EFFECT OF NUTRIENT APPLICATION ON LEAF LONGEVITY IN TREE SEEDLINGS: AN EXPERIMENTAL TRIAL

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KIRAN BARGALI & SINGH, S. P. 1994. **Effect of nutrient application on leaf longevity in tree seedlings: an experimental trial.** Leaf longevities of *Quercus leucotrichophora* and *Pinus roxburghii* seedlings were compared at five levels of nutrient application. Seedlings of both species were grown in polyethylene bags each containing 1 *kg* mixture of sieved forest soil and fine sand in 1:3 ratio. Seedlings were grown both in pure (intra specific) and mixed (inter specific) culture. Nutrient application treatments were 0, 144, 264, 384 and 504 *mg* of 12:32:16 NPK fertilizer per bag. Percentage leaf drop increased significantly with increasing level of nutrient application in both species. However, the values were always higher for *Q. leucotrichophora* than for *P. roxburghii*. In mixed culture, leaf longevity of *P. roxburghii* increased while the reverse was true for *Q. leucotrichophora*. In both species percentage leaf drop showed significant positive correlation with soil nutrients, nitrogen concentration in mature leaves and height of seedling.

Key words: Leaf longevity - *Pinus roxburghii* - *Quercus leucotrichophora* - percentage leaf drop - nutrient application

KIRAN BARGALI & SINGH, S. P. 1994. **Kesan penggunaan nutrien keatas kelanjutan umur daun pada anak benih pokok : satu percubaan.** Kelanjutan umur daun anak benih *Quercus leucotrichophora* dan *Pinus roxburghii* dibandingkan pada lima tahap penggunaan nutrien. Anak benih kedua-dua spesies di tanam dalam karung polyethylene di mana setiap satunya mempunyai 1 *kg* campuran tanih hutan yang di tapis dan pasir halus dalam nisbah 1:3. Anak benih ini di tanam dalam kultur tulin (intraspesifik) dan campuran (interspesifik). Rawatan penggunaan nutrien adalah 0, 144, 264, 384 dan 504 *mg* dalam nisbah 12:32:16 baja NPK per karung. Peratus pengguguran daun bertambah dengan ketara dengan bertambahnya penggunaan nutrien pada kedua-dua spesies. Namun demikian nilai-nilai ini selalunya lebih bagi *Q. leucotrichophora* dari *P. roxburghii*. Dalam kultur campuran, kelanjutan umur daun *P. roxburghii* meningkat manakala kelanjutan umur daun *Q. leucotrichophora* menurun. Bagi kedua-dua spesies peratus pengguguran daun menunjukkan korelasi positif yang ketara dengan nutrien tanih, kepekatan nitrogen dalam daun yang matang dan ketinggian anak benih.

Introduction

Leaf longevity is important as a nutrient cycling process and as a specific plant adaptation to nutrient availability. In nutrient cycling context leaf longevity plays a major role in determining the nutrient turnover rate in an ecosystem because a significant portion of the nutrient capital in the ecosystem

is in leaves (Shaver 1981). As an adaptation to nutrient availability, leaf longevity might: (i) determine the photosynthetic carbon return per unit of nutrient invested in leaf production (Small 1972a); (ii) determine the ability of old leaves to act as a nutrient source in production of new leaves (Small 1972b, Turner & Olsen 1976, Reader 1980), (iii) determine the ability of old leaves to act as sinks for nutrients taken up during periods of no growth (Mooney & Rundel 1980). It has long been recognized that leaf longevity is an important life history trait of plants with respect to their growth and response to nutrient availability, light, drought, herbivory and other factors (Gray & Schlesinger 1983, Reich 1987, Coley 1988).

This study deals with the effects of soil nutrient status and inter specific competition on leaf longevity in *Quercus leucotrichophora* A. Camus and *Pinus roxburghii* Sarg. seedlings. They are the major tree species of Central Himalaya occurring between 1000-2000 *m* a.s.l. and are evergreen with concentrated summer leaf drop and simultaneous renewal of the canopies (Ralhan *et al.* 1985). Trees of these two species have similar leaf longevities of just more than one year. In this region *P. roxburghii* occurs on sites that are nutrient-poor, while *Q. leucotrichophora* is found in nutrient-rich sites. The objectives of this study were: (i) to analyse responses of these two species to changes in nutrient availability in terms of leaf longevity and (ii) to see whether or not this response was affected by interspecific competition as competition for nutrients by various plant parts controls the course of senescence in leaves (Molisch 1938).

Materials and methods

Seedlings of *Q. leucotrichophora* and *P. roxburghii* were raised from the healthy seeds of the current year crop in plastic bags (each containing 1 *kg* prepared soil), and maintained at two seedlings per bag. In one set, each bag contained two individuals of the same species (pure culture) and there were 6 replicate bags per species per treatment (*i.e.*, nutrient level). In the other set each bag contained an individual of each species (mixed culture) and there were 12 replicate bags per nutrient treatment.

Soil used in this experiment was collected to a depth of 15 *cm* from a stand of *Q. leucotrichophora* forest. Soil was air dried and sieved through wire mesh screen (mesh size 1 × 1 *mm*) to remove all plant parts and gravel. After sieving the soil had 0.275% N, 0.098% P and 0.113 % K. This sieved soil was mixed with washed fine commercial sand (having an undetectable level of nutrients) in 1:3 ratio, and filled into plastic bags. The gradient of nutrient availability was developed by adding 0, 144, 264, 384 and 504 *mg* of 12:32:16 NPK fertilizer to the bags once each growing season (hereafter referred to as N1, N2, N3, N4 and N5 nutrient level respectively). Total nutrient concentrations in the soil ($mg\ kg^{-1}$) were determined for each nutrient level (as given in Singh & Bisht 1992). All levels of nutrients were subjected to equal, regular supplies of distilled water. A layer of cotton gauge prevented soil from being washed during watering as the bags contained holes for drainage. The experiment was carried out under glass house conditions for a two-

year duration where the mean maximum temperatures were 1- 5°C higher than the air temperatures (Singh & Bisht 1992) and all the seedling received full sunlight. The temperature in the glass house was within the range of temperature experienced by the species in nature, taking into account that both species occur at elevations of 1000 *m* lower than the site of the experiment (*i.e.*, 2000 *m*. a.s.l.)

Seedlings were selected for the uniformity of size. At the start of experiment the average heights of *Q. leucotrichophora* and *P. roxburghii* seedlings were 4.63 ± 0.02 *cm* and 3.62 ± 0.004 *cm* respectively. Leaves falling during the study period were collected, oven dried and weighed. All the leaves falling during the first year and second year were then summed for each treatment separately and calculated as percentage of total leaves produced that year. Final height values of the seedlings up to shoot apex were measured at the end of the experiment. For nutrient analysis mature leaves (> 50% green) were ground into fine powder and digested using a quick digestion unit for N and nitric acid perchloric acid procedure for P and K. Nitrogen was determined by the micro Kjeldahl technique (Misra 1968). Phosphorus was determined by spectrophotometer and potassium by flame photometer.

Effects of species (*Q. leucotrichophora* versus *P. roxburghii*), competition (pure versus mixed), year (first versus second) and nutrient application (N1-N5) were analysed using analysis of variance (Snedecor & Cochran 1968).

Results and discussion

Nutrient availability versus leaf longevity

The analysis of variance indicates that the percentage leaf drop was significantly affected by species, competition, year, nutrient application and some of their interactions (see Table 1). In both species the percentage leaf drop (of total leaves produced in terms of dry mass) increased with increasing nutrient availability (Figure 1). Such increase was relatively more pronounced in *Q. leucotrichophora*. In *P. roxburghii* it was well marked only in the second year in pure culture. One might expect leaf longevity to decrease with increased nutrient availability because: (i) new evergreen leaves are photosynthetically more efficient than old leaves (Johnson & Tieszen 1976) and maintenance of old leaves may be inefficient when rapid new leaf production is possible, or (ii) increased new leaf production with higher nutrients may simply shade out the old leaves (Shaver 1981). Increase in nutrient availability resulted in increased leaf production but decreased leaf survival, although the effects were significant in the second year only, leading to a positive correlation between soil nutrients (*mg kg* $^{-1}$) and percentage leaf drop (Table 2). This indicates that the life span of leaves decreased in response to increased nutrient availability (Chapin 1980, Shaver 1981, Mooney & Gulman 1982). Increased leaf longevity in low nutrient environments provides greater photosynthetic gain per unit nutrient allocated to the leaf. Thus photosynthesis by long-lived leaves can meet plant energy requirements with little or no leaf production, so that the plants can survive the period when there are

insufficient plant nutrient reserves to sustain leaf production. According to Shaver (1981), the inverse relationship between nutrient concentration in the soil and leaf longevity indicates the direction of the response to a chronic change in nutrient availability, *i.e.*, with increased nutrient availability, a plant community should become more "deciduous" and less "evergreen".

Table 1. F values and probability values (p) from an analysis of variance for percentage leaf drop of *Q. leucotrichophora* and *P. roxburghii*

Source of variation	Degree of freedom	Mean square	F Value	p Value
Species (S)	1	604.5	72.0	0.001
Competition (C)	1	9.5	1.13	0.250
Year (Y)	1	450.4	53.7	0.001
Nutrient (N)	4	179.1	21.3	0.001
S × C	1	138.8	16.5	0.001
S × Y	1	100.9	12.0	0.01
S × N	4	21.9	2.6	NS
C × Y	1	7.3	0.9	NS
C × N	4	1.9	0.2	NS
Y × N	4	5.4	0.6	NS
Error	17	8.4		

Table 2. Parameters of linear regressions relating percentage leaf drop of second year to total soil nutrient ($mg\ kg^{-1}$). Regression equations are of the form $P = a + bN$, where P = percentage leaf drop and N = soil nutrient concentration

| Species	Culture	Nutrient	Intercept (a)	Slope (b)	Correlation coefficient (r)
Quercus leucotrichophora	Pure	Nitrogen	- 123.65	0.21	0.99*
		Phosphorus	2. 86	0.08	0.99*
		Potassium	- 22.42	0.16	0.99*
	Mixed	Nitrogen	-139.82	0.24	0.99*
		Phosphorus	6.32	0.09	0.99*
		Potassium	- 23.48	0.18	0.99*
Pinus roxburghii	Pure	Nitrogen	-182.32	0.29	0.99*
		Phosphorus	-10.10	0.11	0.99*
		Potassium	- 45.65	0.21	0.99*
	Mixed	Nitrogen	- 56.79	0.11	0.94**
		Phosphorus	5.88	0.04	0.94**
		Potassium	- 7.72	0.08	0.94**

*p < 0.01, **p < 0.05.
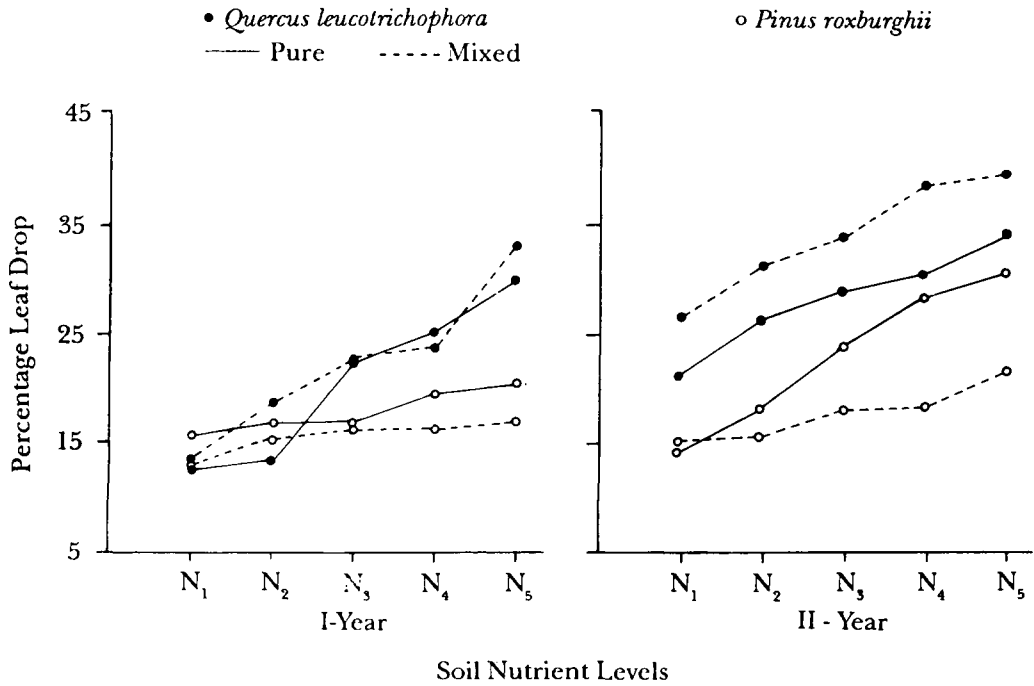
Figure 1. Percentage leaf drop versus nutrient concentration in the soil for *Q. leucotrichophora* and *P. roxburghii*

Foliar nutrient concentration versus leaf longevity

Regression equations between nutrient (N, P, K) concentration (%) in mature leaves and percentage leaf drop show a significant positive correlation for both the species (Table 3). In these two species as nutrient concentration in leaves increased with increasing nutrient level (see Table 4) the leaf life span decreased. Reich *et al.* (1991, 1992) also reported similar correlation between foliar concentrations and leaf life span for different tree species in a diverse ecosystem.

Height of the seedling versus leaf longevity

As the height of the seedling increased (except for *Q. leucotrichophora* in mixed culture) with increasing nutrient level (see Table 5), percentage leaf drop shows a significant positive correlation with height of the seedlings (Figure 2). (However, in mixed culture this relationship was negative for *Q. leucotrichophora*). Coley (1988) also found similar correlation between height growth and leaf life span for a large group of species in Panama. This indicates that within species individual plants grown in environments with high (versus low) nutrient availability have greater leaf nutrient mass (*e.g.*, Klinka & Carter 1990, del Arco *et.al.* 1991), resulting in greater

Table 3. Parameters of linear regressions relating percentage leaf drop of second year
to nutrient concentration in mature leaves (%). Regression equations
are of the form P = a + bN where P = percentage leaf drop
and N = Foliar nutrient concentration

Species	Culture	Nutrient	Intercept (a)	Slope (b)	Correlation coefficient (r)
Quercus leucotrichophora	Pure	Nitrogen	- 37.40	31.75	0.96*
		Phosphorus	- 24.86	367.31	0.97*
		Potassium	- 62.70	116.2	0.99*
.	Mixed	Nitrogen	- 31.29	33.16	0.89*
		Phosphorus	- 28.77	439.40	0.98*
		Potassium	- 85.80	154.6	0.96*
Pinus roxburghii	Pure	Nitrogen	-193.75	123.82	0.99*
		Phosphorus	- 88.14	887.50	0.99*
		Potassium	- 52.27	100.73	0.92*
	Mixed	Nitrogen	- 18.58	21.08	0.89**
		Phosphorus	- 11.70	230.96	0.88**
		Potassium	- 4.71	33.05	0.90**

$*p < 0.01$, $**p < 0.05$.

Table 4. Mean concentration (% ± SE) of nutrients at five nutrient levels for leaves
of *Q. leucotrichophora* and *P. roxburghii*

| | *Quercus leucotrichophora* | | *Pinus roxburghii* | |
	Pure	Mixed	Pure	Mixed
Foliar nitrogen				
N_1	1.880 ± 0.021	1.801 ± 0.013	1.691 ± 0.015	1.690 ± 0.017
N_2	1.962 ± 0.034	1.867 ± 0.026	1.706 ± 0.002	1.698 ± 0.018
N_3	2.012 ± 0.056	1.901 ± 0.052	1.742 ± 0.048	1.885 ± 0.032
N_4	2.021 ± 0.052	2.005 ± 0.045	1.805 ± 0.023	1.899 ± 0.078
N_5	2.281 ± 0.054	2.205 ± 0.052	1.806 ± 0.041	1.905 ± 0.048
Foliar phosphorus				
N_1	1.130 ± 0.003	0.129 ± 0.004	0.116 ± 0.002	0.118 ± 0.007
N_2	0.135 ± 0.008	0.134 ± 0.005	0.121 ± 0.002	0.120 ± 0.002
N_3	0.145 ± 0.011	0.146 ± 0.006	0.127 ± 0.007	0.125 ± 0.006
N_4	0.155 ± 0.012	0.154 ± 0.011	0.132 ± 0.008	0.138 ± 0.007
N_5	0.162 ± 0.013	0.159 ± 0.016	0.135 ± 0.017	0.139 ± 0.018
Foliar potassium				
N_1	0.720 ± 0.008	0.715 ± 0.010	0.650 ± 0.011	0.620 ± 0.012
N_2	0.770 ± 0.012	0.762 ± 0.023	0.742 ± 0.013	0.680 ± 0.017
N_3	0.786 ± 0.012	0.785 ± 0.016	0.781 ± 0.010	0.752 ± 0.008
N_4	0.816 ± 0.032	0.792 ± 0.018	0.820 ± 0.021	0.780 ± 0.020
N_5	0.828 ± 0.031	0.825 ± 0.023	0.825 ± 0.011	0.812 ± 0.012

canopy growth than root growth (*e.g.*, Walters & Reich 1989), and thus a high rate of height growth. High rates of height growth will result in earlier leaf senescence (Hirose & Werger 1987, Field 1988) with associated retranslocation of nutrient to new foliage higher on the stem with faster turnover rates of individual leaves (*i.e.*, shorter leaf life span).

Table 5. Mean values of seedling height (*cm* + 1 SE) of *Q. leucotrichophora* and *P. roxburghii* at five nutrient levels

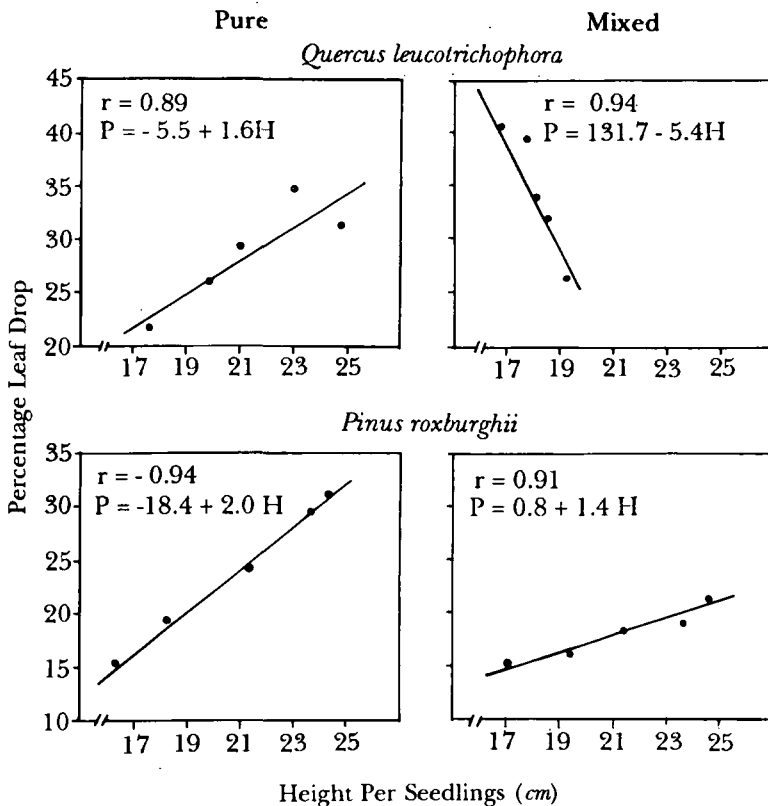|	Quercus leucotrichophora		Pinus roxburghii	
	Pure	Mixed	Pure	Mixed
Height (*cm*)				
N_1	17.6 ± 0.82	19.2 ± 0.92	16.4 ± 0.85	17.0 ± 1.23
N_2	19.8 ± 1.05	18.7 ± 1.02	18.2 ± 0.89	19.3 ± 1.36
N_3	20.9 ± 1.13	18.0 ± 1.01	21.3 ± 1.65	21.5 ± 1.12
N_4	24.6 ± 1.25	17.7 ± 0.83	23.5 ± 1.68	23.5 ± 1.12
N_5	23.1 ± 1.21	16.6 ± 0.92	24.2 ± 1.78	24.5 ± 1.05

Figure 2. Percentage leaf drop in relation to height of the seedlings for *Q. leucotrichophora* and *P. roxburghii*. Relationships were significant at p < 0.05, where P = percentage leaf drop and H = height of the seedling

Interspecific competition versus leaf longevity

Mixed culture of the two species produced contrasting responses. While in *P. roxburghii* it reduced the percentage of leaf drop at all nutrient levels, in *Q. leucotrichophora* it resulted in higher percentage of leaf drop. However, these effects became significant in the second year when values of leaf drop were higher than in the first year (Figure 1). Increased leaf life span in *P. roxburghii* will increase the time period for photosynthesis (Mooney & Dunn 1970). In this regard it can compensate for low rates of photosynthesis (Orians & Solbrig 1977, Schulze *et al.* 1977,) or allow *P. roxburghii* to initiate photosynthesis early in the spring before conditions are suitable for leaf growth, and utilize the longer part of winter season for dry mass accumulation.

Conclusion

The results of the present study help in understanding the role of leaf longevity in nutrient cycling of the ecosystem. For example, the fact that leaf survival decreased with increasing nutrient availability suggests that as nutrient input increases the nutrient movement through leaf fall also increases. As the litter of the fertilized plants would almost certainly have a higher nutrient content (Chapin 1980, Linder & Rook 1984) and would decompose faster (Van Cleve 1974), the turnover of nutrients in the ecosystem would become rapid. However, this study was limited by the fact that it was focussed on seedlings and the experiment was conducted for a short period of the life of long-lived plants. However, the present study could form a basis of a further study dealing with quantitive evaluation of the relationship between leaf life span and other characteristics, *viz.* relative growth rate, net photosynthetic rate, leaf area, ratio, specific leaf area, leaf mass and leaf nutrient mass, *etc.* in different plant species of the Central Himalayan forest.

Acknowledgements

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