Mineral Nutrients in Native Vegetation on Atlantic Coastal Plain Soil Types

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The classification of a soil type or any specified area of soil in terms of the nutritional quality of food or feed crops growing thereon requires the collection and analysis of a large number of samples because of the wide variation in the composition of plants growing under apparently uniform conditions. A study of the vegetation on five soil types in six areas of the Atlantic Coastal Plain showed that sampling leaves of the swamp blackgum, *Nyssa sylvatica* Marsh. var. biflora (Walt.), was an efficient method of estimating the cobalt, copper, manganese, and phosphorus status of native forage types in this region. For this purpose two samples of blackgum leaves were adequate for grouping soils with respect to the mineral nutrients, cobalt and copper. The cobalt content of the soil was not a sufficiently sensitive estimation of that in the plant to permit it to be used to estimate the cobalt concentration in the vegetation.

THE MINERAL NUTRIENT CONTENT in vegetation from the same or similar sites is often difficult to interpret because of the great variability in the composition of the samples collected. The causes of such variability are often obscure, although some factors such as the portion of the plant taken, the relative position of a leaf or other organ of the plant, age of the plant, and microvariations in soil and climate are frequently suspected. Hence, large numbers of samples are sometimes required to establish significant differences between locations (*2*, *5*). The investigation reported here was designed to determine the magnitude and character of such variation and to find means of coping with the problem.

Experimental Plan

This experiment was designed to control as completely as possible such factors as soil, variety, age, and portion of the plant selected, and its location within a uniform area of a well-defined soil type. On the basis of previous studies (*2*, *5*) six plots were selected in 1952 in North and South Carolina so as to provide a wide variation in certain mineral elements in the vegetation. One area, in the Hofmann Forest in North Carolina, was known to produce forage of poor nutritional quality, because cattle could not survive there unless fed supplements of cobalt and copper. In the other areas, no cattle have been observed that could be used as indicators of forage quality. All of the areas were cutover forest lands, and showed no indications of any recent agricultural use.

Within each area a plot of a single soil type was delineated and classified. Within the limits of visual techniques, every effort was made to ensure soil uniformity throughout each plot. The boundaries of the plots were carefully marked and mapped so as to permit their re-establishment at any time in the future. Their exact location will be supplied by the United States Plant, Soil, and Nutrition Laboratory upon request. All of the soils are either poorly or somewhat poorly drained and vary in surface texture from fine sands to loams. All plots lie on either the Talbot or Penholoway terraces of the Atlantic Coastal Plain. The soil types and locations are presented in Table I.

Five plants of each of five species of native browse and grass were located on Plots I and II. A list of the plant species, their common names, and the portion of the plant sampled are presented in Table II. The location of each plant was marked with an iron stake

Table I. Locations of Plots and Classifications of Soils

Plot No.	Location	Soil Type	Terrace[a]	Approx. Size, Acre
I	Hofmann Forest, Jones Co., N. C.	Ona fine sand	Penholoway	1/2
II	Croatan Forest, Craven Co., N. C.	Bladen fine sandy loam, gravelly substratum phase	Talbot	1/8
III	Sampit Forest, Georgetown Co., S. C.	Coxville silt loam	Talbot	1/2
IV	Oak Ridge Bay, Williamsburg Co., S. C.	Rains fine sandy loam	Penholoway	1/2
V	Friendship Church, Georgetown Co., S. C.	Ona fine sand	Talbot	1/3
VI	Bullhead Bay, Berkeley Co., S. C.	Bladen loam	Penholoway	1/3

[a] Talbot terrace lies between 25 and 42 feet; Penholoway between 42 and 70 feet above sea level.

Table II. Plants Selected for Sampling with Description of Part Taken for Analysis

Common Name	Scientific Name	Part Taken for Analysis
Swamp blackgum	*Nyssa sylvatica* Marsh. var. biflora (Walt.) Sarg.	Leaf
Broomsedge	*Andropogon glomeratus* (Walt.)	Leaf blade
Gallberry	*Ilex glabra* (L.) Gray	Leaf with current year's stem
Pepperbush[a]	*Clethra alnifolia* L.	Leaf
Sweetbay[b]	*Magnolia virginiana* L.	Leaf

[a] Found in the Hofmann and Croatan areas only.
[b] Not found in the Bullhead Bay area.

and a numbered tag. Whenever possible, groups of two or more species growing in close proximity were selected to permit comparison under conditions of minimum soil variation. This procedure was repeated on all plots, except that sweetbay was not located on Plot VI and pepperbush was found only on Plots I and II.

Vegetation samples were first collected about June 1, 1952. At that time not more than a third of the leaves of a small plant was collected, the balance being reserved for subsequent collections about the middle of July and finally about October 1. It was not possible to follow this procedure in the case of broomsedge, because one clone was barely sufficient for each sample. Occasionally, to obtain a minimum of 6 to 10 grams of dry matter, it was necessary to select the broomsedge from an area as large as 100 square feet. Thus, 375 samples in 1952 and 345 in 1953, when no pepperbush samples were taken, were selected from the six plots.

Soil samples were taken at the time the soil types were identified and on later dates in 1952 to obtain samples throughout the season. In 1953, additional samples were taken from locations where two plants of a given species within a plot differed significantly in their content of a specific micronutrient element. The first samples in 1952 consisted of two composites of corresponding horizons, each composite prepared from three profiles located at random in each half of a plot. Composited surface samples at each of these profile locations were again taken in July and October of 1952. The samples taken in 1953 represented horizons of a profile adjacent to the plant under study.

All plant material was dried at 70° C. for 24 hours, crushed by hand where possible or cut into small pieces with steel shears. Analyses of the samples were made following published procedures (*1*, *7*, *11*) for cobalt, copper, manganese, calcium, and phosphorus.

Results

All data for each element determined were subjected to an analysis of variance to determine the significance of effects of location or plot, date of sampling, and year of sampling. In addition, the data from each soil type or plot were examined to determine the occurrence of significant variations within plots. The data from different plots were ranked and separated according to the method proposed by Duncan (*6*). In no case was any interpretation based on a level of significance less than 1%. The data for each element can best be presented individually, because there appeared to be no relationship among them.

As a standard of comparison, the combined data on composition of broomsedge for both years (1952 and 1953) were used. It was assumed that broomsedge as a member of the grass family would be representative of most of the common grasses growing in the region and grazed by cattle in the forested areas. By combining all samples of this species a practical estimate of the relative quantities of mineral nutrients derived from each soil and available to the animal was thus provided.

Cobalt The concentration of cobalt in each of the five species collected from Plot I and determined throughout the season is shown in Figure 1. The differences between blackgum and pepperbush leaves on the one hand and gallberry and sweetbay leaves and broomsedge on the other are so large that different scales must be used to represent the levels of cobalt in each group. The species in the former group, for example, contain from 70 to 100 times as much cobalt per unit of plant tissue as do those in the latter group. A similar relationship, varying only in magnitude, was found for the cobalt in these plants obtained from the other plots. Hence, data for Plot I only are presented in Figure 1 and all following figures. Significant increases ($P \leq 0.01$) in the cobalt concentration in blackgum and pepperbush were found as these plants matured (Figure 1), but no significant changes occurred in the other species from this or the other plot.

There is a wide range of cobalt values in relation to the vegetation on different plots (Table III). The mean cobalt content of broomsedge from Plot V is below the minimum level (0.04 to 0.07 p.p.m.) required by cattle (*3*). A consideration of the 2 years of data shows a highly significant difference ($P \leq 0.01$) between Plots V and I and between Plots III and VI, the latter being also the division between minimum and adequate levels of cobalt (*3*). The difference between Plots I, II, and III is not of importance nutritionally, because grass from all these plots contains only a minimum quantity of cobalt.

An examination of the cobalt concentration in blackgum samples collected during the 2 years revealed a relationship among plots similar to that shown by broomsedge. Thus, the ranking and separation of plots on the basis of significant differences in the concentration of cobalt in either of the two species were the same. Furthermore, it was apparent that fewer samples of blackgum leaves than broomsedge were required to recognize these differences among the plots. The data in Table III show, for example, that separation of plots on the basis of two samples of blackgum leaves collected in July 1952 was sufficient for practical nutritional purposes. The six plots were separated by blackgum leaves into two groups one of which in terms of broomsedge supplied insufficient to borderline and the other supplied borderline to sufficient quantities of cobalt. Similar results were obtained on the 1953 samples. Examination of blackgum data for other sampling schemes did not indicate any closer relationship to the broomsedge data.

A further consideration of the broomsedge data revealed no possibility of reducing the number of samples of this material and obtaining sufficient information to permit adequate evaluation of the plots. From the gallberry data for July of either year it was possible to identify the high cobalt plots, but intermediate plots could not be separated (Table III) from Plot V. Gallberry data for all collection dates in either year were not superior in this respect to the July data. No complete separations of plots could be made on the basis of cobalt in the leaves of the sweetbay.

Copper The five species studied fall into two groups with respect to their copper concentration (Figure 2). The range of copper values was very narrow for all species at all locations, and would generally be considered low with respect to nutritional adequacy. In general, the concentration was highest in June and lowest in October ($P \leq 0.01$ for each species).

A consideration of all broomsedge samples permitted a separation of Plots I, IV, and V as being relatively low in the copper concentration in their vegetation and Plots III and VI as relatively high (Table IV). Definite minimum values for nutritionally adequate con-

Table III. Concentration of Cobalt in Vegetation with Significant Differences ($P \leq 0.01$) in Ranked Data

	Cobalt, P.P.M. in Dry Matter		
Plot	Broomsedge[a]	Blackgum[b]	Gallberry[c]
V	0.03 < 0.04	1.7 < 20.0	0.03 < 0.12
I	0.04 < 0.06	3.1 < 20.0	0.04 < 0.12
II	0.06 < 0.08	8.6 < 20.0	0.07 < 0.12
III	0.06 < 0.08	22.5 ...	0.07 < 0.12
VI	0.08 ...	20.0 ...	0.09 ...
IV	0.09 ...	22.0 ...	0.12 ...

[a] Ranked means of 30 samples collected on three dates in each of 2 years (1952 and 1953); neither year nor date of collection was a significant factor in the variation in the data.
[b] Means of first 2 of 5 samples collected in July 1952 (results confirmed in July 1953).
[c] Means of 5 samples collected in July 1952 (results confirmed in July 1953).

centrations of copper in forage have not been established, but a level of 5 p.p.m. in the dry matter may be assumed (*8*). Hence, the separation of these plots on the basis of a significant difference in the copper content of broomsedge samples has a tentative nutritional significance.

Data from both blackgum leaves and gallberry leaves and stems were similar to broomsedge in ranking and characterizing the plots. If all data on these two species obtained in either 1952 or 1953 are considered, the plots can be separated into three groups, but such a separation would have little nutritional significance. A less precise but practical separation of the plots can also be obtained by considering certain July samples of broomsedge, blackgum, and gallberry collected in either year (Table IV). The copper content of the first two samples of blackgum leaves collected in 1952, for example places Plots I, II, and V in a low group, and III, IV, and VI in a high group. It is clear that Plots II and IV are intermediate with respect to the other plots, however, and probably cannot be definitely classified by any sampling scheme studied.

Table IV. Concentration of Copper in Vegetation with Significant Differences ($P \leq 0.01$) in Ranked Data

Plot	Copper, P.P.M. in Dry Matter: Broomsedge[a]	Broomsedge[b]	Blackgum[c]	Gallberry[b]
I	4.3 < 5.1	4.5 < 5.6	6.6 < 10.2	4.1 < 6.0
V	4.3 < 5.1	4.6 < 5.6	6.7 < 10.2	4.3 < 6.0
IV	4.4 < 5.1	4.8 < 5.6	10.2 ...	5.4 ...
II	4.9 ...	4.8 < 5.6	7.5 < 10.2	4.0 < 6.0
III	5.1 ...	5.2 ...	11.3 ...	6.0 ...
VI	5.4 ...	5.6 ...	10.6 ...	4.7 < 6.0

[a] Ranked means of 30 samples collected on 3 dates in each of 2 years (1952 and 1953). The year of collection was not a significant factor in the variation in the data. Date of collection within years was significant ($P \leq 0.01$).
[b] Means of 5 samples collected in July 1952 (results confirmed in July 1953).
[c] Means of first 2 of 5 samples collected in July 1952 (results confirmed in July 1953).

Manganese The data for Plot I presented in Figure 3 show a wide variation in manganese concentration in the different species examined. For all plots there was some variation in the data with respect to date of collection. Thus, there was a significantly higher concentration of manganese in broomsedge on the first date of collection ($P \leq 0.01$) and a higher concentration in gallberry leaves and stems on the last day of collection ($P \leq 0.05$ in 1952, $P \leq 0.01$ in 1953). No effect of date of collection on composition was observed for sweetbay leaves; the variation in blackgum leaves, while significant each year at the 5% level, was difficult to interpret for trends.

The variations in manganese concentration with respect to plots are presented in Table V. No significant differences were found for Plots I, II, III, V, and VI, but the soil of Plot IV can readily be separated as supplying a significantly higher ($P \leq 0.01$) amount of manganese to the vegetation grown on it. The data in Table V show that a separation equal to that obtained with the samples of broomsedge collected on three dates in 2 years was obtained with either broomsedge or blackgum leaves collected only in July of either year.

Table V. Concentration of Manganese in Vegetation with Significant Differences ($P \leq 0.01$) in Ranked Data

Plot	Manganese, P.P.M. in Dry Matter: Broomsedge[a]	Broomsedge[b]	Blackgum[b]
VI	49 < 126	46 < 131	306 < 539
I	52 < 126	53 < 131	246 < 539
III	53 < 126	36 < 131	174 < 539
V	56 < 126	57 < 131	207 < 539
II	57 < 126	63 < 131	166 < 539
IV	126 ...	131 ...	539 ...

[a] Ranked means of 30 samples collected on three dates in each of 2 years (1952 and 1953). Date of collection within years was significant at the 1% level and year of collection was significant at the 5% level.
[b] Means of 5 samples collected in July 1952 (results confirmed in July 1953).

Phosphorus The concentration of phosphorus in all species from any of the six plots is very low. Except for the early June period, the quantity present never exceeded 0.10% and frequently fell below 0.05%. The lowest mean concentration of phosphorus was found in the leaves and current year's stem growth of the gallberry in both 1952 and 1953 (Figure 4). Except at the time of the first collection in June 1952, there was no difference in phosphorus concentration among the other species. In agreement with known behavior of this element, the concentration was highest in the spring, with little further change between July and October.

Based on the analysis of samples collected over the 2-year period, the six plots can be divided into three groups with respect to the concentration of phosphorus in broomsedge (Table VI). Plots II, III, and V fall in the lowest group, IV and VI fall in the medium, and I falls in the high group. Practically the difference between the low and medium group is of no importance. The phosphorus content of broomsedge in

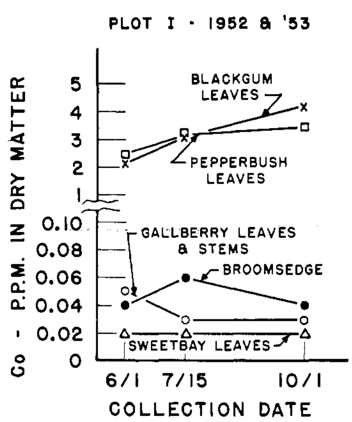

Figure 1. Concentration of cobalt in each of 5 plant species collected on each of three dates

Each datum is mean of 2 years except for pepperbush

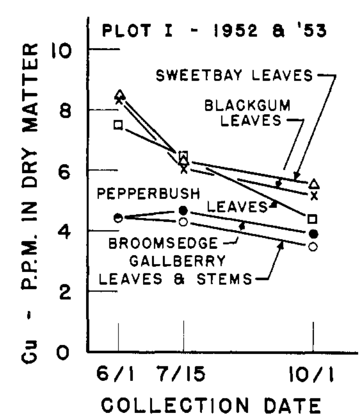

Figure 2. Concentration of copper in each of 5 plant species collected on each of three dates

Each datum is mean of 2 years except for pepperbush

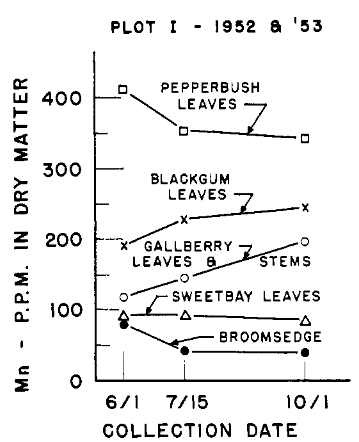

Figure 3. Concentration of manganese in each of 5 plant species collected on each of three dates

Each datum is mean of 2 years except for pepperbush

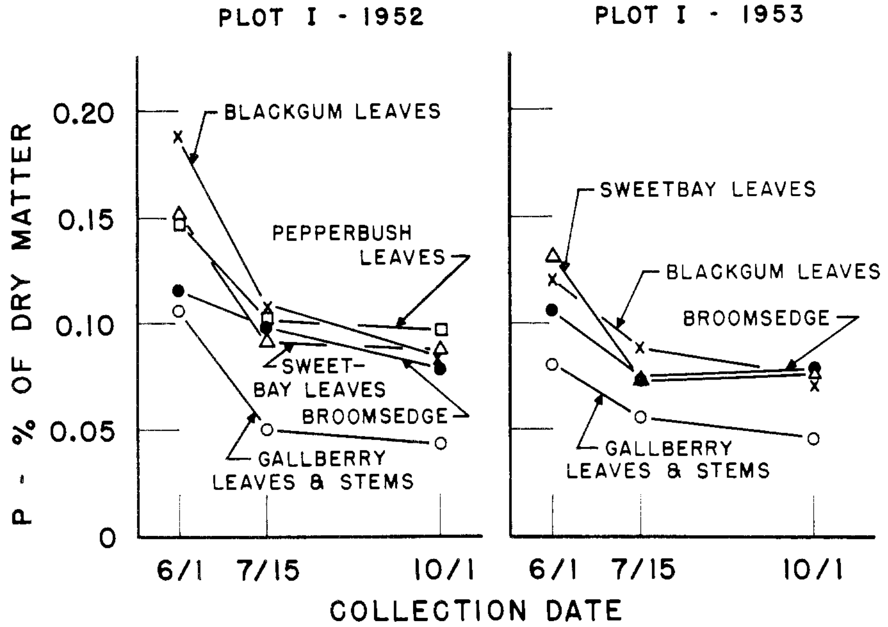

Figure 4. Concentration of phosphorus in each of 5 plant species collected on each of three dates

Each datum is mean of 2 years except for pepperbush

Plot I is borderline and the difference between vegetation from this and the other plots is accepted as being of minor importance nutritionally.

The data in Table VI show that the first two samples of either broomsedge or blackgum leaves collected on each of the three dates in 1952 would also have separated the vegetation from Plot I as having a higher concentration of phosphorus than that from the other five. A number of other sampling schemes were tested, but none gave consistent results in both years. Thus, all five samples of broomsedge or blackgum, collected in July 1952, resulted in as good separation of plots as did a consideration of 15 samples collected on the three dates. However, in 1953, no significant differences in the concentration of phosphorus were found when only the July broomsedge samples were considered, and a difference only at the 5% level of significance was found in the blackgum samples for July 1953. Neither the gallberry nor the sweetbay leaves effected the same separations of the six plots as did the broomsedge.

Calcium The concentration of calcium in the five species showed a 15-fold variation from a low of less than 0.1% in broomsedge to a high of 1.4% in the leaves of the pepperbush. Except in broomsedge, the calcium concentration increased as the leaves matured (Figure 5), the most striking change occurring in the pepperbush. There was no significant difference in the calcium concentration in blackgum, sweetbay, and gallberry leaves, but broomsedge and pepperbush were each clearly different from this group.

From a consideration of the calcium content of all samples of broomsedge collected in both years (Table VII), the plots can be separated into three groups at the 1% level of significance. Thus, broomsedge in Plots I and III contained significantly less calcium than that in II, IV, and V, while the broomsedge in Plot VI contained a significantly higher concentration of calcium than is in this species from either of the other groups.

To determine the minimum number of samples of broomsedge that would have given an adequate separation, data from those collected only in 1952 or 1953 were subjected to analysis (data for 1952 only are shown in Table VII). Plots I and III were again shown to be significantly low ($P \leq 0.01$) but the separation from II and V was not so clear as with the analysis using all data. However, with both sampling schemes, Plot VI stood out as producing a grass with a significantly higher content of calcium. For all practical purposes, it is clear that a collection of samples in either year would have supplied as much information about the classification of plots and the nutritional quality of this forage as measured by calcium as did the 2 years.

A further analysis was made of the samples collected in July of both years. The data given in Table VII show that only Plot I is clearly separated from the higher level Plots IV, V, and VI, while Plot VI stands out as a significantly high calcium plot. For a practical nutritional interpretation, however, this amount of information is of value. No other sampling scheme with broomsedge or with any other species permitted an equally good separation of areas.

Cobalt Content of Soils The total cobalt content and that soluble in 1 to 1 hydrochloric acid in soils from each of the six plots are presented in Table VIII. The soils have been ranked according to the cobalt content of broomsedge growing thereon. The soil cobalt values show a limited relative agreement with those in the vegetation (Tables III and VIII). Plot IV, however, which supports vegetation with the highest cobalt content, has substantially less soil cobalt than has Plot III, which supports vegetation containing significantly less cobalt than does plot IV.

Discussion

Although a number of soil-plant composition studies have been reported (*3, 9, 12*), no adequate estimation of a normal variation in the mineral composition of plants grown under uniform conditions is available. That the range is large, however, is clearly indicated by the number of samples required to characterize a limited area of a single soil type. Furthermore, the usual range of concentration of cobalt in most vegetation is so low, particularly in the grasses, that variations of severalfold in samples from one plot are not uncommon. One advantage of the blackgum leaf lies, therefore, in the larger quantity of cobalt contained in it and the smaller relative variation between samples. Hence, if sufficient relationship can be established

Table VI. Concentration of Phosphorus in Vegetation with Significant Differences ($P \leq 0.01$) in Ranked Data

	Phosphorus, % of Dry Matter		
Plot	Broomsedge[a]	Broomsedge[b]	Blackgum[b]
II	0.063 < 0.071	0.073 < 0.100	0.101 < 0.140
III	0.063 < 0.071	0.065 < 0.083	0.084 < 0.140
V	0.064 < 0.071	0.083 < 0.100	0.108 < 0.140
VI	0.071 < 0.093	0.075 < 0.100	0.105 < 0.140
IV	0.071 < 0.093	0.071 < 0.100	0.092 < 0.140
I	0.093 ...	0.100 ...	0.140 ...

[a] Ranked means of 30 samples collected on three dates in each of 2 years (1952 and 1953). There were highly significant ($P \leq 0.01$) differences between years and between dates within years (see Figure 4).
[b] Means of the first two samples collected on each of three dates in 1952. There were highly significant ($P \leq 0.01$) differences between dates. These results were confirmed in 1953.

between it and the forage species, the blackgum should serve as an excellent indicator for the cobalt status of the forage species. The usefulness of blackgum is limited, of course, by its habitat. Although this appears to be extensive in eastern United States, it is limited locally by moisture conditions. Fortunately, it is most frequently found in the Coastal Plain, where deficiencies of cobalt are commonly encountered (*4*).

While it is clear that some increase in the concentration of cobalt in blackgum occurs as the leaf matures, the change is small. Probably a reasonable period between the collection dates on several plots would not affect a comparison of the data. This observation appears to hold for the other species studied. This fact would permit the study of blackgum leaves collected over a wide region for a period of several weeks—a matter of importance where numbers of personnel are limited.

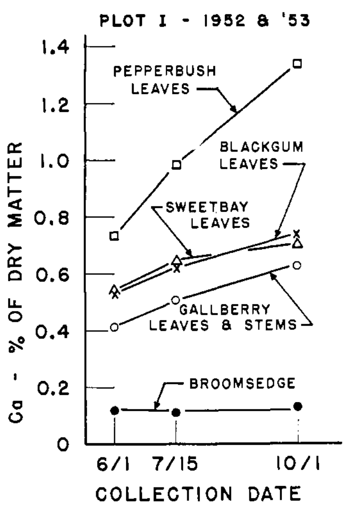

Figure 5. Concentration of calcium in each of 5 plant species collected on each of three dates

Each datum is mean of 2 years except for pepperbush

A further advantage of blackgum lies in the fact that its leaves, remote from direct soil contamination, can be collected in abundance. The size of the tree apparently has no effect on the concentration of cobalt in the leaf, small and large trees in close proximity to each other being similar in this respect.

The composition of the blackgum leaf is more consistent with that of broomsedge in respect to copper, manganese, and phosphorus than were the other species studied. Hence, with the exception of calcium, this species can generally be useful in estimating the quality of native forages with respect to their mineral nutrients. It is apparent that for this purpose not less than two and preferably more samples must be collected from any unit area. Of course, much more experience will be needed in interpreting the mineral status of soils of widely differing characteristics and their vegetation from blackgum composition data. About 0.5 pound of dry black gum or pepperbush leaves from a cobalt deficient area and containing 1.5 to 2.0 p.p.m. of cobalt would, along with the other browse and forage, supply the minimum daily requirement of cattle.

Any variation in the mineral composition of plant tissue due to date or year of collection appears to be no greater than variation among samples from the same soil. Data on the within-plot variation obtained in this study (but not presented in detail here) show that the cobalt content of blackgum leaves sampled on one date from different plants on one plot may differ by as much as twofold. While the magnitude of these variations was often less, similar differences were found on all three sampling dates and in both years. The cause for such variations was not determined, but they were not related to the size or age of the plant, the position of the leaf on the plant, or to the total or acid-soluble cobalt in the soil. Variations of threefold were found in the cobalt content of broomsedge from one plot, but they were not so consistent from one date to another or one year to another as were variations in blackgum.

Microrelief on the plots with slight differences in drainage patterns could account for some variation in uptake of minerals. Thus, on one plot, there was a significantly higher concentration of manganese in the vegetation growing on a narrow strip through the center. The availability of manganese may be greater in the presence of excessive moisture (*10*), and this condition probably obtains in the center of this frequently flooded area.

Important within-plot variations in calcium were observed in many instances. In most cases the plant high in calcium in June was also high throughout the season, but in only a few cases did one plant remain high in the second year. There appeared to be little relation to variation of soil pH within a plot. Differences between plots, except in the extreme cases, were not correlated with pH of the soils. A further investigation of variation in calcium and the other mineral constituents of the plant samples in relation to physical and chemical properties of the soil is now in progress.

In agreement with earlier results (*2*, *5*), it is interesting to note that the concentrations of cobalt and phosphorus are significantly lower in vegetation from the Ona soil on the Talbot terrace than from this soil on the Penholoway terrace.

Table VII. Concentration of Calcium in Broomsedge with Significant Differences ($P \leq 0.01$) in Ranked Data

	Calcium, % in Dry Matter		
Plot	*1952 and 1953*[a]	*1952 only*[b]	*July 1952 and 1953*[c]
I	0.12 < 0.17	0.11 < 0.16	0.11 < 0.19
III	0.13 < 0.17	0.12 < 0.17	0.14 < 0.25
II	0.17 < 0.23	0.17 < 0.23	0.17 < 0.25
V	0.17 < 0.23	0.16 < 0.23	0.19 ...
IV	0.20 < 0.23	0.19 ...	0.19 ...
VI	0.23 ...	0.23 ...	0.25 ...

[a] Ranked means of 30 samples collected on three dates in each of 2 years (1952 and 1953); neither year nor date of collection was a significant factor in the variation in the data.
[b] Means of 15 samples collected on three dates. Date of collection was not a factor at the 1% level of significance.
[c] Means of 10 samples, 5 collected in July 1952, and 5 in July 1953. Year of collection was not a significant factor in variation in the data.

Table VIII. Concentration of Cobalt in Air-Dried Soils Compared to that in Broomsedge

	Cobalt, P.P.M.				
	Surface Soil[a]		Subsurface Soil[b]		
Plot No.	*Total*	*Acid-soluble*[d]	*Total*	*Acid-soluble*[d]	*Broomsedge*[c]
V	0.22	0.06	0.24–0.45	0.16–0.40	0.03
I	0.34	0.09	0.34–0.90	0.05–0.30	0.04
II	0.75	0.27	0.80–2.80	0.50–2.35	0.06
III	2.50	1.37	2.70–2.80	1.80–2.30	0.06
VI	2.14	1.52	2.65–3.90	2.30–3.85	0.08
IV	1.40	0.55	1.40–2.65	0.95–2.15	0.09

[a] The A_1 horizon. The depth varied from 0–4 to 0–8 inches in the different plots. Values are the means of data from two composites of corresponding horizons, each composite prepared from three profiles.
[b] The horizons below the A_1. In general, the cobalt content increased with depth of the profile. The values given are for the second and the lowest horizons.
[c] See Table III.
[d] 1 to 1 hydrochloric acid.

In general, the level of minerals in vegetation from the Talbot terrace is low with respect to higher terraces, although it has not always been possible to show a significant difference from that on the Penholoway terrace.

Conclusions

An intensive study over a period of 2 years of the concentration of cobalt, copper, manganese, calcium, and phosphorus in five native browse and forage species showed that sampling leaves of the swamp blackgum, *Nyssa sylvatica* Marsh. var. biflora (Walt.) Sarg., was, except for calcium, an efficient method of estimating the mineral status of native forage species.

Two samples of blackgum leaves from each soil plot were adequate for grouping soils with respect to the mineral nutrients, cobalt and copper, in broomsedge. Five samples were required for estimating the nutritional adequacy of phosphorus and significant differences in the manganese in broomsedge. Black gum was superior in this respect to other plant species tested.

The best estimate of the calcium status of these soils was obtained from five samples of broomsedge collected from each plot in July of either of two years. Although some relationship of soil cobalt to the cobalt concentration in broomsedge and blackgum leaves was apparent, the cobalt content of the soil was not a sufficiently sensitive estimation of that in the plant to permit its classification in terms of nutrient requirements of animals.

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Literature Cited

(1) Beeson, K. C., *J. Assoc. Offic. Agr. Chemists* **36**, 405–11 (1953).
(2) Beeson, K. C., *Soil Sci.* **80**, 211–20 (1955).
(3) Beeson, K. C., U. S. Dept. Agr., Agr. Inform. Bull. **7**, March 1950.
(4) Beeson, K. C., Lazar, V. A., Boyce, S. G., *Ecology* **36**, 155–6 (1955).
(5) Beeson, K. C., Matrone, G., "Copper Metabolism," pp. 370–98, W. D. McElroy, B. Glass, eds., Johns Hopkins Press, Baltimore, Md., 1950.
(6) Duncan, D. B., Virginia Polytechnic Inst. Tech. Rept. **6a**, mimeo (1953).
(7) Gregory, R. L., Morris, C. J., Ellis, G. H., *J. Assoc. Offic. Agr. Chemists* **34**, 710–16 (1951).
(8) Guilbert, H. R., Gerlaugh, P., Madsen, L. L., "Recommended Nutrient Allowances for Domestic Animals," Natl. Research Council (U. S.), Rept. **4**, Committee on Animal Nutrition (1945).
(9) Hill, A. C., Toth, S. J., Bear, F. E., *Soil Sci.* **76**, 273–83 (1953).
(10) Mulder, E. G., Gerretsen, F. C., *Advances in Agron.* **4**, 221–77 (1952).
(11) Parks, R. Q., Hood, S. L., Hurwitz, C., Ellis, G. H., *Ind. Eng. Chem., Anal. Ed.* **15**, 527–33 (1943).
(12) Price, N. O., Linkous, W. N., Engel, R. W., J. Agr. Food Chem. **3**, 226–9 (1955).

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PESTICIDE RESIDUES

Spectrophotometric Determination of Heptachlor and Technical Chlordan on Food and Forage Crops

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To secure approval from federal and state regulatory agencies for the use of heptachlor and technical chlordan, it was necessary to measure the residue resulting from treatment of food and forage crops with these toxicants. Analytical methods were tested to find which were most specific and which were the least affected by plant extractives and other pesticides. The Davidow method for chlordan and the Polen-Silverman method for heptachlor were modified by microtechniques to detect toxicant in the 2.5- to 5.0-γ range. Methods were developed which used chromatography to separate as little as 2γ (or 0.01 p.p.m.) of toxicant with recovery in the range of 80% from as much as 2 kg. of crop material. Good agreement with bioassay methods was found. Analyses of a large number of crops treated with recommended dosages of heptachlor and chlordan show no significant residue present at harvest time.

The need for accurate methods for the specific determination of micro quantities of chlordan (technical chlordan, the principal component of which is 1,2,4,5,6,7,8,8-octachloro-3a,4,7,7a-tetrahydro-4,7-methanoindan) and heptachlor (1,4,5,6,7,8,8-heptachloro-3a,4,7,7a-tetrahydro-4,7-methanoindene) on food and forage crops has led to the investigation of the published methods of determining these toxicants. The methods based on total chlorine were examined but were not adopted because of their lack of specificity. The colorimetric methods of Allesandrini and Amormino (*1*), Ard (*2*), Davidow (*3*), Harris (*4*), Palumbo (*8*), and Polen and Silverman (*9*) were investigated. Microtechniques were developed to use the Davidow method for chlordan and the Polen-Silverman method for heptachlor.

The crops are ground, dried chemically with anhydrous sodium sulfate, and extracted with "colorimetric" pentane. The pentane extracts are processed to remove plant pigments and waxes and are treated with the color-forming reagent. The intensity of the color formed with the reagent is determined with a spectrophotometer adapted for sampling on a micro scale. The toxicant content is then interpolated from a