

Research Article

Genetic Gain in Wheat Grain Yield and Nitrogen Use Efficiency at Different Nitrogen Levels in an Irrigated Hot Environment

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Improved nitrogen use-efficient cultivars could be the most economically beneficial and environmentally friendly approach to reduce pollution associated with excessive N fertilization. The performance and genetic gain in grain yield and nitrogen use efficiency (NUE) of a historical set of 12 bread wheat cultivars released for a heat-stressed environment were investigated at four N levels (0 (N₀), 43 (N₄₃), 86 (N₈₆), and 129 (N₁₂₉) kg/ha) for two seasons. Averaged across seasons, increasing N level from N₀ to N₄₃, N₈₆, and N₁₂₉ resulted in yield increases ranging from 4–45%, 13–69%, and 34–87% at N₄₃, N₈₆, and N₁₂₉, respectively. These yield increases were associated with increases in biomass ($r = 0.86$, $P < 0.01$). Regressing grain yield of cultivars released during 1960 to 2006 against the year of release showed no trend at N₀ and positive nonsignificant trends at N₄₃; however, significant positive trends were found at N₈₆ and N₁₂₉ with genetic gain rates of 12.65 and 15.76 kg ha⁻¹ year⁻¹, respectively. This gain was associated with progresses in harvest index (HI) at N₄₃, N₈₆, and N₁₂₉ but not at N₀. On the other hand, during the period from 1960 to 1990, the genetic gain in grain yield at N₈₆ was 24.5 kg ha⁻¹ year⁻¹. Regressing NUE against the year of release showed significant linear trends at N₈₆ and N₁₂₉ ($R^2 = 0.511$ and $R^2 = 0.477$, respectively), but not at N₄₃. The results indicate that breeders improved grain yield and NUE over 46 years under the heat-stressed environment of Sudan although the rate of increase in yield has been slowed down in recent years. Further improvement in NUE might require broadening the genetic diversity and simultaneous evaluation at low and high N levels.

1. Introduction

The recent worldwide interest to avoid environmental hazards associated with N losses and to reduce the high cost of production is bringing more emphasis in breeding cultivars possessing improved NUE while maintaining an acceptable yield [1–5]. Sustainable agricultural production requires efficient management of N through agronomic practice and the use of appropriate germplasm that are optimized for traits related to N use efficiency rather than yield alone [6]. In addition, field management tools such as drought priming have been suggested to enhance grain yield and nitrogen use efficiency under multiple abiotic stresses in a future drier and warmer climate [7].

Thousands of modern semidwarf wheat (*Triticum aestivum* L.) cultivars with high-yielding potential, highly responsive to nutrients, especially N, have been released for use in both favorable and marginal environments since the “Green Revolution” era in 1960s. It is generally thought that these modern high-yielding cultivars demand high N level to maximize their yield [8, 9]. However, it would be economically and environmentally beneficial to have high-yielding genotype that could attain maximum yield at low N input especially in the light of global climate changes [10]. It has been reported that some modern cultivars outyielded both old tall and earlier semidwarf cultivars at all nitrogen levels [11, 12].

Considerable genetic diversity has been reported in wheat for nitrogen uptake and utilization efficiencies

[10, 11, 13, 14]. However, further improvements may require exploitation of a wider germplasm pool through utilization of land races and ancestral germplasm [6]. Wheat genotypes with high harvest index and low forage yield showed low N loss and increased N use efficiency [15]. For further genetic improvement, maximizing N capture, partitioning and remobilization of N to the grain, and yield *per se* could be targeted [6].

Nitrogen use efficiency is genetically controlled; however, some environmental factors such as drought and heat stresses could modify the genetic effects. High temperature during grain filling stage is reported to reduce N remobilization from the stems to the grains of wheat [16]. However, N fertilization under heat stress was found to increase biomass production and canopy temperature depression [17, 18]. These traits are known to be important for wheat productivity under heat stress conditions [19–22]. So far, differential genotypic response to different N fertilization levels has been rarely addressed under dry, hot irrigated environments. On the other hand, genetic progress in grain yield and NUE have been reported at different environmental conditions [12, 23, 24]. However, despite the fact that genetic gain in grain yield of $25.6 \text{ kg ha}^{-1} \text{ year}^{-1}$ has been reported at the heat-stressed environment of Sudan using the recommended N dose [25], very little is known about the genetic progress made in wheat grain yield and N use efficiency under different N levels in such environments. Periodic and frequent evaluation of the genotypes and the breeding methodologies followed is essential for attaining progressive genetic gain in grain yield and NUE and identifying traits that might need more emphasis by breeders especially in the light of the changing climate. Moreover, the environment where this study has been conducted is expected to represent the future environment of several areas when the ensuing climate change is considered. Hence, valuable information could be provided for the global wheat community that could provide roadmap for the future crop improvement. This study investigated the genotypic variation in the performance and the genetic progress in grain yield and N use efficiency in response to four N levels of a historical set of bread wheat cultivars released for a hot irrigated environment.

2. Materials and Methods

Twelve bread wheat genotypes were used in this study to represent a historical set of cultivars released during 1960 to 2006 for the dry, hot irrigated environment of Sudan. The two eldest cultivars (Beladi and Giza 155) are tall (full stature), whereas the rest have semidwarf stature. The cultivars were grown in a field experiment conducted at the Gezira Research Station Farm (GRSF), Wad Medani ($14^{\circ}44'N$; $33^{\circ}29'E$; 411 m asl), Sudan, for two seasons 2006/07 and 2007/08. At GRSF, the soil is categorized as fine, heavy cracking montmorillonitic, isohyperthermic soil with pH of 8.0–8.5 and the rooting depth is up to about 0.4 m. The soil is poor in organic matter (0.3%), deficient in total nitrogen (0.03–0.05%), and low in available phosphorus (4–5 ppm; Olsen extractable P).

The 12 cultivars were evaluated under four nitrogen levels; 0 kg N/ha (N_0), 43 kg N/ha (N_{43}), 86 kg N/ha (N_{86}), and 129 kg N/ha (N_{129}) split-applied in the form of urea at three-leaf and tillering stages. The recommended dose of phosphorus fertilizer in the form of triple super phosphate at the rate of 43 kg P_2O_5 /ha was applied by furrow placement prior to sowing. The daily maximum and minimum temperatures have been provided from a meteorological station located within the experimental sites during the two cropping seasons (2006/07 and 2007/08).

Seeds were treated with Gaucho (imidacloprid 35% WP) for the control of pests mainly termites and aphids. Sowing was done mechanically in plots consisting of eight rows, 5 m long and 0.2 m apart in both seasons, using a seed rate of 120 kg ha^{-1} . Sowing was done during the 3rd week of November in both seasons. To avoid any water stress, the experiment was irrigated frequently (every 10–12 days) and kept free from weeds by manual weeding at least twice. The experiment was arranged in a randomized complete block design with three replications in both seasons. It is noteworthy that the experimental site was free from major wheat diseases and no serious lodging was reported that could have affected the grain yield.

Excluding the two border rows, a net area of 2.4 m^2 (4 rows by 3 m) was harvested manually from the ground level in the first season, whereas the area harvested in the second season was 4.8 m^2 (6 rows by 4 m). The harvested material was banded and left to dry, then weighed, and threshed, and the grain was weighed again to give biomass and grain yield. Harvest index was calculated as the ratio of grain yield to biomass weight.

Representative plant samples were taken at harvest to determine the nitrogen contents of the aboveground portions. Samples were oven-dried to constant weight before grinding. Nitrogen was determined by semi micro-Kjeldhal procedure of [26]. Nitrogen uptake in grains (NUTG) and nitrogen uptake in straw (NUTS) were calculated by the multiplication of the N concentration in both fractions by their corresponding dry weights (kg/ha).

Ladha et al. [27] defined 18 different ways to calculate the nitrogen use efficiency. In this study, only three terms of NUE were used, namely:

$\text{NUE} = \text{kg grain per kg of available N from soil and fertilizer.}$

$\text{N utilization efficiency (NUTE)} = \text{kg grains divided by above ground plant N.}$

$\text{N harvest index (NHI)} = \text{proportion of grain N from the above ground plant N (above ground plant N was the summation of grain and straw N uptake).}$

Data were subjected to analyses of variance for each season separately and then combined analysis was carried out after the error mean squares were tested for homogeneity. Genotype and N level were considered as fixed factors, whereas season and replication nested within each N level were treated as random factors. Only the combined data for both seasons are presented since season \times N level \times genotype was not significant for most of the measured traits. Standard ANOVA was performed for the NUE traits. Simple correlation coefficients were calculated among different traits

using genotype means across N levels ($n = 48$). Regression analysis was used to measure the relationship of the year of release with the grain yield and NUE. Time elapsed from the year of release was regarded as a quantitative variable to estimate the progress in genetic gain for grain yield and NUE. Eleven cultivars (excluding Tagana) were used in this analysis. Rates of increase in grain yield and NUE were estimated from the slope of the regression line.

3. Results

The weekly maximum and minimum temperatures at GRSF during the two cropping seasons (2006/07 and 2007/08) as well as a 30-year average (LTA) during 1980–2010 are shown in Figure 1. During most parts of the crop vegetative stage in the second season, the maximum and minimum temperatures were higher compared to the 1st season and the LTA. However, temperatures of both seasons became almost similar from midseason onward. During the growth cycle, temperatures above 35°C were experienced for more than 20 days during both seasons as well as the LTA.

3.1. Genotypic Variation. Significant differences were found between the N levels and genotypes for all studied traits. Addition of N increased biomass and grain yield of all genotypes. However, the magnitude of the response varied among the genotypes.

Mean biomass of the 12 genotypes increased from 5423 kg/ha at N_0 to 6750 kg/ha at N_{43} , 8023 kg/ha at N_{86} , and 8924 kg/ha at N_{129} (Table 1). At N_{43} , Argine showed only 1% increase in biomass compared to that at N_0 , whereas Wadi Elneel showed 43% increase. The percent increase in biomass of Condor was 27, whereas that of Wadi Elneel was 65 at N_{86} . Khalifa and Nesser were the two contrasting genotypes in the percent increase of biomass at N_{129} compared to N_0 .

Significant differences in grain yield among N levels and genotypes were found (Table 2). Across all genotypes, increasing N level from N_0 to N_{43} , N_{86} , and N_{129} resulted in increases in grain yield by 24, 44, and 55%, respectively. Averaged across N levels, increasing N level from N_0 to N_{43} increased grain yield with ranges from 4% in Argine to 45% in Wadi Elneel. Similarly, increasing N level from N_0 to N_{86} and N_{129} resulted in grain yield increases ranging from 13% in Condor to 69% in Nesser at N_{86} and from 34% in Debeira to 87% in Khalifa at N_{129} . Genotypes Khalifa, Imam, Bohaine, and Elnielain showed continuous increases in grain yield with application of more N. In contrast, addition of more N above N_{86} did not improve the grain yield of Wadi Elneel, Tagana, and Nesser. Most of the increase in grain yield of the later genotypes resulted from the addition of 43 kg ha⁻¹.

Genotypes showed mixed reactions in response of harvest index (HI) to increasing N levels. For example, the HI of Nesser increased by 14, 15, and 10% at N_{43} , N_{86} , and N_{129} , respectively, compared to N_0 , whereas Beladi, Giza 155, Condor, and Debeira showed different decreasing trends with addition of more N (Table 3).

Significant differences in NUE among N levels, genotypes, and their interaction were found (Table 4). Across all genotypes, increasing N level resulted in decreases of NUE from 58.7 kg grain/kg N applied at N_{43} to 36.1 kg grain/kg N applied at N_{86} and 27.0 kg grain/kg N applied at N_{129} . Averaged across N levels, NUE of the cultivars ranged from 33.8 kg grain/kg N applied in the old cultivar Beladi to 43.8 kg grain/kg N applied in Elnielain.

Table 5 shows the nitrogen uptake in the straw and grains. Significant ($P < 0.001$) genotypic variations in straw N uptake were detected. Across all nitrogen levels, the highest N uptake in straw was recorded by Beladi, Giza 155, and Tagana. On the other hand, Bohaine, Nesser, and Condor recorded the lowest biomass N uptake (Table 5).

The grain N uptake of the 12 wheat genotypes significantly increased by addition of N. Across all nitrogen levels applied, significant variation among tested genotypes was found. The old cultivar (Beladi), Tanana, and Debeira recorded the lowest N uptake, whereas Bohaine and Argine recorded the highest N uptake (Table 5).

Averaged across all genotypes, the proportion of grain N from the total above ground plant nitrogen (nitrogen harvest index, NHI), also known as translocation coefficient and reutilization efficiency, varied significantly among genotypes. The NHI at N_{129} was significantly higher than that at other N levels. Across all N levels, Bohaine, Argine, and Condor showed significantly higher nitrogen harvest index, whereas Debeira, Beladi, and Tagana recorded the lowest (Table 5).

The grain yield produced from a unit of above ground plant N uptake (i.e., nitrogen utilization efficiency, NUTE) was significantly reduced with each increase in N rate. Significant variation among the 12 wheat genotypes in NUTE was also found. Across all nitrogen levels, NUTE was highest in Debeira (47.4 kg grain/kg N taken up) followed by Wadi Elneel, Khalifa, and Elnielain (Table 5).

3.2. Genetic Gain in Grain Yield under Different Nitrogen Levels. The grain yields under different N levels as well as the mean grain yield across all N levels were linearly regressed against the years elapsed since the release of the cultivars (1960 was considered as the year of Beladi release). Regressing the grain yield at N_0 against the year of release showed no trend ($R^2 = 0.003$, $P = 0.957$) (Figure 2). A positive nonsignificant trend was noticed when the year of release was regressed against the grain yield at N_{43} ($R^2 = 0.17$, $P = 0.208$). On the other hand, positive and significant relations were found when the grain yield at N_{86} and N_{129} was plotted against the year of release ($R^2 = 0.511$, $P = 0.013$ and $R^2 = 0.477$, $P = 0.019$, respectively). Based on the regression of grain yield at N_{86} in the two seasons versus the year of release, the grain yield has increased from 2614.3 kg ha⁻¹ in 1960 to 3196.3 kg ha⁻¹ in 2006 with an annual rate of 12.65 kg ha⁻¹ year⁻¹ (0.48% year⁻¹ on the relative basis). Similarly, grain yield at N_{129} increased from 2804.8 kg ha⁻¹ in 1960 to 3529.6 kg ha⁻¹ in 2006 with an annual rate of 15.76 kg ha⁻¹ year⁻¹ (0.56% year⁻¹ on the relative basis). Likewise, regressing mean grain yield across all N levels in

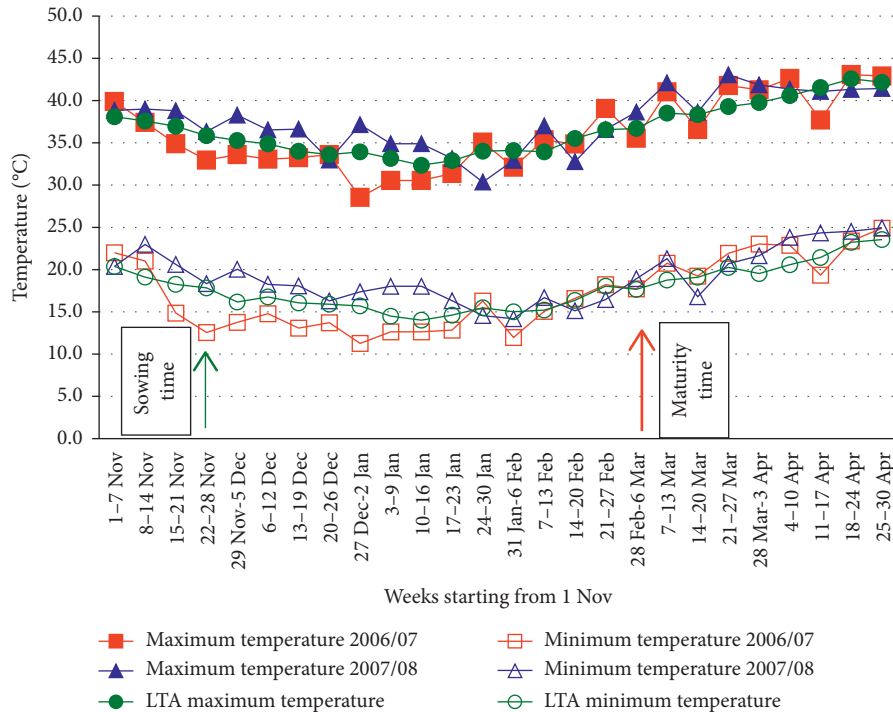


FIGURE 1: Weekly maximum and minimum temperature at Gezira Research Station during two cropping seasons (2006/07 and 2007/08) together with a 30-year average (LTA) temperature (1980–2010).

TABLE 1: Mean biomass (kg/ha) of 12 bread wheat genotypes grown for two seasons under four N levels at Gezira Research Station Farm, Wad Medani, Sudan.

Genotype [†]	Biomass (kg/ha)			
	N ₀	N ₄₃	N ₈₆	N ₁₂₉
Beladi 60	6005	6981 (16)	8447 (41)	9533 (59)
Giza 155 71	5474	7079 (29)	8276 (51)	9679 (77)
Condor 78	5128	6206 (21)	6519 (27)	8661 (69)
Debeira 82	5853	7188 (23)	8101 (38)	8907 (52)
Wadi Elneel 87	4982	7148 (43)	8203 (65)	8522 (71)
Elnielain 90	5854	6934 (18)	8590 (47)	9215 (57)
Nesser 96	5089	6454 (27)	7465 (47)	7363 (45)
Argine 96	5547	5575 (1)	8385 (51)	9081 (64)
Imam 00	5433	7007 (29)	7924 (46)	9187 (69)
Tagana 04	5332	7253 (36)	8657 (62)	9439 (77)
Khalifa 04	4908	6729 (37)	7923 (61)	9402 (92)
Bohaine 06	5470	6444 (18)	7786 (42)	8104 (48)
Mean	5423	6750 (25)	8023 (48)	8924 (65)
SE ± for:				
N level (N)		144.5***		
Genotype (G)		120.9***		
N × G		241.8*		

Number in parentheses is the percent difference relative to N₀. *Significant at $P = 0.05$; ***significant at $P = 0.001$. [†]The numerical suffix (in bold) to the genotype name refers to the year of its release in Sudan.

the two seasons versus the year of release showed that the grain yield has increased from 2473.3 kg ha⁻¹ in 1960 to 2870.2 kg ha⁻¹ in 2006 with an annual rate of 8.63 kg ha⁻¹ year⁻¹ (0.35% year⁻¹ on the relative basis). The genetic gain in grain yield is associated with increases in harvest index at N₄₃, N₈₆, and N₁₂₉ but not at N₀. The HI

TABLE 2: Mean grain yield (kg/ha) of 12 bread wheat genotypes grown for two seasons under four N levels at Gezira Research Station Farm, Wad Medani, Sudan.

Genotype	Grain yield (kg/ha)			
	N ₀	N ₄₃	N ₈₆	N ₁₂₉
Beladi 60	1923	2178 (13)	2558 (33)	2716 (41)
Giza 155 71	2000	2445 (22)	2722 (36)	2994 (50)
Condor 78	2272	2558 (13)	2561 (13)	3188 (40)
Debeira 82	2323	2668 (15)	3023 (30)	3104 (34)
Wadi Elneel 87	1988	2874 (45)	3145 (58)	3167 (59)
Elnielain 90	2306	2730 (18)	3314 (44)	3783 (64)
Nesser 96	1835	2603 (42)	3100 (69)	2889 (57)
Argine 96	2201	2283 (4)	3166 (44)	3364 (53)
Imam 00	2011	2510 (25)	2887 (44)	3352 (67)
Tagana 04	1952	2715 (39)	2979 (53)	2826 (45)
Khalifa 04	1985	2760 (39)	3101 (56)	3704 (87)
Bohaine 06	2129	2525 (19)	3103 (46)	3477 (63)
Mean	2077	2571 (24)	2972 (44)	3214 (55)
SE ± for:				
N level (N)		41.75***		
Genotype (G)		61.8***		
N × G		174.7**		

Number in parentheses is the percent difference relative to N₀. **Significant at $P = 0.01$; ***significant at $P = 0.001$.

increased by 0.16% year⁻¹ ($R^2 = 0.452$, $P < 0.01$), 0.17% year⁻¹ ($R^2 = 0.573$, $P \leq 0.01$), and 0.25% year⁻¹ ($R^2 = 0.731$, $P \leq 0.01$) at N₄₃, N₈₆, and N₁₂₉, respectively (Figure 3).

On the other hand, when the period from 1960 to 1990 was considered, the genetic gains in grain yield at different N levels were higher. For instant, the genetic gain for the mean grain yield across all N levels was estimated to be

TABLE 3: Mean harvest index (%) of 12 bread wheat genotypes grown for two seasons under four N levels at Gezira Research Station Farm, Wad Medani, Sudan.

Genotype	Harvest index (%)			
	N ₀	N ₄₃	N ₈₆	N ₁₂₉
Beladi 60	31.7	31.3 (-1)	30.3 (-4)	28.4 (-10)
Giza 155 71	36.7	34.4 (-6)	32.8 (-11)	30.8 (-16)
Condor 78	44.3	41.4 (-7)	39.7 (-11)	37.5 (-15)
Debeira 82	39.8	37.1 (-7)	37.2 (-7)	34.9 (-12)
Wadi Elneel 87	40.1	39.8 (-1)	38.3 (-4)	37.0 (-8)
Elnielain 90	39.6	39.1 (-1)	38.3 (-3)	41.4 (5)
Nesser 96	35.9	40.7 (14)	41.2 (15)	39.4 (10)
Argine 96	39.6	41.2 (4)	37.8 (-5)	37.6 (-5)
Imam 00	36.8	35.5 (-3)	36.5 (-1)	36.5 (-1)
Tagana 04	36.7	37.7 (3)	34.0 (-7)	30.3 (-17)
Khalifa 04	40.9	41.6 (2)	38.9 (-5)	39.4 (-4)
Bohaine 06	39.3	39.6 (1)	39.9 (2)	42.9 (9)
Mean	38.4	38.3 (0)	37.1 (-3)	36.3 (-5)
SE ± for:				
N level (N)		0.59*		
Genotype (G)		0.74***		
N × G		1.5*		

Number in parentheses is the percent difference relative to N₀. *Significant at $P = 0.05$; ***significant at $P = 0.001$.

20.67 kg ha⁻¹ year⁻¹ (0.89% year⁻¹ on a relative basis) (Figure 4). At N₈₆ (the recommended N dose for wheat production in Sudan), the genetic gain was 24.5 kg ha⁻¹ year⁻¹ (1.0% year⁻¹ on a relative basis).

3.3. Genetic Gain in NUE under Different Nitrogen Levels. Evaluation of the relationship between NUE of the eleven cultivars (excluding Tagana) under different N levels and their year of release was done using regression analysis. The NUE under different N levels as well as the mean NUE was regressed against the year of cultivar release. Regressing NUE at N₄₃ against the year of release revealed nonsignificant positive trend ($R^2 = 0.1695$, $P = 0.208$). However, positive and significant relations were found when NUEs were plotted against year of release at N₈₆ ($R^2 = 0.511$, $P = 0.014$) and N₁₂₉ ($R^2 = 0.477$, $P = 0.019$) as well as the mean value across the N levels ($R^2 = 0.46$, $P = 0.022$) (Figure 5).

4. Discussion

4.1. Genotypic Variation. In this study, a historical set of 12 bread wheat cultivars released for a hot irrigated environment were used to investigate the performance and genetic gain in grain yield and nitrogen use efficiency (NUE) at four N levels. Addition of N increased biomass and grain yield of all genotypes. However, the magnitude of the response varied among the genotypes. The mean biomass of the 12 genotypes increased by 60.7% due to addition of 129 kg N/ha compared to the control (N₀). Similarly, increasing N level from N₀ to N₁₂₉ resulted in increases in grain yield by 55%. The response of some genotypes to addition of 43 kg N/ha was almost negligible (e.g., Argine), whereas others showed more than 40% increase in grain yield (Wadi Elneel and

TABLE 4: Nitrogen use efficiency (NUE) of 12 bread wheat genotypes grown in season 2006/07 and 2007/08 under different N levels at Gezira Research Station Farm, Wad Medani, Sudan.

Genotype	Nitrogen use efficiency (kg grain/kg N applied)			
	N ₄₃	N ₈₆	N ₁₂₉	Mean
Beladi 60	50.7	29.7	21.1	33.8
Giza 155 71	56.9	31.7	23.2	37.2
Condor 78	59.5	29.8	24.7	38.0
Debeira 82	62.0	35.2	24.1	40.4
Wadi Elneel 87	66.8	36.6	24.6	42.7
Elnielain 90	63.5	38.5	29.3	43.8
Nesser 96	60.5	36.0	22.4	39.7
Argine 96	53.1	36.8	26.1	38.7
Imam 00	58.4	33.6	26.0	39.3
Tagana 04	63.1	34.6	21.9	39.9
Khalifa 04	64.2	36.1	28.7	43.0
Bohaine 06	58.7	36.1	27.0	40.6
Mean				
SE ± for:				
N level (N)			0.59***	
Genotype (G)			1.1***	
N × G			1.91*	

*Significant at $P = 0.05$; ***significant at $P = 0.001$.

Nesser). However, addition of more N increased the grain yield of Argine from 4% at N₄₃ to 44% at N₈₆ compared to N₀. Similar results for the responsiveness and efficiency of wheat genotypes to the addition of N have been reported [10, 11, 14]. Increases in biomass and grain yield due to N fertilization have been reported for old and new genotypes [14, 28]. However, other reports found that the modern semidwarf cultivars benefitted more from the high N rate compared to old cultivars [8, 11]. Our results showed that the grain yield of both old and new cultivars increased. Nevertheless, the percent increases in grain yield of the old cultivars were less than the mean value while those of the new cultivars were always higher than the mean value. It was also noticed that the old cultivars (Beladi and Giza 155) had the highest biomass at N₁₂₉; however, this was not reflected in their grain yield and thus resulted in decreased HI. One of the new cultivars (Tagana) showed the same trend of increased biomass and decreased HI at N₁₂₉ probably because of the tallness of this cultivar compared with the modern ones.

The increases in grain yield under the four N levels are associated significantly with the counterpart increases in biomass ($r = 0.86$, $P < 0.001$). It has been observed that some genotypes efficiently used the increases in biomass under high N levels for higher grain yield production (e.g., Khalifa and Elnielain), whereas other genotypes were less efficient in utilizing the high biomass for high grain yield (e.g., Giza 155 and Tagana). Biomass accumulation is regarded as one of the key traits for high yield under heat stress [18, 20]; however, efficient remobilization of these assimilates to grain under heat stress is crucial [16].

Genotypes differentially responded to N levels in terms of various traits suggesting the existence of wide variation in N utilization efficiency among the studied genotypes.

TABLE 5: Straw and grain nitrogen uptake (kg/ha), nitrogen harvest index (NHI), and nitrogen utilization efficiency (NUTE) of 12 bread wheat genotypes grown in season 2008 under four N levels at Gezira Research Station Farm, Wad Medani, Sudan.

	Straw nitrogen uptake	Grain nitrogen uptake	Nitrogen harvest index	Nitrogen utilization efficiency
Genotype:				
Beladi 60	26.5	34.0	0.56	36.1
Giza 155 71	25.3	37.0	0.59	35.2
Condor 78	18.8	44.6	0.70	38.8
Debeira 82	22.9	28.7	0.55	47.4
Wadi Elneel 87	21.2	37.9	0.63	41.8
Elnielain 90	20.2	40.7	0.65	41.0
Nesser 96	18.7	43.6	0.68	39.4
Argine 96	21.3	52.1	0.70	34.1
Imam 00	22.3	37.7	0.62	36.6
Tagana 04	24.6	33.6	0.56	36.1
Khalifa 04	19.7	42.5	0.68	41.3
Bohaine 06	17.7	51.9	0.72	37.8
Significance level	***	***	**	*
SE±	1.55	3.42	0.027	2.47
N level:				
N ₀	15.8	26.8	0.63	—
N ₄₃	19.5	31.3	0.61	43.8
N ₈₆	24.4	42.4	0.62	38.8
N ₁₂₉	26.7	60.9	0.68	33.7
Significance level	***	***	**	***
SE±	0.77	1.71	0.013	1.24
CV %	20.5	23.0	12.5	19.1

*, **, ***Significant at P = 0.05, 0.01, and 0.001, respectively.

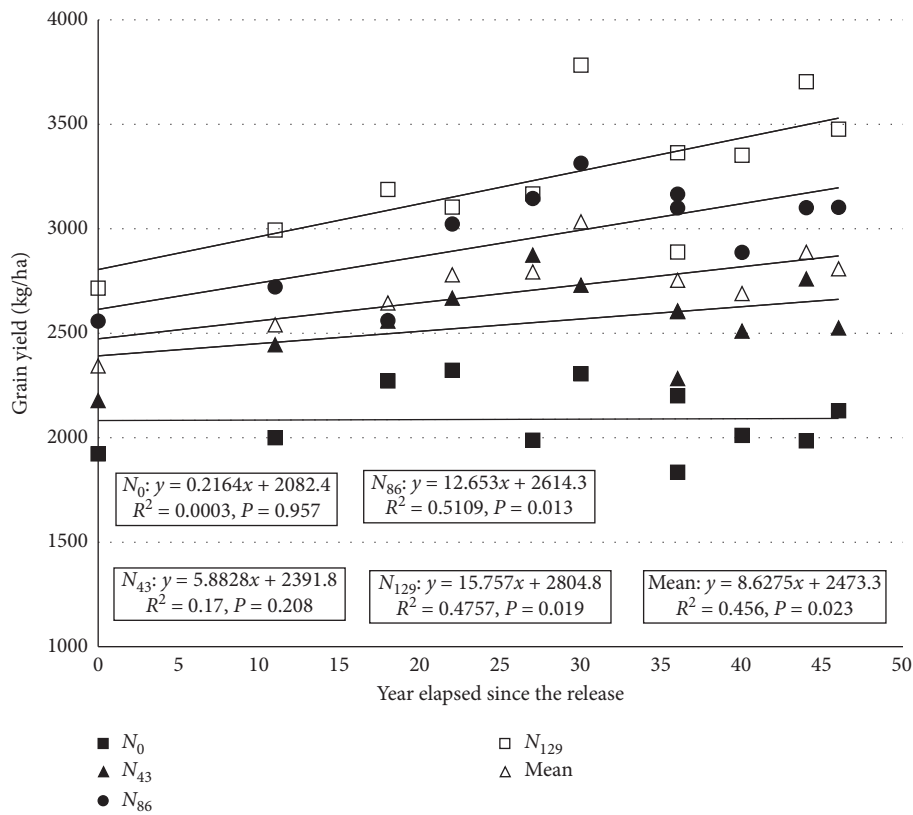


FIGURE 2: Relationship of grain yield (kg ha⁻¹) of 11 bread wheat cultivars released between 1960 and 2006 and grown at four N levels with the time elapsed since their year of release.

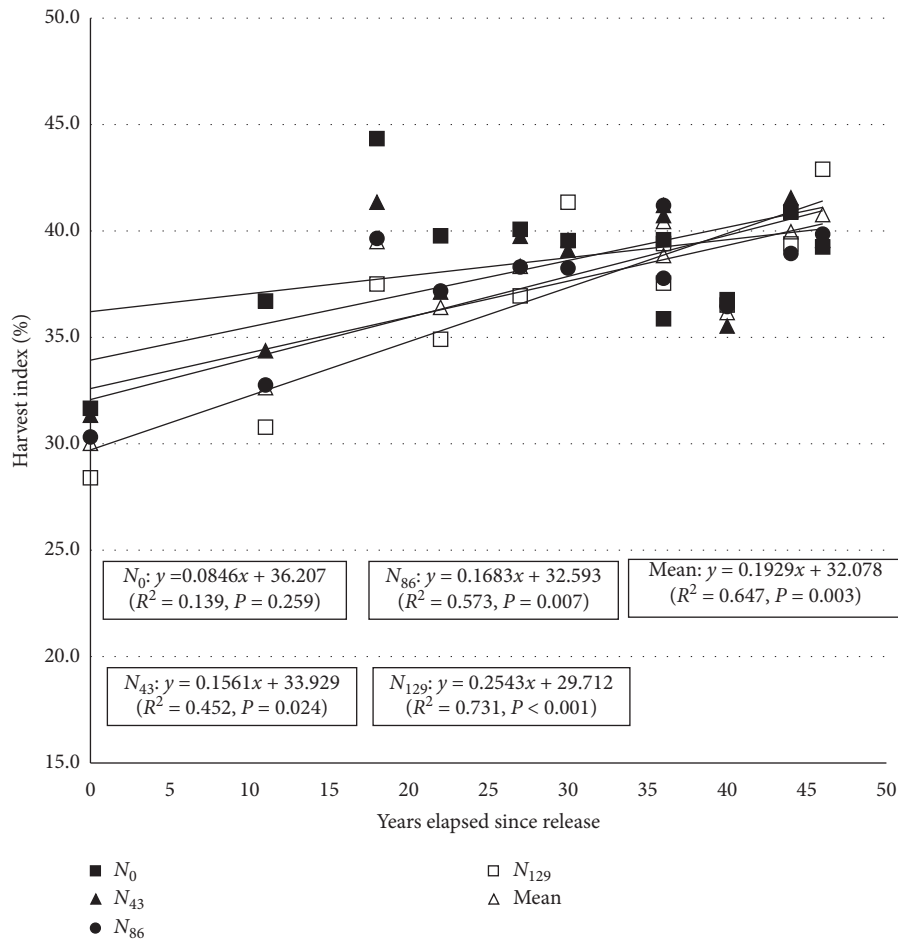


FIGURE 3: Relationship of harvest index (%) of 11 bread wheat cultivars released between 1960 and 2006 and grown at four N levels with the time elapsed since their year of release.

Genotypic variation observed in this study for N utilization efficiency and responses supports the thought of including low-input selection environments to maximize selection gains for breeding programs targeting production with low N input [29]. However, this should not be done at the expense of grain quality which is expected to decrease under low N input.

The grain N uptake of the 12 wheat cultivars differed across N levels. Most of the modern semidwarf cultivars revealed positive and significant increases in grain N uptake. In contrast to the oldest cultivar used in this study (Beladi), the recent cultivars (Argine and Bohaine) recorded the highest N uptake. On the other hand, the highest N uptake in straw across all nitrogen levels was recorded by the tall and old cultivars such as Beladi, Giza 155, and Tagana, whereas the semidwarf cultivars like Bohaine, Nesser, and Condor recorded the lowest biomass N uptake.

Nitrogen capture would appear to be the key underpinning trait aligned to its use efficiency. The Nitrogen uptake efficiency is defined as the amount of N taken up by the crop as a function of the available N. At the crop scale, this includes fertilizer, soil N, and atmospherically deposited N [6]. High yields of high quality grain can only be achieved with high uptakes of N [1]. It has been observed that while

Bohaine recorded higher grain N uptake, it showed significantly the lowest straw N uptake which might be a reflection of high N translocation capacity of Bohaine compared to other genotypes. Nitrogen and carbohydrate translocation is an important trait because it could serve as an alternative source of assimilates and compensate for the reduced photosynthesis under heat stress conditions [16].

Nitrogen harvest index (NHI) is used also as indicator of genotypic efficiency in nitrogen distribution between vegetative plant parts and the grain; it represents the part of plant nitrogen used for protein synthesis. Grain N is for a large part derived from canopy N remobilized during grain filling as the canopy senesces which is more efficient in modern cultivars. Ability of genotype to relocate nitrogen from vegetative parts to grain represents a base for grain protein increase and thus the NHI could be recommended as a selection criterion for nitrogen use efficiency improvement [30]. However, Barraclough et al. [1] reported that the NHI is already high in modern wheat and almost insensitive to N input. Barraclough et al. [31] reported that the development and use of wheat cultivars with higher NUE can contribute to the reduction of the applied N amounts without decreasing grain yield. Our results showed that the modern semidwarf cultivars such

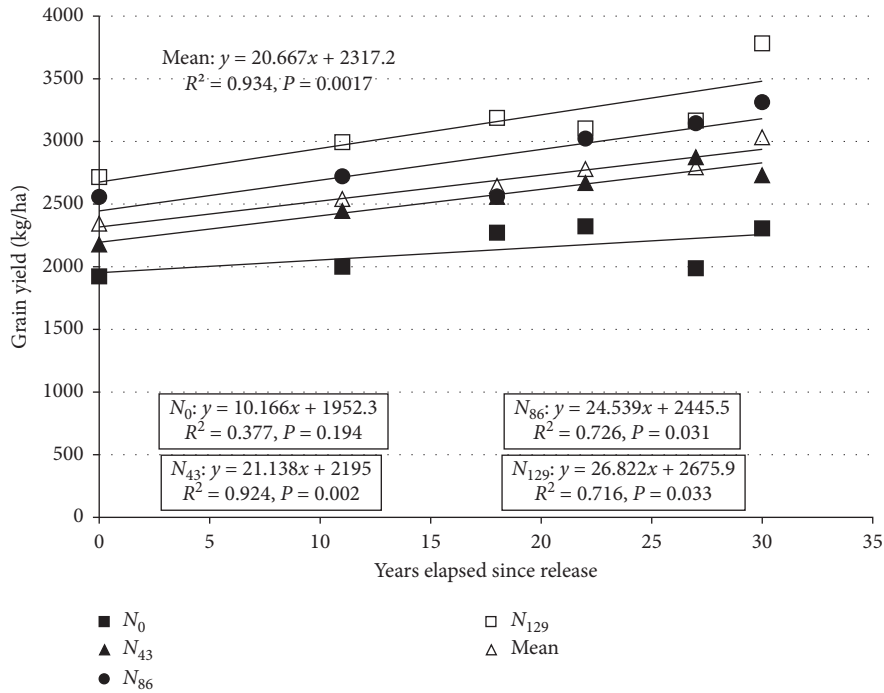


FIGURE 4: Relationship of grain yield (kg ha⁻¹) of six bread wheat cultivars released between 1960 and 1990 and grown at four N levels with the time elapsed since their year of release.

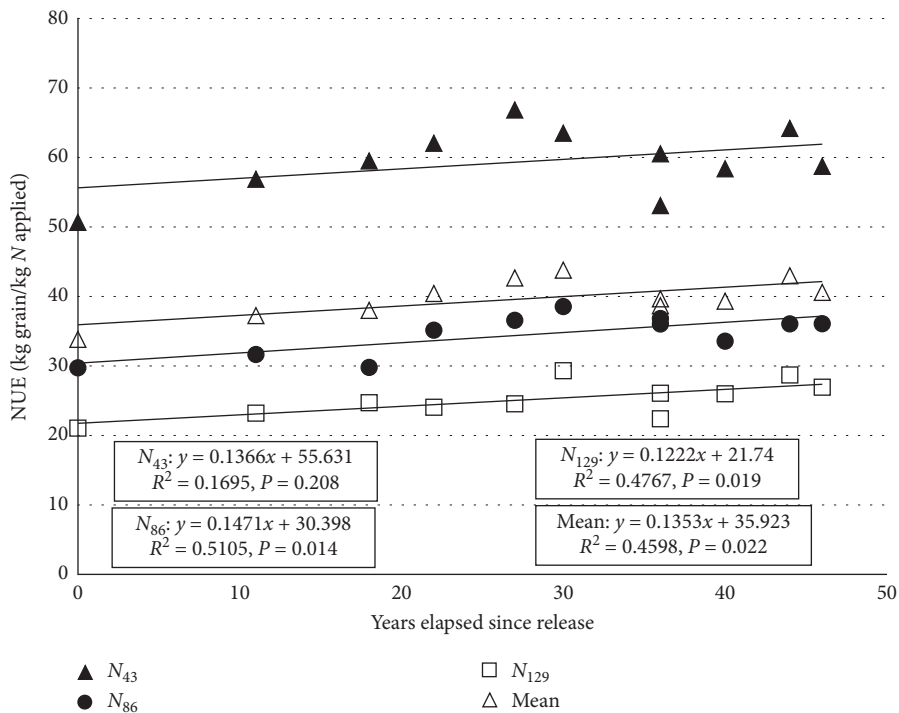


FIGURE 5: Relationship of NUE of 11 bread wheat cultivars released between 1960 and 2006 and grown at different N levels for two seasons (2006/07 and 2007/08) with the time elapsed since their year of release.

as Bohaine, Argine, and Condor showed significantly higher NHI across all N levels, whereas the tall cultivars (Beladi and Tagana) recorded the lowest NHI.

The grain yield produced from a unit of above ground plant N uptake, i.e., nitrogen utilization efficiency, NUTE, was significantly reduced with each increase in N rate.

Across all N levels, the 12 wheat cultivars significantly varied in NUTE. With the exception of few modern cultivars (e.g., Argine and Tagana), NUTE was always higher in the semidwarf cultivars such as Debeira, Wadi Elneel, Khalifa, and Elnielain compared to the old tall cultivars. The aforementioned results showed that modern semidwarf cultivars are more efficient in utilizing the applied N than the old tall cultivars. The variability of modern cultivars' response to NUE has been attributed mainly to NUTE [1]. The differential response of cultivars with different N supply levels, regarding the NUTE, is important when defining the management and choosing the cultivar [32].

4.2. Genetic Gain in Grain Yield under Different Nitrogen Levels. Different rates of genetic gains were found under different N levels. The result of the genetic gain at N_{86} and N_{129} showed clearly that selection of the genotypes under these N levels reflected in the progress made in grain yield. Under different N levels, different rates of genetic gains were obtained. Ortiz-Monasterio et al. [12] reported that the genetic gains in grain yield at no or low N levels (0 and 75 kg of N/ha) were 32 and 43 kg/ha/year, respectively, whereas at high N levels (150 and 300 kg of N/ha) the genetic gains were 59 and 89 kg/ha/year, respectively. Our results under the four N levels did not follow exactly the same trend especially under N_0 and N_{43} treatments, suggesting that the progress made in grain yield was affected by the conditions under which the selection of the cultivars has been made. Under the low N levels, old and modern cultivars responded almost similarly and no significant progress has been found. The significant positive trends at N_{86} and N_{129} showed that addition of N fertilizer was more beneficial to the modern cultivar than the old ones. It has been suggested that in order to identify the most efficient type for N use, the breeders need to test their material in low as well as high N environments [33]. On the other hand, similar genetic progress was reported at both high and low N treatments for 195 European winter wheat varieties [23]. However, direct selection at low N condition was recommended when the low N treatment is targeted.

Regressing the grain yield at N_{86} in the two seasons against the year of release, the annual rate of increase in grain yield was $12.65 \text{ kg ha}^{-1} \text{ year}^{-1}$. This genetic gain in grain yield is comparable with most of the reports in many countries such as Argentina [34], Canada [35], UK [36], and USA [37]. Higher rates of increase were reported under some favorable environments such as Mexico [12, 38, 39] and the United Kingdom [40]. On the other hand, lower rates were reported in Australia [41] and India [42]. It has been reported that increasing grain yield under stressed environments is a difficult task when compared with favorable environments [43]. This is true with the conditions of Sudan, where the major wheat growing areas experience high temperature during most parts of the crop cycle (Figure 1). Despite this, a reasonable level of improvement was achieved. However, when the period from 1960 to 1990 was considered, the genetic gains in grain yield at different N levels were higher. For instance, the genetic gain for the

mean grain yield across all N levels was estimated to be $20.67 \text{ kg ha}^{-1} \text{ year}^{-1}$ (Figure 4). At N_{86} (the recommended N dose for wheat production in Sudan), the genetic gain was $24.5 \text{ kg ha}^{-1} \text{ year}^{-1}$, a figure which is closer to what has been reported earlier under the similar N level [25]. This showed that the rate of the genetic gain was slowed down in the recent years. The decelerated rate of genetic gain in grain yield has been reported elsewhere without being plateaued [38, 44]. Noticeably, significant genetic gain was achieved during the period 1960 to 1990 under the low N level of N_{43} ($21.14 \text{ kg ha}^{-1} \text{ year}^{-1}$, $R^2 = 0.924$, $P = 0.002$) (Figure 4).

4.3. Genetic Gain in NUE under Different Nitrogen Levels. The genetic gain in NUE under different nitrogen levels showed more or less similar trends of that of the grain yield. Although it has been observed that at the three N levels, the recently released semidwarf cultivars exhibited relatively higher NUEs compared to the old and tall ones (Beladi and Giza), the trend was stronger at higher N levels. This might reflect the effect of selecting the new varieties under high N level (mainly under N_{86}). For further improvement in NUE, it would be useful to enrich the genetic variation and test and select genotypes at early stages under different N doses especially low levels. Ortiz-Monasterio et al. [12] measured the genetic progress in grain yield and NUE, particularly uptake efficiency (UPE) and utilization efficiency (UTE) using tall vs semidwarf wheat cultivars and reported that the progress in NUE resulted in the improvement of both UPE and UTE. Significant enhancement of NUE at both low and high N levels was reported [23], but more efficiently at low N level which demonstrated the higher yield stability of recent cultivars. Improvement in nitrogen use efficiency in the Great Plains' hard winter wheat germplasm in the period from 1960 to 2014 was found to be associated with significant trends of grain yield, grain N yield, nitrogen harvest index, nitrogen uptake efficiency, and postanthesis nitrogen uptake with year of release [24].

It is concluded that genotypes differentially responded to N levels in terms of various traits suggesting the existence of some level of variation in NUE among the studied genotypes. The set of released cultivars used in this study demonstrated that breeders improved grain yield and NUE over 46 years of crop improvement under the heat stress conditions of Sudan although the rate of increase in grain yield has been slowed down in recent years. The results also indicate that the best grain yields could be achieved when modern cultivars are used even under limited N levels. The selection of the cultivars under the optimum and high N conditions resulted in the genetic gain in grain yield under these conditions but not under no or low N doses. Therefore, for better improvement in NUE, simultaneous testing of wheat genotypes under different N levels would be useful especially under low doses. Due to the slowdown in genetic gain reported in recent years, new integrated approaches would be needed for further improvement including broadening the genetic base for abiotic stress tolerance especially in the light of the ensuing climate change.

Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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