

Effect of Nitrogen on Yield and Quality of Forage Sorghum Genotypes in Peninsular Malaysia

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ABSTRACT

Sorghum is a potential source of cellulosic biomass; however, selecting suitable genotypes and providing optimal nutritional supplements are necessary to achieve optimal yield. Proper management practices and fertiliser application will increase the yield and quality of the crop. This study aimed to evaluate the agronomic performance and quality of selected forage sorghum genotypes in response to nitrogen application for cultivation under local climatic conditions. Three forage sorghum genotypes (G1, G2, G3) from different source populations were evaluated for their quality and yield-related components, while grain corn (G4) was utilised for agronomic comparison with the sorghum genotypes only. The experiments were conducted at Field 10, Universiti Putra Malaysia, using a randomised complete block design with a split-plot arrangement, with nitrogen doses as treatments: T1 (120 kg/ha) and T2 (0 kg/ha). Under the influence of nitrogen, G2 (17-39-2-4-3) recorded substantially high stem fresh weights (1248.2 g) and stem diameter

(2.87 mm), while G3 (14-9-4-1-2) showed the highest total soluble contents (16.55%) and chlorophyll index (52.46). G2 had the highest levels of neutral detergent fibre (NDF) and acid detergent fibre (ADF) in the stem at T1 (120 kg/ha N), while the leaves of G3 had higher NDF values under the influence of nitrogen. The study identifies G2 and G3 as potential forage sorghum genotypes that generate good yield and quality under nitrogen application. Nevertheless, the study was conducted under limited nitrogen

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regimes; therefore, an extensive field evaluation is recommended for determining optimal yields under varied nitrogen levels.

Keywords: Chlorophyll, NDF, physiology, sorghum, TSS

INTRODUCTION

Sorghum (*Sorghum bicolor* (L.) Moench) is the fifth most important cereal crop worldwide. It originated from North Africa and is cultivated across continents in countries including China, India, and Mexico (Venkateswaran et al., 2019). It can be cultivated in challenging environments with significantly high productivity. Sorghum has been widely introduced as a forage crop in numerous tropical and subtropical regions across the globe (Dicko et al., 2006; Dexuan et al., 2022). It has become a staple food source in the semi-arid tropics of Asia and Africa and is also a preferred fibre food for millions of people worldwide (Hariprasanna, 2015; Mohammed et al., 2019). In Southeast Asia, Thailand is a major sorghum producer, with an annual production of more than 50,000 tons, while China and India are the largest sorghum producers on the Asian continent (Taylor, 2019).

Sorghum can be used for feed and fodder, and it is chosen as a potential animal feed because of its high cellulosic and hemicellulose content (Bartzialis et al., 2020; Hao et al., 2014). Even though water and good soil conditions have been reported among the main factors of sorghum yield limitation, as this crop is typically grown under rain-fed conditions (Bakari et al., 2023). However, for many years, sorghum has also been grown extensively in marginal soils where nitrogen deficiency affects the productivity of the crop, as it is the major component of plant nutrition. The tuning of the nitrogen component of the soil will apparently increase the biomass of sorghum (Franke et al., 2018; Wortmann et al., 2007). Effective fertiliser management practices can enhance growth rate, biomass, plant height, and leaf area index. Suitable genotype selection along with adequate fertilisation application will improve the biomass yield of sorghum. This will help reduce input costs and prevent the loss of available nutrients (Baligar et al., 2001; Silva et al., 2024).

Nutrient efficiency is the most important factor for breeding new cultivars. Nitrogen has been effectively found to affect the growth and productivity of sorghum and other grasses (Rihacek et al., 2020). Sorghum has a very minimal fertiliser requirement, and efficient nitrogen (N) utilisation in grass production is highly recommended to reduce production costs (Afzal et al., 2012; Tasie & Gebreyes, 2020). The cell wall is composed of key traits such as protein content, neutral detergent fibre (NDF), acid detergent fibre (ADF), and acid detergent lignin (ADL), which are all quality indicators for forage applications (Yucel & Erkan, 2020). These components are directly obtained from the nutrients supplemented to the crop, which enhances the quality of the sorghum-based animal feed (Oktem et al., 2021). Protein is one of the most important components of sorghum-based feed, which is rich in amino acids and N. The nitrogen content of the feed samples is used to calculate

the total protein content of the forage (Tasie & Gebreyes, 2020). The type and quality of forage are distinguished from one another based on cell wall materials. The nutritional value of forage is also determined by cell wall composition (Rivai et al., 2021).

The amount of digestible forage for the animals is calculated using NDF and ADF values. The insoluble portion of forage tissue is mainly composed of NDF, which further contains hemicellulose, cellulose, and lignin components of the plant cell wall. ADF is part of the feed that is slightly indigestible under acidic conditions, as it contains cellulose, lignin, and other indigestible components. NDF and ADF are good indicators of feed quality and plant maturity within a specific diet. Better estimation of fodder consumption and diets can be designed using NDF and ADF values (Harper & McNeill, 2015; Singh et al., 2018). Forage plants require various nutrients to grow, and different forage plants have diverse nutrient requirements. Irregular and insufficient nitrogen supplementation usually restricts plant productivity and affects the feed crop quality (Alley, 2001; Sayre et al., 2023). Nitrogen is a scarce nutrient in most tropical soils; therefore, sufficient nitrogen administration showed a great impact on the crops when applied in local cultivation practices (Amin, 2011; Arshad et al., 2020). Monitoring the nitrogen supplementation efficiency of crops can reduce production costs while maintaining environmental quality. Heavy use of nitrogenous fertilisers can result in negative environmental impacts, even though the crop shows an increase in yield (Omara et al., 2019).

Malaysia has an equatorial climate, with an average annual rainfall of 250 centimetres. Heavy rainfall can lead to a significant loss of nutrients in the soil, rendering key nutrients unavailable. Additionally, the nitrogen requirements can vary depending on specific crops (Mustaffer et al., 2024). Sorghum holds potential as a crop for forage production, particularly if appropriate sustainable agricultural practices are employed. It is essential to identify the most effective agronomic practices and tailor them to achieve productive forage cultivation. This study was conducted to assess the yield potential and quality of various forage sorghum genotypes under differing nitrogen regimes in Malaysia. Since sorghum cultivation is still in its nascent stages in the country, it is crucial to identify the necessary agronomic practices to optimise sustainable production in local conditions.

MATERIALS AND METHODS

Experimental Location and Planting Material

This study was conducted at Ladang 10, Faculty of Agriculture, Universiti Putra Malaysia (UPM). The land was ploughed and harrowed twice to a depth of 30 cm using a tractor-mounted moldboard plough. Sorghum seeds of three genotypes, including G1 (21-107-1-3-3), G2 (17-39-2-4-3), and G3 (14-9-4-1-2), were obtained from the Agrobank Faculty of Agriculture, UPM, Malaysia. A grain corn genotype was acquired from Famox Company (Kedah, Malaysia) and was designated as G4 in the study for comparative agronomic analysis only. The field experiment started from August to December 2020.

Seeds were sown in germination trays for 7-10 days before being transplanted to the experimental plot.

Experimental Design

The experimental design applied was a split-plot in a randomised complete block design, with three replications. The first factor was the nitrogen rate, which served as the main plot. It had two levels: 120 kg/ha of nitrogen fertiliser (T1) and 0 kg/ha of nitrogen application (T2). The second factor was genotypes, including three sorghum genotypes and one grain corn, assigned as the subplots. The distance between blocks was 2.5 m, the inter-row spacing was 0.7 m, and the interplant spacing was 0.3 m between plants (Chakravarthi et al., 2017; Naharudin et al., 2021).

Data Collection

Agronomic Parameters

The agronomic parameters, such as days to flowering, were observed when the plant reached the desired stage. Plant height was measured from the ground to the collar of the last leaf, while the number of leaves was counted manually. Relative chlorophyll content was measured from the second-to-last opened leaf using a SPAD meter (Konica Minolta SPAD-502Plus). Stem diameter was recorded 15 cm from the ground using a digital Vernier calliper (Mitutoyo), biomass yield, including the fresh weight of leaves and stem, using an electronic balance, and total soluble content was measured using a digital refractometer post-harvest (Chakravarthi et al., 2017; Farahat et al., 2020; Naharudin et al., 2021).

Chemical Analysis of Feed Quality Traits

Feed quality analysis was conducted for sorghum genotypes only. Composites of stem and leaf samples of each genotype were dried in an oven at 70 °C for 48 hours and ground using a bench-scale Retsch SM2000 (1 mm screen) knife mill. Samples were then analysed for neutral detergent fibre and acid detergent fibre with the Fibertec System, taking their fresh and dried weights into account (Chakravarthi et al., 2017; Van Soest, 1963). Crude protein was determined by the Kjeldahl method in which the resulting nitrogen content of the sample are multiplied values are then multiplied by 6.25 to get the crude protein content (Association of Official Analytical Chemists [AOAC], 1990).

Statistical Analysis

A split-plot ANOVA was used to analyse the effects of nitrogen rate, genotypes, and their interactions on all measured variables. Mean comparison using the LSD method was done to determine the best nitrogen rate and genotype ($p < 0.05$).

RESULTS AND DISCUSSION

Growth and Yield Performance of Sorghum Genotypes

Nitrogen application had a significant effect on the phenology and growth of genotypes ($p < 0.05$). Different genotypes exhibited varying days to reach flowering; however, G1 under T1 had the earliest flowering at 63 days among sorghum genotypes under nitrogen treatment. (Figure 1A). There was a significant difference in the average stem diameter between the two treatments, specifically 2.47 cm and 2.12 cm, respectively. Under the influence of 120 kg/ha nitrogen, the highest stem diameter of 2.87 cm was shown by G2, followed by G3 (2.487 cm) (Figure 1B). A significant difference in plant height was also observed in the experimental groups treated with nitrogen and those without nitrogen treatment. The average plant height under the influence of nitrogen was 278.59 cm, whereas under no nitrogen treatment, it was 228.66 cm. Among the sorghum lines, G1 showed the highest plant height (320.7 cm), followed by G3 (319.7 cm) and G2 (302.5 cm). Grain corn showed the lowest response to nitrogen and was only 172 cm high (Figure 1C). Nitrogen did not have any significant effect on the number of leaves, while genotypic effects were significant ($p < 0.05$). G2 has the highest number of leaves (13.66 leaves), while the lowest number belongs to the grain corn (11.33 leaves) (Figure 1D).

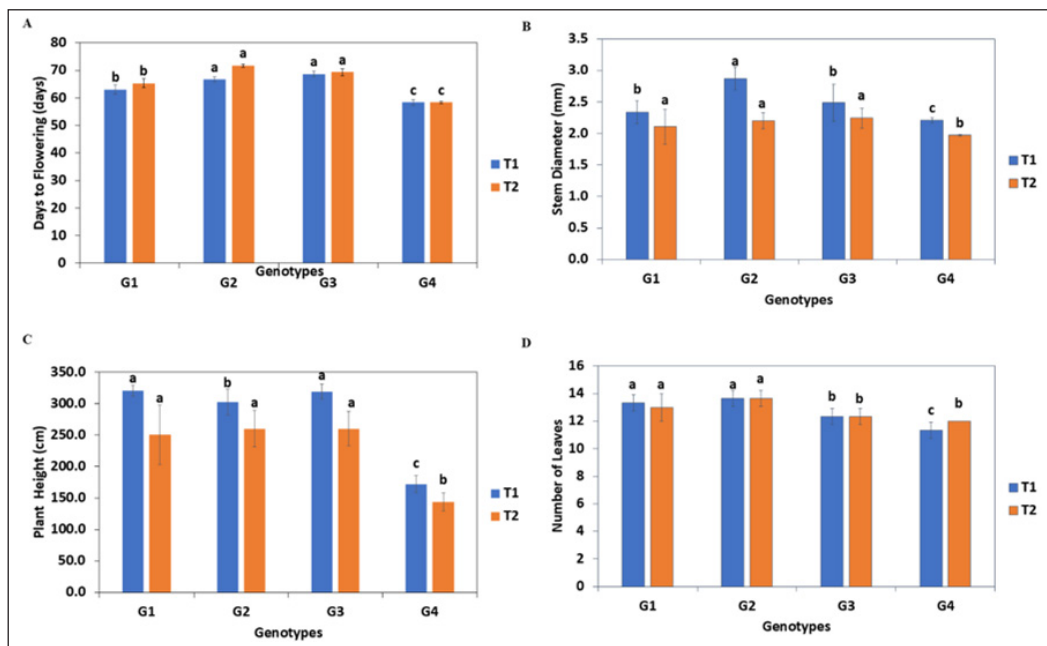


Figure 1. Effect of nitrogen treatments on different characteristics of genotypes: (a) days to flowering (days), (b) stem diameter (mm), (c) plant height (cm), and (d) number of leaves. Small letters show the significant differences ($p < 0.05$) among genotypes within the same treatment groups according to the LSD test

Note. Error bars represent standard deviations among the replicates

Nitrogen application had a significant effect on the fresh weight of stems and leaves ($p < 0.05$). G2 recorded the highest fresh weight of stems, 1248.2 g, followed by G3 with a fresh weight of 995.5 g under nitrogen supplementation, while G1 recorded 963.4 g within the same treatment (Figure 2A). The highest average fresh weight of the leaves under nitrogen treatment was recorded by G1 (140 g), followed by G2 (135.8 g). The average fresh weight of leaves without nitrogen treatment was 67.7 g, where G3 showed the highest fresh weight, 80.8 g, followed by G2 with a fresh weight of leaves of 74.6.5 g (Figure 2B). The stem is a vital component of plant architecture that gives structural support, allows transportation of water and nutrients throughout the plant (Javed et al., 2022). Nitrogen is an integral part of amino acids, proteins, and enzymes that are required for cell division, elongation, and differentiation (Ali et al., 2021). When nitrogen is readily available, it facilitates the protein-building process that supports cell growth and development (Anas et al., 2020). This contributes to stem elongation and diameter, and also enables the plant to gain a larger height and accumulate greater biomass. The biomass of sorghum increased upon nitrogen application (Holman et al., 2019).

Total Soluble Solid Content

Nitrogen application has a significant effect on the total soluble content of genotypes ($p < 0.05$). G3 had the maximal total soluble solid content (16.5%), while grain corn had the lowest total soluble solid content (4.3%) at T1 (Figure 2C). The difference in total soluble solids between G3 and grain maize points to a considerable variation in the composition or accumulation of sugars. The total soluble solids are the weight percent of water-soluble solids in the stem, and it is widely used as an approximation of sugar content due to their significant linear correlation with total sugar content (Sen et al., 2019). The value of sugar content in sorghum can be enhanced to use it as a feedstock (Jebril et al., 2021). Nitrogen is involved in enhancing photosynthetic activity, thereby producing more assimilates, which can enhance the synthesis of carbohydrates. Therefore, the higher sugar content in the stem under the influence of nitrogen is attributed to enhanced carbohydrate production. The increased value of total soluble solids in sorghum is a key trait for quality assessment and also indicates a sweeter genotype with better flavour and higher nutritional value (Zaituniguli et al., 2021).

Chlorophyll Index

Nitrogen significantly affected the chlorophyll index of the genotypes ($p < 0.05$). G3 exhibited the highest chlorophyll index in T1 (52.46) and T2 (42.22), while G1 was the lowest chlorophyll-producing genotype in T1 (44.09) and T2 (29.56) (Figure 2D). Nitrogen is an important element for plant metabolism. It is an essential part of the chlorophyll structure in leaves necessary for photosynthesis. Nitrogen increases the chlorophyll index of the plant under optimum conditions (Buah & Mwinkaara, 2009; Dembele et al., 2021).

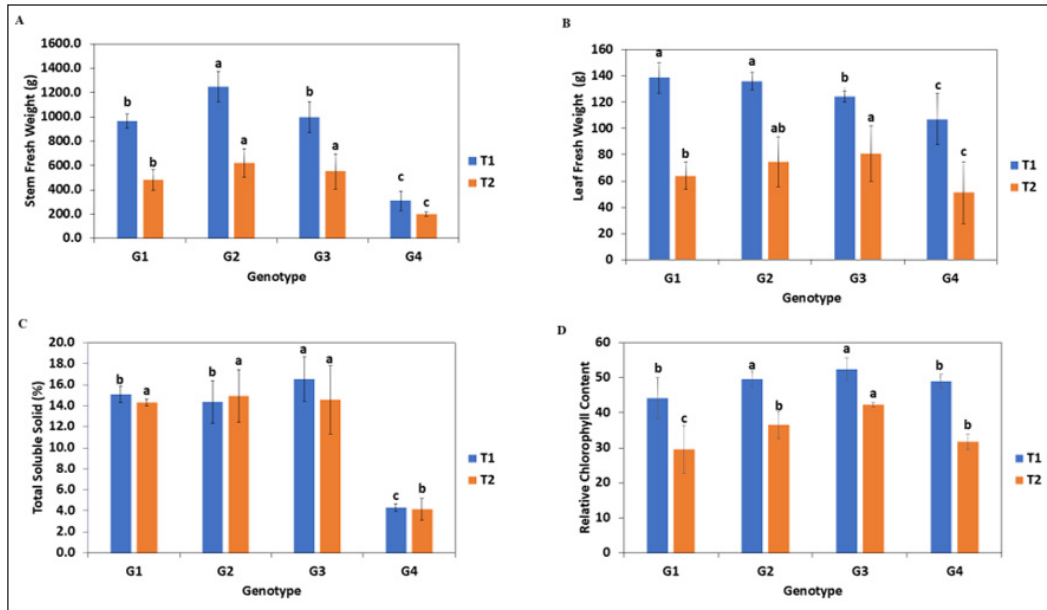


Figure 2. Effect of nitrogen supplementation on different characteristics of genotypes: (a) stem fresh weight (g), (b) leaf fresh weight (g), (c) total soluble contents (%), (d) chlorophyll content. Small letters show the significant differences ($p < 0.05$) among genotypes within the same treatment groups according to the LSD test. Note: Error bars represent standard deviations among the replicates.

Although grain corn (G4) was included as a comparative genotype for agronomic evaluation, its performance was generally intermediate relative to the sorghum genotypes across most measured traits. Therefore, the discussion focused more on the contrasting responses of the sorghum genotypes, particularly those showing superior agronomic and forage quality characteristics under the study conditions.

Feed Quality Parameters

Crude Protein Content

The effect of nitrogen on crude protein content in sorghum stem and leaf was highly significant ($p < 0.05$). Among the observed genotypes, G2 produced the highest crude protein level of 4.24% under T1 and 2.14% at T2 in the stem. However, G1 produced the lowest crude protein of 2.08% under 120 kg/ha nitrogen supplementation, and 1.75% at no nitrogen application (Figure 3A). The crude protein in leaf ranged from 11.1 to 7.64% across treatments, with G2 producing the highest in T1 (11.1%) and G1 recording the maximum values in T2 (7.74%) (Figure 3B). The crude protein contents of leaf and stem in G2 were the highest in nitrogen application treatments. This also pertains to the highest nitrogen efficiency of G2. The crude protein yield decreased significantly with a

reduction in nitrogen doses across all sorghum genotypes. Similarly, Mut et al. (2017) also reported a quadratic response in crude protein content to increasing nitrogen rates across all tested samples. Rihacek et al. (2020) reported similar crude protein ranges of 11.1 to 16.5% while studying the nitrogen efficiency of sorghum genotypes. Likewise, Xu et al. (2021) and Damian et al. (2017) observed that crude protein yield rose significantly with increasing nitrogen doses across all maize and sorghum crops. The crude protein content in leaves observed in the current study was higher than the minimum recommended by Van Soest (2018) for optimal rumen fermentation. Therefore, the evaluated genotypes can be recommended for further selection of high-quality forage. These results were in line with the findings of Toum et al. (2018), who also reported that high nitrogen efficiency increased plant crude protein content. This can possibly be explained by the fact that nitrogen is frequently involved in protein synthesis (Chen et al., 1994; Singh et al., 2018).

Neutral Detergent Fibre Content

The nitrogen application had a significant effect on the stem and leaf neutral detergent fibre content of sorghum genotypes ($p < 0.05$). The fertilised forage had a higher mean NDF concentration in the stem (61.3%) compared with the non-fertilised ones (59.08%). The study also observed that stem NDF content varied among the genotypes as well. G2 produced the highest NDF content with 62.77% in T1, while G3 recorded the lowest (58.03%) (Figure 3C). This result is related to the high stem girth of G2, which raises the amount of cell-wall material in forage. This result aligns with the findings of Carpici et al. (2010), who reported a correlation between forage NDF content and stem diameter. Collectively, these results suggest that increasing plant stem diameter could play a role in elevating NDF content (Sher et al., 2016). Although an increased amount of fibre in forage reduces digestibility. Highly digestible fibre enables greater dry matter intake in animals whose intake is limited by physical fill (Hoffman et al., 2001). Eltelib and Eltom (2006) reported similar results that increased nitrogen concentration enhances the fibre quantity of sorghum. In contrast, Delevatti et al. (2019) reported that an increase in nitrogen levels decreases the NDF concentration. It is worth noting that the high NDF content of the forage is still a dominant factor in determining overall forage quality.

Leaves of sorghum shared contrasting results as compared to the stem, where the mean NDF value for T2 (68.85 %) was higher than T1 (65.95 %), suggesting that nitrogen application did not increase the NDF content in leaves. G3 produced the highest NDF content at T1 (67.22%) and T2 (70.81%), whereas the lowest NDF values in leaves were recorded by G2 (63.9%) at T1 and G1 (66.8%) at T2 (Figure 3D). Nitrogen application promotes protein and chlorophyll content, resulting in less fibre content in leaf tissues (Carpici et al., 2010). Similar findings were reported by Bélanger & McQueen (1999), where the leaf NDF concentration decreased with increasing rates of N fertilisation.

Additionally, the range of NDF found in this study was higher than the National Research Council's dairy cow diet recommendation, affirming their utility as animal feed (Mohammed et al., 2019).

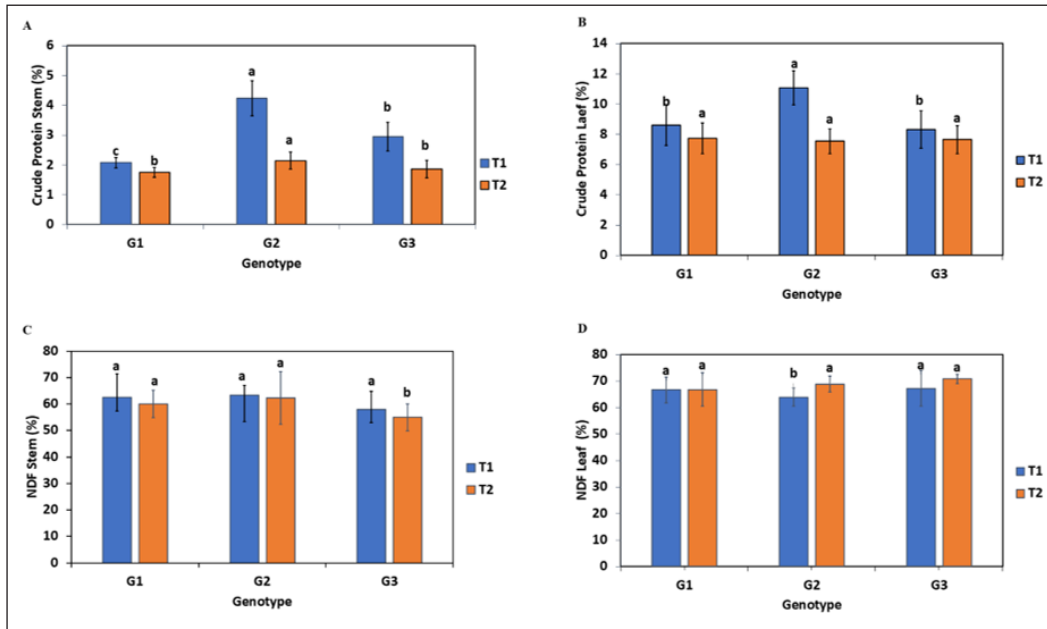


Figure 3. Effect of nitrogen feed on quality parameters of various sorghum genotypes: (a) crude protein in stem (%), (b) crude protein in leaf (%), (c) neutral detergent fibre in stem (%), (d) neutral detergent fibre in leaves (%). Small letters show the significant differences ($p<0.05$) among genotypes within the same treatment groups according to the LSD test

Note. Error bars represent standard deviations among the replicates

Acid Detergent Fibre Content

Nitrogen application significantly affected the ADF content in the stems and leaves of sorghum genotypes, while genotypic differences were observed. The highest ADF content in the stem was observed in G2 (40.81%) at T1 and T2 (39.57%), while G3 produced the lowest values across treatments (Figure 4A). This was probably corresponding to the fact that G2 showed the highest stem diameter. Thus, it contained the highest proportion of cell walls. Cell wall degradability was highly correlated with chemical composition in plants (He et al., 2020). Leaves of G1 produced the highest ADF content of 33.83%, followed by G2 with 33.03% at T1, while G3 recorded the maximum values at T2 (36.21%), and the lowest was generated by G1 (35.09%) within the same treatment (Figure 4B). Johnson et al. (2001) reported similar findings, indicating that the application of nitrogen had a minimal effect on the ADF concentration of plants. This trend is in disagreement with the findings

by Bélanger & Gastal (2000), which indicate that the increase in nitrogen has significantly affected the stem ADF concentration of the forage.

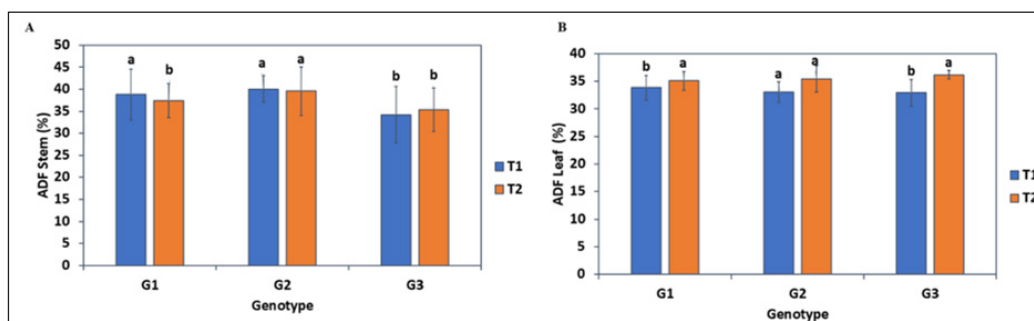


Figure 4. Effect of nitrogen on different characteristics of sorghum genotypes, (a) acid detergent fibre in stem (%), (b) acid detergent fibre in leaves (%). Small letters show the significant differences ($p < 0.05$) among genotypes within the same treatment groups according to the LSD test

Note. Error bars represent standard deviations among the replicates

CONCLUSION

Among all sorghum genotypes studied, G2 showed higher biomass yield, stem diameter, and the highest crude protein values in the stem under the influence of nitrogen. Whereas G3 recorded the maximum total soluble solids and relative chlorophyll content, and also generated the maximum NDF content in leaves in response to nitrogen application. The study concludes by identifying G2 and G3 as potential forage sorghum genotypes that yield well and maintain quality in response to nitrogen application and are therefore suitable for development as nitrogen-efficient sorghum genotypes. The experiment was preliminary and involved a dose-specific trial with limited observations; therefore, extensive field evaluations are required to assess performance under varied nitrogen regimes.

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CONFLICTS OF INTEREST

The authors do not have any conflict of interest related to the design, data collection, analyses, interpretation or writing of the manuscript.

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