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Efficient rates of nitrogenous fertiliser for irrigated sweet sorghum cultivation during the post-rainy season in the semi-arid tropics

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ABSTRACT

Sorghum (*Sorghum bicolor* (L.) Moench) is a multipurpose crop with high tolerance to environmental stresses. To meet the increased demand for food and biofuel, current agricultural practices rely on the excessive use of inorganic nitrogen (N) fertiliser. However, excessive N fertiliser has resulted in negative environmental effects. In view of the varied N use efficiency (NUE) of plants under different environmental conditions, the aim of this study was to evaluate the efficient rates of N fertiliser in semi-arid tropics for sweet sorghum cultivation during post-rainy season by maximising NUE without compromising yield. Field experiments were conducted on two sweet sorghum cultivars with four different N fertilisation rates (0, 63, 90 and 150 kg N ha^{-1}) during the post-rainy season in India. Grain and stalk yields increased with N fertiliser, but significantly only up to 90 kg N ha^{-1}. The observed increases in grain yield were attributed by increases in kernel numbers. Corresponding with the differences in biomass, both relative growth rate (RGR) and crop growth rate (CGR) increased with N fertilisation rate up to 90 kg N ha^{-1}. Component analyses of RGR and CGR revealed that both net assimilation rate (NAR) and leaf area index (LAI) significantly contributed with increasing rates of N fertiliser applications. Furthermore, studies of NUE indices showed that agronomic N use efficiency (ANUE, indicating yield production per unit of fertiliser N) responded comparably up to 90 kg N ha^{-1}, and decreased significantly thereafter. Analysis of ANUE components showed that the decline in ANUE at 150 kg N ha^{-1} was due to a decrease in physiological N use efficiency (PNUE), indicating that the absorbed N was not utilised efficiently for biomass and yield production, but merely accumulated. These results together suggest that 90 kg N ha^{-1} is an efficient N fertilisation rate suggested among the tested treatments for sustainable sweet sorghum cultivation during the post-rainy season in the semi-arid tropics.

1. Introduction

Sorghum (*Sorghum bicolor* (L.) Moench), the fifth major food worldwide after maize, wheat, rice and barley (Paterson, 2008), is also staple food for half a billion people, particularly in the semi-arid tropics such as south Asia and sub-Saharan Africa (Mace et al., 2013). Sorghums, as a C_4 crop with high tolerance to environmental stresses, such as drought and heat, grow relatively well under adverse conditions (Teetor et al., 2011). For this reason, sorghum is cultivated mainly as an off-season crop (Filho et al., 2014) during the post-rainy season mainly in developing countries, including India, where day length is short and only residual moisture from the previous rainy season is used for cultivation. Sweet sorghum is a type of sorghum in which sucrose is accumulated in the stems in high concentrations, up to 15.5% (Regassa and Wortmann, 2014; Vinutha et al., 2014). These features, together with a relatively short growing time (3–5 months) as compared with sugar cane (7–12 months) (Almodares and Hadi, 2009) and higher energy use efficiency as compared with maize (Regassa and Wortmann, 2014) increase the attraction of sweet sorghum as an energy crop. Furthermore, sweet sorghum is a multipurpose crop with other uses such as grains for food and bagasse and leaves for fodder (Uchino et al., 2013).

With the rise in global population and the consequent food demand, there is an increasing need for agricultural production of biofuel as an alternative energy resource. To meet these increased

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Abbreviations: ANRE, apparent nitrogen recovery efficiency; ANUE, agronomic nitrogen use efficiency; C, carbon; CGR, crop growth rate; DAS, days after sowing; DM, dry matter; EONR, economically optimum nitrogen rate; HI, harvest index; LAI, leaf area index; LAR, leaf area ratio; N, nitrogen; NAR, net assimilation rate; NHI, nitrogen harvest index; NUE, nitrogen use efficiency; PFP, partial factor productivity; PNUE, physiological nitrogen use efficiency; RGR, relative growth rate; Yp, the maximum yield potential.

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demands, current agricultural practices rely on the use of inorganic nitrogen (N) fertiliser (Kurai et al., 2011). N is a crucial nutrient for maximising the yield for farmers, particularly in developing countries where there is a large yield gap (Good and Beatty, 2011). Consequently, farmers tend to apply N fertilisers in excess for assuring yield (Sheriff, 2005). However, the use of large amounts of N fertiliser has resulted in negative environmental effects, such as pollution by nitrate leaching and nitrous oxide emission (Ramu et al., 2012) as well as ecological imbalance (Miller and Cramer, 2004).

One of the possible solutions to this situation is to enhance N use efficiency (NUE) of crops while maintaining the yield, and thus increasing the financial income of farmers. Among the multiple indices available for NUE assessment, agronomic NUE (ANUE) is the most informative in this case, as it describes grain yield per unit of N applied as fertiliser, indicating benefit-to-cost ratio (Cassman et al., 1996). As ANUE is a function of N uptake and N utilisation efficiency, component analysis of ANUE reveals whether uptake and/or utilisation of N is affected in a given environment. In maize, NUE was strongly affected by the utilisation of absorbed N under low N conditions, whereas N uptake efficiency was unaffected under high N conditions (Moll et al., 1982).

N availability is known to affect plant growth (Aerts and Chapin, 2000), and plants respond plastically to the surrounding environment both morphologically and physiologically (Useche and Shipley, 2010). Growth rate differences due to experimental treatments are often compared in the form of relative growth rate (RGR), which is a product of net assimilation rate (NAR) and leaf area ratio (LAR), responding physiologically and morphologically respectively. Changes in plant growth, particularly under limited N, affect plant biomass and grain yield. Crop growth rate (CGR) is an index of crop dry matter (DM) production, and can be divided into two components, NAR and leaf area index (LAI). LAI is another plant growth parameter known to be affected by N availability, and N uptake is reported to be directly proportional to LAI in many crops (Hirel et al., 2007). These component analyses of plant growth can be used to quantitatively interpret the effects of N fertilisation on plant development.

Muchow (1998) reported sorghum's variable response to N fertiliser depending on climatic, soil and genotypic conditions. We have previously reported optimum N fertiliser rates on sorghum during rainy season (Uchino et al., 2013). However, due to common practices of sorghum cultivation during post-rainy season in southern India, it is important to optimise N-fertilizer rates for the post-rainy season sorghum cultivation as well. In addition, no study has conducted NUE assessment in relation to plant growth analysis of sorghum in the semi-arid tropics to date. The aim of this study was to identify the efficient N fertiliser application rates for sustainable sorghum cultivation during the post-rainy seasons in the semi-arid tropics with focuses on assessments of growth analysis and multiple NUE indices in addition to yield in response to multiple N rates.

2. Materials and methods

2.1. Field experimental sites and conditions

The field experiment was conducted at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) at Patancheru (17.53° N, 78.27° E, 545 m above sea level) near Hyderabad, India, during the post-rainy season (from late October to early March) of 2011–12 and 2012–13. The climate of experimental site is semi-arid with an average annual rainfall of 750–800 mm, approximately 80% of which is received between June and October (Ramu et al., 2012). Average air temperatures of the maximum and the minimum as well as amounts of precipitation for the first year and the second year of the experiments are illustrated in Fig. 1A and B, respectively. The same experimental plot was used throughout the experiment. Maize (*Zea mays* L.) was pre-cultivated each year for few months prior to the experiments without any fertiliser to deplete residual N, and the whole maize plants including roots were completely removed from the experimental site before the initiation of experiments each year. The main soil properties at various depths at the experimental sites before the initiation of experiments were measured according to Sahrawat and Wani (2013) and summarised in Table 1. In brief, available phosphate, potassium, sulphur, boron, and zinc were measured using sodium bicarbonate, ammonium acetate, calcium chloride, hot water and diethylenetriaminepentaacetic acid (DTPA) as extractants respectively. The soil of the experimental site is categorised to an Alfisol [Udic Rhodustalf by USDA classification (Soil Survey Staff, 1999) or Ferric Luvisols by FAO classification (FAO-UNESCO, 1977)]. The soil of the experimental site consists of sand [73.0% (0–18 cm) and 43.3% (18–71 cm), 2.0–0.2 mm in size], silt [6.0% (0–18 cm) and 5.6% (18–71 cm), 0.02–0.002 mm in size], and clay [(21.0% (0–18 cm) and 51.1% (18–71 cm), less than 0.002 mm in size] (El-Swaify et al., 1985).

2.2. Experimental design

The experiments were arranged as completely randomised block designs with three replications. Each plot had a size of 4.8 m × 9 m with a row spacing of 60 cm and plant-to-plant spacing of 20 cm, resulting in an average planting density of 8.3 plants m^{-2}. A promising sweet-sorghum hybrid in India, CSH 22SS, and a widely and commercially cultivated sweet-sorghum variety, ICSV

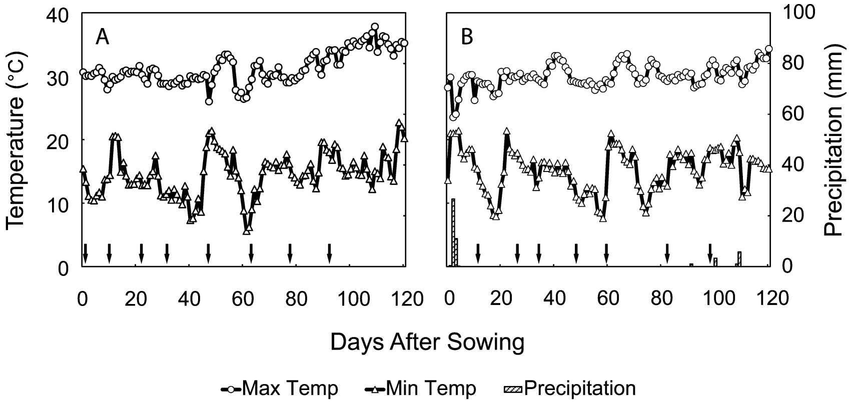

Fig. 1. Daily maximum and minimum air temperatures as well as precipitations were recorded in the experimental sites during the growing periods for the years 2010–2011 (A) and 2011–2012 (B). Black arrows indicate furrow irrigations provided every 2 weeks at an approximate depth of 90 mm.

Table 1
General characteristics of the soil at the experimental site before sowing.

Soil depth (cm)	0–15	15–30	30–60	60–90
pH (H_2O; 1:2)	6.86 ± 0.06	7.20 ± 0.12	7.42 ± 0.07	7.51 ± 0.16
EC (dS m^{-1})	0.12 ± 0.01	0.09 ± 0.01	0.10 ± 0.02	0.11 ± 0.02
Organic C (%)	0.51 ± 0.04	0.30 ± 0.04	0.33 ± 0.07	0.22 ± 0.12
Total N (mg kg^{-1})	690.98 ± 57.24	706.54 ± 113.98	716.94 ± 11.13	532.82 ± 114.39
Olsen P (mg kg^{-1})	6.07 ± 1.00	3.54 ± 0.75	3.28 ± 1.17	3.19 ± 0.49
Extractable K (mg kg^{-1})	150.92 ± 15.28	125.48 ± 13.58	135.62 ± 3.66	129.81 ± 23.63
Extractable S (mg kg^{-1})	4.04 ± 0.78	3.79 ± 1.14	3.82 ± 1.08	4.70 ± 1.49
Extractable B (mg kg^{-1})	0.33 ± 0.08	0.47 ± 0.14	0.51 ± 0.09	0.41 ± 0.05
Extractable Zn (mg kg^{-1})	13.33 ± 0.81	6.07 ± 2.04	3.97 ± 0.85	5.17 ± 1.68

Values are the means ± SD of 4 replicates.

93046, were selected for the experiments. In addition to high risks and instability associated with sorghum cultivation during post-rainy season in the semi-arid tropics, where precipitation is little, high costs of fertilizers relative to average income and lower returns in the Indian market makes it necessary for farmers to attain the economical maximum efficiency of N fertilizer (Jodha, 1973). Our previous work on sorghum in rainy seasons showed that net income was maximised between 90 kg to 120 kg N ha^{-1}, and both biomass and grain yield were found to be increased up to 90 kg N ha^{-1} (Uchino et al., 2013). Accordingly, four different N rates (0, 63, 90 and 150 kg N ha^{-1}, designated as 0 N, 63 N, 90 N and 150 N, respectively) were tested in this study.

2.3. Crop management

Sorghum seeds were sown on 15 November in 2011 and 31 October in 2012. All N fertiliser was applied in the form of urea (46% N) by split application of basal and two topdressings at 30 and 60 days after sowing (DAS). Basal N fertiliser was fixed at 30 kg N ha^{-1} across the treatments, and the remainder was equally split for two topdressed applications, except for 63 N, where 18 and 15 kg N ha^{-1} was applied at the first and second topdressings, respectively (Supplementary Table 1). Other macro- and micronutrients were also applied before sowing as follows: 40 kg P ha^{-1} as triple superphosphate (46% P), 30 kg S ha^{-1} as Gypsum (15% S), 0.475 kg B ha^{-1} as sodium pentaborate (19% B) and 20 kg Zn ha^{-1} as zinc sulphate (40% Zn). All fertilisers including N fertiliser were applied and mixed on the top of ridges at a depth of 5–10 cm. Irrigation was applied as furrow irrigation every two weeks at an approximate irrigation depth of 90 mm (Fig. 1A and B). Insecticide treatment was applied as needed. No plant residue was introduced or carried over from the cultivation of the previous year.

2.4. Plant sampling and physiological data collection

Nine sorghum plants were randomly selected from each plot and harvested from ground level at 30, 60 and 90 DAS and physiological maturity. Samplings at physiological maturity were performed on 20 March in 2012 and 5 March in 2013, which corresponded to 125 DAS and 124 DAS respectively. For each plant, the plant height and leaf area were measured with a leaf area metre (LI-3100 model; LI-COR, Lincoln, NE). Samples were then separated into leaf blades, stems and grains, and dried to a constant weight in an 80 °C oven for a minimum of 72 h for DM measurement. Total DM was determined for the above-ground parts of the whole plant and calculated as the sum of DMs of leaf blades, stems and grains (if any).

Furthermore, eight sorghum plants were separately and randomly harvested from each plot at a physiological maturity for sugar yield analysis. Immediately after the harvest, plants were stripped of panicles and leaves. The fresh weights of the remaining stalks were measured and the stalks were then crushed using a three-roller mill to extract juice. Juice volume was recorded and sugar concentration in the juice was individually measured with a digital refractometer (PAL-1 model; Atago, Tokyo, Japan). Sugar yield was estimated using the following equation (Uchino et al., 2013):

$$\text{Sugar yield}\,(\text{t ha}^{-1}) = \{(\text{Brix}\,(\%) \times 0.8746) + 0.1516\}/100 \times \text{juice yield}\,(\text{m}^3\ \text{ha}^{-1}) \quad (1)$$

Similarly, potential ethanol yield was calculated by the following equation (Spencer and Meade, 1963):

$$\text{Potential ethanol yield}\,(\text{m}^3\ \text{ha}^{-1}) = \text{juice yield}\,(\text{m}^3\ \text{ha}^{-1}) \times \text{Brix}\,(\%)/100 \times (0.85/1.76) \quad (2)$$

For yield analysis, sorghum plants were harvested from an area of 18 m^2, equivalent to the area for 150 plants. Seeds were carefully threshed from each plant and kernel numbers were determined by seed-counter, followed by drying to a constant weight in a 60 °C oven for a minimum of 120 h. Grain DM was measured for the estimation of yield per hectare. Yield response curves were drawn based on these measured grain DM and rates of N fertilisers. Economically optimum N rate (EONR) was calculated by the following equation (Bachmaier, 2012).

$$\text{EONR}\,(\text{kg ha}^{-1}) = (r \cdot \text{p}^{-1} - \beta_1)/2\beta_2 \quad (3)$$

where r is N fertiliser price (in € kg^{-1}) and p is the product price of sorghum (in € t^{-1}), and € 0.8 kg^{-1} and € 160 t^{-1} are applied respectively. Two constant, β_1 and β_2, are from quadratic yield functions ($Y = \beta_0 + \beta_1(N\,\text{rate}) + \beta_2(N\,\text{rate})^2$) derived from yield response curves.

2.5. Growth analysis

Using measured leaf DM, total DM and leaf area, RGR, NAR and LAR were calculated (Hunt et al., 2002). CGR was calculated as the product of NAR and LAI.

2.6. N content determination and NUE analyses

N concentrations of leaf, stem and grain were determined by digesting dried plant materials collected at physiological maturity with sulphuric acid–selenium mixture, followed by ammonium determination with autoanalyzer (San^{++} model; Skalar Analytical B.V., Breda, The Netherlands; Sahrawat et al., 2002). The N content of each tissue was calculated as the product of the measured N concentration and DM. Total N was calculated for the above-ground parts of the whole plant as the sum of N contents of leaf blades,

Table 2
Results of analysis of variances for combined years.

Source	df	Plant Height	Total DM[a]	Grain DM	1000-Seed weight	Kernel number	HI[b]	Brix	Stalk yield	Juice yield	Sugar yield	Potential ethanol yield	Total N	Grain N	NHI[c]
C	1	ns	**	**	**	**	**	**	ns	ns	*		ns	ns	**
Y	1	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
N	3	**	**	**	**	**	ns	ns	**	**	**	**	**	**	ns
C×Y	1	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
C×N	3	ns	ns	*	ns	*	**	ns	ns	ns	ns	ns	ns	ns	*
Y×N	3	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
C×Y×N	3	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Error	200														

The effects of cultivar (C), year (Y), nitrogen fertilizer rates (N), and their interactions were analysed and displayed. The single asterisk (*) indicates the statistically significant effect at $P < 0.05$. The double asterisk (**) indicates the statistically significant effect at $P < 0.01$. The letter 'ns' indicates statistically not significant.

[a] DM: dry matter.

[b] HI: harvest index.

[c] NHI: nitrogen harvest index.

stems and grains (if any). Various types of NUE were calculated using the following equations:

$$\text{N harvest index (NHI)} = \text{total N in grain/total N of above-ground parts} \quad (4)$$

$$\text{Partial factor productivity (PFP)}\,(\text{kg}\cdot\text{kg N}^{-1}) = \text{grain yield/amount of N fertiliser} \quad (5)$$

$$\text{Agronomic N use efficiency (ANUE)}\,(\text{kg}\cdot\text{kg N}^{-1}) = (\text{grain yield with N fertiliser} - \text{grain yield without N fertiliser})/\text{amount of N fertiliser applied} \quad (6)$$

$$\text{Apparent N recovery efficiency (ANRE)} = (\text{total plant N absorbed with N fertiliser} - \text{total plant N absorbed without N fertiliser})/\text{amount of N fertiliser applied} \quad (7)$$

$$\text{Physiological N use efficiency (PNUE)}\,(\text{kg}\cdot\text{kg N}^{-1}) = (\text{grain yield with N fertiliser} - \text{grain yield without N fertiliser})/(\text{total plant N absorbed with N fertiliser} - \text{total plant N absorbed without N fertiliser}) \quad (8)$$

2.7. *Statistical analysis*

All data were statistically evaluated for normality and homogeneity of variance by Shapilo–Wilk test and Levene's test, respectively. Statistical significance of differences between mean values was determined using analysis of variance (ANOVA), followed by post hoc comparisons using Tukey's tests on the basis of $P < 0.05$. Regression analyses were performed to determine relationship between rates of N fertilizers and grain DM or ANUE. Statistical significance of a correlation was evaluated by ANOVA. Values of coefficient of determination (R^2) were used to evaluate the fitness of the model. All statistical analyses were conducted using SPSS 13.0 for windows (SPSS Inc., Chicago, IL).

3. Results

No significant effects ($P < 0.05$) on 'year', 'year × N treatment' and 'cultivar × year × N treatment' interactions were found in any of the parameters/attributes measured at harvest in our study (Table 2). Therefore, all the data over the two years were pooled and are presented.

3.1. *Plant height, biomass production and yield attributes*

Plant height under the 0 N treatment was significantly lower than that under all other N treatments for both CSH 22SS and ICSV 93046 at physiological maturity (Table 3). However, no significant difference was observed among other N fertiliser rates for either cultivar (Table 3).

Similar to plant height, total DM and grain DM at physiological maturity were both lowest under the 0 N treatment compared to that under any other N treatment in both CSH 22SS and ICSV 93046 (Fig. 2A–D). Mean values of total DM, grain DM and kernel numbers increased in response to N input (Fig. 2A–D, Table 3). Although no statistical difference was found between the 90 N and 150 N treatments, yield response curve drawn against N fertiliser rate displayed quadratic responses for both cultivars tested (Fig. 2C and D). Calculated EONR using regression equation were found as 133.5 and 144.5 kg N ha^{-1} for CSH 22SS and ICSV 93046, respectively (Fig. 2C and D). Total DM and grain DM were proportional

Table 3
Physiological properties of sweet sorghum cultivars grown under multiple N regimes during the post-rainy seasons in the semi-arid tropics.

Cuitivar	N Treatment (kg N ha^{-1})	Plant height (kg N ha^{-1})	1000 seed weight (g)	Kernel number (plant^{-1})	Harvest index (%)
CSH 22SS	0	175.5 ± 11.1a	30.9 ± 1.5a	1145 ± 408a	53.2 ± 3.0a
	63	203.8 ± 13.4b	31.2 ± 2.3a	2124 ± 133b	55.3 ± 4.5a
	90	197.8 ± 11.1b	32.2 ± 1.2a	2787 ± 454bc	56.6 ± 3.9a
	150	204.9 ± 15.7b	31.7 ± 1.8a	3171 ± 199c	54.0 ± 3.7a
ICSV 93046	0	171.5 ± 8.2a	26.4 ± 0.8a	934 ± 88a	43.8 ± 5.0a
	63	200.5 ± 5.4b	28.4 ± 1.4a,b	1366 ± 126b	41.5 ± 4.2a
	90	196.5 ± 10.5b	29.2 ± 1.1b	1936 ± 218c	43.6 ± 3.3a
	150	195.6 ± 18.8b	29.5 ± 0.4b	2227 ± 192c	43.3 ± 3.9a

Values are the means ± SD of 27 replicates. Letters indicate statistically significant differences ($P < 0.05$), when compared with 0 kg N ha^{-1} treatment.

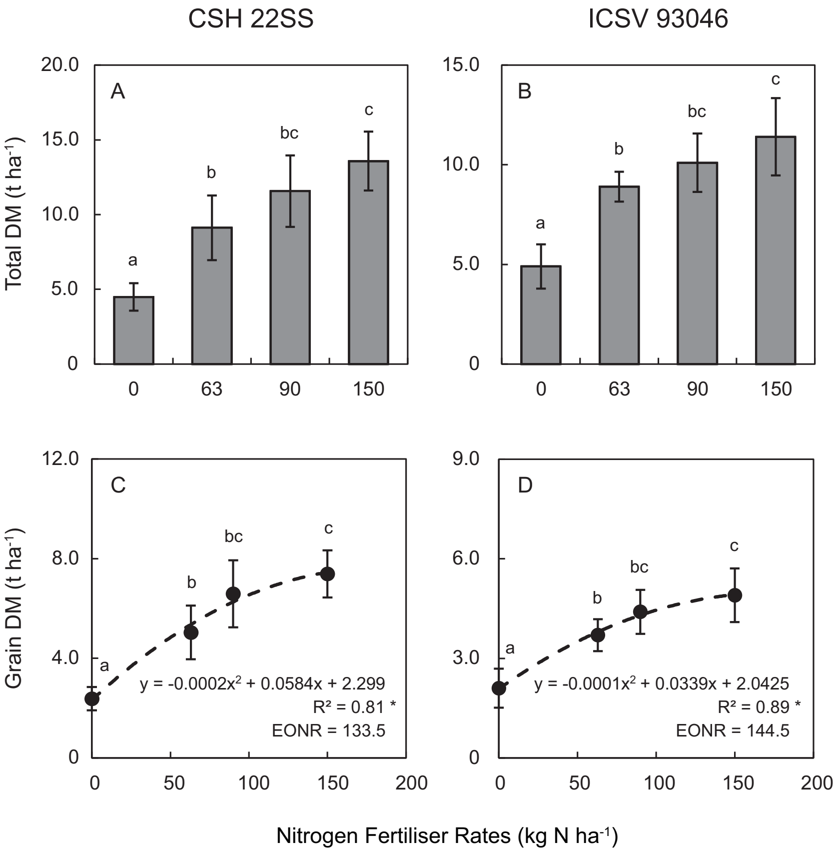

Fig. 2. Effects of N fertiliser rates on total dry matter (DM) (top: A and B) and grain DM (bottom: C and D) of two different sweet sorghum cultivars, CSH 22SS (left half) and ICSV 93046 (right half) were examined at physiological maturity. Values represent mean ± SD of at least 27 replicates. Letters indicate statistically significant differences ($P < 0.05$) between treatments. Furthermore, relationships between N fertilizer rates and grain DM of CSH 22SS (C) and ICSV 93046 (D) were determined. The slopes of regression lines are indicated together with the coefficient of determination (R^2) for each relationship. Economically optimal nitrogen rate (EONR) is calculated based on the slopes of regression with market values of urea and sorghum as € 0.8 kg^{-1} and € 160 t^{-1} respectively. Asterisks indicate statistically significant relationships at $P < 0.01$.

to each other, and this relationship was further confirmed by the almost identical values of harvest index (HI, Table 3). Despite the changes in kernel numbers, 1000-seed weights were comparable among the treatments, except for the 0 N treatment of ICSV 93046, for which it was slightly lower than under the other N treatments (Table 3). These results indicated that although grain filling was unaffected by N treatments, sink size as kernel number was affected, resulting in differences in yield.

3.2. *Growth analysis*

Given that basal N fertiliser was equally applied to all treatments, growth analysis was performed for further evaluation of N-fertiliser effects at two different time periods, 30–60 and 60–90 DAS, representing the effect of the first and second topdressings, respectively. Both cultivars, CSH 22SS and ICSV 93046, exhibited trends of similar response to the amount of N fertiliser at both time periods (Fig. 3A–T).

During 30–60 DAS, RGR increased in response to N fertiliser up to 90 N treatment (Fig. 3A and B). This increase in RGR could be attributed to NAR, as LAR was similar between all treatments (Fig. 3I, J, Q, and R). This result was further supported by a strong correlation of RGR with NAR, but not with LAR, in both cultivars tested (Fig. 4A, B, E, and F).

During 60–90 DAS, RGR showed no significant difference between treatments (Fig. 3C and D). Although NAR was significantly higher in the 90 N and 150 N treatments (Fig. 3K and L), LAR was much lower in the 90 N and 150 N than in the 0 N and 63 N treatments (Fig. 3S and T), resulting in comparable RGRs. Nonetheless the strong correlation between RGR and NAR, but not with LAR, was maintained at this developmental stage in both cultivars (Fig. 4C, D, G, and H).

CGR response to N was similar to RGR during 30–60 DAS (Fig. 3E and F). Similarly, despite the comparable RGR, CGR during 60–90 DAS showed a significant increase up to 90 N treatment (Fig. 3G and H). This response was due to increases in both LAI and NAR in response to increased N (Fig. 3I–P). Irrespective of cultivar, no statistical difference was observed between the 90 N and 150 N treatments for any of the tested parameters (i.e. RGR, CGR, NAR, LAI and LAR; Fig. 3A–T). This finding, together with the CGR results, was in accordance with the response of total DM (Fig. 2A and B), which increased with the amount of N supplied, but not statistically differently between the 90 N and 150 N treatments.

3.3. *Sugar yield*

The Brix values of all treatments regardless of cultivar were found to be low at approximately 7% and not statistically different between N treatments (Table 4). In correspondence with the results of DM (Fig. 2A and B), stalk biomass increased with N fertiliser (Table 4). As both sugar yield and potential ethanol yield depends on Brix value and juice yield (Eqs. (1) and (2)), and the juice yield is determined largely by biomass (i.e. stalk yield), all of juice, sugar, and potential ethanol yield were consequently increased with N fertiliser rates (Table 4). Mean values of these properties were significantly different between the 0 N and 90 N/150 N treatments, but no statistical differences were detected between 90 N and 150 N in either CSH 22SS or ICSV 93046 except potential ethanol yield

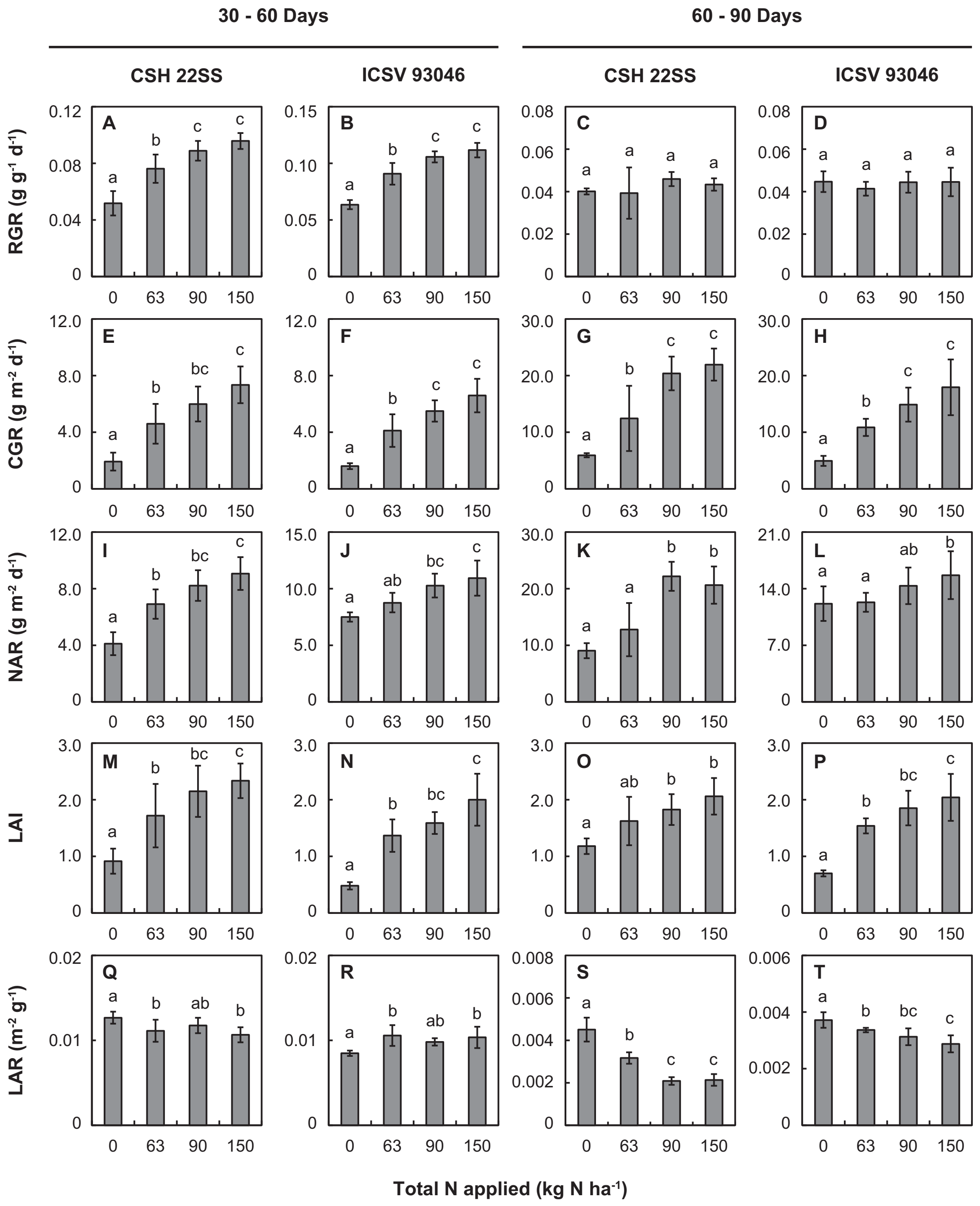

Fig. 3. Effects of N fertilisers on growth parameters were examined as relative growth rate (RGR; A-D), crop growth rate (CGR; E-H), net assimilation rate (NAR; I-L), leaf area index (LAI; M-P) and leaf area ratio (LAR; Q-T) at two different developmental stages, 30–60 days after sowing (DAS) (left half) and 60–90 DAS (right half). For each developmental stage, every growth parameter was analysed on two different sweet sorghum cultivars, CSH 22SS (left) and ICSV 93046 (right). For LAI and LAR, mean values at 60 DAS (left) and 90 DAS (right) are shown for each cultivar. Values represent mean ± SD of at least 27 replicates. Letters indicate statistically significant differences ($P < 0.05$) between treatments.

(Table 4). For the ethanol yield, there was a significant difference detected even between 90 N and 150 N in both CSH 22SS and ICSV 93046 (Table 4).

3.4. N uptake and distribution

It is shown in Table 5 that the total N content of plants increased significantly in both cultivars as N fertilisation rates increased. Results of total N under the 0 N treatment indicated that substantial amounts of N were acquired from native soil N (Table 5). For both CSH 22SS and ICSV 93046, the N surplus, defined as N application rate subtracted by above-ground N uptake (total N in our study), under 63 N and 90 N treatments were relatively small, whereas total N absorbed was equivalent to approximately 80% that of the fertiliser N for the 150 N treatments, resulting N surplus being large in excess. For example, the N fertiliser applied was 63, 90 and 150 kg N ha^{-1}, but total N absorbed from both indigenous and supplied N of ICSV 93046 was approximately 72, 86 and 113 kg N ha^{-1}, respectively (Table 5).

Total grain N contents also increased with N fertiliser rates, and were found to be highly correlated with total N across all N treatments and cultivars (Table 5). This finding was confirmed by

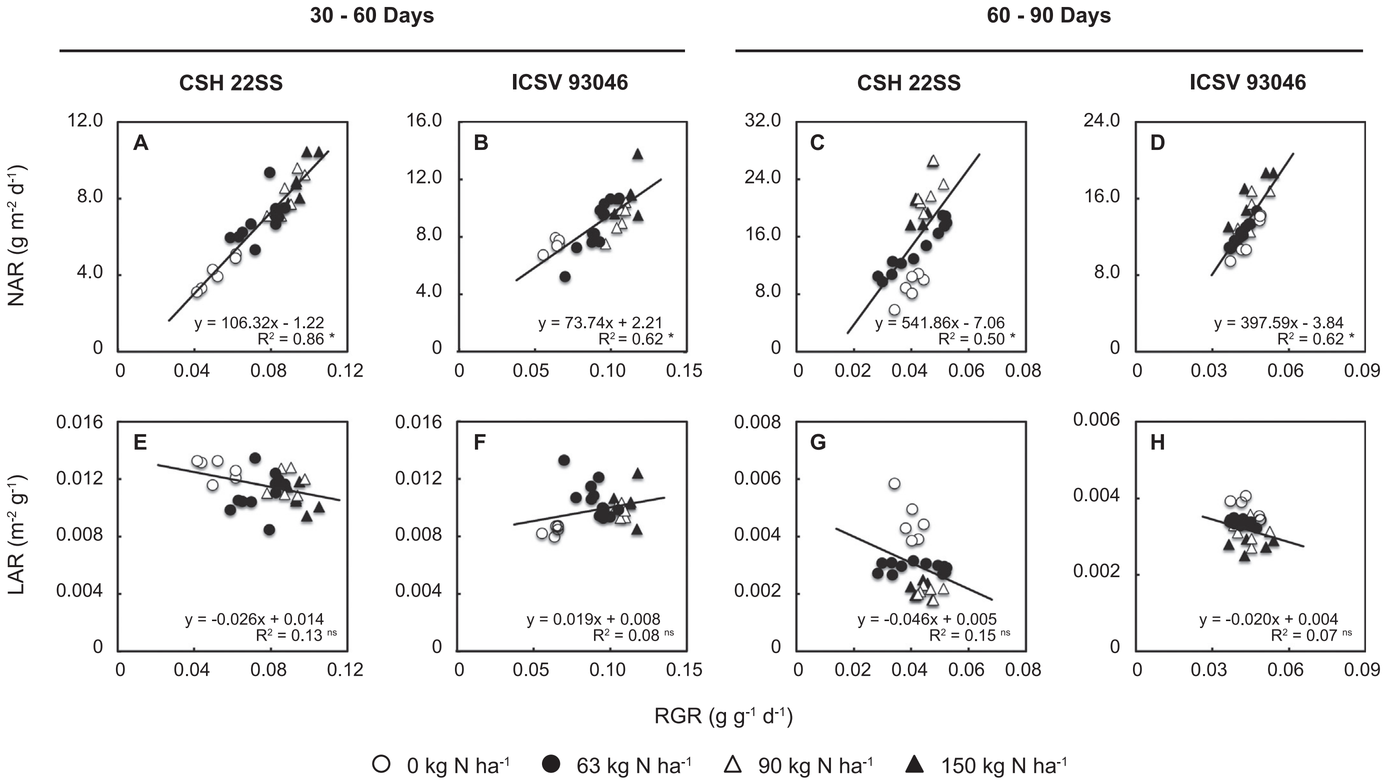

Fig. 4. Relationships between relative growth rate (RGR) and net assimilation rate (NAR; A–D) or leaf area ratio (LAR; E and F) were examined at two different developmental stages, 30–60 days after sowing (DAS) (left half) and 60–90 DAS (right half). For each developmental stage, each relationship was analysed on two different sweet sorghum cultivars, CSH 22SS (left) and ICSV 93046 (right). Plants were grown under 0 kg N ha^{-1} (open circle), 63 kg N ha^{-1} (filled circle), 90 kg N ha^{-1} (open triangle) and 150 kg N ha^{-1} (filled triangle). The slopes of regression lines are indicated together with the coefficient of determination (R^2) for each relationship. Asterisks indicate statistically significant relationships at $P < 0.05$, and the letter "ns" indicates not statistically significant.

Table 4
Sugar-related properties of sweet sorghum cultivars grown under multiple N regimes during the post-rainy seasons in the semi-arid tropics.

Cultivar	N Treatment (kg N ha^{-1})	Brix value (%)	Stalk yield (t ha^{-1})	Juice yield (m^3 ha^{-1})	Sugar yield (t ha^{-1})	Potential ethanol yield (m^3 ha^{-1})
CSH 22SS	0	6.1 ± 1.2a	9.9 ± 3.0a	3.75 ± 1.47a	0.22 ± 0.11a	0.11 ± 0.01a
	63	7.7 ± 1.5a	15.0 ± 3.2a,b	5.32 ± 0.79a,b	0.36 ± 0.07a,b	0.20 ± 0.01b
	90	6.8 ± 1.4a	18.3 ± 2.2b,c	6.59 ± 1.88b,c	0.40 ± 0.05b	0.22 ± 0.01b
	150	6.0 ± 0.5a	22.1 ± 0.8c	8.49 ± 0.24c	0.46 ± 0.02b	0.25 ± 0.00c
ICSV 93046	0	6.9 ± 1.5a	9.9 ± 2.2a	1.94 ± 0.64a	0.13 ± 0.06a	0.07 ± 0.00a
	63	8.8 ± 0.8a	14.7 ± 2.3a,b	4.54 ± 1.66a	0.36 ± 0.15a,b	0.20 ± 0.01b
	90	7.7 ± 0.6a	19.0 ± 3.6b,c	7.72 ± 2.29b	0.53 ± 0.16b,c	0.29 ± 0.01c
	150	7.7 ± 0.8a	22.1 ± 2.8c	9.70 ± 1.09b	0.67 ± 0.14c	0.36 ± 0.00d

Values are the means ± SD of 24 replicates. Letters indicate statistically significant differences ($P < 0.05$), when compared with 0 kg N ha^{-1} treatment.

the calculations of N harvest index (NHI), which was comparable regardless of N treatment except for a slight decrease under 0 N treatment of CSH 22SS, indicating that the relative distribution of N was unaffected by N fertiliser rates (Table 5).

3.5. NUE

Partial factor productivity (PFP), as it is a ratio, tends to decline with increases in N application rates, and our results followed the

Table 5
Nitrogen content and nitrogen harvest index of sweet sorghum cultivars grown under multiple N regimes in post-rainy seasons.

Cultivar	N Treatment (kg N ha^{-1})	Total plant N (kg ha^{-1})	Grain N (kg ha^{-1})	Nitrogen harvest index (%)
CSH 22SS	0	34.7 ± 6.5a	27.0 ± 5.5a	77.7 ± 3.2a
	63	71.4 ± 6.5b	58.4 ± 13.2b	81.4 ± 3.4b
	90	93.9 ± 24.0c	77.6 ± 19.1c	82.9 ± 2.6
	150	128.4 ± 27.0d	105.8 ± 19.2d	82.9 ± 4.1
ICSV 93046	0	42.3 ± 10.2a	33.7 ± 8.6a	79.7 ± 3.1a
	63	72.3 ± 5.7b	56.9 ± 5.3b	78.7 ± 1.3a
	90	88.0 ± 9.2c	70.0 ± 4.6c	79.5 ± 2.5a
	150	113.2 ± 22.6c	88.8 ± 10.3d	78.4 ± 3.2a

Values are the means ± SD of 27 replicates. Letters indicate statistically significant differences ($P < 0.05$), when compared with 0 kg N ha^{-1} treatment.

Table 6
Multiple nitrogen use efficiency indices of sweet sorghum cultivars grown under multiple N regimes during the post-rainy seasons in the semi-arid tropics.

Cultivar	N Treatment ($kg\,N\,ha^{-1}$)	PFP[a] ($kg\,kg\,N^{-1}$)	ANUE[b] ($kg\,kg\,N^{-1}$)	ANRE[c]	PNUE[d] ($kg\,kg\,N^{-1}$)
CSH 22SS	0				
	63	80.3 ± 8.5b	41.7 ± 8.5a,b	0.58 ± 0.12a	86.3 ± 12.7b
	90	72.7 ± 4.2b	45.7 ± 3.1b	0.66 ± 0.06a	76.3 ± 7.4b
	150	49.3 ± 3.7a	33.1 ± 2.6a	0.62 ± 0.07a	54.7 ± 3.9a
ICSV 93046	0				
	63	56.8 ± 3.9b	25.3 ± 2.2b	0.48 ± 0.05a	53.3 ± 5.2b
	90	48.9 ± 5.1b	25.6 ± 1.9b	0.51 ± 0.04a	50.5 ± 4.0b
	150	32.7 ± 2.7a	18.6 ± 0.7a	0.47 ± 0.08a	39.4 ± 3.8a

Values are the means ± SD of at least 27 replicates. Letters indicate statistically significant differences ($P < 0.05$), when compared with other treatments within the same index.

[a] PFP: partial factor productivity.
[b] ANUE: agronomic nitrogen use efficiency.
[c] ANRE: apparent nitrogen recovery efficiency.
[d] PNUE: physiological nitrogen use efficiency.

same trend regardless of cultivar (Table 6). However, the decline was gentle between 63 N and 90 N compared to 90 N and 150 N, indicating an acute drop of NUE beyond 90 N (Table 6).

Agronomic NUE (ANUE), one of the components of PFP and an indicator of an efficiency of yield derived from fertiliser N, was the highest under 90 N treatment, and was significantly higher than that under 150 N treatment in both CSH 22SS and ICSV 93046 (Table 6), indicating the highest N-fertiliser use efficiency under 90 N treatment. Regression analysis further illustrated that ANUEs under 90 N treatment for both cultivars were the highest of tested treatments and close to the peak values (Fig. 5A and B).

Further analysis of ANUE components revealed that apparent N recovery efficiency (ANRE; Eq. (7)) was comparable between all treatments, but physiological N use efficiency (PNUE; Eq. (8)) was significantly lowered under 150 N compared to 63 N and 90 N treatments for both CSH 22SS and ICSV 93046 (Table 6).

4. Discussion

4.1. Post-rainy season compared to rainy season

Post-rainy season in India is not ideal for sorghum cultivation compared to rainy season due to shorter day-length, lower temperature and little/no precipitation. Because of these different agro-climatic conditions, seasonal adaptations as well as responses of sorghums are distinctively different (Reddy et al., 2012), which raises an absolute necessity of investigating N response analysis in each season independently. The present study also displayed substantial reduction in plant height and biomass compared to rainy seasons (Uchino et al., 2013). Because this study was conducted with irrigation, water was not the limiting factor, hence not contributing this lower growth. Therefore, this decrease in growth could be mainly attributed to lower temperature and shorter day length. Under such low productive environment, efficient use of precious investment such as fertilizers is critical especially for small farmers to maximise income. Thus this study was conducted in a scope of sustainable and efficient N management.

4.2. Determination of efficient N fertiliser rates

With rise in fertiliser prices and unstable sorghum cultivation due to unfavourable climatic reasons during the post-rainy season in India, increasing fertiliser efficiency to maximise returns from fertiliser investment is rather more important than achieving the maximum production. The regression analysis of ANUE displayed the peaks of curves at 96 $kg\,N\,ha^{-1}$ and 77 $kg\,N\,ha^{-1}$ for CSH 22SS and ICSV 93046, respectively (Fig. 5A and B). Furthermore, the calculated EONR was found to be 133.5 $kg\,N\,ha^{-1}$ and 144.5 $kg\,N\,ha^{-1}$ for CSH 22SS and ICSV 93046 respectively (Fig. 5A and B). In our study, the plateau of the N response curve, hence the maximum yield, could not be shown explicitly (Fig. 2C and D), but these maximum ANUE and EONR values were found within the highest fertiliser rate tested, 150 $kg\,N\,ha^{-1}$. Considering these results together with the fact that there was no statistical difference on grain yield or DM production between 90 N treatment and 150 N treatment for both cultivars tested (Fig. 2A–D), it is plausible to

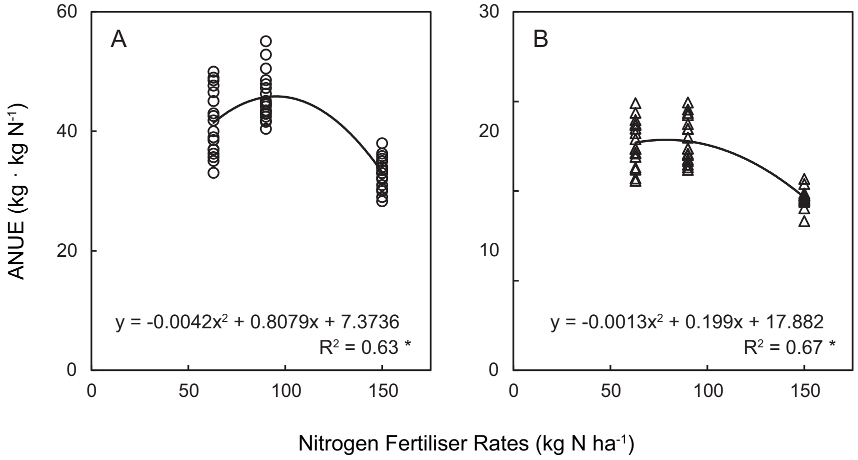

Fig. 5. Relationship between N fertilizer applied and agricultural nitrogen use efficiency (ANUE) of two different sweet sorghum cultivars, CSH 22SS (A) and ICSV 93046 (B). The slopes of regression lines are indicated together with the coefficient of determination (R^2) for each relationship. Asterisks indicate statistically significant relationships at $P < 0.01$.

consider that the most efficient rate among our experimental treatments was 90 kg N ha^{-1}.

4.3. *Effects of N treatments on plant growth and biomass productions*

Validity of 90 kg N ha^{-1} as the most efficient rate in our experiments was further observed from plant growth analysis (Fig. 3). Our study showed NAR as the major contributor to the observed increase in RGR in response to N fertilisation during the early period of development (Fig. 3I and J). There were significant correlations between NAR and RGR observed for both varieties tested (Fig. 4A–D), in agreement with a previous study under irradiance higher than 25 mol m^{-2} day^{-1} such as India (Shipley, 2006). However, the correlation between RGR and NAR appears to become weaker during the late developmental stage, owing to reduction in LAR due to shift on their assimilate distributions more to non-leaf tissues during development (Fig. 3S and T; Wadas et al., 2009). Consequently, although NAR was maintained to be increased with the N fertiliser rates (Fig. 3K and L), the RGRs became comparable regardless of N treatment (Fig. 3C and D).

However, the earlier differences observed in RGR were reflected in CGR and thereby affected biomass at harvest. Because plant biomass often increases exponentially with time in sorghum (Gerik et al., 2004), early seedling vigour has marked effects on crop growth, biomass and yield (Cisse and Ejeta, 2003; Ellis, 1992). In addition, the observed changes in CGR could also be explained by increased NAR and LAI. We found that LAI also increased with increasing N rates up to 90 kg N ha^{-1} (Fig. 3M–P), which is consistent with those of a previous study of grain sorghum in which increased N fertiliser resulted in increased LAI, but only up to 100 kg N ha^{-1} (Locke and Hons, 1988).

4.4. *N treatments and grain yield components*

Our results displayed that differences in yield were due to kernel numbers, but not HI or grain filling indicated by 1000-seed weight (Table 3). The total numbers of kernels are the major contributor to yield in grain sorghum (Saeed et al., 1986) and an important indicator of sink size, which accounts for 60–87% of total grain yield (Gerik et al., 2004). Seed numbers are determined between panicle initiation and flowering (approximately 35–90 DAS in our study) (Saeed et al., 1986; Gerik et al., 2004), and this period overlaps with that when the effects of topdressing were inferred in the present study. Thus, lower N rates may have resulted in reduced numbers of total seeds owing to insufficient N availability. In addition, Rego et al. (1998) under similar environmental conditions reported a linear increase in seed numbers of grain sorghum in response to N rates, but maximum seed numbers were attained at 90 kg N ha^{-1} with no significant differences thereafter, further supporting our findings.

4.5. *N treatments and sugar and ethanol yield*

Brix values were only approximately 7% in this study (Table 4) and independent of N treatments and biomass, similar to Uchino et al. (2013). The two cultivars tested have Brix values above 15% under rainy season cultivation in India (Dalvi et al., 2013). These results indicated seasonal variations on sugar accumulation in sweet sorghums, and in fact sugar yield and subsequent ethanol production tends to be decreased during post-rainy season in India (Dalvi et al., 2013; Vinutha et al., 2014). Although our results indicated that the yield levels of all tested treatments were too small to be a competitive energy crop, calculated ethanol yield increased with increasing N rates mainly due to increase in juice volumes (Table 5; Eq. (2)). Under irrigated conditions where sorghum moisture is stable, juice volumes and hence total amounts of non-structural carbohydrates correlate with biomass (Slewinski, 2012). Thus, in order to compete with sugarcane as an efficient energy crop, elite sweet sorghum varieties capable of producing higher biomass and Brix values in Indian post-rainy season are awaited. Additionally, further elucidation on the mechanism of sugar accumulation in sweet sorghum could facilitate this objective.

4.6. *N treatments and NUE*

ANUE is an indirect indicator of benefit-cost ratio, as it describes yield made from an investment in the form of N fertiliser (Cassman et al., 1996). In our study, the highest ANUE was found to be approximately 45.0 for CSH 22SS and 25.0 for ICSV 93046 (Table 6). This result, together with previous reports on ANUE of sorghum ranging from 5.0 and 30.0 (Ouedoraogo and Mando, 2010; Prasad, 1996), rice (Quezada et al., 2013) and maize (Vanlauwe et al., 2011), suggests the presence of large genotypic variation in ANUE. Yet, regression analysis on ANUE clearly displayed the rate of 90 kg N ha^{-1} has the highest efficiency between all treatments tested (Fig. 5A and B). Additional investigations of the components of ANUE revealed that the reduction of PNUE rather than ANRE showed a larger impact on ANUE, indicating PNUE rather than ANRE is the major factor for the observed differences on PFP and ANUE between tested treatments. Lower PNUE values under 150 N treatment indicated that absorbed N was not utilised efficiently for grain production, but merely accumulated. A strong relationship reported between grain yield and plant biomass (Mahama et al., 2014), was used as the underlying basis to assume that the absorbed N was not used efficiently for biomass production as well. Given that the concentrations of N in both biomass and grains of sorghum are known to correspond to the rate of N fertilisation (Hons et al., 1986), the observed reduction in PNUE under 150 N treatment implied that the N fertiliser was supplied in excess of that needed for efficient utilisation.

5. Conclusions

Growth analysis of two sorghum cultivars, CSH 22SS and ICSV 93046, indicated that NAR and LAI were affected by rates of N fertilisation, resulting in increased RGR and CGR with increasing N fertiliser rates. However, significant positive effects of N fertilisation on these growth parameters as well as on the resulting plant biomass and grain yield were detected only up to 90 kg N ha^{-1}. Further investigation of NUE revealed that ANUE was the highest at 90 kg N ha^{-1}. This result was explained by the increased PNUE at this rate compared to others. These findings were similar for both cultivars of sweet sorghum tested. Thus, the most efficient rate of N fertilisation among the tested treatments was concluded to be 90 kg N ha^{-1} for sweet sorghum cultivation during the post-rainy season in the semi-arid tropics.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.eja.2015.07.010.

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