

The Effect of Drought on Growth and Dry Matter Allocation in Seedlings of *Vigna ambacensis* L.

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Abstract. This study describes drought effects on the growth and performance of seedlings of cowpea (*Vigna ambacensis* L.), to evaluate possible adaptive mechanisms for drought. Seedlings were grown in plastic pots filled with a mixture of sandy-loam soil and peat (3:1 ratio) and were subjected to four drought levels, in glasshouse conditions. Plant growth analysis was conducted over three consecutive harvesting dates. Growth rates of leaf area and plant height during the course of the experiment were either slowed down or reduced by drought. Six weeks of the severest droughting resulted in progressively smaller leaf area (23%), shoot (22%) and total dry weight (29%), and RGR, but increased root length (285%), all compared to the well-watered control. Root:shoot ratio increased from 0.16 to 0.53 with greatest droughting. These responses were partly responsible for enabling the plant to tolerate drought.

Introduction

Soil water stress is one of the most important edaphic factors limiting crop production all over the world where irrigation is practised or rainfed crops are grown [1], causing acute problems for agriculture in arid and semi-arid regions and imposing constraints on the distribution of cultivated plant species [2].

Droughting can significantly influence plant performance and survival and can lead to major constraints in plant functioning, inducing a series of morphological, physiological and metabolic changes [3-5].

Furthermore, droughting affects photosynthesis directly and indirectly and consequently dry matter production, and its allocation to various plant organs [6], and final plant yield; droughting determines time of germination [7-9], influences relative growth rate and root:shoot ratios [10; 6-11], causes 'absolute' root growth [12-13] and may cause premature death [14]. The occurrence of drought at the vegetative stage causes reduction in plant growth and leaf number [15]; it also reduces grain yield and dry matter production [16].

Drought is particularly important in hot semi-arid parts of Saudi Arabia where *Vigna ambacensis* L. is grown in a hostile environment. Low soil water and atmospheric moisture stress are the main cause of low crop production resulting from high air temperature, low and erratic rainfall and high evapotranspiration rate [17].

Vigna ambacensis L. is a wild legume species native to south-western Saudi Arabia [18]. Food legume crops are widely grown as a source of protein in semi-arid and tropical regions [11]. Since there is no information available regarding the response of species of *Vigna* at the vegetative stage to drought, this study was initiated to describe the growth performance and the adaptive drought mechanisms of *V. ambacensis* at the early vegetative growth stage.

Materials and Methods

Seeds were collected from the Jizan area of south-western Saudi Arabia in summer 1994, stored in a closed container and kept in a laboratory refrigerator at 5 °C. Ten seeds of *Vigna ambacensis* were sown in 25 cm diameter pots filled with a mixture of sandy-loam soil and peat (3:1). These pots were irrigated daily with tap water till germination was complete and additional watering was given weekly using half-strength Hoagland nutrient solution [19] to ensure adequate nutrition. Four weeks after complete germination, seedlings were thinned to four per pot and on the sixth week seedlings were subjected to four irrigation treatments with four replicates, each randomized in a glasshouse (approximate light intensity as photosynthetically active radiation (PAR) \cong 454 $\mu\text{mol m}^{-2} \text{s}^{-1}$; day and night temperatures \cong 25/20 °C and relative humidity \cong 45%). The water regime was as follows: $T_0 = 230 \text{ ml}$; $T_1 = 160 \text{ ml}$; $T_2 = 110 \text{ ml}$ and $T_3 = 46 \text{ ml}$ per pot applied at two-day intervals. These regimes corresponded to about 100, 70, 50 and 20% of extractable soil water relative to field capacity. Each pot also received 230 ml half-strength Hoagland solution once a week.

There were four harvesting dates: $H_0 = 6$ weeks ; $H_1 = 8$ weeks ; $H_2 = 10$ weeks; and $H_3 = 12$ weeks after seedling emergence; H_0 was used as the initial harvest. At each harvest 16 plants from each treatment were harvested and the leaf area of each plant was determined using a portable leaf area meter (Li-3000 (Li-Cor)); stem and root lengths were measured and oven-dry weight was taken for all plant parts at 85°C after 72 hr. Relative growth rate was calculated using data of plants harvested at H_0 as an initial growth measure; also calculated were root: shoot ratio (R:S), specific leaf area (SLA), relative growth rate (RGR) and net assimilation rate (NAR) according to Evans [20].

Results

The results of different measures of growth of *Vigna ambacensis* cultured under different soil moisture stress are presented in Tables (1 & 2) and Figs. (1 & 2). Soil moisture stress at the first harvest H_1 had only a small effect on most of the measurements up to two weeks from the initiation of the droughting regime, except for the driest treatment T_3 which compared to the control treatment (T_0) resulted in a significantly shorter stem height (49% of T_0 value), smaller leaf area (45%) and lower stem dry weight (75%), and increased root length (117%); there was also yellowing of the older leaves. Other measures were also influenced by droughting to different extents (Table 1).

Table 1. Effect on measures of growth (mean values) of *Vigna ambacensis* (R:S; SLA; RGR and NAR), under different droughting treatments. Mean values within a column with different lower case letters differ significantly at the $P < 0.05$ level, as determined by LSD test

Harvest & treatment		R:S ratio		SLA	RGR	NAR
		Weight	Length	$\text{cm}^2 \text{mg}^{-1}$	$\text{mg mg}^{-1} \text{day}^{-1}$	$\text{mg cm}^{-2} \text{day}^{-1}$
H_0		0.17	0.54	0.064		
H_1	T_0	0.19 a	0.30 d	0.47 a	0.024 a	0.008 c
	T_1	0.12 bc	0.34 c	0.30 b	0.035 b	0.0155 b
	T_2	0.11 c	0.42 b	0.26 c	0.041 c	0.0199 a
	T_3	0.14 b	0.71 a	0.31 b	0.046 d	-0.0023 d
	LSD	0.03	0.02	0.017	0.0054	0.0015
H_2	T_0	0.17 cd	0.19 d	0.29 ns	0.051 a	0.024 a
	T_1	0.14 d	0.29 c	0.29 ns	0.047 b	0.022 b
	T_2	0.24 b	0.56 ab	0.28 ns	0.037 c	0.021 c
	T_3	0.39 a	0.88 a	0.29 ns	0.025 d	0.015 d
	LSD	0.02	0.04	0.016	0.0028	0.012
H_3	T_0	0.16 d	0.13 d	0.24 a	0.041 a	0.019 a
	T_1	0.17 c	0.41 c	0.23 a	0.038 b	0.020 a
	T_2	0.27 b	0.77 b	0.22 a	0.032 c	0.021 d
	T_3	0.53 a	1.74 a	0.33 b	0.011 d	0.0066 b
	LSD	0.01	0.04	0.021	0.0016	0.0008

Table 2. Drought effect on plant growth measures as percentage changes relative to the control for harvests H₁, H₂ and H₃

Treatment & harvest	Leaf per plant		Stem per plant		Shoot dry Weight	Root per plant		Total dry Weight	NAR	RGR
	Area	Dry weight	Number	Height		Dry weight	Dry weight			
T ₀	100	100	100	100	100	100	100	100	100	100
H ₁ T ₁	84	125	102	84	124	80	106	117	176	145
T ₂	79	145	99	69	137	78	98	128	226	170
T ₃	45	67	99	49	72	55	117	67	-26	191
T ₀	100	100	100	100	100	100	100	100	100	100
H ₂ T ₁	88	85	73	59	90	93	89	87	92	92
T ₂	58	60	60	43	64	93	125	68	85	72
T ₃	37	35	44	38	40	95	172	48	62	49
T ₀	100	100	100	100	100	100	100	100	100	100
H ₃ T ₁	72	83	81	39	88	89	123	88	107	92
T ₂	45	50	79	36	64	103	218	69	109	78
T ₃	23	19	48	21	22	71	285	29	35	26

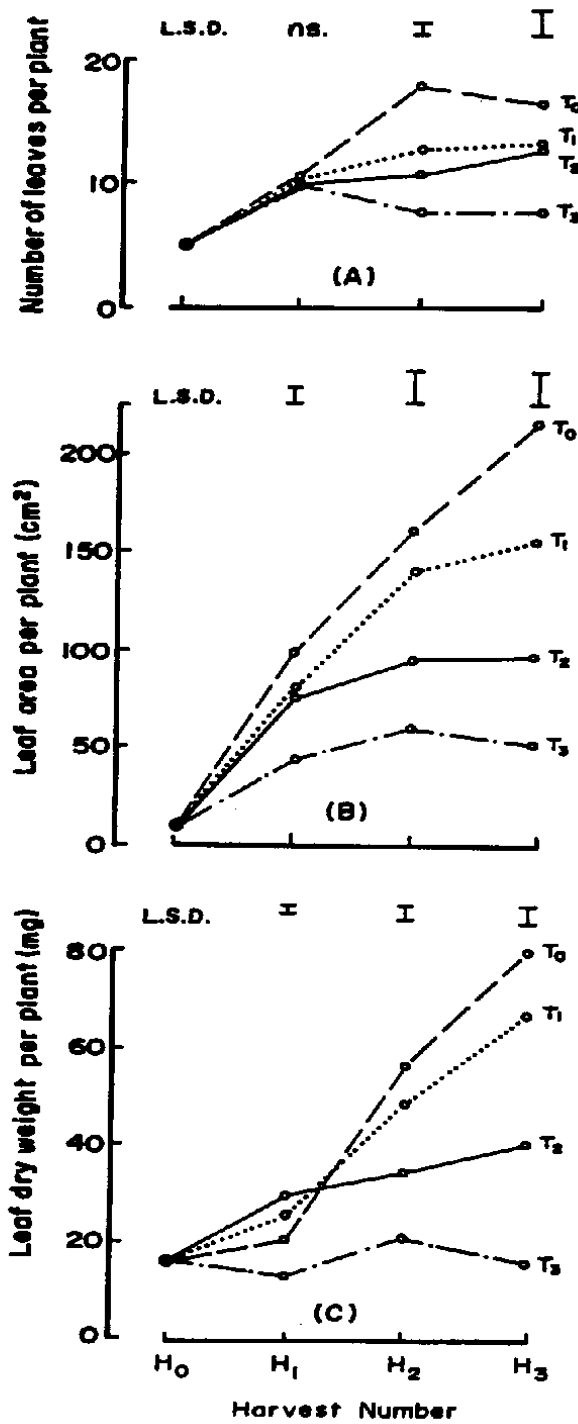


Fig. 1. Growth response of *Vigna ambacensis* seedlings cultured under four soil moisture regimes. A number of leaves per plant; B. leaf are per plant; C. leaf dry weight per plant; (o—o T₀; 0...0 T₁; o-o T₂; 0 0 T₃). Anova was performed separately. Vertical bars represent LSD values at 0.05 level. Treatments T₀-T₃ refer to water applications (ml per 2 days); T₀, 230; T₁, 160; T₂, 110 and T₃, 46.

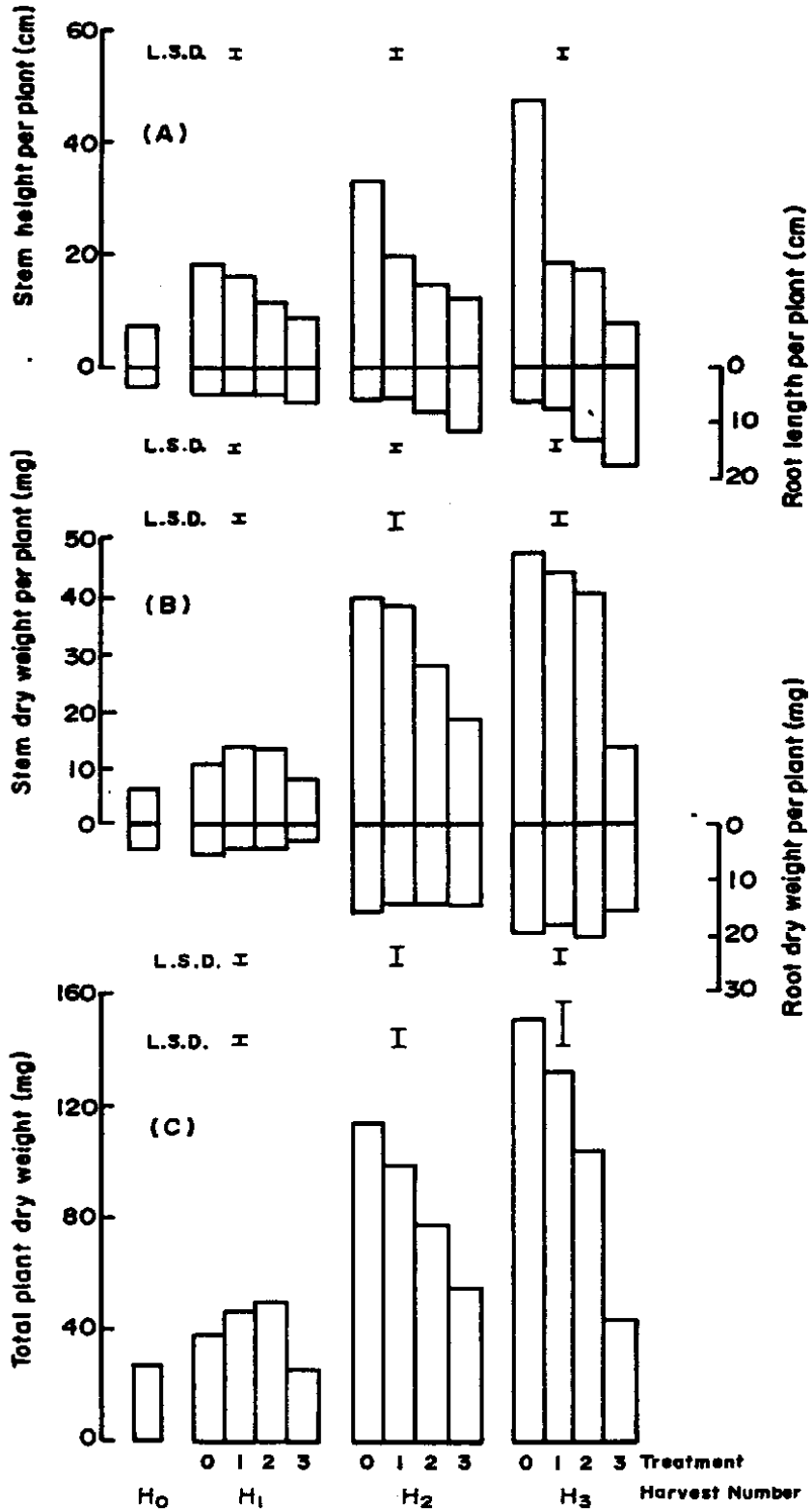


Fig. 2. Growth response of *Vigna ambacensis* seedlings to four soil moisture regimes. A Stem height and root length. B. Stem and root dry weight per plant. C. Total dry weight per plant. Vertical bars represent LSD values at P = 0.05 level. Treatments and statistical analysis are as in Fig. 1.

As the water stress developed over an extended period of four and six weeks (H_2 and H_3), the droughted plants showed a gradual decrease in most of the measures of growth except for the roots (Figs. 1 and 2). Progressive increase in root length was observed at H_2 and H_3 , but soil water stress produced smaller and fewer green leaves (Fig. 1 (A,B)) and shorter stem height (Fig. 2A). Leaf area at H_2 and H_3 in T_3 was drastically reduced by about 63 to 77% respectively compared to the wettest treatment T_0 . The average response of leaf number indicated that there was a significant decline at H_3 with increase in soil moisture stress from 100% at T_0 to 81, 79 and 48% at T_1 , T_2 and T_3 respectively (Table 2). Differences in leaf dry weight (LDW) also occurred between treatments; percentage values of LDW at H_3 were lowest at T_3 being only 19%, 50% for T_2 , and 83% for T_1 compared to T_0 (Table 2); at T_3 values for LDW at all the harvests also indicated a significant effect of droughting (Fig. 1C).

Stem height was significantly decreased in seedlings treated at T_1 , T_2 and T_3 compared with those at T_0 . The values of stem height during the entire drought treatments (42 days) are presented in Figure 2A and Table 2 (84, 69, 49% for H_1 , 59, 43, 38% for H_2 and 39, 36 and 21% for H_3 respectively). Stem dry weight (SDW) was lower in most of the drought treatments compared to the control. However, the value at harvest H_1 was not correlated with the drought treatment, being 25% lower at T_3 but 20% greater in the other two treatments relative to T_0 . Larger and significant differences were found at the later harvesting times H_2 and H_3 ; at H_3 stem dry weight at T_3 was only 27% of the value at T_0 .

Water stress also resulted in a lower percentage of total shoot dry weight (SH.DW) and total plant dry weight in T_3 at H_1 by about 30%. The decrease of SH.DW was much greater at H_2 and H_3 (T_3 compared to T_0). This response of SH.DW to all the drought treatments for all harvests (H_1 , H_2 and H_3) is shown in Table 2. There was a significant drop in SH.DW values in H_2 and H_3 for T_2 and T_3 compared to T_0 ; a similar response to water stress was found for total plant dry weight (Fig. 2C).

Root responses to drought treatment were different from those of the shoot, in both dry weight and length; values are given in Fig. 2 (A & B) and Tables (1 & 2). No marked difference in root length was found at H_1 between treatments. However, root length at H_2 and H_3 increased significantly with severity of drought treatment (Fig. 2 (A) Drought at T_2 (125% at H_2 and 218% at H_3) had a substantial effect, being highest at T_3 (172% at H_2 and 285% at H_3). There was little effect of drought treatment on root dry weight at H_1 except that of T_3 plants in which there was a 45% decrease compared to T_0 . However, some differences were found between treatments at H_3 but values for root dry weight are not significantly different at H_2 (Figure 2B). Relative growth rate (RGR) increased at H_1 with increase of water stress from 100% at T_0 to 145%, 170% and 191% at T_1 , T_2 and T_3 respectively; but at H_2 , there was a significant drop in RGR values by

exposure to severe droughting. RGR values decreased at H₂ for the greatest drought treatment from 0.051 for T₀ to 0.025 mg mg⁻¹ day⁻¹ for T₃ indicating about 50% decrease; a similar response was also observed at H₃ (Table 1 and 2). A similar response was found for NAR. There were also significant effects of water stress on NAR value in T₃ for all harvests. At H₁ there was a significant increase in NAR values at T₁ but Table 1 shows a significant decrease in NAR for T₂ compared to T₀. The percentage values for all the harvests are presented in Table 2.

Drought treatment had a significant effect on dry matter allocation on shoot (leaves, stem) and root at H₃. The R:S ratio (Table 1) indicated that at H₂ less dry matter was allocated to shoot than to root at T₁ (0.14) than at the T₂ (0.24) drought treatment. A further increase in this ratio was observed at T₃ (0.39). This result indicates that the shoot was more adversely affected by drought than the root. The R:S ratio increased significantly with droughting at H₃ to a higher value at T₃ (0.53). SLA values at H₁ were affected by droughting showing lower values at T₃ compared to T₀ (Table 1), but no clear response was found at H₂. At H₃ SLA was significantly higher at T₃ than in the other treatments.

Discussion

Exposure of seedlings of *Vigna ambacensis* L. to water stress during the vegetative growth stage led to reduced leaf area and stem length but to greater root length during the droughting period compared to well-watered seedlings. This observation suggests that *V. ambacensis* is a sensitive species, as drought symptoms developed even at mild water stress in T₂. This response to drought can be considered as a major adaptive mechanism avoiding water stress.

Similar results were reported for other legume species by Turk and Hall [21-22]; furthermore they reported the ability of these plants to maintain relatively high water content under drought conditions. The present results showed that moderate to severe water stress inhibits plant growth, resulting in smaller total dry weight, number of leaves, leaf area and stem length. These results agree with earlier reports [22-23]. Wien, *et al.* [24], using field-grown cowpea, reported a large reduction in leaf area and number of leaves, but shoot dry weight was only slightly affected by moderate water stress. Reduction in leaf area by severe water stress can be considered as an adaptive mechanism which helps to reduce water loss from the plant [22]. Smaller leaf area may be ascribed to acceleration of leaf senescence and abscission [25] or to the sensitivity of leaf expansion to water stress [26-28]. Whiteman and Wilson [28] reported that severe non-lethal water stress during the early vegetative growth stage of sorghum delayed rate of leaf appearance and reduced area of individual leaves. The observations recorded here at T₃ are in agreement with their findings.

Drought adversely affects total dry weight, causing a reduction which varies in different plant species. Pandey *et al.* [11] reported a reduction in total shoot weight in four legume species which varied between 78% in mungbean, 52% in soybean, 60% in cowpea and 37% in peanut compared with unstressed plants. Figure 2 clearly shows that water stress during the early stage of growth brings about a large reduction in dry weight, as found by Pandey, *et al.* [11].

The effect of increasing water stress on the root is an increase in root length and increased R:S ratio, which can be considered to represent an adaptive mechanism. Plants subjected to limited water supply show high R:S [6]. Comparison of total root length in the T₃ treatment compared to T₀ at H₃ (Table 2) indicated a nearly three-fold increase in root length, the balance between shoot and root development being greatly dependent on water stress as reported by other workers [6; 22]. Root length and/or density differences among legume crop species may be associated with their drought adaptation. Begg and Turner [27] pointed out that water stress usually leads to a greater root:shoot ratio. However, soil water stress also leads to increase in 'absolute' root length found in cotton by Malik, *et al.* [12] and in maize by Sharp and Davies [13]. These authors also suggested that under extreme low root frequency, the plant fails to extract the water available in the lower half of the root zone, limiting water uptake, causing water stress. Increase in root:shoot ratio is usually accompanied by a decrease in RGR [29;10]. The extension of roots in a deeper soil layer is a function of both genotype and environment [3]. It is suggested here that the adverse effect on growth at the vegetative stage may be due to the combination of two factors: reduction in water available in the soil owing to high osmotic potential and slow root growth in exploiting a greater depth of soil. This investigation has shown the interplay of morphological adjustment that enables *Vigna ambacensis* to grow under drought conditions, by reducing leaf area, lowering transpiration losses and increasing root length which helps to balance the demand for water in maintaining its uptake.

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أثر الجفاف على النمو وتوزيع المادة الجافة لبادرات اللوبيا البرية

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ملخص البحث. تمت دراسة أثر الجفاف على النمو وتوزيع المادة الجافة على البادات لنبات اللوبيا البرية وذلك للتحقق من وجود آليات لتجنب الإجهاد الجفافي عن طريق زراعتها داخل الصوبات الزجاجية في تربة مختلطة وتعريفها إلى مستويات مختلفة من الجفاف. أوضحت الدراسة أن هنالك تغيرات في المعايير المستخدمة لقياس النمو حيث أدى تعريض النباتات للجفاف لمدة ستة أسابيع (الحصاد الثابت للمعاملة الثالثة) إلى نقص في مساحة (٢٣٪)، أعداد الأوراق (٤٨٪) وأوزانها الجافة (٢١٪) كما انخفض الوزن الجاف الكلي للنبات إلى ٢٢٪، وإلى زيادة في كل من الوزن النسبي للأوراق (١٢٦٪) وفي أطوال الجذور (٢٨٥٪) ونسبة الوزن الجاف للجذور إلى السيقان (٥٧, ٠). عملت هذه الاستجابات جزئياً على مساعدة النبات للحد من آثار الجفاف.