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## Nutrient limitation in the Chesapeake Bay : nutrient bioassays in the Virginia Bay system: final report

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NUTRIENT LIMITATION IN THE CHESAPEAKE BAY:  
NUTRIENT BIOASSAYS IN THE VIRGINIA BAY SYSTEM

FINAL REPORT

to

VIRGINIA COASTAL RESOURCES MANAGEMENT PROGRAM  
(Grant No. NA17OZ0359-01)

by

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and

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March 1993



## EXECUTIVE SUMMARY

Nutrient enrichment bioassays were conducted on water samples collected from six stations in the Virginia portion of the Chesapeake Bay system on a monthly basis over a year. Two stations were located in the tidal freshwater portions of the Rappahannock and James Rivers, at the mouths of these rivers and in the mainstem of the Bay. The purpose of the experiments was to determine the spatial and temporal pattern of nutrient limitation of phytoplankton growth.

Phytoplankton at the tidal freshwater stations appear to be light-limited throughout the year, probably a result of the high turbidity characteristic of these waters. Reduced nutrient uptake by the light-limited phytoplankton combined with the proximity of these stations to nutrient-rich, freshwater inputs results in high concentrations of dissolved inorganic nitrogen and phosphorus. To the extent that nutrient limitation is expressed at these stations, it is a weak phosphorus limitation, which is consistent with the observation of dissolved N:P ratios in excess of 30:1.

The two mainstem bay stations and the station at the mouth of the Rappahannock River exhibited a similar pattern of limitation characterized by nitrogen limitation throughout the year except for a period of phosphorus limitation during March-May, a time period that might be expected to coincide with the spring bloom. This pattern of sequential P and N limitation is consistent with observations in the mainstem Maryland portion of the Bay (Fisher et al., 1992) and the lower York River (Webb, 1988).

The station in mouth of the James river was strongly nitrogen limited throughout the year with no indication of phosphorus limitation. Two possible explanations for the lack of a spring period of P limitation are local conditions which maintain an adequate year round supply of phosphorus or the relative proximity of the station to marine waters of the continental shelf which are considered to be N limited.

A comparison of enrichment bioassay indexes to the ratios of dissolved inorganic N and P for all stations were in general agreement, suggesting that short term bioassays are suitable indicators of longer term processes which drive nutrient limitation. Thus, patterns of nutrient limitation seem to be driven primarily by the ratio of dissolved inorganic N:P in the Bay-tributary system. Such ratios are typically in excess of 30 throughout the year in the tidal freshwater regions and decrease to a range of 10-5:1 in the lower tributaries and mainstem Bay stations. The processes responsible for this change are not known.

## ACKNOWLEDGEMENTS

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## INTRODUCTION

Eutrophication resulting from anthropogenic inputs of nutrients is increasingly recognized as having a deleterious impact on the water quality of the Chesapeake Bay and its tributaries (US EPA 1982). Potential or actual effects from over-enrichment with nitrogen (N) and phosphorus (P) include: 1. a change in the phytoplankton species constituting the base of the food web with possible implications for the nature of the food web and harvestable resources (Verity 1988); 2. a decline in submerged aquatic vegetation resulting from reduced submarine irradiance caused by increased phytoplankton and epiphyte abundance (Orth and Moore 1983); 3. the exacerbation of hypoxia and anoxia in subpycnocline waters of the Bay resulting from increased phytoplankton abundance which is transported to the bottom waters and sediments and depletes oxygen during decomposition (Malone et al. 1986, 1988).

Nutrient inputs to the Bay vary significantly in both time and space and with respect to their nature and source (Magnien et al. 1982), and given the size of the Bay and its hydrographic complexity it is difficult to clearly determine where and when phytoplankton are being limited by nutrients and which nutrients are the primary cause of the limitation. For remediation efforts to be successful and cost effective it is necessary to have a better understanding of the nutrients which limit phytoplankton growth or abundance and how this limitation changes temporally and spatially within the Chesapeake Bay.

The purpose of the research reported here is to extend the investigations of the role of N and P in controlling the abundance of algal biomass in the Virginia portion of the Bay system beyond the initial studies conducted in the lower York River (Webb 1988). Toward this end, nutrient enrichment bioassays were conducted monthly on water samples collected from six sites in the Bay and its lower tributaries and which represent a diverse range of hydrodynamic, chemical and ecological conditions.

## METHODS

Six stations were sampled monthly, except for December, for a year. Two of the stations are in the tidal freshwater portions of the James and Rappahannock Rivers (TF), two are located near the mouths of these same tributaries (LE), and two are located in the mainstem of the Virginia portion of the Bay (CB) (see Fig. 1, Table I). All of the stations, except TF3.2, coincide with stations routinely sampled for the Chesapeake Bay Program phytoplankton monitoring program. Station TF3.2 is routinely sampled as part of the nutrient monitoring program. Samples (ca. 20 L) were collected from 0.5 m depth by the same personnel and using the same protocol as for the ongoing monitoring programs. The TF stations were sampled from March 1992 through February 1993, while the remaining stations were sampled from February 1992 through January 1993.

Samples from stations TF3.2, TF5.5, LE3.6 and CB6.1 normally were transported to Gloucester Point within 2-3 hours of collection. Samples from LE5.5 and CB6.4 were stored overnight in a cool, dark environment and transported to Gloucester Point the morning after collection.

Upon arrival at Gloucester Point, subsamples from each water sample were taken for the following analyses: chlorophyll "a" (fluorometric determination following acetone/DMSO extraction, Webb and Hayward, unpublished manuscript); dissolved nitrate ( $\text{NO}_3$ ), nitrite ( $\text{NO}_2$ ), reactive phosphate ( $\text{PO}_4$ , all Parsons et al., 1984) and ammonia ( $\text{NH}_4$ , Solorzano, 1969); particulate organic nitrogen and carbon (PON, POC, Perkin Elmer 240 elemental analyzer, Perkin Elmer, 1981); and particulate organic phosphorus (POP, Solorzano and Sharp, 1980). The remaining water was then filtered through 90  $\mu\text{m}$  Nitex to remove larger grazers and subdivided into paired, 1l, clear polycarbonate bottles. Each pair then received one of the following enrichment treatments: +N (25  $\mu\text{M}$   $\text{NH}_4$  as  $\text{NH}_4\text{Cl}$ ); +P (5  $\mu\text{M}$   $\text{PO}_4$  as  $\text{NaH}_2\text{PO}_4$ ); +Si (30  $\mu\text{M}$   $\text{SiO}_4$  as  $\text{NaSiO}_3$ ); +N+P; +N+P+Si. One pair of bottles received no nutrient additions and served as a control. The two mainstem stations (CB) and the two river mouth stations (LE) did not receive either the +Si or +N+P+Si treatment from July through November. The bottles were then placed in a shallow (ca. 50 cm water depth), flow-through water table (ambient York River water). The bath was kept covered with a plastic screening to provide a 50% reduction in ambient irradiance. The bottles were sampled in the morning every day for chlorophyll "a" and every other day for POC, PON and POP. When a maximum response of the phytoplankton to enrichment was reached (based on chlorophyll "a" and taking 4-8 days depending on time of year) the incubations were terminated. In those instances when an incubation was anticipated to last more than 4 days, no chlorophyll "a" sample was collected on day 1 of the incubation.

For each incubation, the phytoplankton response to enrichment was quantified as an enrichment index which was derived by dividing the phytoplankton biomass as chlorophyll "a" or particulate C, N or P on the day of peak response by the value of the control on that same day. To simplify this determination, the day of peak response was considered to be the same for all treatments for a particular incubation. An enrichment index  $>1$  indicates an enhancement of phytoplankton biomass relative to the control for a particular nutrient and thus is an indication of a limiting nutrient. An enrichment index of  $<1$  indicates a reduced phytoplankton biomass relative to the control and thus no limitation by that nutrient. A value of 1 indicates no difference with regard to the control and thus no limitation.

## RESULTS AND DISCUSSION

All required water samples were collected and transported to Gloucester Point. One water sample (LE5.5, January 1993) was lost before it could be analyzed.

Chlorophyll concentrations observed at the six stations are shown in Table II and Fig 2A & B. The highest concentrations were observed at Station TF5.5 which was characterized by a prolonged summer bloom with chlorophyll in excess of 75 ug/l from July through October. Chlorophyll values at TF3.2 generally ranged between 10-30 ug/l with a maximum concentration of 40 ug/l observed in August. Chlorophyll concentrations were similar at stations CB6.1, CB6.4 and LE3.6. and were generally lowest in February 1992 (ca. 5 ug/l) and increased to a maximum of 22-27 ug/l in October (Fig. 2B). Chlorophyll concentrations at station LE5.5 showed a greater degree of temporal variability although the annual average (12.9 ug/l) was not appreciably greater than the annual average for the other lower river and bay stations (10.1-11.8 ug/l).

The concentrations of inorganic nutrients at each station are shown in Table III and Figs. 3A-E. In general, concentrations of all nutrients were greater at the two tidal freshwater stations than at the other stations, ranging from 20-70  $\mu\text{M}$  for nitrate, 5-30  $\mu\text{M}$  for ammonia, and 0.7-1.8  $\mu\text{M}$  for phosphate. Ammonia was generally greater than nitrate at TF5.5, while at TF3.2 the reverse was true. However, total dissolved inorganic nitrogen (DIN =  $\text{NO}_3 + \text{NO}_2 + \text{NH}_4$ ) was essentially the same at both stations and markedly greater in the freshwater stations than at the lower river and bay stations (Fig. 3E). There appeared to be no consistent or obvious seasonal trends for nutrients at these stations except for nitrate at TF3.3 which was greatest in winter-spring and least in summer. Nitrate values at both CB and LE stations were consistently less than 10  $\mu\text{M}$  with higher concentrations in the spring and lower concentrations in the summer (Fig. 3A). Ammonia concentrations at these four stations were, with one exception, less than 4  $\mu\text{M}$  with values greatest in February 1992 and decreasing gradually for the remainder of the study (Fig 3B). Phosphate concentrations at the two CB stations and at LE3.6 were low in February-March, increased in April and remained constant for the remainder of the year in the range of 0.4-0.6  $\mu\text{M}$  (Fig. 3D). Phosphate at LE5.5 was similar to that of the bay and lower river stations from February through June but then increased substantially for the remainder of the year. An N:P ratio was calculated as the ratio of DIN:PO<sub>4</sub> and the values are shown in Table III.

The results of the bioassay incubations are shown in Figs. 4-15. Enrichment indexes were calculated for each enrichment



treatment at each station and are shown in Table IV and Figs. 16A-F. As a general rule, and with the exception of Si enrichment, all treatments at the CB and LE stations elicited much greater response, compared to the controls, than at the TF stations (note change of scale for TF enrichment indexes compared to other stations). There was little if any response to Si enrichment at any of the six stations. The greatest positive response observed for Si (ca. 20% greater than control) was in October at the two TF stations and even then the single addition of N or P elicited an equal or greater response than observed for Si. Silica added in combination with N and P generally did not elicit a greater response than N and P combined. These results indicate that Si is not a limiting nutrient at any of the stations during the time when Si enrichments were used.

For the most part the addition of N and P together elicited a greater response than N and P added singly. This suggests a relatively close balance between inorganic P and N, with respect to the Redfield proportion (see below), such that when the limiting nutrient is added as a single enrichment, the non-limiting nutrient becomes limiting and retards the growth of the phytoplankton. When both nutrients are added and there is no opportunity for limitation by either nutrient, then biomass accumulation can be expected to exceed that of either nutrient added singly. Fisher et al., (1992) refer to this as primary N or P limitation as compared to exclusive N or P limitation in which the effect of a single added nutrient equals or exceeds that of both nutrients added simultaneously. In only one instance was there continuing, exclusive limitation and it was for N at LE5.5 from August to November (Fig 16D).

Despite the small magnitude of response to enrichment observed at both of the TF stations, in 14 of 22 observations the response to P was greater than the response to N. On only three occasions was the response to N clearly greater than the response to P and they occurred in August-November once at TF3.2 and twice at TF5.5. There appears to be a weak limitation by phosphorus that persists throughout the year at both the tidal freshwater stations with a possibility of N limitation in the fall.

At station LE5.5 there was no response to P addition during the year, while there was a consistent, large response to nitrogen enrichment (annual mean enrichment index = 2.31, n=10). As mentioned previously, for four months in the late summer-fall, the N limitation was characterized as exclusive. This is clear evidence of a persistent and strong limitation by nitrogen at this station.

The response to N and P enrichment was similar at the remaining three stations (CB6.1, CB6.4 and LE3.6) and was characterized by either a response to N or no response to either nutrient in February-March, a response to P in April-May and a response to N greater than the response to P for the remainder of the calendar year. Thus all three stations appear to shift to P

limitation during the period of the spring bloom but are strongly to weakly N limited during the remainder of the year.

Both tidal freshwater stations exhibit a weak but generally consistent pattern of P limitation throughout the year. The weakness of the enrichment response is consistent with both the persistently high concentrations of PO<sub>4</sub> and DIN at both these stations and with the prevailing belief that freshwater systems are generally P limited (Hecky and Kilham 1988; Howarth 1988). The reduced response to enrichment at these stations is also consistent with the location of these stations at or near the turbidity maximum region of the respective rivers. Our observations indicated water samples from these stations typically contained high concentrations of detrital-particulate material which hampered filtration. Given the high turbidity and lack of vertical stratification (the absence of salt) in these regions, it is likely that phytoplankton are light-limited and are not able to make full use of available nutrients. Evidence for light limitation can be found in the bioassay results from the TF stations in which all 22 enrichment experiments resulted in significant and persistent increases in the chlorophyll content of the unenriched controls during the 4-8 days of incubation (see Figs. 5-15A&B). Both the relatively high light level of the water bath, even with the 50% attenuation screening, and the rapid settling of particles in the incubation bottles while in the water bath, exposed the suspended phytoplankton to sufficiently increased light level that substantial phytoplankton growth was possible even in the absence of added nutrients. Increases in control chlorophylls at the other four stations were the exception rather than the rule (only 10 of 43 incubations) and occurred primarily in November and January when shorter days and increased vertical mixing due to wind and decreased thermal input might be expected to reduce light availability to the phytoplankton.

The similarity of stations CB6.4, CB6.1 and LE3.6 in terms of nutrients, chlorophyll and enrichment index suggests a commonality of function for these three stations. While this might be expected for the two CB stations, it is less expected for the lower Rappahannock River station and suggests that this station is more influenced by bay conditions than by river conditions on the upestuary side. The observation that all three stations are nitrogen limited except for a short but pronounced period of P limitation during the spring is consistent with the results of Webb (1988) working at a station in the lower York River at Gloucester Point. Based on nutrient bioassays conducted in 50l microcosms he determined that at this station, which ranged in salinity from 12 to 24.5 ps during an 18-month period, the phytoplankton were nitrogen limited for most of the year except during the late winter-early spring (February-April) when P limitation replaced N limitation. Our results are also consistent with the observations of D'Elia et al. (1986) using similar nutrient-enrichment microcosms to those of Webb (1988) in

the Patuxent estuary over a 13 month period (salinity 3.2-10.5 psu). They observed P limitation in the fall and winter (October-February) and N limitation in the late spring and summer (June-September).

Webb(1988) has proposed that these observations of varying indices of limitation over salinity gradients were consistent with a pattern of persistent P limitation in tidal freshwater which progresses down tributary during the fall and winter reaching its most downstream extent in late spring. This limitation is in turn displaced by N limitation up estuary in the late spring and summer. He proposed that the seasonal cycle of PO<sub>4</sub> abundance in estuaries (high in summer and low in winter) and a large input of non-point source nutrients with a high N:P ratio from winter-spring runoff as the main cause of this pattern. Our observation of a short period of P limitation at station LE3.6 is consistent with this scenario. Our observation of a concurrent period of P limitation at the two mainstem stations extends the downestuary phenomenon of P limitation from the tributary mouths to the mainstem of the Bay, and confirms recently published work by Fisher et al. (1992) in which several different indices of nutrient limitation, including 31 nutrient enrichment bioassays, indicated that phytoplankton accumulation in the mainstem of the Maryland portion of the Bay was sequentially limited by P in the spring and N in the summer.

Station LE5.5 at the James River mouth does not follow the same temporal pattern of limitation exhibited by the other river mouth and lower bay stations in that this station was consistently N limited. It is possible that local conditions of nutrient inputs and processing in this region of the estuary have created a situation which ameliorates the relative lack of phosphorus during the spring season and maintains N limitation throughout the year. Another possibility is that because of the proximity of this station to the Bay mouth, it is sufficiently influenced by those conditions which act to maintain marine systems under persistent N limitation (Howarth 1988)

While indications of nutrient limitation can be derived from short term enrichment bioassays it is really the in situ nutrient conditions which dictate the physiological state of the phytoplankton. A criticism of the enrichment bioassay is that the small temporal and spatial scales of the experiments (liters of water incubated over a few days usually once a month) is not compatible with environmental (e.g. nutrient concentrations) and biological (e.g. phytoplankton growth and accumulation) processes which constitute limitation and are typically governed at much larger time and space scales. One approach to resolving these differences in scale is to relate the short term bioassay responses to processes governed at larger scales such as nutrient concentrations.

Actively growing phytoplankton contain N and P in approximately a 16:1 atomic ratio (i. e. Redfield proportion).

If the dissolved inorganic N:P ratio is less than 16:1 then nitrogen is in short supply relative to phosphorus and nitrogen will limit any increase in phytoplankton biomass. If the N:P ratio is greater than 16:1 then phosphorus may be expected to ultimately limit any increase in phytoplankton biomass and thus it is limiting. In order to relate nutrient concentrations to the bioassay results, we plotted the inorganic N:P ratio for each station against the ratio of the N enrichment index:P enrichment index (Table III, Fig. 17) If the value of this latter index is  $> 1$ , it indicates that the enrichment index for nitrogen was greater than for phosphorus at that station and that N is the limiting nutrient. If the ratio is  $< 1$  then the P enrichment index is greater than the N enrichment index and P is the limiting nutrient.

If smaller scale enrichment bioassay results are compatible with the larger scale nutrient data, one can expect phytoplankton in an environment with a nutrient ratio  $>16:1$  to exhibit an enrichment index ratio of  $<1$ , since both are indications of P limitation. Conversely, phytoplankton taken from an environment with a nutrient ratio of  $<16:1$  should be expected to show an enrichment index ratio  $>1$ , since both are an indication of N limitation. Inspection of Fig. 17 indicates that 81% (50 out of 62) paired observations fall in the appropriate quadrant. That is, samples with N:P ratios  $>16$  generally have an enrichment index ratios  $<1$  (quadrant III), while samples with N:P ratios  $<16$  generally have index ratios  $>1$  (quadrant I). Three bioassay could not be used since there was not available nutrient data for them. Of the eight data points with N:P ratios  $>16$  which fall into the "wrong" quadrant, six occur at stations CB and LE during February and March. A review of the data for these six samples indicates that both the individual N and P enrichment indexes and DIN values appear to be appropriate for the time and place of the sample. However, the phosphate concentrations at these stations was low for the two months in question (mean = 0.05  $\mu\text{M}$ ,  $n = 8$ , Fig. 3D) which resulted in a high N:P ratio and placed these points in quadrant II instead of quadrant I. Of the seven lower estuary and bay samples which exhibit P limitation (enrichment index ratio  $<1$ ), only three exhibit the appropriate N:P nutrient ratio (i.e.  $<16$  and occur in quadrant III). That more do not fall into the appropriate quadrant is perhaps not surprising considering the short-lived nature of P limitation at these stations. In general, the comparisons of enrichment indexes and inorganic nutrient ratios are in good agreement and support the conclusion that the results of short term enrichment bioassays reflect the effects of standing stock nutrient ratios on phytoplankton physiology and are valid indicators of nutrient limitation.

The data presented in Fig. 17 also illustrates the differences in nutrient patterns among the various stations. For example, despite the difference in the dominant form of DIN (ammonia at TF5.5 and nitrate at TF3.2) both stations consistently exhibit N:P ratios greater than ca 30:1, which

considerably exceed the Redfield ratio. Considering the position of these stations at the head of their respective estuaries, it indicates that the nutrient loadings from freshwater input are typically high in dissolved N compared to dissolved P, an observation that is consistent with other observations in the Bay region (Fisher et al., 1992; Magnien, et al., 1992). However, as one moves down estuary the N:P nutrient ratio decreases significantly, generally to below 10:1 and in many instances to below 5:1. Apparently biogeochemical processing along the estuarine gradient is depleting the dissolved N pool to a greater extent than the dissolved P pool. Alternatively, or in addition, biogeochemical processes in the lower estuary could be enriching inorganic P relative to N. Such processes could include denitrification and the mobilization of phosphate from the sediments to the water column during summer anoxia. In any case the N:P ratios are lower, and for the most part the appropriate limitation response is obtained.

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## TABLES

- I. Station location and description.
- II. Station chlorophyll "a" (ug/l).
- III. Station nutrient concentrations (uM), N:P molar ratios and N:P enrichment index ratio.
- IV. Enrichment indexes.



Table I. Station location and description

Station	Latitude	Longitude	River	Salinity
	DD MM SS	DD MM SS		
TF 5.5	37 18 46	77 13 59	James	Fresh
LE 5.5	36 59 48	76 18 12	James	Mesohaline
TF 3.2			Rappa	Fresh
LE 3.6	37 35 48	76 18 12	Rappa	Mesohaline
CB 6.1	37 35 18	76 09 45	Chesa	Mesohaline
CB 6.4	37 14 11	76 12 30	Chesa	Polyhaline

Table II. Station chlorophyll "a" (ug/l).

Station	FEB	MAR	APR	MAY	JUNE	JUL	AUG	SEP	OCT	NOV	JAN93	FEB	AVCHL
CB6.1	3.5	6.3	5.5	10	8.4	8.83	12.53	13.2	24.45	7.04	14.67		10.40
CB6.4	6.24	7.4	8.64	8.67	9.6	12.96	13.2	8.93	26.61	10.93	16.27		11.77
LE3.6	4.83	5.97	8.8	9.87	10.27	11.01	14.08	16.35	21.12	6.77	1.65		10.07
LE5.5	24.53	9.87	5.65	8.8	21.33	12.85	12.53	19.23	7.52	6.51			12.88
TF3.2		8.53	11.47	23.47	10.93	23.2	40	19.87	10.61	5.15	2.51	5.01	14.61
TF5.5		5.87	15.2	37.6	17.2	84.27	100.53	78.93	80.72	49.87	4.88	4.83	43.63

Table III. Station nutrient concentrations ( $\mu\text{M}$ ), N:P molar ratios and N:P enrichment index ratio.

station	month	NH4 $\mu\text{M}$	PO4 $\mu\text{M}$	NO2 $\mu\text{M}$	NO3 $\mu\text{M}$	T_DIN	N:P	N:P index
CB6.1	FEB	2.03	0	0.24	1.89	4.16		0.82
CB6.4		3.12	0.04	0.12	0.28	3.52	88	2.11
LE3.6		2.78	0.05	0.24	1.4	4.42	88.4	0.94
LE5.5		3.05	0.1	0.18	0.15	3.38	33.8	1.85
CB6.1	MAR	1.49	0.02	0.23	1.14	2.86	143.00	1.22
CB6.4		1.89	0.06	0.12	0.2	2.21	36.83	3.31
LE3.6		2.34	0.12	0.17	1.03	3.54	29.50	1.26
LE5.5		1.53	0.03	0.2	0.52	2.25	75.00	2.82
TF3.2		13.8	0.91	0.75	68.91	83.46	91.71	0.76
TF5.5		15.57	1.4	0.61	30.85	47.03	33.59	0.96
CB6.1	APR	1.45	0.3	0.31	6.33	8.09	26.97	0.44
CB6.4		1.13	0.34	0.11	0.21	1.45	4.26	1.80
LE3.6		3.68	0.45	0.24	4.73	8.65	19.22	0.78
LE5.5		1.95	0.38	0.13	0.58	2.66	7.00	3.19
TF3.2		8.84	0.89	0.42	47.97	57.23	64.30	0.67
TF5.5		26.97	1.39	0.79	26.05	53.81	38.71	0.92
CB6.1	MAY	1.79	0.42	0.32	5.1	7.21	17.17	0.70
CB6.4		1.79	0.41	0.24	3.76	5.79	14.12	0.59
LE3.6		0.93	0.38	0.48	9.87	11.28	29.68	0.47
LE5.5		3.07	0.5	0.38	2.88	6.33	12.66	2.52
TF3.2		3.39	0.87	0.47	47.36	51.22	58.87	0.91
TF5.5		10.92	1.04	1.51	40.36	52.79	50.76	1.02
CB6.1	JUN	1.36	0.39	0.09	0.1	1.55	3.97	2.08
CB6.4		1.6	0.37	0.18	0.2	1.98	5.35	4.62
LE3.6		1.33	0.41	0.18	1.53	3.04	7.41	1.27
LE5.5		0.83	0.5	0.41	3.87	5.11	10.22	2.64

TF3.2		11.79	0.86	0.63	30.68	43.1	50.12	0.79
TF5.5		17.81	1.36	1.19	26.11	45.11	33.17	0.94
CB6.1	JUL	0.84	0.49	0.38	0.24	1.46	2.98	2.24
CB6.4		0.27	0.68	0.03	0.08	0.38	0.56	8.54
LE3.6		0.15	0.62	0.08	0.16	0.39	0.63	2.35
LE5.5		0.33	0.9	0.2	0.8	1.33	1.48	2.64
TF3.2		7.19	0.73	0.68	35.39	43.26	59.26	0.67
TF5.5		19.69	0.95	2.53	20.59	42.81	45.06	0.92
CB6.1	AUG	0.84	0.49	0	0.37	1.21	2.47	0.95
CB6.4		0.73	0.49	0	0.18	0.91	1.86	2.10
LE3.6		1.15	0.47	0.05	0.28	1.48	3.15	2.05
LE5.5		6.25	1.24	0.4	1.85	8.5	6.85	1.55
TF3.2		3.5	0.85	0.57	21.65	25.72	30.26	0.92
TF5.5								1.19
CB6.1	SEP	0	0.48	0.44	0.2	0.64	1.33	2.23
CB6.4		1.48	0.6	0.35	0.23	2.06	3.43	1.59
LE3.6		0.11	0.46	0.11	0.24	0.46	1.00	1.73
LE5.5		0.03	0.87	0.14	0.26	0.43	0.49	3.04
TF3.2		7.63	1.13	0.57	49.78	57.98	51.31	0.86
TF5.5		4.3	1.18	2.16	32	38.46	32.59	0.98
CB6.1	OCT	0.72	0.6	0.5	0.48	1.7	2.83	2.64
CB6.4		0.49	0.52	0.13	0.16	0.78	1.50	2.79
LE3.6		0.42	0.48	2.07	8.97	11.46	23.88	1.79
LE5.5		6.91	1.75	0.16	0.33	7.4	4.23	2.24
TF3.2		4.1	1.28	0.45	56.08	60.63	47.37	0.84
TF5.5		12.21	1.21	1.99	18.61	32.81	27.12	0.83
CB6.1	NOV	1.06	0.6	0.14	1.41	2.61	4.35	1.95
CB6.4		0.59	0.75	0.23	1.3	2.12	2.83	1.22
LE3.6		0.23	0.49	0.22	1.5	1.95	3.98	4.49

LE5.5		0.28	0.67	0.21	2.64	3.13	4.67	1.43
TF3.2		4.27	1.32	0.48	37.3	42.05	31.86	1.06
TF5.5		26.45	1.77	2.14	34.3	62.89	35.53	1.17
CB6.1	JAN93	0.24	0.52	0.37	3.46	4.07	7.83	0.54
CB6.4		1.27	0.59	0.34	3.03	4.64	7.86	1.02
LE3.6		0.72	0.94	0.36	3.9	4.98	5.30	0.75
TF3.2		4.33	1.43	0.47	60.87	65.67	45.92	0.81
TF5.5		4.94	1.84	0.34	43.39	48.67	26.45	0.95
TF3.2	FEB93	34.24	1.89	0.46	35.23	69.93	37.00	0.91
TF5.5		6.7	1.17	0.44	66.18	73.32	62.67	1.02

Table IV. Enrichment indexes.

STA/TRT	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	JAN93	FEB93
TP3.2/N		0.94	1.02	1.24	1.2	1.1	0.97	1.31	1.45	1.09	0.92	1.72
P		1.24	1.52	1.57	1.52	1.64	1.06	0.86	1.73	1.03	1.13	1.88
SE		1.03	1.05	0.95	1.04	1.06	0.92	0.91	1.2	0.92	0.49	1.11
NP		1.38	1.74	1.67	1.85	1.79	1.31	1.72	2.38	1.07	1.24	2
NPSE		1.21	1.66	1.47	1.66	1.72	1.26	1.82	2	1.01	0.71	1.08
TP5.5/N		1.09	1.09	1.06	1.01	1.08	1.19	1	1.13	1.18	0.97	0.86
P		1.14	1.19	1.04	1.08	1.17	1	1.02	1.36	1.01	1.02	0.84
SE		0.89	0.97	0.93	1.04	1.11	1.04	0.98	1.16	0.97	0.72	0.74
NP		1.37	1.36	1.18	1.31	1.37	1.22	1.26	1.48	1.21	1.25	0.95
NPSE		1.26	1.42	1.13	1.08	1.39	1.21	1.28	1.55	1.04	0.98	0.77
LE3.6/N	1.06	1.7	1.31	1.23	1	2.35	1.76	1.52	1.74	5.43	1.11	
P	1.13	1.35	1.69	2.64	0.79	1	0.86	0.88	0.97	1.21	1.48	
SE	1.11	1.01	1.23	1.13	1.04						0.92	
NP	2	8.75	3.44	3.83	2.07	2.96	3.3	6.98	5.84	6.14	3.94	
NPSE	9.38	10.5	5.58	6	2.09						3.69	
LE5.5/N	2.05	3.27	2.63	2.07	2.22	3.04	1.6	2.71	1.99	1.5		
P	1.11	1.16	0.83	0.82	0.84	1.15	1.03	0.89	0.89	1.05		
SE	1.06	1.29	1.06	1.02	1.08							
NP	4.25	12.07	8.6	4.18	3.73	5.32	1.83	1.98	2.1	1.69		
NPSE	5.57	12.6	9.04	5.21	3.78							
CB6.1/N	0.84	1.04	1.01	1.34	1.85	2.31	1.06	1.63	2.43	1.46	0.84	
P	1.03	0.85	2.27	1.91	0.89	1.03	1.12	0.73	0.92	0.75	1.55	
SE	1.21	0.98	0.91	1.12	0.97						0.86	
NP	1.21	7.63	3.36	7.32	8.92	5.64	1.69	2.66	5.93	1.95	4.11	
NPSE	8.31	8.54	10.13	4.88	13.5						3.84	
CB6.4/N	1.75	3.14	1.62	1.17	4.2	5.81	5.85	1.61	1.87	1.09	1.04	
P	0.83	0.95	0.9	2	0.91	0.68	2.79	1.01	0.67	0.89	1.02	
SE	0.97	1.03	1.02	1	1.12						0.74	
NP	3.42	12.58	3.13	3.59	8.8	4.63	2.56	2.09	3.47	1.23	3.03	
NPSE	13.3	12.38	6.43	3.36	10.2						3.03	

## FIGURES

1. Map of region and station locations.
2. Chlorophyll "a" concentrations (ug/L) on day 0 at stations (A) TF3.2 and TF5.5 and (B) LE3.6, LE5.5, CB6.1 and CB6.4.
3. Nutrient concentrations (uM) at six stations. (A) Nitrate, (B) Ammonia, (C) Nitrite, (D) Phosphate, (E) Total DIN.
- 4-15. Chlorophyll "a" concentrations (ug/L) for bioassay each month for (A) TF3.2, (B) TF5.5, (C) LE3.6, (D) LE5.5, (E) CB6.1, (F) CB6.4. Arrow on x-axis indicates day for which enrichment index was calculated.
16. Enrichment indexes at stations (A) TF3.2, (B) TF5.5, (C) LE3.6, (D) LE5.5, (E) CB6.1, (F) CB6.4.
17. Nitrogen:phosphorus enrichment index ratio vs nitrogen:phosphorus molar nutrient ratio for each bioassay at each station. An index ratio  $>1$  indicates nitrogen limitation,  $<1$ , phosphorus limitation. A nutrient ratio  $> 16$  indicates phosphorus limitation,  $<1$ , nitrogen limitation.

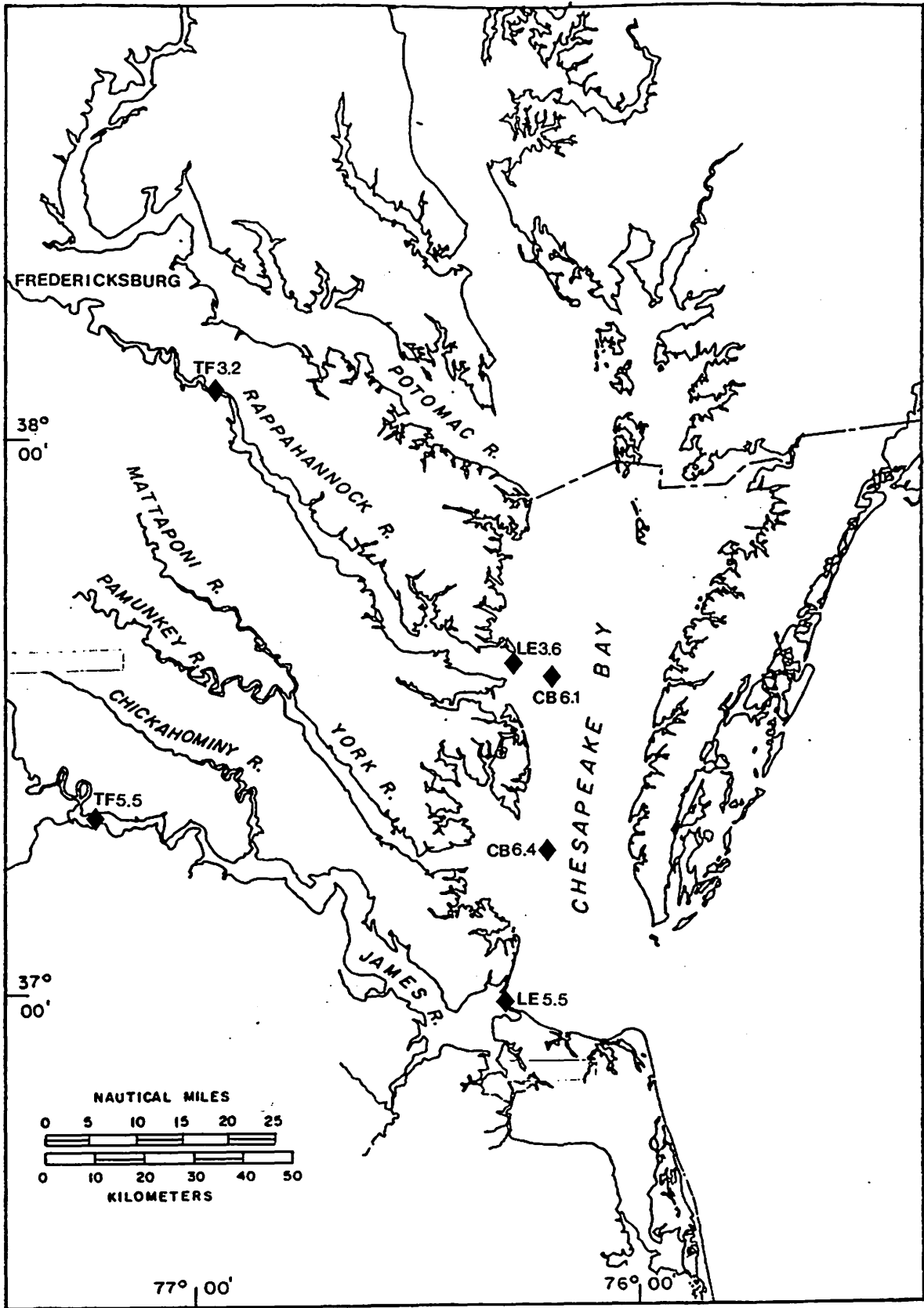
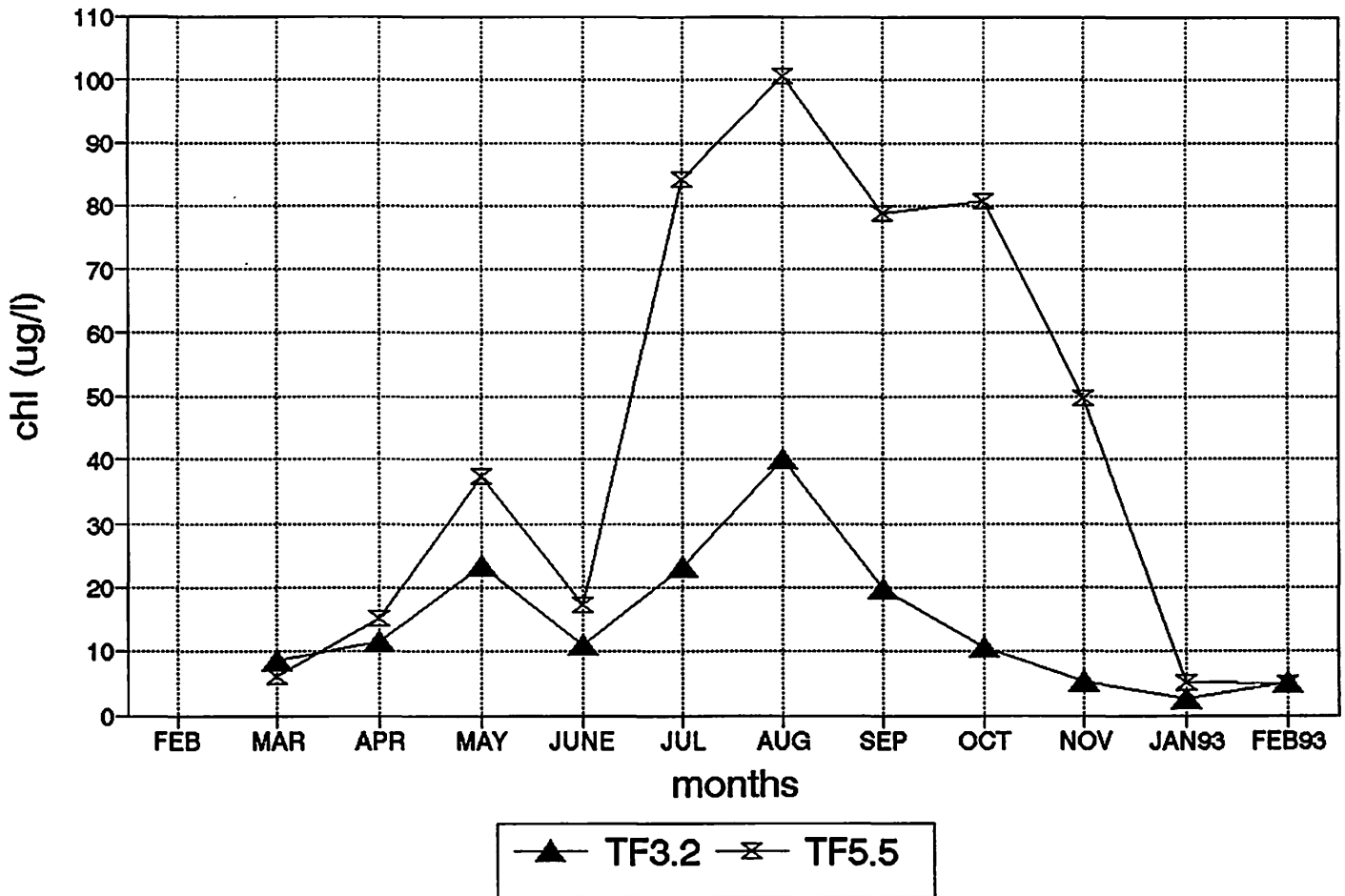


Figure 1



Fig 2A. Chlorophyll concentration Day 0



# Fig 2B. Chlorophyll concentration day 0

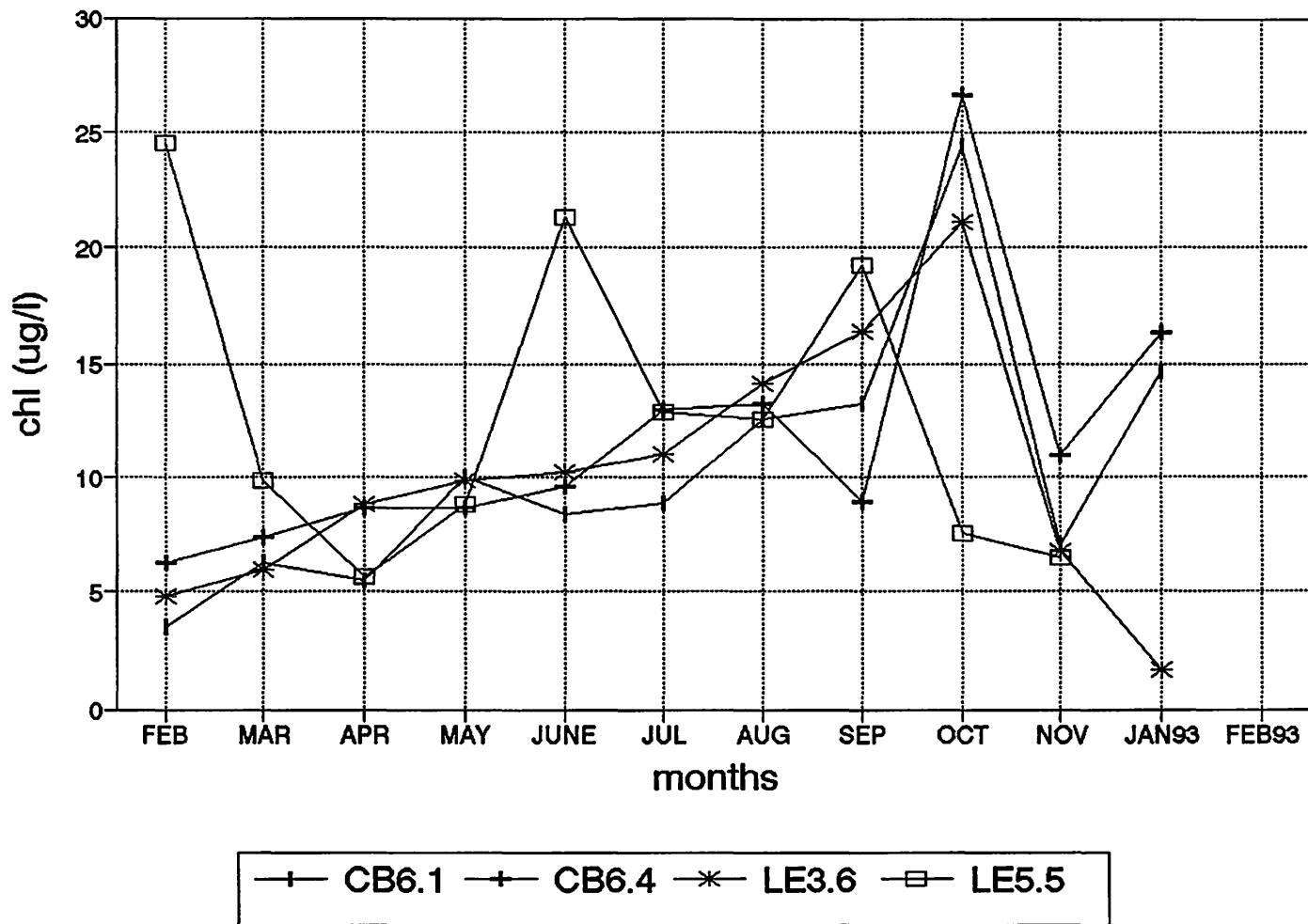


Fig 3A. NO<sub>3</sub> (uM) concentration.

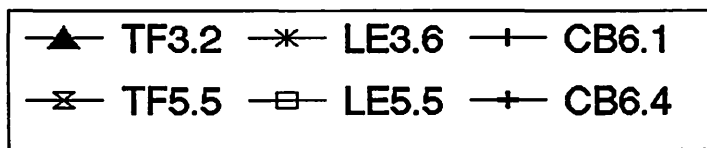
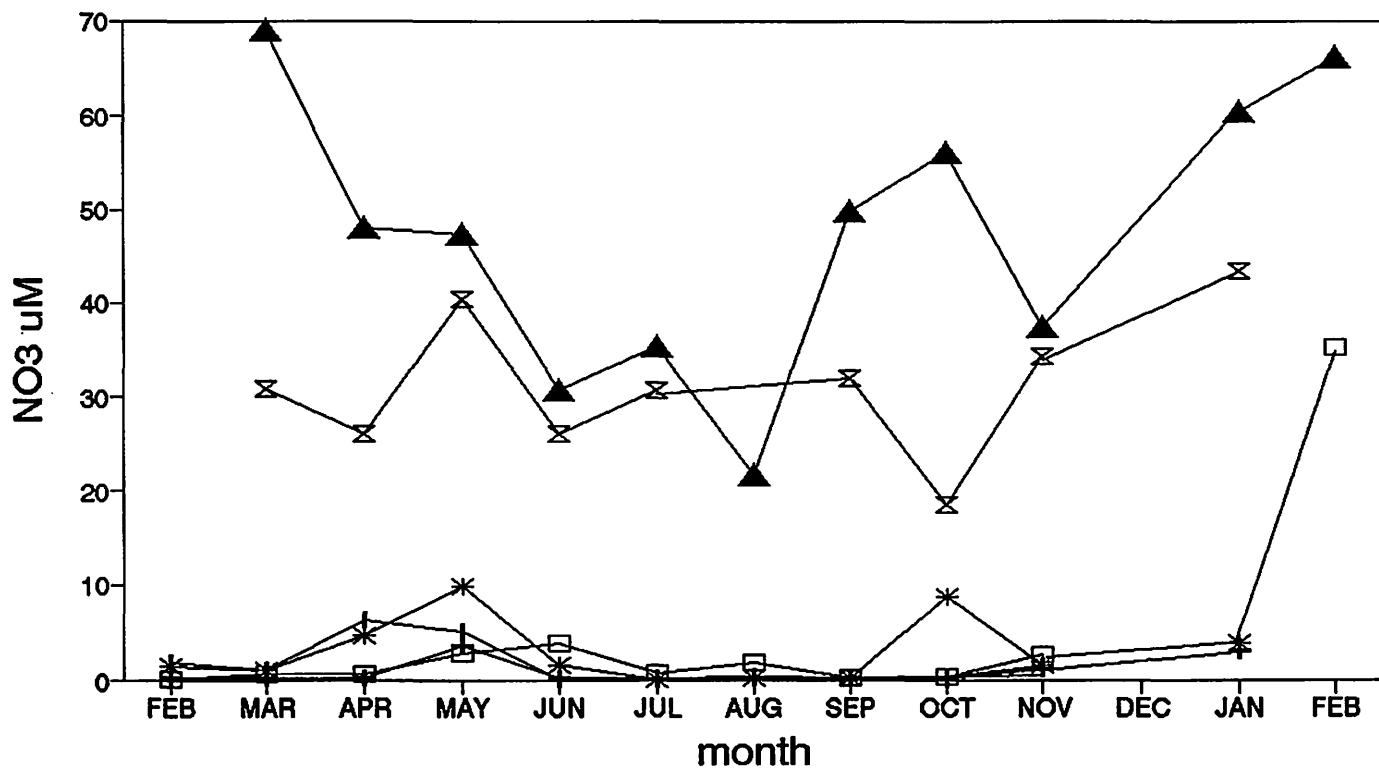


Fig 3B. NH4 (uM) concentration.

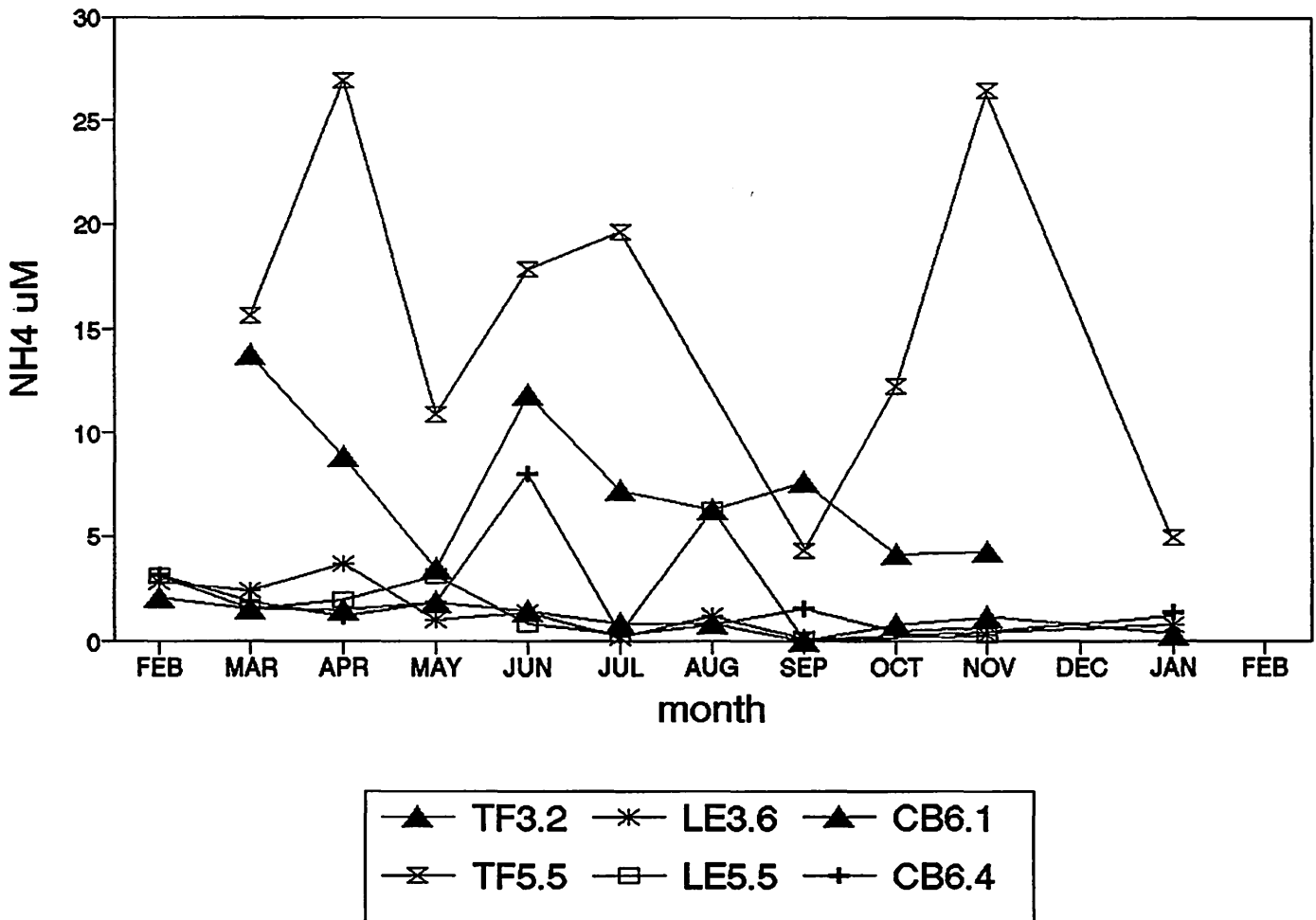


Fig 3C. NO<sub>2</sub> (uM) concentration.

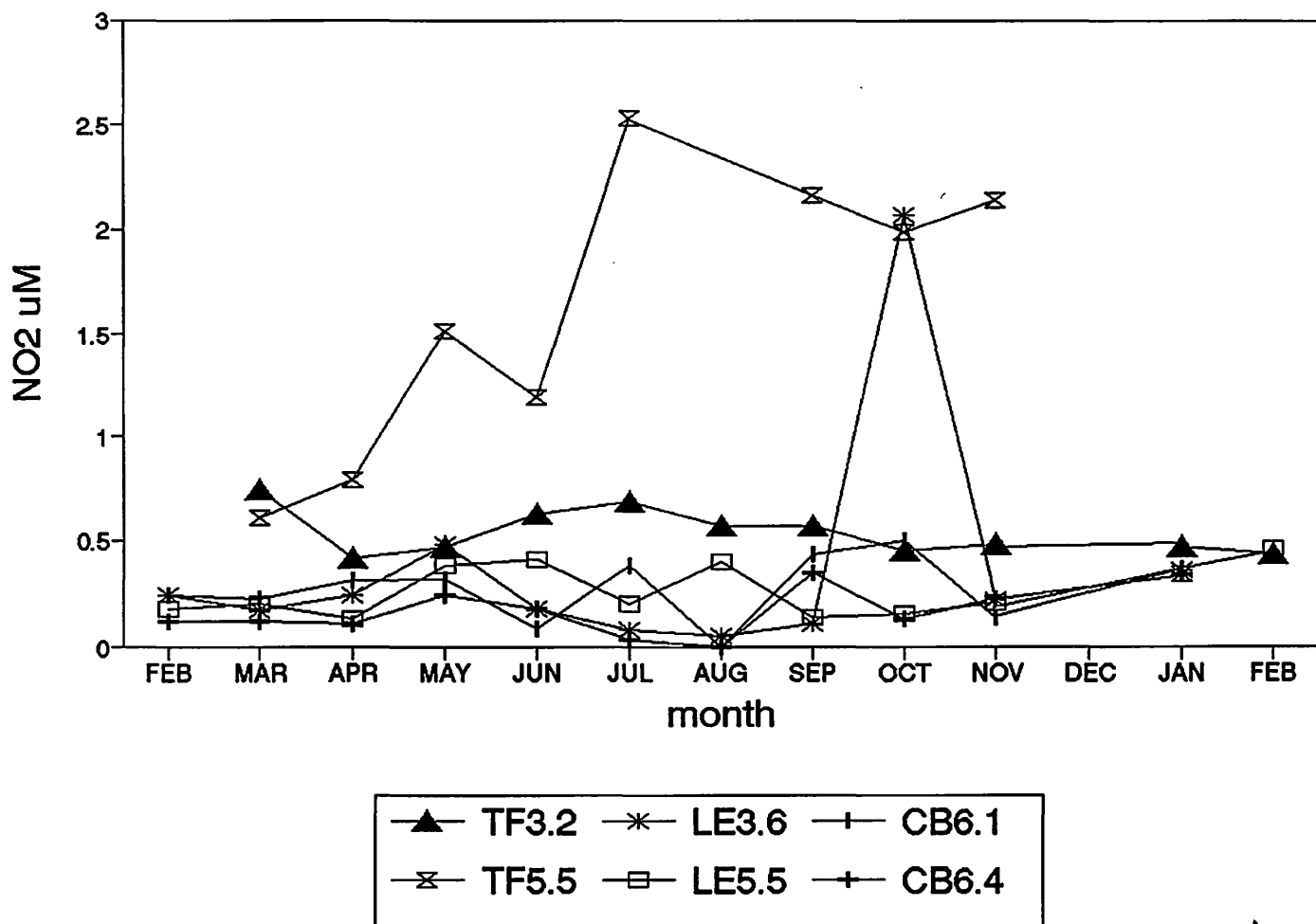


Fig 3D. PO<sub>4</sub> (uM) concentration.

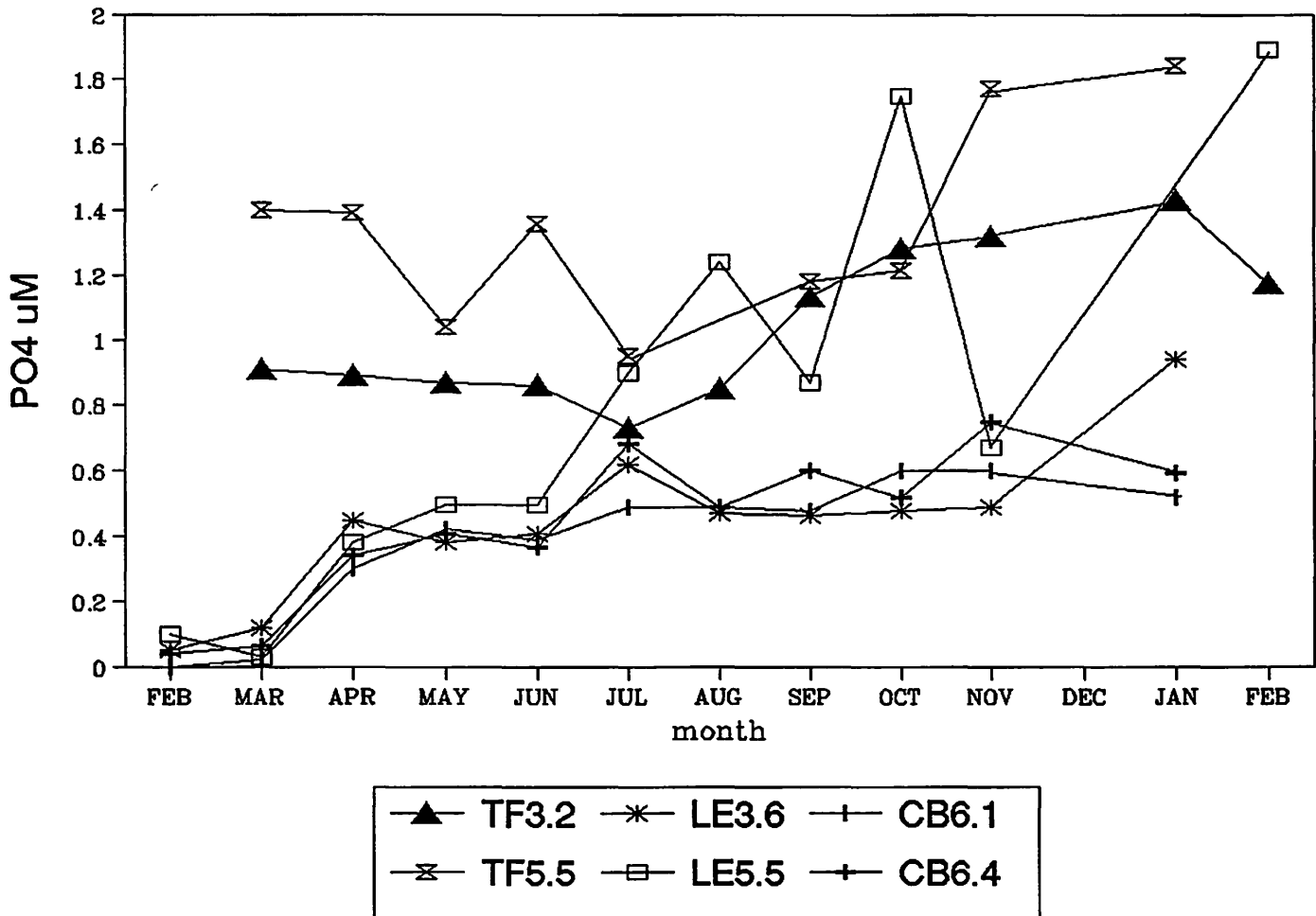


Fig 3E. Total DIN (uM) concentration.

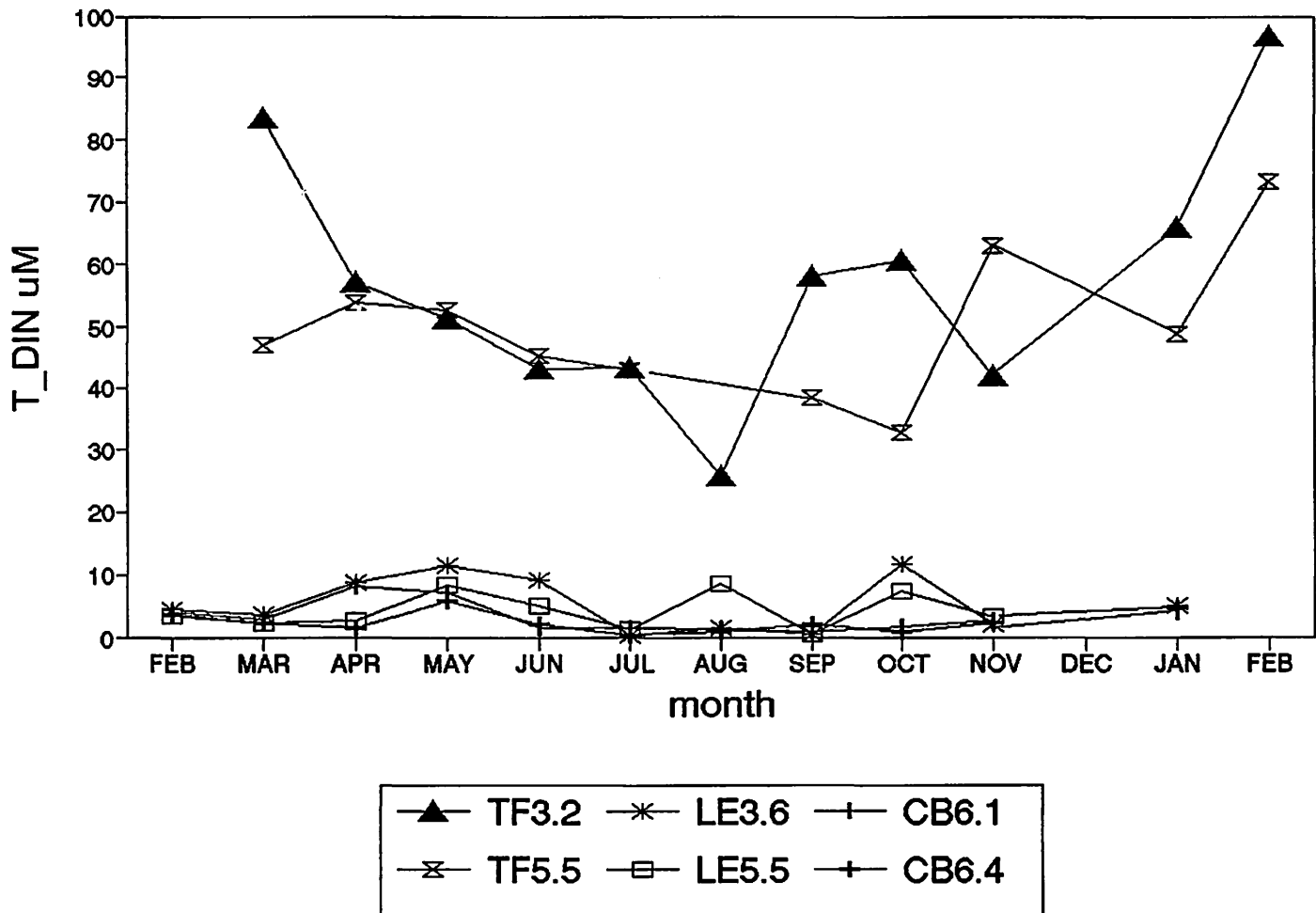


Fig 4C. February LE3.6

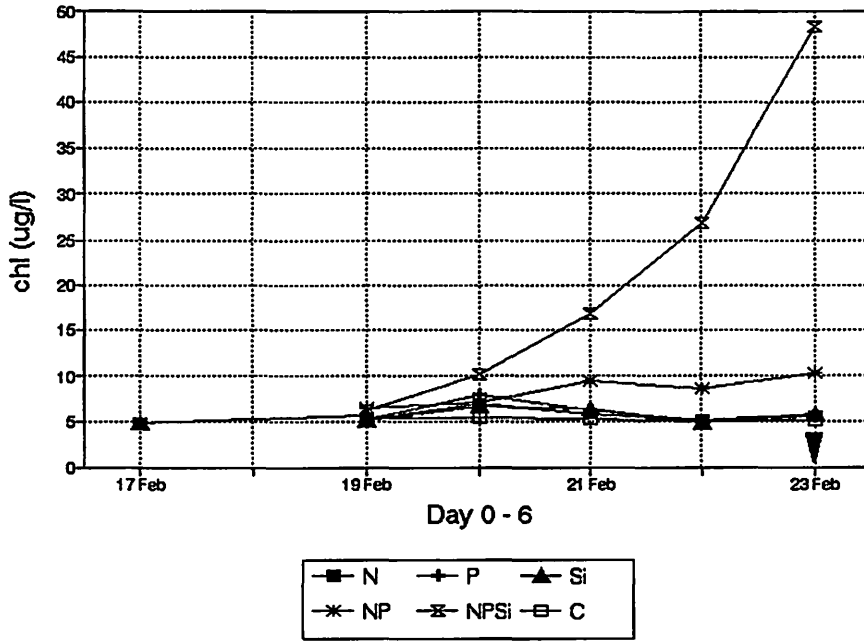


Fig 4D. February LE5.5

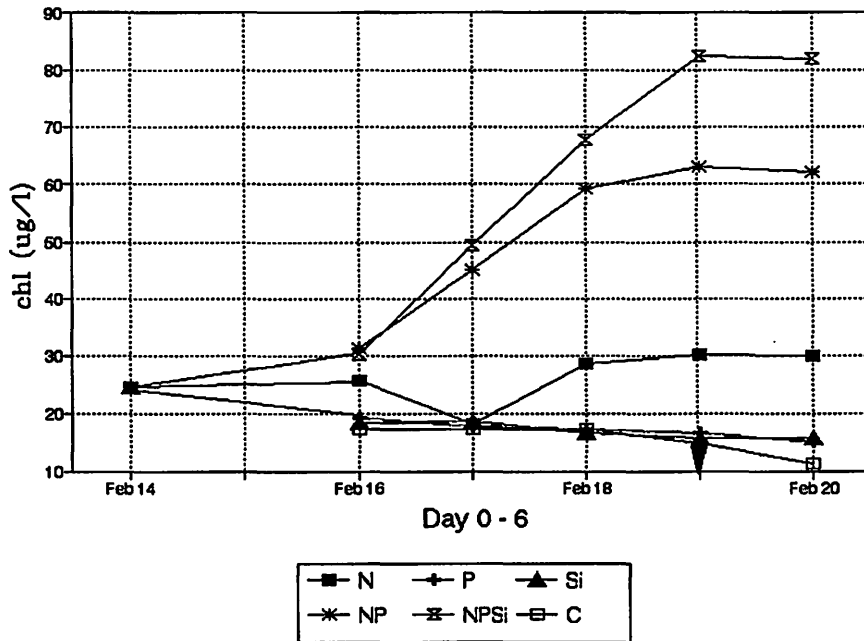




Fig 4E.

February CB6.1

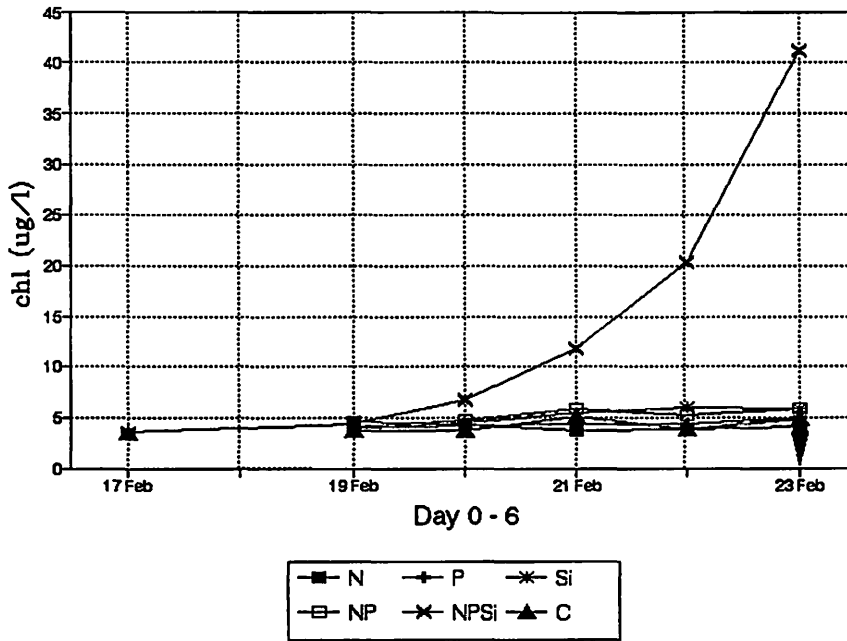


Fig 4F.

February CB6.4

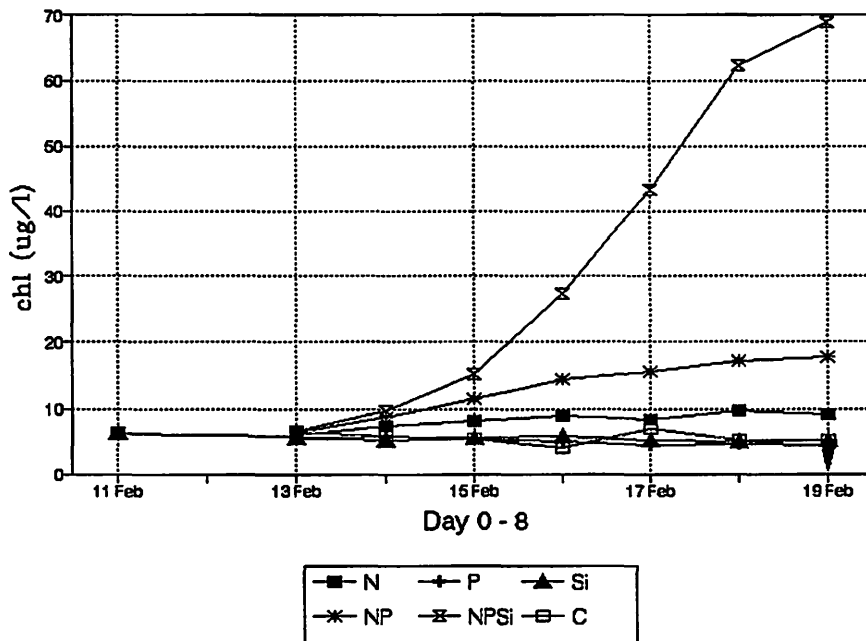


Fig 5A. March TF3.2

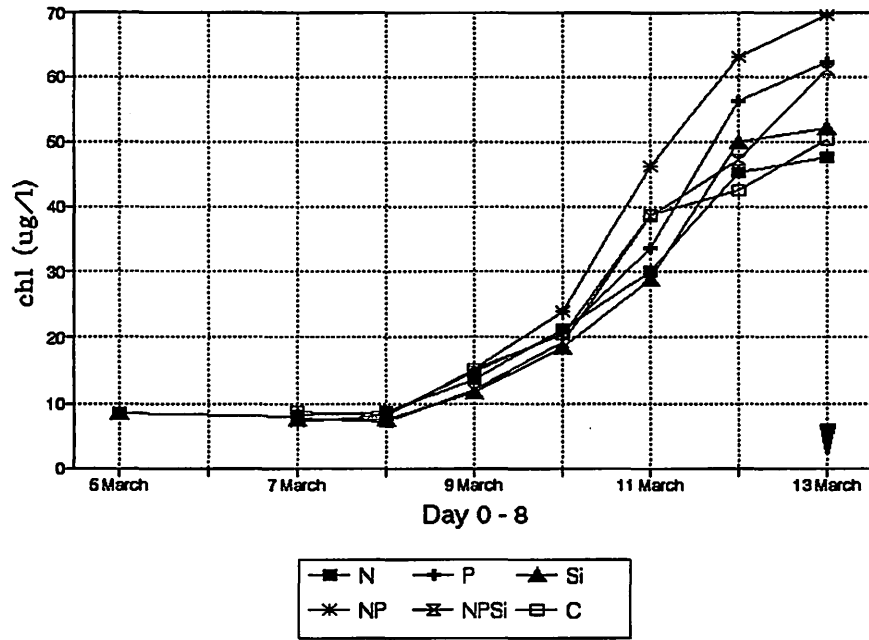


Fig 5B. March TF5.5

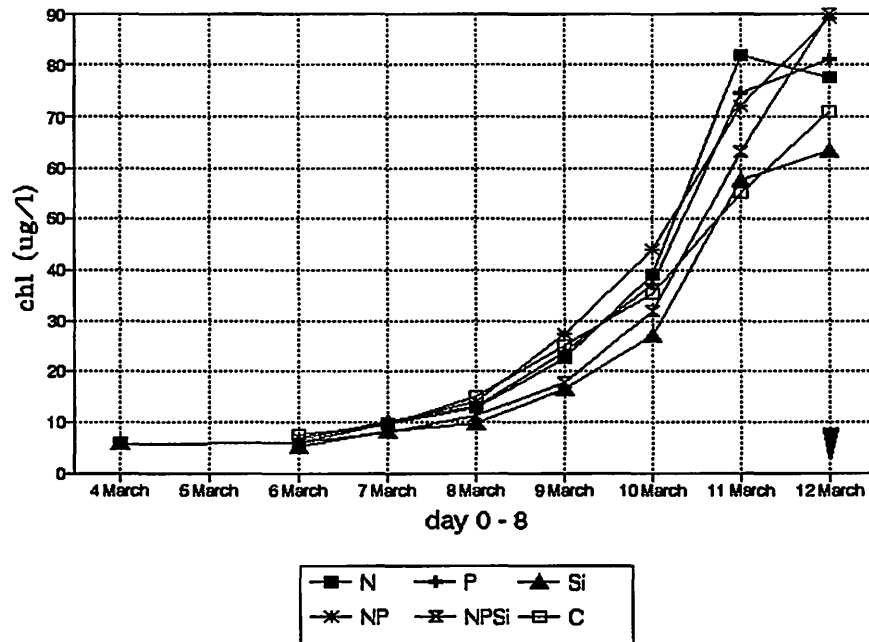


Fig 5C.

March LE3.6

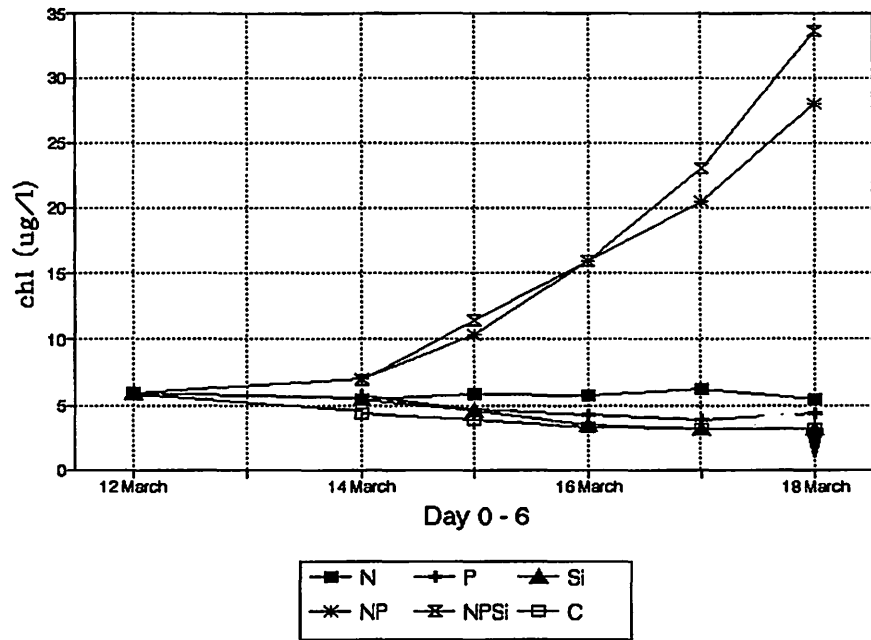


Fig 5D.

March LE5.5

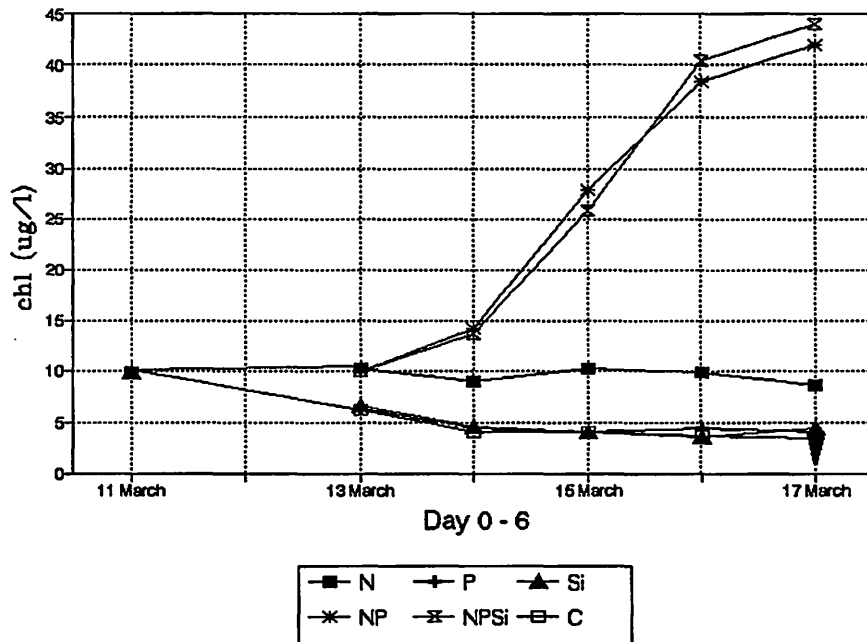


Fig 5E.

March CB6.1

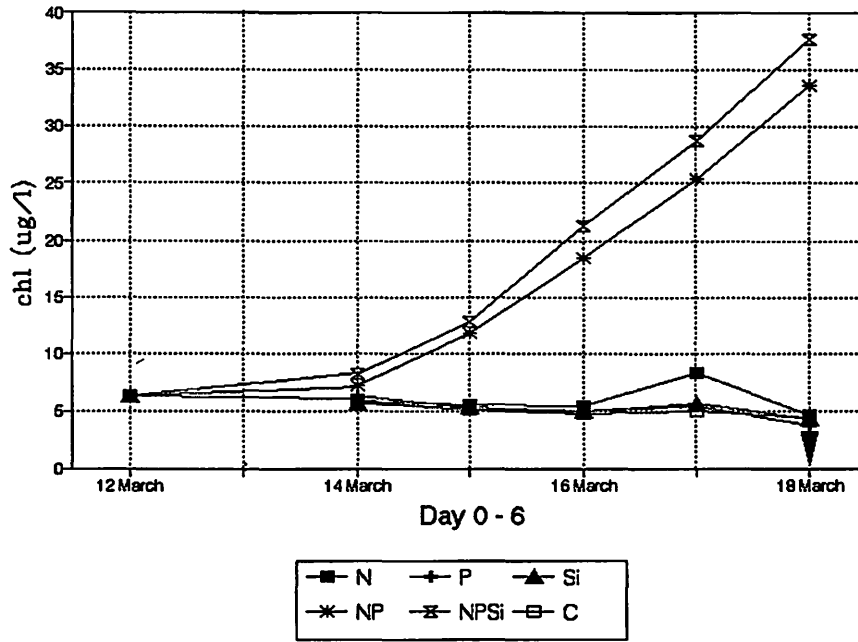


Fig 5F.

March CB6.4

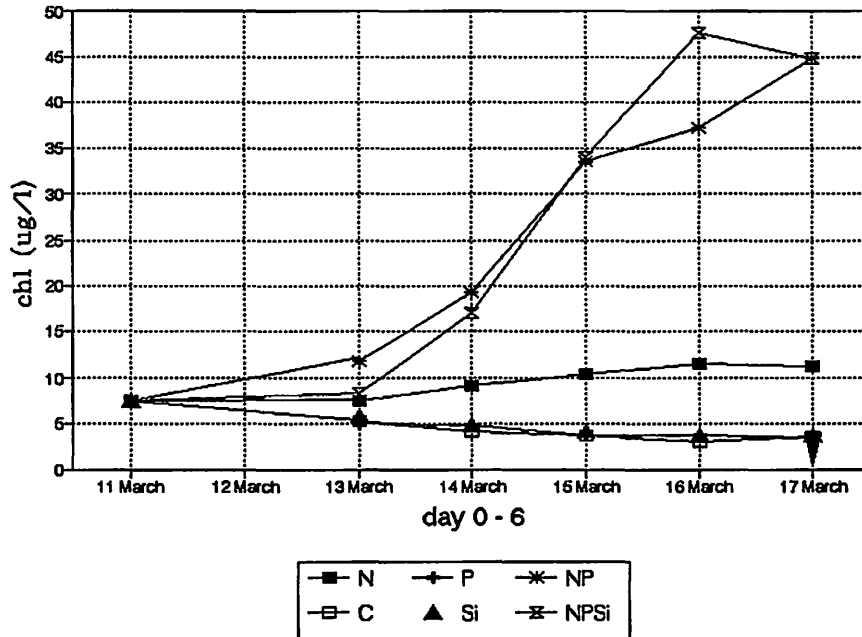


Fig 6A.

April TF3.2

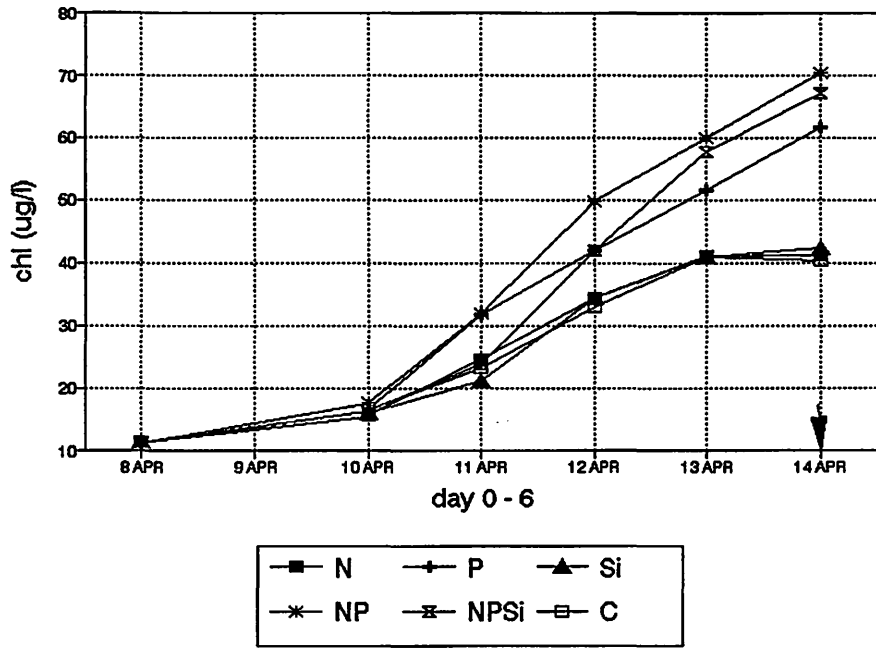


Fig 6B.

April TF5.5

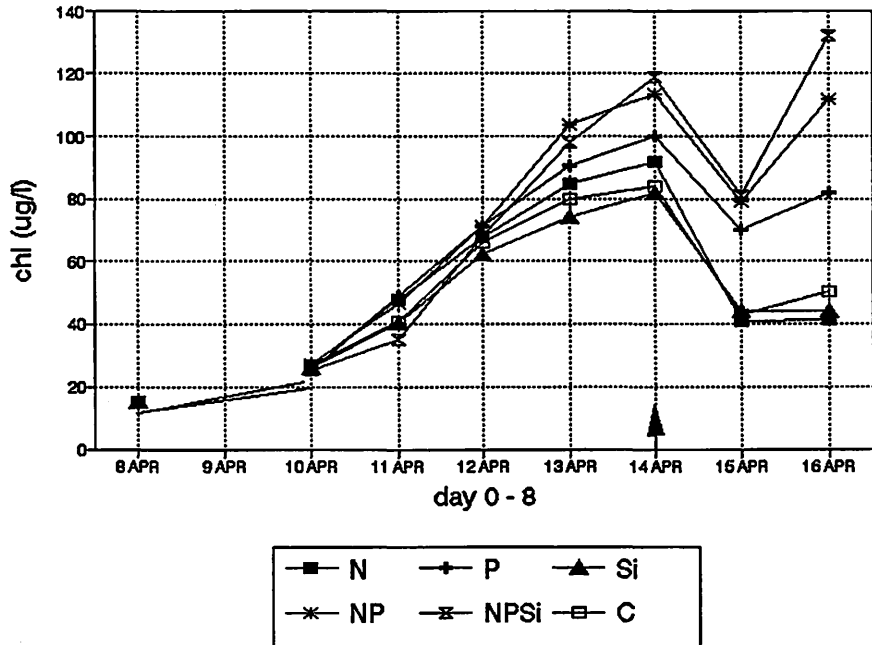


Fig 6C.

April LE3.6

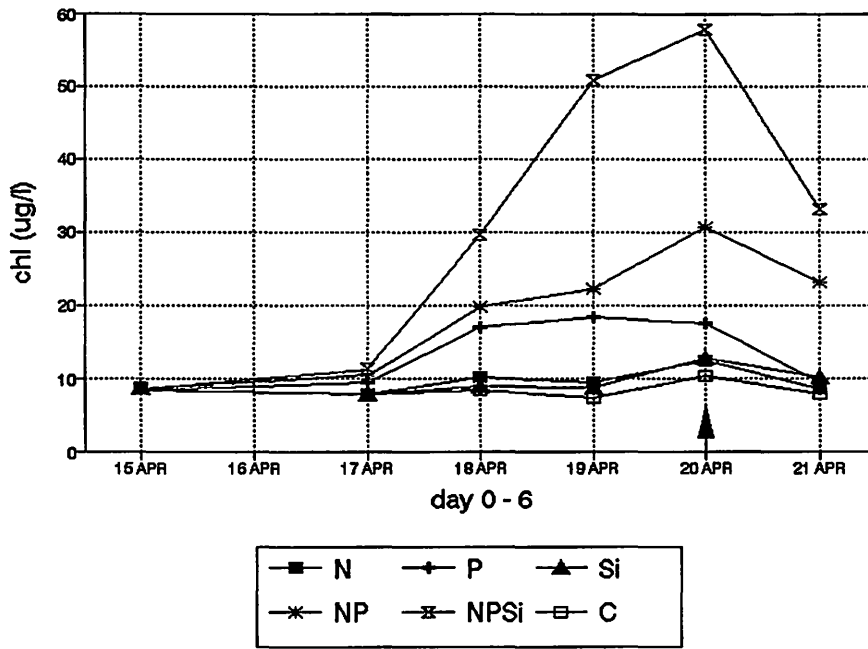


Fig 6D.

April LE5.5

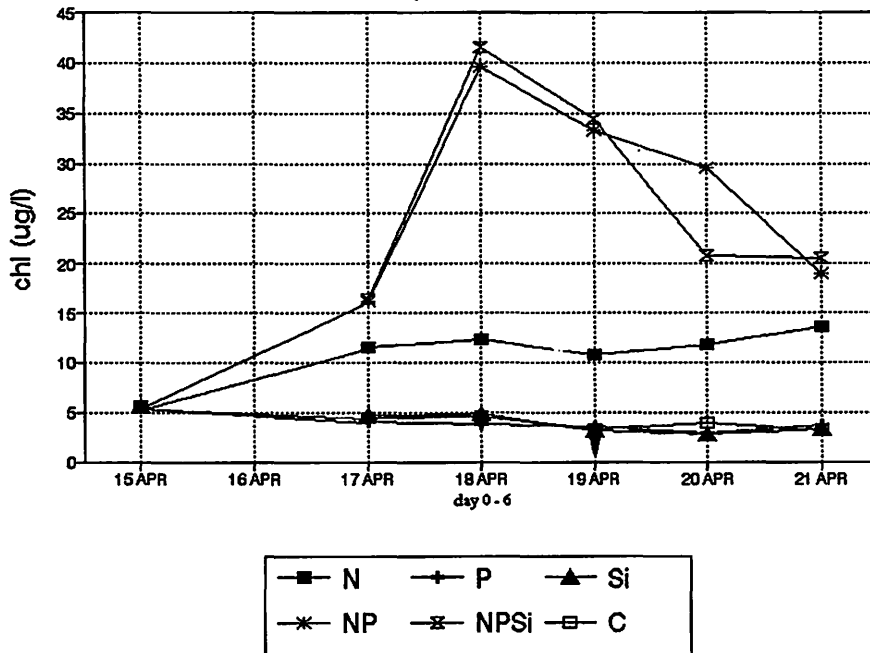


Fig 6E.

April CB6.1

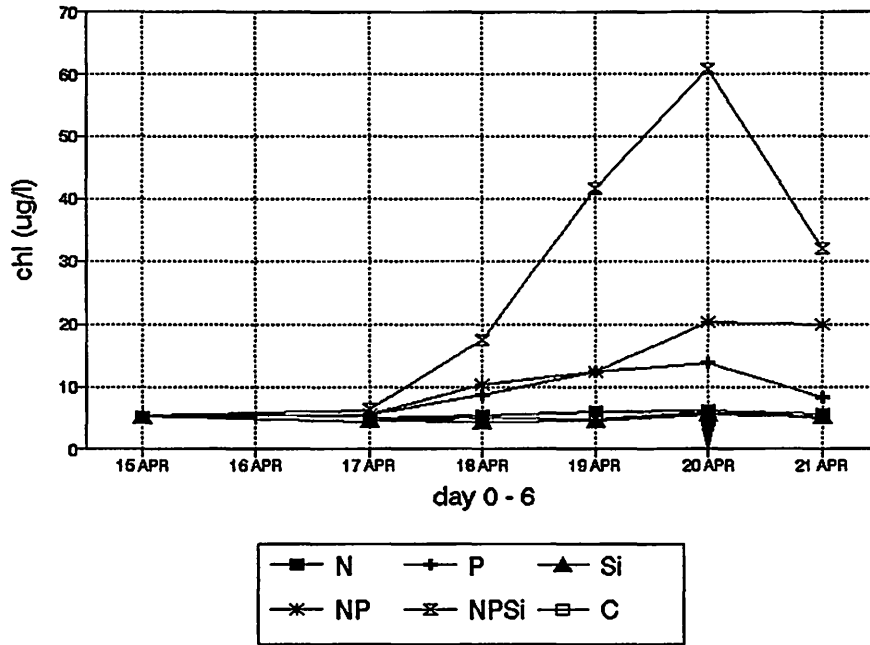


Fig 6F.

April CB6.4

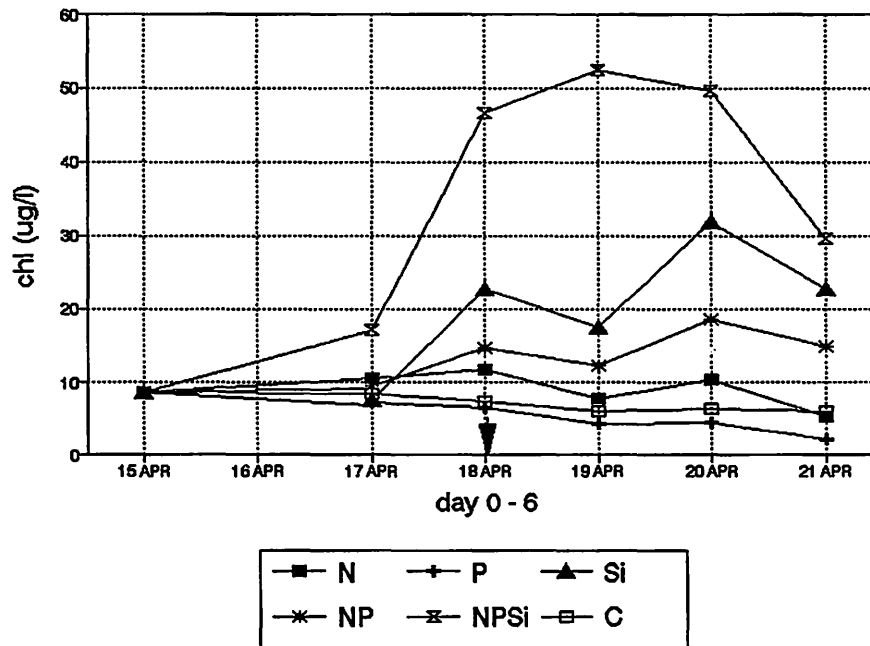


Fig 7A.

May TF3.2

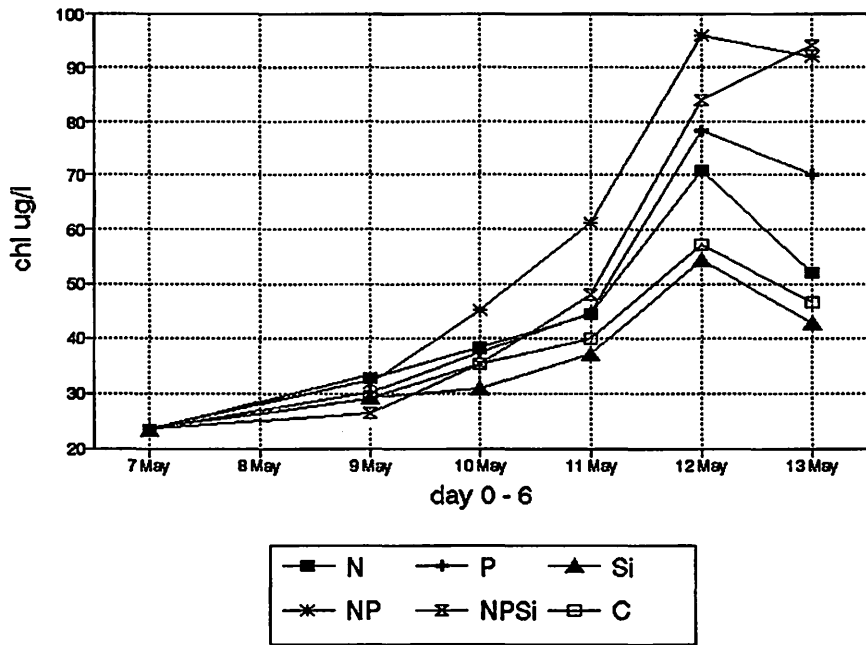


Fig 7B.

May TF5.5

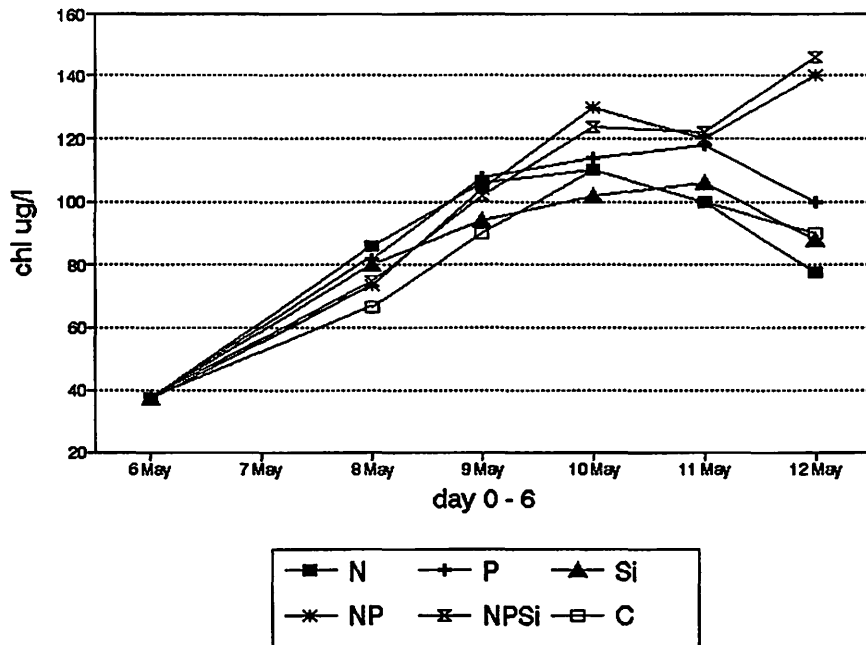




Fig 7C.

May LE3.6

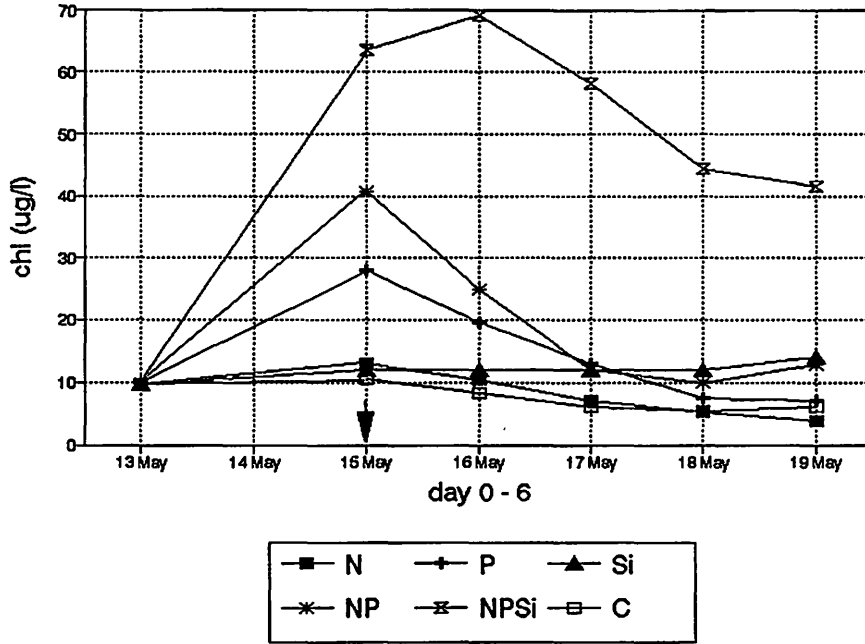


Fig 7D.

May LE5.5

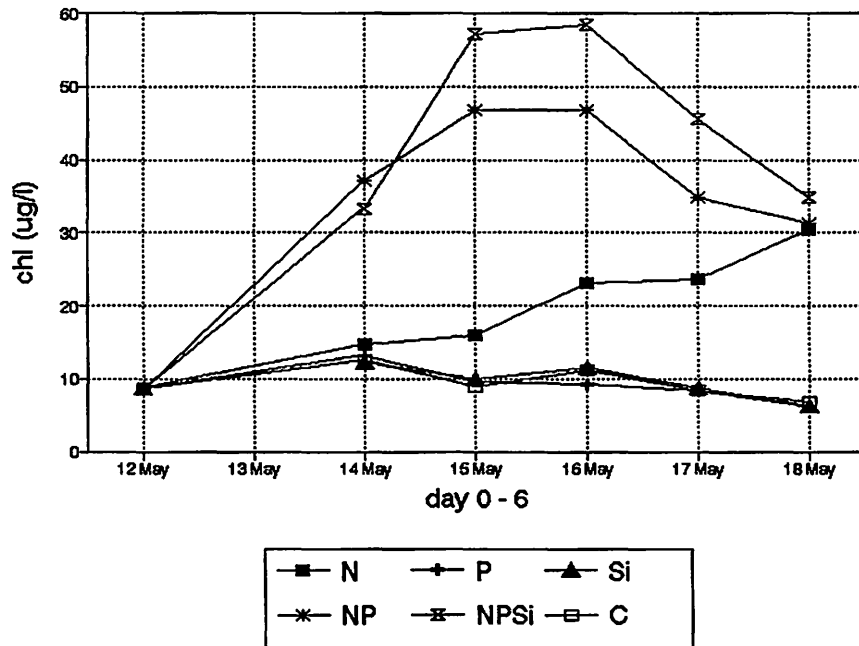


Fig 7E.

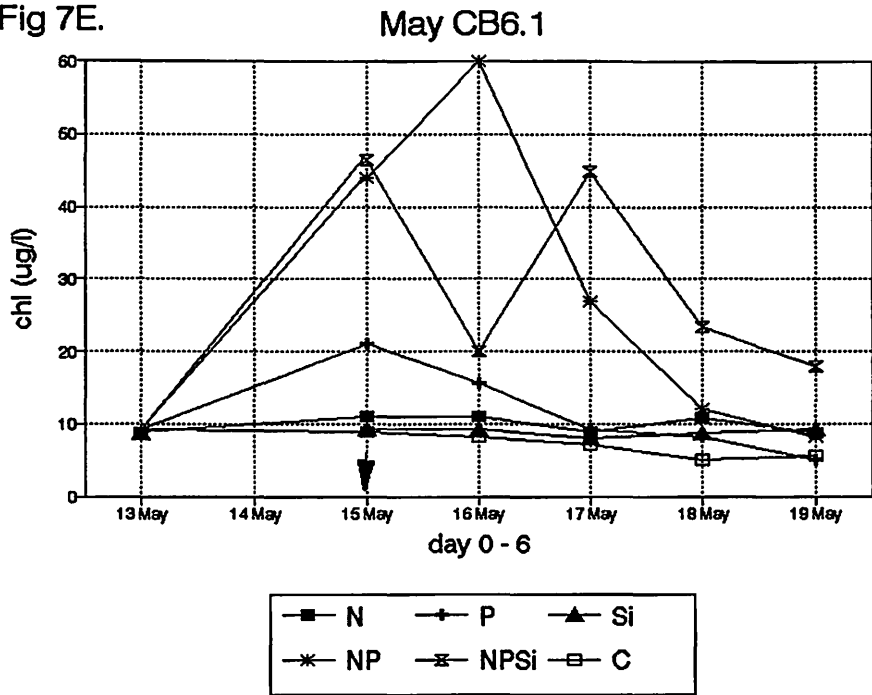


Fig 7F.

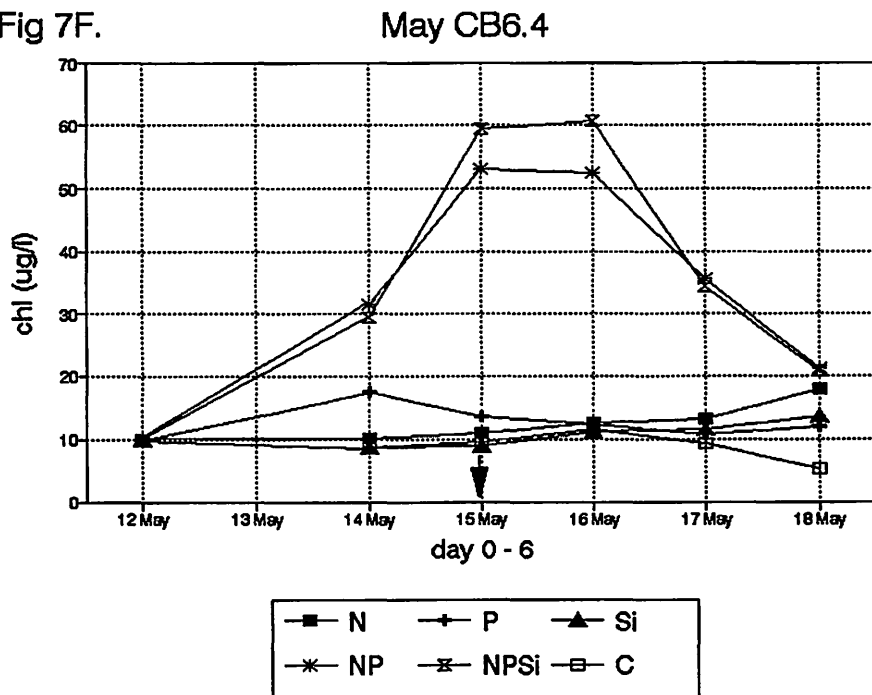


Fig 8A.

June TF3.2

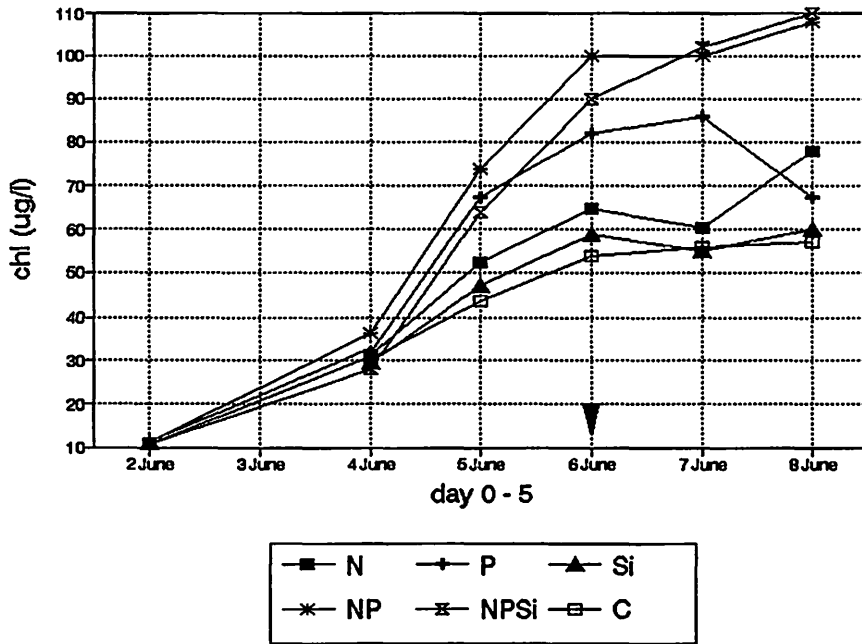


Fig 8B.

June TF5.5

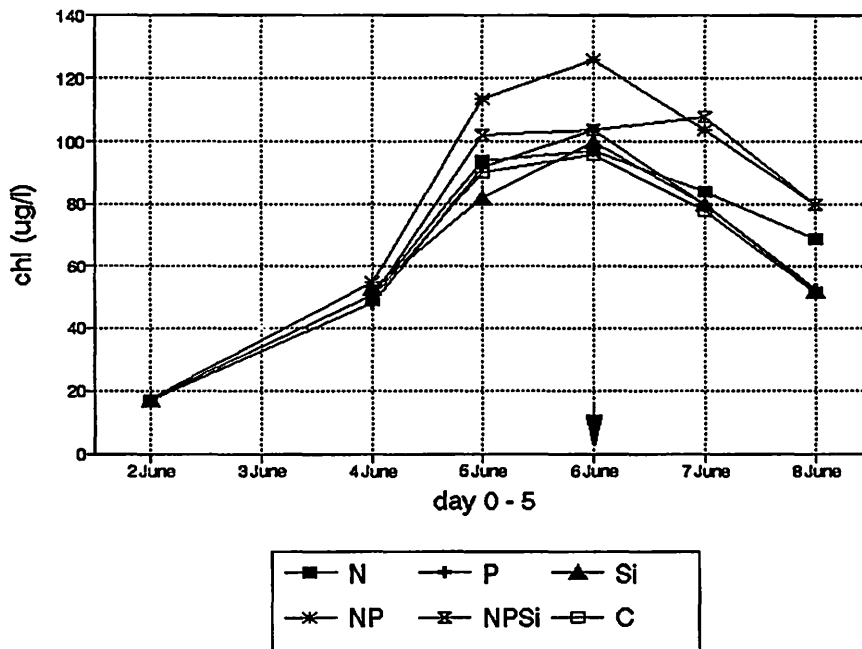


Fig 8C.

June LE3.6

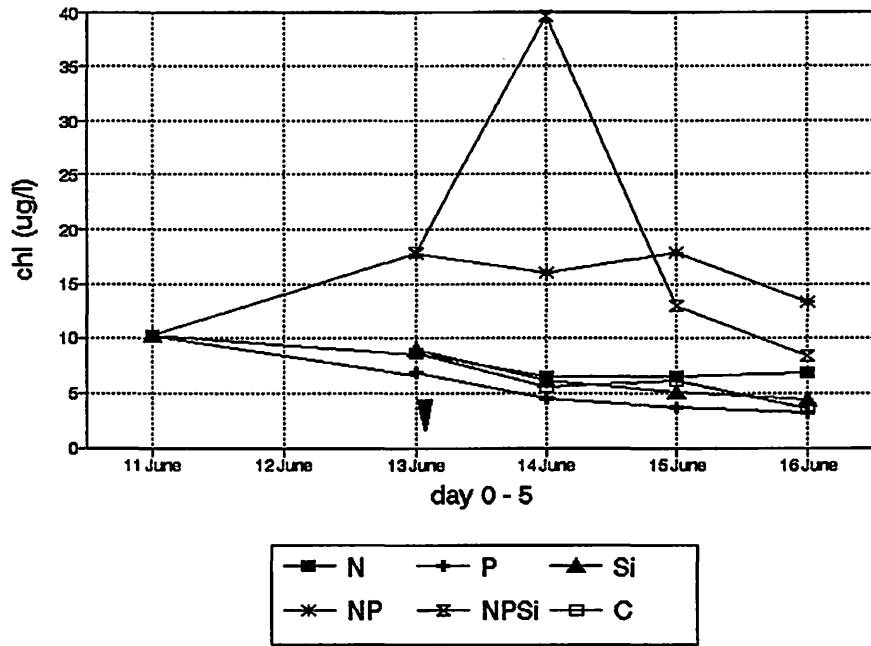


Fig 8D.

June LE5.5

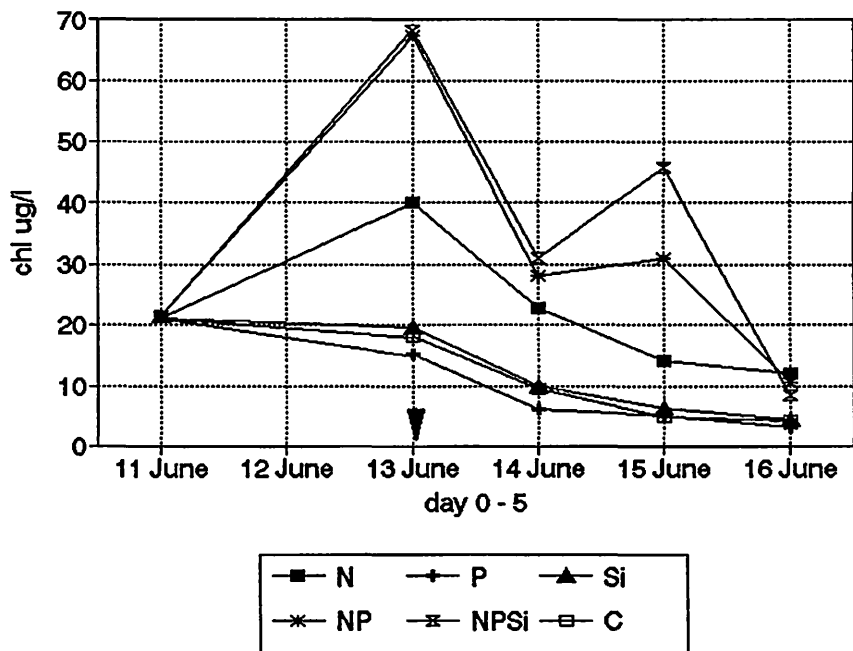


Fig 8E.

June CB6.1

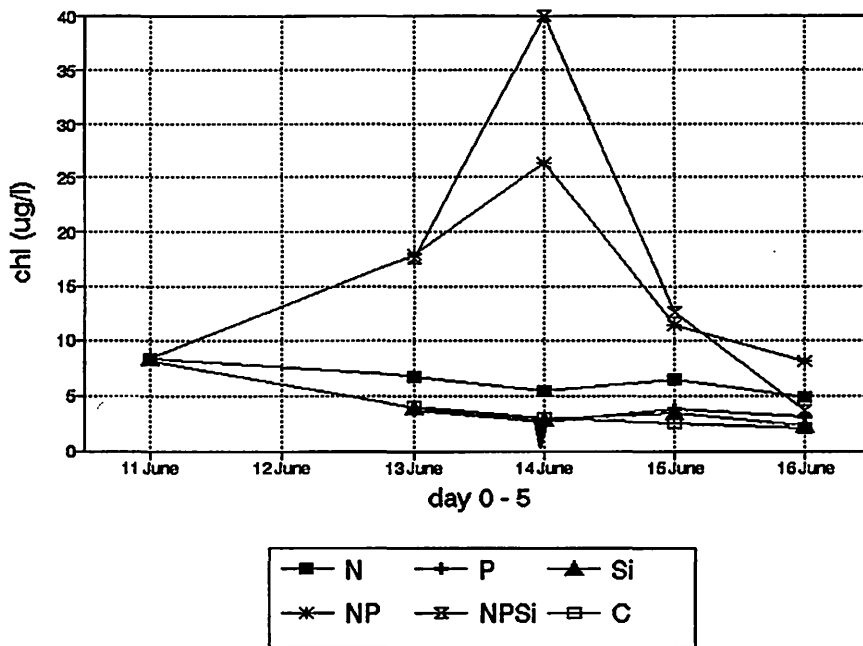


Fig 8F.

June CB6.4

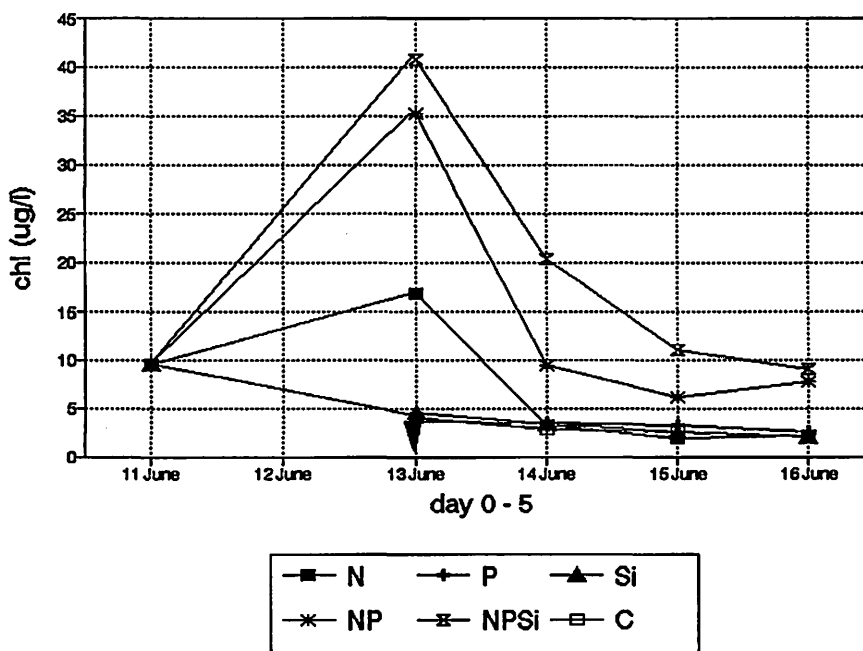


Fig 9A.

July TF3.2

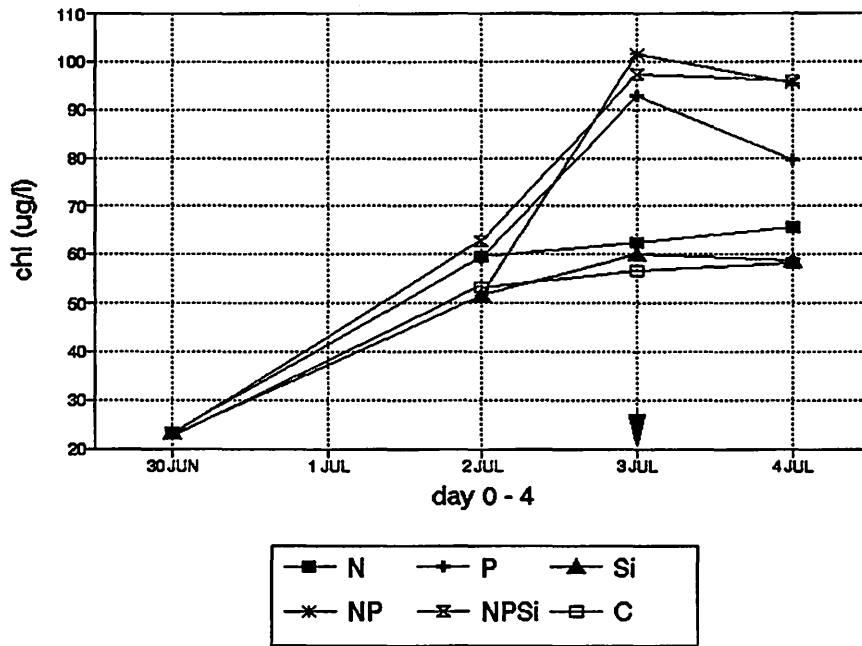


Fig 9B.

July TF5.5

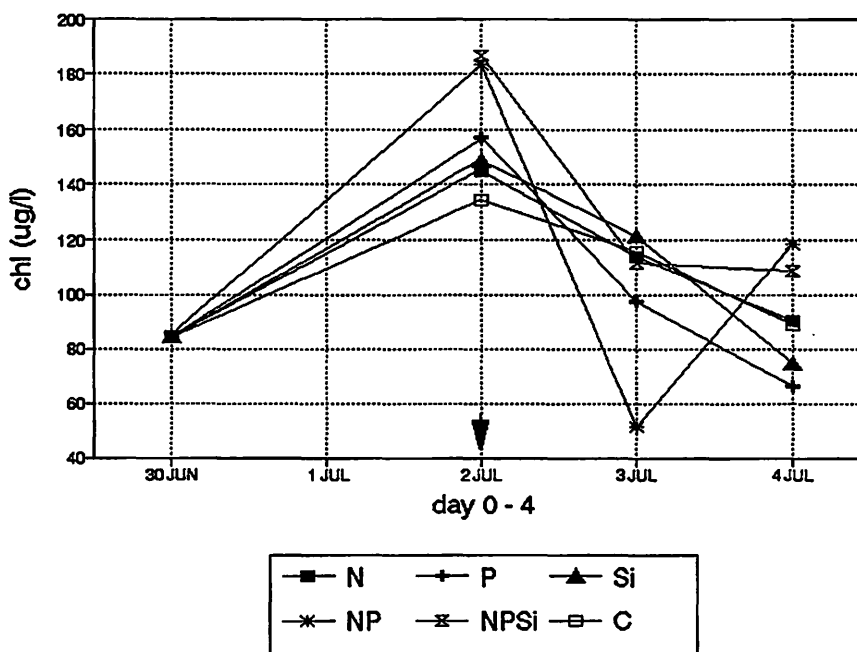


Fig 9C.

July LE3.6

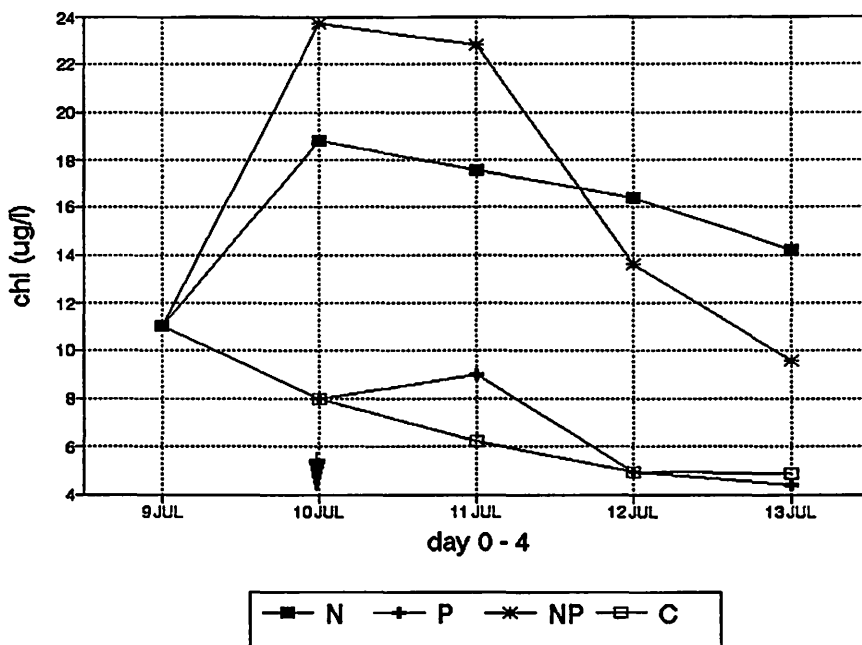


Fig 9D.

July LE5.5

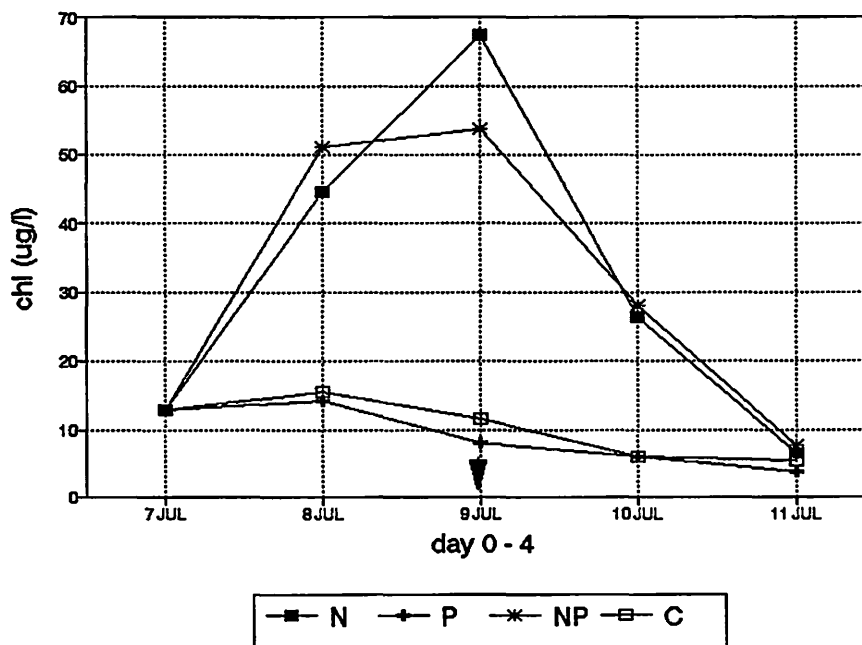


Fig 9E.

July CB6.1

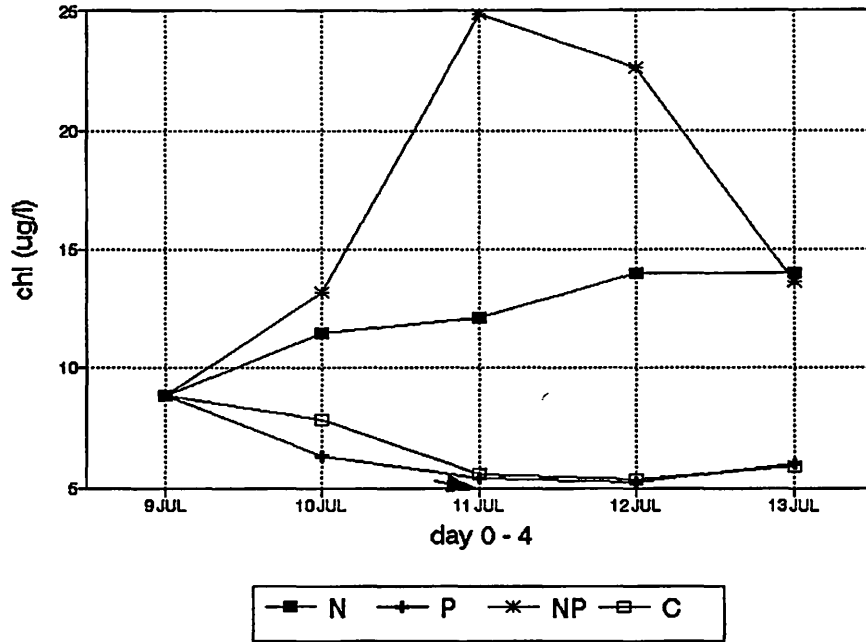


Fig 9F.

July CB6.4

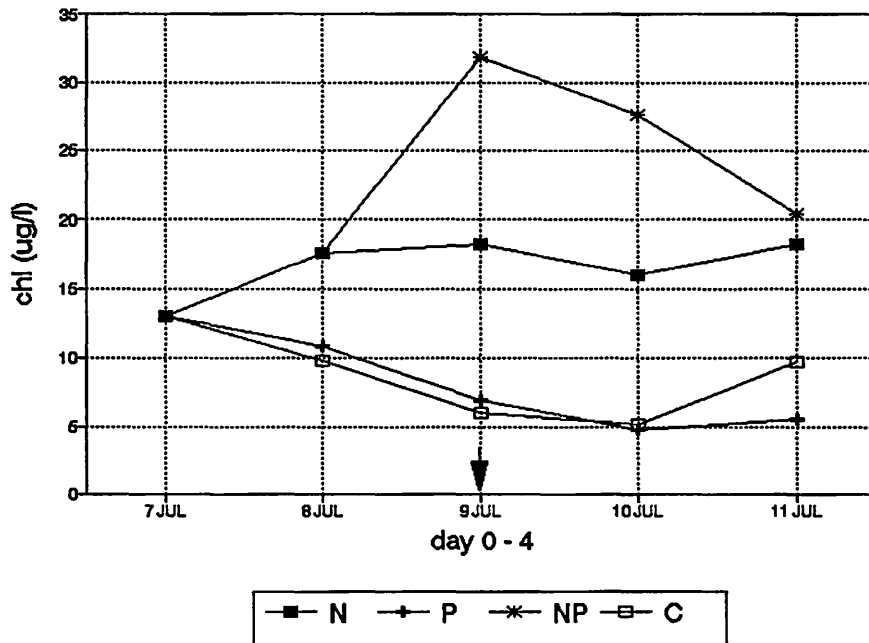




Fig 10 A. August TF3.2

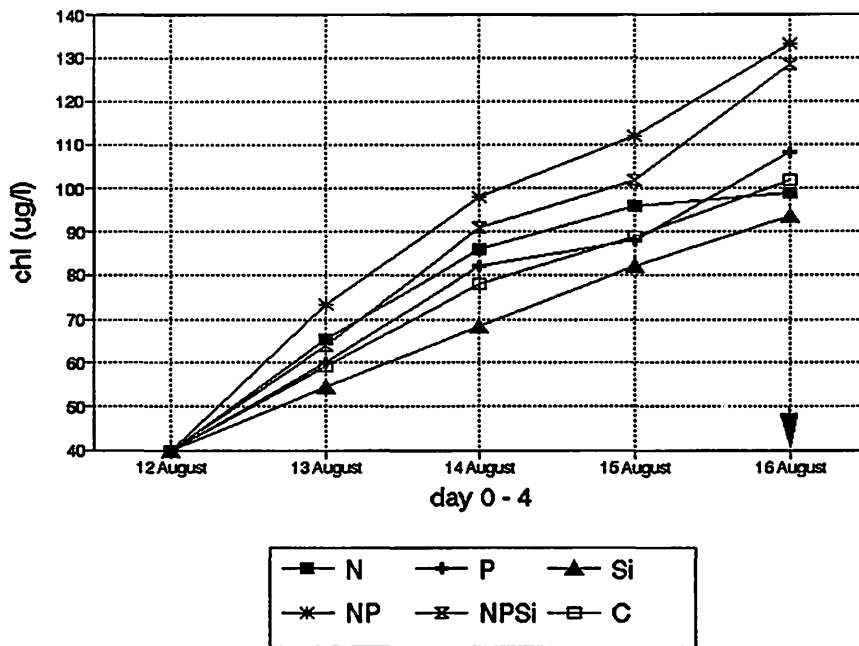


Fig 10B. August TF5.5

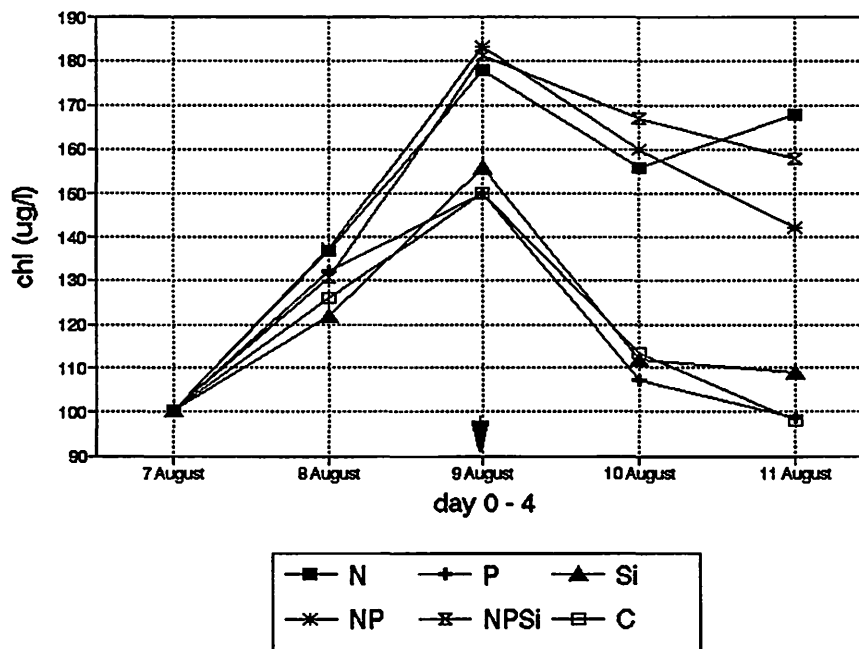


Fig 10C.

August LE3.6

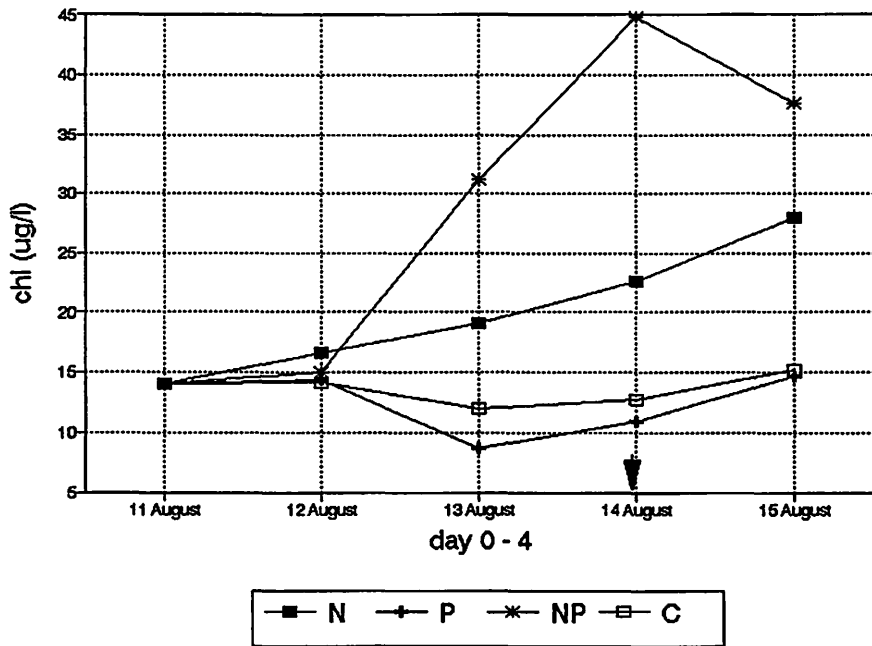


Fig 10D.

August LE5.5

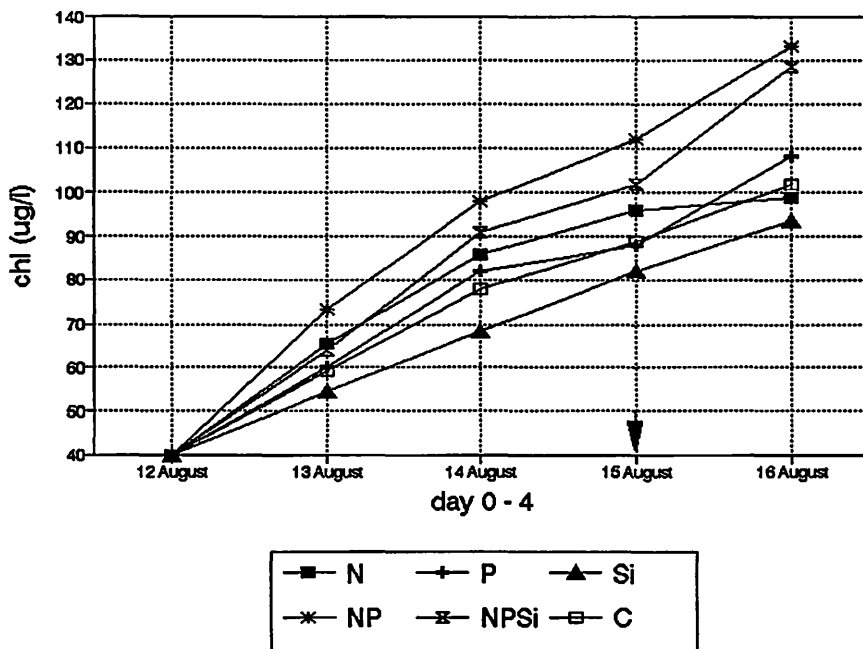


Fig 10E.

August CB6.1

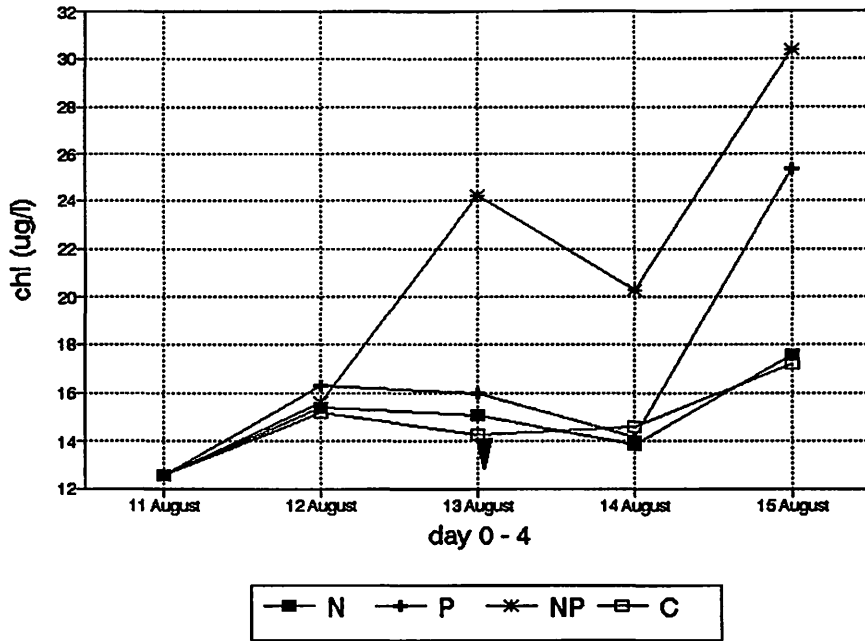


Fig 10F.

August CB6.4

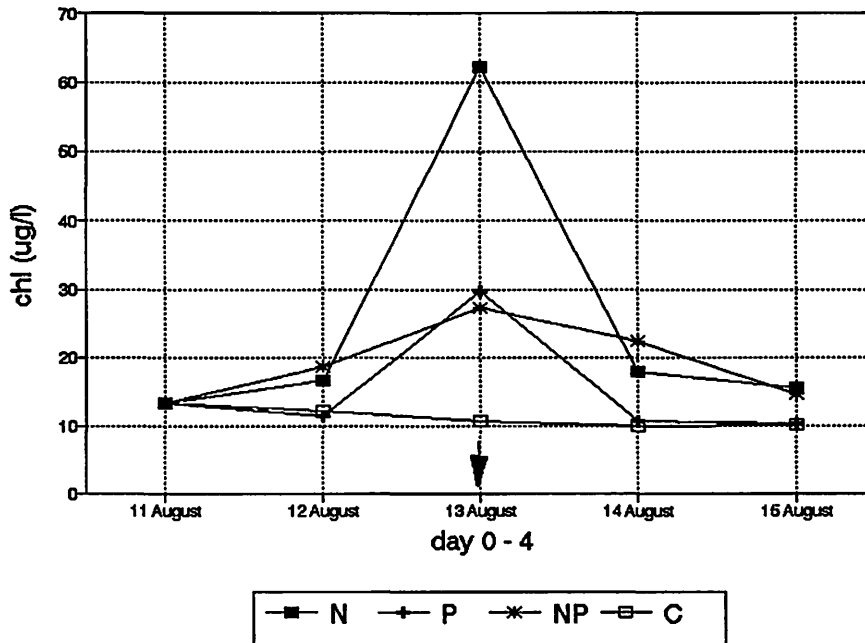


Fig 11A.

September TF3.2

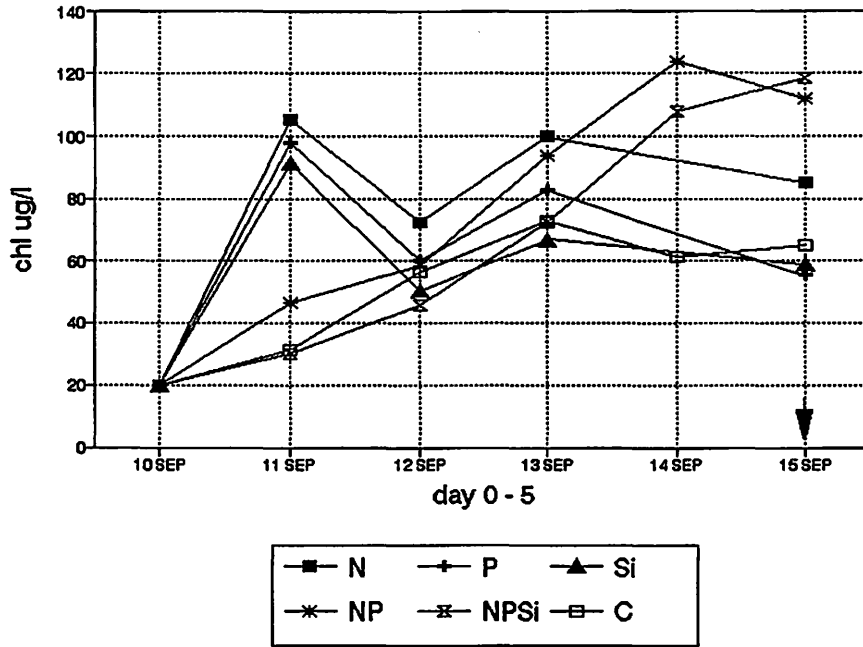


Fig 11B.

September TF5.5

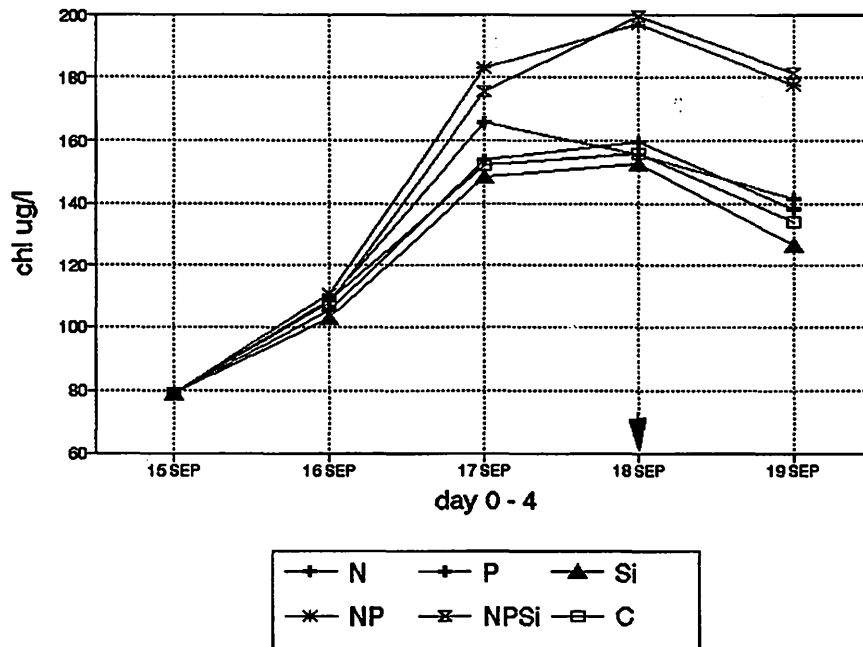


Fig 11C. September LE3.6

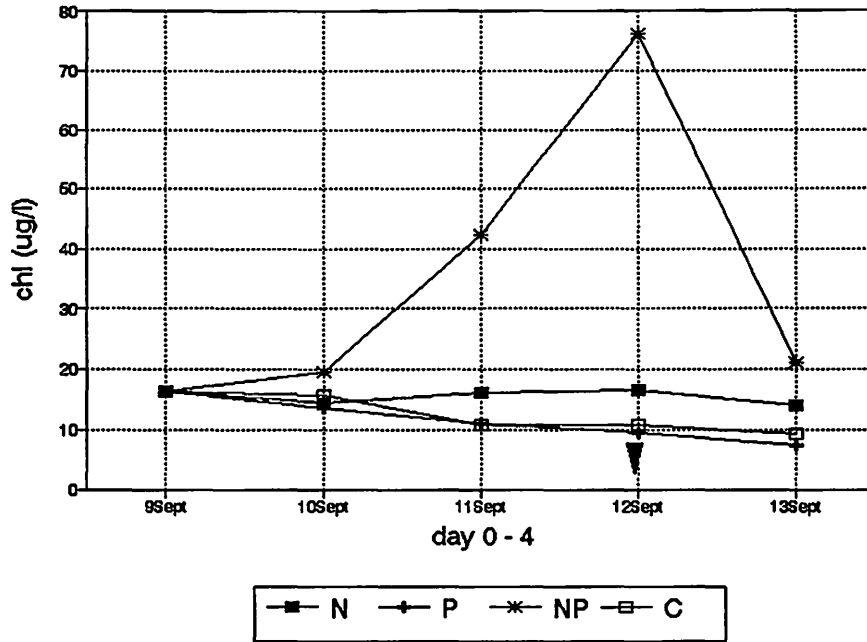


Fig 11D. September LE5.5

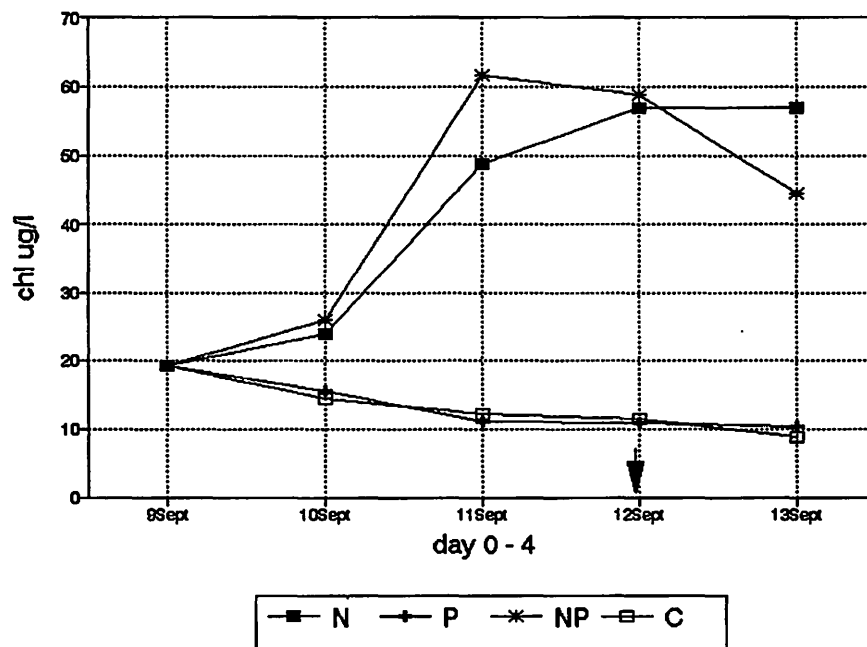


Fig 11E.

September CB6.1

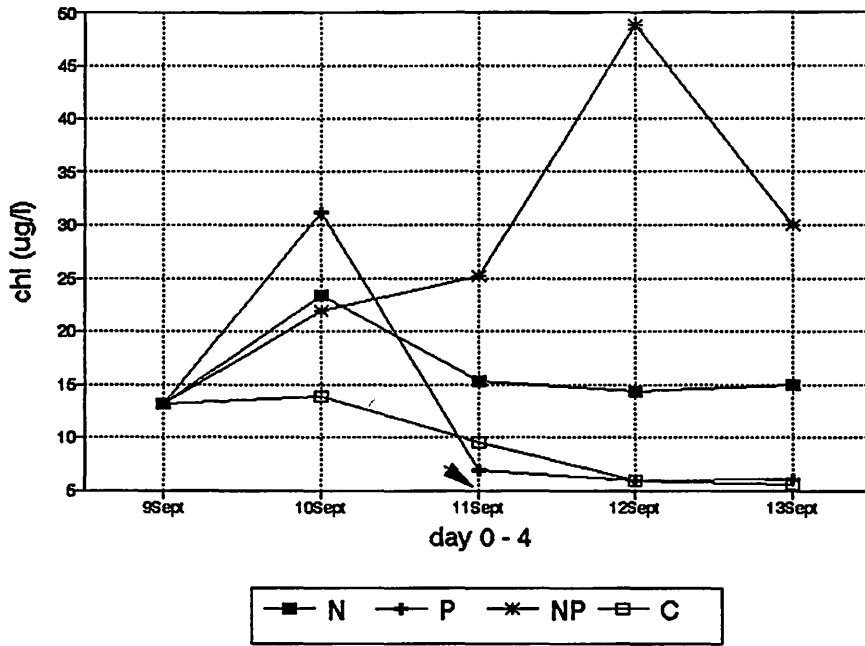


Fig 11F.

September CB6.4

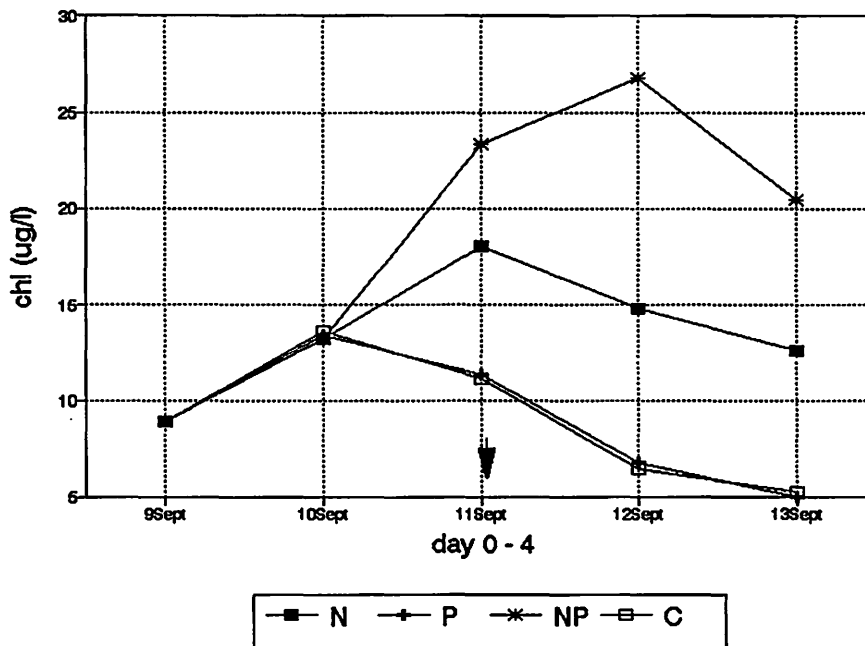


Fig 12A.

October TF3.2

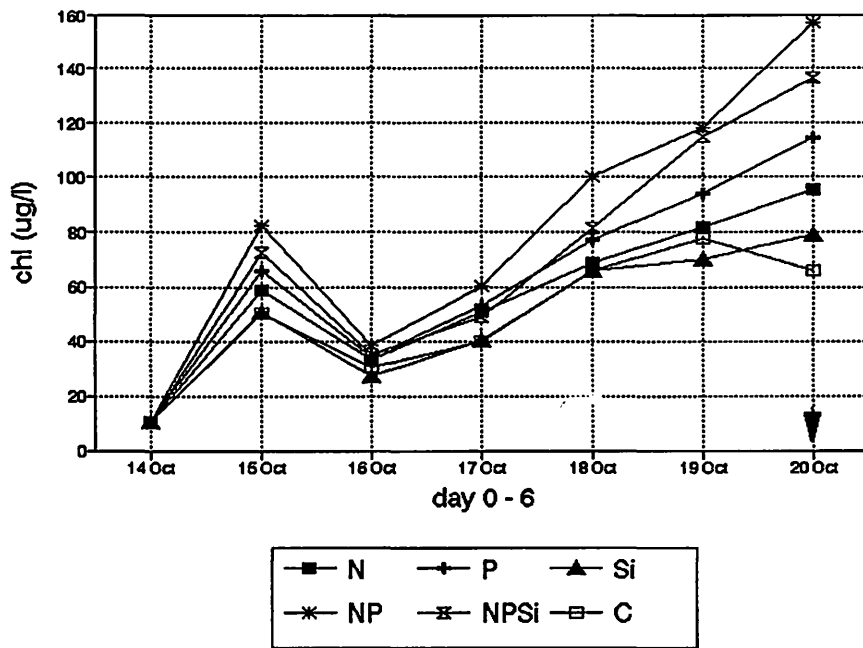


Fig 12B.

October TF5.5

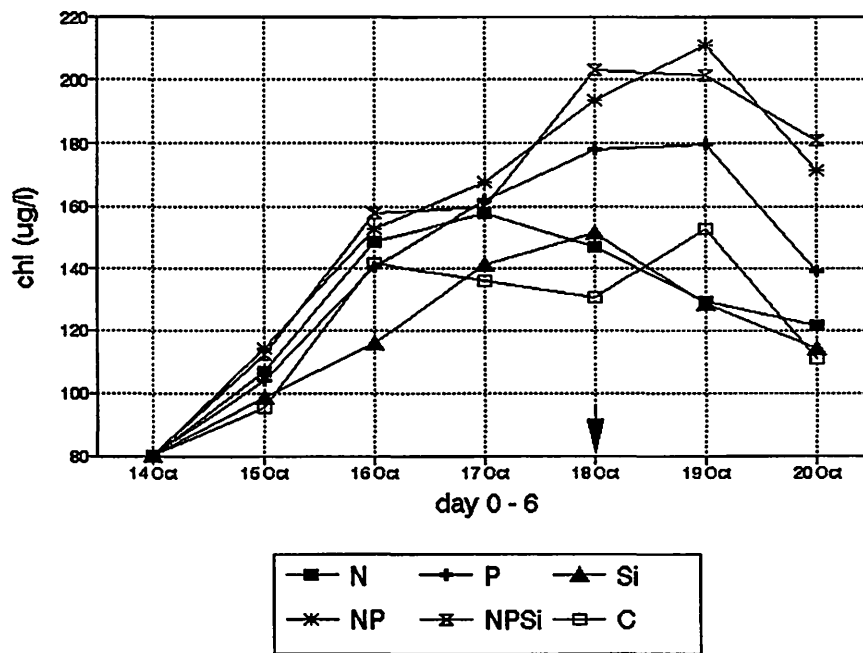


Fig 12C.

October LE3.6

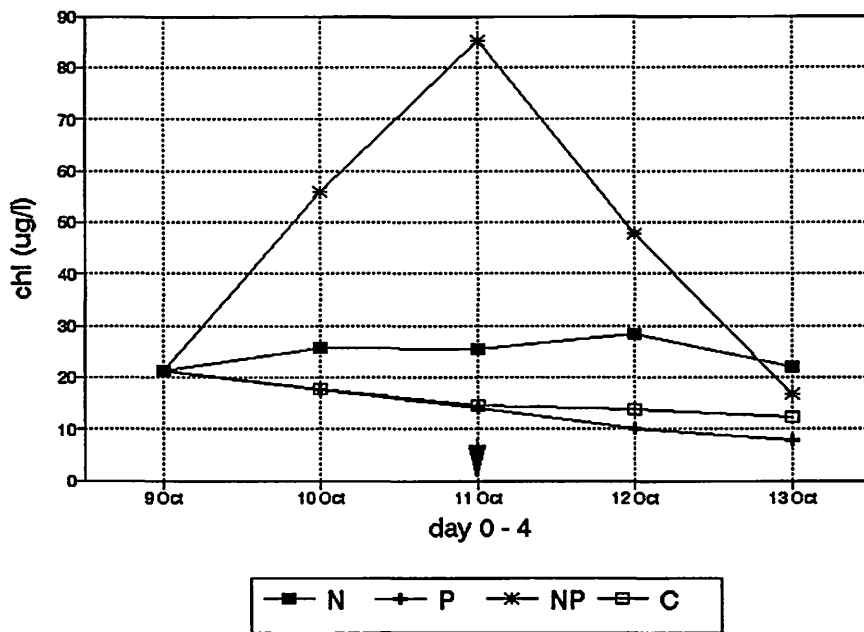


Fig 12D.

October LE5.5

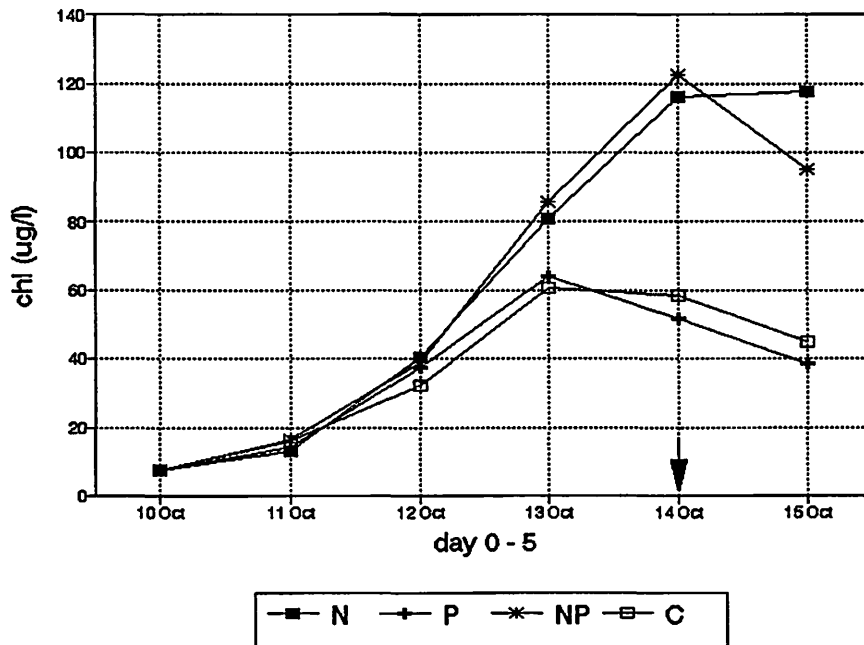




Fig 12E.

### October CB6.1

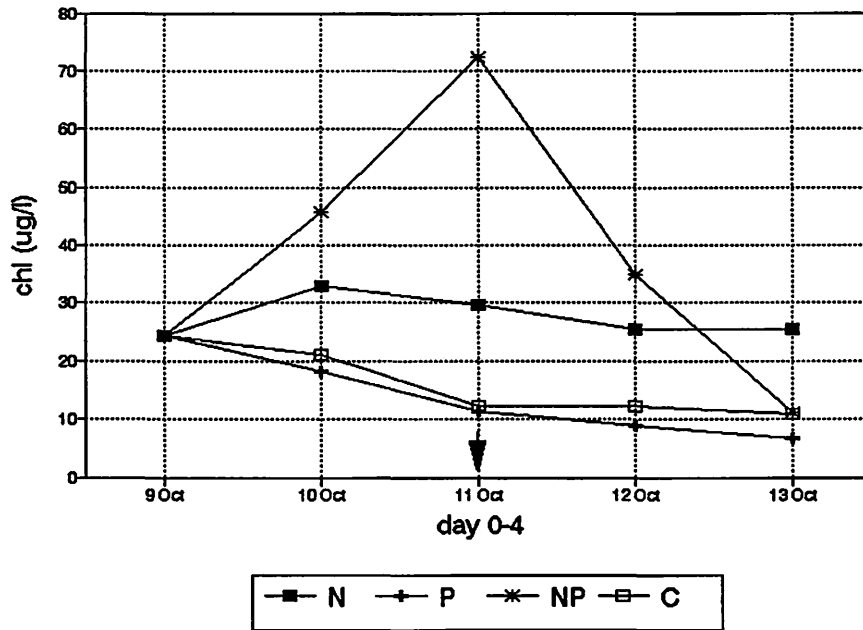


Fig 12F.

### October CB6.4

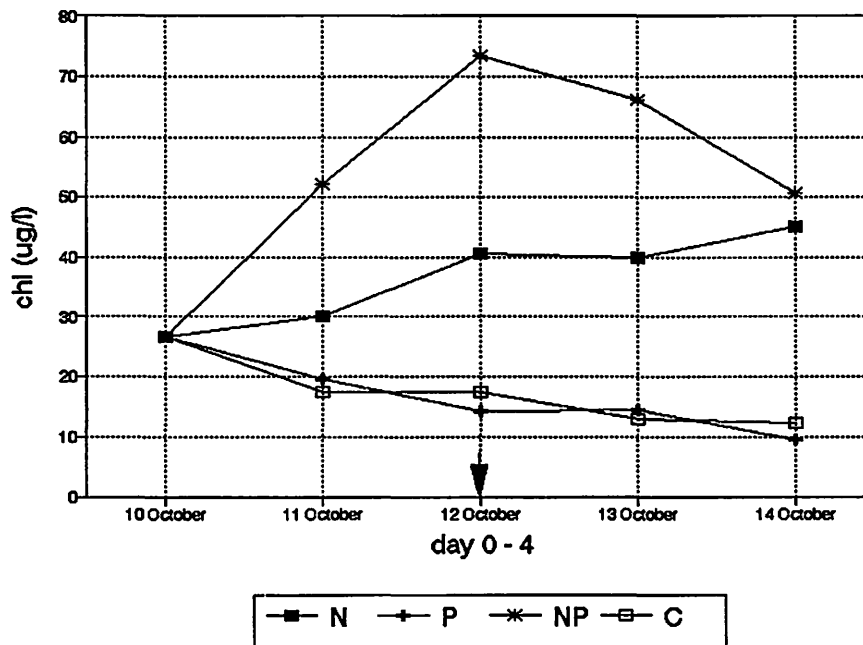


Fig 13A.

November TF3.2

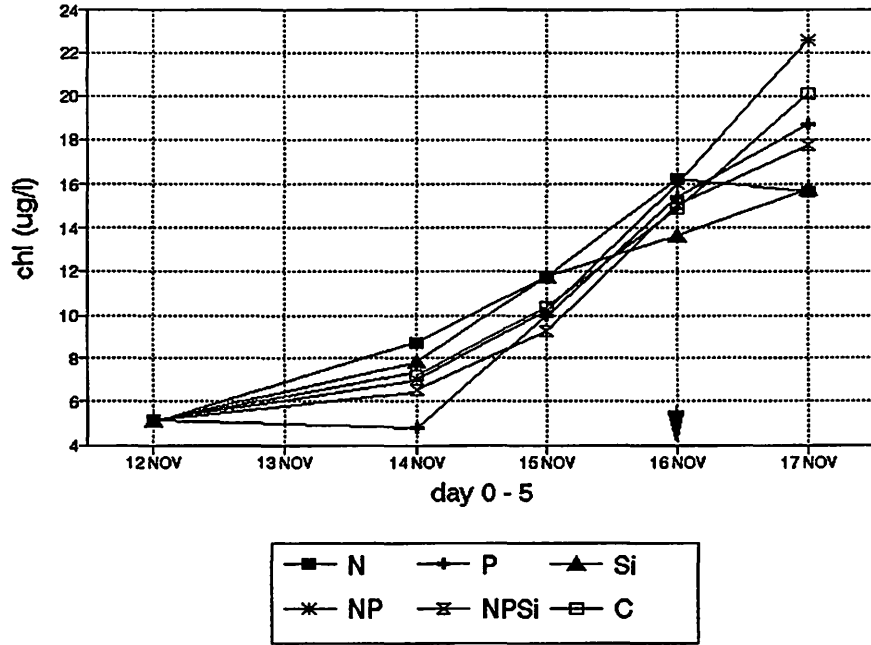


Fig 13B.

November TF5.5

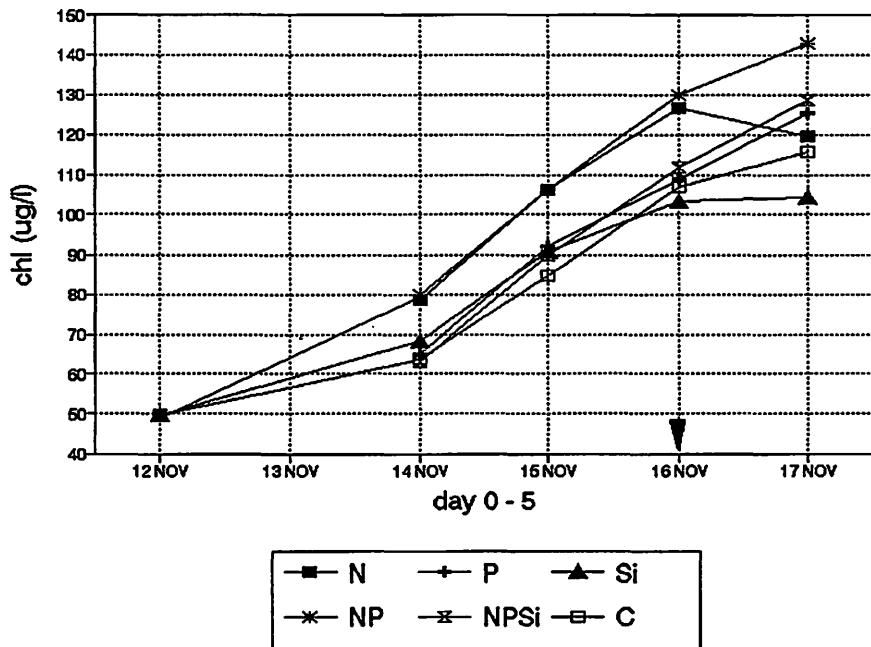


Fig 13C.

November LE3.6

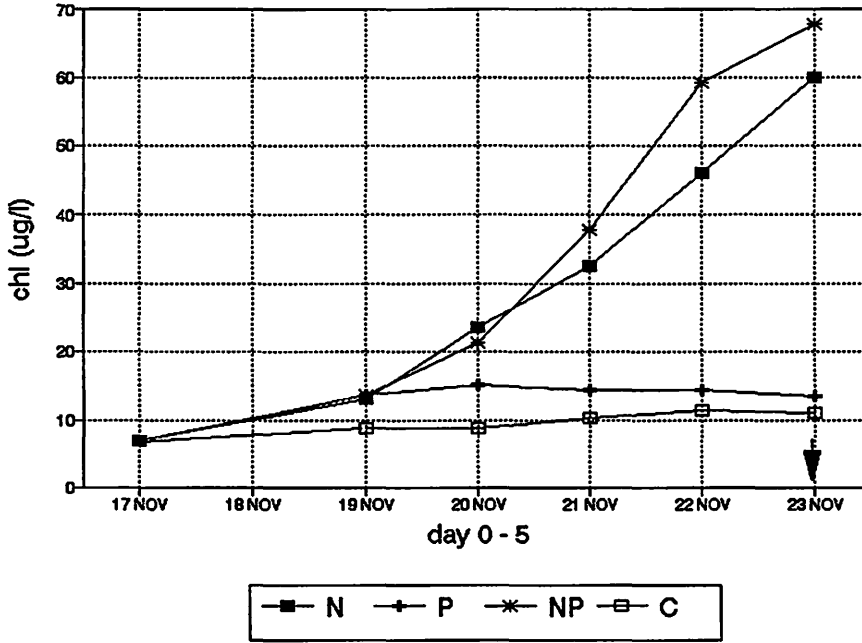


Fig 13D.

November LE5.5

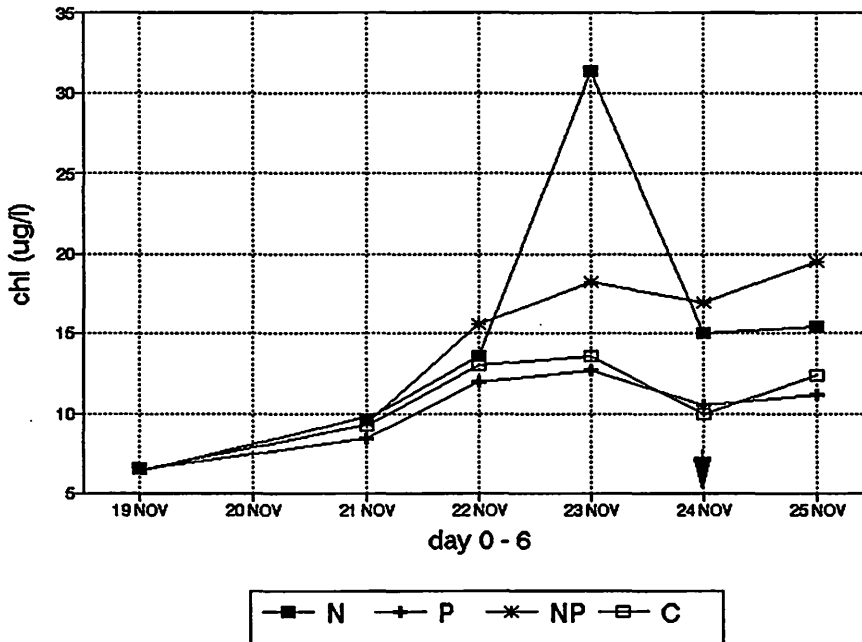


Fig 13E.

November CB6.1

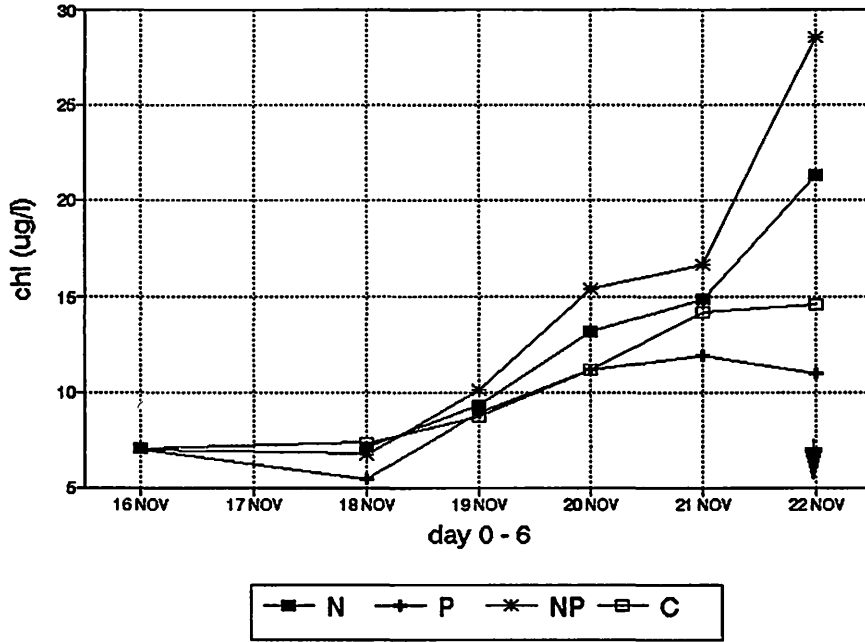


Fig 13F.

November CB6.4

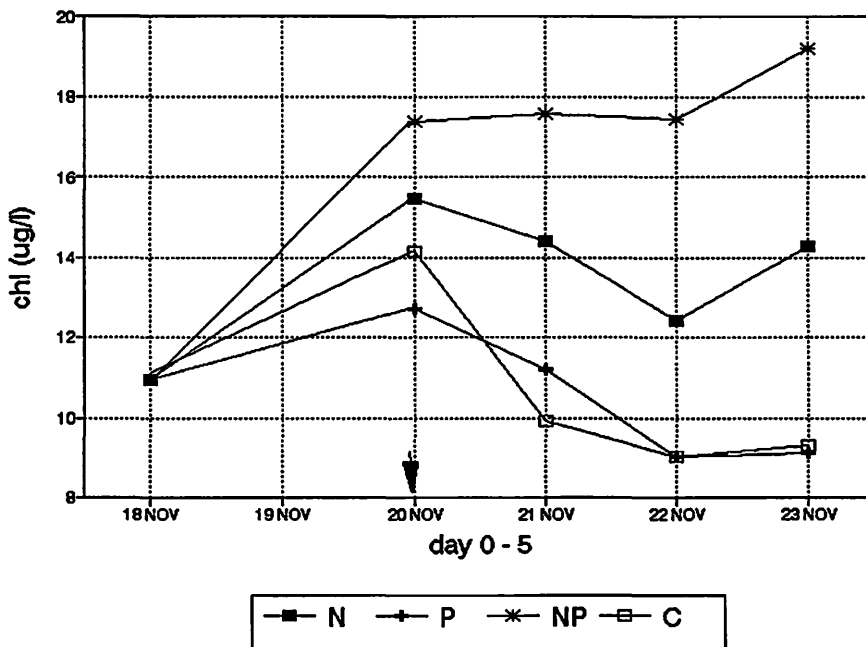


Fig 14A.

January TF3.2

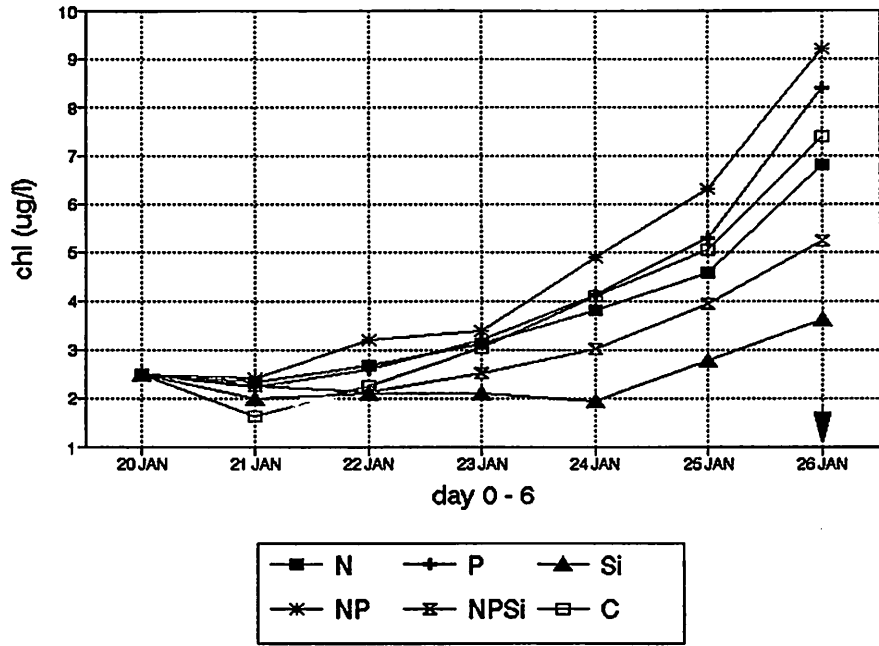


Fig 14B.

January TF5.5

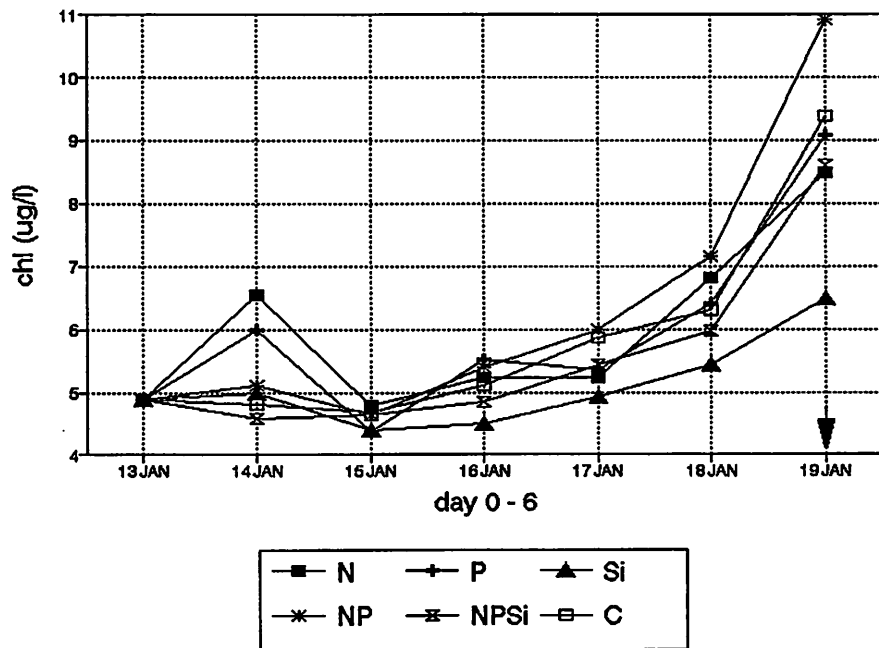


Fig 14C.

January LE3.6

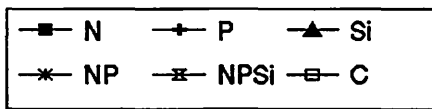
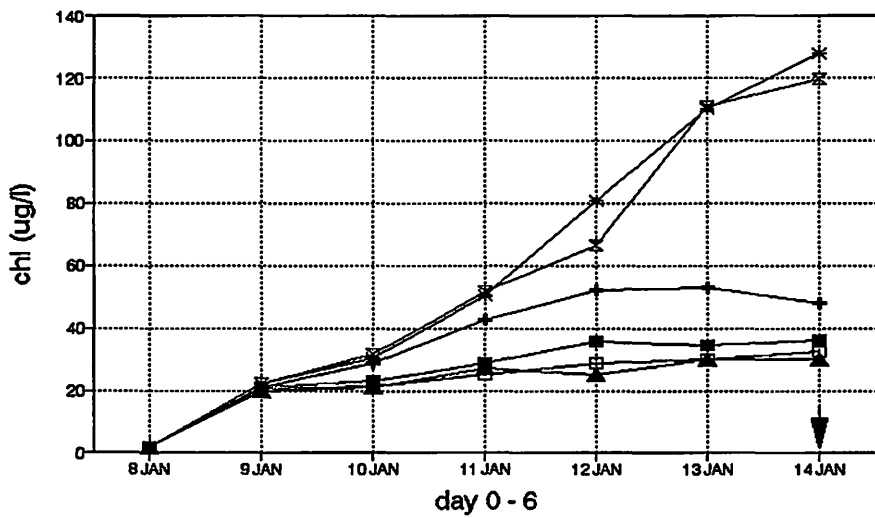


Fig 14E. January CB6.1

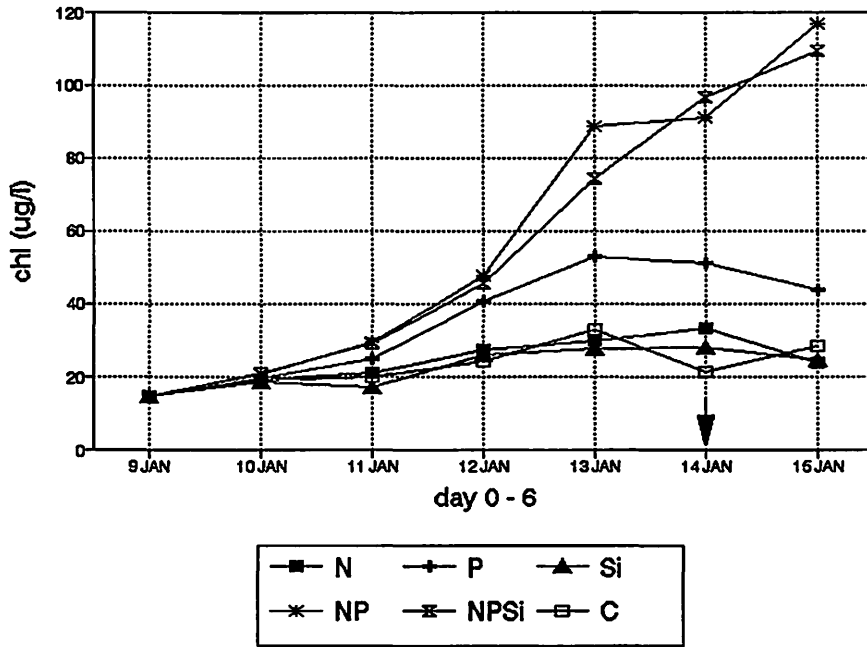


Fig 14F. January CB6.4

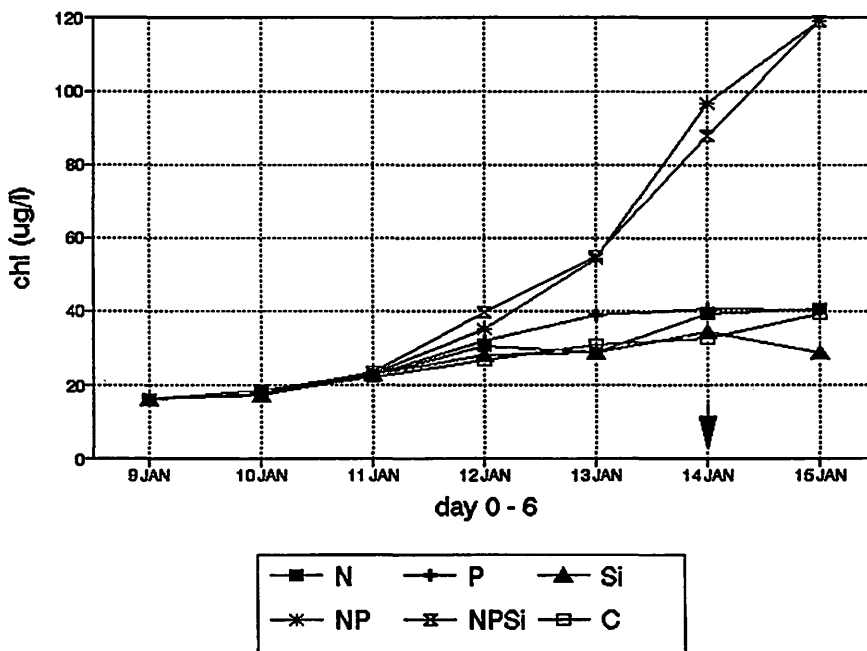


Fig 15A. February TF3.2

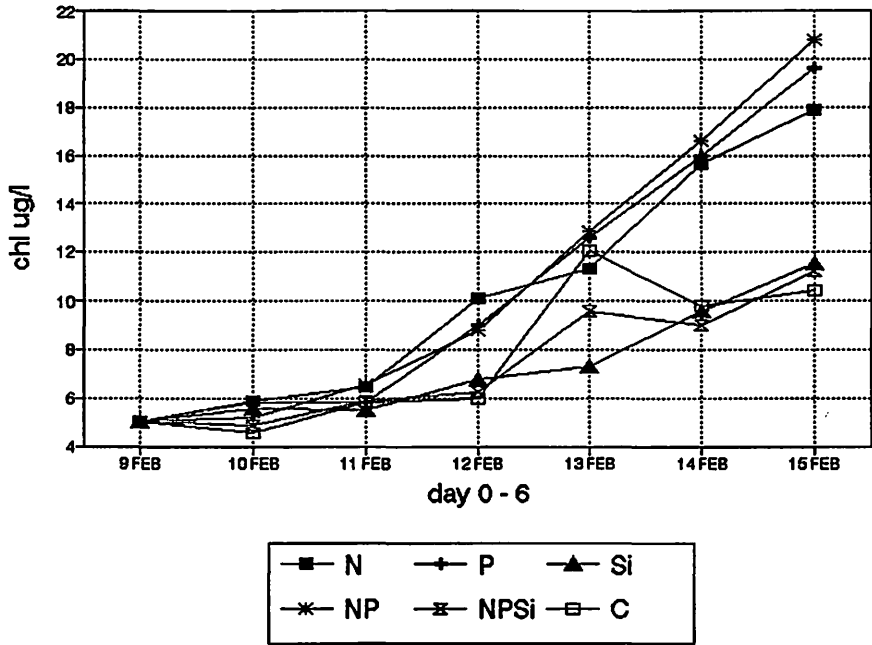


Fig 15B. February TF5.5

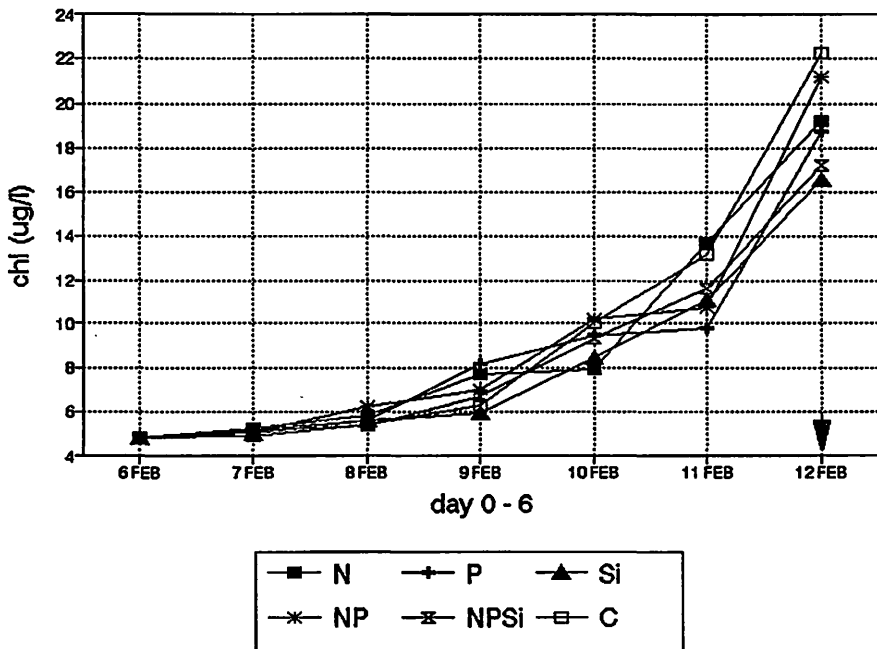




Fig 16A. Enrichment Index  
TF 3.2

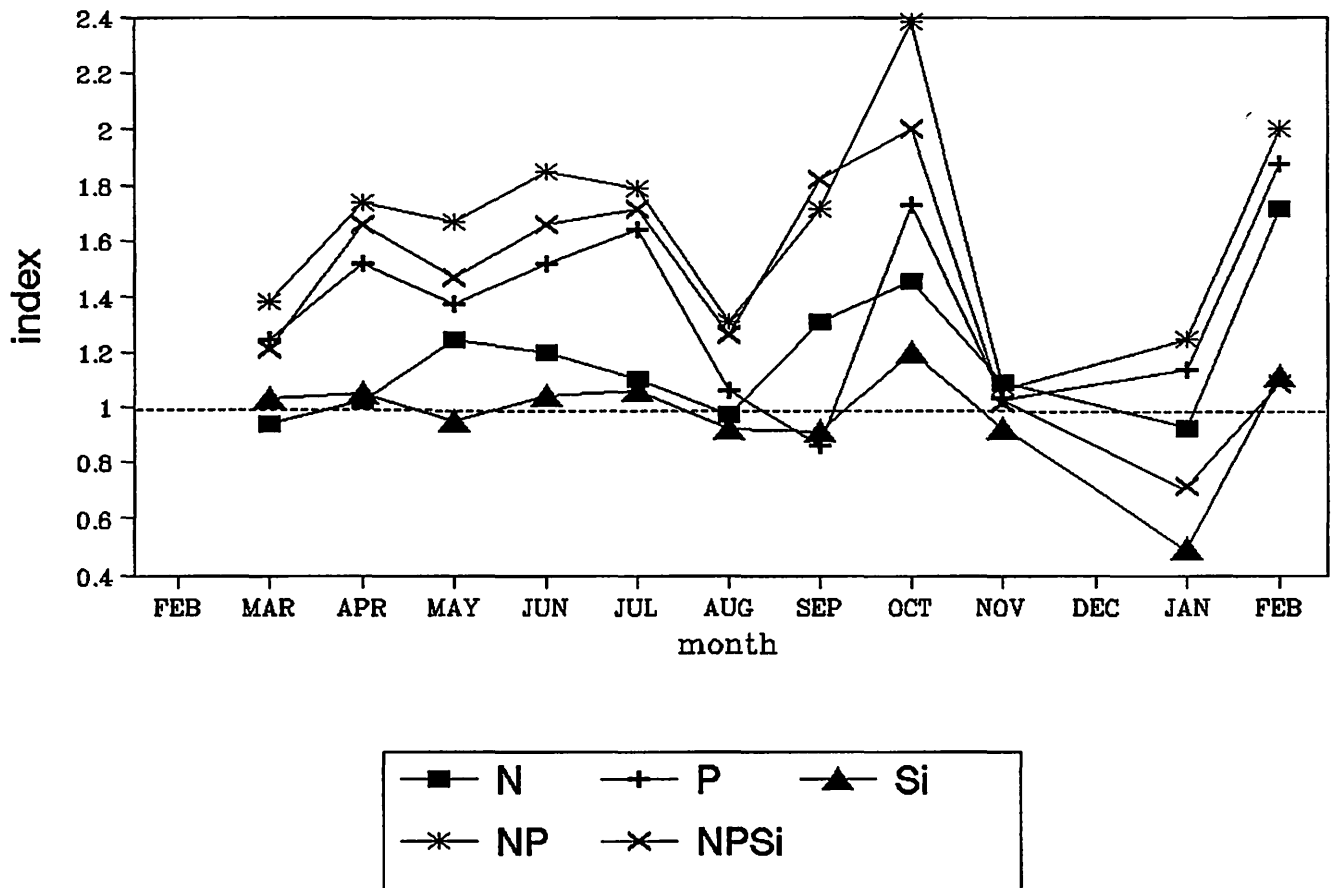


Fig 16B. Enrichment Index  
TF 5.5

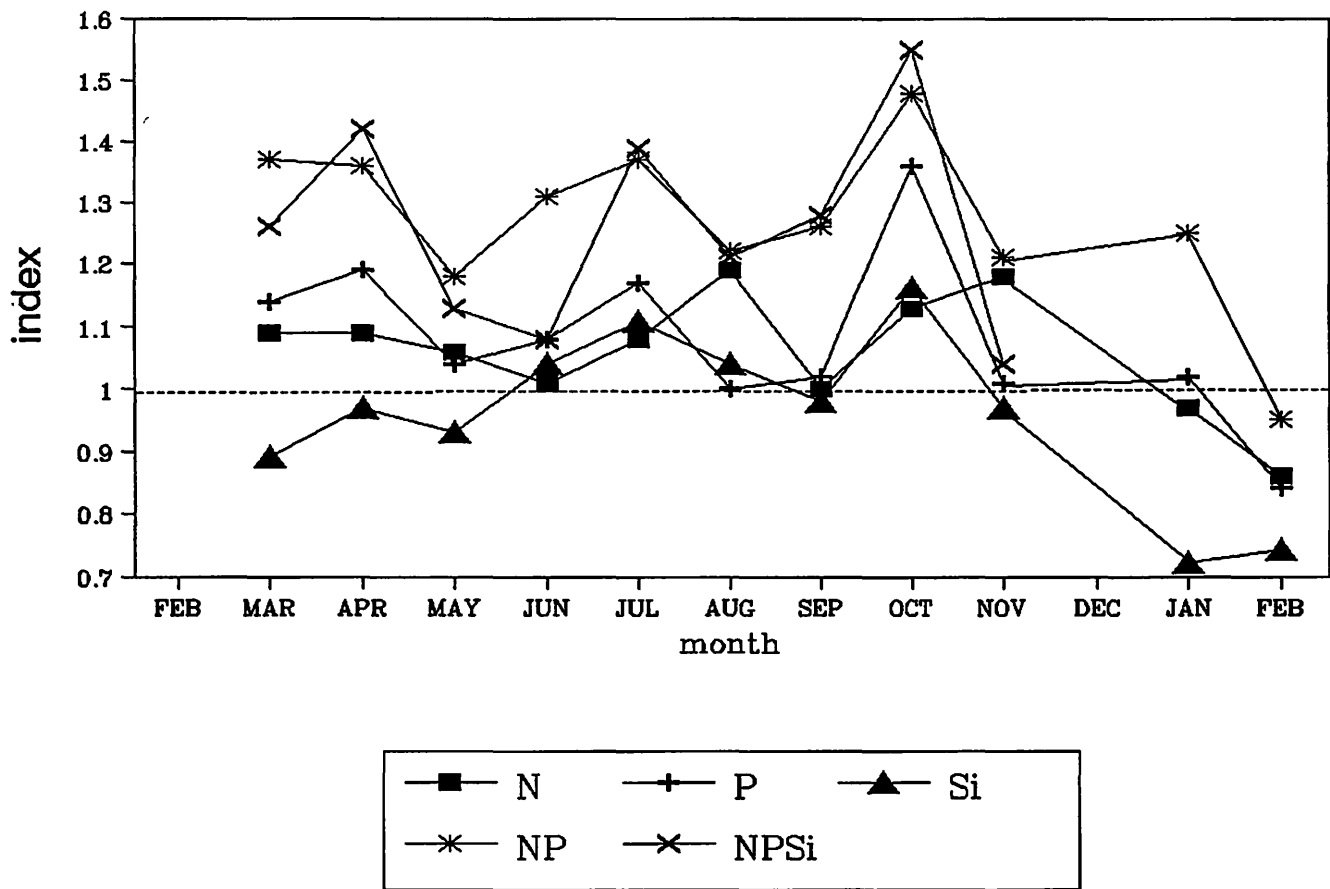


Fig 16C. Enrichment Index  
LE 3.6

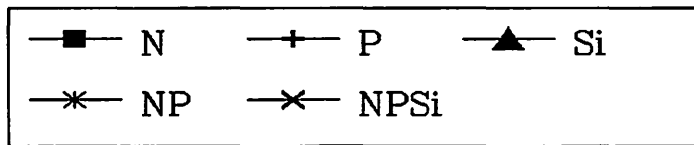
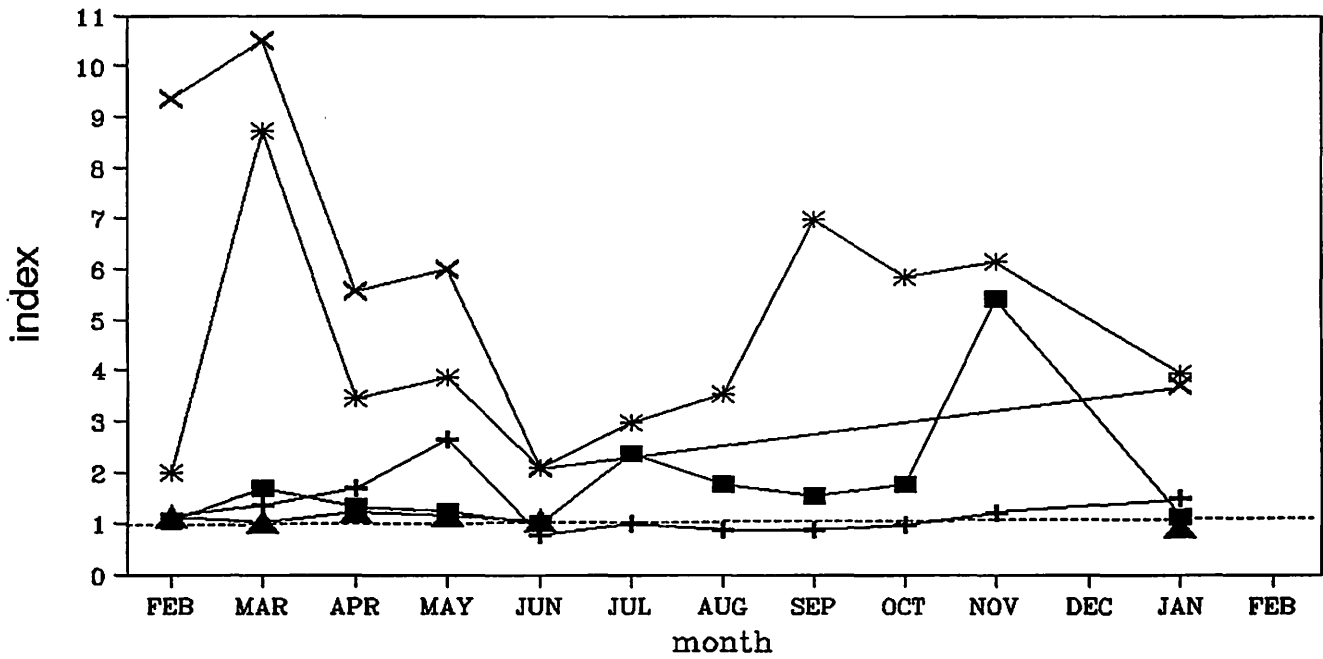


Fig 16D. Enrichment Index  
LE 5.5

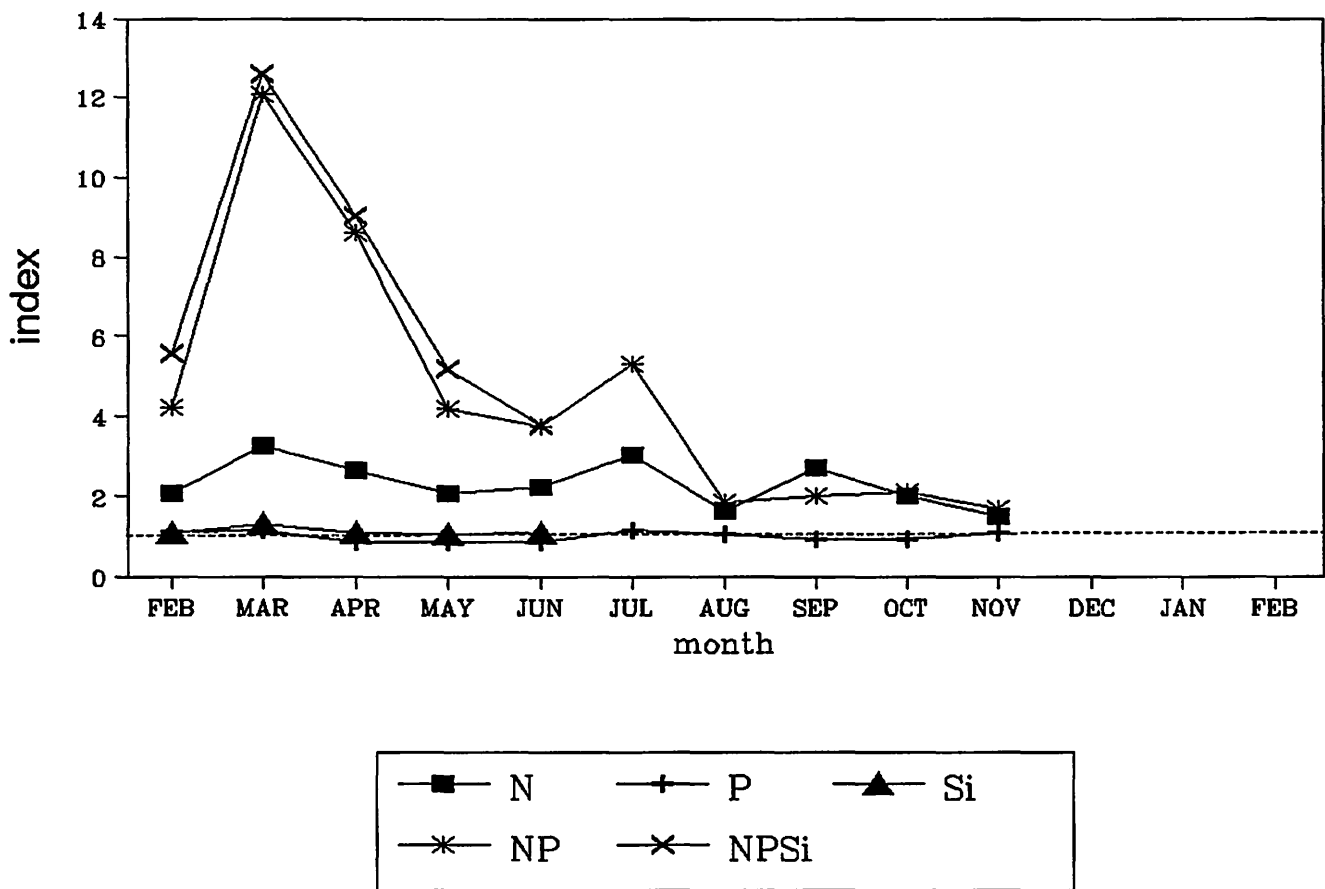


Fig 16E. Enrichment Index  
CB 6.1

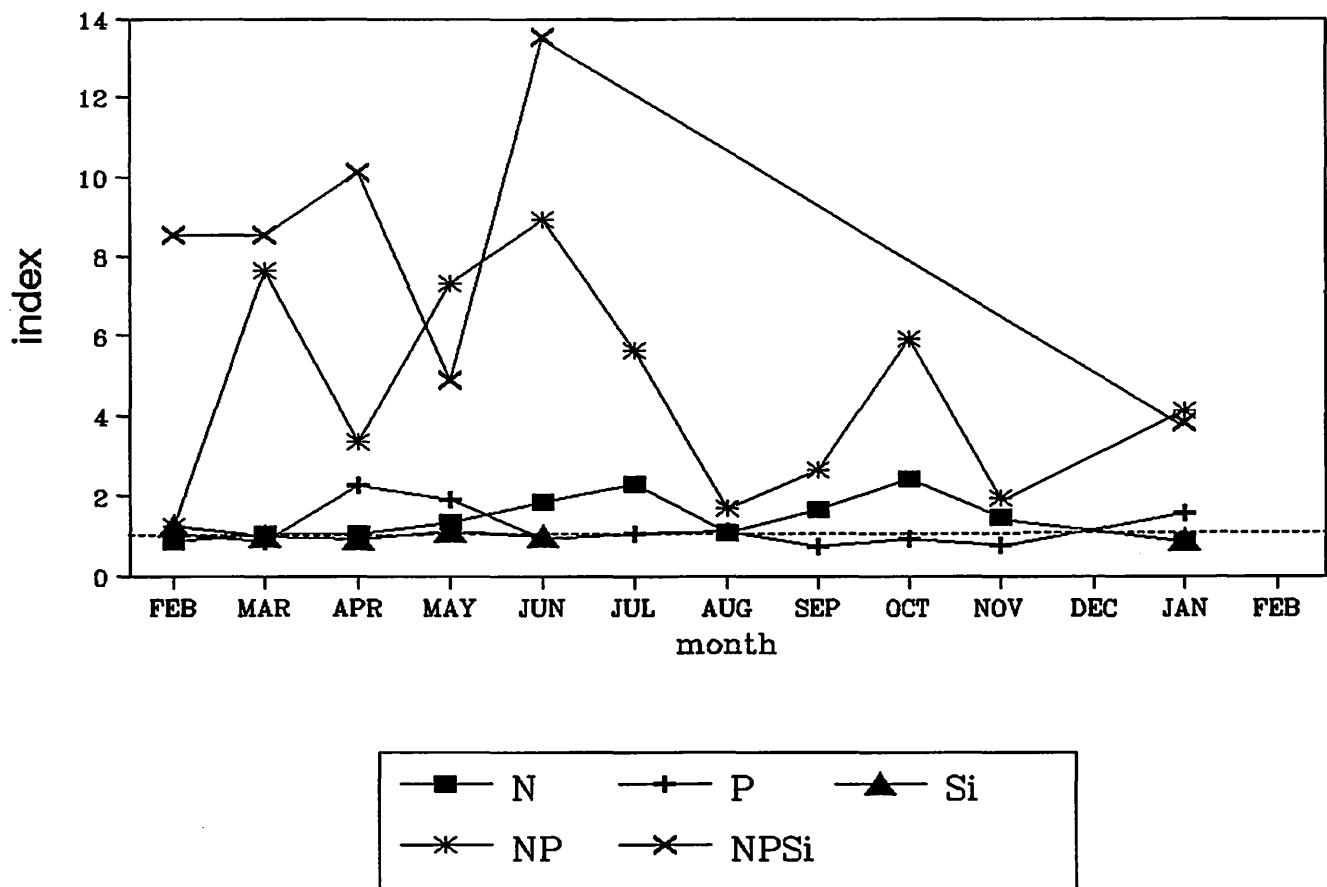


Fig 16F. Enrichment Index  
CB 6.4

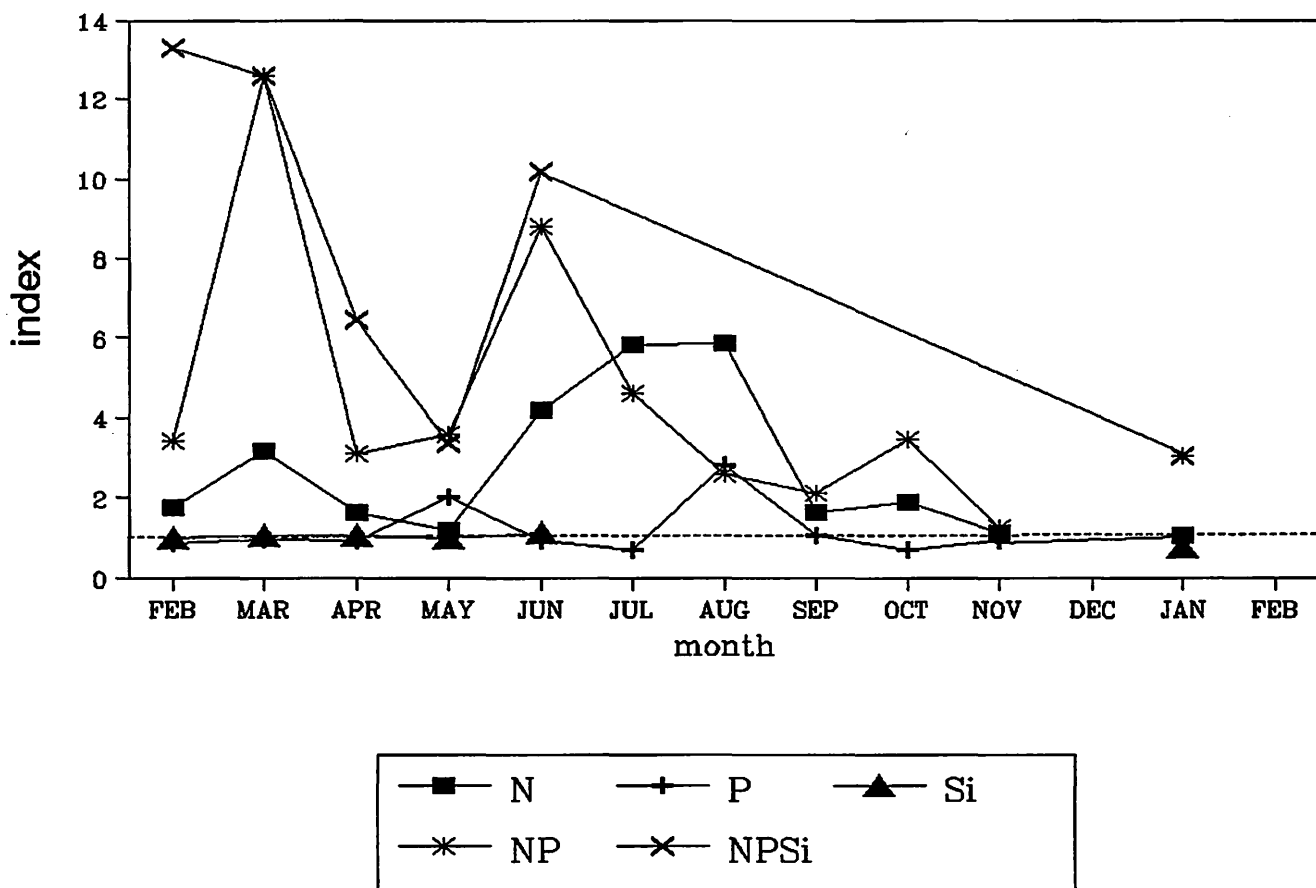


Fig 17.

