SOME FACTORS AFFECTING FOLIAR ANALYSIS IN SUGARCANE

By J. M. GOSNELL and A. C. LONG

Rhodesia Sugar Association Experiment Station,
Chiredzi, Rhodesia

Abstract

During the morning, time of sampling did not materially affect foliar N, P, K, Ca, Mg analyses, but there was a decline in N and Mg and a rise in K when sampling was done in the afternoon. Delays of up to 24 hours between sampling and removal of the midrib did not affect N, P, Ca, Mg analyses, but there was a rise in K content when delays occurred.

Increasing age (eliminating seasonal variability) caused very large reductions in foliar N, and fairly large reductions in P, K, Ca, Mg and Mn. In most cases the rate of decline was fairly slow after five months. Age did not have marked effects on Fe, Cu or Zn. When age was eliminated, season had marked effects on N and Ca, both of which declined markedly between August and April; there was a similar effect with P and Mg, but no seasonal effect on K.

Substantial differences were found between different varieties, with a tendency for the values of K, Ca and Mg to be correlated. Moisture stress caused a decline in N, K and P and a rise in Ca. The soil moisture deficit since the last irrigation may be a useful guide here.

Increasing applications of nitrogen produced an exponential rise in foliar nitrogen, and a useful relationship with sugar yield/hectare was found. Nitrogen deficiency caused a marked reduction in uptake of P, K, Ca, Mg, Mn, Cu and Zn.

Where soil phosphate was high, massive applications of phosphate produced no increase in foliar P or yield; at medium values of soil phosphate, foliar P was increased but yields unaffected, and at low soil values, both foliar P and yields were increased by phosphate application. Phosphate deficiency also resulted in reduced uptake of N, K, Ca and Mg.

On most soils, potash application has had no effect on either foliar K or yields; however, in one case on a soil with low K analysis, application of potash resulted in marked increase in foliar K with a decrease in Ca and Mg.

Tables of correction factors for season and variety have been compiled.

Introduction

Foliar analysis is used in many parts of the world as an adjunct to, or instead of soil analysis for fertilizer recommendations. It has one clear advantage over soil analysis: by analysing what nutrients the plant has actually taken up, it eliminates doubts as to whether certain nutrients are in an "available" form. However foliar analysis introduces a whole new set of variables such as age, season and variety which are not present in soil analysis. It is therefore necessary to standardize as many variables as possible in sampling for foliar analysis and to determine in advance the effects which other variables may have.

To this end, foliar samples were taken from a large number of experiments and some special sampling trials were instituted. The effects of various factors on the analyses for the major elements (N, P, K, Ca, Mg) are given below together with trace elements in certain cases.

In order to facilitate sampling on a large scale, it was decided to use only one tissue, and the top visible dewlap leaf was selected as it is the quickest to sample and sampling errors are less likely than with other tissues. Primary stalks were selected from vigorously grown cane of age 5 months $\pm$ 2 weeks. Damaged or diseased leaves were rejected and 32 leaves were taken from each experimental plot. The leaves were placed in polythene tubes and removed to the laboratory.

As the true centre of length of the leaf of a large bundle of leaves is very time consuming to determine, and may be affected by broken leaf tips, it was decided to follow Farquhar* and use the centre of gravity of the leaf bundle as a simple and practical standard sampling point. A length of 20 cm from the centre of gravity towards the tip was cut out. The midribs were then removed and the laminae oven dried and finely ground. Details of the analytical methods are given by Long (1971).

Effect of time of sampling

Little factual evidence is available on the effect of sampling sugarcane at various times of day; while recommendations have been made to complete sampling by 8 a.m. (Halais 1962) or by 10 a.m. (Farquhar 1965), it is not easy to carry this out on a large scale where routine foliar sampling of many fields is carried out.

The effect of sampling at various times of day was investigated in a fully irrigated field of five month old third ratoon NCo 376 in December 1970. Triplicate samples were taken at two hourly intervals from 6 a.m. to 4 p.m., placed in plastic bags and removed to the laboratory where the midribs were immediately removed. Good agreement was achieved between the triplicate samples as shown by Long (1971). The effects of time of day on the foliar analysis for N, P, K, Ca and Mg are shown in Figure 1.

*Personal communication. R. H. Farquhar, Colonial Sugar Refining Co., Australia (1968).

There was little change in the nitrogen content of samples taken between 6 a.m. and 12 noon, but there was a reduction in the afternoon. Phosphorus content was apparently unaffected by the time of day of sampling; however there was a marked rise in potassium content in the afternoon. The calcium content rose to a peak at midday, dropping again in the afternoon while magnesium was steady in the morning, dropping off in the afternoon. It is interesting to note that the drop in magnesium was mirrored by the rise in potassium over the same period.

From these analyses it may be concluded that the time of sampling does not materially affect the result provided that sampling is carried out in the morning. There are appreciable fluctuations in K and Mg and a significant decline in N during the afternoon.

Effect of delay in midrib removal

Advice is generally given that the midribs should be removed immediately after taking the foliar sample. This is presumably because of evidence of potassium migration between lamina and midrib (Evans 1965). However, midrib removal in the field by unskilled operators is not desirable, and an investigation was therefore undertaken to ascertain the effect of delay in midrib removal on nutrient content of leaves. The samples were taken in a field of five month old third ratoon NCo 376. Twelve samples were taken at 9 a.m., and divided into four treatments, as follows:

0 hrs. Midribs removed in the field within two minutes of taking the sample.
2 hrs. Midribs removed in the laboratory after two hours.
6 hrs. Midribs removed in the laboratory after six hours.
24 hrs. Midribs removed in the laboratory after 24 hours.

The results are given in Table I.

TABLE I

Effect of delay from sampling to removal of midrib on foliar N, P, K, Ca, Mg.

Delay	0 hrs.	2 hrs.	6 hrs.	24 hrs.	L.s.d. 5%	L.s.d. 1%	C.V.%
% N	2,20	2,15	2,21	2,21	0,10	0,15	2,2
% P	,247	,240	,243	,253	,051	,077	3,3
% K	1,46	1,51	1,55	1,55	0,12	0,19	4,1
% Ca	,230	,220	,233	,217	,024	,037	5,4
% Mg	,180	,160	,143	,173	,017	,026	5,1

The results confirm that there was a migration of potassium from the midribs which was complete some 2-6 hours after sampling. No effect of delay in midrib removal was evident on any other nutrient. The practical conclusions from these two investigations are that all sampling should be carried out between 6 a.m. and 12 noon and the samples brought into the laboratory where they are processed in the afternoon, some 4-8 hours after sampling. By this time, the migration of K should have finished.

Effect of age

Reduction in leaf nitrogen content with increasing age has been frequently observed, e.g. Clements (1959), Bishop (1965) and Samuels (1969). An exact evaluation of the effect of age has not always been possible, however, because of the confounding effect of age and season with a single time of planting.

We established an experiment in which four plots (one each of our main commercial varieties NCo 376, NCo 310, CP 29-116 and Co 462) were planted on the 10th of each month for 18 successive months. Upon reaching the age of 27 months (commencing February 1970) each set of four plots was ratooned, and foliar samples were then taken at monthly intervals starting at one month. Thus foliar analyses are available for any age of cane at any month in the year.

In order to ascertain the true effect of age, eliminating seasonal variability, data was extracted from four months of the main foliar sampling season, December, January, February and March, and meaned over the four varieties to give the information shown in Figure 2, each point representing the mean of 16 samples.

The effect of age on nitrogen is extremely marked, especially over the first three months, after which the rate of decline lessens until it is fairly slow from six months onwards. Foliar potassium declined fairly sharply until three months of age after which there was little or no change. Phosphorus, calcium and magnesium all declined sharply until about five months, after which the values levelled off (with calcium) or declined slowly (with P and Mg).

The effect of age on some trace elements is shown in Table II.

TABLE II

Effect of age on foliar Fe, Mn, Zn, Cu, B (ppm)

Age	Fe	Mn	Zn	Cu	B
1	121	63	8,5	10,8	3,2
2	120	52	9,1	14,2	4,2
3	121	42	7,8	13,8	4,1
4	117	44	9,2	16,1	5,0
5	127	40	10,8	15,0	3,4
6	129	34	10,5	12,2	5,0
7	132	32	10,2	11,0	4,8
8	139	27	9,5	11,8	3,6
9	132	26	9,0	10,4	3,9
Critical level	5	20	15	4	1
L.s.d. 5%	33	15	2,1	4,5	—
1%	44	20	2,8	6,1	—
C.V. %	17,7	25,0	15,1	23,9	—

Manganese showed a marked and steady drop with increasing age; iron was erratic but showed a slight increase with increasing age; copper and to a smaller extent zinc rose to a peak at 4-6 months after which there was a decline. However, all values with the exception of zinc were substantially above the critical levels proposed by Evans (1965) and these trends are therefore of little practical importance. The possibility of an incipient zinc deficiency is currently being investigated.

Seasonal effect

Fluctuations of foliar nutrient contents with season have been observed, e.g. by Holford (1968). However, the seasonal variability has usually been confounded with variation in age and plant moisture status.

We used data from the experiment described above, and eliminated age as a variable by grouping analyses of fixed ages (mean 3, 4, 5 and 6 months) over the various months of the sampling season. As the crop was fully irrigated, variations in plant moisture status should be eliminated. Figure 3 shows the effect of season on the major nutrients, each point being the mean of 16 samples.

There was a very marked reduction in nitrogen content as the season advanced, dropping from over 2,1% in August to around 1,6% at the end of the season. Calcium content also showed a very marked reduction from 0,4% to 0,2% over the same period. There was little, if any, effect of season on potassium, while phosphorus and magnesium showed slight drops over the period.

Although chronological age has been eliminated as a variable in this seasonal comparison, it is possible that part of the effect described above is due to increasing physiological age of the crop as the season advances. It is obvious that a five month old crop in September, (which was ratooned in April and grown over the winter months), is much less advanced than a five month old crop in March (which was ratooned in October and grown over the summer months).

It is therefore clear that different critical values must be applied during the various months or that sampling should be carried out at a younger age in summer than in winter.

Varietal effect

The influence of variety on nutrient content has been noted by a number of workers, e.g. Farquhar and Lee (1962), Halais (1965).

The wide range of levels obtaining in the varieties is exemplified by foliar analyses from a variety trial in which promising and pre-release varieties were compared with the standard NCo 376. All samples were taken at five months and results are given in Table III.

TABLE III

Influence of Variety on Foliar N, P, K, Ca, Mg

Variety	N	P	K	Ca	Mg
Co 1001	1,92	0,172	1,56	,285	,195
NCo 376	1,82	0,170	1,46	,212	,148
N 52-219	1,78	0,140	1,58	,302	,165
M 31-45	1,77	0,165	1,70	,285	,158
Co 462	1,74	0,162	1,49	,358	,172
N 55-805	1,73	0,172	1,43	,275	,152
Pindar	1,66	0,170	1,58	,325	,162
CB 40-77	1,54	0,155	1,80	,280	,212
L.s.d. 5%	0,11	0,023	0,12	,060	,019
1%	0,15	0,031	0,16	,082	,026
C.V. %	4,2	9,6	5,0	14,1	7,7

There was a very large variation in nitrogen level from a high value of 1,92 for Co 1001 to a low value of 1,54 for CB 40-77, these differences being very highly significant. Large and significant differences between varieties were also observed for P, K, Ca and Mg analyses, although the variability in the case of Ca was rather high.

There is some evidence of a correlation between the K, Ca and Mg levels in a particular variety, e.g. NCo 376 has the lowest Ca and Mg values and second lowest K values; N 55-805 has the lowest K, and second lowest Ca and Mg values; CB 40-77 has the highest K and Mg levels with moderately high Ca.

Table IV, based on results from a third ratoon trial in which each variety was replicated 30 times, gives a more precise comparison of the three major commercial varieties.

TABLE IV

Foliar analysis of NCo 310, NCo 376 & CP 29-116

Variety	N	P	K	Ca	Mg
NCo 310	1,92	0,219	1,37	,307	,185
NCo 376	1,98	0,227	1,38	,264	,180
CP 29-116	1,86	0,199	1,47	,245	,235
L.s.d. 5%	0,04	0,007	0,05	,020	,012
1%	0,05	0,009	0,06	,026	,017
C.V. %	3,9	5,9	6,3	14,1	12,0

Effect of moisture stress

It is generally accepted that moisture stress affects foliar nutrient analyses, especially nitrogen, and that sampling should only be carried out where moisture is non-limiting (Halaïs 1962). Little factual evidence is available, however, on the effects of various degrees of moisture stress on foliar analysis; in most cases, e.g. Samuels (1965), comparisons were made simply of irrigated and unirrigated cane.

Foliar samples were taken in a third ratoon experiment of NCo 376 on which six irrigation treatments were applied ranging from 1,0 × Pan to 0,37 × Pan. Figure 4 shows the effect of irrigation on foliar analysis.

Severe moisture stress caused a marked reduction in potassium content with no difference between well irrigated and moderately stressed treatments. There was a marked increase in calcium content with severe moisture stress presumably due to smaller cells with higher proportion of calcium pectate rich cells walls. Phosphorus content showed a slight but steady drop with increasing moisture stress, while magnesium showed a drop followed by a rise which could not be interpreted.

The most interesting effect is that of nitrogen which shows a steady decline with increasing moisture stress, except for a sharp rise at the driest treatment. However, if the treatments are rearranged according to the number of days from irrigation prior to sampling, a pattern emerges, as shown in Table V.

TABLE V

Foliar nitrogen content as affected by moisture deficit at time of sampling

Days between Irrigation & Sampling	Pan Moisture Deficit (mm)	% N	Treatment (Pan factor)
5	24	1,96	1,00
5	24	1,95	0,37
7	35	1,92	0,84
7	35	1,91	0,84/0,60
8	40	1,92	0,68
13	66	1,84	0,53

It is clear that the nitrogen content declines with increasing soil moisture deficit, as measured by the pan evaporation deficit. A further investigation is at present being carried out to ascertain whether a critical deficit can be established, which will facilitate field sampling procedures.

Effect of nitrogen

Foliar nitrogen

Foliar analyses from 10 typical nitrogen trials are shown in Figure 5; in all cases the crops were ratoon NCo 376. There is a marked exponential relationship between increasing N fertilizer levels and foliar N, which is clearly depicted in the line shown in Figure 5 for two trials in the same field which cover the range 0 to 168 and 101 to 258 kg N/ha. This curve is similar to but more marked than the relationship shown by Bonnet (1965).

Foliar analyses from these two trials are plotted in Figure 6 against sugar yield/hectare. It appears that the quadratic relationship between foliar N and sugar yield becomes asymptotic until at luxury levels of N, both yield and N content decline.

It would appear that a level of about 1,9 should be taken as the critical value of N for ratoon NCo 376. This is somewhat higher than the value currently used (1,8%). In practice, a high proportion of all samples received lies between 1,8 and 1,9%.

Other elements

The influence of nitrogen deficiency on the foliar analyses of other elements has been noted by Farquhar (1965) who found that nitrogen deficiency severely depressed uptake of P; the use of a P : N ratio was suggested as an index of phosphate deficiency where a shortage of N is suspected.

We found in several experiments that nitrogen deficiency caused large reductions in the analyses of most elements. Typical are the two trials mentioned above which covered the range from 0 to 258 kg N/ha. Results of P, K, Ca and Mg foliar analyses from these experiments are shown in Figure 7.

There was a substantial increase in P, K, Ca and Mg foliar values with an increase in N application from 0 to 100 kg N/ha. Above 100 kg N/ha increasing N level had no further effect, and it is apparent that the extreme nitrogen deficiency associated with N levels below 100 kg/ha depressed uptake of the other major nutrients.

Very similar trends were observed with the trace elements copper, zinc and manganese as shown in Figure 8. Foliar iron content was apparently unaffected by nitrogen applications, however.

Effect of phosphate

Foliar phosphorus

Useful relationships between leaf phosphorus and sugar yield per acre are given by Gonzalez-Velez and Samuels (1962), who found that foliar phosphorus continued to increase with applied phosphate beyond the point where yield increases occurred. In our experiments, three distinct zones of response to applied phosphate could be detected: where soil phosphate was high (above 50 ppm P_2O_5 on the resin extract vanadomolybdate method), neither foliar P nor yield were affected by even massive applications of up to 400 kg P_2O_5/ha. In such instances foliar values of P were generally between 0,24 and 0,28%.

Where the soil phosphate level was intermediate (20-50 ppm P_2O_5), phosphate applications produced an increase in foliar P, but no yield response. Foliar analyses of P ranged from 0,19 to 0,25% in this category.

Where the soil phosphate level was low (below 20 ppm P_2O_5), increases in both foliar P and sugar yields have been obtained; foliar P values were generally 0,18 or lower in this category.

It is noteworthy that in the last category, the increase in foliar P was not very large, but it did rise above the critical value set at 0,18 for five month old NCo 376. This is shown in Figure 9, which includes results meaned two ratoons in some instances shown.

Other elements

Where soil phosphate is sufficiently high that no yield response is observed, phosphate applications at rates up to 400 kg P_2O_5/ha have not affected foliar N, K, Ca or Mg values. However, where soil phosphate is deficient, application of phosphate has resulted in increases in all these values, as shown in Table VI.

TABLE VI

Effect of phosphate fertilizing on foliar N, K, Ca, Mg (mean of 2 years)

	% N		% K		% Ca		% Mg	
	6300/4	NPK	6300/4	NPK	6300/4	NPK	6300/4	NPK
P_2O_5 0	1,67	1,59	1,27	0,98	,248	,273	,208	,164
kg/ 34	1,72		1,30		,266		,220	
ha 67	1,72		1,35		,252		,230	
101	1,72		1,34		,250		,230	
112		1,71		1,03		,288		,195
224		1,70		1,04		,314		,201

In general, the rises are not large, although they are appreciable in some cases. The increases in foliar N, K, Ca and Mg normally occur mainly between O and a low level of applied phosphate; thus indicating that when acute phosphate deficiency has been alleviated, applied phosphate has little further effect on these elements.

Effect of potash

In the majority of Lowveld soils, the potash status is adequate to very high and no yield response has been obtained, nor has the foliar analysis for K or other elements been significantly affected by potash application. However, some interesting results were obtained in a N P K trial where foliar sampling was carried out in sixth and seventh ratoons on a soil of moderately low K status (0,30 m.e. %). There was a slight sugar yield response of about 6% to potash application which also resulted in an appreciable increase in foliar K. This confirms work from Natal reviewed by Stewart (1969) that the critical level is about 1,10%. On the other hand, potash application caused a marked reduction in foliar Ca and Mg, presumably an example of cationic antagonism. This effect is shown in Figure 10 which shows the mean foliar values in sixth and seventh ratoon for plots receiving adequate nitrogen. These effects were not discernible in the no nitrogen plots where a severe nitrogen deficiency prevented normal growth.

Effect of other elements

It has already been observed (Gosnell and Long 1969) that an acute sulphur deficiency reduced not only the foliar sulphur content, but also drastically reduced potash uptake, and also caused reduced Mn, B, Cu and increased Fe and Al contents.

In the subsequent ratoon of the same trial, adequate sulphur was supplied to all treatments which now compared with the application of a number of major and trace elements (Ca, Mg, Cu, Zn, B, Mo). Results of foliar analyses on this trial are given in Table VII, which shows that none of the applied nutrients affected the foliar concentration of any nutrient. This was not surprising, since no yield responses were obtained to any treatment.

TABLE VII

Effects of application of some major & trace elements on the foliar concentration of the element concerned

Element	Rate kg/ha	Foliar Concentration	Control	Unit	L.s.d. 5%	C.V. %
Ca	56	,305	,300	%	,034	7,7
Mg	34	,218	,213	%	,028	9,3
Cu	8,5	12,5	13,3	ppm	3,7	19,5
Zn	7,6	17,3	15,5	ppm	2,4	10,3

Discussion

The overall variability caused by interacting factors on foliar analysis is almost overwhelming, and great care must be taken with interpretation of results.

Some of the variables can be satisfactorily eliminated by standardization of procedure. Thus all our sampling is carried out between 6 a.m. and midday with midribs being removed in the afternoon. All samples are taken from 5 months (± 2 weeks) old cane and any visual moisture stress is avoided. Further improvements may be possible here by sampling up to a certain evaporation deficit. The effect of nitrogen deficiency on other elements can be guarded against by interpreting the analyses of all other elements in conjunction with the nitrogen analysis. The same could be done for P and K deficiencies, but these are in practice uncommon.

The remaining variables (variety and season) can be allowed for by using separate criteria for different varieties and months or by establishing correction factors to be applied. Table VIII and IX give correction factors which have been derived from the results obtained in the experiments described above and others. It is expected that the factors will be altered as further information comes to hand.

TABLE VIII

Proposed seasonal correction values

	N	P	K	Ca	Mg
September	– ,20	– ,015	0	– ,10	– ,020
October	– ,15	– ,010	0	– ,08	– ,015
November	– ,10	– ,010	0	– ,05	– ,010
December	– ,05	– ,005	0	– ,03	– ,005
January	0	0	0	0	0
February	+ ,05	+ ,005	0	+ ,02	+ ,005
March	+ ,10	+ ,010	0	+ ,05	+ ,010
April	+ ,15	+ ,015	0	+ ,07	+ ,015
May	+ ,20	+ ,020	0	+ ,10	+ ,020

TABLE IX

Proposed varietal corrections

Variety	N	P	K	Ca	Mg
NCo 310	+ ,06	+ ,012	0	– ,025	0
CP 29-116	+ ,11	+ ,028	– ,10	– ,014	– .060
NCo 376	0	0	0	0	0

These corrections factors are applied to the following standard critical values which refer to ratoon NCo 376 five months old sampled in January:

N	1,8%
P	0,18%
K	1,10%
Ca	0,18%
Mg	0,08%

Acknowledgements

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Discussion

Mr. du Toit (in the chair): Many factors such as age, season etc., can be accounted for but with varieties the position is more difficult. However, the authors have shown that there are large differences between varieties. These are for average figures—what effect would there be on the critical level? Apparently the critical level must be found for each variety.

Dr. Gosnell: If a variety is fit to be released after years of hard work by the plant breeding department then it is at least entitled to a nitrogen level trial.

Dr. Sumner: The data in figure 4 gives excellent support to the contention of various workers that potash and phosphate are diffusion controlled and that calcium is controlled by mass flow. As the amount of water supplied increased so did the amount of phosphate taken up by the plant.

With calcium, by increasing water we are diluting the soil solution and therefore the amount of calcium being presented to the roots will be less, and less will be taken up. Depending whether the nitrogen is ammonium or nitrate you get mass flow or diffusion.

Mr. du Toit: I prefer Dr. Gosnell's explanation regarding nitrogen as it fits the data perfectly.

Mr. Meyer: Did Dr. Gosnell standardise sampling times and what were seasonal effects?

Dr. Gosnell: All samples were taken before noon.

In table VIII we have introduced proposed seasonal correction values which will be modified in the light of later data that will be collected.

A study of field data collected last season shows that fluctuations are somewhat less marked than are reflected in these figures.

We are still considering what correction, if any, to make for irrigation.

Dr. Thompson: Earlier work done at the Experiment Station by Mr. du Toit and myself at Sezela and Illovo on foliar diagnosis corroborates the findings in this paper. Regarding the effect of season, we got depressions of N, P and K levels during winter. Regarding the effect of age, we got lower values for nutrients in the second summer than in the first. Under dry conditions N and P levels dropped more markedly than did the K levels. There were also instances where applications of N fertilizer caused P levels to rise as well.

Varieties certainly have their own threshold values, particularly the K value for NC_o 293.

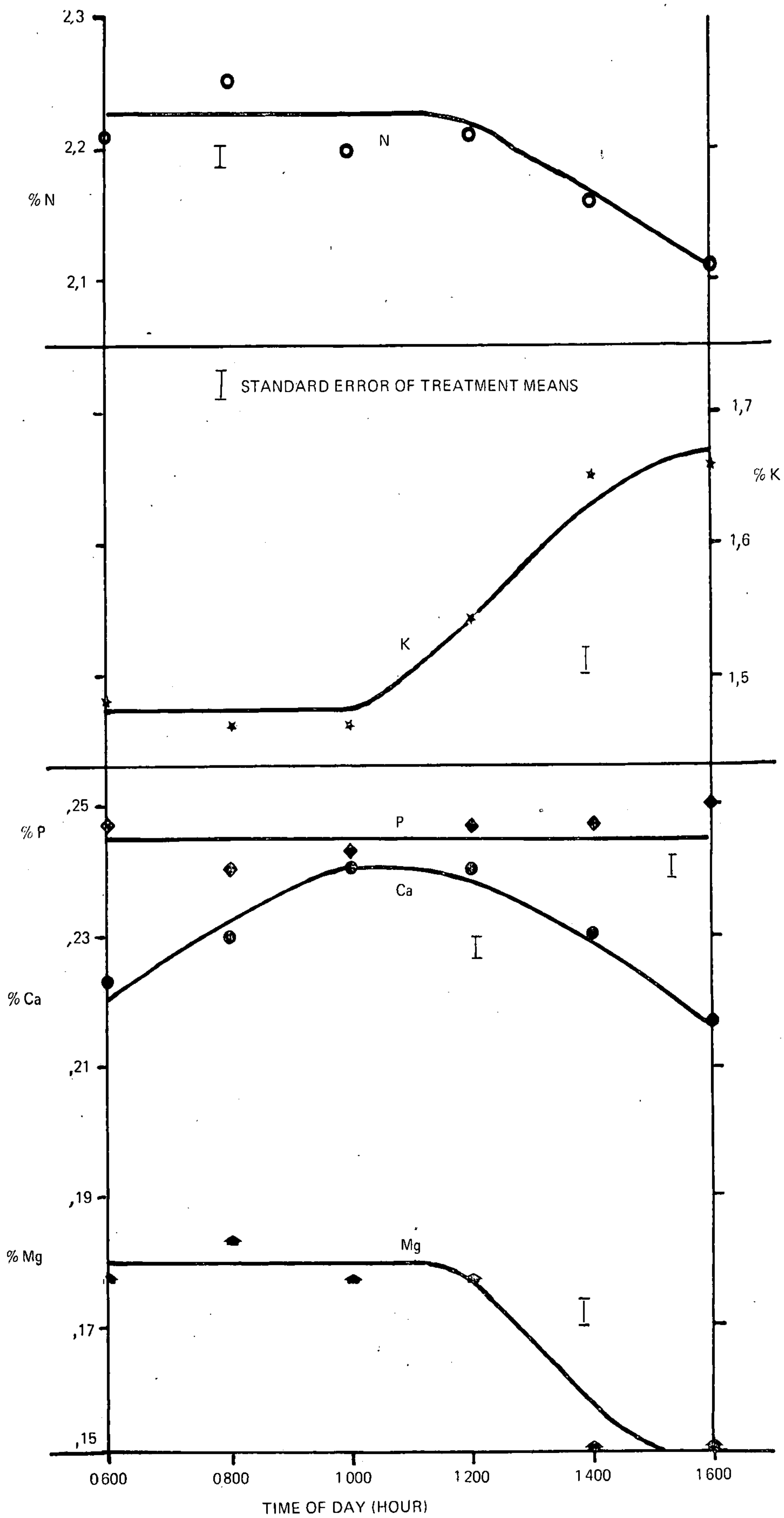

FIGURE 1: Effect of time of sampling on foliar N, P, K, Ca, Mg.

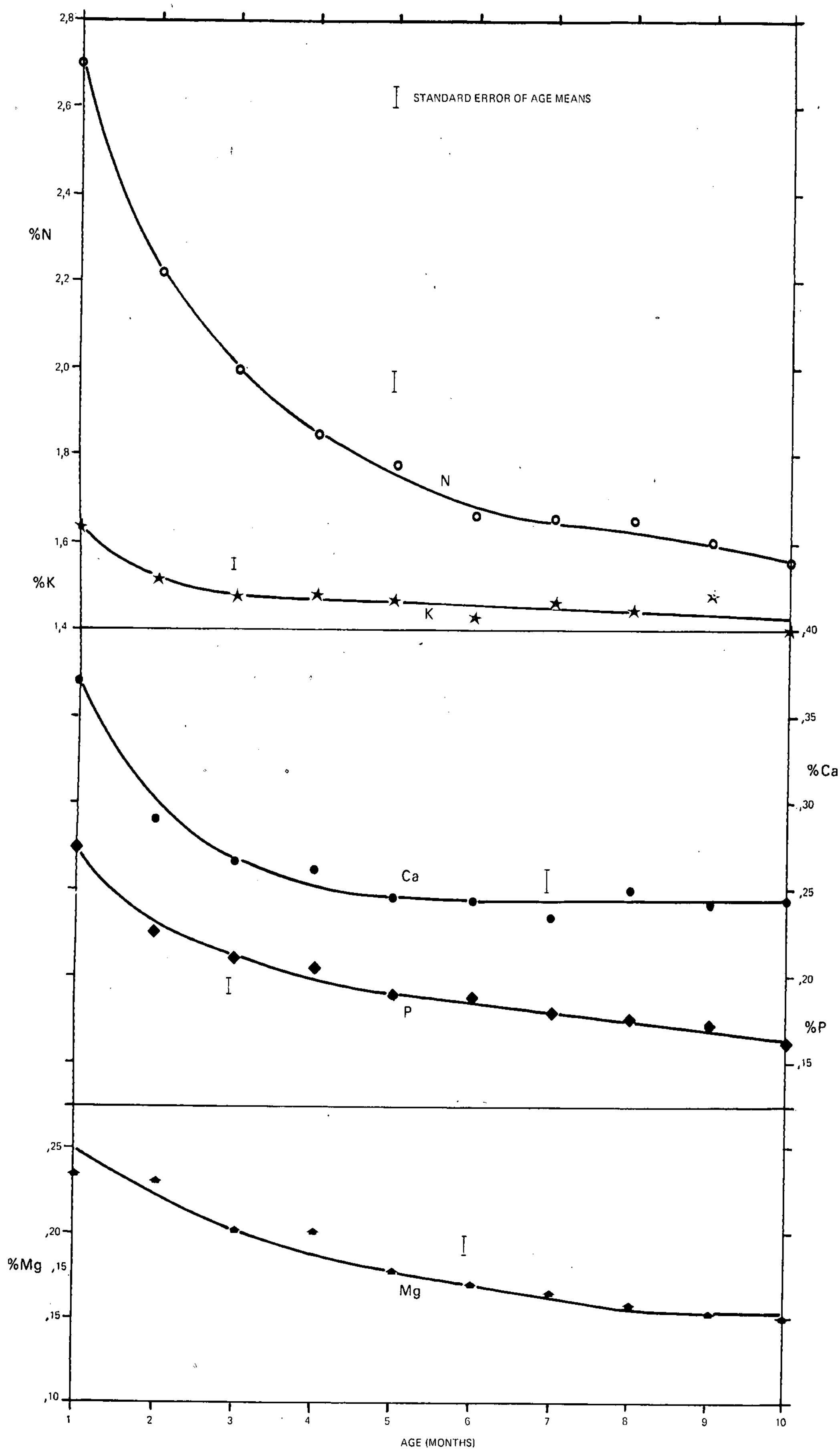

FIGURE 2: Effect of age on foliar N, P, K, Ca, Mg.

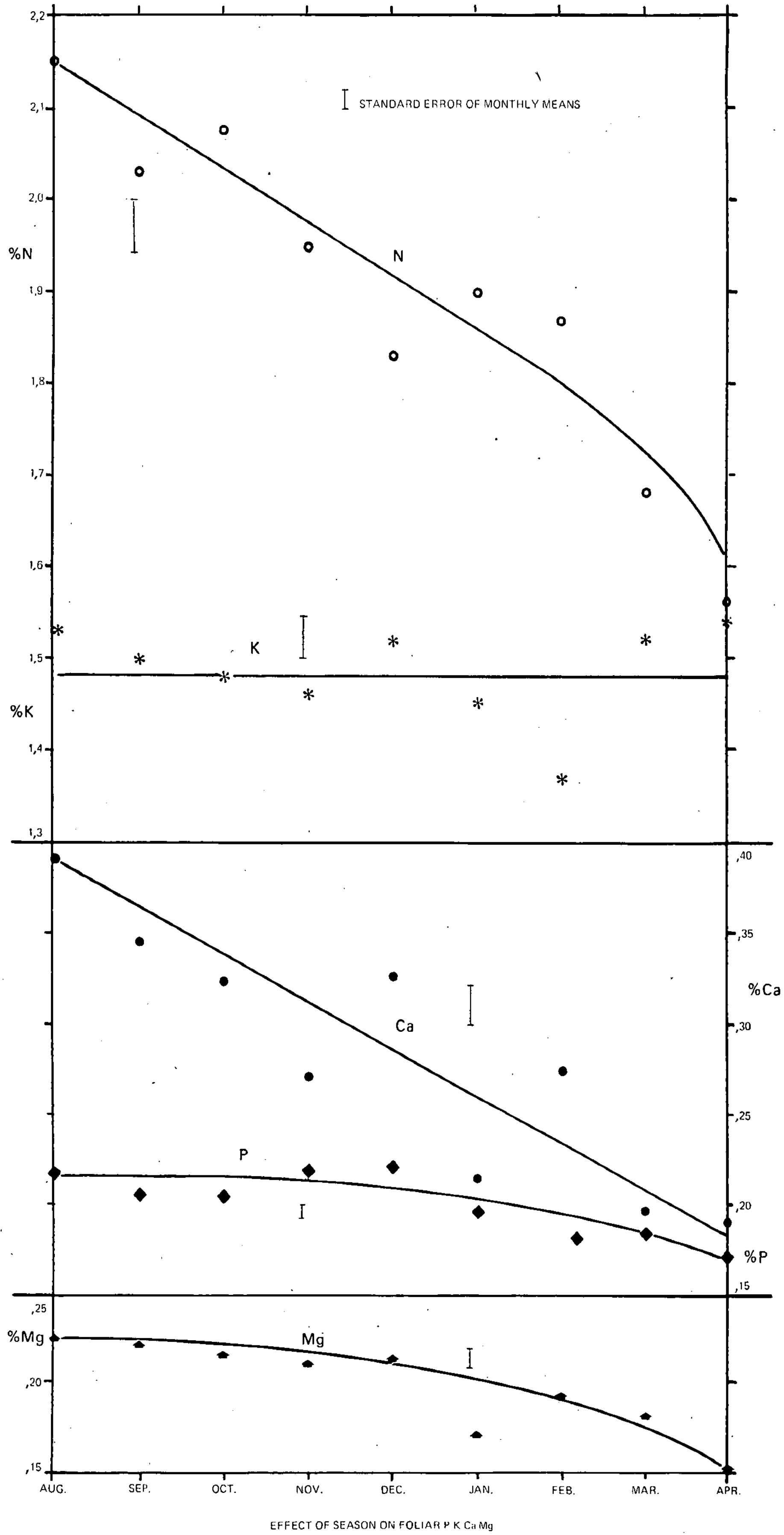

FIGURE 3: Effect of season on foliar N, P, K, Ca, Mg.

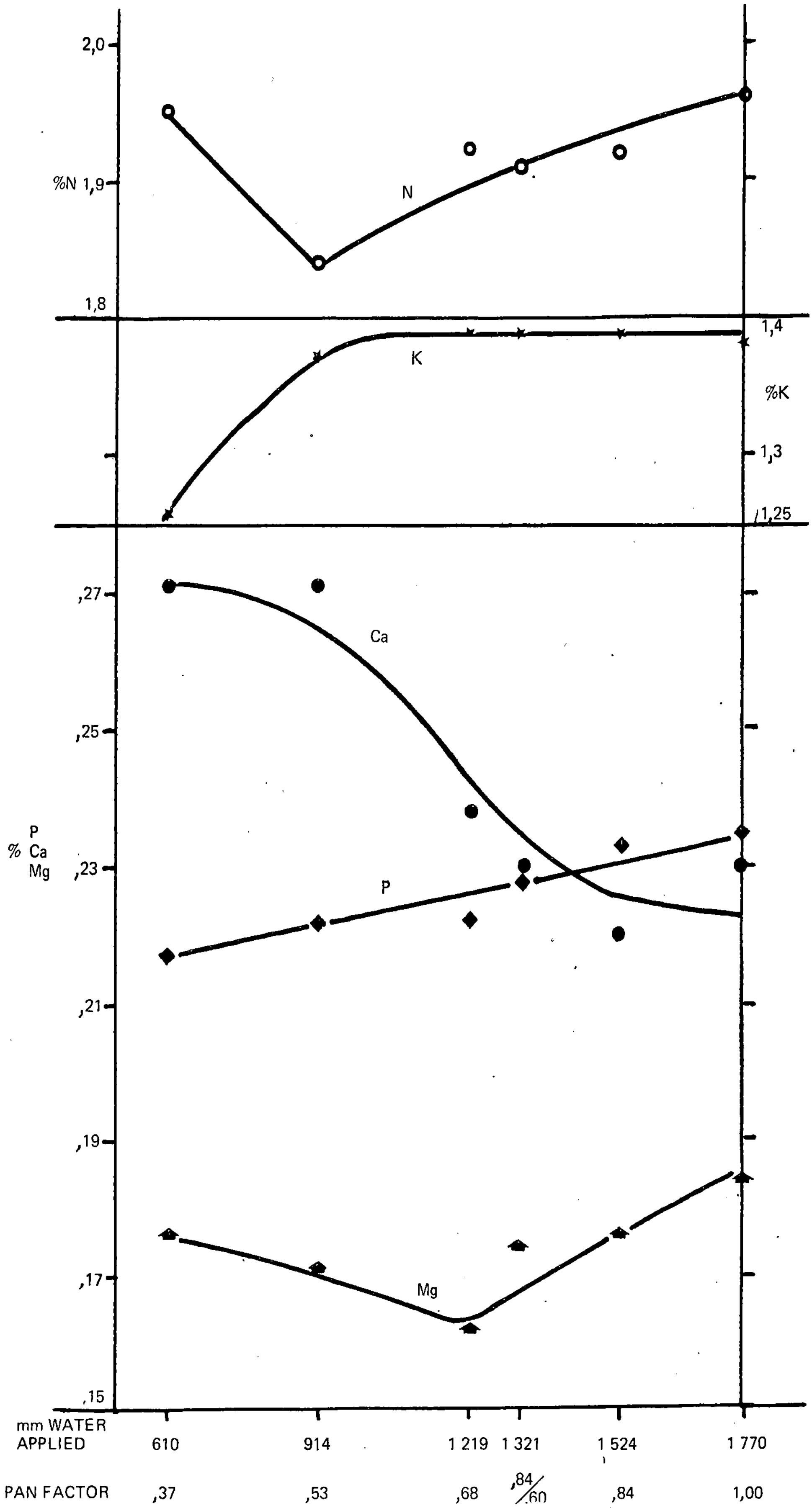

FIGURE 4: Effect of irrigation on foliar N, P, K, Ca, Mg.

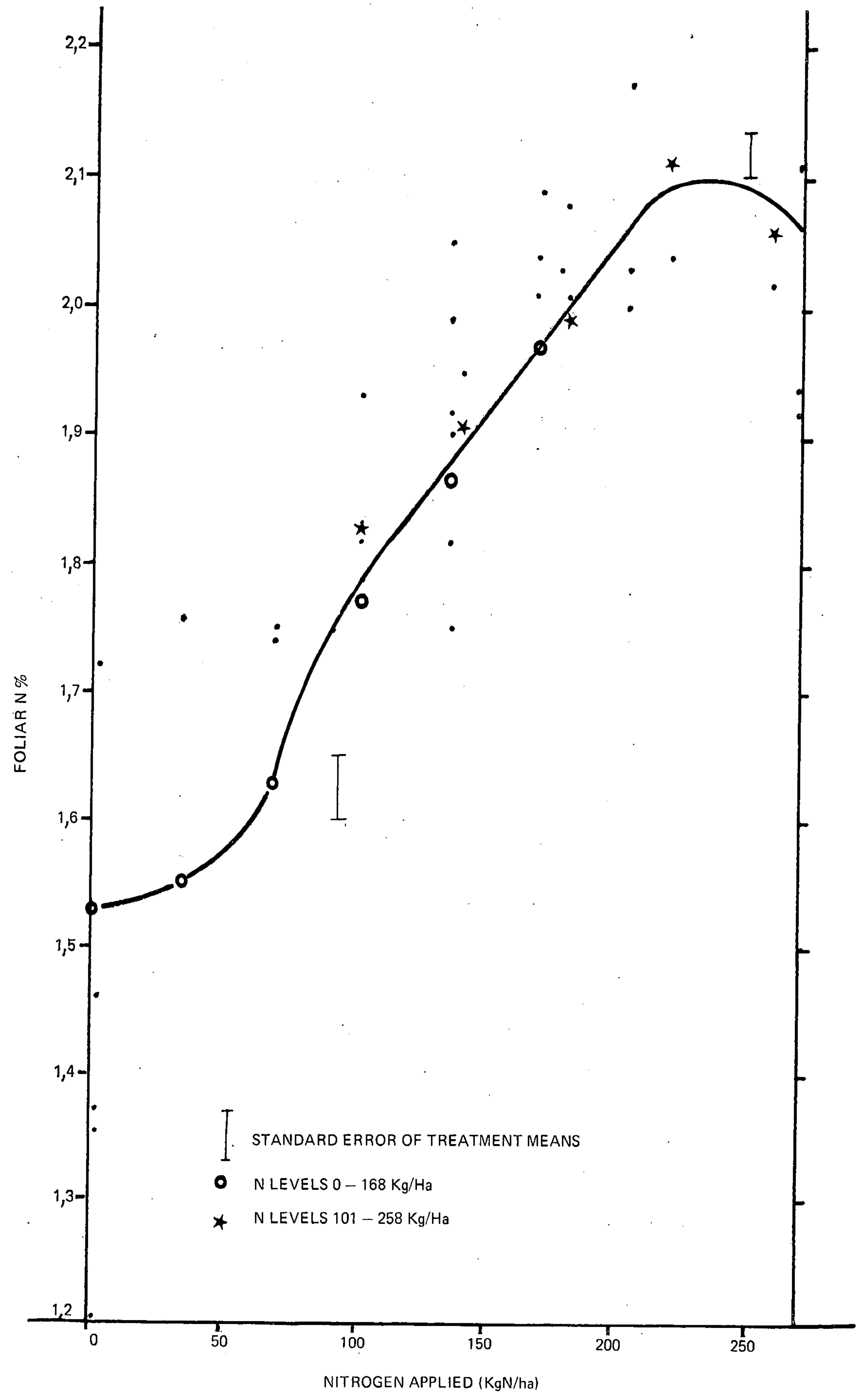

FIGURE 5: Effect of nitrogen applied on foliar N.

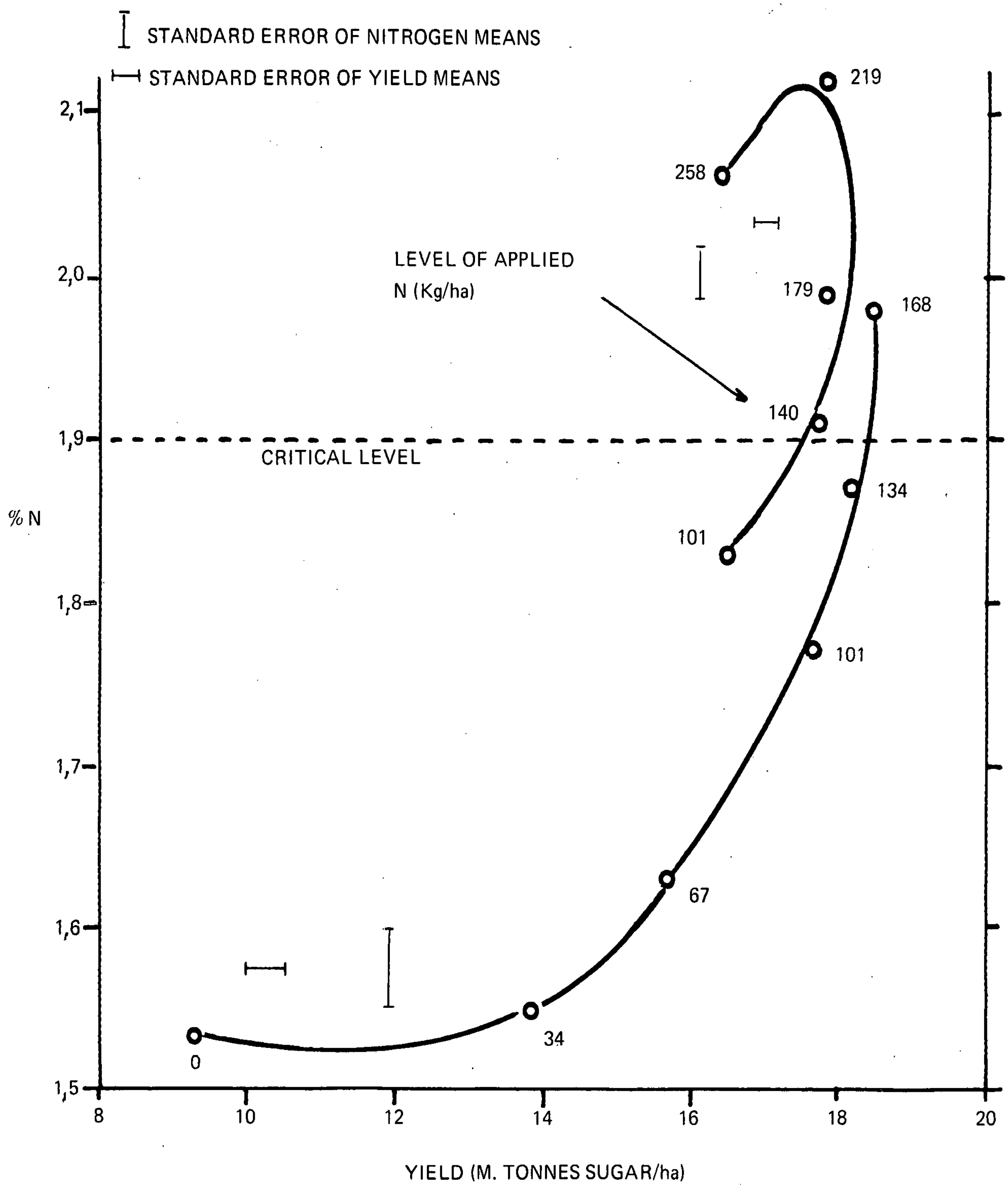

FIGURE 6: Relationship between foliar N and sugar yield.

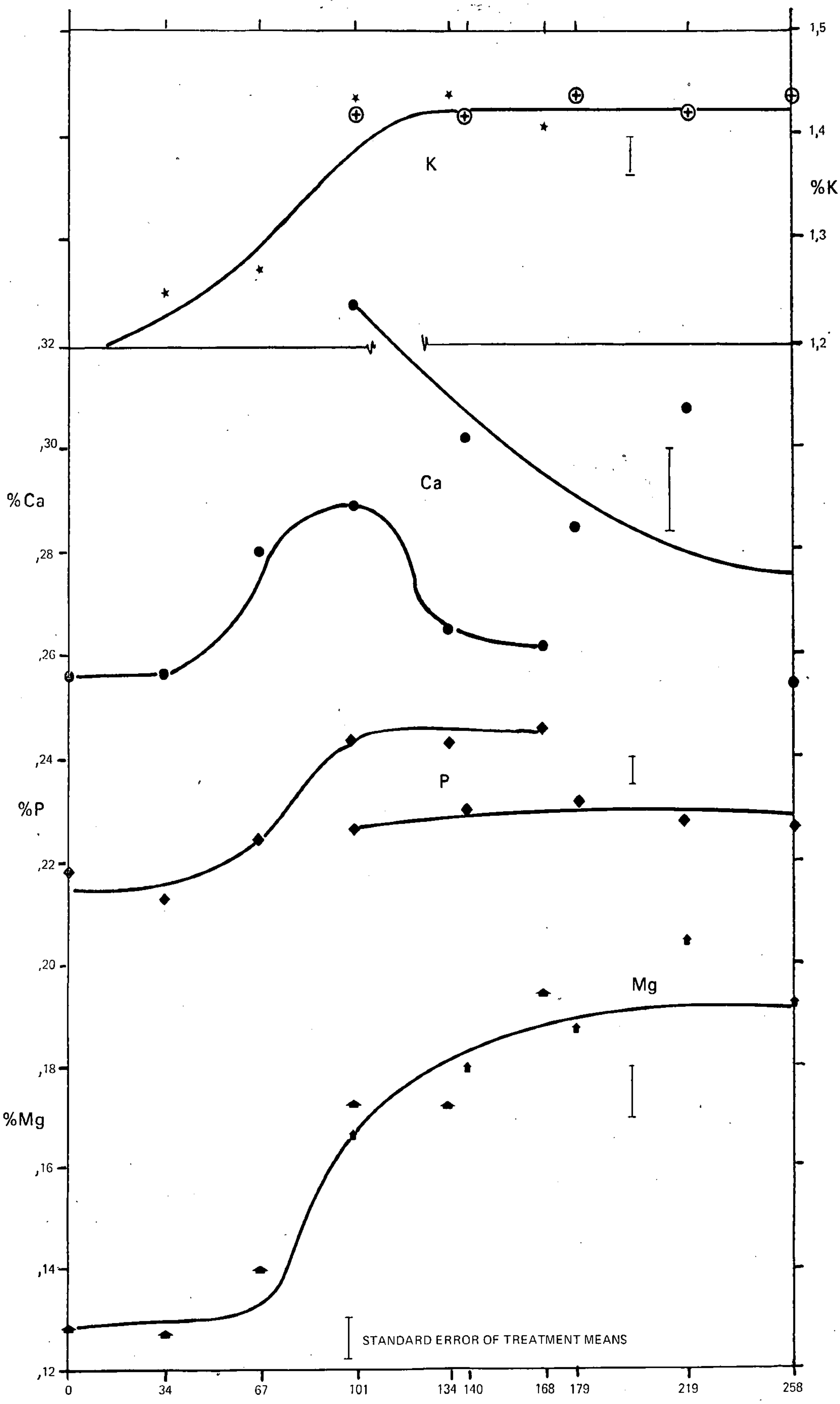

FIGURE 7: Effect of nitrogen applied on foliar P, K, Ca, Mg.

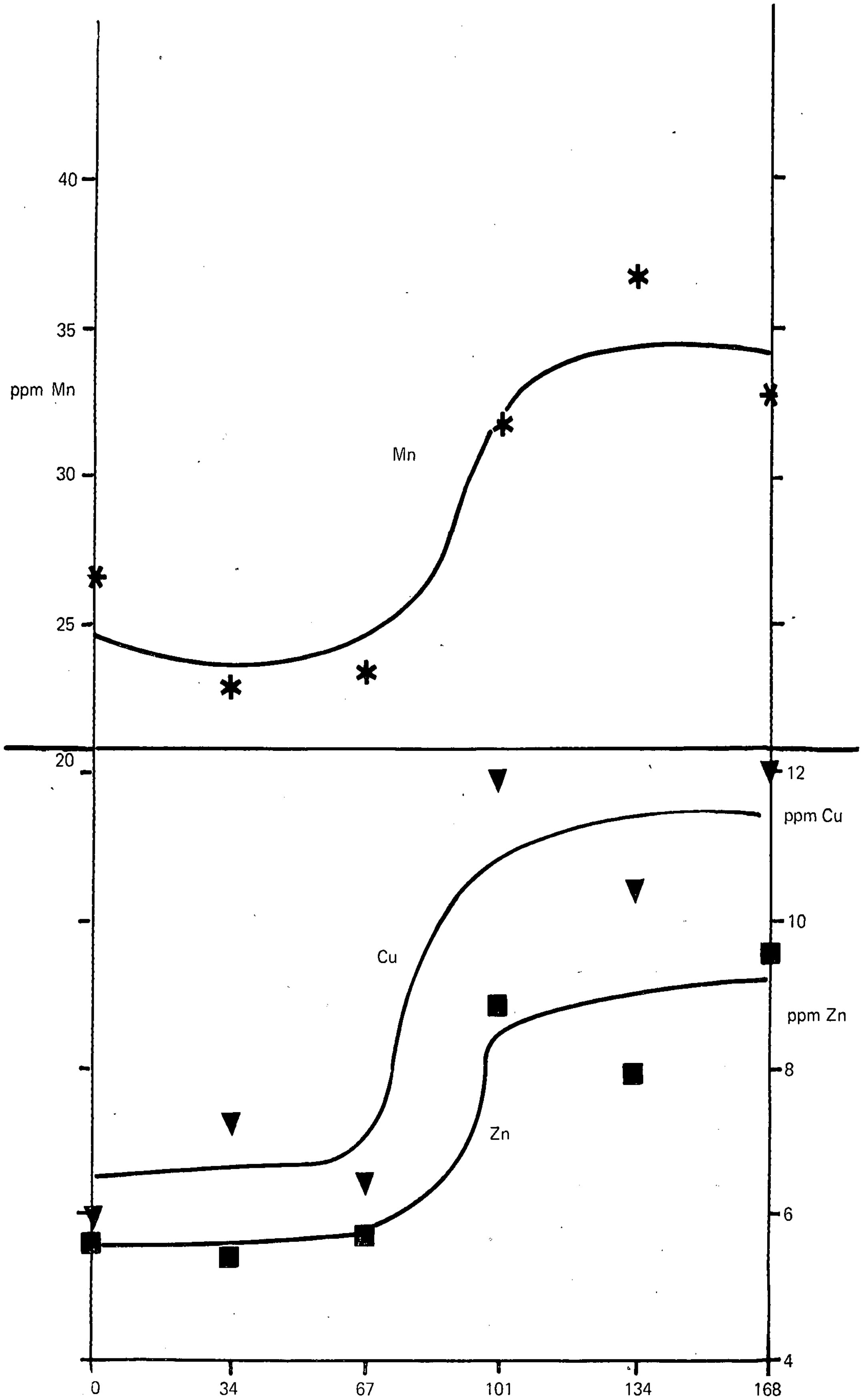

FIGURE 8: Effect of nitrogen level on foliar Cu, Zn, Mn.

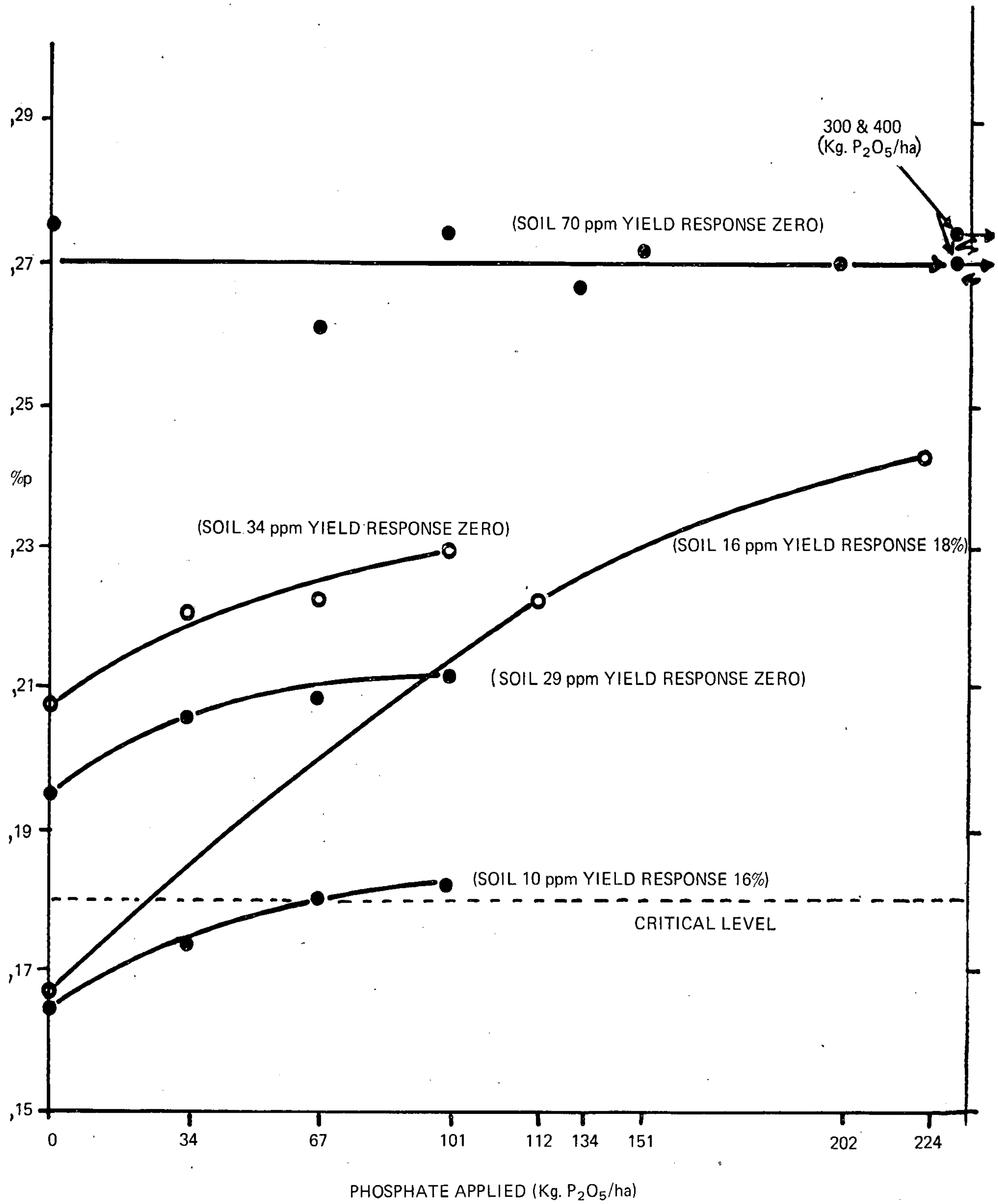

FIGURE 9: Effect of phosphate applied on foliar P.

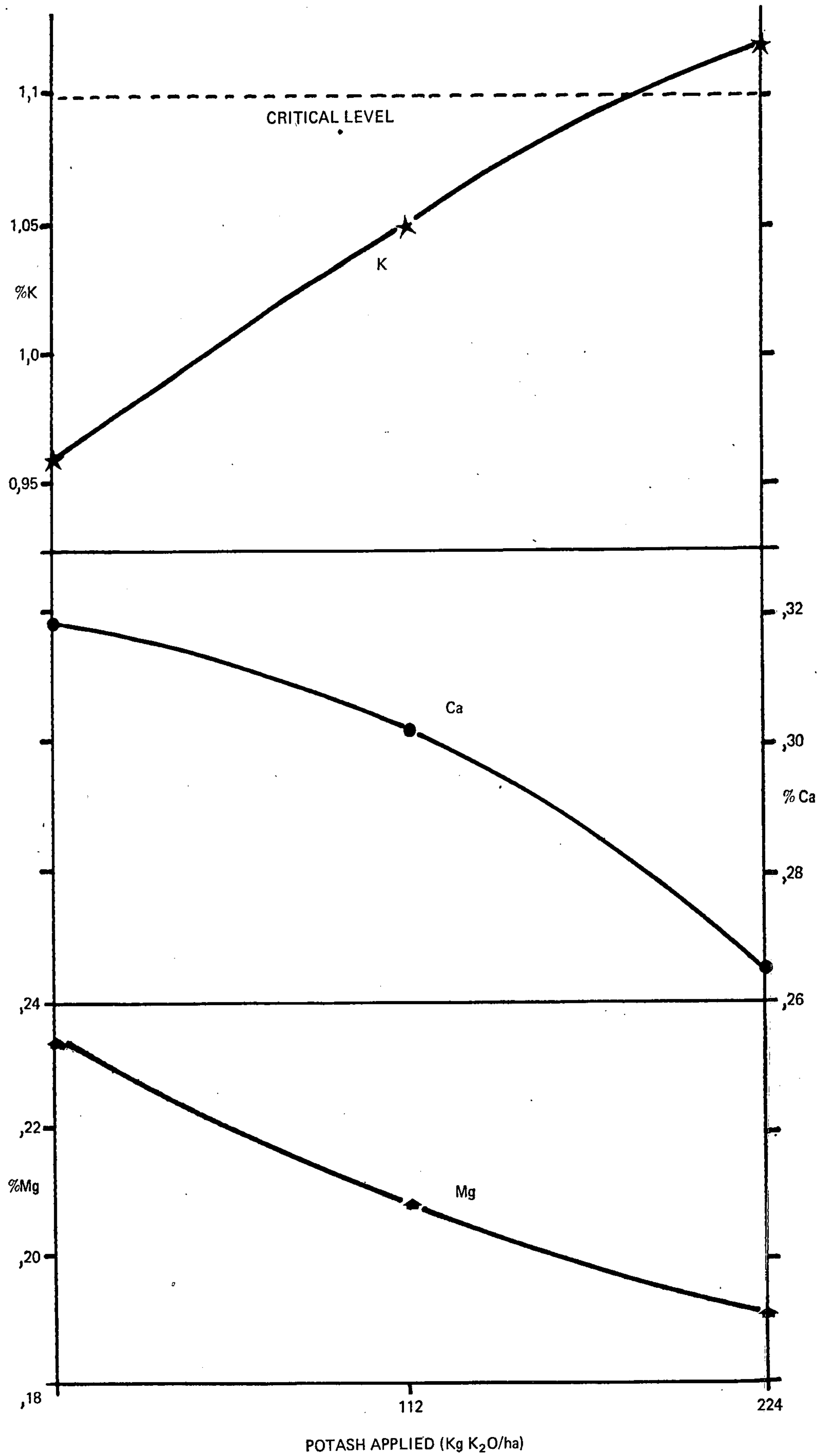

FIGURE 10: Effect of potash applied on foliar cations.