Journal of Tropical Crop Science Vol. 12 No. 3, October 2025
www.j-tropical-crops.com

Received 16/07/2024; Revised 13/06/2025; Accepted 01/08/2025
https://doi.org/10.29244/jtcs.12.03.483-491

RESEARCH ARTICLES

Boosting Soybean Growth: The Role of Soil Moisture Conservation Technique and Vesicular Arbuscular Mycorrhizae in Limpopo Province, South Africa

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Abstract

Low soil fertility and moisture stress are the primary factors affecting soybean productivity. The study aimed to evaluate the effect of vesicular arbuscular mycorrhizae (VAM) and soil moisture conservation techniques on the performance of a promiscuous soybean variety during the 2018/2019 and 2019/2020 growing seasons. Two levels of soil moisture conservation techniques (flat and closed ridges) and two levels of VAM (0 and 100 kg.ha^{-1}) were arranged in a split-plot design within a completely randomized block design with four replications. The data collected include growth, physiological, grain yield, and yield attributes of the promiscuous soybean variety. Sole application of soil moisture conservation techniques (SMCT) and VAM showed no significant influence ($p>0.05$) on the number of leaves, plant height, and number of branches of the promiscuous soybean variety. Soil moisture conservation techniques showed significant differences ($p<0.05$) in pod length. The interaction effect between soil moisture conservation techniques and the application of VAM had a significant impact on plant vigor (0.75), while non-significant variations were observed in the number of branches (5.31), plant height, and the number of leaves (20.81). The interaction effect of soil moisture conservation techniques and VAM was significant ($p<0.05$) on shelling percentage, while a non-significant difference was observed in grain yield (1382.7 kg.ha^{-1}). Flat planting and non-VAM application had a higher shelling percentage (40.53%). The study demonstrated that flat planting, in combination with VAM, improved plant vigor under moisture-limited conditions. Flat planting can be recommended for soybean production at Syferkuil farm in Limpopo province.

Keywords: biofertilizer, closed ridges, flat, grain yield, growth parameters, soybean

Introduction

The current global population of 7 billion is expected to increase to 10 billion by 2050, with global food demand projected to rise by 50% and protein demand by 70% (Danquah et al., 2022). Soybean, being a viable pulse crop advocated for alleviating acute protein shortages worldwide, also contain oil and is a good source of unsaturated fatty acids, minerals calcium (Ca) and phosphorus (P), and vitamins A, B, C, and D (Rathwa et al., 2023). Soybeans are a versatile crop that can be used to make a wide range of products, including soy-based cuisines, soy milk, and animal feed. The crop can also improve soil fertility by fixing atmospheric nitrogen (N) with a compatible rhizobium in the soil. It can fix up to 300 kg N.ha^{-1} with the help of symbiosis (Peoples et al., 2021), which is less susceptible to leaching and volatilization in the soil (Kumar et al., 2022). Additionally, 30-80 kg N.ha^{-1} is left behind in the soil after crop harvest, which is beneficial for the subsequent crop (Shah et al., 2021). However, the absence of N could have a negative impact on the growth and yield characteristics of soybeans (Israilov et al., 2023). In semi-arid conditions, the use of Rhizobium spp. and mycorrhizal fungi is advised to produce and maintain high-quality soybeans (Igiehon et al., 2021).

Plant-microbe interactions are an essential component of the living ecosystem and are recognized as natural partners that modulate local and systemic mechanisms in plants, providing defence at the molecular, physiological, and biochemical levels

Received 16/07/2024; Revised 13/06/2025; Accepted 01/08/2025
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under various external stress conditions (Chauhan et al., 2023). Most ecosystems have mycorrhiza, which forms a symbiotic relationship with most crops, including legumes such as soybean. The arbuscular mycorrhizal fungal (AMF) symbiosis enhances plant growth by increasing water uptake, facilitating osmotic adjustment in roots, maintaining high leaf water potential, reducing oxidative damage to lipids, and supplying Phosphorus from the soil (Bahadur et al., 2019; Tang et al., 2022). Furthermore, Mycorrhiza enhances photosynthesis and water use efficiency, decreases soil erosion risk, minimizes P leaching, and increases tolerance to both biotic and abiotic constraints (Tang et al., 2022; Bahadur et al., 2019). Vesicular Arbuscular Mycorrhizae produces specific organic acids, and phosphatase enzymes, and improves the root system to increase the amount of plant-available P (Aswitha and Malarvizhi, 2022). Despite the importance of mycorrhizae, the effectiveness of commercial AMF inoculants under certain soil and ecological conditions is a significant concern as well; native AMF species are frequently thought to be more symbiotic than exotic strains (Querejeta et al., 2006). The inability of certain commercial inoculants to establish mycorrhizal associations may be due to the exotic AMF inoculants' poor adaptability to local edaphic conditions, including soil nutrient concentrations and other abiotic variables (Schreiner, 2007).

Abiotic stress, especially drought, can severely impact soybean yield and productivity (Begna, 2020). Due to the unreliable and inconsistent rainfall, the crop may experience moisture stress at various developmental stages, leading to yield loss (Begna, 2020). Soybean is less vulnerable to water deficiencies during the vegetative stage but is more vulnerable during flowering, pod set, and pod filling. In this scenario of unprecedented and reliable rainfall, soil moisture conservation strategies can be used to conserve soil water in the field. Therefore, the development of affordable methods to mitigate drought stress and enhance seed production, particularly in semi-arid and arid environments, is urgently needed (Igiehon et al., 2021).

The increasing effects of climate change present enormous problems for agriculture in arid and semi-arid countries, endangering the resilience and sustainability of agri-food systems (Dzvene et al., 2025). Low rainfall conditions and soil fertility prevalence adversely affect crop growth and yield, nutritional security, and livelihoods of mainly rural households dependent on dryland production. In addition to decreasing soil and runoff losses, the use of moisture conservation techniques are helpful for effectively growing rainfed crops in dry and semiarid regions (Singh et al., 2018). Most soils in the smallholder farming systems are inherently low in nutrients. One such endeavor is the application of biological fertilizers, like arbuscular mycorrhizae. Arbuscular mycorrhizal fungi are one example of a biological antioxidant booster that has been used to increase plants' resistance to drought stress. In ecologically conscious agricultural techniques, arbuscular mycorrhizae fungi have the potential to enhance crop yield and soil condition, especially in nutrient-deficient soils (Rasouli et al., 2023). There is still a need to assess VAM inoculation and soil moisture conservation techniques on promiscuous soybeans under field conditions in dryland areas. These studies will help smallholder farmers to produce soybeans using moisture-saving techniques and growth-promoting in regions with marginally low rainfall. It is essential to capitalize on the potential role that VAM and soil moisture conservation measures can play in enhancing plant production, as climate change is already affecting agricultural yields in drylands. This study aimed to evaluate the interactive effects of AMF inoculation and different soil moisture conservation techniques on the growth, physiological performance and yield of a promiscuous soybean variety in the Limpopo Province of South Africa. The findings of this research will provide valuable insights into sustainable and eco-friendly approaches for enhancing soybean production in water-limited environments, contributing to food security and sustainable agricultural practice in the region.

Materials and Methods

Experimental Site

The experiments were conducted at the University of Limpopo Experimental farm (Syferkuil farm), Limpopo province, South Africa, during 2018/2019 and 2019/2020. The experimental site GPS coordinates are 23°50'02.7"S and 29°41'25.5"E, 1324 m above sea level. The experimental site received rainfall of 40-97 mm during the growing season. The average temperature ranged from 11°C and 30°C during the growing season (Figure 1). The highest rainfall pattern was observed during December 2018 and January 2019 (Figure 1). The soils are classed as Chromic Stagnic Plinthic Cambisol/Regosol and have sandy loam textures (average 15% clay content) with an effective depth of 900 mm (World Reference Base for soil resource, 2014; Van Huyssteen, 2020).

Treatment and Design

The experimental plot consisted of the following treatments: soil moisture conservation techniques

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(flat (control) and closed ridges) and vesicular-arbuscular mycorrhizae (VAM) (with and without). The experiment was laid out in a split-plot arrangement fitted within a completely randomized block design with four replications. Soil moisture conservation techniques (flat (control) and closed ridges) were assigned to the main plots, while VAM (*Glomus* spp.) (with and without) were assigned to the subplots. A promiscuous soybean variety (TGx1085-1F) was used as the test crop. The plot size was 3.6 m × 3 m (10.8 m²), with an inter- and intra-row spacing of 60 cm × 15 cm, the alley between the replications and the soil moisture conservation techniques.

Data Collection

Soil sampling and analysis
Soil samples were collected at the planting and harvesting stages at 0-30 cm depth. The soil samples were air-dried, sieved (5 mm sieve and 2 mm), and stored in airtight plastic bags for soil nutrient analysis. The soil pH (KCl) was determined using the 1:10 KCl method and measured using a pH meter (Mettler Toledo). The Mclean titration method was used to calculate exchangeable acidity. Available phosphorus (P) and potassium (K) were extracted using the Bray 1 and Mehlich-3 methods, respectively (Matcham et al., 2023). Magnesium (Mg) and calcium (Ca) were determined using atomic absorption spectrophotometry (Diwakar et al., 2023). Nitrogen (N), zinc (Zn), and manganese (Mn) were determined using the Kjeldahl method (Aguirre, 2023). Soil organic carbon was determined using the Walkley-Black (Mustapha et al., 2023) (Table 2).

Growth parameters

Plant height was measured using a meter ruler, and the number of leaves and branches per plant was counted on five randomly selected plants within each plot during the growing season.

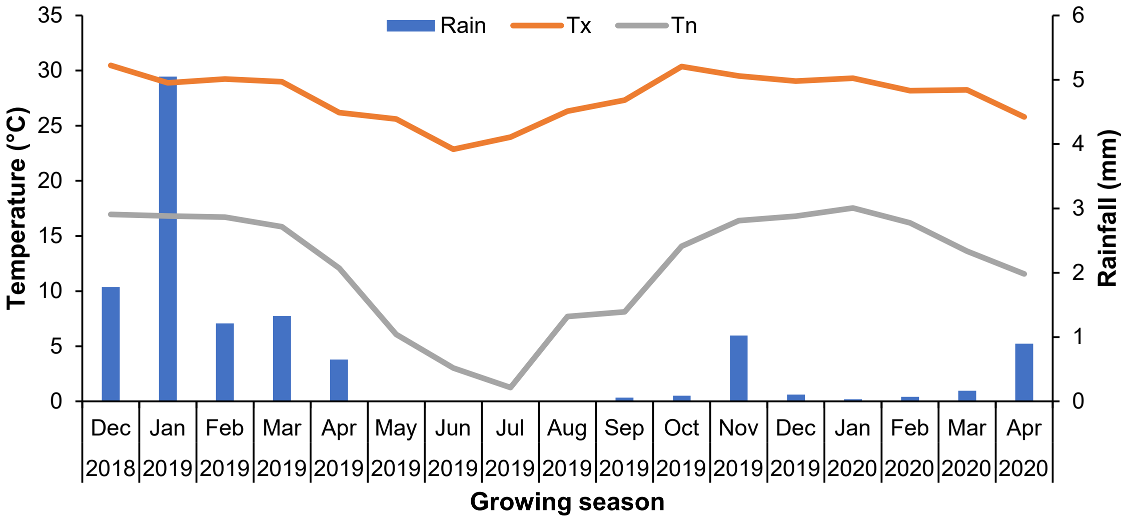

Figure 1. The weather data and average climatic conditions at Syferkuil. Tx = maximum temperature; Tn = minimum temperature

Table 1. The experimental design in the field

Rep 1		Rep 2		Rep 3		Rep 4
Flat planting		**Closed ridges**		**Flat planting**		**Closed ridges**
TGx1085-1F X Without VAM	Alley	TGx1085-1F X With VAM	Alley	TGx1085-1F X Without VAM	Alley	TGx1085-1F X With VAM
TGx1085-1F X With-VAM		TGx1085-1F X Without VAM		TGx1085-1F X With-VAM		TGx1085-1F X Without VAM
Alley						
Closed ridges		**Flat planting**		**Closed ridges**		**Flat planting**
TGx1085-1F X Without VAM	Alley	TGx1085-1F X With VAM	Alley	TGx1085-1F X Without VAM	Alley	TGx1085-1F X With VAM
TGx1085-1F X With-VAM		TGx1085-1F X Without VAM		TGx1085-1F X With-VAM		TGx1085-1F X Without VAM

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Physiological parameters

Chlorophyll content was measured using a chlorophyll meter (Opti-sciences, 8 Winn Avenue, Hudson, NH 03051, USA) during the vegetative stage. Plant vigor was estimated using a Trimble Green Seeker (Trimble Inc.) during the vegetative stage on sunny days.

Harvesting and yield determination

The net plot area for harvesting was 5.4 m^2, consisting of four rows. At maturity, the plants were uprooted and weighed to determine their total biomass. Yield parameters such as the number of pods per plant and number of seeds per pod were counted manually on 10 pods per treatment, 100 seeds weight, plant biomass, and grain yield were weighed using a weighing scale.

Experimental plant maintenance, VAM culture preparation, and inoculation procedures

The mycorrhizae used mycoroot (new root dimension, supergro). Mycoroot (Pty) Ltd, a Rhodes-based company, manufactures biofertilizers such Mycoroot Super Booster, Mycoroot Green, and Mycoroot SuperGold. At the time of planting, 100 $kg.ha^{-1}$ of mycorrhizal inoculum was uniformly distributed inside the row, and the soil was covered after the two seeds were planted. Two weeks after the emergence, the seedlings were thinned to one. The experimental sites were ploughed and harrowed to prepare a good soil tilth. The field was manually weeded to maintain a clean field. Scouting for diseases and pest damage was done regularly.

Data Analysis

The growth and yield attributes data were subjected to analysis of variance using STATISTIX version 10. The least significant difference (LSD) test at a 5% probability level was used to compare means among treatments (Gomez and Gomez, 1984).

Results and Discussion

Interaction of Soil Moisture Conservation and VAM on Soybean Growth

The interaction between soil moisture conservation techniques and vesicular-arbuscular mycorrhizae showed significant variation ($p<0.05$) in plant vigor. In contrast, non-significant differences ($p>0.05$) were observed in plant height, number of leaves, and chlorophyll content. The interaction between flat planting and application of vesicular arbuscular mycorrhizae (100 $kg.ha^{-1}$) recorded higher plant vigour (0.75) than the closed ridge and application of VAM (100 $kg.ha^{-1}$) (Table 3). In the current study, a rise in plant vigour may be attributed to enhanced nutrients and water uptake by mycorrhizae. Arbuscular Mycorrhizal (AM) fungi play a crucial role in the uptake of nutrients from the soil, particularly phosphorus (P), which enhances plant vigor and photosynthesis. VAM inoculation provides a safe, natural method to mitigate drought stress, promoting plant growth and reducing the need for chemical inputs. Another reason is that these could be associated with soil type and soil moisture content in the flat planting rather than the closed ridge. Soil disturbance decreases the benefits of mycorrhizae to crops and soil quality because it negatively impacts AM fungi (Wilkes, 2021). Future studies should investigate genotype-specific responses to AMF and focus on field research under various soil and climate conditions. Encouraging integration of AMF into climate-smart agricultural systems to protect crops,

Table 2. Physico-chemical properties at pre-planting for soil collected from Syferkuil during the 2018/2019 and 2019/2020 growing seasons

Soil parameters	Extraction methods	Value	Status
pH	KCl	7.09	Neutral
N%	Kjeldahl	<0.050	Low
Bulk density $g.ml^{-1}$)	Cylinder method	1.47	Favourable
P ($mg.kg^{-1}$)	Bray 1 method	0.002	Low
K ($mg.kg^{-1}$)	Atomic absorption spectrophotometer	0.20	Low
Ca ($mg.kg^{-1}$)	Atomic absorption spectrophotometer	0.75	Low
Mg ($mg.kg^{-1}$)	Atomic absorption spectrophotometer	0.48	Low
Zn ($mg.kg^{-1}$)	Atomic absorption spectrophotometer	0.00	Low
Mn ($mg.kg^{-1}$)	Atomic absorption spectrophotometer	0.018	Low
Exch acidity ($cmol.L^{-1}$)	Mclean titration method	0.07	Low
Organic C %	Walkley and Black	0.55	Low

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www.j-tropical-crops.com

Received 16/07/2024; Revised 13/06/2025; Accepted 01/08/2025
https://doi.org/10.29244/jtcs.12.03.483-491

improve drought resistance, and advance resource conservation and sustainable soil health.

Soil Moisture Conservation and VAM Effects on Soybean Yield

The interaction effect between VAM and SMCT showed no significant difference ($p<0.05$) on grain yield and yield attributes of the promiscuous soybean variety (Table 4). The interaction effect of soil moisture conservation techniques and VAM application showed significant differences ($p<0.05$) in shelling percentage (40.53%). Planting on flat without VAM application (0 kg.ha^{-1}) increased shelling percentage. This demonstrates the presence of indigenous or native AMF which formed a symbiotic relationship with soybean roots. The non-response of VAM can be associated with the infectiveness of the native strains in the soil, cropping history, abiotic factors and timing of application (Berruti et al., 2016; Mukhongo et al., 2023). The findings match those of Hindumathi and Reddy (2011) where the soil in the soybean field showed fluctuation in propagule number, and spore number at the time of harvest in the two fields. It is challenging to identify a consistent host response to inoculation because the effectiveness of the AM symbiosis is determined by soil fertility inoculation timing, site disturbance level, and maybe partner co-adaptation (Adeyemi et al., 2020). AMF inoculants are currently rarely used in commercial farming, and the majority of research is conducted in laboratory and greenhouse environments (de Souza Buzo et al., 2023). More research is needed in the field conditions to explore the effect of VAM on grain yield of soybean.

Correlation Between Growth, Yield Attributes, and Grain Yield of Promiscuous Soybean Variety

Plant height had a significant ($p<0.05$) and positive correlation with the number of leaves (r=0.5997), number of branches (r=0.2892), and plant vigour (r=0.6368) (Table 5). This implies that an increase in plant height values leads to a rise in the number of leaves, branches, chlorophyll content, plant vigour, number of pods, grain yield, and plant biomass. The results align with those of Jadhav et al. (2022), who reported a positive correlation (r=0.265) between stem height and the number of leaves.

The number of leaves showed a significant and positive correlation with the number of branches (r=0.2756), chlorophyll content (r=0.3528), plant vigor (r=0.5388), number of pods per plant (r=0.5388), and plant biomass (r=0.2621). According to this study, the growth characteristic significantly influencing soybean plant biomass is the number of leaves, while plant biomass is the most predictable parameter (Kasu-Bandi et al., 2019). There was a significant and positive correlation among the number of branches and chlorophyll content (r=0.3193), plant vigour (r=0.332), number of pods per plant (r=0.3372), number of seeds per pod (r=0.3824), and hundred-seed weight (r=0.3205). Plant vigour had a significant and positive correlation with the number of pods per plant (r=0.2327), grain yield (r=0.2894), and biomass (0.5084). An increasing number of leaves is a practical strategy for improving the total number of pods. This is explained by the part that leaves play in photosynthesis, which would have favored the development of pods while disadvantageously affecting the weight of the seeds within the pods (Kasu-Bandi et al., 2019).

The number of pods per plant showed a significant and positive correlation with pod length (r=0.3388), number of seeds per pod (r=0.9850), hundred-seed weight (r=0.3558), grain yield (r=0.4286), and harvest index (r=0.3712) (Table 5). The results are similar to Esan et al. (2023) who reported that the total number of pods had a strong, positive and highly significant correlation with the number of seeds per pod, yield per plant, hundred seed weight, yield per plot,

Table 3. Interaction effect between SMCT and VAM on growth attributes of promiscuous soybean variety (2018/2019 and 2019/2020 growing seasons)

SMCT	VAM	Number of leaves	Number of branches	Chlorophyll content (CCI)	Plant vigour
Flat	With-VAM	20.81[a]	5.08[a]	20.16[a]	0.75[a]
Flat	Without VAM	19.20[a]	5.09[a]	21.08[a]	0.73[ab]
Ridge	Without VAM	17.80[a]	4.76[a]	20.24[a]	0.72[ab]
Ridge	With-VAM	16.44[a]	5.31[a]	19.06[a]	0.70[b]
p value		ns	ns	ns	*
$LSD_{0.05}$		7.5855	2.2639	5.3510	0.1420

Notes: Means followed by the same letter in a column differ significantly at $p\leq0.05$. ns= not significant. SMCT= Soil moisture conservation techniques, *= significant. LSD= Least significant difference, VAM= Vesicular arbuscular mycorrhizae.

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https://doi.org/10.29244/jtcs.12.03.483-491

Table 4. Interaction effect between soil moisture conservation techniques and vesicular arbuscular mycorrhizae on grain yield and yield components of promiscuous soybean variety (2018/2019 and 2019/2020 growing seasons)

SMCT	VAM	Number of pods per plant	Pod length (cm)	Seeds per pod	Number of pods per plant	Hundred seeds weight (g)	Grain yield ($kg.ha^{-1}$)	Plant Biomass ($kg.ha^{-1}$)	Harvest index (%)	Shelling percentage
Flat	Without VAM	219.31^{a}	3.87^{a}	2.90^{a}	638.56^{a}	8.93^{a}	1382.7^{a}	6273.3^{a}	23.69^{a}	40.53^{a}
Ridge	With-VAM	207.38^{a}	3.92^{a}	2.93^{a}	608.57^{a}	8.67^{a}	1135.9^{a}	4997.5^{a}	23.65^{a}	38.44^{ab}
Ridge	Without VAM	205.40^{a}	3.88^{a}	2.93^{a}	601.98^{a}	8.94^{a}	1010.4^{a}	4964.3^{a}	22.20^{a}	37.78^{ab}
Flat	With-VAM	202.72^{a}	3.89^{a}	2.91^{a}	592.95^{a}	9.05^{a}	1190.4^{a}	5609.1^{a}	22.79^{a}	36.66^{b}
p value		ns	ns	ns	ns	ns	ns	ns	ns	*
$LSD_{0.05}$		60.307	0.2100	0.1282	179.94	1.4900	462.92	3649.6	3.9301	4.3485

Notes: Means followed by the same letter in a column differ significantly at p≤0.05. ns= not significant. SMCT= Soil moisture conservation techniques, *= significant. LSD= Least significant difference, VAM= Vesicular arbuscular mycorrhizae.

Table 5. Correlation analysis between growth, yield and yield attributes of promiscuous soybean variety (2018/2019 and 2019/2020 growing seasons)

	PH	NL	NB	CC	PV	NPPA	PL	SPPA	NSSP	HSW	GRA	BIO	HI
PH	1												
NL	5.5997**	1											
NB	0.2892**	0.2756*	1										
CC	0.1750	0.3528**	0.3193**	1									
PV	0.6368**	0.5388**	0.3322**	0.3002**	1								
NPPAV	0.1201	0.2621*	0.3372**	0.2763*	0.2327*	1							
PL	-0.0547	0.0470	0.1548	0.0757	0.0947	0.3388**	1						
SPPA	-0.0361	0.0392	0.0935	0.0200	0.0181	0.0848	0.4311**	1					
NSSP	0.1013	0.2573	0.3824**	0.2653*	0.2288*	0.9850**	0.4127**	0.2412*	1				
HSW	0.0743	0.1664	0.3205**	0.2633*	0.2460*	0.3558**	0.0703	0.1150	0.3619**	1			
GRA	0.1488	0.1764	0.0941	0.0909	0.2894**	0.4286**	0.1919	0.1005	0.4334**	0.5262**	1		
BIO	0.3113	0.2669*	0.0406	0.0990	0.5084**	0.2087	0.1797	0.347	0.2107	0.3171**	0.5772**	1	
HI	-0.1110	-0.0839	0.0656	-0.0628	-0.1539	0.3712**	0.1134	0.1139	0.3787**	0.4024**	0.5460**	-0.1610	1
SP	-0.1507	-0.0668	0.0765	-0.0431	-0.1390	0.2002	-0.0862	0.0108	0.1921	0.3580**	0.5404**	0.009	0.6735

Note: PH-Plant height, NL-number of leaves, NB-number of branches, CC-Chlorophyll content, PV-plant vigour, NPPAV-Number of pods per plant, PL-Pod length, SPPA-Seeds per pod, HSW-hundred seeds weight, SP-shelling percentage, HI-Harvest index, GRA Grain yield and Bio- plant biomass, *-significant at p<0.05 and **-significant at p<0.01.

Received 16/07/2024; Revised 13/06/2025; Accepted 01/08/2025
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shelling percentage, harvest index, and yield per plot of unshelled on Bambara groundnut. This suggests that increasing the value of one of these parameters causes the parameter with which it has a strong correlation to increase as well (Malik et al., 2011). Pod length has a significant and positive correlation with the number of seeds per pod (r=0.4311) and the number of pods per plant (r=0.4127). The number of seeds per pod had a significant and positive correlation with the number of pods per plant (r=0.2412). Number of pods per plant had a positive and significant correlation with hundred-seed weight (r=0.3619), grain yield (r=0.4334), and harvest index (r=0.3787). Pod length, number of seeds per pod, and number of pods per plant are the major contributors to grain yield. The two characteristics that contributed most to grain yield were the number of pods and the amount of chlorophyll (Abderemane et al., 2023). Hundred-seed weight had a strong positive and significant correlation with grain yield (r=0.5772), plant biomass (r=0.3171), harvest index (r=0.4024), and shelling percentage (r=0.3580). The correlation matrix revealed that yield was positively and strongly associated with pod length (r=0.5772), harvest index (r=0.5460), and shelling percentage (r=0.5404) (Malik et al., 2023).

Conclusions

The interaction effect between soil moisture conservation techniques and VAM had a significant impact on plant vigor ($p<0.05$). In contrast, no significant variation ($p>0.05$) was observed in the number of branches, plant height, grain yield, and yield attributes. The study demonstrated that flat planting, in combination with VAM, improved plant vigor under moisture-limited conditions. However, the interaction effects on grain yield were not significant, likely due to indigenous AMF activity. Future studies should focus on identifying and utilizing native AMF strains to enhance soybean productivity in semi-arid regions.

Acknowledgements

The National Research Foundation is acknowledged for the bursary of SS Thosago. Prof D Maposa is recognized for his contribution to the study.

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Received 16/07/2024; Revised 13/06/2025; Accepted 01/08/2025
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