

Note

Nutrient concentrations in *Pinus radiata* D Don needles in the Basque Country (Spain): a preliminary classification of parameters and sites

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Summary — This work presents the first results on nutrient concentrations in *Pinus radiata* needles in the Basque Country (Spain). Current year and 1-yr-old needles collected at different sites were analyzed for calcium, magnesium, potassium, phosphorus, nitrogen and manganese content. Our results indicate some differences between sites, seasons and needles of different ages. Nutrient levels are sufficient, in general, for tree nutritive requirements, but some low concentrations of magnesium, and especially phosphorus found in 1-yr-old needles, indicate the possibility that some trees may show signs of deficiency in the near future. In turn, nitrogen and manganese concentrations are, in many cases, higher than those usually found in this species and may give rise to growth disorders (nitrogen) or toxicity (manganese).

We present here a preliminary classification of sites and parameters based on the principal component analysis. Our results show the utility of this method to differentiate appropriate sites for the adequate development of *Pinus radiata*.

nutrient / cation / needle / *Pinus radiata* / deficiency / classification

Résumé — Concentration en éléments nutritifs des aiguilles de *Pinus radiata* D Don dans le Pays Basque (Espagne) : classification préliminaire des paramètres et des sites. Ce travail présente les premiers résultats concernant la concentration en éléments nutritifs des aiguilles de *Pinus radiata* dans le Pays Basque (Espagne). Des éléments nutritifs (calcium, magnésium, potassium, phosphore, azote et manganèse) ont été analysés dans des aiguilles de *Pinus radiata* de l'année courante et de 1 an prélevées dans différentes stations. Les résultats indiquent quelques différences selon les stations, les saisons et l'âge des aiguilles. Les niveaux d'éléments nutritifs sont, en général, suffisants pour les besoins nutritifs des arbres, mais quelques faibles concentrations en magnésium, et surtout en phosphore, observées dans les aiguilles de 1 an, pourraient indiquer une possible déficience dans le futur. D'un autre côté, les concentrations d'azote et de manganèse sont, dans beaucoup des cas, plus élevées que d'habitude pour cette espèce et pourraient provoquer

ainsi des désordres de croissance (azote) ou toxiques (manganèse). La classification des stations et des paramètres réalisée sur la base de l'analyse des composantes principales montre que cette méthode est utile pour différencier les stations favorables à un développement adéquat de Pinus radiata.

éléments nutritifs / cations / aiguilles / Pinus radiata / déficiences / classification

INTRODUCTION

Of the total surface area planted with *Pinus radiata* in Spain (> 200 000 ha, ie ≈ 10% of the world total (Turner and Lambert, 1986) > 70% is located in the Basque Country (Departamento de Agricultura y Pesca, Gobierno Vasco, 1986). The economic importance of this coniferous species in this region is thus evident, as is the concern about the sanitary situation of these trees, at the moment that a general deterioration due to unknown reasons has been observed in many forested areas in Europe and North America.

Since investigations began, many hypotheses have been considered to explain the phenomenon of forest decline. Air pollution and acid rain were for a long time considered as the main causes of the decline, but at present the phenomenon is attributed to many different causes that often act synergistically. As many types of damage are associated with nutritional problems (Zöttl and Mies, 1983; Hüetl *et al*, 1987) the study of the nutrient concentration of pine needles is of great importance.

In order to determine the effect of air pollution on the plantations of *Pinus radiata* in the Basque Country and their present sanitary state, a study is being carried out within the context of the EPOCA programme (Estudio en el Pirineo Occidental de la Contaminación Acida). This study includes inventories of forest damage, soil

analysis and nutrient concentration analysis in needles of trees from different sites. The results concerning this last section are presented here.

MATERIALS AND METHODS

Ten sites of *Pinus radiata* distributed throughout the territory of the Basque Country were chosen according to location of urban and industrial areas, symptoms of damage, age of trees and their geographical situation. The physiographical and tree growth data corresponding to the sites studied can be seen in table I.

Foliar samples were collected in November 1989 and November 1990. The sites Irún and Lezama were not sampled in 1990 because both of them were victims of fire and abusive pruning. In 1990 the site Odiaga was added to increase the information on the area near the industrial town of Llodio. In autumn 1990 it was not possible to take samples from the Ispazter site as the trees were considerably damaged by parasites (*Thaumetopoea pityocampa*).

Healthy needles were harvested from 3 trees representative of each site. Current-year and 1-yr-old needles were collected from branches in the upper third of the tree (fifth whorl in mature trees, or third whorl in young trees). The needles collected were taken to the laboratory and oven-dried at 80 °C for 48 h. A dry mineralization technique was then used before analysis.

Calcium, magnesium, potassium and manganese concentrations were measured by atomic absorption, nitrogen by Kjeldahl and Büchi apparatus and phosphorus colorimetrically. The methods have been reported elsewhere (MAFF, 1981). The needles of each tree were analysed separately.

Table I. Values of some physiographical and tree growth parameters of sites.

Sites	Altitude (m)	Slope (%)	Age (yr)	Height (m)*	Height/ age*	DBH (cm)*
Irún	100	23	30	25 (2.1)	0.83	46 (14.1)
Durango	220	25	29	30 (3.3)	1.03	57 (6.8)
Murueta	150	0	24	26 (4.8)	1.08	39 (10.3)
Odiaga	450	30	23	22 (2.0)	0.91	34 (14.1)
Urkiola	620	15	17	12 (1.2)	0.71	31 (9.0)
Ansorregi	320	55	29	29 (1.4)	1.00	49 (13.8)
Lezama	450	10	9	12 (0.5)	1.33	27 (8.0)
Cruceta	850	35	18	16 (2.8)	0.89	34 (8.3)
Olaeta	600	0	40	25	0.63	58
Ispazter	150	43	23	24 (1.0)	1.04	41 (3.5)

DBH : diameter breast height; * Results are the mean of 3 trees; () : SE.

RESULTS AND DISCUSSION

The results obtained in the analysis of foliar samples are shown in table II. Although the number of measurements (3) at each site was not enough for ideal statistical accuracy, the results indicate some differences depending on the age of the needles, the time of collection and the sites sampled.

As can be seen in table I, some of the sites studied are located at altitudes which exceed or approach 500 m, the altitude at which the growth of *Pinus radiata* is limited by low temperatures and strong winds (Scott, 1961; Gandullo *et al*, 1974).

NUTRIENT CONCENTRATIONS

Calcium concentration

In all cases the values exceed the deficiency levels indicated by several authors

(Turner and Lambert, 1986). The results show how calcium is accumulated in the needles as these grow older (comparing current year vs 1-yr-old needles)*. The values of needles of the same age collected in the autumn of 1989 and 1990 indicate a certain tendency of the calcium concentration to drop, with the exception of trees of the Ansorregi site, which is situated on a limestone soil.

Magnesium concentration

Considering a magnesium concentration of 0.07% as the critical level and considering 0.06–0.08% as the range of low values (Will, 1966, 1978); low or critical levels in 1-yr-old needles were observed at some sites (table II). These levels appeared in Irún in autumn 1989 and in Cruceta, Olaeta, Urkiola y Odiaga in autumn 1990. These levels lead us to envisage a possible magnesium nutritional problem in the areas affected in the future, especially if this situation becomes worse. Current

* Statements followed by this symbol have been verified by the Mann—Whitney *U* test (Centre d'Enseignement et de Recherche de Statistique Appliquée, 1986).

Table II. Nutrient concentrations (% dry weight) of needles from each of the 10 sites. Mean values and SE.

Element	Ca (%)				Mg (%)				K (%)			
	0.1 (Turner and Lambert, 1986)		0.07 (Will, 1966, 1978)		0.4 (Hall and Purnell, 1961)							
Deficiency level	Nov 1989 Mean	Nov 1990 SE	Nov 1989 Mean	Nov 1990 SE	Nov 1989 Mean	Nov 1990 SE	Nov 1989 Mean	Nov 1990 SE	Nov 1989 Mean	Nov 1990 SE	Nov 1989 Mean	Nov 1990 SE
<i>Current year needles</i>												
Ansorregi	0.24	0.04	0.27	0.00	0.15	0.04	0.15	0.00	0.70	0.11	0.65	0.15
Cruceta	0.19	0.02	0.17	0.04	0.12	0.01	0.11	0.04	1.03	0.10	1.04	0.17
Olaeta	0.17	0.04	0.12	0.03	0.10	0.03	0.11	0.02	0.79	0.10	0.74	0.17
Urkiola	0.24	0.02	0.20	0.05	0.10	0.01	0.11	0.01	1.07	0.22	0.69	0.32
Durango	0.24	0.03	0.20	0.06	0.14	0.01	0.14	0.01	1.05	0.47	0.26	0.05
Murueta	0.28	0.02	0.30	0.03	0.14	0.02	0.19	0.07	0.76	0.11	0.61	0.11
Izpaster	0.18	0.05			0.17	0.03			1.02	0.04		
Irún	0.21	0.02			0.11	0.01			0.95	0.07		
Lezama	0.32	0.07			0.11	0.02			0.92	0.07		
Odiaga			0.19	0.06			0.12	0.02			0.69	0.21
<i>1 yr-old needles</i>												
Ansorregi	0.35	0.08	0.39	0.03	0.14	0.06	0.15	0.07	0.72	0.08	0.61	0.04
Cruceta	0.43		0.26	0.06	0.10	0.00	0.08	0.02	0.75		0.72	0.07
Olaeta	0.34	0.09	0.24	0.03	0.12	0.04	0.08	0.01	0.64	0.05	0.53	0.03
Urkiola	0.49	0.03	0.40	0.09	0.11	0.01	0.07	0.00	0.67	0.06	0.54	0.08
Durango	0.37	0.09	0.35	0.11	0.13	0.01	0.10	0.02	0.80	0.27	0.34	0.11
Murueta			0.51	0.11			0.09	0.01			0.54	0.09
Izpaster	0.24	0.06			0.11	0.01			0.91	0.15		
Irún	0.33	0.02			0.07				0.59	0.39		
Lezama	0.29	0.02			0.09	0.00			0.98	0.15		
Odiaga			0.24	0.07			0.06	0.01			0.34	0.25

year needles, however, do not show deficiency levels of magnesium in any site (table II).

The results show that magnesium levels also drop with age*. The 1-yr-old needles contain \approx 25% less magnesium than the current year needles.

Potassium concentration

Potassium concentration tends to decrease in needles as they grow older. A reduction of the levels at all sites was also observed from autumn 1989 to autumn 1990 (table II)*. This decrease may lead to

P (%)				N (%)				Mn (ppm)				N/P			
0.12 (Turner and Lambert, 1986)				1.1 (Will, 1971)				10 (Lange, 1969)							
Nov 1989		Nov 1990		Nov 1989		Nov 1990		Nov 1989		Nov 1990		Nov 1989		Nov 1990	
Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
0.16	0.01	0.13	0.01	2.05	0.11	1.73	0.04	181.1	96.0	158.4	89.6	12.6	1.5	13.3	1.1
0.11	0.00	0.12	0.01	1.83	0.06	2.05	0.14	281.2	83.2	320.7	105.7	16.1	0.9	17.5	1.2
0.13	0.01	0.13	0.02	1.91	0.03	1.69	0.23	385.2	203.3	424.9	221.2	14.7	1.1	13.0	1.6
0.12	0.01	0.13	0.01	1.78	0.08	1.83	0.15	196.0	54.5	220.5	57.4	14.8	1.3	13.7	1.0
0.12	0.01	0.13	0.01	1.79	0.11	1.77	0.16	400.2	245.4	422.1	161.8	14.9	2.5	13.6	1.1
0.14	0.01	0.15	0.03	1.78	0.15	1.54	0.09	147.8	46.7	226.3	176.1	12.7	0.6	10.1	2.7
0.15	0.02			1.83	0.20			272.3	94.9			12.2	0.2		
0.12	0.02			1.86	0.12			758.0	273.0			15.9	2.4		
0.15	0.00			1.79	0.15			365.7	76.9			11.8	1.1		
		0.12	0.01			1.71	0.20			671.2	264.4			14.7	2.8
0.14	0.01	0.12	0.02	1.88	0.13	1.53	0.06	275.0	137.0	376.5	304.2	13.3	0.5	13.3	
0.09	0.00	0.11	0.02	1.86		2.14	0.20	606.0		493.2	67.9	20.6		19.5	3.4
0.12	0.01	0.10	0.01	1.90	0.11	1.70	0.22	762.8	234.8	726.3	371.6	15.8	0.7	16.5	1.3
0.12	0.02	0.08	0.02	1.79	0.03	1.40	0.09	243.8	34.8	359.7	182.3	14.9		16.8	3.3
0.10	0.01	0.09	0.01	1.63	0.38	1.44	0.26	636.8	436.0	609.9	298.1	16.5	2.4	16.0	1.6
		0.09	0.01			1.22	0.18			216.3	137.9			14.1	0.9
0.11	0.01			1.64	0.19			332.5	151.3			14.7	0.4		
0.10	0.01			1.64	0.24			799.5	229.5			16.4	0.8		
0.14				1.71	0.04			415.5	30.0			12.2	0.3		
		0.08	0.01			1.47	0.20			902.4	402.3			19.1	4.2

problems in the coming years. The lack of potassium affects the vitality and growth of the trees (Raupach and Clarke, 1972). The deficiency levels indicated by Hall and Purnell (1961) ($K < 0.4\%$) or those indicated

by Hall and Raupach (1963) (0.25–0.35%) were not reached in any of these cases, although the levels found in Durango and Odiaga in autumn 1990 were surprisingly low.

Phosphorus concentration

Phosphorus deficiency in plantations of *Pinus radiata* is a well-known problem in many countries where this tree is planted. Successive generations of this species occupying the same plot can use up the scarce reserves of phosphorus in the soil and suffer from a growth deficiency which, in many cases, must be corrected by fertilizing the soil (Raupach, 1967; Gentle and Humphreys, 1968; Raupach *et al*, 1975; Baker and Attiwill, 1985). On the other hand, forestry practices can cause the loss of a large part of the phosphorus available by removing the organic material from the surface of the soil in which this nutrient is found (Daniel *et al*, 1982; Nambiar, 1985).

In our case, we have observed low levels of phosphorus which, nevertheless, did not appear to produce visual symptoms of deficiency. However, it should be pointed out, as observed in other countries (Turner and Lambert, 1986), that the trees from the vast majority of plantations in the Basque Country do not retain 3-yr-old needles on their branches, and that the 2-yr-old needles are very scarce. Consequently, there is a lack of mature foliage in many trees.

The comparison of our results with the critical phosphorus levels indicated by several authors (0.1%; Humphreys and Truman, 1964; 0.11%, Will, 1965; or 0.12%, Turner and Lambert, 1986), shows that the levels of phosphorus found in 1-yr-old needles reach deficiency levels in the majority of needles collected in 1990, and many in 1989 (table II). Only the sites at Ansozregi, Lezama and Ispazter situated in rural areas near the coast show values higher than or equal to deficiency values. Values close to deficiency levels warn of a possible appearance of deficiency problems in new plantations in the future or in the following generations of *Pinus radiata* in certain areas of the Basque Country.

Nitrogen concentration

The nitrogen concentration values largely exceed the critical level indicated by Will (1971). In fact, the large number of branches and their strong growth observed in many trees in the sites studied can be related to the high nitrogen concentration values found in the trees (Stone and Will, 1965).

Manganese concentration

The manganese levels are quite high and exceed the deficiency levels indicated by different authors (10 ppm; Lange, 1969; 24–63 ppm, Marcos de Lanuza, 1966). Although an evident toxic effect due to manganese accumulation in leaves has not been described (Kazda and Zvacek, 1989), damage can be caused by manganese acting synergistically with other elements such as aluminium (Kazda and Zvacek, 1989).

The manganese levels, as observed in the case of calcium, are greater in 1-yr-old needles* due to accumulation with aging (table II).

The existence of steel mills and foundries near the sites of Durango, Olaeta and Odiaga may be related to the rather high levels of manganese concentrations in the needles.

Nitrogen/phosphorus (N:P) ratio

According to several authors, this ratio is considered to be a good marker of the nutritional status of *Pinus radiata* (Raupach, 1967; Raupach *et al*, 1969). It ranges from 5 to 16 (Raupach, 1967) and a mean value of 10 is considered as good (Raupach *et al*, 1969).

In our case all values exceed 10 and in most 1-yr-old needles are > 16. These high values confirm the excess of nitrogen and the low phosphorus concentration found in needles.

The ratio values in Ansorregi, Murueta and Ispazter are close to 10, indicating a better nutritional balance of nitrogen and phosphorus than that of other sites.

CLASSIFICATION OF SITES AND PHYSIOGRAPHICAL AND NUTRITIONAL PARAMETERS BY MEANS OF CORRELATION ANALYSIS AND PRINCIPAL COMPONENT ANALYSIS (PCA)

We have made statistical analysis of correlations and principal components analysis in view of determining the relationships between parameters and the classification of sites.

Correlation analyses

The correlation coefficients between nutrient concentrations, growth and physiological parameters in current-year needles are shown in table III. The positive correlation between phosphorus and magnesium is especially noteworthy. Calcium shows significant positive correlations with magnesium and phosphorus (as already observed by Gentle *et al*, 1968; and Turner and Lambert, 1986), as well as with the height/age (H:A) ratio. Magnesium is negatively correlated with altitude and with the N:P ratio, indicating low levels of this nutrient in the mountainous sites studied. The negative correlation between altitude and phosphorus is also outstanding, indicating, as in the case of magnesium, the low values of phosphorus found in trees growing in the mountains.

The positive correlation between phosphorus and the H:A ratio indicates the importance of phosphorus for the height growth of the trees. The N:P ratio is positively correlated with altitude and negatively correlated with the H:A ratio. These results indicate that high N:P ratio values are observed at higher altitudes and correspond to lower height growth rates of the trees. The negative correlation of altitude with the H:A ratio confirms the negative effect of greater altitudes on height growth.

Table IV shows the correlation coefficients for 1-yr-old needles. Here again, the positive correlation between magnesium and phosphorus and their negative correlation with the N:P ratio are noteworthy. These nutrients are very important in photosynthesis processes and show, as we have already seen, low levels in 1-yr-old needles of some sites. We have to note that, in this case, the phosphorus level is positively correlated with nitrogen and potassium, as Raupach *et al* (1978) have already pointed out. These nutrients seem to take a special importance in the more mature needles of this species and are probably involved in retranslocation processes from the more mature needles to those in development (Turner and Lambert, 1986).

The results of the correlation analysis have been used to classify the parameters into 2 types, the "healthy" and the "unhealthy" parameters. The "healthy" parameters are magnesium, phosphorus, potassium and the H:A ratio, which are positively correlated each other. High values of these parameters are related with high rates of tree growth. The "unhealthy" parameters are nitrogen, manganese, the N:P ratio, altitude and age which are positively correlated each other. High values of these parameters are related to low rates of tree growth and nutritional imbalances (high values of N:P ratio).

Table III. Correlation coefficients significant at $P < 0.05$ between nutrient concentrations, growth and physiological parameters. Current year needles.

	Calcium	Magnesium	Potassium	Phosphorus	Nitrogen	Manganese	Nit/phos	Altitude	Age	Height/age
Calcium	1.000									
Magnesium	0.372	1.000								
Potassium			1.000							
Phosphorus	0.312	0.657		1.000						
Nitrogen					1.000					
Manganese						1.000				
Nit phos	-0.354	-0.468		-0.780	0.497		1.000			
Altitude	-0.383	-0.456		-0.343			0.394	1.000		
Age	-0.390								1.000	
Height/age	0.574	0.344		0.326			-0.341	-0.543	-0.570	1.000

Table IV. Correlation coefficients significant at $P < 0.05$ between nutrient concentrations, growth and physiological parameters. 1-yr-old needles.

	Calcium	Magnesium	Potassium	Phosphorus	Nitrogen	Manganese	Nit/phos	Altitude	Age	Height/age
Calcium	1.000									
Magnesium		1.000								
Potassium			1.000							
Phosphorus		0.455	0.607	1.000						
Nitrogen	-0.418		0.336	0.569	1.000					
Manganese						1.000				
Nit phos		-0.351	-0.243	-0.570			1.000			
Altitude					0.452		0.399	1.000		
Age									1.000	
Height/age								-0.543	-0.570	1.000

Principal component analysis (PCA)

Two principal component analyses (PCA) (Lebart *et al*, 1979) corresponding to current year and 1-yr-old needles, respectively (figs 1, 2), have been used to classify the sites and parameters. Only parameters correlated with the axes at $P < 0.05$ have been considered.

Current year needles

The percent of variance explanation is 38.7% for axis 1 and 19.1% for axis 2 (fig 1).

At the positive end of axis 1 are situated the calcium, magnesium, phosphorus and H:A ratio parameters whereas at the negative end are situated the altitude and the N:P ratio. In other words, the so-called healthy are opposed to the unhealthy parameters (fig 1a).

The distribution of sites shows, at the positive end of axis 1, the Ansorregi, Is-pazter and Murueta sites, located in the same position as the healthy parameters. At the negative end of the axis are situated the Cruceta, Olaeta and Urkiola sites, located in the same position as the unhealthy parameters (fig 1b). Comparing the position of sites in 1989 and 1990 we can see a certain tendency towards healthy values in 1990.

1-yr-old needles

The percent of variance explanation is 29.1% for axis 1 and 23.9% for axis 2 (fig 2).

At the positive end of axis 1 are situated the potassium, phosphorus, magnesium and the H:A ratio parameters, coinciding to a large extent with the healthy parameters already observed in the case of current-

year needles. At the negative end of the axis are situated the N:P ratio, manganese and altitude (*ie* the unhealthy parameters) (fig 2a).

The distribution of sites on the axes is similar to that of current year needles. As we can see, the Urkiola site occupies an intermediary position. Comparing the position of sites in 1989 and 1990, we can see a certain tendency towards unhealthy values in 1990, contrary to what happened in current year needles. This fact could be related to a possible retranslocation of nutrients, like magnesium and phosphorus, from the more mature needles to the younger ones taking place in Autumn 1990.

CONCLUSIONS

Our results concerning the nutrient level of the needles do not indicate very serious deficiencies. However, some cases of low levels of magnesium and especially phosphorus, which seem to increase with time, constitute a warning to be kept in mind during the coming years.

Soil nutrient level is the main factor influencing element content in the needles. Results from soil analysis (data not shown) confirm our results of needle analysis and show that sites like Ansorregi, Murueta and Lezama maintain a better nutritional balance than the other sites. However, sites like Odiaga and Cruceta have lower levels of soil cations.

The principal component analysis (PCA) has been confirmed to be an efficient instrument to separate sites according to nutrient concentrations and physiographical and tree growth parameters. PCA can provide a better knowledge of nutrient requirements of sites and facilitate the diagnosis of nutritional problems.

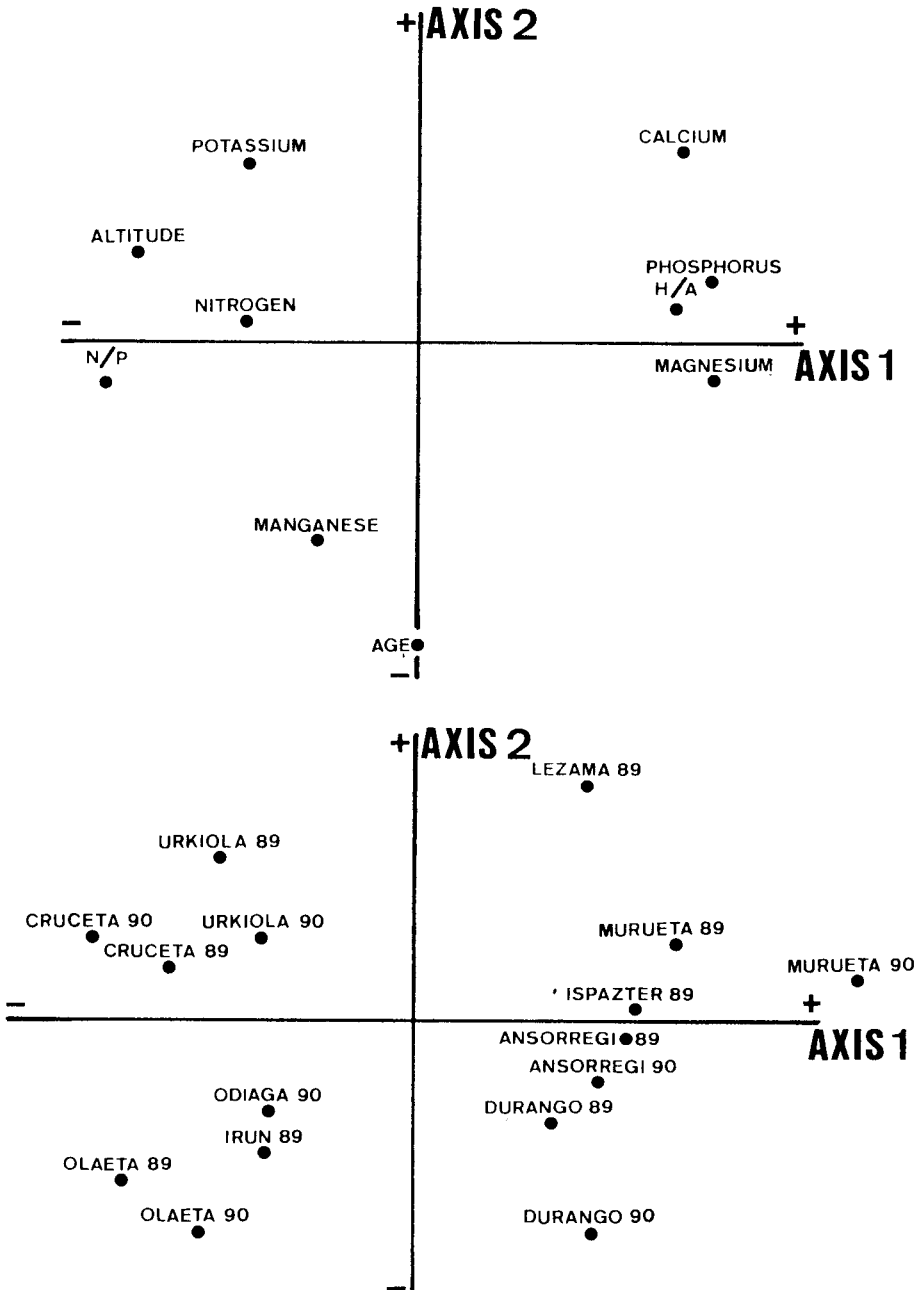


Fig 1. Principal component analysis (PCA). Current-year needles. Axis 1 and 2: **a.** Distribution of nutrients and physiological and tree growth parameters. **b.** Distribution of sites in November 1989 and November 1990.

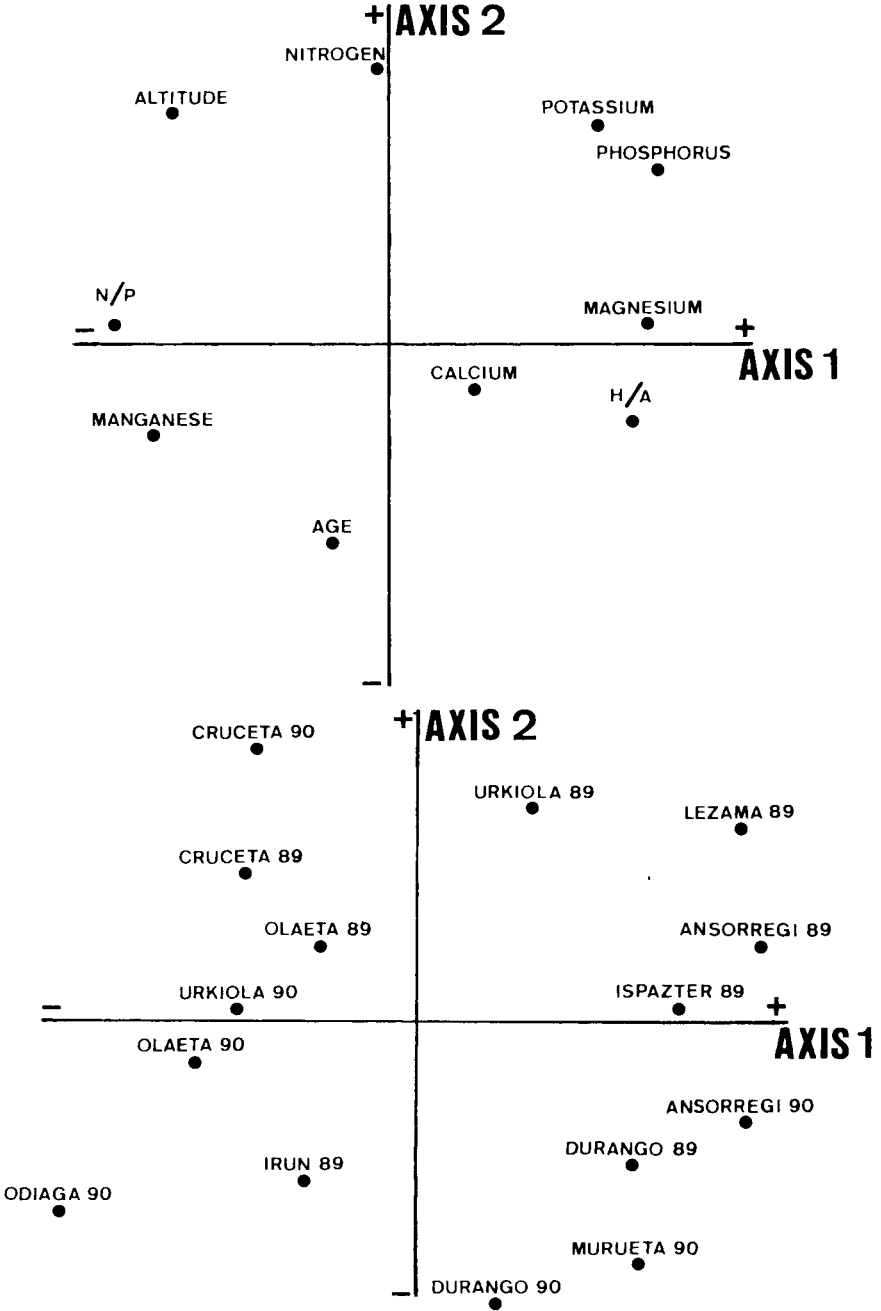


Fig 2. Principal component analysis (PCA). 1-yr-old needles. Axis 1 and 2: **a.** Distribution of nutrients and physiographical and tree growth parameters. **b.** Distribution of sites in November 1989 and November 1990.

This analysis is now being completed with the study of the relationship between nutrient concentration of needles, atmospheric conditions (pollution, climate), biochemical parameters and nutrient levels in the soil.

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