SUST Journal of Agricultural and Veterinary Sciences (SJAVS)
Vol. 18 No.(2)
ISSN: 1858 6775

December 2017

SUST
Journal of Agricultural and Veterinary Sciences
Journal homepage:
http:// journals.sustech.edu

Variation in Seedling Growth and Morphology of *Adansonia digitata* in the Sudan: A way Forward for Promoting Species Cultivation

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Article History: Received: 01/02/2017
Accepted: 15/04/2017

Abstract
The scope of variation in seedling characters of the baobab trees were evaluated at environmental variables level in the Blue Nile and North Kordofan states in Sudan. The present study was conducted in deeming (2014 and 2015) where Baobab seedlings were grown in the nursery of the Faculty of Natural Resources, University of Sinnar. Seedlings were grown for 18 weeks and several morphological characteristics (Stem, hypocotyl, epicotyl, medial leaf, taproot and Root length, stem diameter, number of leaves, Medial leaf Width and thickness, tap root diameter and stomata density, among others) were recorded at different harvesting times, their growth and morphology were studied. The study revealed significant differences (P≤ 0.05) in seedling growth and morphology parameters. In general seedlings from North Kordofan State (drier area), were smaller in the overall measurements. They showed fewer leaves, higher number of stomata, characteristics frequently linked to drought adaptation, in order to accumulate more water as well as avoiding water loss. Results from this study show that there is a great variation in seedling growth and morphology of baobab, which gives opportunities for selection of superior planting material.
Keywords: Seedling characteristics, Domestication, Adansonia digitata, Native fruit trees.

Introduction
African baobab (*Adansonia digitata* L.), belongs to the family *Bombacaceae* Muchvalued tree for the livelihood of rural population in arid zones of tropical Africa, which has multiple uses; numerous medicinal properties, income generation, fodder and food uses (Jamnadass *et al*., 2009; Leakey *et al*., 2007). The fruits are an important source of cash income for the certain tribes living in Central and South Sudan (Bonkoungou *et al*., 1999; Gebauer *et al*., 2002). Therefore domestication of indigenous fruit trees can play an important role in the achievement of the Millennium Development Goals in developing countries (Leakey, 2003).

In fact, this species has been recognized as one of the most important edible forest trees to be domesticated in Africa (Eyog-Matig *et al*., 2002). The species stands have been exposed to over exploitation, because of the increasing international trade of baobab products and the growing human population in Africa which would have negative impacts on local livelihoods in Africa (Buchmann et al., 2010). Therefore domestication and cultivation of the baobab tree is necessary, and this may help to protect the species, need to screen more material to get super-trees as well as enhance the food security and income generation for the rural population.

Considerable variability in fruit phenotypic, characters of leaves morphology, seedling growth, of baobab has already been reported for this species, offering selection opportunities for conservation, domestication and improvement (Cuní *et al*., 2010, 2011; De Smedt *et al*., 2011; Soloviev *et al*., 2004)

The baobab tree is tenderly called the "Tree of Life". Baobab has traditionally been propagated by transplanting naturally regenerated seedlings. However, seedlings are rare due to intensive browsing by livestock. In the field, seedlings do not emerge immediately after seeds are released from fruits due to a dormancy imposed by the hard seed coats which appear to be non-permeable. (Sidible and William 2002).

Seedlings produce a strong prominent taproot but this is soon replaced by laterals. Seedlings have big, flat and paired cotyledons, and the first leaves are petiolate, generally narrow, simple and linear. Effective protection against livestock is essential after planting the seedlings (Gebauer 2002). Baobab seedlings and saplings are absent from many populations in southern Africa, suggesting that there are significant recruitment bottle necks (Chirwa *et al.*, 2006).

Recent work has shown that the substrate used for germination and the climatic zone from which seeds are collected both have a significant influence on both germination and seedling growth (Assogbadjo *et al.*, 2010; Cuni *et al.*, 2011).

Germination is hypogeal (Baum, 1995). Soon after germination, seedlings develop simple leaves and a swollen carrot-like tap root. The tap root functions as a water storage organ until the stem develops water-storage capacity, after which it reduces over time. Once this is developed, usually within the first three months, the young plant is referred to as a 'sapling' (Wickens and Lowe, 2008).

It is believed that the baobab tree has long seed dormancy (Owen 1974, Wickens 1982). In nature, this dormancy is believed to be broken by fire, a long period of rain or digestion by elephant or other big mammals (Wickens 1982, Wickens and Lowe 2008). Baobab seedling's first leaves are simple, followed by digitated leaves with progressively more leaflets, up to 5-7. Seedlings and young saplings lack the characteristic swollen trunk of the adults. The sapling develops a deep taproot (Cuni 2010).

Little is known about the viability and persistence of seeds within natural or human-modified environments, or the factors that limit seedling establishment (Wilson1988).

Differences in baobab growth rates in different countries have been related to differences in climate (Wilson 1988, Wickens and Lowe 2008). Regardless of the environment, it is accepted that baobabs grow quickly during the early part of their lives while the rate of growth slows later. Patrut *et al*., (2007). Baobab seeds (*Adansonia digitata*) have very hard seed coats and germination is usually under 20% as recorded by Danthu P., (1994).

Surprisingly little is known about natural regeneration rates, but to a large degree this could be because seedlings are not readily recognized since they lack the obvious palmately digitat leaves and swollen trunks (sidble and William 2002)

In general, trees are selected based on leaves. For this reason wild trees are chosen with a desired quality and seedlings (wildlings), occasionally cuttings, are transplanted to fields near homes where they can receive 'protection'. (Sidble and William 2002). It has been reported that seedling morphology and leaf anatomy was linked to drought tolerance (Teklehaimanot *et al*. 1998). However, no studies have been undertaken on baobab leaf size and shape variation.

Domestication and cultivation of the baobab tree is needed to protect natural stands and to improve livelihood to local populations in Sudan. Pye-Smith, (2010) reported that the domestication process seeks to capture and multiply trees with desirable characteristics, thus taking advantage of variations found in the wild.

In fact, The considerable size differences which exist in quality of fruit and leaf morphology, and nutritional value has already been reported for this species, which gives the chance to select high quality superior sources of planting material (Gurashi 2015, De Smedt *et al*., 2011, Cuní Sanchez *et al*., 2011, Chadare *et al*., 2009 Assogbadjo *et al*., 2005, Soloviev *et al*., 2004).

Consequally, it is probable that there are also differences in seedling growth and morphology between baobab trees provenance from Blue Nile and North kordofan states in Sudan.

Therefore aim of this study was to investigate the differences in baobab seedling growth and morphology between Blue Nile and North Kordofan States from Sudan, and determine if superior seedling characteristics of baobab tree can be selected.

Materials and Methods

***Site description*:**

The study was performed on baobab (*Adansonia digitata* L.). Population established within five locations from each North Kordofan and Blue Nile states in Sudan. North Kordofan state located roughly between latitude 10.5˚ and 15˚ north and longitude 27.5˚ and 32˚ east, it occupies a total area of about 185302 km². Blue Nile state located between latitude 10˚ and 13˚ North and longitude 33˚and 36˚ east, and occupies a total area about 38500km² (Figure 1 & Table 1).

***Study Methodology*:**

The present study was conducted two times partly late 2014 and then early 2015 in two states namely *Blue Nile* and *North Kordofan States* Sudan. The Different locations in study sites were selected based on their distribution along a rainfall gradient and the basis of the presence of baobabs trees with well-established and fruit-bearing as well as the domination of the species.

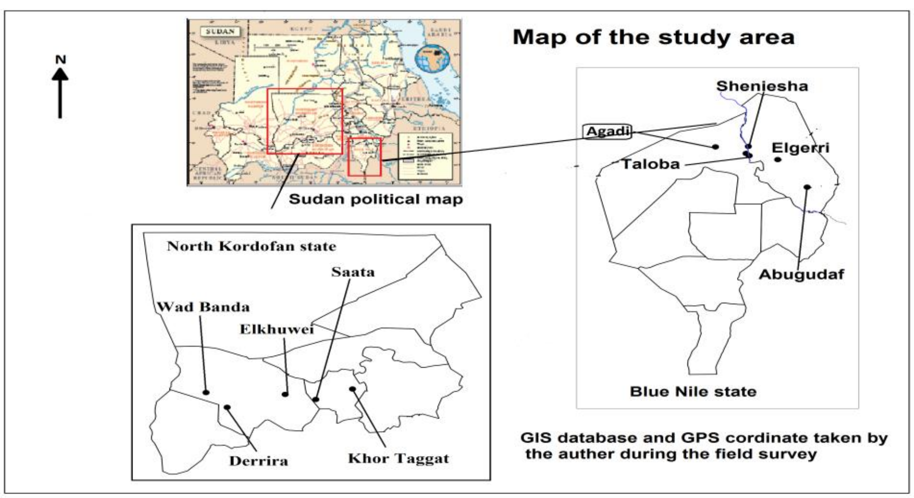


Figure 1: Map of Blue Nile and North Kordofan states.

Table 1: Details of the geographical location of Adansonia digitata

State	Locations and code	Altitudes (m)	Soil	Lat. (N°)	Long. (E°)	Average rainfall (mm)
Blue Nile	Taloba (T) (n = 20)	474	Stony hill side	11°49'	34°23'	700
	Sheneisha (Sh) (n = 12)	465	Silty clay	11°57′58.0	34°22′09.5	700
	El Gerri (G) (n = 20)	538	Stony hill side	11°49′00.1	34°36′31.3	700
	Abugudaf (AG) (n = 7)	550	Clay	11°31′04.4	34°50′52.2	700
	Agadi (A) (n = 5)	522	Stony hill side	11°57′25.8	34°06′23.4	700
North Kordofan	Kour Taggat (KT) (n = 20)	560	Sandy Loam	13°11′	30°18′	350
	El Saata (S) (n= 10)	512	Sandy Loam	12°58′ 014	29°52′	350
	ElKhuwei (Kh) n = 20)	530	Sandy Loam	13° 04′	29° 13′	350
	Darrira (D) (n = 10)	565	Sand	12°47′ 20.5	28°16′ 99	350
	Wad Banda (W) (n = 7)	623	Sandy loam	13°06′401	27°56′62	350

Source: Ministry of Agriculture of Blue Nile and North Kordofan states (2013), General Department of Planning and information

Fruits from ten populations (five form each state) were collected. Then the fruits were cracked and washed to remove away the dry powder and the seeds were dried for 2 hours under direct sun light, for this experiment, seeds with similar weight were selected. To facilitate the germination, the seeds were soaked in hot water and then left to soak for 24 hours without heating (Cass 1995).The seeds were planted in polythene bags with river sand using CRD (randomized block design) with 20 repetition (one seed per repetition) per treatment in the nursery of the Faculty of Natural Resources and Environmental Studies, Elsuki Campus, University of Sinner and for 18 weeks of following up. The seedlings were irrigated twice a week. Six healthy looking seedlings were selected per a treatment and then growth parameters (stem- epicotyl and hypocotyl, taproot and total root length, stem length and taproot diameter at largest width) using ruler and using digital caliper as well as number of leaves was recorded at different time intervals (week 1, week 3, week 6, week 10, week 14 and week 18) (Figures 2 and 3).

The data was analyzed using Statistical Package for Social Sciences (SPSS) version16. To detect the significant variations of seedlings growth parameters in relation to the fruit shape types. ANOVA analysis was applied.

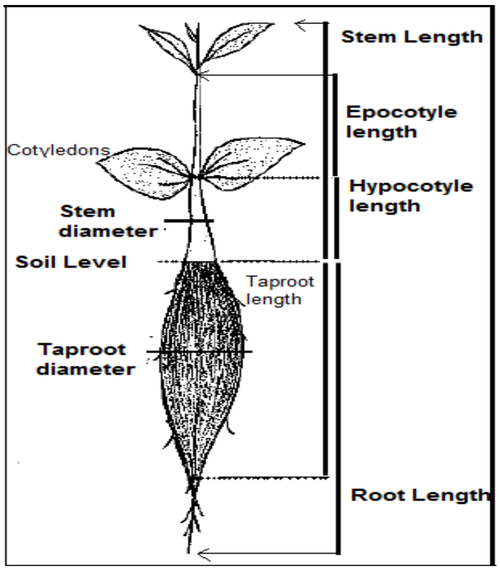


Figure 2: Characteristics record

Figure 3: Seedling of *Adansonia digitata* from each seedling Cuni (2010)

Results and Discussion

The results indicated that there were significant differences (P≤ 0.05) between seedlings cotyledon leaf from Blue Nile and North Kordofan state in relation to their length and width. The study findings also illustrated that seedlings from Blue Nile (wetter sites) had larger cotyledon length (6.74±1.19cm) and width (4.73±0.86cm) (Table 2).

Table 2: Variation on cotyledon of Baobab seedling from BN and NK

	Blue Nile state	North Kordofan state
Cotyledon length (cm)	6.74±1.19 a	5.51±1.23b b
Cotyledon width (cm)	4.73±0.86 a	4.42±0.69 b

Means followed by the different letter within arrow for each state are significantly different from each other at p<0.05)

Whereas cotyledon leaf size from the drier sites in North Kordofan (Elkhuwei) was shorter with the length (5.51±1.23cm) and width (4.42±0.69cm) Figure (4). These results were due to the fact that differences in cotyledon morphology may be related to drier area adaptation (Fujita and Humphreys 1992 and Fisher 2008). Similarly, Pock Tsy *et al*. (2009) reported that seedlings from Mali (West Africa baobab population) have their cotyledons at a lower height than those from Malawi (East Africa baobab population).

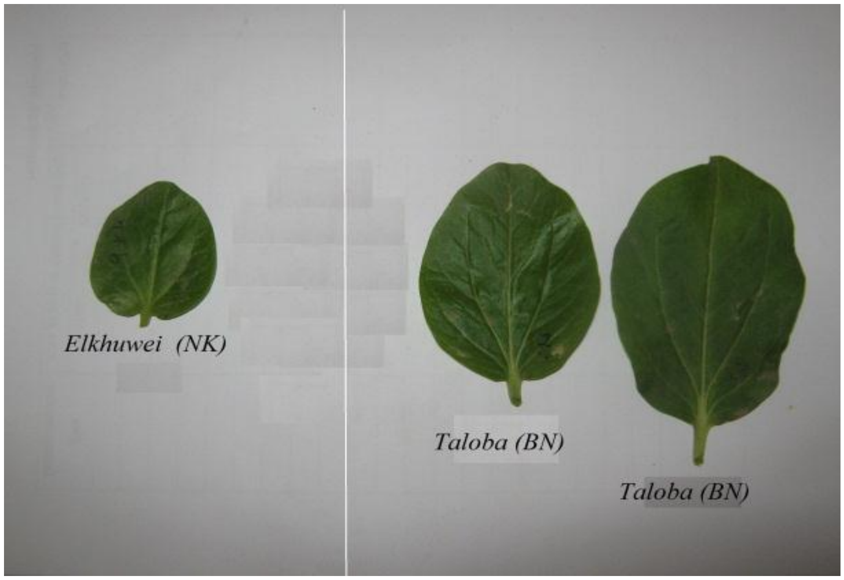


Figure 4: Cotyledon leaf variations of Taloba (BN) and Elkhuwei (NK) Seedlings.

After 6 weeks, it was observed that there were significant differences between seedlings medial leaf thickness, taproot length, taproot diameter and root length. Seedling from the Blue Nile state has thicker leaves than those from North Kordofan state. Whereas taproot length, taproot diameter and root length from Blue Nile were longer than those from North Kordofan in 6 weeks (Table 4.11).

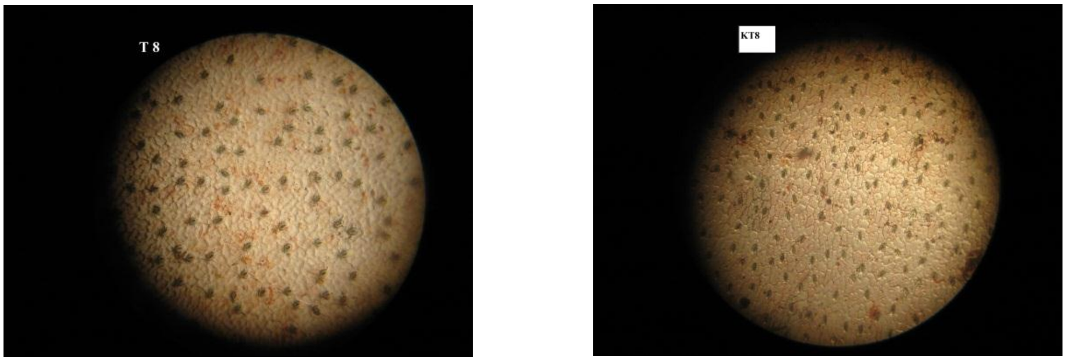
After 10, 14, and 18 weeks, seedling stem and taproot lengths from Blue Nile states (wetter areas) were found to be longer than those from North Kordofan state (drier area). Also the number of leaves from Blue Nile State after 10, 14 and 18 were found to be higher than those from North Kordofan state. After 14 and 18 weeks ratio (taproot length/taproot width) from Blue Nile state were found to be higher than those from North Kordofan state (Table 3). These findings agreed with the results of Pock-Tsy *et al*. (2009) who reported that the differences in seedling morphology may support the genetic differences between baobab populations from West Africa and south-eastern Africa. In Malawi, Cuni, (2010) also reported significant differences between study sites regarding leaf and fruit characteristics. Similarly, Assogbadjo *et al*. (2011) argued that there were differences between study sites within the one country. The results were attributed to the fact that the climatic conditions (rainfall, temperature, humidity, and etc…) have influences on seedling characteristics (e.g. stem height, diameter and number of leaves) growth within and between geographical locations. Additionally, differences in seedlings growth are related to variation in seeds size which is ultimately affected by geographical locations. In their study, Parker *et al*. (2006) observed a positive influence of large seed size on seedlings establishment and early growth.

Table 3: Seedling characteristics of *A. digitata* from the two states after 6, 10, 14 and 18 weeks after germination

Characters	6Week		10Week		14Week		18 week	
	BN	NK	BN	NK	BN	NK	BN	NK
Stem length (cm)	34.7± 6.39 a	32.5± 5.65 a	40.6± 7.19 a	35.7± 8 b	42.6± 9.36 a	34.0± 7.71 b	46.3± 8.37 a	35.7± 9.14 b
Stem diameter (mm)	5.4± 2.46 a	5.82± 0.94 a	8.65± 1.54 a	7.56± 1.1 b	9.32± 1.8 a	9.7± 10.9 a	10.0± 1.72 a	8.69± 1.37 b
Hypocoty length(cm)	2.7± 0.62 a	2.69± 0.64 a	2.86± 0.55	2.76± 0.66 a	3.38± 0.77 a	2.71± 0.42 b	4.01± 1.19 a	3.44± 1.97 a
Epocotyl length (cm)	4.76± 1.31 a	4.69± 0.87 a	4.61± 0.9 a	4.53± 1.03 a	4.5± 1.21 a	5± 0.67 a	3.97± 1.08 a	3.39± 0.73 a
Number of leaves	12.77± 2.87 a	11.79± 2.15 a	18.71± 3.82 a	12.85± 3.26 b	19.49± 5.21 a	13.28± 2.95 b	21.13± 4.6 a	13.09± 3.82 b
Medial leaf length(cm)	10.73± 1.52 a	11.15± 1.66 a	10.9± 1.82 a	11.9± 1.99 a	11.98± 1.71 a	11.56± 1.83 a	11.13± 2.1 a	12.91± 3.13 a
Medial leaf width(cm)	4± 0.76 a	4.06± 0.62 a	4.09± 0.57 a	4.28± 0.82 a	4.23± 0.72 a	4.12± 0.88 a	4.02± 0.53 a	4.5± 1.25 a
Medial leaf Thickness (mm)	0.26± 0.18 a	0.13± 0.08 b	0.16± 0.13 a	0.13± 0.13 a	0.11± 0.02 a	0.1± 0.01 a	0.1± 0.01 a	0.1± 0.01 a
Taproot length (cm)	6.37± 2.54 a	4.56± 1.01 b	9.73± 2.73 a	8.92± 2.18 b	13.9± 2.89 a	11.43± 2.11 b	16.93± 2.89 a	14.07± 3.84 b
Taproot diameter(cm)	0.995± 2.5 a	0.82± 1.13 b	1.81± 0.48 a	1.89± 0.67 a	2.42± 0.32 a	2.37± 0.38 a	2.46± 0.27 a	2.46± 0.27 a
Ratio L/D	6.32± 1.42 a	5.55± 0.89 a	5.59± 1.53 a	8.33± 14.1 a	5.74± 1.13 a	4.91± 1.05 b	6.93± 1.44 a	5.78± 1.6 b
Root length (cm)	18.0± 3.38 a	14.5± 5.23 b	20.9± 5.11 a	22.7± 4.7 a	23.4± 4.87 a	23.2± 3.44 a	31.6± 5.98 a	27.2± 6.09 a

Mean with the same letter in a column are not significantly different from each other at $p<0.05$ using Duncan New Mull. Range Test.

The findings revealed that there was a significant difference ($P\leq 0.05$) in stomata density after 3 weeks between the studied states (Table 3). The same table indicated that stomata density was found to be lower ($93.94\pm21.6mm^2$) in Blue Nile state than those from North kordofan state ($110\pm13.5mm^2$) (Figure 5). This result may be due to the fact that baobab seedlings are adapting drought in order to accumulate more water as well as avoiding water loss. This fact agreed with Cuni (2010) who reported that high stomata density with small size is thought to be a characteristic of drier area adaptation.



Blue Nile state *North Kordofan state*

Figure 5: Stomata density variation on the abaxial surface of baobab seedlings.
Picture taken at under a light microscope at x100 magnification

The study findings in the same table indicated that there were no other significant differences between states in recorded morphological characteristics after 3 weeks (Table 4). This was due to the fact that in the first week after germination, the seedlings directed their growth towards fine roots and leaves, and later in the taproot formulation which functioning as the main organ for water store (Table 4).

Table 4: Significant differences in seedling characteristics of *A. digitata* from the two states after 3, 6, 10 and 14 weeks

Character	3 Week	6 Week	10 Week	14 Week
Stem length (cm)			*	*
Stem diameter (cm)			*	
Hypocotyl length (cm)				*
Epicotyl length (cm)				
Number of leaves			*	*
Medial leaf length (cm)				
Medial leaf width (cm)				
Medial leaf thickness (mm)	*	*		
Taproot length (cm)		*	*	*
Taproot diameter (cm)		*		
Root length (cm)		*		
Ratio				*
Number of stomata	*			

* indicates significant differences between study sites within BN and NK at $p<0.05$ (ANOVA).

Conclusion and Recommendations

A great variation in baobab seedling growth and morphology has been recorded for this species, which have suggestions for baobab domestication. Within the seedlings growth, it was observed that the wetter study areas recorded long length of the growth parameters under investigation in relation to those in drier study areas. Seedlings from drier state (North Kordofan) allocated they had higher number of stomata, a way often related to drought adaptation. Differences in some characteristics could be suggested to be related with genetic differences between baobab populations from the study area.

The current study recommended more research on genetic variation of baobab (biotechnology research work) are a necessity prerequisite to selection and conservation programs.

Acknowledgements
We would like to thanks the local people who deal with baobab product activities in the study area (*Elgerri*, *Taloba*, *Elkhuwei* and *Derrira*) for their welcome reception and provision of the necessary data. Administrative Units and colleagues at Faculty of Forestry, University of Khartoum, especially, Dr. Yahia Omar Adam for providing space during the write up.

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الاختلافات في نمو وشكل شتلات التبلدى (*Adansonia digitata* L.) في السودان في اتجاه تطوير زراعة الانواع

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المستخلص

تم تقييم نطاق الاختلافات في صفات شتول اشجار التبلدى علي مستوى التباين البيئي بولايتى النيل الازرق وشمال كردفان. اجريت هذه الدراسة خلال عامى 2014 – 2015، زرعت شتول التبلدى في مشتل كلية الموارد الطبيعية والدراسات البيئية جامعة سنار، تمت دراسة النمو و الشكل الخارجي للشتول، زرعت الشتلات لمدة 18 اسبوع تم تسجيل عدة صفات شكلية هى طول كل من: (الساق، الساق التحت فلقي، الفوق فلقي، الجزور، الجزر الوتدى، الوريقة في المنتصف)، قطر الجزر الوتدى وكثافة الثغور من بين الصفات الاخرى والتى تم تسجيلها في اوقات مختلفة، تمت دراسة النمو والشكل الخارجي لها. اوضحت الدراسة أن هنالك فروقات معنوية (احتمالية ≤ 0.05) في قياسات نمو وشكل البادرات. بصوره عامة كانت الشتلات من شمال كردفان (المنطقة الاكثر جفافا) كانت اصغر في القياسات عامة، اقل عدد اوراق، اكبر عدد من الثقور، الخصائص هذه كثيرا ما ترتبط بالتكيف علي الجفاف، من اجل مراكمة المزيد من المياه فضلا عن تجنب فقدان المياه. اظهرت النتائج من هذه الدراسة ان هنالك اختلافات كبيرة في نمو وشكل البادرات مما يساعد علي مزيد من الفرص لاختيار العينات الزراعية الجيدة.