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Soil Acidity, Temperature, and Water Relationships of Four Clovers in Sierra Nevada Meadows

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Sites in meadows of the Sierra Nevada near Fresno, California, were studied to learn whether Bolander's (*Trifolium bolanderi* Gray.), long-stalked (*T. longipes* Nutt.), carpet (*T. monanthum* Gray.), and mountain (*T. wormskioldii* Lehm.) clovers occurred under the same soil acidity, temperature, and water conditions. In selected meadows, along transects within its community, 20 plants of a clover species were randomly selected. Due to a small population, only 10 plants of mountain clover were selected in one community. In soil next to each plant, soil temperature was measured at the 10-cm depth, and soil was subjectively classed as saturated, wet, moist, or dry. Plants were extracted for an independent study of their rhizobia relationships, and a sample of soil from the 5-cm to 15-cm depth was taken for laboratory analysis of pH. Across the sites examined, species distribution of clover appeared to be independent of soil pH. Carpet clover occurred in warmer soil than the other three species. Clover distribution was dependent on soil water category. Soil water relations among the four species, from wettest to driest, were mountain, Bolander's, long-stalked, and carpet clover.

Retrieval Terms: *Trifolium bolanderi*, *T. longipes*, *T. monanthum*, *T. wormskioldii*, soil properties, California

A plant's persistence in an area over time indicates success in responding to environmental factors that limit its growth and reproduction. Clovers (*Trifolium* spp.), as legumes, benefit from symbiotic association with nitrogen-fixing bacteria of the genus *Rhizobium*. Environmental factors may control the range and distribution of different clover species by acting on the plants, on their specific rhizobia, or on the interaction between the plants and the bacteria.

We are studying the relationships of four perennial clover species to meadow microenvironments, and the diversity and distributions of their indigenous rhizobia in the Sierra Nevada of California. Our overall objective is to determine whether management activities on our National Forests can reasonably be expected to affect clover population maintenance and expansion. This note addresses the questions of whether the four species occur under the same conditions of soil pH, temperature, and water.

The clover species and the elevations at which they are found are: *T. bolanderi* Gray. (Bolander's clover; 2,134 m), *T. longipes* Nutt. (long-stalked clover; below 2,744 m), *T. monanthum* Gray. (carpet clover; 1,524 to 3,354 m), and *T. wormskioldii* Lehm. (mountain clover; below 3,049 m).¹ Bolander's clover is of special interest because it is listed as a sensitive species by the Sierra National Forest and as a Federal Category 2 candidate species.² Such species are those for which information is insufficient to support their listing as threatened or endangered. Bolander's clover is restricted in distribution to meadows in Mariposa, Madera, and Fresno Counties, in Califor-

nia. The other three clover species are more widespread, occurring throughout the Sierra Nevada.

Several cultivated clovers and other legumes do well on soils with a pH of about 6.0, but some species do better under slightly more acidic or basic conditions.³ Soil pH may influence plant distributions more as a result of its indirect effect on other soil properties. For example, the availability of nitrogen and phosphorus to plants tends to be lower in acidic soils than in neutral soils, whereas aluminum, manganese, and iron concentrations in solution tend to increase as acidity increases.^{4,5} Rhizobia also vary in pH optima; tolerance of acidity in culture correlates with the ability to survive in acidic soils⁶ and nodulate the host plant under acidic conditions.^{7,8,9}

Plant species differ in temperatures at which various biological processes are most active, and temperature affects the activity of rhizobia strains.¹⁰ Our knowledge of soil temperature and plant relationships is not well developed. Growth of pasture herbage stops at temperatures below 5.5 °C at a depth of 10 cm in the soil.⁴ The maximum reported growth temperature for clover rhizobia is 39 °C, although some strains of alfalfa rhizobia grow at temperatures as high as 42.5 °C.¹¹

In meadows it is unusual for soil water availability to limit the survival and distribution of plant species. Nevertheless, plants may encounter periods of unusual water stress, as during the recent drought in California. Length of time that soil water is in excess of the field capacity may limit growth of plants or rhizobia by affecting soil aera-

tion. Microbiological activity is reduced in extremely wet or dry soils.¹²

The extent to which environmental factors can control clover distribution through effects on the symbiotic bacteria will depend in part on the level of specificity of the interaction between the plants and particular bacterial strains. Environmental factors can complicate the plant-microbe interaction. For example, pH¹³ and temperature¹⁴ can alter the outcome of competition between rhizobial strains for nodulation of the host plant.

METHODS

Field Sampling

Because of its sensitive status, sampling of Bolander's clover was restricted to five meadows, randomly selected from among those known to contain populations of that species. From July 16 to July 31, 1991 (during the flowering period), 20 plants of Bolander's clover were selected at each meadow.

Our intent was to sample each of the four clover species at those five meadows. Not all species occurred at all of the meadows, however. When a population of one or more of the other three clover species was found within one of the five meadows, 20 plants of those species were also selected. When this was not the case, plants of missing species were selected in nearby meadows known to contain them.

In all cases, plant selection was at random along transects positioned within the different clover communities present. Total transect lengths varied depending on size of the site and density of the species being sampled. Space between possible sample points along a transect was determined by dividing its total length in steps by 100. The plant of the clover species nearest a random point was the one selected. Only 10 mountain clover plants were selected in one case so as not to unduly disturb a relatively small population.

Adjacent to each plant, soil water was subjectively classed as: saturated (S)—water is at or on the surface or comes to the surface when stepped on; wet (W)—free water is near the surface or can be squeezed from the soil by hand; moist (M)—free water is not present, the soil feels cool and damp like a well-wrung face cloth, and clods do not easily crumble unless sandy;

or dry (D)—soil feels dry and clods crumble easily. Respectively, these water categories were similar to the very poorly and poorly drained, imperfectly or somewhat poorly drained, moderately well and well drained, and somewhat excessively drained classes of the U.S. Department of Agriculture.¹⁵

Soil temperature was measured at the 10-cm depth. The plant was then extracted, and a sample of soil from the 5-cm to 15-cm depth was placed in a soil can for laboratory analysis of pH. Soil pH was estimated by the saturated paste technique with a combination electrode.

Statistical Analysis

Because only one of the five meadows where Bolander's clover was collected contained all four species and because of the possibility of heterogeneous variances in the species, the Games and Howell test¹⁶ (the modified Tukey's test) with the overall alpha level at 0.05 was applied for paired comparisons of clover species. Values of pH were converted to relative acidity (arithmetically equivalent values of hydrogen ion normality), and soil water categories were assigned scores (Saturated = 1, Wet = 2, Moist = 3, and Dry = 4) to permit analysis. Also, percentages of all plants of each species were computed by soil water category.

RESULTS

Soil Acidity

Across the sites involved in this study, distribution of the four clover species appeared to be independent of relative acidity (table 1). Corresponding average pH values (all extremely acidic) and the extremes for each species were:

Clover species	Avg. pH	Extreme values	
		Low pH	High pH
Bolander's	4.26	3.4	5.4
Long-stalked	4.37	3.8	5.2
Carpet	4.29	3.7	6.6
Mountain	4.36	3.8	4.8

Soil Temperatures

On the average, a difference of only 1 °C or less in soil temperature was found between species (table 2). Nevertheless, across the sites involved in this study, the other species occurred in slightly cooler soil than carpet clover.

Soil Water

All four clover species preferred moist or more wet soils to dry soils (table 3). They differed from each other in their average soil water categories, however.

Mountain clover appeared to prefer saturated sites, whereas Bolander's clover appeared to prefer wet sites. Long-stalked clover and carpet clover both appeared to prefer moist sites. Even though the numbers of plants occurring on dry sites were few, carpet clover appeared best suited to grow there. Percentages of the clovers under the four soil water conditions were:

Clover species	Soil water category			
	Saturated	Wet	Moist	Dry
	percent			
Bolander's	22.0	47.0	31.0	0.0
Long-stalked	6.0	30.0	61.0	3.0
Carpet	0.0	17.5	66.3	16.2
Mountain	55.7	28.6	15.7	0.0

DISCUSSION

Additional study of long-stalked clover and carpet clover should show that they tolerate relatively wide pH ranges. In this study, those species grew at soil acidities 4.3 and 12.8 times greater than those previously reported.¹⁷ The low pH's encountered may, however, be related to sampling activity, because pH values can be decreased as much as one unit by trampling the meadow surface.¹⁸

Three of the clover species occurred under cooler, wetter conditions than carpet clover, which suggests an ameliorating effect of soil water on soil temperature. Nevertheless, additional study is needed to determine the extent that soil temperature influences clover success. Below-ground plant parts tend to be more sensitive to low temperatures than above-ground parts.⁵ Minimum temperature or temperature fluctuation in the root zone may therefore better define a species' ecologic amplitude than ambient soil or above-ground cardinal temperatures.

A basic premise is that where other environmental minimums are met, soil water is the most important variable controlling plant distribution. Soil water associations among the four clover species from wettest to driest are mountain clover, Bolander's clover, long-stalked clover, and carpet clover. With erosion and meadow drying, therefore, we would expect abundance of clover

Table 1—Averages and differences in soil acidity at the 5-cm to 15-cm depth for four perennial clover species of the Sierra Nevada, California

Clover species	Average (SE) ²	Between-species differences ¹		
		Long-stalked	Carpet	Mountain
		-----Relative acidity ³ -----		
Bolander's	545.7 (44.9)	117.2	32.7	102.4
Long-stalked	428.5 (27.1)		84.5	14.8
Carpet	513.0 (37.9)			69.7
Mountain	443.3 (24.6)			

¹No significant differences by Games and Howell test at $\alpha = 0.05$.

²SE = Standard error of the mean.

³Arithmetically equivalent values of hydrogen ion normality.

Table 2—Averages and differences in soil temperatures at the 10-cm depth for four perennial clover species of the Sierra Nevada, California

Clover species	Average (SE) ²	Between-species differences ¹		
		Long-stalked	Carpet	Mountain
		-----°C-----		
Bolander's	16.08 (0.11)	0.47	0.54*	0.45
Long-stalked	15.60 (0.18)		1.02*	0.03
Carpet	16.62 (0.17)			0.99*
Mountain	15.63 (0.19)			

¹Between-species differences with an asterisk (*) are significantly different by the Games and Howell test at $\alpha = 0.05$.

²SE = Standard error of the mean.

Table 3—Averages and differences in soil water scores (1 = saturated, 2 = wet, 3 = moist, and 4 = dry) for four perennial clover species of the Sierra Nevada, California

Clover species	Average (SE) ²	Between-species differences ¹		
		Long-stalked	Carpet	Mountain
Bolander's	2.090 (0.068)	0.520*	0.898*	0.352*
Long-stalked	2.610 (0.058)		0.378*	0.872*
Carpet	2.988 (0.050)			1.250*
Mountain	1.738 (0.068)			

¹Between-species differences with an * are significantly different by the Games and Howell test at $\alpha = 0.05$.

²SE = Standard error of the mean.

species to decrease in that order. Whether there are corresponding distributions and changes in distributions of rhizobial strains with soil water remains to be determined.

Given the extremes of pH and temperature under which Bolander's clover occurs, the hypothesis that Bolander's clover has a high ecologic amplitude with regard to environmental variables¹⁹ is supported by this study. On average, however, temperature and soil water relationships tend to support the observation that Bolander's clover prefers cool wet sites. The negative association between Bolander's clover and long-stalked clover previously reported¹⁹ is supported by this study, because they occur under somewhat different conditions of soil water.

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END NOTES AND REFERENCES

¹Munz, Philip A.; Keck, David D. 1959. *A California flora*. Berkeley, CA: Univ. of California Press; 1681 p.

²California Department of Fish and Game. 1991. *Natural diversity data base, special plants*. Sacramento, CA. Natural Heritage Division, Department of Fish and Game; 65 p.

³Hughes, H. D.; Heath, Maurice E.; Metcalfe, Darrel S., eds. 1953. *Forages, the science of grassland agriculture*. Ames, IA: The Iowa State College Press; 724 p.

⁴Black, C. A. 1957. *Soil-plant relationships*. New York: John Wiley and Sons; 332 p.

⁵Fitter, A. H.; Hay, R. K. M. 1987. *Environmental physiology of plants*, 2d ed. New York: Academic Press; 423 p.

⁶Lowendorf, Henry S.; Alexander, Martin. 1983. *Identification of Rhizobium phaseoli strains that are tolerant or sensitive to soil acidity*. Applied and Environmental Microbiology 45:737-742.

⁷Damirgi, S. M.; Frederick, L. R.; Anderson, J. C. 1967. *Serogroups of Rhizobium japonicum as affected by soil types*. Agronomy Journal 59:10-12.

⁸Keyser, H. H.; Munns, D. N.; Hohenberg, J. S. 1979. *Acid tolerance of rhizobia in culture and in symbiosis with cowpea*. Soil Science Society of America Journal 43:719-722.

⁹Thornton, Frank C.; Davey, C. B. 1983. *Response of the clover-Rhizobium symbiosis to soil acidity and Rhizobium strain*. Agronomy Journal 75:557-560.

¹⁰Waldon, Hollis B.; Jenkins, Michael B.; Virginia, Ross A.; Harding, Ethelynda E. 1989. *Characteristics of woodland rhizobial populations from surface- and deep-soil environments of the Sonoran desert*. Applied and Environmental Microbiology 55 (12): 3058-3064.

¹¹Lowendorf, Henry S. 1980. *Factors affecting survival of Rhizobium in soil*. Advances in Microbial Ecology 4:87-124.

¹²Bartholomew, W. V. 1957. *Maintaining organic matter*. In: Stefferud, Alfred, ed. *Soil: the yearbook of agriculture 1957*. Washington, DC: U.S. Department of Agriculture; 245-252.

¹³Jones, D. G.; Morley, S. J. 1981. *The effect of pH on host plant 'preference' for strains of Rhizobium trifolii using fluorescent ELISA for strain identification*. Annals of Applied Biology 97:183-190.

¹⁴Hardarson, G.; Jones, D. Gareth. 1979. *Effect of temperature on competition amongst strains of Rhizobium trifolii for nodulation of two white clover varieties*. Annals of Applied Biology 92:229-236.

¹⁵Soil Survey Staff. 1951. *Soil survey manual*. Agric. Handb. 18. Washington, DC: U.S. Department of Agriculture; 503 p.

¹⁶Games, P. A.; Howell, J. F. 1976. *Pairwise comparison procedures with unequal n's and/or variances: A Monte Carlo study*. Journal of Educational Statistics 1:113-125.

¹⁷Ratliff, Raymond D. 1982. *A meadow site classification for the Sierra Nevada, California*. Gen. Tech. Rep. PSW-60. Berkeley, CA: Pacific Southwest Forest and Range Experiment Station, Forest Service, U.S. Department of Agriculture; 16 p.

¹⁸Leonard, Robert; Harkin, Donald; Zinke, Paul. 1968. *Ecological study of meadows in lower Rock Creek, Sequoia National Park: progress report for 1967*. Unpublished draft supplied by authors.

¹⁹Ratliff, Raymond D.; Denton, Renee G. 1992. *Bolander's clover in the central Sierra Nevada: a sensitive species?* Unpublished draft supplied by authors.

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