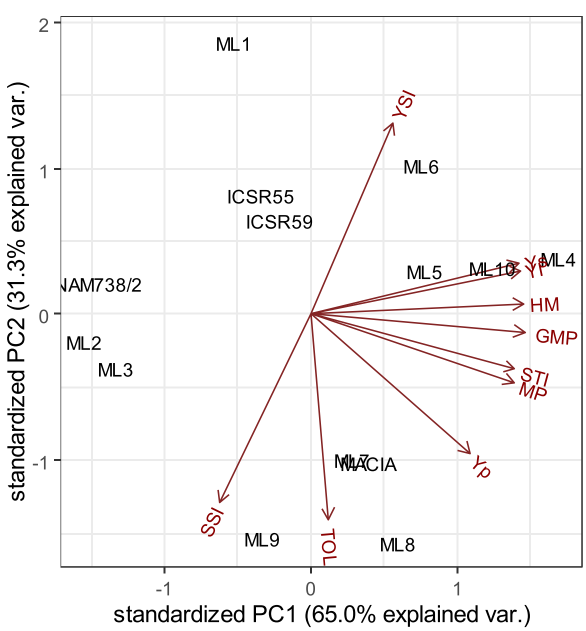

Fig. 7. Biplot showing grouping of 14 sorghum lines under drought-stressed, non-stressed conditions and drought tolerance indices. PC, principal component; Y_p, mean genotype yield under non-stressed condition; Y_s, mean genotype yield under drought-stressed condition; MP, mean productivity; HM, harmonic mean; GMP, geometric mean productivity; TOL, tolerance index; SSI, stress susceptibility index; YI, yield index; YSI, yield stability index; STI, stress tolerance index; red line denote vectors; red fonts denote drought tolerance indices.

associated with drought adaption which indicates that transpiration is maintained under water-limited condition [41]. Cooler canopy temperature under water-limited condition indicates relates to effective use of the available soil moisture for photosynthesis and transpiration to avoid excessive dehydration [43]. In the present study, a negative relationship was detected between canopy temperature and above-ground biomass in drought-stressed conditions (Table 5) suggested that high biomass yielders may have better water use efficiency by maintaining key physiological and biochemical processes under water-limited conditions. Canopy temperature negatively correlated with SPAD indicating that lines with enhanced chlorophyll formation under water-limited conditions are drought-tolerant. Higher grain yielder lines such as ML6 and ML10 were identified as sources of useful alleles for high chlorophyll production with low canopy temperatures. The selected mutant lines are recommended for breeding of drought-adapted genotypes combining desirable agronomic traits for cultivation in dry-land agro-ecologies in Namibia or related agro-ecologies.

8. Conclusions

Mutation breeding using gamma-radiation aided the development of 10 promising sorghum mutant lines suited for cultivation in water-limited conditions. Four mutant lines including ML4, ML10, ML6 and ML5 were selected with high yields (>1.64 t/ha) compared to the parental variety Macia (1.38 t/ha) under drought stress condition. These are recommended for cultivation in water-limited agro-ecologies in Namibia or similar regions or for breeding targeting high grain yield potential and drought tolerance. Also, the selected lines are sources of secondary yield-improving traits including short plant height, early flowering, high shoot biomass production, enhanced panicle weight, high harvest index, high chlorophyll production, and cooler leaf canopy temperatures.

CRediT authorship contribution statement

Maliata Athon Wanga: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Software, Visualization, Writing – original draft, Writing – review & editing. **Hussein Shimelis:** Conceptualization, Writing – review & editing, Data curation, Formal analysis, Funding acquisition, Resources, Validation, Visualization, Writing – original draft. **Jacob Mashilo:** Conceptualization, Data curation, Validation, Visualization, Writing – review & editing. **Lydia N. Horn:** Writing – review & editing. **Fatma Sarsu:** Writing – original draft, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.repbre.2023.10.005.

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