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Advancing while identifying constructive lines from West African pearl millet accessions for future hybrid breeding

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ABSTRACT: The identification of parental genetic material that will provide crosses with high heterosis for grain yield and its stability is the most important factor in hybrid millet breeding in the arid zones of Asia and Africa. Self-pollination method along with directional selection was used to develop advanced lines from four improved varieties and landraces: Gamoji, HKB, HKP-GMS, and Moro-R. A total of 131 progenies of S_5 to S_{11} generation were screened along four years under natural downy mildew infestation in Niger. The screening activities occurred in the course of rainy seasons 2021 at Bengou and Tara. In 2022, one treatments' set (set-1) was evaluated at Bengou and Tara while the second set was tested at Bengou and Kalapaté. Field activities were carried out at Bengou and N'Dounga in 2023; and lastly in 2024 at the four mentioned experimental stations. Entries were laid in RCBD in three replicates regardless the year and the location. There was significant difference between tested treatments for the numbers of days to blooming in the face of their genetic level guarantying hence a selection of good nicking hybrid parents. Forty seven (47) lines including 13 with null disease incidence, 26 highly resistant, and eight resistant yielded as high as the average mean according the experiments. Although the Gam-B lines' cluster revealed the earliest in general, issued individual plants could play the role of female parents in the development of the future hybrids. Indeed, the group was the shortest in matter of plant height while presenting the maximum grain yield and inherited best resistance to the most disastrous disease of pearl millet. Moro-R offspring would be source of male parents for its overall greatest plant height. Favorable HKP-GMS descendants will be taken as female parents to cross to Moro-R side whereas approving others will be crossed onto the female Gam-B. The variety HKB, a key component in our breeding program should be improved through reciprocal recurrent selection prior to its exploitation in hybrid development.

KEYWORDS: pearl millet, hybrid breeding, lines, cluster, inbreeding, genetic admixture, standard deviation, downy mildew, disease resistance, trait, correlation.

1 INTRODUCTION

Pearl millet [*Pennisetum glaucum* (L.) R. Br] is the next-generation crop holding the potential of nutritional richness and the climate resilience. Being a climate-resilient crop is important to minimize the adverse effects of climate change. The millet plant produces tillers at staggered developmental stages. This ability prevents the plant from complete lack of yield after an intra-cropping stress like drought. That is, when some tillers are at the vulnerable stage, others are at vegetative stage. In summary, the plant can provide reliable crop where other cereals cannot (Bidinger and Hash, 2003; Prem *et al.,* 2006; Srivastava *et al*., 2022). Pearl millet as well has the potential to increase income and food security of farming communities in arid regions (Serba *et al*., 2017; Satyavathi *et al*., 2021; Yadav *et al*., 2021; Gaoh *et al*., 2023). Nonetheless, the Sahel Africa is currently facing an increasing food deficit problem because of insignificant yield increases of pearl millet. Indeed, breeding effort in Sahel was focused on developing OPVs mostly using mass selection, a method which is inappropriate for yield enhancement (Niangado and Ouendeba, 1987; Falconer and Mackay, 1996, Rattunde *et al*., 1997; De Rouw, 2004; Kanfany *et al*., 2020). It is thus crucial that productivity of this key crop is increased to ensure at least the minimal food security. Other approaches to address the yield limitation barrier should be explored. One challenge to developing the commercial potential of pearl millet is the capacity of growers to provide a consistent product that meets market standards. Pearl millet varieties with high yield and disease resistance are an integral part of meeting this challenge. According to Choudhary *et al*. (2023) hybrid development in pearl millet is of paramount importance, especially in the context of increasing food demand and the need for climate-resilient crops. Ausiku *et al.* (2022) found pearl millet hybrid to have substantially higher water use efficiency characteristics for grain yield compared to improved varieties and landraces. It offers the promise of enhancing the yield potential, nutritional quality, resistance breeding and overall agronomic performance. Hybrid breeding is therefore one of the most effective approaches for maximizing the potential of the crop and the development of high yielding hybrids is the major target of pearl millet researchers globally (Devos *et al*., 2006; Kapila *et al*., 2008; Wilson *et al*., 2008; Ambawat *et al*., 2020). India subcontinent is an illustration where hybridization has become a focused approach of all the pearl millet breeders due to the adaptive and yield advantage of hybrids (Rakshith *et al.,* 2023). Although Africa is the top pearl millet producer in the world in terms of area occupied, farmers have not benefited from the exploitation of hybrid potential. There is no millet hybrid grown on commercial scale anywhere in Africa while a range of improved OPVs and CMS sources are available for long. The challenge is to develop hybrids for the Sub-Sahara continent (Pucher *et al*., 2018; Sattler *et al*., 2019; République du Niger, 2021; Sharma *et al.,* 2023; Papanna *et al.,* 2024). Being a highly cross-pollinated crop pearl millet offers huge amount of genetic variation for traits that are constitutive and responsive to multiple breeding objectives. Therefore, there is great potential to improve the cereal production through hybrid breeding. The aim of the breeder of cross-pollinated crops is to increase the amount of heterosis over the level already found in random mating populations. This can be achieved by crossing two selected genetically different populations. In this regard, INRAN in charge of agricultural research programs in Niger has developed manually 16 variety-cross hybrids from West African germplasm belonging to two putative heterotic groups. The highest yielding hybrid, Gamoji x HKB showed also the best SCA for grain yield of 163%. It was developed from parents who had high and positive GCA for grain yield. Crosses that involve two good general combiner parents should be promoted to generate the desired transgressive segregants. In contrast Moro and HKP-GMS had negative GCA for grain yield. But the cross between Moro and HKP-GMS showed high and positive SCA (155%) for this trait. For the other possible crosses, Gamoji x Moro at our knowledge is not explored so far while Gamoji x HKP-GMS and Moro x HKB showed higher and subordinate yield compared to the average mean, respectively (Srivastava, 2022; Gajjar et al., 2023; Rasitha *et al*., 2023; Rakshith *et al.,* 2023; Issaka *et al*., 2024a). Since the amount of heterosis depends on the levels of homozygosity in the parents, the type of reproduction, and the genetic distance between the parents, its effect can be increased further if specific inbred lines are developed and crossed. Indeed, inbred lines provide maximum heterosis effects when they are fixed at loci involved in the hybrid reaction. Thus, a hybrid breeding program starts with the production of inbred lines (Melchinger *et al*., 2007; Sattler and Haussmann, 2020). Still, for a large scale production of hybrid seed, an efficient pollination through the use of synchronous flowering parents - the pollinator should preferably flower before the female parent - will be necessary. In this regard, the direction of the cross Moro x HKP-GMS should be changed. Indeed, the female parent which is Moro flowered before the male parent HKP-GMS. The change is further of worth value because the female parent yielded less than HKP-GMS. Whereas, in seed production it is economically important to use the higher yielding parent as the female to increase the production per unit area (Andrews *et al*., 1997; Talukdar *et al*., 1999; Khair and Bhanvadia, 2019). Besides synchronous flowering, plant height is another important factor in the choice of hybrid parents as the height of the male parent should be greater than that of the female. Taller male parent allows for free flow of pollen across the rows and thus cuts down the seed production cost by giving better seed set, and consequently higher hybrid seed yield per unit area. This allows as well planting greater female/male ratio, which also leads to higher hybrid seed yield (Prem *et al*., 2006; Papanna *et al.,* 2024). Else, a specific character that is usually sought in pollinators is high tillering capacity which contributes to abundant and continued pollen shed (Andrews *et al*., 1997; Talukdar *et al*., 1999). Otherwise, pearl millet germplasm of any genetic formula is subjected to attack from the obligate biotrophic oomycete parasite, *Sclerospora graminicola* (Sacc.) Schroet. Yet, yield stability is a major objective in breeding cultivars because in pearl millet producing areas of Asia and Africa farmers are largely dependent on the crop as their main source of staple food grain (Bollam *et al*., 2018; République du Niger, 2020). In Africa, the initial F_1 hybrids developed from A_1 and A_2 CMS systems were completely destroyed by downy mildew and this discouraged hybrid breeding on the continent (Niangado and Ouendeba, 1987). It should be noted the 16 variety-cross hybrids developed showed a similar disease incidence with their parents, that is, the inheritance of downy mildew resistance in pearl millet is here additive. For the reason, both hybrid parents shall carry the resistant gene for the offspring to struggle against the oomycete. The objective of the current research activity was to produce inbred lines by pedigree method while identifying constructive ones to develop hybrids in the close future.

2 MATERIAL AND METHODS

Advanced lines developed from four OPVs (Table 1) belonging to two putative heterotic groups were used in this study. The eastern Niger group is characterized by ecotypes with short panicle having large diameter and includes landraces, namely Gamoji and Moro. Yet, it should be noted that Moro is of thin panicle and has further the highest content in iron and zinc among the whole germplasm. HKB and HKP-GMS belong to the western Niger heterotic group. This cluster covers genotypes with long and thin panicles. HKP-GMS is the most widely grown variety in Niger due to its large adaptation, acceptable grain yield, and short vegetative cycle, but it is afterward susceptible to downy mildew attack. In terms of head compactness, Moro is naturally the best throughout the country and may belong also to the 2nd heterotic group due to its thin panicle. On the opposite side Gamoji and HKB had the largest panicle girth and thus the loosest spikes while HKP-GMS is of intermediate caliber. The western cluster was developed through mass selection from local landraces. Gamoji was successfully converted into A_4 CMS carrier while Moro and HKP-GMS proved to encompass good number of restorer plants over the same sterility system. Among the checks, HKP of 80-85 days to maturity was obtained by pedigree mass selection from the local land race HK in 1967 following drought years. HKP-GMS was developed by ICRISAT from HKP through gridded mass selection – "GMS".

HKP, the widest grown variety in Niger like its relative HKP-GMS, showed to be the most susceptible to mildew attack amid the country pearl millet diversity. On the contrary, SoSaT-C88 with around 60 days for DFL is known for its high resistance to the disease and subsequently its good grain yielding capacity. Really, it is one of if not the highest yielding cultivar in West Africa. Original Zatib, a variety-cross hybrid is of medium maturity cycle of 90-95 days with midway behavior in matter of disease resistance.

Table 1. List of lines' parents and some of their interesting characteristics

Parents	DMI	DFL	SPL	SPD	SPN	GCA$_{GY}$	Hybrids	SCA$_{GY}$
Gamoji	7	63	43	15	60	37	Gamoji x HKB	163%
HKB	8	56	62	12	59	40		
Moro	10	57	43	11	66	-72	Moro x HKP-GMS	155%
HKP-GMS	16	60	54	12	70	-3		

Source: Issaka A., 2012

The line breeding process began in the off season of October to January 2014 and continuously run so far. Sometimes we advanced lines twice in the long off season and once during screening operation in the normal campaign. To produce inbred lines, main shoots of concerned genotypes were covered with glassine bags after exertion of the tip apex. During the evaluation in the normal season, only plants free from mildew attack are each time selected to constitute the next generation of self-fertilization. The selection of promising lines in hotspots was also based on other phenotypic traits such as the tillering propensity, the plant height, and especially the characteristics of the ear. Bagged diseased plants which were rated as healthy before were rejected at maturity. At harvest time spikes were labeled and threshed individually thereafter, and grains were dried under natural ventilation in a laboratory for the cycle to start again. Additional selection traits encompassed the grain characteristics such as its size, color, uniformity, and the quantity of seed available to cover the necessary number of trials. Once reaching the fifth generation of inbreeding in 2021, 22 lines were selected among hundreds developed for evaluation and data field record. The number and types of checks added varied according to the tests before being focused on the main appropriate ones during the most advanced assessment. Trials were implemented in Bengou (11° 58’ 44, 4” N; 3° 33’ 25, 6” E) and Tara (11° 90’ 357” N; 3° 30’ 25, 6” E) under natural disease infestation. Both experimental stations are located in the rainiest (rainfall in 2021 was 796.9 mm in 61 days and 962.4 mm in 51 days in Bengou and Tara, respectively) part of the country. Knowing the most favorable period reaction of inoculums, field assessment was conducted on July. The reason for planting at this period was seedling stage to coincide with the most regular and highest moisture time for optimal disease growth. In the following year a larger number of lines all at eighth to ninth generation of selfing were screened and the location of Kalapaté (13° 12’ N & 2° 56’ E) in the Sahelian agro zone was added to the list of testing environments. In the three sites, two sets of 25 lines different from one to another were assessed. One set consisting of progenies derived from the two parents involved in the cross Moro x HKP-GMS was appraised in Bengou and Tara. The second set composed of a mixture of offspring extracted from the two former varieties added to those issued from Gamoji, was evaluated in Bengou and Kalapaté. Only one check, SoSat-C88 was used during this evaluation. In 2023, 24 lines developed from Gamoji, thus considered as female lines were evaluated in Bengou and N’Dounga (11° 22’ N & 2° 14’ E). But trial establishment was poor in the latter environment and then discarded. Variety SOSAT-C88 was added to Gamoji, the original parent of all the Gam-B lines, as check. Finally, for the most advanced trial in 2024, thirty lines were tested in the four environments against three checks: HKP, SOSAT-C88, and Zatib. All the evaluation tests were run following the same protocol along the years. Indeed, accessions without any seed treatment were planted in a RCBD in three replications. Each plot was comprised of 20 hills arranged in two rows of 7.2 m length. Spacing between rows as well as between hills was 0.8 m. In sum the plot was 12.8 m^2. The trials were hand weeded and the crop grown as single plant per hill with recommended fertilization. Data were recorded on (i) days to flowering (DFL): number of days from sowing to the time at which 50% of plants within a plot flowered; (ii) plant height (PLH): height of the plant from the ground level to the spike tip in centimeter; (iii) main shoot diameter (MSD): main stem girth at half height in millimeter; (iv) spike length (SPL): interval between the tip of the spike and its bottom in centimeter; (v) spike diameter (SPD): spike girth at its middle distance in millimeter; (vi) number of spikes harvested (NSH): number of spikes per plot; (vii) grain yield (GY): grain weight per plot. Grain yield was measured as kg per plot, and then converted to kg per hectare; (viii) number of hills (NHH): number of hills harvested per plot; (ix) downy mildew incidence (DMI): number of diseased plants by the total one within the plot in percentage at 90 days after sowing. Pearl millet genotypes were classified according to Ball (1983) as (1) Highly Resistant (HR) for 0-5% disease incidence, (2) Resistant (R) for 5-10% disease incidence, (3) Moderately Resistant (MR) for 10-25% disease incidence, (4) Moderately Susceptible (MS) for 25-50% disease incidence, (5) Susceptible (S) for 50-80% disease incidence, and (6) Highly Susceptible (HS) for > 80% disease incidence. Data (ii), (iii), (iv), and (v) on entries were collected on three plants taken at random within each plot. Analysis of variance was carried out using GenStat to sort out the difference and correlation between variables.

3 RESULTS

3.1 CORRELATION BETWEEN VARIABLES

Although the tested entries varied from one experiment to another, there was a positive connection generally highly significant between NHH and NSH not considering the self-fertilization level. We had the same kind of association between MSD and SPD. But here the relationship was becoming more significant with increased cycles of inbreeding. Similarly, PLH was positively and sometimes significantly coupled to SPL. NSH was usually positively and highly correlated with PLH while it showed the preeminent positive correlation with GY. This link was also the highest in terms of significance, followed by that between GY and plant height. Yet, the relationship between GY and MSD as well as that between NSH and MSD was highly significant and negative in the first selfing cycles whereas it became positive with increased inbreeding. GY and PLH were always negatively correlated with DFL at significant level. The association between SPL and DFL was in general negative as well. Moreover it was weak and non significant. DFL was positively associated with MSD. This association of high level of significance became lower with advanced generations. Through all the five case studies DMI was positively and weakly linked four times to GY, NSH, and SPL against one negative correlation, three times to NHH, twice to MSD and PLH, and lastly once to SPD and DFL. The overall computation gave DMI lowly and inconsistently connected to the leftover parameters whereas most of connections were not significant.

Table 2. Correlation between variables for the different sets of entries along four years

DFL							**HKB/HKP-GMS/Moro-R in 2021**		
DMI	-0.19 ^{ns}								
GY	-0.52**	0.06 ^{ns}							
MSD	0.60****	-0.34 ^{ns}	-0.70*****						
NHH	-0.53**	0.13 ^{ns}	0.81*****	-0.50**					
NSH	-0.58***	0.10 ^{ns}	0.92*****	-0.67*****	0.91*****				
PLH	-0.26 ^{ns}	0.23 ^{ns}	0.26 ^{ns}	-0.38 ^{ns}	0.00 ^{ns}	0.19 ^{ns}			
SPD	0.02 ^{ns}	-0.29 ^{ns}	0.11 ^{ns}	0.23 ^{ns}	0.29 ^{ns}	0.10 ^{ns}	-0.77*****		
SPL	-0.38 ^{ns}	0.55***	0.10 ^{ns}	-0.34 ^{ns}	0.12 ^{ns}	0.13 ^{ns}	0.55***	-0.41*	
DMI	0.29 ^{ns}						**Set-1: Moro-R/HKP-GMS in 2022**		
GY	-0.65*****	-0.13 ^{ns}							
MSD	0.68*****	0.46*	-0.41*						
NHH	-0.35 ^{ns}	-0.12 ^{ns}	0.44*	-0.39*					
NSH	-0.59****	-0.28 ^{ns}	0.75*****	-0.54***	0.77*****				
PLH	-0.55***	-0.10 ^{ns}	0.79*****	-0.38*	0.15 ^{ns}	0.49**			
SPD	0.25 ^{ns}	-0.14 ^{ns}	-0.25 ^{ns}	0.31 ^{ns}	0.06 ^{ns}	-0.13 ^{ns}	-0.31 ^{ns}		
SPL	-0.06 ^{ns}	-0.15 ^{ns}	0.03 ^{ns}	-0.01 ^{ns}	-0.35 ^{ns}	-0.21 ^{ns}	0.25 ^{ns}	-0.45*	
DMI	-0.03 ^{ns}						**Set-2: Gam-B/Moro-R/HKP-GMS in 2022**		
GY	-0.41*	0.12 ^{ns}							
MSD	0.14 ^{ns}	-0.09 ^{ns}	-0.20 ^{ns}						
NHH	-0.30 ^{ns}	0.06 ^{ns}	0.33 ^{ns}	-0.44*					
NSH	-0.23 ^{ns}	0.15 ^{ns}	0.20 ^{ns}	-0.30 ^{ns}	0.38*				
PLH	-0.24 ^{ns}	-0.11 ^{ns}	0.36 ^{ns}	0.10 ^{ns}	0.06 ^{ns}	-0.11 ^{ns}			
SPD	0.09 ^{ns}	0.20 ^{ns}	0.06 ^{ns}	0.07 ^{ns}	-0.17 ^{ns}	-0.32 ^{ns}	0.36 ^{ns}		
SPL	-0.07 ^{ns}	0.07 ^{ns}	0.04 ^{ns}	0.16 ^{ns}	-0.38*	0.34 ^{ns}	-0.06 ^{ns}	-0.43*	
DMI	-0.36**						**Gam-B-37-30/Gam-B-11-1-14 in 2023**		
GY	-0.54****	0.09 ^{ns}							
MSD	0.22 ^{ns}	-0.27 ^{ns}	0.19 ^{ns}						
NHH	-0.53****	0.27 ^{ns}	0.60****	-0.24 ^{ns}					
NSH	-0.60****	0.30*	0.80****	0.02 ^{ns}	0.67****				
PLH	-0.50****	0.15 ^{ns}	0.63****	-0.22 ^{ns}	0.59****	0.43***			
SPD	0.34*	-0.10 ^{ns}	-0.01 ^{ns}	0.40***	0.02 ^{ns}	0.02 ^{ns}	-0.42***		
SPL	-0.08 ^{ns}	0.04 ^{ns}	0.18 ^{ns}	0.31*	-0.02 ^{ns}	0.03 ^{ns}	0.34*	-0.44***	
DMI	-0.08 ^{ns}						**Gam-B/HKP-GMS/Moro-R in 2024**		
GY	-0.39*****	0.02 ^{ns}							
MSD	0.15 ^{ns}	0.19*	0.18*						
NHH	-0.24**	-0.31*****	0.35*****	-0.46*****					
NSH	-0.48*****	0.13 ^{ns}	0.79*****	0.15 ^{ns}	0.35*****				
PLH	-0.28****	-0.16 ^{ns}	0.49*****	-0.02 ^{ns}	0.27***	0.30*****			
SPD	0.09 ^{ns}	-0.04 ^{ns}	0.30*****	0.44*****	-0.15 ^{ns}	0.15 ^{ns}	0.09 ^{ns}		
SPL	0.00 ^{ns}	0.04 ^{ns}	-0.09 ^{ns}	0.32*****	-0.06 ^{ns}	0.04 ^{ns}	0.05 ^{ns}	-0.39*****	
	DFL	**DMI**	**GY**	**MSD**	**NHH**	**NSH**	**PLH**	**SPD**	**SPL**

3.2 ANOVA OF VARIABLES

Because of highly significant correlation between NHH and NSH we dropped the variable NHH during the analysis of variance. For the first trials run in 2021, HKB lines group had the highest DMI of 25%, which is more than the double of the average mean (table 3). Consequently it illustrated one of the lowest grain yields in the middle of varietal bunches, even confirming its longest spike. Furthermore, the four HKB lines were significantly different only for two variables against the total of eight registered. Otherwise, three lines attested good resistance with a null disease incidence amongst Gam-B-37-30-2-5-2 presenting the top grain yield amid the whole entries. It had also together with the local variety the best tillering capacity. On the whole, the female Gam-B cluster recorded the peak tillering ability and the maximum grain yield. Talking about PLH, it was shorter than possible corresponding males of any other genotypes' group whereas it was of the earliest flowering.

Table 3. Mean performance of pearl millet genotypes for 8 traits across sites over 2021 to 2024

Year	Cluster	DMI	DFL	PLH	MSD	SPL	SPD	NSH	GY
2021	Gam-B	7*	61*	183*****	13*****	46^{ns}	32*****	44 ^{ns}	779*****
	HKB	25 ^{ns}	62 ^{ns}	212 ^{ns}	14 ^{ns}	56 ^{ns}	26*****	34 ^{ns}	452*
	HKP-GMS	6 ^{ns}	65**	214*****	14 ^{ns}	48*****	26 ^{ns}	26*****	393*****
	Moro-R	3**	64 ^{ns}	230 ^{ns}	14 ^{ns}	49 ^{ns}	23*	40 ^{ns}	646 ^{ns}
	Checks	13 ^{ns}	61 ^{ns}	234 ^{ns}	13 ^{ns}	55 ^{ns}	25 ^{ns}	55 ^{ns}	1124 ^{ns}
	Average	10*****	63*****	211*****	14*****	50*****	27*****	35*****	588*****
2022	Moro-R	2 ^{ns}	66 ^{ns}	221 ^{ns}	16 ^{ns}	43 ^{ns}	26 ^{ns}	42 ^{ns}	725 ^{ns}
	HKP-GMS	2 ^{ns}	68 ^{ns}	220 ^{ns}	16 ^{ns}	44 ^{ns}	26 ^{ns}	38 ^{ns}	684 ^{ns}
	Check	3	62	227	15	37	26	44	659
	Average	**2 ^{ns}**	**67 ^{ns}**	**221 ^{ns}**	**16 ^{ns}**	**43 ^{ns}**	**26 ^{ns}**	**40 ^{ns}**	**704 ^{ns}**
	Gam-B	3 ^{ns}	66**	224 ^{ns}	14 ^{ns}	43*****	25****	46 ^{ns}	763 ^{ns}
	Moro-R	6*	66*	227 ^{ns}	14 ^{ns}	48*	24***	42 ^{ns}	735 ^{ns}
	HKP-GMS	6 ^{ns}	68 ^{ns}	209 ^{ns}	14 ^{ns}	38 ^{ns}	25 ^{ns}	41 ^{ns}	612 ^{ns}
	Check	5	59	252	14	40	27	50	1058
	Average	4**	66*****	**225 ^{ns}**	14*	**48 ^{ns}**	24*****	**44 ^{ns}**	**754 ^{ns}**
2023	Gam-37-30-2	1^{ns}	72*****	174*****	15*	44*	33****	49*****	766****
	Gam-11-1-14	1****	68*****	196*****	14 ^{ns}	50*****	28*****	57*****	1050 ^{ns}
	Checks	1 ^{ns}	59 ^{ns}	232 ^{ns}	13 ^{ns}	36 ^{ns}	29 ^{ns}	75 ^{ns}	1540*
	Average	1***	70*****	187*****	15***	46*****	31*****	53*****	922*****
2024	Gam-B	4 ^{ns}	67*	191*	12 ^{ns}	49*****	19 ^{ns}	27*	551 ^{ns}
	HKP-GMS	6 ^{ns}	71 ^{ns}	228 ^{ns}	14 ^{ns}	49 ^{ns}	23***	29**	737**
	Moro-R	6***	69*	204***	14 ^{ns}	50*****	24*****	28*****	653*****
	Checks	7 ^{ns}	66 ^{ns}	215 ^{ns}	14 ^{ns}	45*****	25***	36 ^{ns}	842 ^{ns}
	Average	6****	68*****	206*****	**14 ^{ns}**	49*****	24*****	29*****	666*****

ns = non significant; * significant at 0.05; ** significant at 0.01; *** significant at 0.005; **** significant at 0.001***** significant at <0.001

According to Student-Newman-Keuls test at the 5% level, a larger sub-cluster of HKP-GMS – nine lines next to the total of eleven – thus at fifth generation of selfing and even being issued from different S_4 parents recorded together a lower amount of hills harvested, direr tillering capability, and consequently the poorest grain yield. Among the two leftover genotypes HKP-GMS-128-1-196-1-1 with long spike, high tillering ability, and subsequently good grain yield revealed promising. Nonetheless it should undergo self-fertilization process to better enhance its resistance to downy mildew. Else the difference relating lines was significant, particularly in matter of DFL, PLH, and GY inside Gam-B and HKP-GMS offspring giving thus a large choice in selecting hybrid parental material. The distance between the two most remote lines contained by cluster or the standard deviation was of (64-58) = 6, and (70-62) = 8 points for the Gam-B and HKP-GMS in that order. Whilst Gam-B is considered as female part for future hybrid development it flowered three and four days before the supposed male blocks Moro-R and HKP-GMS (table 3), respectively. The complex Moro-R was one day earlier than HKP-GMS and confirmed its thinnest spike diameter where Gam-B proved its largest one.

The HKB cluster was useless because of its lack of performance in 2021, so not considered for the 2022 research work. For the set-1 of genetic material evaluated in 2022 six lines: Moro-R-68-17-2-9-3-1, Moro-R-68-17-2-9-1-23-3, Moro-R-68-17-2-9-1-23-4, HKP-GMS-128-1-31-2-3-1, HKP-GMS-128-1-196-1-1-6 and HKP-GMS-128-1-196-4-27-1 were evidence for good grain yield and null disease incidence. In sum the mean disease incidence was smaller than that of the resistant check. What was very specific for the set-1 was lines were not different from each other within and between the two genotypes' groups for any of the eight traits. Although HKP-GMS issued lines were all of S_6 class, the Moro-R division was a mixture of S_6 and S_7 lines, but in both patterns the whole material attained similar genetic standing at the same time. Furthermore, some genotypes inside clusters were derived from different relatives. The standard deviation for DFL inside lines' groups was superior to that between groups. Moro-R and HKP-GMS were very similar for all the traits with the former group being slightly earlier.

Whilst the HKP-GMS faction going towards inbreeding depression was also excluded, two derivatives from the highly resistant HKP-GMS-R-128-1-14-2-1 were included for the 2022 trial set-1. Any statistical difference for any variable was recorded between the two lines of the seven generation of self-fertilization. The Gam-B group was enlarged with a different sub-group. Checks taken apart, the Gam-B lines kept a merely advantage of better tillering capacity and as result that of grain yield (table 3). For the essential, entries were significantly different within Gam-B and Moro-R clusters for DFL and SPD. Therefore, many Moro-R lines could be used to cross with the Gam-B germplasm. Indeed, while displaying the same DFL average mean of 66 days, Moro-R and Gam-B sets recorded a within standard deviation of ten and nine days, respectively. Here also, the variability intra-cluster was thus higher than that between clusters concerning the DFL parameter. Additionally, crosses become naturally possible between individual plants of HKP-GMS and Moro-R in one side and amid Gam-B and HKP-GMS in another side because of lower intergroup standard deviations. Really the male parental material Moro-R was slightly earlier and taller than the female one, but the future pollinator HKP-GMS was shorter than its corresponding female Gam-B. So, dwarfing gene should be backcrossed in the female background for large scale hybrid exploitation. There was no significant difference for the variables NSH and GY, may be due to the fact that spikes counting and threshing operations were made manually. Actually the related coefficients of variation were high. One female line Gam-B-37-30-2-5-8-1-2 was added to the list of genotypes with null downy mildew incidence.

In 2023, trials were implemented mainly to check whether the disease resistance of the female parent was improved and differences between the two Gam-B sub-groups exist. Lines were highly different inside clusters in regard to five traits amid the eight recorded. Nine genotypes: Gam-B-37-30-2-5-8-1-1-1, Gam-B-37-30-2-5-8-1-3-3, Gam-B-37-30-2-5-8-1-3-6, Gam-B-37-30-2-5-7B-1-4-2, Gam-B-11-1-14-32-12-1-3-6, Gam-B-11-1-14-32-12-1-2-1, Gam-B-11-1-14-32-12-1-2-4, Gam-B-11-1-14-32-4-1-1-3, and Gam-B-11-1-14-32-4-1-1-5 showed null disease incidence and three from the band were not significantly different from the known high yielding SoSat-C88 in matter of GY. At comparable values of DMI, DFL, and NSH the sub-group with the longer spike and thinner stem produce better grain yield. Again this demonstrated the positive correlation connecting grain yield and spike length observed in almost all experiment cases. The DFL variable recorded the highest standard deviation of 23 and 28 for the sub-group Gam-B-11-1-14 and Gam-B-37-30 correspondingly, compared to only four points involving the two progenies' sub-groups.

Concerning the last and most advanced trial, the Moro-R cluster showed the highest variability. It flowered two days after and before Gam-B and HKP-GMS, respectively. Except for MSD the variability inter-cluster was in general highly significant. Apart from the Gam-B and the checks ensembles, lines were significantly different within and between clusters in view of the most important trait in breeding staple cereals that is the grain yield. The inside standard deviation of DFL varied from three points, eight to 13 for HKP-GMS, Gam-B, and Moro-R clusters, respectively.

In general, the group Gam-B showed the lowest value in terms of DMI and PLH, but the highest value of GY. These qualifications constitute a strong proof of the female status that can play Gamoji's offspring. Nonetheless, the Gam-B formula had generally the earliest vegetative cycle what may hinder its role of female since in normal hybrid production the female parent should be the late flowering component indoor the couple. Still, the DFL parameter was enlarged within groups through inbreeding generations making crosses more feasible between groups or at least between individual plants of counterpart clusters. Also, the genetic material developed in the course of the research activities revealed added resistance. Twenty-two parental lines and their derivates showed a null disease incidence, but these entries were not necessary the best in matter of grain yield. All told, 64 lines of the total germplasm evaluated amongst nine with null disease incidence, 42 highly resistant and thus 13 resistant; had lower grain yield against the mean average. Fortunately 47 others yielded as high as the average mean according the experiments. These include 13 accessions which were never present of downy mildew, 26 highly resistant genotypes, and eight lines with disease incidence < 10%. Four lines: **N°1** Gam-B-37-30-1-2-1; **N°43** Moro-R-68-17-2-1-5-1-3; **N°44** Moro-R-68-17-2-1-5-1-4; and **N°131** Moro-R-68-17-2-1-5-1-1-1-27 even though moderately resistant demonstrated acceptable grain yields above the average mean. The check cluster showed the lowest value of DFL during the whole field evaluation whilst this parameter was more dynamic with line clusters under breeding procedure (table 3). MSD was the variable with a reduction of significant differences.

4 DISCUSSIONS

4.1 CORRELATION BETWEEN VARIABLES

Before starting breeding, knowledge of association among traits enables the breeders in deciding a suitable selection criterion for simultaneous genetic improvement of complex and associated traits. Grain yield was positively and significantly correlated with the number of harvested hills as well as the number of harvested spikes per plot as reported by Pucher *et al.* (2016). That is, genotypes with higher tillering capacity would be more productive. It means also grain yield could be improved by increased plant density, one reason to breed for hybrid pearl millet to cope with the growing population. In fact, OPVs do not tolerate higher plant density due to elevated interplant competition (Fasoula and Fasoula, 2002). On the contrary GY was always negatively correlated with DFL at a significant point, meaning high grain yield is related to early flowering. Similar results were reported by Serba *et al*. (2020) in contrast with the findings by Kumar *et al*. (2021). Plant height was positively and significantly linked to spike length in accord with the findings of Pucher *et al.* (2016), whom revealed SPL was sequentially also highly significantly correlated to number of grains per panicle. Nonetheless, the link between GY and the latter trait was not significant. But it was according to Serba *et al*. (2020). Still, the link between PLH and SPD was negative and significant denoting that plants with larger panicle diameter are shorter, thus favorable to be female parents. Issaka *et al.* (2024a) reported that the shortest traditional variety in Niger, namely Ankoutess had spikes with the lowest length and largest girth values amid the whole country millet diversity. So, in hybrid breeding one should be tempted to convert into CMS carrier those plants which are naturally shorter for a good hybrid seed set when crossed to taller male plants. According to Papanna *et al.* (2024) B-lines are commonly bred for shorter height (<100 cm) and larger seed size, lodging resistance, good exertion and seed set, and compact panicles. DFL was positively and significantly associated with MSD which in turn was positively connected to SPD in all the cases study. Then, plants with large panicle girth are actually supposed to be the later flowering, another argument supporting again the female nature of those genotypes because in hybrid breeding the female should flower after the male parent. Still, Ankoutess with the largest spike girth, but having the thinnest MSD in Niger pertains to the earliest flowering material. The remark here is that with Ankoutess MSD and SPD are specifically inversely connected. Conversely, taller plants and thus bearing long panicles are supposed to play the role of the male parent in breeding hybrid cultivar. This is reinforced by the fact that SPD was negatively and significantly correlated with the SPL which was positively and significantly linked to PLH. It is desirable to breed pollinators of 150–180 cm in height, but no shorter than the A-line with built-in attributes that will be preferred by farmers in the hybrids (Rasitha *et al*., 2023; Papanna *et al.,* 2024). Again positive correlation between MSD and SPD suggests certainly the male plants in pearl millet were of thin stem and panicle girth while female at the opposed side should have larger stem girth to support the hybrid seed. The relationship was becoming more significant with increased cycles of inbreeding due to unambiguous segregation of spikes towards the two types. Co-segregation of genes controlling characteristics expression might be the basis of inter-trait correlation in a positive direction. Hence, the correlation analysis suggested that by careful selection of parental alleles controlling increasing or decreasing combinations of character, one can improve the traits simultaneously (Kumar *et al.,* 2021).

NSH was all in all positively and highly correlated with PLH, the latter trait in turn positively and sometimes significantly linked to SPL would refer to the superior capacity of male parental material to develop more tillers. According to Prem *et al*. (2006) a specific character that is usually sought in pollinators is high tillering capacity which contributes to profuse production of pollen. It seems likely that abundant pollen production in hybrids will confer some protection from ergot and smut infection (Andrews *et al*., 1997). The relationship between GY and MSD like that between NSH and MSD was highly significant and negative in the first selfing cycles whereas it became positive with increased inbreeding. Like mentioned earlier the increased GY would arise from an increased proportion of plants with high tillering ability along the self-fertilization process, plants which are in reality the male part. Genotypes of high tillering ability showed in general thin stem diameter – NSH was negatively and highly correlated with MSD – one of the reasons why the association between DFL and MSD, initially of high level of significance may become lower with advanced generations. Additionally, the positive correlation between MSD and SPD meant thin spikes are longer and hence supporting improved grain yield. According to De Rouw (2004) grain yield in pearl millet is primarily a function of grain or panicle number per unit area. Long panicle containing as many as fertile florets ending up in large seed numbers, were preferred by farmers in the Sahel zone of Africa (Bidinger and Hash, 2003; Omanya *et al*., 2007; Drabo *et al*., 2019; Kanfani *et al*., 2020; Bassirou *et al*., 2023). In different words, GY being a necessary character in cereal improvement, breeders have tendency to primary select for traits like profuse tillering ability, panicle compactness both precursors of higher grain yield. Spike girth is definitively determined by the length of the peduncle of the spikelet. More this peduncle is longer, more the spike girth is enlarged giving loose panicle. In contrast, thin spikes result from spikelets developed on short peduncles. This may lead to head compactness equivalent to increased grain yield supported by the greater number of grains per panicle (Pucher *et al*., 2016; Serba *et al.,* 2020; Kumar *et al*., 2021; Issaka *et al.,* 2024a). Really, we observed a general trend to compactness of even the spikes of larger diameter following the advancement of lines under directional selection.

Downy mildew is the most important disease of pearl millet in Africa and Asia. This requires the use of sources of resistance in the breeding material. DMI was lowly and inconsistently connected with the eight other agronomic traits in the current study. Its sign changed from one experiment to another for same variables, and most of the correlations were not significant. For illustration it was positively correlated to GY as well as to its principal components like NSH and SPL four times over the total five of experiment cases. Whereas, downy mildew is the most disastrous disease of pearl millet, how can one approve a positive correlation between the disease incidence and the cereal production? According to Prakash *et al.* (2014), the inheritance of downy mildew resistance in pearl millet is highly variable and inconsistent. This could be attributed to a lack of homozygous resistant/susceptible genotypes, a lack of genetically pure pathogen isolate, and variable environmental conditions. According to Raj *et al.* (2018), the complexity in the inheritance of the disease resistance may be due to the out-crossing nature of both the pathogen and the cereal host. Also, it was negatively connected to DFL for four cases among the total five. That is, late flowering was associated with relatively lower downy mildew susceptibility in agreement with the findings of Pucher *et al.* (2016). Hence, with increased generation of selfing synonymous of augmented DFL value, appeared more genotypes with enlarged disease resistance. Twenty-two parental lines and their derivates showed a null disease incidence while out of the HKB cluster, the whole tested germplasm proved resistant with a disease incidence < 10%. So, even though the determinism of genetic control to the oomycete is generally under additive gene effects (Issaka *et al*., 2024b) what implies two resistant parents will be necessary for the hybrid to prosper adjacent to the oomycete, the entire diversity developed would be valuable in consequence. Nonetheless, the most resistant hybrid may be produced by crossing two lines with the highest general combining ability for the resistance (Ouendeba *et al*., 1993; Rasitha *et al*., 2023; Papanna *et al.,* 2024). Among the fresh material, Gamoji derivatives beyond demonstrating the inherited GCA for GY and resistance to the fungus auxiliary supported by the lowest DFL would represent a key female part for building pearl millet hybrids. But, it should be noted that some few lines even revealing adequate grain yield were moderately resistant and therefore should probably undergo additional selfing cycles to align with the disease. For the same reason even some resistant ones particularly those showing high grain yield should go forward generations of selfing to update the resistance level as well as the yielding capacity.

4.2 ANOVA OF VARIABLES

HKB which complete designation is Haini Kirey of Bengou is a local variety constantly cultivated in the area. Even though, it was reported as resistant cultivar and this even from trials' results recorded at Bengou (Wilson *et al*., 2008; Issaka *et al*., 2024b). Nonetheless, its offspring showed a high disease incidence out of loose panicles, after five generations of selfing each coupled with selection of only resistant plants. Determinism of the resistance to the oomycete may be controlled by additive gene effect. In fact, studying the resistance of pearl millet to four isolates collected in Niger, Issaka *et al*. (2024b) found the genetic control of the resistance to the isolate of Tara, a village close to that of Bengou was under additive gene action. This is the reason why isolated lines became so susceptible compared to their original parent. Thus, a performing OPV does not necessary yield good offspring, and therefore shall not be considered based on its performance *per se*. Still, among the genetic material tested, HKB cluster had being the tallest and one of the earliest genotypes could be considered as pollinator in future breeding work. This role was reinforced by the fact that it had the best spike length too. Indeed, the correlation between spike length and GY was positive from our study. Moreover long spikes were preferred by Sahelian pearl millet growers wherever the commerce and transport were based on bundles (Bezançon *et al.,* 2001; Buerkert *et al*., 2001; Omanya *et al.,* 2007; Drabo *et al.,* 2019). Therefore, HKB derivatives could be used in breeding program whenever the spike length constitutes a desirable trait. Furthermore, the wild variety registered good GCA for 6 agronomic parameters over 7 measured (Issaka *et al*., 2024a). Although, the genetic treasure HKB may be, it should be noted that resistance to downy mildew is a prerequisite in breeding pearl millet hybrid parents. Yet, one option to get benefit from the HKB genotypes in the future would be to test the material in more environments as the disease could vary from one locality to another (Issaka *et al*., 2024b). HKB was developed by mass selection, a method expected to be more effective for qualitative traits such as plant height, panicle length, and seed mass. However, mass selection technique is less efficient for low heritable traits and another option is to improve the original population, for instance via recurrent selection. This method has proven very effective as a result of concentrating favorable genes ending up in increased mean of the population and superior performance of the best families for the target attribute. It could by the way boost the valuable use of non-elite source materials, where the greater opportunities for recombination could break linkages between genes for the desired trait and unfavorable agronomic characteristics. Moreover, through its tandem cycling of selection and recombination, recurrent selection is a means particularly useful for improvement of polygenic traits at once of several qualitative others like disease resistance (Rattunde and Witcomb, 1993; Weltzien *et al*., 1995; Rattunde *et al.,* 1997; Weltzien *et al.,* 2020). It would be therefore a proper approach of upgrading varieties like HKB that already showed good GCA for quantitative traits akin to grain yield. For breeding programs emphasizing the development of inbred lines and hybrids from populations derived from distinct heterotic groups, reciprocal recurrent selection for a few cycles is suggested to help derive superior heterotic inbred lines. To do so, the population from one heterotic pool will serve as a tester for the population from the other heterotic pool to improve the GCA and SCA of both the populations in different heterotic pools. Thus, HKB could specifically be subjected to recurrent selection to improve the frequency of restorer genes in the male parent for the definite CMS system (Patil *et al*., 2021; Papanna *et al.,* 2024).

An HKP-GMS series showed a general reduction of NHH and consequently of NSH which outcome was a lesser grain production. NSH underlying the number of tillers per hectare had presently the highest positive and significant correlation with grain yield. Because high grain yield and its stability is the top priority in breeding pearl millet in the arid zones of Asia and Africa, the HKP-GMS faction that showed weak NSH value revealed then useless (Bidinger and Hash, 2003; Ambawat *et al*., 2020; Weltzien *et al*., 2020). But two elements of the ensemble that are HKP-GMS-128-1-14-2-1 and HKP-GMS-128-1-196-1-1 of comparable seedling establishment level showed better tillering propensity. The high significance observed between entries for most variables inside HKP-GMS block in 2021 would arise simply from the partition in the two contrasting classes of low potential and promising genotypes. It was not a matter of poor germination, but lines extracted from the same genotype would reach inbreeding depression at different generations of selfing. Indeed, in out crossing cereals like pearl millet, stigmata inside one spike are usually pollinated by different male gametes of diverse genetic level, or more precisely of different DFL values. According to Khair and Bhanvadia (2019) pearl millet florets stigmas emerge first and anthers appear 3–4 days later. Practically, most of the standard deviation values obtained stuck between one to four days inside clusters and along all experiments, the same span time for all stigmata of a given ear to come out day by day and to be available for cross-fertilization.

Yet, the two Gam-B groups evaluated in 2023 scored the premier intra-cluster standard deviation of above 20 days followed by Moro-R from the final experiment. This result pointed at the landrace nature of both Gamoji and Moro characterized by huge genetic load. The important genetic mixture was difficult to be fully uncovered even after numbers of inbreeding generations. For instance, the Gam-B lines while being at higher level of self-fertilization (S_8 to S_9) were very much different in regard to six to seven traits amid the eight recorded. Moro-R S_6 to S_{11} descendants concerning the 2024 trial were significantly different about seven variables over eight. Pucher *et al*. (2016) and Sattler *et al*. (2019) reported that high levels of genetic admixture in West African naturally occurring populations or released OPVs as the cause for failure to identify heterotic pools. The lack of differentiation into distant genetic clusters among such populations was probably due to the high out crossing nature of pearl millet leading to the concomitant high rate of pollen-mediated gene flow within the region. One may think that DFL is governed by polygene, and it is thus a vast quantitative trait. Its average mean was even more variable than that of the grain yield supposed to be the most quantitative of the parameters under the study. According to Mendelien laws more the number of genes controlling a trait are important more the trait requires extra generations of self-fertilization to reach homozygous status. But things should be viewed differently. For extreme deviations noted up, one plant's offspring may come from different tillers of diverse stages. In pearl millet, the first tiller generally appears in the axial of the third leaf nearly 12 days after seedling emergence. Subsequent primary tillers continue to develop alternatively at approximately every three days. Secondary tillers are produced from primaries at a similar rate, resulting in a potentially very large number of tillers, all at different phases of development (Bidinger and Hash, 2003). Furthermore, some varieties like Gamoji has another type of branching tillers which appear in the upper level, source of second to third harvest to occur. Such circumstances are very convenient for millet accessions of even different maturing group to cross-fertilize each others because of synchronous flowering with specific tillers of the corresponding parents.

Although pearl millet is a highly cross pollinated crop, the amount of heterozygosis observed within some clusters was not significant for variable characters. This may be due to the moving forward to homogeneous or even homozygous nature of the measured traits, acquired after several generations of directional selection and selfing. In regard to the experiment set-1 implemented in 2022, both Moro-R and HKP-GMS clusters comprised of S_6 lines reached same genetic standing at once in spite of the variables. Whereas, another Moro-R group comprised of lines of higher inbreeding level demonstrated larger heterozygosis for seven traits out of the eight of concern in 2024. In general, the presence of significant phenotypic differences particularly for DFL and secondary for grain yield between the tested entries suggested the existence of sufficient phenotypic and genetic variation in the experimental material for selection.

Definitively lines were significantly dissimilar in matter of DFL between and inside offspring's groups in spite of the self-fertilization cycles. Except for small subsets of Moro-R, HKB, and HKP-GMS related to 2021 and 2024 testing; the standard deviation intra-cluster was always higher to the inter-cluster. But similar sample of no more than 2 HKP-GMS lines in 2022 illustrated may be a hazardous standard deviation for DFL greater than that observed between the corresponding clusters. For large scale production of hybrid seed efficient pollination method will be necessary. The use of synchronous flowering parents was made possible through the larger inside standard deviations obtained and sustained by the weaker inter-clusters DFL differences, in agreement with Bidinger and Hash (2003), and Serba *et al*. (2017). Thus, Moro-R could play the role of pollinators against both HKP-GMS and Gam-B. Indeed, in addition to its thinnest spikes it revealed being taller than both female progenies, another important factor in the choice of hybrid parents. In opposition and at overall the Gam-B group with the highest grain yield, the best inherited GCA for resistance to downy mildew, the largest panicle girth, and the shortest plant height may confirm really its great role of female parent (Prem *et al*., 2006; Papanna *et al*., 2024; Issaka *et al*., 2024a). What's more and though, the mean grain yield recorded by the three different clusters was almost similar from one experiment to another, the Gam-B families were slightly better in general. In seed production it is economically important to use the higher yielding parent as the female to increase the production per unit area (Andrews *et al*., 1997; Talukdar *et al*., 1999). Indeed, the high grain yield potential of A-lines, both as lines per se as well as in combining ability is still the most important consideration. GCA is heritable from one progeny to another and A-lines with high GCA for grain yield confer stability of hybrid production over more diverse environments (Hanna and Rai, 1999; Issaka *et al.,* 2024a). High general combiners are likely or even more likely to occur in lines with average to high grain yield per se. Selecting high GCA lines in high-yielding lines increases the profitability of seed production. The selected lines can then be evaluated for GCA and SCA at the later stages. Hybrid per se performance can be predicted based on parental GCA, which is attributable to additive gene effects (Falconer and Mackay, 1996). As a result, the GCA of parents can be used as a predictive tool for developing hybrids with superior per se, reducing the use of input resources and increasing breeding efficiency. Thus, the restorers should also be highly productive from the viewpoint of the seed production economy. According to Gajjar *et al*. (2023) there was high degree of correspondence between *per se* performance and SCA effects of hybrids as well as estimates of heterosis.

Being the tallest genotype, the earliest individual Moro-R plants could be involved in developing early flowering and high yielding hybrids which could escape terminal drought very frequent in Sahel. As reminder, GY was always negatively correlated with DFL at a significant level, meaning that high grain yield is related to early flowering. According to Bidinger and Hash (2003), early genotypes that flowered 20 days before the onset of a terminal drought stress had four-fold lower yield reduction than later-flowering genotypes that flowered only ten days before the onset of such stress. Else, the direction of the variety-cross hybrid Moro x HKP-GMS manually made should change with regard to the future exploitation of CMS hybrids. Even though the two parents showed comparable grain yield with minor advantage for Moro-R, this adjustment would be of worth value because the conversion of Moro edifice into stable A_4 CMS carrier was not successful. But this was at population level and things may change when dealing now with advanced lines. Really it will be plausible to exploit from now the cross in both directions.

Coming again to the variable DFL, it got increased with recurrent inbreeding in the present study. Really, in the course of selfing process lines become weaker, a reason why the checks previously of similar DFL with the lines parental material, revealed earlier in any trialing case thereafter. However, DFL was negatively correlated with grain yield, the most significant objective in breeding pearl millet. Still, crossing between distant inbred lines can result in an early offspring because of the heterosis effect which can be controlled by epistatic effect for days to blooming trait. Shull (1908) observed in maize as result of self-fertilization, a reduction or a deterioration of plant vigor succeeding a physiological infirmity, competitive weakness, and susceptibility to disease. Still, early establishment of leaf area sufficient to intercept a majority of the incoming radiation is essential for producing a large crop biomass. Total biomass productivity in pearl millet, as in any crop, is a product of growth rate and growth duration (Bidinger and Hash, 2003; Srivastava *et al*., 2020). Yet, the maize plant vigor was restored by crossing and heterosis can manifest by virtue of improvement of several traits during plant growth and development. In pearl millet, considerable growth differences between hybrids and their parents was observed during different stages of growth and development (Srivastava *et al*., 2020).

5 CONCLUSIONS AND PERSPECTIVES

Self-fertilization followed by selection is really one of the best ways to discover previously hidden genetic combinations and get rid of those are undesirable. One of the great results was the obtaining of lines completely free of mildew during this research activity. Indeed, the absence of resistant parents was the first blocking point in the development of millet hybrids in Niger. A step has therefore been taken with the advent of these new genotypes. Another key result was the method also allowed a segregation of populations into lines sometimes very distant in terms of flowering date. This makes the choice of synchronous parents for the production of hybrids more flexible, unlike with OPVs where the flowering date is fixed. Working with several varieties also increases the chance in the qualitative choice of future parents who will produce good hybrid vigor. One of the most significant breeding objectives for pearl millet is to develop lines with higher grain yield. Certain lines showed a grain yield close to that of the check SOSAT-C88, one of the best pearl millet varieties if not the best in terms of productivity on the African continent. It is therefore the fundamental trait and any genotype is only really valid in view of the contribution it can make in building the grain yield. For the reason, some genotypes though reaching plausible stability level and even being resistant may not be challenging for hybrid development because of their low grain yield. On the contrary, some corresponding lines that are close to homozygosity with traits of interest can be utilized in breeding program straightaway as parents for hybrid production or may be used in the line development programs. Nevertheless, the use of molecular markers would be of worth value to better decode the lines' genetic purity and to know exactly which germplasm is fixed and thus to be ready for use. Other lines of interest and showing a disease incidence close to null should be pushed to next generation of inbreeding. The crop productivity was positively associated with the tillering capacity, the number of hills harvested, and the length and diameter of the ear. The association is even greater with the tillering capacity and the number of hills harvested. This last trait would mean that crop productivity could be improved by adjusting population density. On the contrary, the date of 50% flowering and the incidence of downy mildew illustrated the highest values in terms of negative correlation with the grain yield. Genetic admixture may be the cause of low heterosis with crosses like Gamoji x HKP-GMS between heterotic parents for both spike length and its girth. With the now available constructive lines such crosses could be reinvestigated to check whether improved grain yield can be. Gamoji and Moro as well pertained to two heterotic groups according only the panicle girth. It was not the general case through the current results, but it was evident when considering various individual and nicking lines. Then, it's time to explore such cross. On the whole, the role of Gam-B cluster as female parent was strengthened by its highest grain yield and resistance to downy mildew inherited from the original parent. Additional favorable traits encompass its lowest PLH standard supplemented by divergent DFL values following inbreeding cycles. Even though the putative B-lines were in general shorter, the new developed material still taller compared to the recommended height of 100 cm. So, backcrossing with a dwarf CMS source will be necessary to introduce at once the sterility system, the dwarfing gene as well as the panicle exertion trait into the female background.

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ABBREVIATIONS

INRAN: Institut National de la Recherche Agronomique du Niger, OPV: open-pollinated variety, CMS: cytoplasm male sterility, GCA: general combining ability, SCA: specific combining ability, ANOVA: analysis of variance, RCBD: randomized complete block design.

APPENDIX

APPENDIX 1: MEAN PERFORMANCE OF 25 PEARL MILLET GENOTYPES FOR 8 TRAITS ACROSS 2 SITES OVER 2021

Entry	Genotype/Cluster mean	DMI	DFL	PLH	MSD	SPL	SPD	NSH	GY
1	Gam-B-37-30-1-2-1	25	58	215	12	50	24	45	792
2	Gam-B-37-30-1-2-10	7	64	185	13	46	34	40	943
3	Gam-B-37-30-2-5-2	0	62	191	13	43	33	61	1188
4	Gam-B-37-30-2-5-4	2	62	161	14	46	34	37	504
5	Gam-B-37-30-2-5-6	2	62	164	15	45	35	36	467
6	HKB-1-7-6-4-3	25	63	199	14	52	30	46	598
7	HKB-1-7-9-1-2	21	61	209	14	58	23	28	383
8	HKB-1-7-9-1-4	42	63	229	13	59	24	26	319
9	HKB-1-7-9-1-5	10	63	213	14	58	26	36	498
10	HKP-GMS-128-1-14-2-1	5	63	223	14	44	24	50	680
11	HKP-GMS-128-1-196-1-1	12	62	241	13	50	24	51	1018
12	HKP-GMS-128-1-196-1-5	11	70	202	14	50	26	30	376
13	HKP-GMS-128-1-196-4-15	3	63	237	14	53	27	18	275
14	HKP-GMS-128-1-196-4-19	7	65	212	15	44	26	19	348
15	HKP-GMS-128-1-196-4-25	9	66	214	15	51	26	15	285
16	HKP-GMS-128-1-196-4-27	0	63	225	14	54	25	13	202
17	HKP-GMS-128-1-22-1-2	4	64	220	14	47	26	30	387
18	HKP-GMS-128-1-22-1-3	2	64	205	14	41	25	17	262
19	HKP-GMS-128-1-31-1-1	4	63	192	13	49	25	33	369
20	HKP-GMS-128-1-31-1-3	9	70	179	15	41	27	10	121
21	Moro-R-68-17-2-1-4	0	65	226	14	49	24	31	547
22	Moro-R-68-17-2-1-5	7	63	234	13	50	22	49	746
23	HKP	13	60	230	12	55	25	56	1103
24	Zatib	17	64	226	13	52	25	48	1096
25	Local	10	59	247	13	57	25	62	1173
	Gam Cluster	7*	61*	183*****	13*****	46^{ns}	32*****	44^{ns}	779*****
	HKB Cluster	25^{ns}	62^{ns}	212^{ns}	14^{ns}	56^{ns}	26*****	34^{ns}	452*
	HKP-GMS Cluster	6^{ns}	65**	214*****	14^{ns}	48*****	26^{ns}	26*****	393*****
	Moro-R Cluster	3**	64^{ns}	230^{ns}	14^{ns}	49^{ns}	23*	40^{ns}	646^{ns}
	Checks Cluster	13^{ns}	61^{ns}	234^{ns}	13^{ns}	55^{ns}	25^{ns}	55^{ns}	1124^{ns}
	Average mean	**10***	**63***	**211***	**14***	**50***	**27***	**35***	**588***

APPENDIX 2: SET-1 MEAN PERFORMANCE OF 26 MILLET GENOTYPES FOR 8 TRAITS ACROSS 2 SITES OVER 2022

Entry	Genotype/Cluster mean	DMI	DFL	PLH	MSD	SPL	SPD	NSH	GY
52	Moro-R-68-17-2-9-1-23-1	5	69	212	18	46	27	37	635
53	Moro-R-68-17-2-9-28-7-2	2	67	203	16	37	24	34	396
54	Moro-R-68-17-2-1-8-1	3	71	213	16	47	25	31	569
55	Moro-R-68-17-2-1-8-2	4	66	215	17	45	30	47	899
56	Moro-R-68-17-2-9-3-1	0	66	217	16	40	28	43	789
57	Moro-R-68-17-2-9-1-23-2	0	68	212	17	48	27	37	599
58	Moro-R-68-17-2-9-6-3	2	66	214	15	40	26	46	668
59	Moro-R-68-17-2-9-1-23-3	0	66	232	15	45	27	48	854
60	Moro-R-68-17-2-9-1-23-4	0	64	213	15	42	25	41	821
61	Moro-R-68-17-2-3-1-3-3	3	68	222	17	42	26	45	937
62	Moro-R-68-17-2-9-28-7-1	1	65	217	16	43	26	50	896
63	Moro-R-68-17-2-9-28-7-3	6	66	287	16	41	25	42	633
64	Moro-R-68-17-2-9-28-7-4	5	63	221	14	40	24	47	736
65	HKP-GMS-128-1-22-1-1-2	3	69	226	18	46	28	40	676
66	HKP-GMS-128-1-31-2-3-1	0	72	217	17	46	27	38	798
67	HKP-GMS-128-1-31-2-3-4	3	66	237	17	50	26	43	753
68	HKP-GMS-128-1-31-2-3-6	1	65	223	14	44	24	46	968
69	HKP-GMS-128-1-196-1-1-1	7	68	210	16	43	26	37	569
70	HKP-GMS-128-1-196-1-1-3	3	70	225	16	47	27	35	635
71	HKP-GMS-128-1-196-1-1-4	2	71	226	18	42	28	39	813
72	HKP-GMS-128-1-196-1-1-6	0	67	223	15	47	27	38	710
73	HKP-GMS-128-1-196-1-1-7	1	65	208	16	44	24	35	446
74	HKP-GMS-128-1-196-1-1-9	1	69	218	16	43	27	30	569
75	HKP-GMS-128-1-196-2-1-2	1	68	214	16	42	25	35	492
76	HKP-GMS-128-1-196-4-27-1	0	67	218	14	39	29	45	778
51	SOSAT-C88	3	62	227	15	37	26	44	659
	Moro-R Cluster	2 ^{ns}	66 ^{ns}	221 ^{ns}	16 ^{ns}	43 ^{ns}	26 ^{ns}	42 ^{ns}	725 ^{ns}
	HKP-GMS Cluster	2 ^{ns}	68 ^{ns}	220 ^{ns}	16 ^{ns}	44 ^{ns}	26 ^{ns}	38 ^{ns}	684 ^{ns}
	Check Cluster	3	62	227	15	37	26	44	659
	Average mean	2 ^{ns}	67 ^{ns}	221 ^{ns}	16 ^{ns}	43 ^{ns}	26 ^{ns}	40 ^{ns}	704 ^{ns}

APPENDIX 3: SET-2 MEAN PERFORMANCE OF 26 MILLET GENOTYPES FOR 8 TRAITS ACROSS 2 SITES OVER 2022

Entry	Genotype/Cluster mean	DMI	DFL	PLH	MSD	SPL	SPD	NSH	GY
26	Gam-B-11-1-14-5-2-4-2-1	6	66	211	15	48	25	42	617
27	Gam-B-11-1-14-32-2-2-1-2	1	65	214	13	37	26	42	713
28	Gam-B-11-1-14-32-13-3-2-2	7	67	211	14	95	23	55	820
29	Gam-B-37-30-1-2-1-1-1	7	66	207	11	44	24	51	859
30	Gam-B-37-30-1-2-1-5-4	2	67	217	12	37	27	51	678
31	Gam-B-37-30-2-5-3-1-1	2	64	357	14	48	27	45	973
32	Gam-B-37-30-2-5-3-1-2	2	67	232	13	44	23	45	699
33	Gam-B-37-30-2-5-3-1-4	9	64	232	16	49	27	34	657
34	Gam-B-37-30-2-5-3-1-5	5	63	209	12	44	22	42	851
35	Gam-B-37-30-2-5-3-1-6	9	68	211	14	44	23	55	737
36	Gam-B-37-30-2-5-7-1-1	4	62	199	14	81	21	57	691
37	Gam-B-37-30-2-5-8-1-1	0	65	198	14	35	24	51	788
38	Gam-B-37-30-2-5-8-1-2	2	65	225	14	42	23	45	749
39	Gam-B-37-30-2-5-8-2-1	3	71	216	16	46	27	29	910
40	Gam-B-37-30-2-5-8-2-2	1	68	214	14	46	25	42	705
41	Moro-R-68-17-2-1-4-4-3	3	67	237	14	56	22	44	705
42	Moro-R-68-17-2-1-5-1-2	2	61	230	14	49	22	38	884
43	Moro-R-68-17-2-1-5-1-3	20	65	220	13	45	26	43	838
44	Moro-R-68-17-2-1-5-1-4	12	66	226	13	43	26	49	872
45	Moro-R-68-17-2-1-5-3-4	4	69	229	17	47	25	41	490
46	Moro-R-68-17-2-1-4-3-2	1	65	235	13	45	26	32	536
47	Moro-R-68-17-2-9-8-3-1	1	66	222	15	53	21	43	837
48	Moro-R-68-17-2-9-12-1-1	5	59	252	14	40	27	50	1058
49	HKP-GMS-128-1-14-2-1-5-1	4	64	221	16	45	24	47	720
50	HKP-GMS-128-1-14-2-1-3-1	3	70	203	13	38	24	38	626
51	SOSAT-C88	5	59	252	14	40	27	50	1058
	Gam Cluster	3 ^{ns}	66**	224 ^{ns}	14 ^{ns}	43*****	25****	46 ^{ns}	763 ^{ns}
	HKP-GMS Cluster	6 ^{ns}	68 ^{ns}	209 ^{ns}	14 ^{ns}	38 ^{ns}	25 ^{ns}	41 ^{ns}	612 ^{ns}
	Moro-R Cluster	6*	66*	227 ^{ns}	14 ^{ns}	48*	24***	42 ^{ns}	735 ^{ns}
	Check Cluster	5	59	252	14	40	27	50	1058
	Average mean	**4****	**66*******	**225** ^{ns}	**14***	**48** ^{ns}	**24*******	**44** ^{ns}	**754** ^{ns}

APPENDIX 4: MEAN PERFORMANCE OF 26 PEARL MILLET GENOTYPES FOR 8 TRAITS ACROSS ONE SITE OVER 2023

Entry	Genotype/Cluster mean	DMI	DFL	PLH	MSD	SPL	SPD	NSH	GY
77	Gam-B-37-30-2-5-8-2-1-1-1	0	70	192	16	45	37	65	938
78	Gam-B-37-30-2-5-8-2-1-1-2	1	66	175	15	44	37	55	1191
79	Gam-B-37-30-2-5-8-1-1-1	0	62	172	15	47	30	58	945
80	Gam-B-37-30-2-5-8-1-3-3	0	62	148	13	44	32	53	546
81	Gam-B-37-30-2-5-8-2-2-1	1	69	219	14	41	35	46	816
82	Gam-B-37-30-2-5-8-1-3-5	1	74	178	15	41	31	54	785
83	Gam-B-37-30-2-5-8-1-3-6	0	80	164	15	48	33	35	418
84	Gam-B-37-30-2-5-3-1-6-1	2	75	143	18	40	34	30	383
85	Gam-37-30-2-5-3-1-6-2	1	77	176	16	44	33	48	586
86	Gam-B-37-30-2-5-3-1-1-1	1	63	218	15	46	30	66	1301
87	Gam-B-37-30-2-5-3-1-1-2	1	65	165	15	36	36	58	941
88	Gam-B-37-30-2-5-7-1-4-2	0	72	156	14	50	31	56	702
89	Gam-B-37-30-2-5-3-1-2-1	3	90	142	19	47	30	17	472
90	Gam-B-37-30-2-5-8-1-3-4	1	79	192	16	49	31	40	624
91	Gam-B-11-1-14-32-12-1-3-6	0	77	124	13	38	31	46	419
92	Gam-B-11-1-14-32-12-1-2-1	0	81	169	16	51	30	41	318
93	Gam-B-11-1-14-32-12-1-2-4	0	63	236	15	58	31	34	1335
94	Gam-B-11-1-14-32-4-1-1-1	2	80	173	15	44	31	67	1032
95	Gam-B-11-1-14-32-4-1-1-2	1	58	210	12	57	21	44	847
96	Gam-B-11-1-14-32-4-1-1-3	0	65	209	13	54	27	67	1388
97	Gam-B-11-1-14-32-4-1-1-4	3	74	198	16	46	25	47	935
98	Gam-B-11-1-14-32-4-1-1-5	0	68	235	14	56	29	67	1470
99	Gam-B-11-1-14-32-4-1-1-6	1	59	188	15	50	25	70	1299
100	Gam-B-11-1-14-32-4-1-1-7	1	68	218	15	49	29	73	1207
101	Gamoji	2	56	225	12	43	30	67	1227
51	SOSAT-C88	0	62	239	13	29	29	83	1854
	Gam-B-37-30-2-5 Cluster	1^{ns}	72*****	174*****	15*	44*	33****	49*****	766****
	Gam-B-11-1-14-32 Cluster	1****	68*****	196*****	14^{ns}	50*****	28*****	57*****	1050^{ns}
	Check Cluster	0	62	239	13	29	29	83	1854
	Average mean	1***	70*****	187*****	15***	46*****	31*****	53*****	922*****

APPENDIX 5: MEAN PERFORMANCE OF 33 PEARL MILLET GENOTYPES FOR 8 TRAITS ACROSS FOUR SITES OVER 2024

Entry	Genotype/Cluster mean	DMI	DFL	PLH	MSD	SPL	SPD	NSH	GY
102	Gam-B-11-1-14-32-2-2-1-1-4-4	7	61	174	11	44	19	35	601
103	Gam-B-11-1-14-38-18-2-1-1	2	69	214	13	53	19	21	497
104	Gam-B-11-1-14-38-18-2-1-2	4	69	186	12	49	21	24	557
105	HKP-GMS-128-1-196-2-1-1-1-3-3	12	73	199	14	48	22	21	460
106	HKP-GMS-128-1-196-2-1-1-1-3-4	1	70	258	14	50	24	36	1015
107	Moro-R-68-17-2-1-4-3-3	4	66	224	14	55	23	31	783
108	Moro-R-68-17-2-1-4-3-3-2	6	65	199	12	46	21	32	803
109	Moro-R-68-17-2-1-4-3-3-5	6	70	192	14	54	22	16	284
110	Moro-R-68-17-2-1-4-3-3-9	2	69	201	14	51	21	30	691
111	Moro-R-68-17-2-1-4-3-3-11	0	69	223	14	56	20	28	603
112	Moro-R-68-17-2-1-4-3-3-15	9	68	207	13	50	21	36	734
113	Moro-R-68-17-2-1-4-3-3-15-1	5	69	214	13	50	21	32	651
114	Moro-R-68-17-2-1-4-3-3-19-1	4	68	203	14	56	22	28	576
115	Moro-R-68-17-2-1-4-3-3-27	5	71	209	15	60	24	32	728
116	Moro-R-68-17-2-1-4-3-3-32	5	67	192	14	53	23	28	578
117	Moro-R-68-17-2-1-4-3-3-38	5	64	199	13	50	22	33	793
118	Moro-R-68-17-2-1-4-3-3-39	23	68	189	14	51	22	25	546
119	Moro-R-68-17-2-1-4-3-3-42	8	68	205	15	55	22	30	685
120	Moro-R-68-17-2-1-5-1-1-1-21	2	68	220	14	43	25	33	954
121	Moro-R-68-17-2-1-5-1-1-1-22	2	68	204	13	44	24	25	630
122	Moro-R-68-17-2-1-5-1-1-1-24	3	71	203	15	46	27	23	560
123	Moro-R-68-17-2-9-1-23-1-1-1	2	67	205	14	48	27	31	847
124	Moro-R-68-17-2-9-1-23-1-1-1-1	9	66	227	14	44	28	31	940
125	Moro-R-68-17-2-9-1-23-4-70	9	73	192	12	43	23	29	502
126	Moro-R-68-17-2-9-3-1	2	77	186	15	46	28	17	269
127	Moro-R-68-17-2-9-8-3-1-1-2-2	8	69	182	14	50	30	34	612
128	Moro-R-68-17-2-9-8-3-1-1-2-4	4	70	207	14	51	25	26	672
129	Moro-R-68-17-2-9-8-3-1-1-2-10-1	2	72	206	14	49	26	25	572
130	Moro-R-68-17-2-1-4-3-3-45	1	72	203	14	51	22	21	521
131	Moro-R-68-17-2-1-5-1-1-1-27	18	68	215	13	41	27	31	788
23	HKP	14	64	209	14	54	22	42	944
51	SOSAT-C88	2	68	210	14	28	31	33	859
24	Zatib	5	67	226	14	54	23	33	723
	Gam Cluster	4^{ns}	67*	191*	12^{ns}	49*****	19^{ns}	27*	551^{ns}
	HKP-GMS Cluster	6^{ns}	71^{ns}	228^{ns}	14^{ns}	49^{ns}	23***	29**	737**
	Moro-R Cluster	6***	69*	204***	14^{ns}	50*****	24*****	28*****	653*****
	Checks Cluster	7^{ns}	66^{ns}	215^{ns}	14^{ns}	45*****	25***	36^{ns}	842^{ns}
	Average mean	6****	68*****	206*****	14^{ns}	49*****	24*****	29*****	666*****

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