

# THE NUTRITIONAL STATUS OF THE COTTON PLANT AS INDICATED BY TISSUE TESTS<sup>1</sup>

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(WITH ONE FIGURE)

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## Introduction

Tissue tests applied to corn and various truck crops have afforded a convenient means of estimating the nutritional status of the plant. The application of suitable tissue tests to cotton should be of similar value. The determination of the portion of the plant to be used, the relation existing between the availability of certain elements and the accumulation of these elements in the sampling tissue, are among the problems which must be solved so that results obtained from tissue tests may be interpreted properly.

Tissue tests developed at Purdue University are being used in diagnoses of nutrient deficiencies in corn (8, 12, 13, 14). According to HOFFER (8) the best time to make tests on corn is when the plants are well along in maturity. The value of information obtained late in the season is limited, however, by the fact that corrective measures cannot be taken until the following growing season.

CAROLUS (1) employed quantitative tissue-testing methods using cabbage and potatoes as his experimental plants. He noted that a deficient supply of nitrogen was reflected in a high concentration of soluble phosphorus and a low concentration of soluble nitrate nitrogen in the experimental tissue. A reverse of the above situation was observed when phosphorus was the limiting element. A potassium deficiency resulted mainly in a low concentration of that element and in some cases was accompanied by increased amounts of soluble nitrate nitrogen. Magnesium deficiency was generally associated with a low concentration of nitrogen. WOLF and ICHISAKA (16) drew similar conclusions as to the relationship existing between nitrogen and phosphorus.

EMMERT (5) utilized plant tissue tests as a guide in fertilizing tomatoes. He concludes that determinations of soluble nutrients are of greater value as tissue tests than total analyses, which often obscure important variations in the nutrients concerned.

## Analytical methods

The tissue to be analyzed was collected in small glass vials, stoppered and held in a refrigerator until extracted. Fresh samples were chopped,

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weighed (1 gram fresh or 200 mg. dry weight), placed in 50-ml. Erlenmeyer flasks, and allowed to stand for 48 hours in 25 ml. of sodium acetate solution (9). When dried samples were used it was found that a few milliliters of ethyl alcohol as a wetting agent facilitated the extraction. After the extraction period, measured quantities of activated carbon were added to each flask (11). The flasks were shaken and the contents filtered through Whatman's number 12 filter paper. The resulting clear solutions were then used for chemical analysis. Soluble nitrate nitrogen, phosphorus, potassium, calcium, and magnesium concentrations in the cleared extracts were determined by colorimetric and turbidimetric measurements using a Klett-Summerson photoelectric colorimeter. Nitrate nitrogen was determined by a modification of the brucine hydrochloride method as described by PEECH and ENGLISH (11). It was found necessary to allow the samples to stand 10 minutes on a steam bath after the addition of the sulphuric acid. Phosphorus was determined by the aminonaphtholsulphonic acid method given by WOLF (15).

The sodium cobaltinitrite method for potassium as given by WOLF (15) was found satisfactory for this work. The addition of a few drops of a 1% solution of gum arabic to the aliquots taken for the potassium determination stabilized the suspension. Magnesium measurements were made by the titan yellow method and calcium by the ammoniacal citrate procedure as given by PEECH and ENGLISH (11).

### Experimental

#### THE SELECTION OF SAMPLING TISSUE

It was necessary at the beginning of this work to determine which portion of the cotton plant would be most suitable for tissue testing. The selection of the tissue to be employed would depend upon: (a) ease of handling, (b) concentrations of the various nutrients within the tissue and (c) response of these concentrations to variations in the substrate.

On June 20, 1947, Stoneville 2-B upland cotton was planted in 3-gallon glazed pots filled with washed creek sand. After emergence of the seedlings one liter of a half strength Hoagland solution (7) was applied daily to each pot. On July 3, the plants were thinned to three per pot. From that date on the plants were watered with tap water in which the pH had been adjusted to 5.5 with a few drops of  $\text{HNO}_3$  (4). On each of the following dates—August 1, 8, 15, and 22—typical plants were harvested and divided into their component parts. Analyses for soluble  $\text{NO}_3$  nitrogen, phosphorus and potassium were made and these results are presented graphically in figure 1.

Flowering started on August 10 and it may be observed that this change in the physiological activity of the plant is reflected in reduced concentrations of nitrogen and phosphorus in all tissue analyzed. Except for the main stem petioles, the same was true for the concentration of potassium in the various plant parts (figure 1).

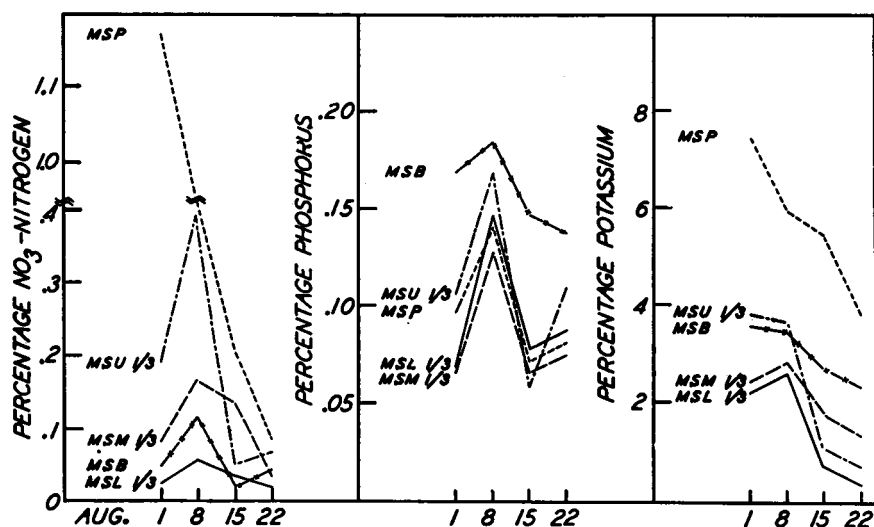


FIG. 1. Soluble  $\text{NO}_3$  nitrogen, phosphorus, and potassium in various portions of the cotton plant harvested at weekly intervals. Flowering started August 10. MS, main stem; L, lower; M, middle; U, upper; P, petioles; B, blades.

On examination of the data presented in figure 1, it can be seen that the petioles of the main stem were consistently high in  $\text{NO}_3$  nitrogen and potassium. The  $\text{NO}_3$  nitrogen of the main stem increased in concentration as the stem apex was approached. The leaf blades were noticeably higher in phosphorus although the petioles maintained a concentration of phosphorus which was favorable for analysis by the procedures employed. As the petioles satisfied the first two criteria for the selection of the sampling tissue set forth earlier in this paper, they were used in further investigations as the experimental portion of the plant.

#### INFLUENCE OF NUTRIENT SUPPLY ON THE ACCUMULATION OF IONS IN THE PETIOLES

Stoneville 2-B upland cotton was planted in 2-gallon glazed pots filled with washed creek sand in the greenhouse. Eight days after planting the seedlings were thinned to one per pot and the first nutrient applications were made. Nutrients were added every 15 days and the quantities applied were determined by the absorption curves for cotton grown on Cecil sandy loam as found by OLSON and BLEDSOE (10). Nutrient applications were made 15 days in advance of these absorption curves. The plants were divided into 13 groups corresponding to the 13 nutrient treatments employed in this investigation. The base treatment contained twice the quantities of nitrogen, phosphorus, potassium, calcium, and magnesium found in the plants analyzed by Olson and Bledsoe, thus insuring a sufficient supply of nutrients for good growth. Sulphur was added at approximately the same

rate as phosphorus. Finely ground commercial magnetite was added to the sand as a source of iron at the rate of 0.2%. EATON (3) and later CHAPMAN (2) have shown that ample iron is supplied in sand cultures by this method. Other trace elements were applied during the course of the investigation. Plants receiving the base or "high" treatment were supplied with the following total amounts of nutrients during the course of the investigation:  $\text{NO}_3$  nitrogen 5.300 gm., phosphorus 1.116 gm., sulphur 1.172 gm., calcium 4.566 gm., potassium 4.174 gm., and magnesium 1.614 gm. The "medium" treatment for each element contained one-half of the quantity of that element in its respective high level. For the "low" treatment each element was dropped separately to one-sixth that quantity contained in the high series. This arrangement gave a high, medium, and low nutrient treatment for each element in which the quantities of the other elements employed did not vary from the base treatment.

This experiment was carried out during the winter of 1947-48 and again in the winter of 1949-50. In the experiment of 1947 the plants were watered carefully to avoid leaching. Although no serious wilting was noted it is felt that insufficient watering accounted for the relatively poor growth obtained. In the 1949 experiment each pot was provided with an enameled pan to catch the leachate, thus making it possible to water the plants freely. The leachate was returned to the pots periodically. Tissue test results of the two experiments closely parallel one another and in view of this only the 1949 data are presented in this paper.

Four tissue harvests were made, at 30, 60, 90, and 145 days after emergence. As the plants were too small at 30 days to provide sufficient petiole material, the entire aboveground portion was harvested. In order to standardize the selection of the sampling tissue, petioles from the third and fourth nodes from the main stem apex were taken on the other harvest dates. The samples were dried and chemical determinations were made as previously described.

Thirty days after emergence, plants receiving the low treatment of nitrogen, phosphorus, and potassium were significantly shorter than those grown in the high series as seen in table I. No real differences were noted in dry weight at this time. Results of the tissue tests on the 30-day sampling are given in table II. The soluble  $\text{NO}_3$  nitrogen, phosphorus, and potassium content of the aboveground portion of the plants had significant, positive correlation coefficients when compared with their respective supplies in the substrate. Several interactions were noted at this date. Both the soluble calcium and magnesium content of the plant tops were correlated inversely with the potassium supply. Soluble magnesium in the tops displayed the same correlation when compared with substrate calcium. These observations are in accord with results obtained by GARMAN (6) on the absorption of cations by the cotton plant. Nitrate nitrogen varied directly with the substrate potassium.

The analyses of height measurements taken 60 days after emergence

TABLE I  
HEIGHT, DRY WEIGHT AND YIELD AS INFLUENCED BY NUTRIENT TREATMENTS

Treatment	30-Day harvest			60-Day harvest			90-Day harvest			145-Day harvest			Average dry wt. bolls per plant gm.
	Height cm.	Dry wt. gm.		Height cm.	Dry wt. gm.		Height cm.	Dry wt. gm.		Height cm.	Dry wt. gm.		
Low nitrogen .....	15.9*	.390		51.5	5.11**		103			132*	36.1**		29.1*
Med. " .....	17.9	.460		69.6	9.61		122			131	65.8		59.8*
High " .....	19.0	.532		64.4	10.90		108			117	72.4		69.2
Low phosphorus ....	14.8**	.355		56.7	6.66**		129			144**	66.1		60.4
Med. " .....	17.9	.455		62.4	7.92**		115			125	65.0		65.8
High " .....	19.0	.532		64.4	10.90		108			117	72.4		69.2
Low sulphur .....	18.0	.455		62.8	6.72**		117			129	68.1		59.6*
Med. " .....	18.5	.450		65.4	8.66**		117			129	81.9*		65.5
High " .....	19.0	.532		64.4	10.90		108			117	72.4		69.2
Low calcium .....	17.8	.511		68.1	7.83**		125			137**	81.0*		62.2
Med. " .....	20.2	.509		65.5	10.59		113			122	73.0		62.5
High " .....	19.0	.532		64.4	10.90		108			117	72.4		69.2
Low potassium .....	15.8**	.427		61.3	8.34**		109			115	62.4*		59.7*
Med. " .....	18.4	.567		64.1	7.94		122			131	77.2		68.0
High " .....	19.0	.532		64.4	10.90		108			117	72.4		69.2
Low magnesium .....	17.8	.526		65.7	8.45**		124			129	75.8		69.3
Med. " .....	20.2	.605		71.6	9.56*		112			123	75.4		60.5
High " .....	19.0	.532		64.4	10.90		108			117	72.4		69.2

\* Significant to .05 level.

\*\* Significant to .01 level.

showed no effect of treatment, however, differences in dry weight were highly significant. Table III contains the results of tissue tests on the 60-day petiole samples. No direct comparison can be made between the 30- and 60-day harvests since the entire aboveground portion of the plants were taken on the former date. Soluble  $\text{NO}_3$  nitrogen and potassium in the petioles varied directly with their supply in the substrate. Positive inter-

TABLE II  
RESULTS OF TISSUE TESTS ON 30-DAY HARVEST  
(RESULTS BASED ON OVEN-DRY WEIGHT)

Treatment	Percentage in tops*				
	$\text{NO}_3 \text{ N}$	P	K	Ca	Mg
Low nitrogen .....	.138**	.342	4.04	3.40	.363
Med. " .....	1.187	.323-	3.92	3.71	.541
High " .....	1.623	.315	4.61	3.46	.536
$r =$	.874 §	...	...	...	...
Low phosphorus ...	1.520	.172	4.60	3.33	.595
Med. " ...	1.865	.249	3.76	3.44	.539
High " ...	1.623	.315	4.61	3.46	.536
$r =$	...	.858 §	...	...	...
Low sulphur .....	1.668	.332	4.71	3.56	.645
Med. " .....	1.376	.340	4.65	3.48	.565
High " .....	1.623	.315	4.61	3.46	.536
Low calcium .....	1.462	.268	4.44	3.65	.885
Med. " .....	1.412	.324	4.62	3.28	.648
High " .....	1.623	.315	4.61	3.46	.536
$r =$	...	...	...	...	-.885 §
Low potassium ....	1.225	.321	2.77	4.11	.808
Med. " ....	1.402	.335	3.84	3.96	.712
High " ....	1.623	.315	4.61	3.46	.536
$r =$	.547 ††	...	.882 §	-.771 §	-.905 §
Low magnesium ...	1.181	.319	4.48	3.57	.520
Med. " ...	1.110	.322	4.47	3.49	.573
High " ...	1.623	.315	4.61	3.46	.536

\* Entire aboveground portion of the cotton plant was harvested at this sampling.

\*\* Each figure is an average of six replications.

†  $r$  indicates significant correlation coefficients between the concentration of elements in the petioles and nutrient treatment.

†† Significant to .05.

§ Significant to .01 level.

relations can be seen in table III for petiole potassium and substrate nitrogen on one hand and substrate sulphur on the other. The soluble calcium and magnesium in the petioles again displayed significant negative correlation coefficients when compared with substrate potassium. Petiole calcium had a similar correlation when compared with substrate phosphorus.

Since entire plant samples were not taken at the 90-day harvest, only height measurements were made as an index of growth. No significant effect of treatment on plant height was noted. Chemical analyses of the

petioles showed that each element was correlated directly with its supply in the substrate (table IV). Again negative interactions can be seen in comparing petiole calcium and magnesium with substrate potassium. Petiole calcium was inversely correlated with substrate nitrogen, phosphorus, and sulphur. The inverse interaction between petiole phosphorus and substrate nitrogen is noted in table IV for the first time in the 1949 investigation. This relation was observed in the 60-day harvest of the 1947 experiment.

TABLE III  
TISSUE TEST RESULTS ON PETIOLE FROM 60-DAY HARVEST  
(RESULTS BASED ON OVEN-DRY WEIGHT)

Treatment	Percentage in petioles				
	NO <sub>3</sub> N	P	K	Ca	Mg
Low nitrogen .....	.009*	.183	6.45	2.04	.421
Med. " .....	.631	.131	7.59	1.79	.367
High " .....	2.210	.173	9.67	1.71	.501
$r^{**} =$	.887 ††	...	.898 ††	...	...
Low phosphorus ...	2.022	.149	8.62	2.84	.536
Med. " ...	2.021	.137	8.13	2.58	.464
High " ...	2.210	.173	9.67	1.71	.501
$r =$	...	...	...	-.730 ††	...
Low sulphur .....	2.124	.157	8.09	2.67	.488
Med. " .....	1.673	.165	8.99	2.76	.620
High " .....	2.210	.173	9.67	1.71	.501
$r =$	...	...	.685 †	...	...
Low calcium .....	2.421	.177	8.13	2.52	.591
Med. " .....	1.040	.192	7.98	2.32	.504
High " .....	2.210	.173	9.67	1.71	.501
Low potassium ....	1.687	.158	4.17	3.16	1.018
Med. " ....	1.233	.176	6.04	3.05	.648
High " ....	2.210	.173	9.67	1.71	.501
$r =$	...	...	.904 ††	-.801 ††	-.872 ††
Low magnesium ...	1.549	.164	8.12	2.89	.465
Med. " ...	2.131	.180	8.33	2.88	.602
High " ...	2.210	.173	9.67	1.71	.501

\* Each figure is an average of four replications.

\*\*  $r$  indicates significant correlation coefficients between the concentration of elements in the petioles and nutrient treatment.

† Significant to .05.

†† Significant to .01 level.

The final harvest was made 145 days after emergence. From table I it can be seen that height and dry-weight measurements were significantly altered by some treatments. Tissue test results on the 145-day harvest are shown in table V. Again, as in the 90-day harvest, the petiole concentrations of each of the five nutrient elements varied directly with their respective substrate levels. Soluble calcium in the petioles was inversely correlated with substrate magnesium. Petiole magnesium varied directly with

the nitrogen and calcium levels of the substrate. Potassium in the petioles showed negative correlations with substrate sulphur and magnesium. Petiole phosphorus varied inversely with substrate nitrogen and calcium. No interactions were noted for petiole nitrogen.

Yield of seed cotton was reduced by the low and medium nitrogen treatments and the low levels of sulphur and potassium. A significant

TABLE IV  
TISSUE TEST RESULTS ON PETIOLES FROM 90-DAY HARVEST  
(RESULTS BASED ON OVEN-DRY WEIGHT)

Treatment	Percentage in petioles				
	NO <sub>3</sub> N	P	K	Ca	Mg
Low nitrogen .....	.447*	.356	6.10	2.31	.436
Med. " .....	.610	.258	4.94	1.57	.368
High " .....	1.042	.218	5.83	1.47	.454
$r^{**} =$	.918 ††	-.862 ††	...	-.817 ††	...
Low phosphorus ...	1.002	.130	5.42	1.81	.362
Med. " ...	.829	.175	5.41	1.63	.460
High " ...	1.042	.218	5.83	1.47	.454
$r =$	...	.703 ††	...	-.682 ††	...
Low sulphur .....	.873	.249	4.70	2.60	.431
Med. " .....	.704	.217	4.86	1.52	.438
High " .....	1.042	.218	5.83	1.47	.454
$r =$	...	...	...	-.560 †	...
Low calcium .....	1.036	.245	5.55	1.29	.369
Med. " .....	.721	.227	4.92	1.39	.400
High " .....	1.042	.218	5.83	1.47	.454
$r =$	...	...	...	.606 †	...
Low potassium ....	.703	.211	2.29	2.58	.972
Med. " ....	.700	.215	4.13	1.85	.624
High " ....	1.042	.218	5.83	1.17	.454
$r =$	...	...	.954 ††	-.916 ††	-.760 ††
Low magnesium ...	.911	.204	5.70	1.96	.286
Med. " ...	.758	.207	5.08	1.93	.380
High " ...	1.042	.218	5.83	1.47	.454
$r =$	...	...	...	...	.621 †

\* Each figure is an average of five replications.

\*\* $r$  indicates significant correlation coefficients between the concentration of elements in the petioles and nutrient treatment.

† Significant to .05.

†† Significant to .01 level.

correlation was obtained ( $r = .804$ ) when petiole nitrogen of the 90-day harvest from the low, medium, and high nitrogen treatments were compared with the yield of seed cotton for those treatments.

#### FIELD STUDY

After completion of the 1947-48 greenhouse experiment, tissue tests were applied to cotton grown under field conditions. Through the cooperation of



the South Carolina Agriculture Experiment Station tissue tests were run on their field plots at the Pee Dee Experiment Station. These fertilizer plots have been maintained for the past 28 years, and deficiencies are known to exist in nitrogen, phosphorus, and potassium. Petiole harvests were made 90 and 120 days after planting on petioles taken from the third and fourth node from the apex of the main stem.

TABLE V  
TISSUE TEST RESULTS ON PETIOLES FROM 145-DAY HARVEST  
(RESULTS BASED ON OVEN-DRY WEIGHT)

Treatment	Percentage in petioles				
	NO <sub>3</sub> N	P	K	Ca	Mg
Low nitrogen .....	.0048*	.126	4.37	1.07	.291
Med. " .....	.0048	.068	4.86	1.10	.620
High " .....	.0458	.053	4.73	1.03	.794
<i>r</i> ** =	.658 ††	-.777 ††	...	...	.909 ††
Low phosphorus ...	.047	.035	4.58	.97	.743
Med. " ...	.070	.052	4.63	1.01	.821
High " ...	.046	.053	4.73	1.03	.794
<i>r</i> =	...	.826 ††	...	...	...
Low sulphur .....	.102	.059	6.38	1.22	.719
Med. " .....	.026	.057	4.76	.98	.727
High " .....	.046	.053	4.73	1.03	.794
<i>r</i> =	...	...	-.569 †	...	...
Low calcium .....	.056	.075	5.32	.59	.482
Med. " .....	.029	.071	4.90	.73	.783
High " .....	.046	.053	4.73	1.03	.794
<i>r</i> =	...	-.960 ††	...	.990 ††	.624 †
Low potassium ....	.085	.072	.25	1.15	.894
Med. " ....	.023	.051	3.00	1.07	1.082
High " ....	.046	.053	4.73	1.03	.794
<i>r</i> =	...	...	.834 ††	.562 †	...
Low magnesium ...	.031	.045	5.63	1.30	.270
Med. " ...	.039	.055	5.03	1.14	.519
High " ...	.046	.053	4.73	1.03	.794
<i>r</i> =	...	...	-.609 †	-.640 †	.921 ††

\* Each figure is an average of five replications.

\*\**r* indicates significant correlation coefficients between the concentration of elements in the petioles and nutrient treatment.

†Significant to .05.

††Significant to .01 level.

Table VI contains the summarized results of the 90-day harvest. These data confirm to a large extent the results of the previous greenhouse work. The level of soluble NO<sub>3</sub> nitrogen, phosphorus, and potassium within the petioles varied directly with their respective concentrations in the fertilizer; however, interrelations between these elements which influenced petiole concentrations were noted. The negative correlation between substrate nitrogen and petiole phosphorus was apparent. The nitrogen level of the fertilizer

influenced the petiole calcium in an inverse manner; however, the petiole magnesium varied directly with the nitrogen applied. An inverse correlation was shown between phosphorus supply and nitrogen in the petioles, and a direct relation can be seen between fertilizer phosphorus and petiole calcium. This latter relation may be due to the use of superphosphate in the fertilizer. Negative correlations between potassium supplied and the

TABLE VI  
TISSUE TEST RESULTS ON PETIOLES HARVESTED  
FROM 90-DAY OLD FIELD GROWN COTTON  
(RESULTS BASED ON FRESH WEIGHT)

Treatment*			Yield in lbs. seed cotton per acre	Percentage soluble elements in petioles				
N	P	K		NO <sub>3</sub> N	P	K	Ca	Mg
0	8	4	1,266	.0012**	.0933	.760	.359	.0458
2	8	4	1,674	.0025	.0310	.996	.265	.0358
3	8	4	2,046	.0055	.0200	1.021	.254	.0509
4	8	4	2,376	.0336	.0162	.992	.219	.0511
5	8	4	2,427	.0510	.0132	.799	.258	.0660
6	8	4	2,406	.0927	.0160	.789	.213	.0369
$r^{\dagger} =$			...	.620 \$	-.785 \$	...	-.655\$	.523 \$
4	0	4	546	.1495	.0085	.887	.146	.0532
4	2	4	1,215	.0923	.0087	1.011	.188	.0384
4	4	4	1,806	.0978	.0149	1.976	.212	.0415
4	6	4	1,967	.0855	.0090	1.199	.216	.0382
4	8	4	2,376	.0336	.0162	.992	.219	.0511
4	10	4	2,234	.0625	.0213	1.012	.233	.0492
$r =$			...	-.438 ††	.555 \$	...	.716\$	...
4	8	0	665	.1177	.0332	...	.399	.1196
4	8	1	1,412	.0516	.0239	.234	.311	.0859
4	8	2	1,874	.0386	.0249	.599	.314	.0563
4	8	3	2,076	.0388	.0183	.722	.298	.0609
4	8	4	2,376	.0336	.0162	.992	.210	.0511
4	8	5	2,274	.0429	.0180	.828	.239	.0410
$r =$			...	-.522 \$	-.639 \$	.808\$	-.840\$	-.788 \$

\*1,000 lbs. of fertilizer indicated applied per acre.

\*\*Each figure is an average of four replications.

† $r$  indicates significant correlation coefficients between the concentration of elements in the petioles and nutrient treatment.

††Significant to .05.

\$ Significant to .01 level.

petiole calcium and magnesium were obtained. The actual differences in petiole calcium brought about by fertilizer potassium were small, ranging from .399% for the zero potash plot to .239% for the 5% level of potassium. Variations in the petiole magnesium due to fertilizer potassium were of a larger order than those of calcium, as shown in table VI. Inverse relations were obtained between fertilizer potassium and petiole nitrogen and phosphorus. The latter interaction was noted only in the 90-day sampling.

Table VI contains yield data in pounds per acre of seed cotton for each treatment. Too few observations are available to compare yield with petiole nitrogen, phosphorus or potassium. Some generalizations, however, seem obvious. If the levels of  $\text{NO}_3$  nitrogen in the petioles of those plants in the nitrogen series are compared with yield, it is seen that plots having an average  $\text{NO}_3$  nitrogen content lower than .0055% of the fresh petiole weight produced substantially lower yields. Critical levels for these plants can be observed for phosphorus and potassium in their respective series. In the case of phosphorus this level seems to be in the neighborhood of .016%, and for potassium 0.7% of the fresh petiole weight.

### Discussions and conclusions

The petioles of the main-stem leaves most nearly satisfied the requirements of the sampling tissue. The level of soluble nutrients in the petioles reflected the nutrient status of the entire plant. The ease of handling the material, the responses to variations in substrate, and the reflection of interactions between nutrient elements are factors in support of the use of petioles as the experimental tissue. The accumulation of soluble potassium in the petioles is another factor in favor of their selection. It is necessary to standardize as far as possible the collection of petiole material. In these investigations, petioles were collected from the third and fourth nodes from the apex of the main stem.

Highly significant, positive correlations were obtained when the concentrations of soluble nutrient elements in the petioles were compared with their respective levels in the substrate. The age at which deficiencies of nutrients could be detected by tissue tests is difficult to establish. Under favorable growing conditions it may be possible to determine a deficient supply of nitrogen by tissue tests within two or three weeks after emergence. On the other hand, a level of nitrogen which is not sufficient under rapid growing conditions may be ample under conditions not favoring rapid growth. In the 1947-48 greenhouse experiment it is believed plant growth was retarded due to insufficient watering. It is probably for this reason significant correlations between treatment and ion accumulation in the petioles were not obtained until the 90-day harvest. In the 1949-50 greenhouse experiment such correlations were obtained 30 days after emergence when comparing the nitrate nitrogen, phosphorus, and potassium in the tops with their respective substrate levels. The plants of the latter experiment were watered freely and this was reflected in their more rapid growth. Thus the present data indicates that deficiencies of nutrients may be detected by tissue testing methods in advance of the appearance of typical deficiency symptoms. The results of field studies presented here indicated the above conclusion also applies to field-grown cotton as well as greenhouse plants.

The accumulation of nutrient ions in the petioles is complicated by interrelations between these ions. A high concentration of a particular ion in

the petiole does not necessarily indicate an excess of this ion in the substrate but may be associated with a deficiency of some other essential element. A knowledge of these interrelations is of value in the interpretation of results obtained from tissue tests. Petioles of cotton plants grown on low nitrogen substrates were found to contain high amounts of soluble phosphorus. Potassium deficiencies were accompanied by accumulations of calcium and magnesium in the petioles. Substrate calcium and petiole phosphorus seem to be related in an inverse manner as were substrate magnesium and petiole calcium. Other interactions appeared sporadically throughout the course of these investigations but their importance is questionable.

Tissue test may be of considerable value as an early means of predicting yields. In the 1949-50 greenhouse experiment the  $\text{NO}_3$  nitrogen content of the tops 30 days after emergence had a correlation coefficient of  $r = .852$  when compared with the final yield of seed cotton. This value was significant beyond the .01 level. The 30-day sampling indicated that the  $\text{NO}_3$

TABLE VII  
CRITICAL CONCENTRATIONS OF PETIOLE NITROGEN, PHOSPHORUS,  
AND POTASSIUM OBSERVED IN 90-DAY FIELD GROWN  
AND GREENHOUSE COTTON  
(RESULTS BASED ON FRESH WEIGHT)

	Percentage in petioles		
	$\text{NO}_3$ N	P	K
Field grown .....	0.03	.016	0.7
Greenhouse .....	0.10	.019	0.7

nitrogen content of high-yielding plants should presumably have been above 1.2% and potassium above 3.8% of the dry top weight. The  $\text{NO}_3$  nitrogen content in the petioles of these plants remained above 0.7% at the 60- and 90-day harvest, and did not drop below .04% by maturity. The potassium content of dried petioles of high-yielding plants for the 60-, 90-, and 145-day samplings were 6.0, 4.1, and 3.0%, respectively. Direct comparison can be made between the critical concentrations of nitrogen, phosphorus and potassium observed in the 90-day field-grown plants and those noted for the 90-day sampling of the greenhouse experiment by conversion of the latter data to fresh-weight bases. Table VII shows these critical concentrations. A wide difference is noted between the  $\text{NO}_3$  nitrogen levels, but it must be remembered that the field-grown plants had other supplies of nitrogen than the nitrates shown by the tests. The phosphorus level of the greenhouse plants appears high. The reduction in yield due to phosphorus in the greenhouse experiment was not statistically significant, thus the .019% phosphorus may be somewhat higher than the true critical concentration.

### Summary

Colorimetric and turbidimetric methods were used for the determination of soluble  $\text{NO}_3$  nitrogen, phosphorus, potassium, calcium, and magnesium. The procedures were slightly modified to meet the conditions necessary for their application to cotton.

It was found that petioles from the main stem taken from the third and fourth nodes from the apex most nearly satisfied the requirements of sampling.

The level of five nutrients in the petioles reflected the availability of their individual ions in the substrate. Thus, in plants 90 days old positive correlations were obtained when petiole nitrogen, phosphorus, potassium, calcium, or magnesium were compared with their respective concentrations in the substrate. In the 1949-50 greenhouse experiment significant correlations were obtained between tissue concentrations and substrate levels for  $\text{NO}_3$  nitrogen, phosphorus, and potassium 30 days after emergence.

A knowledge of interactions between nutrient elements is essential for proper evaluation of tissue test results. Negative correlations were observed between petiole phosphorus and substrate nitrogen, petiole calcium and substrate potassium, and petiole magnesium and substrate potassium. Other interactions were noted.

From these studies it is evident that tissue tests can play an important part in determining the nutritional status of cotton plants. Indications of deficiencies by tissue test can be obtained in advance of the appearance of typical deficiency symptoms.

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