

COMPARATIVE STUDIES ON GROWTH, NODULATION AND NITROGEN
FIXATION OF TWO LEGUMINOUS PLANTS GROWING
UNDER DIFFERENT LEVELS OF WATER SUPPLY
AND IRRIGATION INTERVALS

By

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Abstract:

A greenhouse study was conducted at Heliopolis using loamy soil characterized by low water capacity retention to compare between growth, nodulation and nitrogen fixation of (*Lupinus termis* cv. Giza 4) and chickpea (*Cicer arietinum* cv. Giza 2) when subjected to different levels of water supply and irrigation intervals.

Four water amounts; 400, 300, 200 and 100 cm³ and four water intervals, 2-day, 4-day, 6-day and 8-day intervals were applied for a period of five weeks in a factorial randomized block design with three replications.

Growth of the two plants were greatly affected by deficiency in water supply when they were grown either at low water amount (100 cm³) or increased irrigation intervals.

Lupin gave best growth (dry matter) at water amount (300 cm³) and 2-day irrigation interval, while in case of chickpea growth was excellent at 300 cm³ and irrigated every 4-day interval. On the other hand, nodulation and nitrogen fixation were optimum for the two investigated plants irrigated every 2-day. Lupin shoot looked thick and short, with few nodes at low water amount (100 cm³) and increased irrigation interval (4-day).

Furthermore, chickpea responded to water regime by formation of new lateral branches and long tap roots at water amount (100 cm³) and irrigation interval of 4-day which manifested in rich dry matter accumulation at the same level of water amount.

Generally application of 100 cm³ water at any irrigation interval resulted in very poor crop performance with respect to Lupin but not for chickpea.

Introduction:

Water is a very important factor affecting crop production in arid and semi arid regions (Labanauskas *et al.*, 1981), it affects non legumes (Thakur and Ria, 1984; El-Zeiny and Kortam, 1985 and Batanouny *et al.*, (1991), as well as several legumes; (Morandi *et al.*, 1981 and Egli *et al.*, 1983). Not only legumes growth but also legumes varieties, (Babalola, 1980 and Abou Bakr *et al.*, 1993) and nodulation and nitrogen fixation (Finn and Brun, 1980; Dejong and Phillips, 1982; Mbagwa and Osuigwe, 1984 and Sprent and Sprent, 1990).

Some data indicate that water stress disrupts interactions between Rhizobium and host plant directly by altering nodule fine structure or enzyme activity (Sprent, 1976), other evidence suggested that it affects photosynthesis in plants (Finn and Brun, 1980) and root system in non legumes (Batanouny *et al.*, 1991). It decreases nitrate reductase activity (Srivastava, 1980) and negatively affects various other aspects of N₂-metabolism (Hsiao, 1973).

Water stress decreases, plant heights and causes stem dwarfism (Taylor *et al.*, 1982; Hutchnison *et al.*, 1986; Kandil *et al.*, 1988 and Abou Bakr *et al.*, 1993).

Because Lupin and chickpea are very important crops in the developing countries and in semiarid regions where water is very important factor affecting plant growth, the present study was performed to through light upon growth behaviour, nodulation and nitrogen fixation of these two legumes which grow in many areas have deficient water supply to maximize their growth under sever water conditions, also to show the optimum amount of water used together with irrigation intervals so as to avoid wastage of water in those semiarid regions.

Materials and Methods:

Seeds of Lupin (Luninus termis cv. Giza 4) and Chickpea (Cicer arietinum cv. Giza 2) were obtained from Agriculture Research Centre Ministry of Agriculture, Giza.

Seeds of each plant were inoculated separately with its specific Rhizobium: Rhizobium lupini and Rhizobium leguminosarum (local isolates) for Lupinus termis and Cicer arietinum respectively. Rhizobia were obtained from Microbial Culture Collection Centre (MIRCEN), the unit of Bio-fertilizer, Faculty of Agriculture, Ain Shams University, Shoubra, Egypt. The experiment was performed in the experimental garden of the Botany Department, Women's College, Ain Shams University. The soil was air dried, passed through a 2 mm sieve and adjusted at pH 7.2. Clay pots (30 cm diameter and 25-cm depth) were chosen for cultivation. In each pot, 4 Kg of clay loam soil were placed. Mechanical soil analysis were carried out following the method described by Jackson (1973), and given in Table (1). The soil have a 21% moisture content at field capacity, 1.04% organic matter, 1.40% total nitrogen and 0.6 ds/m electrical conductivity. Each pot was fertilized with 60 mg/Kg⁻¹ N₂, 15 mg/Kg⁻¹ p and 60 mg/Kg⁻¹ K. Ten seeds of each plant Lupinus termis and Cicer arietinum were sown per pot. Two weeks after germination, plants were thinned to five plants per pot, then subjected to the treatments which consist of four water amounts (400, 300, 200 and 100 cm³), and four irrigation intervals (2-day, 4-day, 6-day and 8-day), keeping three replication for each treatment.

Plants were uprooted weekly, washed and dried at 70°C to constant weight, growth parameters including root and shoot height and dry weight were made. Plants were ground and used for determination of nitrogen according to the method described by Jackson (1973). Nitrogen determination was determined by kjeldahl method. All the nodules per pot were carefully counted, dried and weighted. The obtained data were subjected to analysis of variance (Snedecor and Cochran, 1980). Because data were more obvious at 2-and 4-day intervals than 6-and 8-day intervals, comparative studies of the 2-tested plants were made at 4-day interval.

Results and Discussions:

Shoot length:

The obtained data (Fig. 1&2) show that the various growth criteria measured in the present study was affected by extending the period between the successive irrigation intervals as well as the amount of water used

The average shoot length of Lupinus termis after five weeks from the initiation of treatments was 9, 14.5, 14 and 10 cm in plants receiving water amount of 100, 200, 300 and 400^{cm³} respectively, and irrigated every 4-day interval, while it was 19, 19.5, 18 and 17.5 cm in shoot height of Cicer arietinum irrigated at the same level of field capacity and irrigation interval. These results showed that Lupinus termis shoot, growth was short, thick and had few nodes but the shoot growth of Cicer arietinum was long and had normal shape at the same level of water amount and irrigation interval. Kramer (1969) pointed out that water deficit reduces enlargement and stem elongation. This observation was confirmed by Levitt (1980) who stated that decrease in cell enlargement must be the most sensitive response of the plant to water stress. Similar results were, however, obtained by other investigators (Morandi *et al.*, 1983) and Abou Bakr *et al.*, 1993). They also found that soil moisture deficit retarded plant growth, resulting in short stems and few nodes. Furthermore, our results on Lupinus termis were in agreement with the finding of the following workers (Hutchinson *et al.*, 1986 and Kandil *et al.*, 1988) and were in disagreement with our results on Cicer arietinum. The above mentioned investigators noticed that the increase in water stress by elongation of irrigation intervals resulted in stem dwarfism of many legumes.

Root length:

Data obtained (Fig. 1&2) on root length revealed that root system has been greatly affected by irrigation regime, thus root length of Cicer arietinum after five weeks from the initiation of treatment was 14, 12, 11.5, 10 cm in plant irrigated with 100, 200, 300 and 400^{cm³} respectively, and every 4-day interval (long period interval) comparing with Lupinus termis root length which was 6, 6.5, 6.3 and 6.3^{cm} irrigated at the same level of water amount and days intervals. It is evident that Cicer arietinum root length was much longer than Lupinus termis root length under the same level of water supply. Batanouny *et al.*, (1991) found that there is a close relationship between water supply and root system characters in Zea mays plant. They mentioned that decrease in water supply resulted in increase in the depth of penetration, increase in

lateral extension and increase in fresh and dry weight. Those authors attributed such increment to mechanisms by which plants increase the absorbing surface and exploiting large volume of soil surface.

Dry matter Content

It is clear from Fig. (3&4) that the dry matter of the whole plant exhibited considerable variation with depletion of water supply and irrigation intervals either with Lupinus termis or Cicer arietinum, but in general the first plant has increased amounts of dry matter accumulation than the second, one this is due not to the treatment itself but to the nature of plant growth and leaf area. On the other hand at the end of the experiment (5 weeks), Cicer arietinum showed the highest dry matter whereas Lupinus termis demonstrated the lowest dry weight at all irrigation ^{treatments} and at period intervals (4-day).

Cicer arietinum responded to water stress as mentioned previously by increase in lateral branches and increase in root length which intern enhance dry matter accumulation.

If we compare between the dry matter of the two investigated plants at the end of the experiment, it was found that Lupinus termis dry matter was best. with the application of . 300 cm³ applied every 3-day, whereas best Cicer arietinum dry matter was obtained by application of . 300 cm³ applied every 4-day interval. These data revealed that Cicer arietinum could sustain water stress condition much better than Lupinus termis. This might be due to many factors such as increase in root depth and increase in lateral branch number which reflect increase in dry matter accumulation. Batanouny et al., (1991), found results similarly on Zea mays plant. They claimed that decrease in water supply, increase fresh and dry weights of root system. On the other hand, many authors attributed decreased yield in case of Lupinus termis associated with reduced amount of water to water stress condition which invariably resulted in stomatal closer. This has the overall effect of reducing the photosynthetic efficiency of the leaves with consequent reduction in yield.

Reduction in dry matter accumulation as a result of water deficiency was pronounced by many workers on soybean (Scott, 1984 and Kandil, 1988) and cowpea (Mbagwa and Osuigwe, 1984) as well as on Medicago sativa and its variety (Abou Bakr *et al.*, (1993) in addition to Zea mays (Thakur and Ria, 1984; El-Zeiny and Karam, 1985).

Nodulation and nitrogen fixation:

It is clear from Tables(2&3) that water depletion affects formation of nodules on Lupinus termis and Cicer arietinum roots. Optimum nodulation was obtained by the application of 400 cm³ water and irrigated every day for the two tested plants, but the number of nodule or their weights were varied according to the nature of the plant itself. Water stress affects root growth and the magnitude was differed, being increased in Cicer arietinum and decreased in Lupinus termis.

Lupinus termis root looked short and thick, while Cicer arietinum root was long and penetrated deeper at 100 cm³ and 4-irrigation intervals. All nodules were aggregated on the main root in case of Lupinus termis but in Cicer arietinum were on both main root and lateral roots, moreover, their number were more or less identical under the same treatment of water amount and irrigation intervals. Our results were disagreement, however, with the results obtained by Mbagwa and Osuigwe (1984) on cowpea nodulation and water deficiency. They reported that good nodulation was obtained by the application of 100% field capacity and irrigated every day while Habbish and Mahdi (1976) observed poor nodulation when watering bean and cowpea at 15% soil moisture, these results are in accordance with our results. We found bad nodule formation at 100 cm³ at all irrigation intervals either in Lupinus termis or Cicer arietinum. Moreover, Karama (1976) reported that watering at field capacity or excess increased cowpea nodulation, although excess water than field capacity interferes with aeration (Abou Bakr *et al.*, (1993).

Nitrogen fixed by two investigated plant were greatly affected by the treatments as well as by the nature of the plants themselves. Thus optimum nitrogen content was obtained by the application of 400 cm³ water and irrigated every day for the two

tested plants. This holds true with the number of nodules. Nitrogen decreased by the decrease of water amount used (100 cm^3) and also by increase of irrigation interval (4-day).

Regrading Lupinus termis nitrogen content at the fifth week it was 4.82% at 400 cm^3 and 2-day interval, while Cicer arietinum nitrogen content at the same level of water amount and irrigation interval was 3.32% . On the other hand nitrogen content of Lupinus termis at the same week but at 100 cm^3 and 8-day of irrigation interval was 2.56% . Furthermore in case of Cicer arietinum, nitrogen content at the same levels of field capacity and irrigation interval was 1.75% .

From the above mentioned data, it was found that, low water amount (100 cm^3) and long irrigation interval (8-day) drastically affected nitrogen content of either Lupinus termis or Cicer arietinum.

Our results were in agreement with many other authors (water stress decreased symbiotic nitrogen fixation and growth of nodulated legumes) Dejong and Phillips, 1982; Finn and Brun, 1980 and Sprent and Sprent, 1990). Some others found that it disrupts fine structure of nodule membrane permeability and enzyme activity (Sprent, 1976).

Water stress is also known to decrease nitrate reductase activity (Srivastava, 1980) and negatively affects other aspects of N_2 -metabolism (Hsiao, 1973).

Conclusion:

Cicer arietinum could sustain draught much better than Lupinus termis when grown in semiarid regions with depletion in water supply and elongation of irrigation interval. It is also inferred that Cicer arietinum could be a popular choice for inter cropping because its drought susceptibility is intermediate, the closer of its stomates under stress allows it to conserve moisture for the main crop and its deeper rooting habit is also an advantage under stress conditions in semiarid regions.

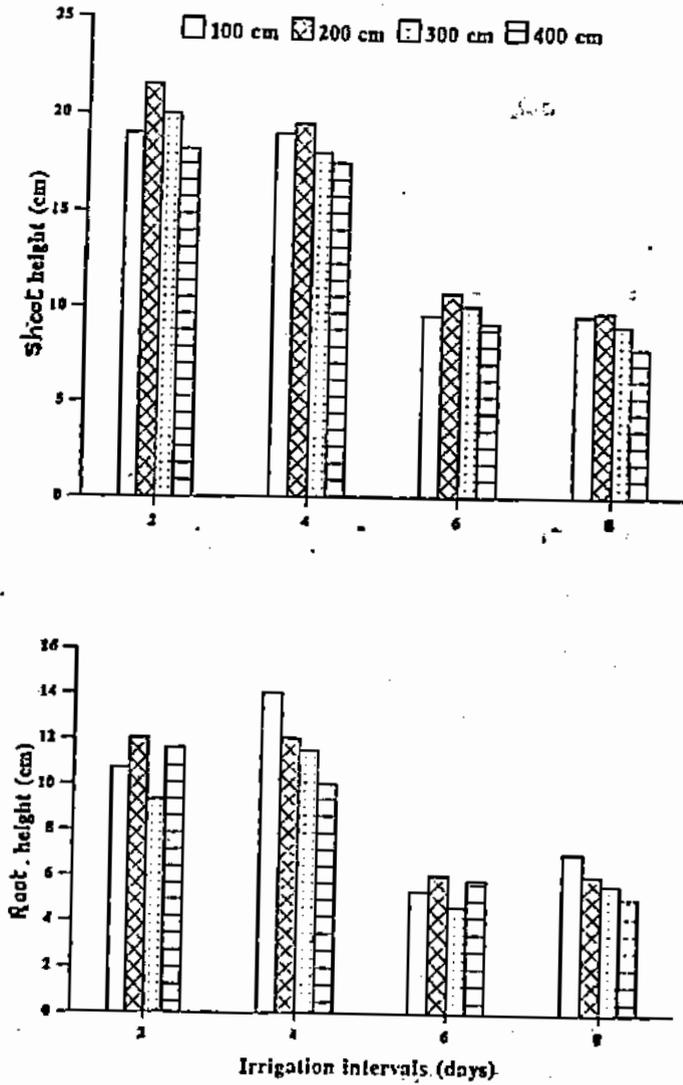


Fig. (1): Effect of irrigation intervals (days) and water amount (cm³) on plant height (cm) of *Cicer arietinum* at 5th week

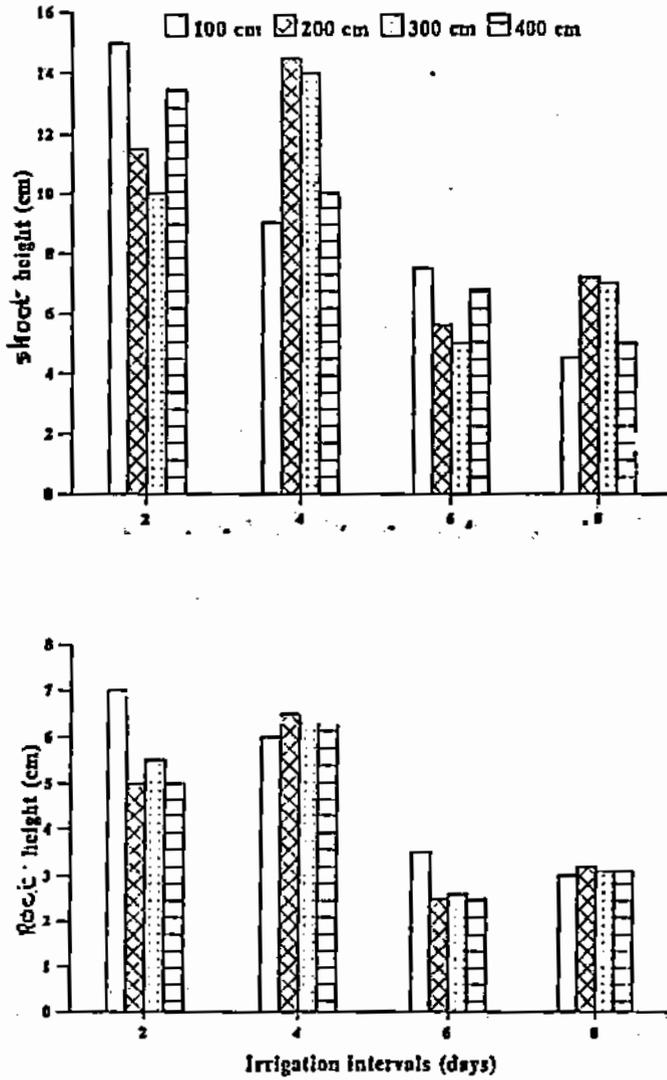


Fig. (2): Effect of irrigation intervals (days) and water amount (cm) on plant height (cm) of Lupinus termis at 5th week

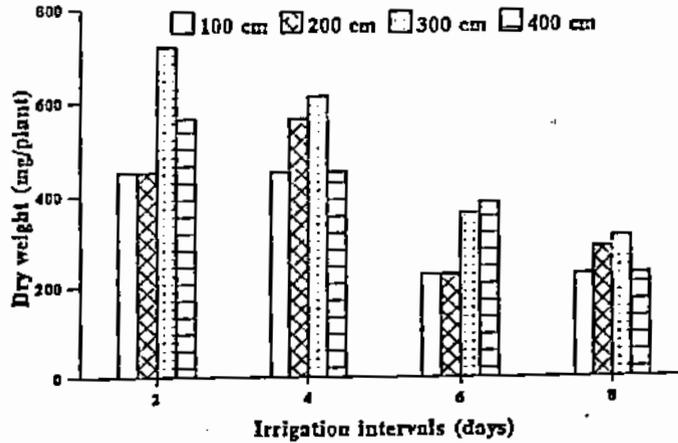


Fig. (3): Effect of irrigation intervals (days) and water amount (cm³) on dry weight (mg/plant) of Lupinus termis at 5th week

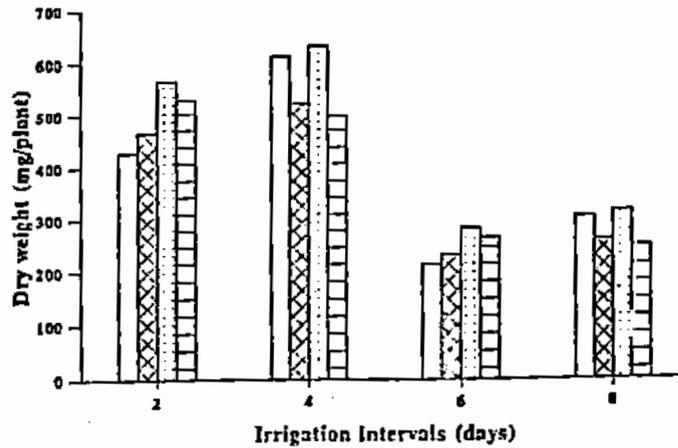


Fig. (4): Effect of irrigation intervals (days) and water amount (cm³) on dry weight (mg/plant) of Cicer arietinum at 5th week

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Table (1)

Mechanical analysis of the soil used in the
experiment

Total sand	37.2%
Silt	30.6%
Clay	32.2%
Soil texture	clay loam

Table (2) : Effect of irrigation intervals and water amounts on nodulation and nitrogen content of Lupinus termis (at 5 th. week).

Irrigation intervals (days)	3 Applied water amount (cm)											
	400			300			200			100		
	Nt	Nodule no./pot	Nodule wt. mg./pot	Nt	Nodule no./pot	Nodule wt. mg./pot	Nt	Nodule no./pot	Nodule wt. mg./pot	Nt	Nodule no./pot	Nodule wt. mg./pot
2	4.82	70	0.12	4.54	30	0.03	4.20	18	0.02	3.77	10	0.001
4	3.90	35	0.08	4.67	24	0.02	3.20	15	0.02	3.67	6	0.001
6	4.50	30	0.02	4.12	19	0.01	3.11	8	0.01	2.87	4	0.001
8	4.50	16	0.01	3.84	13	0.01	2.92	5	0.01	2.81	2	0.001
Means	4.43	38	0.03	4.29	22	0.02	3.36	12	0.015	3.05	5	0.001

* L.S.D for nitrogen content at 0.05 = 0.6274

* L.S.D. for nodule no./pot at 0.05 = 14.5500

* L.S.D for nodule wt. (mg./pot) at 0.05 = 0.0283

Table (3) . Effect of irrigation frequency and water amounts on nodulation and nitrogen content of Cicer arietinum (at 5 fl. week).

Irrigation intervals (days)	3 Applied water amount (cc.)											
	400			300			200			100		
	N2	Module no./pot	Module wt. mg./pot	N2	Module no./pot	Module wt. mg./pot	N2	Module no./pot	Module wt. mg./pot	N2	Module no./pot	Module wt. mg./pot
2	3.32	50	0.10	2.37	28	0.03	3.17	20	0.010	2.43	8	0.002
4	3.26	30	0.02	2.31	28	0.02	3.00	17	0.005	2.00	5	0.002
6	3.20	25	0.03	2.17	17	0.01	3.30	14	0.005	2.10	4	0.001
8	3.01	20	0.02	2.00	12	0.01	2.19	8	0.002	1.75	2	0.001
MEANS	3.22	34	0.04	2.12	21	0.02	2.80	15	0.005	2.00	5	0.002

C.L.S.D for nitrogen content at 0.05 = 0.31932

C.L.S.D for module (no./pot) at 0.05 = 11.9509

C.L.S.D for module wt. (mg./pot) at 0.05 = 0.0316