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Full Length Article

Comparison of Gas Exchange in *Moringa oleifera* and other Drought Tolerant Tree Species for Climate Change Mitigation under Semi-arid Condition of Northern South Africa

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Abstract

Climate change has modified rainfall and temperature patterns especially in semi-arid areas. One practical way to cope with this challenge is to plant trees that could have a great influence on environmental perturbations. Plants act as carbon sink minimize carbon dioxide (CO_2) from the atmosphere during photosynthesis and stores excess carbon as biomass in their parts. This study was conducted under semi-arid condition of northern South Africa to evaluate the physiological parameters of three drought tolerant tree species for their comparative tendency to mitigate climate change. An existing moringa (*Moringa oleifera*) trial, planted at a population of 5000 plants ha^{-1} was used in this study. The other two naturally growing tree species were mopane (*Colophospermum mopane*) and marula (*Sclerocarya birrea*) growing within the moringa trial vicinity were included in the study as a control. Eight trees from each species were tagged for data collection. The photosynthetic rate (A), stomatal conductance (gs), transpiration rate (E) and sub-stomatal CO_2 (Ci) measurements were measured using a fully-expanded leaf on the abaxial side of each selected leaf using a non-destructive method. Furthermore, three fully expanded leaves were sampled from one tree of each species to determine leaf stomatal density in each replication. The tree species differed significantly in gas exchange, although moringa showed highest activity of all the parameters measured. Stomatal density was also different among the species, being 281.8, 355.2 and 930.6 per unit area for marula, mopane and moringa species, respectively. Moringa maintained good leaf yield even under drought condition, indicated its potential to act as a good sink for CO_2 assimilation. The results strongly showed the superiority of moringa in capturing more carbon among the three species. Moringa can therefore be recommended for climate change mitigation in semi-arid areas of Limpopo province and possibly other areas.

Keywords: Climate change; Gas exchange; Marula; Mopane; Moringa; Stomatal density

Introduction

Climate change is negatively affecting smallholder farmers in the developing countries (Daba, 2016). The effect of the change on food security is difficult to predict at present and research is needed to deepen the understanding in this issue. Gedefaw (2015) indicated that a single step to compensate for the several unpreventable carbon dioxide emissions is to plant trees, with their ability for greater CO_2 uptake and the release oxygen in return. Plants act as a carbon sink to remove CO_2 from the atmosphere (IPCC, 2000) during photosynthesis and storing excess carbon as biomass in their parts (Johnson and Coburn, 2010).

Trees source a huge carbon quantity and regulate the carbon cycle by exchange of CO_2 from the atmosphere (Johnson and Coburn, 2010; Suryawanshi *et al.*, 2014). It is therefore, important to identify different plants species which can remove concentrated CO_2 from the atmosphere in photosynthesis and store carbon in their leaves, branches, stem, bark and roots as part of the climate change mitigation process. Trees used or targeted for carbon sequestration do not need to be of a particular kind or to be located at a particular site (Johnson and Coburn, 2010; Hof *et al.*, 2017). However, one of the main essential characteristic is adaptability to drought conditions. In semi-arid regions, the rainfall is seasonal and highly unpredictable (Clifford *et al.*, 1997).

Moringa (*Moringa oleifera*) is fairly a new crop in the Limpopo province while, mopane (*Colophospermum mopane)* and marula (*Sclerocarya birrea)* trees are known to be dominant in the far northern parts of South Africa can with stand harsh conditions better than many tree species. They also have the potential to contribute to food security for either livestock or humans (Mashabane *et al.*, 2001;

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Muok *et al.,* 2009). According to Mabapa *et al.* (2017), *M. oleifera* proved to be a good source of fodder for livestock, able to survive harsh growing conditions and can be recommended to resource-poor farmers in regions such as Limpopo province of South Africa. The nutritional benefits of *M. oleifera* are tremendous with the leaves being rich in essential nutrients required by both human and livestock (Thurber and Fahey, 2009; Anjorin *et al.,* 2010; Omotesho *et al.,* 2013).

Several studies have been conducted on physiological activity of plants some of which reveal that, salinity and soil water stress affect some physiological characteristics of plants (Chaves *et al.,* 2009; Rivas *et al.,* 2013; Wafa, 2015). However, there is no documented information on stomatal activity or gas exchange in moringa, marula and mopane tree species under semi-arid condition, even though these species are known to survive harsh conditions. The aim of this study therefore, was to evaluate the photosynthetic rate (A), stomatal conductance (gs), transpiration rate (E) and sub-stomatal CO_2 (Ci) of the three drought tolerant tree species as well as their stomatal density for climate change mitigation and adaptation planning.

Materials and Methods

Study Location

The study was conducted for 12 months during Summer (December - February), Autumn (March - May), Winter (June - August) and Spring (September - November) during 2014 – 2015, at NTL Baraka Eco-Farming Organic Farm (23°57.691'S and 30°35.205'E) at Eiland. The farm is situated approximately 50 km east of Tzaneen in the Limpopo province of South Africa. The area is a tropical region and receives about 429 mm of rain per annum, with most rainfall occurring mainly during mid-summer. The annual rainfall data were derived from the total monthly values for Eiland averaged in the past seven years. The area received the lowest average rainfall (<0.5 mm) in June and July months; the highest in December and January of more than 120 mm. The monthly temperature distribution showed that the average maximum monthly temperatures for Eiland could rise above 31°C while minimum temperatures could range between 7 to 25°C, during winter and summer seasons, respectively.

Experimental Design

An existing moringa trial which was planted during December 2013 at a population of 5000 plants ha^{-1} was used in this study. The other two naturally growing tree species of mopane and marula trees were included in the study. The naturally growing trees were used as control within the vicinity of moringa trial (Fig. 1). Marula trees are widely spread at the farm, while mopane trees are dominant and occur in pure stand with an estimated population of 1300 plants ha^{-1}. Eight trees were used in each replication and five leaves were randomly measured and averaged per tree.

Leaf Gas Exchange Measurements

Leaf gas exchange parameters were measured on a fully-expanded leaf on the abaxial side of each selected leaf, using a broad leaf chamber of a portable photosynthesis system (ADC Bio Scientific, UK). The photosynthetic rate (A), stomatal conductance (gs), transpiration rate (E) and sub-stomatal CO_2 (Ci) were simultaneously determined for each species using a non-destructive method. All the measurements were carried out under steady-state ambient conditions in full sun between 10:00 am and 14:00 pm (Clifford *et al*., 1997).

Leaf Stomatal Density

During harvesting, three fully expanded leaves were sampled from each tree species to determine leaf stomatal density, which was expressed as number of stomata per unit area of a leaf (Radoglou and Jarvis, 1990). The leaf was cleaned using cotton and slightly smeared with clear glue in the mid area between the central vein and the leaf edge at approximately 5 x 10 mm. The film was applied three times in each leaf and allowed to dry for about thirty minutes. A drop of glycerol was applied on a glass slide and the film was peeled off from the leaf surface using a fine point tweezer, mounted on a glass slide and immediately covered with a cover slip. Number of stomata for each film strip was counted under a photomicroscope system (CH-9435, Leica Microsystems, Heerbrugg, Germany) with a computer attachment.

Statistical Analysis

Data were subjected to analysis of variance using Statistix 10.0 to compare the response of tree species on measured variables. Where significant F-values from treatment effect were observed, means were separated by Least Significant Difference (LSD) at probability level of 0.05. Correlation and regression analyses were performed on the sampled data using Microsoft Excel to determine the relationship between stomatal density and gas exchange.

Results

Weather Data of the Study Area

Data on weather parameters measured during the study and anomaly in rainfall distribution are presented in Fig. 2 and 3, respectively. The rainfall declined to below 100 mm during summer season of 2014/15, as compared to the past two years where it exceeded 200 mm. The temperature was elevated during summer season with the maximum temperature exceeding 35^{o}C (Fig. 2). The drought

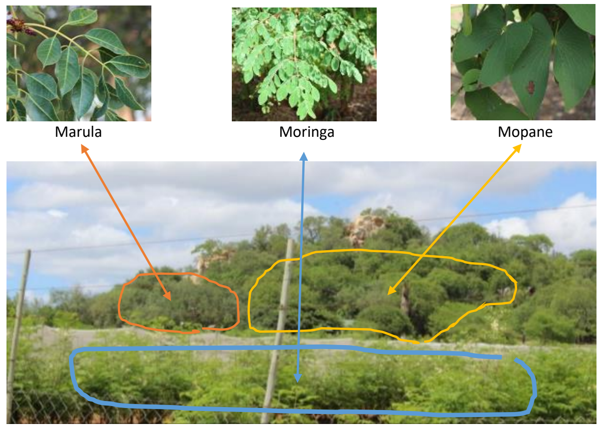

Fig. 1: Marula and Mopane trees growing within the vicinity of *M. oleifera* trial at Eiland

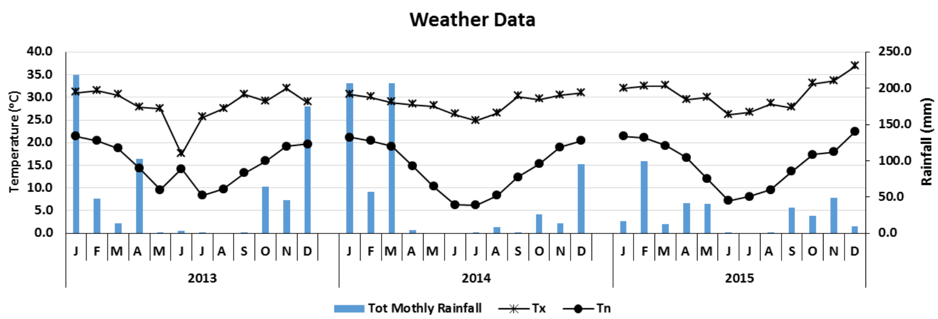

Fig. 2: Total monthly rainfall (mm), average maximum (Tx) and minimum (Tn) temperatures collected at Eiland from during the production seasons

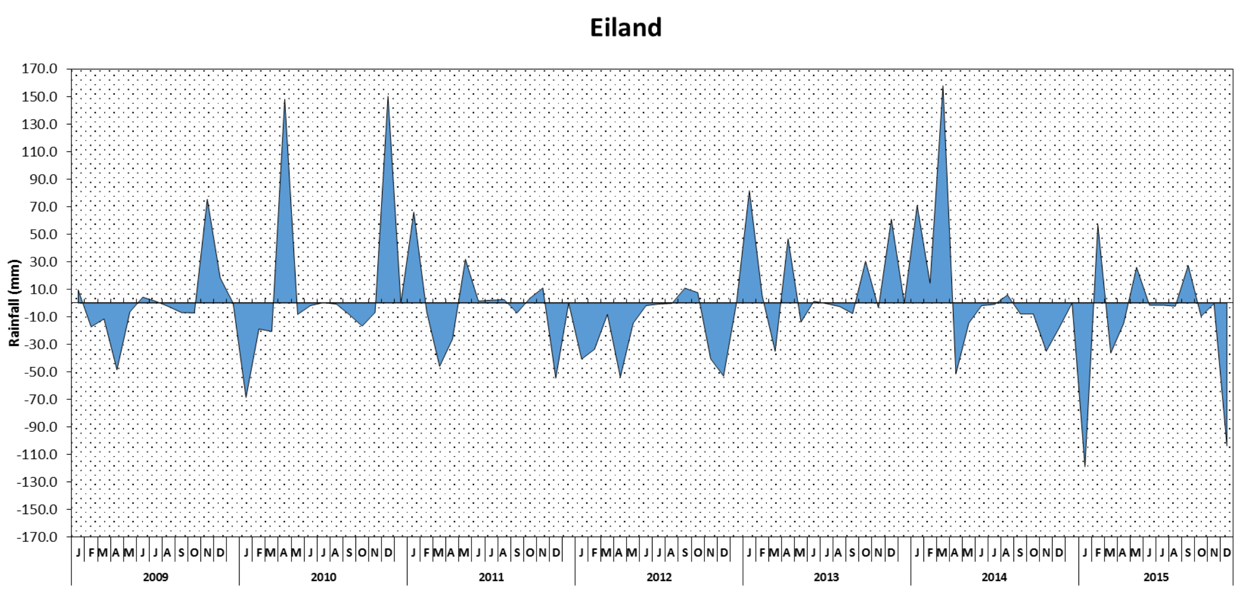

Fig. 3: Rainfall (mm) anomaly at Eiland as compared to long term average rainfall from 2009 to 2015

experienced during 2015 had a significant influence on rainfall pattern. The increased heat accompanied by moisture deficit led to below average rainfall was mainly during the year 2015 as compared with the years 2013 and 2014 where the erratic rainfall was above average in many instances.

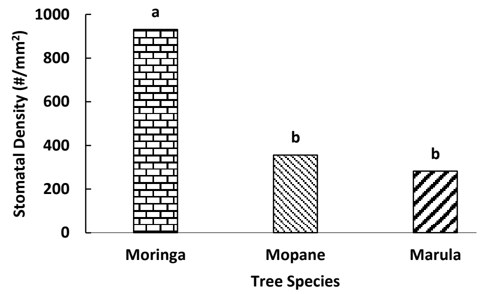

Fig. 4: Stomatal density of the measured tree species recorded from an area of 0.09 #/mm^2

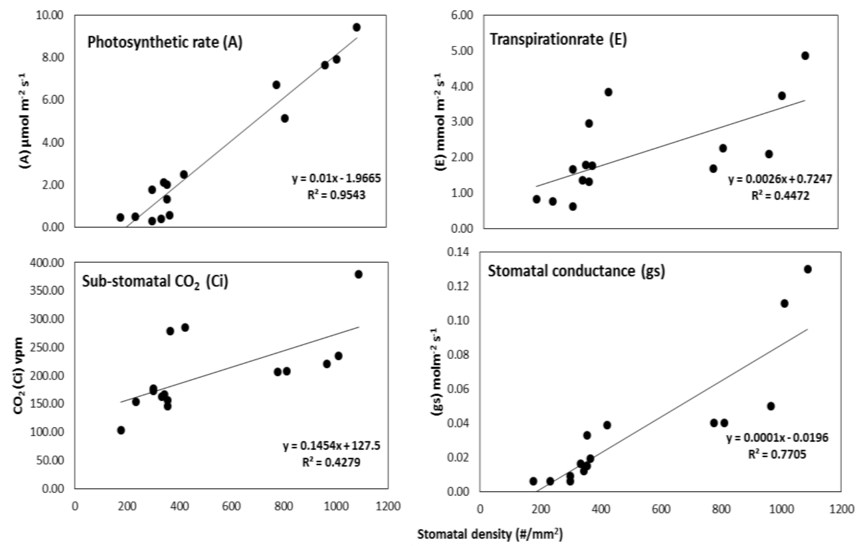

Fig. 5: Correlation and linear relationship effect of stomatal density with gas exchange irrespective of the tree species

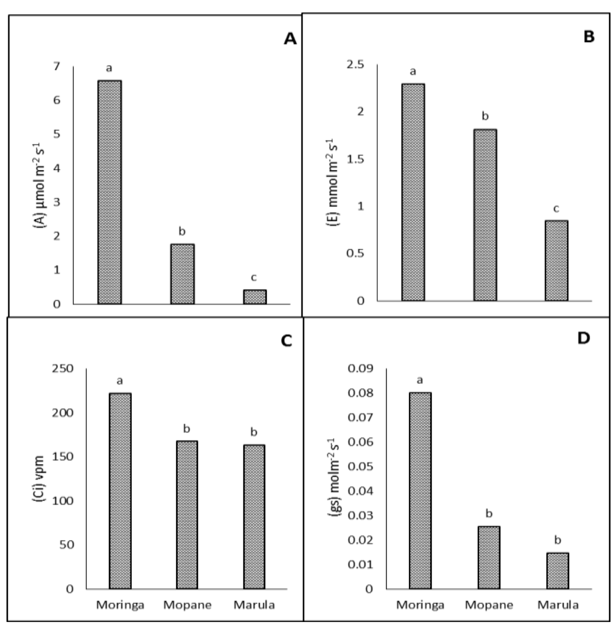

Fig. 6: Gas exchange parameters of moringa, mopane and marula tree species. (A) photosynthetic rate, (B) transpiration rate, (C) sub-stomatal CO_2 and (D) stomatal conductance. Means with different letters are statistically significant

Table 1: Pearson correlation between measured gas exchanges of *M. oleifera*

	Photosynthesis	Transpiration	Sub-stomatal CO_2
Transpiration	0.48***		
Sub-stomatal CO_2	-0.45***	-0.09^{ns}	
Stomatal Conductance	0.46***	0.70***	0.16^{ns}

Significance levels: *P<0.05, ** P<0.01, *** P<0.001, ns: not significant

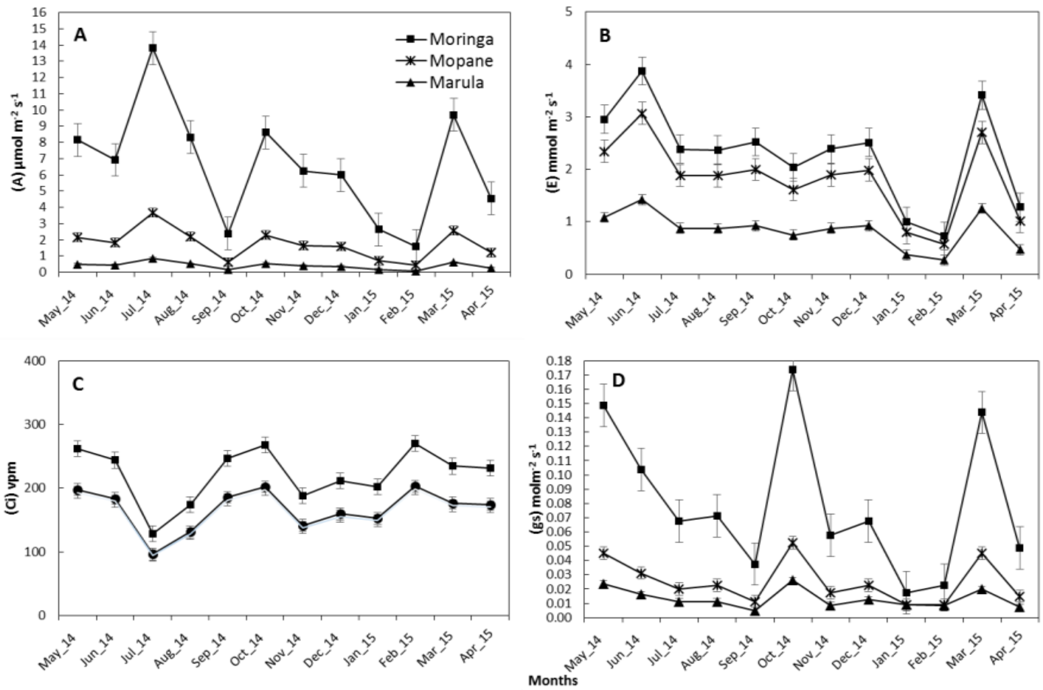

Fig. 7: Monthly gas exchange parameters of moringa, mopane and marula tree species. (A) photosynthetic rate, (B) transpiration rate, (C) sub-stomatal CO_2 and (D) stomatal conductance

Stomatal Density of Moringa, Mopane and Marula Tree Species

Measurement of stomatal density showed significant differences among the investigated species. The stomatal density per unit area ranged between 281.8 - 930.6 with the highest occurring in moringa followed by mopane and then marula (Fig. 4). The relationships between stomatal density and gas exchange are presented in Fig. 5. Photosynthetic rate and stomatal conductance were positive and linearly related to stomatal density (Fig. 5). However, the transpiration rate and sub-stomatal carbon dioxide were also positive and linearly related to stomatal density but with a scattered associations.

Gas Exchange Measurement

Comparing gas exchange among the three tree species, moringa was found to exhibit the highest in all gaseous exchange parameters measured, all of which were significantly higher than the two other species. No significant difference in sub-stomatal CO_2 and stomatal conductance were found between mopane and marula tree species, but mopane had higher photosynthetic rate and transpiration than marula tree (Fig. 6). When comparing the monthly influence on gaseous exchanges among the tree species, moringa was again found to exhibit the highest among the three species (Fig. 7). Photosynthetic rate in moringa was quite higher relative to the other species throughout the data collection period except in September 2014 and February 2015, where the differences were not significant (Fig. 7A). Transpiration rate in moringa and mopane tree species were higher as compared to marula species. However, a drastic decrease was measured in all tree species between January and February 2015 (Fig. 7B). Once again, a highest sub-stomatal CO_2 concentration was noted in moringa compared to the other two species which were similar (Fig. 7C). Stomatal conductance was again higher in moringa during May, October 2014 and March 2015 months compared to the other species during the same period (Fig. 7D).

Photosynthesis was significantly correlated with transpiration rate, sub-stomatal CO_2 and stomatal conductance (Table 1). A stronger correlation was found between transpiration rate and stomatal conductance. However, transpiration and stomatal conductance had a non-significant correlation with sub-stomatal CO_2.

Discussion

During data collection between 2014 and 2015, the study area, Eiland experienced extreme drought and high temperatures. A total rainfall of 432 mm was received during this period. Anjum *et al.* (2011) reported that environmental abiotic stresses mainly drought, salinity and extreme temperatures, could impair plant growth and development and as such limit the productivity.

It was observed from 2014 and 2015 summer season, that Eiland received below average rainfall with the situation worsening during 2015 where the rainfall anomaly was greater than 100 mm below average. This situation affected many agricultural crops and the impact of climate change within this region was evident. Frosi *et al.* (2017) reported that increased irregularities of rainfall and temperature mainly in the semi-arid conditions major climate concern for agricultural productivity. Low rainfall and prolonged dry spells affect crop production as well as physiological activities (Reddy *et al.,* 2003; Chaves *et al.,* 2009; Behrouzyar and Yarnia, 2014).

The stomatal density of moringa was significantly higher as compared to mopane and marula trees. This presented an added advantage to moringa in gaseous exchange. According to Xu and Zhou (2008), the increase in leaf stomatal density leads to significant increase in stomatal conductance and net CO_2 assimilation rate. Camargo and Marenco (2011) reported that stomata are turgor-operated valves that control water loss and CO_2 uptake during the process of photosynthesis. From our results, in addition to possessing the highest stomatal density, moringa was also superior in stomatal conductance. These two morphological and physiological traits could partly explain the relatively highest photosynthetic activity recorded in moringa.

The reduction in leaf conductance due to lower stomatal density lowered photosynthesis as it is positively related to stomatal conductance (Fig. 5). Similar results were observed in this study whereby, moringa had a significantly higher stomatal conductance and consequently, higher photosynthetic rate (Fig. 4 and 5) as compared to the other tree species. This could be due to moringa's ability to survive in all types of soils and various climatic conditions in the semi-arid tropics (Kumar *et al.,* 2017).

The superiority of moringa in terms of gaseous exchange as compared to the other tree species could be due to adaptation strategies utilised by the pant under harsh conditions. As indicated earlier, moringa uses various survival mechanisms against drought and temperature effects. Based on the observations from this study, it was revealed that moringa does not use surrounding soil moisture within the root zone immediately after rainfall rather stores the absorbed water in its succulent root tissues and utilize it at a later stage when there is moisture deficit. The sub-stomatal conductance allows diffusion of CO_2 and other gases in and out of the plant cells. The sub-stomatal CO_2 of marula and mopane were not different between the two species but that of moringa was significantly higher than the other two species, showing that moringa has the potential to sequester more CO_2 from the atmosphere.

From this study, it was evident that prolonged drought had a more severe impact on gas exchange of mopane and marula trees relative to moringa. The two species kept the stomata closed during the time of drought and high temperatures. In this perspective, Anjum *et al.* (2011), Frosi *et al.* (2017) reported that drought stress leads to progressive stomatal closure with a parallel decline in gas exchange. Farooq *et al.* (2009) further reported that, stomata close progressively as drought progresses with a consequent decline in photosynthesis. When water is severely limiting, the first strategy of plants to avoid drought is to close the stomata (Chaves *et al.,* 2009; Farooq *et al.,* 2009; Hussein *et al.,* 2014).

Clifford *et al.* (1997) reported that, under drought conditions, photosynthetic rate and stomatal conductance decreased but the sub-stomatal CO_2 tended to increase, showing that stomatal conductance was not a limiting factor for changes in sub-stomatal conductance. Furthermore, Rivas *et al.* (2013) reported that, as drought prolonged with time, moringa showed a declined gas exchange. This was evident in this study mainly on photosynthesis and stomatal conductance.

It was observed that, moringa maintained equitable leaf canopy even under drought conditions and this gives an added advantage of the plant to act as a good sink for CO_2 absorption. This gives the nutritional benefits of *Moringa oleifera* both for humans and livestock (Maroyi, 2006; Thurber and Fahey, 2009; Anjorin *et al.,* 2010; Omotesho *et al.,* 2013; Mabapa *et al.,* 2017), and the plant can be considered for climate change mitigation and adaptation plans in South Africa. Study by Jagadheesan *et al.* (2011), indicated that moringa can maintain continuous growth even under severe drought conditions but still be able to produce satisfactorily yields. The plant survives harsh conditions and its growth is not easily hampered by restricted availability of water in the soil. It has the capability of modifying the internal structure to resist drought conditions through maintenance of higher water levels (Jagadheesan *et al.,* 2011). After period of drought, the tree shows faster recovery, which enable competitive advantage under semiarid conditions (Frosi *et al.,* 2017).

Based on the prevailing weather condition during the study period, Marula and Mopane trees showed symptoms of heat and drought injury, but the tolerance of moringa to this condition was evidenced by the maintenance of satisfactorily gas exchange. This might be due to the ability of the tree to store water in the bloated trunk, as well as its tuber roots during periods of soil moisture availability for later use (Yamato *et al.,* 2009). It is reported that most terrestrial plants have symbiosis with arbuscular mycorrhizal (AM) fungi and such colonization has been reported in moringa (Yamato *et al.*, 2009). The AM fungal association contributes to soil nutrients uptake, disease resistance and improved drought tolerance of host plant (McGonigle *et al.,* 1990; Jeffries *et al.,* 2003; Yamato *et al.,* 2009). This may also contribute to the drought tolerance abilities of moringa relative to the other species studied.

Conclusion

Moringa can survive harsh conditions such as high temperatures and moisture deficit, which occur under the semi-arid climates, relative to mopane and marula trees. Moringa was superior in terms of controlling gas exchange for adaptation to harsh environmental condition compared to the other two tree species. The tree has more number of stomata which is advantageous to maintaining high photosynthetic rate, stomatal conductance, transpiration rate and sub-stomatal CO_2. Moringa can be a good crop for the semi-arid conditions of Limpopo province; where drought prevails frequently. Furthermore, the superiority of the moringa to capture carbon even under harsh growing conditions is a good attribute for climate change mitigation. The moringa is recommended to be for cultivation in Limpopo province for contribution to national climate change mitigation and adaptation plans.

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