# Drought and Phosphorus Affect Growth of Annual Forage Legumes[1]

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## Highlight

**Three annual forage legumes, apparently differing in their drought resistance in the field, were grown in controlled environments to better understand mechanisms of their drought resistance and to determine relationships between phosphorus nutrition and drought. Phosphorus fertilization stimulated growth of the annual legumes and decreased water use (ml/g dry weight of top growth). Relative top growth and phosphorus uptake of Spanish clover tended to confirm observations of its drought resistance in the field. Water use was higher in Spanish clover than in subterranean clover and therefore does not appear to contribute to its drought resistance. This study provides information that will be helpful in future research on the morphological and physiological traits that contribute to drought resistance in these and other range plants.**

Annual legumes apparently differ in their adaptation to arid conditions. Mt. Barker subterranean clover (*Trifolium subterraneum* L.) is well adapted to the moist coastal range of California. Rose clover (*Trifolium hirtum* All.) is better adapted and often more productive than subterranean clover in the semi-arid central valley of California. Subterranean and rose clover usually complete their life cycle before summer drought and high temperatures, but Spanish clover (*Lotus purshianus* [Benth.] Clements & Clements) often continues vegetative and reproductive growth during the dry summer months.

A study was established under controlled conditions to better understand drought resistance in these annual legumes and to determine the role that phosphorus might play in their response to soil moisture stress.

Drought resistance in plants includes 2 components—avoidance and tolerance (Levitt, 1964). Drought avoiding plants escape drought by means of deep root systems, water storage, or low transpiration. Drought tolerant plants lose water from their tissues in dry environments and yet are able to (1) survive, or (2) continue growth, development, and metabolism.

Since in this study moisture stress in plant tissues was not measured, the more general term *drought resistance* is used. Nevertheless, it is realized that annual legumes, growing in pots in small volumes of soil, have little opportunity for drought avoidance.

This study concerns the ability of annual legumes to continue growth in dry environments rather than their ability to survive in dry environments.

## Materials and Methods

A dilute solution of $H_3PO_4$, in amounts equivalent to 0, 44, or 175 lb P/acre, was thoroughly mixed with 550 g of Auburn silty clay loam, a phosphorus deficient soil (McKell et al., 1962). Six germinated seeds were planted in the soil and were allowed to grow through small holes in the container lid. Cotton was placed in the hole around each plant to minimize evaporation from the soil. During a 25-day period for seedling establishment, plants were grown in a greenhouse at a soil moisture stress below 1 atmosphere. Plants were then transferred to a controlled environment with a light intensity of 1000 ft-c, day length of 16 hr, temperature of 21 C, and relative humidity of 60 to 80% during the day and 80 to 95% during the night. At the same time, soil moisture regimes of 1, 3, and 15 atmospheres were established. A calibration curve relating soil moisture percentage to soil moisture stress was prepared by the pressure membrane method (Richards, 1947). The containers in which plants were growing were weighed twice daily and the soil moisture percentage was calculated by using the total weight and the known weights of the soil and container. When soil moisture decreased to the percentage corresponding to 1, 3, or 15 atm, water was added to increase soil moisture to the percentage corresponding to 0.3 atm (approximately field capacity). The amount of water added at each irrigation was recorded. After 6 weeks of growth under the 3 moisture regimes, top growth was harvested and roots were washed from the soil.

The experiment was conducted with 4 replications. Statistical comparisons were made by using the Duncan multiple range test. Plant material from the 4 replications was pooled for the determination of total phosphorus in tops and roots.

## Results and Discussion

*Soil Moisture Regimes.*—The maximum moisture stress reached before irrigation and the duration of moisture stress levels which restrict growth

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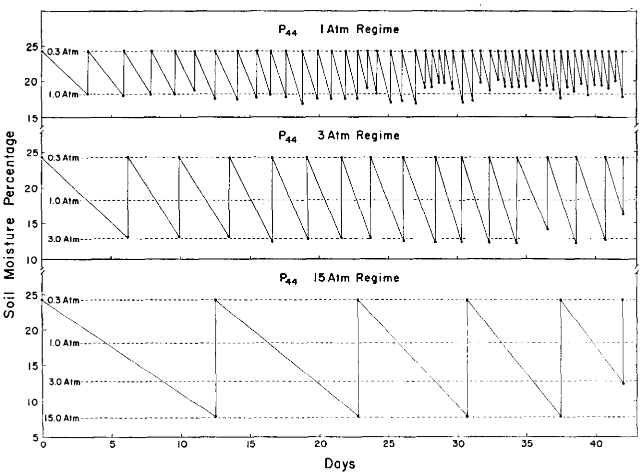


FIG. 1. Drying cycles of plants fertilized with 44 lb P/acre and held in the 1, 3, and 15 atmosphere soil moisture regimes, including all 3 annual legumes. Points on lower side of curves represent average dates of irrigation and average soil moisture percentages prior to irrigation. Assumption of a linear loss of soil moisture with time was made in plotting the data. Horizontal broken lines indicate soil moisture percentages corresponding to a soil moisture stress of 0.3, 1, 3, and 15 atmospheres.

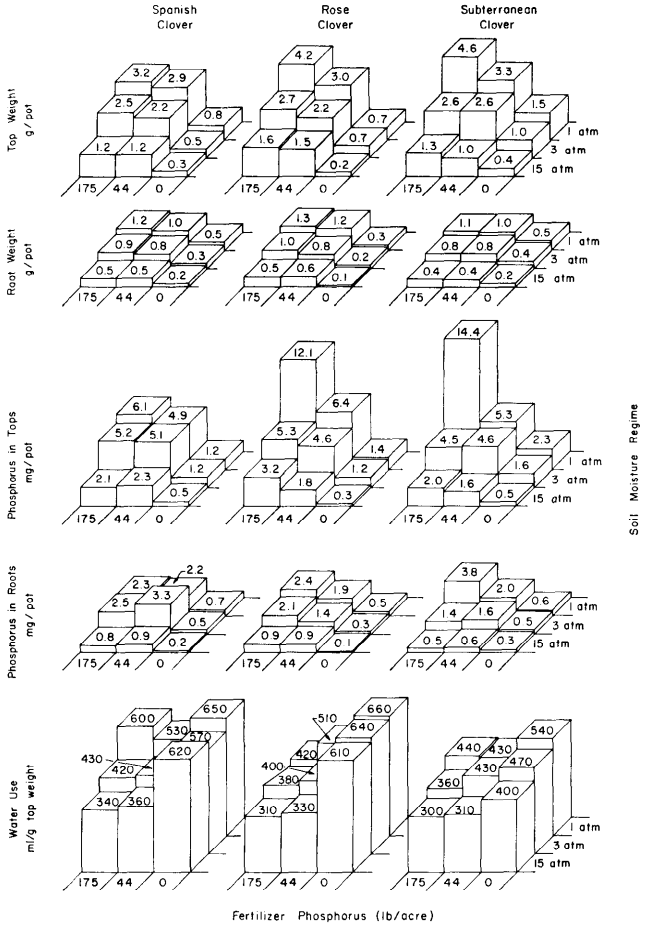


FIG. 2. Top and root growth, phosphorus uptake, and water use of annual legumes in response to soil moisture regimes and phosphorus fertilization.

need to be considered in the interpretation of plant growth in response to a series of drying cycles. Fig. 1 gives the drying cycles of plants fertilized with 44 lb P/acre and held in the 1, 3, or 15 atm regimes. The maximum soil moisture stress to which plants were subjected was approximately constant within each moisture regime. However, because of increased top growth and transpiration, the duration of a single drying cycle decreased as the experiment progressed. When compared with plants in the 44 lb P/acre treatment, small plants in the 0 lb P/acre treatment required less frequent irrigation and large plants in the 175 lb P/acre treatment required more frequent irrigation (Table 1).

The performance of species growing in moisture regimes can be validly compared if the regimes and frequencies of irrigation are similar. In this study the similarity of moisture regimes of the 3 annual legumes, particularly in treatments receiving 44 and 175 lb P/acre (Table 1), permits meaningful comparisons of their responses to drought.

The detailed illustration of top and root growth, phosphorus uptake, and water use is given in Fig. 2. Statistical evaluations of relationships between species, moisture regimes, and phosphorus fertilization are presented in the tables.

*Plant Growth.*—Spanish clover was lowest and subterranean clover was highest in top growth production in the 1 atm regime (Table 2). In the 3 and 15 atm regimes, top growth of the 3 annual legumes did not differ significantly.

Comparisons of relative top growth of plants in the 1 and 3 atm regimes suggest that the growth of Spanish clover was inhibited less by moderate

**Table 1. Number of times annual legumes were irrigated during a 6-week exposure to 3 moisture regimes.**

| Moisture regime | Spanish clover | | | Rose clover | | | Subterranean clover | | |
|---|---|---|---|---|---|---|---|---|---|
| | $P_{175}$ | $P_{44}$ | $P_0$ | $P_{175}$ | $P_{44}$ | $P_0$ | $P_{175}$ | $P_{44}$ | $P_0$ |
| 1 atm | 54 | 48 | 18 | 54 | 49 | 16 | 56 | 48 | 24 |
| 3 atm | 18 | 16 | 5 | 17 | 15 | 6 | 15 | 16 | 8 |
| 15 atm | 5 | 4 | 2 | 6 | 6 | 2 | 5 | 4 | 2 |

**Table 2. Top growth (g/pot dry weight) of 3 annual legumes in response to 3 moisture regimes.**[1]

| Moisture regime | Spanish clover | Rose clover | Subterranean clover |
|---|---|---|---|
| 1 atm | 2.32[c] | 2.65[b] | 3.11[a] |
| 3 atm | 1.75[d] | 1.83[d] | 2.04[cd] |
| 15 atm | 0.89[e] | 0.89[e] | 1.11[e] |

[1] These data include all 3 phosphorus fertilization treatments. Numbers having the same letter in the superscript do not differ significantly at the 5% level.

**Table 3. Relative top growth (percent[1]) for 3 annual legumes as influenced by moisture regime.**

| Moisture regime | Spanish clover | | | Rose clover | | | Subterranean clover | | |
|---|---|---|---|---|---|---|---|---|---|
| | P175 | P44 | P0 | P175 | P44 | P0 | P175 | P44 | P0 |
| 1 atm | 100 | 91 | 25 | 100 | 71 | 17 | 100 | 72 | 33 |
| 3 atm | 78 | 69 | 16 | 64 | 52 | 17 | 57 | 57 | 22 |
| 15 atm | 38 | 38 | 9 | 38 | 36 | 5 | 28 | 22 | 9 |

[1] Values given are in relation to yields of plants fertilized with 175 lb P/acre and held in the 1 atm regime.

stress than the growth of rose and subterranean clover (Table 3).

In the 1 atm regime, the average top growth of the 3 annual legumes increased with each increase in phosphorus (Table 4). In the 3 and 15 atm regimes, the application of 175 lb P/acre did not produce greater top growth than 44 lb P/acre.

The pattern of root growth response to moisture stress for the 3 annual legumes was essentially similar. The application of 44 lb P/acre increased root growth in all 3 moisture regimes (Table 4). But the application of additional phosphorus did not result in a further increase in root growth.

*Phosphorus Uptake.*—Subterranean and rose clover apparently have a greater ability than Spanish clover to absorb phosphorus in the 1 atm regime, but do not maintain this superiority in the 3 atm regime (Table 5). Comparisons of relative phosphorus uptake by plants fertilized with 175 lb P/acre and held in the 1 or 3 atm regimes showed that moisture stress reduced phosphorus in Spanish, rose, and subterranean clover by 8, 49, and 68%, respectively.

*Water Use.*—Efficient water use by fertilized annual legumes (Table 6) suggests that phosphorus increased photosynthesis per unit leaf area more than transpiration per unit leaf area. In experiments where moisture was not a limiting factor, Watson (1947) showed that phosphorus deficiency reduced net assimilation rate in barley and mangolds during the period when leaf area was increasing.

**Table 4. Effects of phosphorus fertilization and moisture regime on top and root growth (g/pot dry weight) of annual legumes.**

| Moisture regime | Top growth[1] | | | Root growth[1] | | |
|---|---|---|---|---|---|---|
| | P175 | P44 | P0 | P175 | P44 | P0 |
| 1 atm | 3.97[a] | 3.09[b] | 1.01[e] | 1.19[a] | 1.10[a] | 0.43[cd] |
| 3 atm | 2.61[c] | 2.35[c] | 0.71[e] | 0.89[b] | 0.82[b] | 0.32[d] |
| 15 atm | 1.37[d] | 1.20[de] | 0.32[f] | 0.49[c] | 0.49[c] | 0.17[e] |

[1] Data include all 3 annual legumes. Values having the same letter in the superscript do not differ significantly at the 5% level. Separate statistical comparisons are made for top and root growth.

**Table 5. Phosphorus uptake on an absolute basis and on a relative basis by species.**

| Moisture regime | Spanish clover | | | Rose clover | | | Subterranean clover | | |
|---|---|---|---|---|---|---|---|---|---|
| | P175 | P44 | P0 | P175 | P44 | P0 | P175 | P44 | P0 |
| Total P uptake, mg/pot[1] | | | | | | | | | |
| 1 atm | 8.4 | 7.1 | 1.9 | 14.5 | 8.3 | 1.9 | 18.2 | 7.3 | 2.9 |
| 3 atm | 7.7 | 8.4 | 1.7 | 7.4 | 6.0 | 1.5 | 5.9 | 6.2 | 2.1 |
| Relative P uptake (%) | | | | | | | | | |
| 1 atm | 100 | 85 | 23 | 100 | 57 | 13 | 100 | 40 | 16 |
| 3 atm | 92 | 100 | 20 | 51 | 41 | 10 | 32 | 34 | 12 |

[1] Total P in roots and tops.

More efficient water use in the 3 and 15 atm regimes than in the 1 atm regime suggests that during moderate stress, transpiration is reduced more than photosynthesis. This more efficient use in dry regimes might not occur in field conditions because evaporation from the soil contributes to total evapotranspiration.

The ability of Spanish clover to continue growth during dry summer months is not explained by more efficient water use (Table 6). Spanish clover apparently possesses some other means of avoiding drought.

*Drought Resistance.*—The stimulation of growth by phosphorus fertilization in the drier regimes does not necessarily mean that phosphorus enhances the drought resistance of annual legumes. Supplying phosphorus certainly results in more rapid root and top growth during the portion of the drying cycle when moisture is favorable. Also, rapid recovery of plants from the effects of moisture stress may be assisted by phosphorus fertilization. But it is uncertain whether plants having adequate phosphorus would continue growth under drier conditions than phosphorus deficient plants.

Moisture stress has been shown to seriously disturb the metabolism of phosphorus-containing nucleic acids in tomato (Gates and Bonner, 1959), corn (West, 1962), and sugar beet (Shah and Loomis, 1965). Concentrations of phosphate esters

**Table 6. Effects of main treatments on water use (ml/g dry weight of top growth).[1]**

| Phosphorus treatment[2] | | | Moisture regime[3] | | | Species[4] | | |
|---|---|---|---|---|---|---|---|---|
| P175 | P44 | P0 | 1 atm | 3 atm | 15 atm | Spanish clover | Rose clover | Sub. clover |
| 396[a] | 416[a] | 573[b] | 531[a] | 455[b] | 398[c] | 501[a] | 473[a] | 411[b] |

[1] Statistical comparisons are made within each main treatment. Values having the same letter in the superscript do not differ significantly at the 5% level.
[2] Includes all species and moisture regimes.
[3] Includes all species and phosphorus treatments.
[4] Includes all moisture regimes and phosphorus treatments.

drop to low levels in wilted subterranean clover plants (Wilson and Huffaker, 1964). Research is needed to learn whether phosphorus deficiency intensifies these injurious effects of drought.

In studies of drought resistance, comparisons of performance on a relative basis may be more meaningful than on an absolute basis. In this way the performance of a species under drought is compared with its own performance under favorable moisture. Top growth (Table 3) and phosphorus uptake (Table 5), on a relative basis, suggest that Spanish clover possesses greater drought resistance than rose and subterranean clover. However, this evidence of drought resistance in Spanish clover must be considered as being preliminary. Additional work is needed to clearly establish its resistance and to discover the morphological and physiological traits that contribute to its resistance.

## LITERATURE CITED

GATES, C. T., AND J. BONNER. 1959. The response of the tomato plant to a brief period of water shortage. IV. Effects of water stress on the ribonucleic acid metabolism of tomato leaves. Plant Physiol. 34:49–55.

LEVITT, J. 1964. Drought. *In*: Forage plant physiology and soil range relationships. American Society of Agronomy, Madison.

MCKELL, C. M., A. M. WILSON, AND W. A. WILLIAMS. 1962. Effect of temperature on phosphorus utilization by native and introduced legumes. Agron. J. 54:109–113.

RICHARDS, L. A. 1947. Pressure-membrane apparatus, construction and use. Agr. Eng. 28:451–454, 460.

SHAH, C. B., AND R. S. LOOMIS. 1965. Ribonucleic acid and protein metabolism in sugar beet during drought. Physiol. Plantarum 18:240–254.

WATSON, D. J. 1947. Comparative physiological studies on the growth of field crops. II. The effect of varying nutrient supply on net assimilation rate and leaf area. Ann. Bot. N. S. 11:375–407.

WEST, S. H. 1962. Protein, nucleotide, and ribonucleic acid metabolism in corn during germination under moisture stress. Plant Physiol. 37:565–571.

WILSON, A. M., AND R. C. HUFFAKER. 1964. Effects of moisture stress on acid-soluble phosphorus compounds in *Trifolium subterraneum*. Plant Physiol. 39:555–560.