EFFECTS OF IRON ON CHLOROPHYLLOUS PIGMENTS, ASCORBIC ACID, ACIDITY AND CARBOHYDRATES OF *ANANAS COMOSUS* (L.) MERR., SUPPLIED WITH NITRATE OR AMMONIUM SALTS[1]

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(WITH SIXTEEN FIGURES)

Introduction

In a former paper (**12**) data were presented on the effects of iron on growth, certain physiochemical properties, and ash constituents of *Ananas comosus* (L.) Merr. The present paper is concerned with data from the same plants pertaining to the amounts and distribution of chlorophyll, carotenoid pigments, titrable acidity, ascorbic acid, dextrose, levulose, sucrose, starch, the pentosan and hexosan fractions of hemicellulose, cellulose and lignins in different sections of the leaves, stem, and roots.

Methods

Sixteen plants were grown for 12 months in constantly aerated solution cultures supplied with equal amounts of nitrate- or ammonium-nitrogen and either with or without iron (5 mg. per liter of solution). At the end of this period 8 were harvested, weighed, sectioned, and analyzed following the technique reported in a former paper (**10**). Methods employed for the chemical analyses of the substances mentioned in this paper were as follows.

The chlorophyllous pigments were determined according to SCHERTZ'S methods (**8, 9**). The titrable acidity of the tissues, expressed as percentage of citric acid, was determined with a 0.1 N solution of sodium hydroxide on a 10-ml. aliquot representing 2 grams of fresh tissue. Ascorbic acid was determined with 2, 6-dichlorophenolindophenol in acetic acid according to the method of BESSEY and KING (**3**). Sugars in the aqueous extracts of fresh tissues were determined by the method of QUISUMBING and THOMAS (**7**) after treatment with lead acetate and deleading with disodium phosphate. Dextrose was determined by the hypoiodite method of WIDDOWSON (**14**), and levulose was calculated from the difference between the values of reducing sugars and dextrose. The colorimetric method of PUCHER and VICKERY (**6**) was employed for the determination of starch. Hemicelluloses were determined after hydrolysis with 0.75 N hydrochloric acid for 2.5 hours on the residue of the sample employed for the analysis of starch following the removal of all dissolved substances. The sugars in the above hydrolysate, reported as total hemicelluloses, contained mixtures of pentose and hexose sugars. The furfural content of the total hemicellulose sugars was made the basis for the separation of the pentose from the hexose sugars. The residue

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of the sample of tissues from the hemicellulose determination was employed for determining lignin and cellulose. Lignin was determined by the method of PETERSON, WALDE and HIXON (5). Cellulose was calculated as the difference between the weights of the dried residue (after the extraction of hemicelluloses) and lignin.

The significance of the differences between the amounts of the various items of the different cultures was calculated, as reported in a former publication (12), from CONRAD'S modification of BESSEL'S formula, and the values of z were obtained from Student's Table as modified by LOVE. Table VII reports the statistical significance of the difference of the amounts of the various items mentioned.

Results

Synoptic expressions have been introduced in the presentation of the results and in the discussion to replace the longer ones for the appellation of

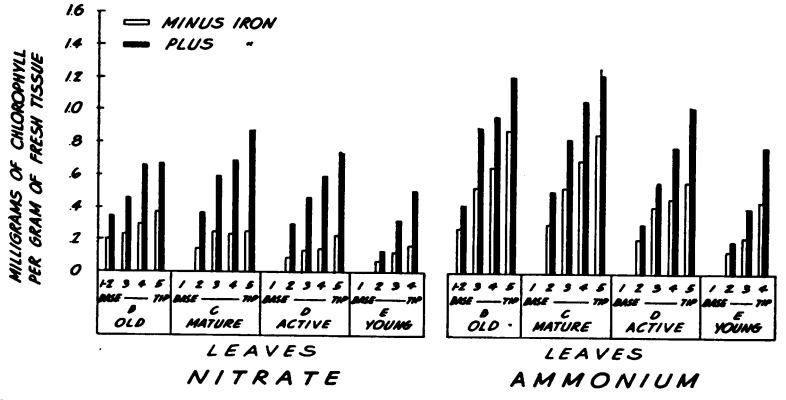

FIG. 1. Chlorophyll in pineapple tissues of plants grown with and without iron, and with nitrate or ammonium as source of nitrogen.

the various treatments. The series Ammonium-nitrogen will be designated by A-n and Nitrate-nitrogen by N-n. Also, the cultures "Plus-iron" by F and Minus-iron" by 0.

CHLOROPHYLLOUS PIGMENTS

Chlorophyll was separated from the carotenoid pigments but no attempt was made to separate either chlorophyll alpha from chlorophyll beta or carotene from xanthophyll. The data in table I and in figure 1, show that the plants of the F cultures contained amounts of chlorophyll considerably in excess of those of the 0 cultures regardless of the source of nutrient nitrogen. They show, also, that the plants of the A-n series contained greater quantities of chlorophyll than those of the N-n series. This relationship has already been discussed (10).

Carotenoids were higher in the plants of the F than 0 cultures of both series. They ranged from 10 to 30 per cent. in the A-n and from 70 to 180 per cent. in the N-n series. Such comparisons show that the distribution of carotenoids paralleled that of chlorophylls. Since the amounts of both types

TABLE I

MILLIGRAMS OF CHLOROPHYLL, CAROTENOIDS, CITRIC ACID (TITRABLE ACIDITY), AND ASCORBIC ACID PER GRAM OF FRESH TISSUE AND pH VALUES OF THE EXTRACTED SAP IN DIFFERENT SECTIONS OF THE LEAVES, STEM, AND ROOTS OF ONE-YEAR-OLD PLANTS OF *A. comosus* GROWN IN PLUS- OR MINUS-IRON SOLUTION CULTURES AND SUPPLIED WITH NITRATE SALTS AS SOURCES OF NITROGEN

PLANT SECTIONS	MINUS IRON					PLUS IRON				
	CHLOROPHYLL	CAROTENOIDS	CITRIC ACID	ASCORBIC ACID	pH	CHLOROPHYLL	CAROTENOIDS	CITRIC ACID	ASCORBIC ACID	pH
	mg.	*mg.*	*mg.*	*mg.*		*mg.*	*mg.*	*mg.*	*mg.*	
Leaves										
Old (B) 1 + 2 (base)	0.20	0.020	2.16	0.29	5.2	0.33	0.033	1.08	0.02	5.3
3	0.23	0.034	7.54	3.03	4.1	0.46	0.054	9.34	0.91	3.9
4	0.30	0.042	10.43	3.90	3.9	0.66	0.079	12.92	1.49	3.7
5 (tip)	0.37	0.051	11.12	4.73	3.9	0.67	0.079	14.00	1.49	3.6
Mature (C) 1 (base)	.....	.....	0.72	0.01	5.5	.....	.....	0.38	0.01	5.6
2	0.12	0.012	3.60	0.53	4.4	0.35	0.031	2.88	0.05	4.3
3	0.25	0.031	9.00	2.31	4.0	0.59	0.067	8.58	0.57	3.9
4	0.23	0.036	11.50	3.48	3.8	0.69	0.083	10.80	1.85	3.7
5 (tip)	0.25	0.038	11.50	5.28	3.9	0.86	0.106	13.64	2.09	3.5
Active (D) 1 (base)	.....	.....	0.71	0.01	5.5	.....	.....	0.38	0.01	5.6
2	0.08	0.006	2.88	0.14	4.4	0.28	0.022	1.79	0.01	4.5
3	0.13	0.011	10.42	0.88	3.8	0.46	0.047	11.15	0.10	3.6
4	0.18	0.025	14.50	3.35	3.6	0.60	0.073	16.50	2.12	3.4
5 (tip)	0.23	0.037	16.20	5.61	3.4	0.74	0.104	20.15	2.87	3.4
Young (E) 1 (base)	.....	.....	1.09	0.01	5.5	.....	.....	0.70	0.01	5.7
2	0.07	0.004	2.52	0.04	4.6	0.12	0.009	2.52	0.01	4.4
3	0.13	0.013	11.90	0.77	3.4	0.33	0.032	13.70	0.16	3.4
4 + 5 (tip)	0.17	0.026	16.56	2.97	3.3	0.50	0.067	22.65	1.10	3.3
Stem										
Base	0.0	0.0	1.44	0.0	5.3	0.0	0.0	1.44	0.0	5.4
Middle	0.0	0.0	2.16	0.0	5.4	0.0	0.0	2.52	0.0	5.3
Apex	0.0	0.0	2.52	0.0	5.3	0.0	0.0	2.16	0.0	5.5
Roots	0.0	0.0	0.36	0.0	5.4	0.0	0.0	0.32	0.0	5.6

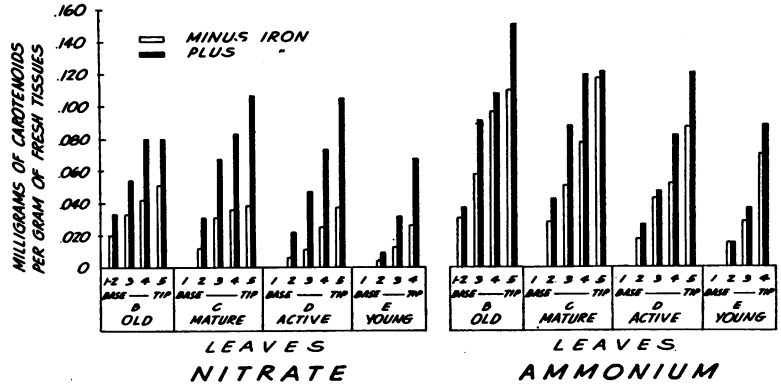

FIG. 2. Carotenoid content of pineapple tissues of plants grown with and without iron, and with nitrate or ammonium nitrogen.

of pigments present in a given section were not proportional to the amounts of iron supplied in the nutrient solution, it is probable that the degree of availability rather than the total amounts of iron within the plants were the determinant factors for the concentrations of chlorophyll and carotenoids. Iron availability was greater in the A-n than N-n series because of more favorable H-ion concentrations in the solution cultures of the former series as shown in an earlier publication (**12**).

ACIDITY

Titrable acidity is reported as milligrams of citric acid per gram of fresh tissue in tables I and II, and in figure 3. The values differed in different sections and also varied to some extent in comparable sections of the plants of the different cultures. They were generally greater in the chlorophyllous

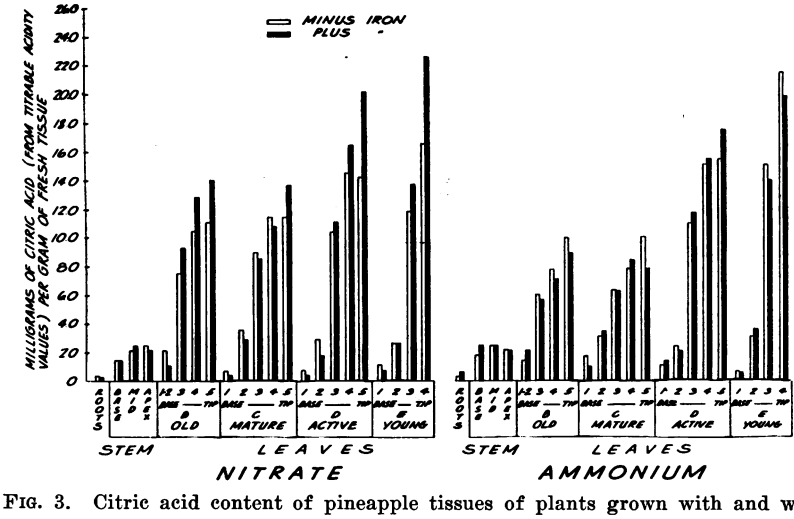

FIG. 3. Citric acid content of pineapple tissues of plants grown with and without iron, and with nitrate or ammonium nitrogen.

TABLE II

MILLIGRAMS OF CHLOROPHYLL, CAROTENOIDS, CITRIC ACID (TITRABLE ACIDITY), AND ASCORBIC ACID PER GRAM OF FRESH TISSUE AND pH VALUES OF THE EXTRACTED SAP IN DIFFERENT SECTIONS OF THE LEAVES, STEM, AND ROOTS OF ONE-YEAR-OLD PLANTS OF *A. comosus* GROWN IN PLUS- OR MINUS-IRON SOLUTION CULTURES AND SUPPLIEDWITH AMMONIUM SALTS AS SOURCES OF NITROGEN

PLANT SECTIONS	MINUS IRON					PLUS IRON				
	CHLOROPHYLL	CAROTENOIDS	CITRIC ACID	ASCORBIC ACID	pH	CHLOROPHYLL	CAROTENOIDS	CITRIC ACID	ASCORBIC ACID	pH
	mg.	*mg.*	*mg.*	*mg.*		*mg.*	*mg.*	*mg.*	*mg.*	
Leaves										
Old (B) 1+2 (base)	0.27	0.031	1.44	0.01	5.3	0.42	0.037	2.16	0.02	5.0
3	0.53	0.057	6.12	1.77	4.1	0.90	0.091	5.77	1.23	4.2
4	0.66	0.096	7.90	2.75	3.9	0.96	0.107	7.17	1.84	3.9
5 (tip)	0.89	0.109	10.05	3.36	3.7	1.21	0.150	8.96	2.27	3.8
Mature (C) 1 (base)	.....	.....	1.79	0.01	5.4	.....	.....	1.02	0.01	5.2
2	0.30	0.028	3.23	0.28	4.6	0.50	0.042	3.59	0.04	4.6
3	0.53	0.050	6.46	1.22	4.0	0.83	0.087	6.40	1.35	4.2
4	0.70	0.077	7.92	2.93	3.8	1.05	0.118	8.57	2.65	3.9
5 (tip)	0.86	0.116	10.10	4.34	3.6	1.22	0.120	7.90	2.88	3.8
Active (D) 1 (base)	.....	.....	1.09	0.01	5.4	.....	.....	1.44	0.01	5.4
2	0.21	0.017	2.51	0.01	4.3	0.30	0.026	2.16	0.01	4.6
3	0.41	0.042	11.13	0.51	3.6	0.56	0.046	11.85	0.86	3.8
4	0.47	0.051	15.10	2.93	3.5	0.78	0.081	15.50	3.39	3.7
5 (tip)	0.56	0.086	15.50	5.04	3.4	1.02	0.119	17.60	6.23	3.7
Young (E) 1 (base)	.....	.....	0.72	0.01	5.6	.....	.....	0.64	0.01	5.6
2	0.13	0.015	3.23	0.01	4.3	0.20	0.014	3.74	0.01	4.7
3	0.22	0.028	15.10	0.25	3.4	0.40	0.036	14.00	0.30	3.4
4+5 (tip)	0.44	0.069	21.60	2.75	3.3	0.78	0.087	19.90	2.47	3.4
Stem										
Base	0.0	0.0	1.79	0.0	5.2	0.0	0.0	2.52	0.0	5.4
Middle	0.0	0.0	2.52	0.0	5.2	0.0	0.0	2.52	0.0	5.2
Apex	0.0	0.0	2.22	0.0	5.3	0.0	0.0	2.18	0.0	5.2
Roots	0.0	0.0	0.32	0.0	6.0	0.0	0.0	0.64	0.0	5.7

TABLE III

MILLIGRAMS OF TOTAL AND REDUCING SUGARS AND THEIR FRACTIONS OF DEXTROSE, LEVULOSE, SUCROSE, AND STARCH PER GRAM OF FRESH TISSUE IN DIFFERENT SECTIONS OF THE LEAVES, STEM, AND ROOTS OF ONE-YEAR-OLD PLANTS OF *A. comosus* GROWN IN PLUS- AND MINUS-IRON SOLUTION CULTURES AND SUPPLIED WITH NITRATE SALTS AS SOURCES OF NTROGEN

PLANT SECTIONS	MINUS IRON						PLUS IRON					
	TOTAL SUGARS	REDUCING SUGARS	DEXTROSE	LEVULOSE	SUCROSE	STARCH	TOTAL SUGARS	REDUCING SUGARS	DEXTROSE	LEVULOSE	SUCROSE	STARCH
	mg.	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*
Leaves												
Old (B) 1 + 2 (base)	20.7	18.8	12.6	6.2	1.9	1.3	19.6	15.7	11.0	4.7	3.9	1.9
3	19.6	18.5	9.9	8.6	1.1	1.2	18.1	14.3	9.0	5.3	3.8	2.1
4	19.3	18.2	10.8	7.4	1.1	1.1	19.3	15.6	10.4	5.2	3.7	1.4
5 (tip)	19.3	18.0	11.7	6.3	1.3	1.2	21.6	17.6	12.8	4.8	4.0	1.7
Mature (C) 1 (base)	24.0	22.5	15.8	6.7	1.5	1.5	25.9	21.4	13.3	8.1	4.5	2.2
2	21.0	19.8	12.2	7.6	1.2	3.5	22.1	17.3	12.4	4.9	4.8	6.3
3	12.5	9.6	6.1	3.5	2.9	2.5	16.2	11.3	7.0	4.3	4.9	2.9
4	14.1	10.6	7.5	3.1	3.5	2.3	16.8	11.7	8.4	3.3	5.1	1.7
5 (tip)	18.7	14.7	9.9	4.8	4.0	2.9	21.5	13.7	10.8	2.9	7.8	2.9
Active (D) 1 (base)	24.7	22.7	15.8	6.9	2.0	3.4	25.5	22.4	16.2	6.2	3.1	3.5
2	21.3	19.5	11.9	7.6	1.8	4.2	22.9	18.1	12.6	5.5	4.8	4.7
3	17.4	14.0	7.2	6.8	3.4	3.0	21.9	15.2	8.1	7.1	6.7	3.5
4	17.0	12.5	6.8	5.7	4.5	1.8	22.1	15.7	10.4	5.3	6.4	1.5
5 (tip)	17.7	12.2	8.1	4.1	5.5	1.8	24.6	16.5	11.7	4.8	8.1	1.8
Young (E) 1 (base)	14.1	13.1	8.1	5.0	1.0	0.7	15.5	12.5	9.5	3.0	3.0	0.8
2	14.3	14.3	8.3	6.0	0.0	0.2	15.3	14.1	9.0	5.1	1.2	0.2
3	19.8	18.6	8.1	10.5	1.2	1.5	21.4	17.5	8.1	9.4	3.9	1.4
4 + 5 (tip)	23.3	19.4	9.0	10.4	1.9	1.1	26.9	18.6	9.2	9.4	8.3	1.4
Stem												
Base	16.6	12.6	14.2	0.0	4.0	17.7	17.0	13.6	14.4	0.0	3.4	19.9
Middle	12.2	7.6	8.4	0.0	4.6	38.6	13.1	8.3	8.7	0.0	4.8	43.7
Apex	17.7	10.4	9.6	0.8	7.3	25.8	19.6	12.1	11.0	1.1	7.5	45.9
Roots	4.4	1.2	1.8	0.0	3.2	0.0	4.3	1.8	1.6	0.2	2.5	0.0

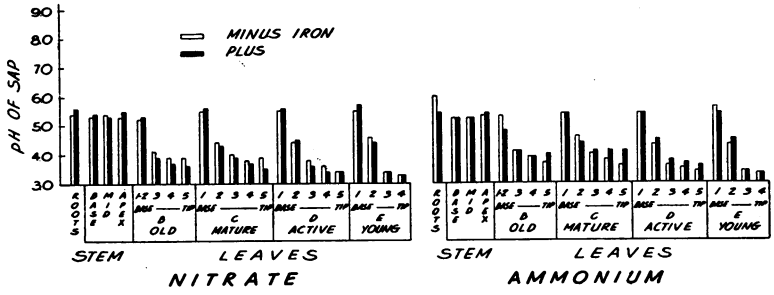

FIG. 4. Active acidity (pH) of sap of pineapple tissues of plants grown with and without iron, and with nitrate or ammonium nitrogen.

than non-chlorophyllous sections of the leaves, a fact also reported in a previous publication (**10**). The values of titrable acidity fluctuated considerably between comparable sections in the plants of the different cultures. They were not consistent with respect to either the amounts of iron or the source of nitrogen. In the N-n series, however, the cholorphyllous sections of the leaves of the F cultures contained significantly greater amounts of titrable acidity than those of the 0 cultures, yet in the A-n the differences in acidity between the F and 0 plants were not statistically significant.

The pH values of the sap, reported in tables I and II and figure 4, were in relative agreement with those of the acidity for corresponding sections although the great amounts of buffer substances in certain sections made perfect agreement between titrable acidity and pH values impossible.

ASCORBIC ACID

The ascorbic acid content of the sap, reported in tables I and II, and in figure 5, was higher in the chlorophyllous than in the non-chlorophyllous sections, thus approximately paralleling the distribution of citric acid. The stem and roots either completely lacked ascorbic acid or the amounts were very small and insignificant.

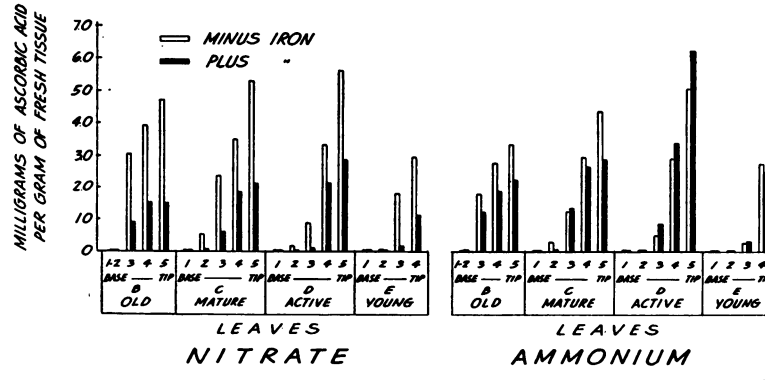

FIG. 5. Ascorbic acid content of pineapple tissues from plants grown with and without iron, with nitrate or ammonium nitrogen.

Ascorbic acid values were considerably greater in the leaves of the 0 than F cultures of the N-n series, the differences being highly significant, as reported in table VII. In the plants of the A-n series ascorbic acid was apparently higher in the "old" (B) and "mature" (C) groups of leaves of the 0 than F cultures, but not in the "active" (D) and "young" (E) groups. Ascorbic acid showed in many sections of the N-n series an inverse relationship to citric acid whereas in the A-n series the relationship was direct.

TOTAL SUGARS

The amounts of total sugars, reported in tables III and IV, and in figure 6, were, with few exceptions, higher for the plants of the F than 0 cultures

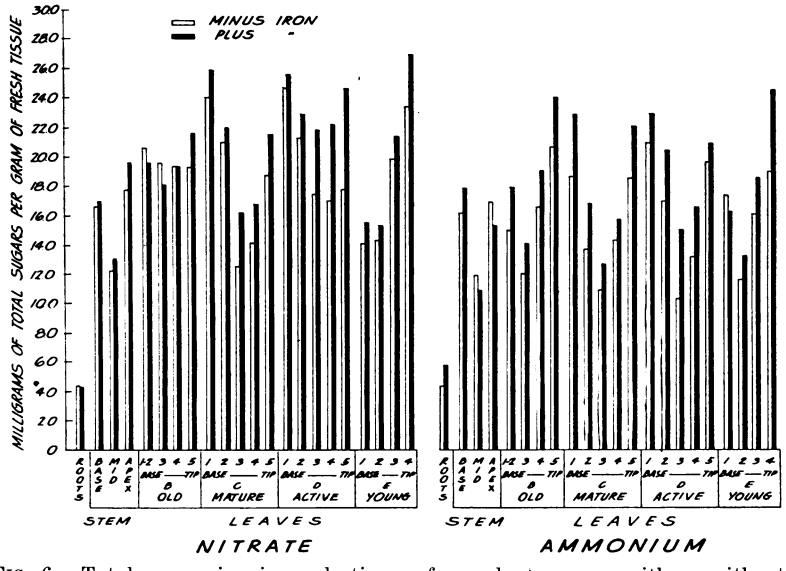

FIG. 6. Total sugars in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

of both series. They were higher in the basal and apical sections of the stem than in the medial sections. The amounts were greater in the N-n than A-n series. Contrasts between the amounts of sugars and chlorophyll in the "mature" (C) and "active" (D) leaves of the F and 0 cultures of the N-n series show that mean chlorophyll values were 212 per cent. (F = 0.572 and 0 = 0.183) and sugars 16.6 per cent. (F = 27.5 and 0 = 23.5) higher in the F than 0 cultures. Variability in the chlorophyll values was 30.8 per cent. $\left(\text{CV} = \frac{0.12 \times 100}{0.39}\right)$, whereas in the sugars it was 57.4 $\left(\text{CV} = \frac{2.236 \times 100}{3.9}\right)$. Accordingly, sugar values were 1.86 times ($57.4 \div 30.8 = 1.86$) more variable, on the basis of these calculations, than chlorophyll values. The results suggest, therefore, that sugar accumulations in the leaves had been influenced by other factors intimately associated with the metabolic and photosynthetic activities of the plant besides chlorophyll concentrations.

TABLE IV

MILLIGRAMS OF TOTAL AND REDUCING SUGARS AND THEIR FRACTIONS OF DEXTROSE, LEVULOSE, SUCROSE, AND STARCH PER GRAM OF FRESH TISSUE IN DIFFERENT SECTIONS OF THE LEAVES, STEM, AND ROOTS OF ONE-YEAR-OLD PLANTS OF *A. comosus* GROWN IN PLUS- AND MINUS-IRON SOLUTION CULTURES AND SUPPLIED WITH AMMONIUM SALTS AS SOURCES OF NITROGEN

Plant sections	Minus iron						Plus iron					
	Total sugars	Reducing sugars	Dextrose	Levulose	Sucrose	Starch	Total sugars	Reducing sugars	Dextrose	Levulose	Sucrose	Starch
	mg.	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*
Leaves												
Old (B) 1 + 2 (base)	15.0	12.0	9.6	2.4	3.0	0.1	17.8	16.5	10.1	6.4	1.3	0.4
3	12.0	8.3	6.6	1.7	3.7	0.3	14.1	11.8	7.8	4.0	2.3	0.6
4	16.6	11.7	10.0	1.7	4.9	0.3	19.1	14.3	10.4	3.9	4.8	0.8
5 (tip)	20.7	14.6	12.4	2.2	6.1	0.3	24.0	18.2	13.3	4.9	5.8	1.0
Mature (C) 1 (base)	18.6	16.6	12.2	4.4	2.0	0.1	22.9	21.7	13.7	8.0	1.2	0.3
2	13.7	11.1	8.2	2.9	2.6	0.3	16.8	15.0	9.5	5.5	1.8	1.0
3	10.9	7.6	7.4	0.2	3.3	0.6	12.7	9.1	6.1	3.0	3.6	1.3
4	14.3	9.9	8.7	1.2	4.4	1.0	15.7	11.0	7.9	3.1	4.7	1.9
5 (tip)	18.5	12.6	11.3	1.3	5.9	1.5	22.1	15.1	11.7	3.4	7.0	3.0
Active (D) 1 (base)	20.9	18.9	12.6	6.3	2.0	0.2	22.9	22.3	13.3	9.0	0.6	0.7
2	16.9	15.3	9.7	5.6	1.6	0.2	20.4	18.9	10.6	8.3	1.5	1.1
3	10.3	6.9	4.6	2.3	3.4	0.5	15.0	11.0	5.5	5.5	4.0	1.2
4	13.2	7.0	5.1	0.9	6.2	0.8	16.8	10.2	7.8	2.4	6.6	1.1
5 (tip)	19.6	11.1	9.6	1.5	8.5	1.4	20.9	12.1	10.2	1.9	8.8	1.9
Young (E) 1 (base)	17.3	16.1	10.4	5.7	1.2	0.2	16.3	14.5	9.0	5.5	1.8	0.3
2	11.6	10.7	5.8	4.9	0.9	0.1	13.2	12.7	6.9	5.8	0.5	0.0
3	16.1	11.9	5.7	6.2	4.2	0.3	18.5	15.3	7.0	8.5	3.2	0.3
4 + 5 (tip)	18.9	11.1	6.6	4.5	7.8	0.7	24.5	17.1	9.7	7.4	7.4	0.5
Stem												
Base	16.2	13.6	13.5	0.1	2.6	2.5	17.8	14.4	13.7	0.7	3.4	5.9
Middle	11.9	7.8	7.7	0.1	4.1	7.8	10.8	7.5	6.9	0.6	3.3	17.7
Apex	17.0	10.8	8.7	2.1	6.2	10.5	15.3	9.8	8.5	1.3	5.5	18.3
Roots	4.3	2.5	2.5	0.0	2.0	0.0	5.7	3.3	2.5	0.8	2.4	0.0

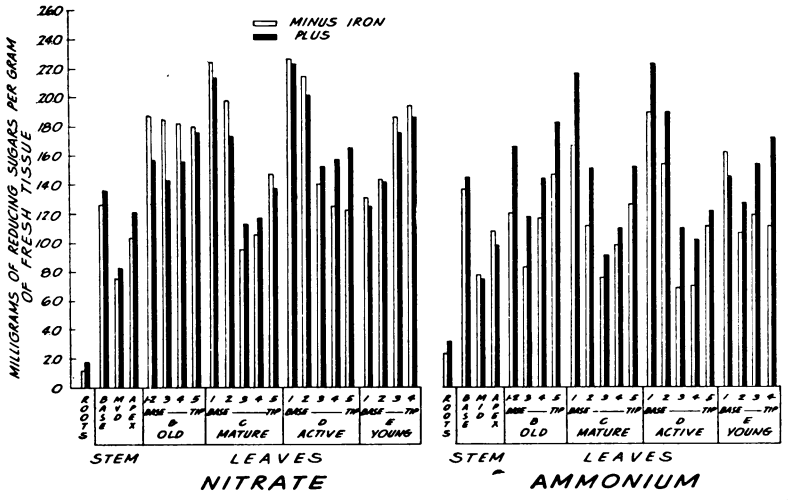

FIG. 7. Reducing sugars in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

REDUCING SUGARS

Data for reducing sugars are presented in figure 7 and for dextrose and levulose in figures 8 and 9, and in tables III and IV. Dextrose occurred in much greater amounts in practically all the sections of the leaves, stem, and roots than did levulose. In the stem, levulose was either absent or present in very small amounts while dextrose made up the bulk of the amounts of

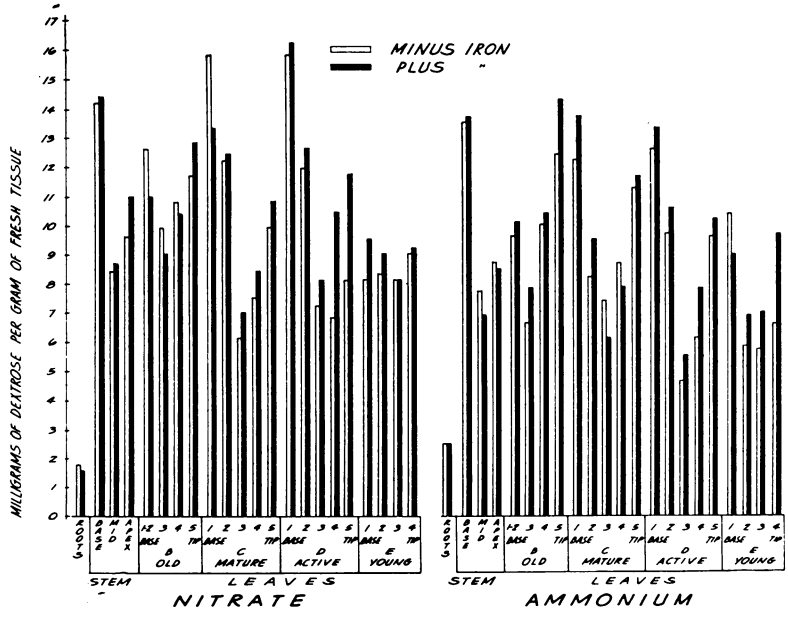

FIG. 8. Dextrose in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

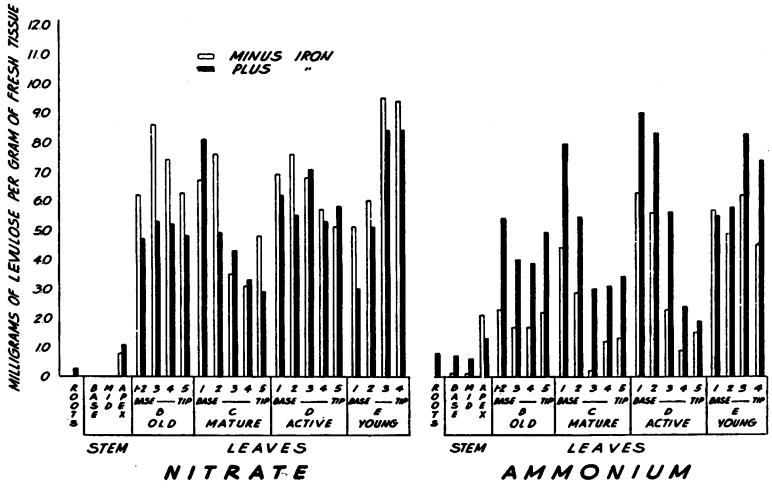

FIG. 9. Levulose in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

reducing sugars. Dextrose accumulated mostly in the basal and terminal sections of the leaves of the "old" (B), "mature" (C), and "active" (D) groups, whereas levulose accumulations were highest in the basal sections. In the leaves of the "young" (E) group the differences between the amounts of either dextrose or levulose of the basal and terminal sections were not as great as in the other groups of leaves. Dextrose occurred in greater amounts in the chlorophyllous (2, 3, 4, 5), while levulose was more abundant in the basal (1), non-chlorophyllous sections of the leaves. The low amounts of levulose in the chlorophyllous and the high in the non-chlorophyllous sections might indicate that either the rate of levulose synthesis was consider-

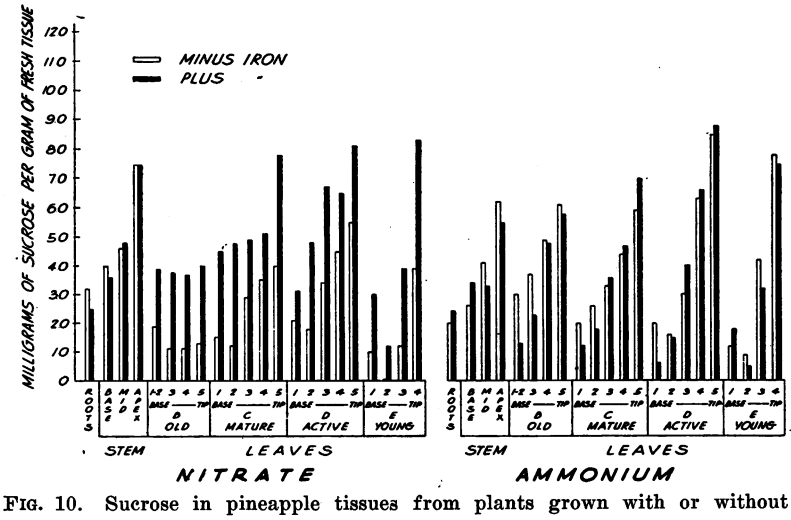

FIG. 10. Sucrose in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

ably lower than that of dextrose or that it was utilized more in the syntheses of various types of substances.

Sucrose

The data in tables III and IV, and in figure 10 show that sucrose was more abundant in the plants of the F than 0 cultures of the N-n series. In the A-n series sucrose did not accumulate in the F cultures and in many sections the 0 cultures had slightly higher sucrose values. There can be no doubt, however, that the greater amounts of sucrose in the plants of the F than 0 cultures of the N-n series resulted from different rates of carbohydrate synthesis due to appreciable differences in chlorophyll content. The lack of

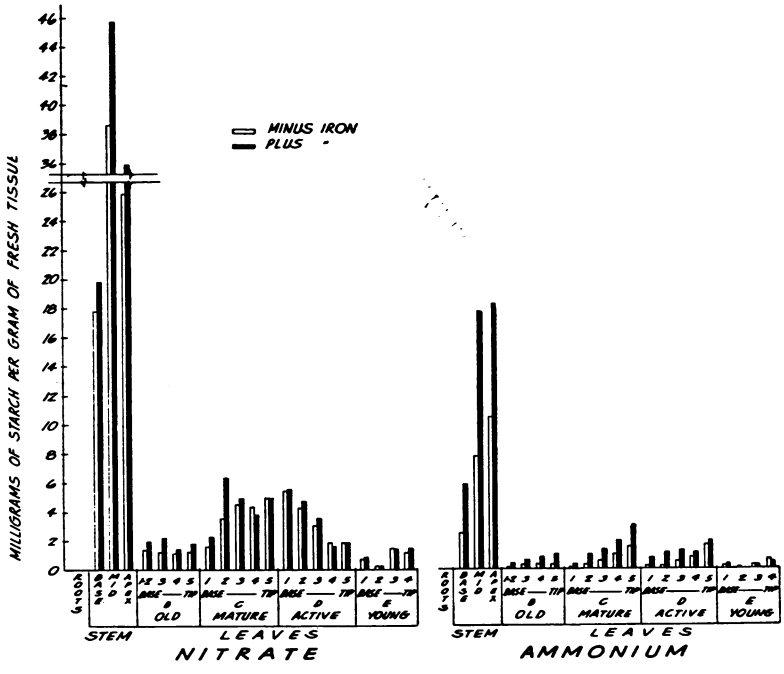

FIG. 11. Starch in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

appreciable difference in the sucrose content of the F and 0 cultures in the A-n series may be attributed to the small differences in the chlorophyll content and possibly to different rates of utilization of sucrose.

Starch

Iron availability and the source of nitrogen had affected the amounts of starch more than any other carbohydrates, as reported in tables III and IV, and in figure 11. The plants of the F cultures of both series contained, with few minor exceptions, more starch than those of the 0 cultures. Also, starch accumulations were enhanced more by nitrate- than ammonium-nitrogen under the same concentrations of iron.

Starch accumulations were restricted mostly to the tissues of the stem and were appreciably greater in the N-n than in the A-n series, whereas reducing sugars and sucrose accumulated mostly in the leaves although some stem sections contained as high amounts of sugars as did leaf sections. The "mature" (C) and "active" (D) leaves contained greater amounts of starch than did those of the "old" (B) and "young" (E) groups. The small amounts of starch in the leaves of the "young" (E) group can be explained on the basis of a relatively rapid utilization of carbohydrates for the synthesis of new tissues, while those in the leaves of the "old" (B) group may be explained by a decreased rate of photosynthetic activity or by a possible hydrolysis of starch resulting from tissue senility.

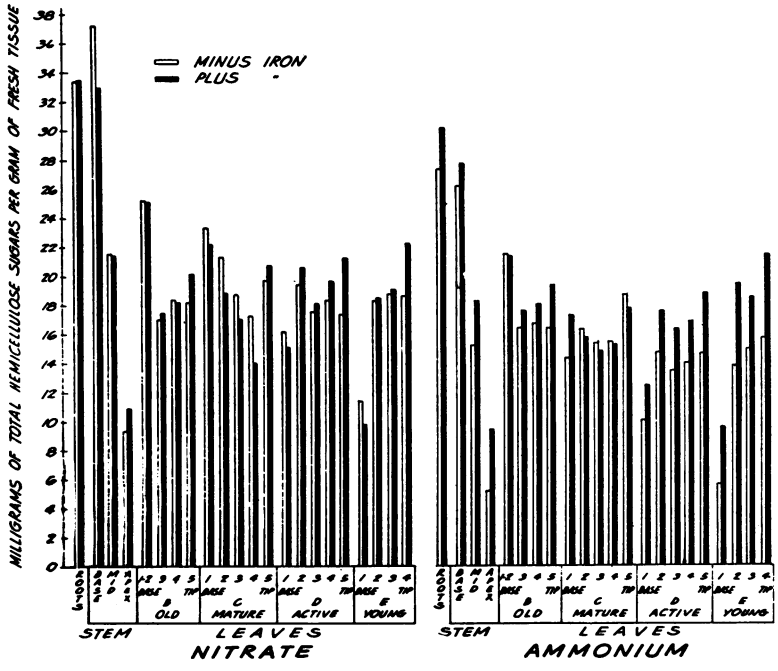

FIG. 12. Total hemicellulose sugars in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

A synoptic analysis of the effects of iron and nitrogen sources on starch shows that nitrate-nitrogen versus ammonium-nitrogen increased the average starch values in the stem 161.6 per cent. for the plus-iron and 295.0 per cent. for the minus-iron cultures. The gains of starch in the stem of the plus-iron versus minus-iron cultures were 33.2 per cent. for the nitrate series and 101.3 per cent. for the ammonium series.

HEMICELLULOSES

The amounts of total hemicellulose sugars, reported in tables V and VI, and in figures 12, 13 and 14, were slightly greater in the F than 0 cultures.

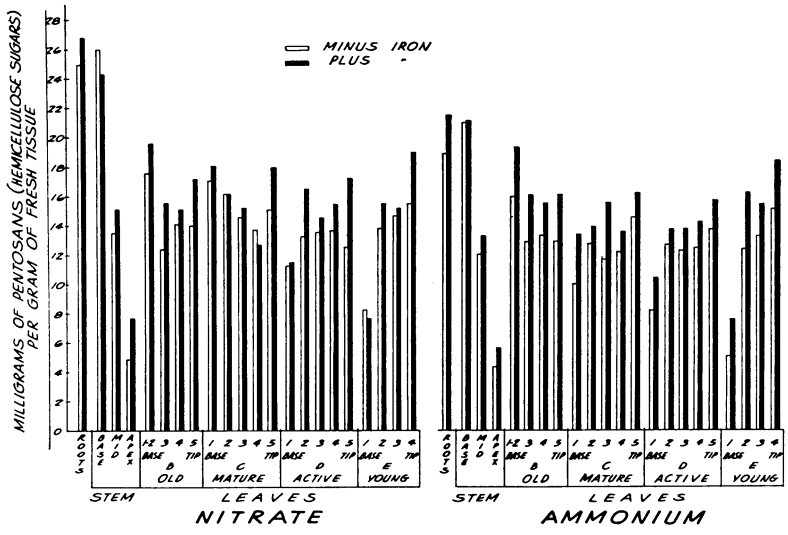

FIG. 13. Pentosans in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

Pentose sugars, constituting more than three-fourths the amounts of total hemicellulose sugars, were generally higher in most leaf sections of the F than 0 cultures of both series. The amounts of hexose sugars showed decided deviations from pentose sugars, being generally higher in the leaves of the 0 than F cultures of the N-n series. In the A-n series the amounts of hexose sugars were higher in the "old" (B) and "mature" (C) and lower in the "active" (D) and "young" (E) groups of leaves of the 0 than in the F cultures. The amounts of hexose sugars in the stem were higher in the 0 than in the F cultures of the N-n series, while in the A-n series the values

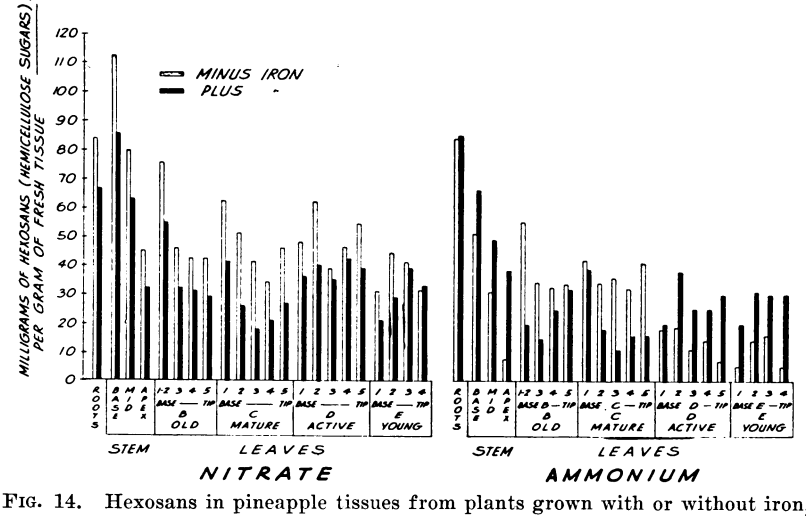

FIG. 14. Hexosans in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

TABLE V

MILLIGRAMS OF TOTAL HEMICELLULOSES AND THEIR FRACTIONS OF PENTOSANS AND HEXOSANS, CELLULOSES, AND LIGNIN PER GRAM OF FRESH TISSUE IN DIFFERENT SECTIONS OF THE LEAVES, STEM, AND ROOTS OF ONE-YEAR-OLD PLANTS OF *A. comosus* GROWN IN PLUS- AND MINUS-IRON SOLUTION CULTURES AND SUPPLIED WITH NITRATE SALTS AS SOURCES OF NITROGEN

Plant sections	Minus iron					Plus iron				
	Hemicelluloses			Celluloses	Lignin	Hemicelluloses			Celluloses	Lignin
	Total	Pentosans	Hexosans			Total	Pentosans	Hexosans		
	mg.	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*
Leaves										
Old (B) 1+2 (base)	25.2	17.6	7.6	39.1	8.9	25.1	19.6	5.5	39.6	9.6
3	17.0	12.4	4.6	27.8	7.7	17.4	15.6	1.8	28.8	8.4
4	18.3	14.1	4.2	28.2	9.1	18.2	15.1	3.1	27.4	8.0
5 (tip)	18.2	14.0	4.2	29.0	10.3	20.1	17.2	2.9	30.8	9.9
Mature (C) 1 (base)	23.3	17.1	6.2	34.8	5.5	22.2	18.1	4.1	33.2	6.0
2	21.3	16.2	5.1	37.0	6.9	18.8	16.2	2.6	34.3	6.9
3	18.7	14.6	4.1	30.0	7.5	17.0	15.2	1.8	29.8	7.6
4	17.2	13.8	3.4	28.5	7.7	14.8	12.7	2.1	25.6	7.5
5 (tip)	19.7	15.1	4.6	30.9	8.9	20.7	18.0	2.7	33.8	10.6
Active (D) 1 (base)	16.1	11.3	4.8	30.2	4.6	15.1	11.5	3.6	25.4	4.9
2	19.4	13.3	6.2	37.1	6.5	20.6	16.6	4.0	39.7	8.2
3	17.5	13.6	3.9	31.3	7.5	18.1	14.6	3.5	34.2	8.4
4	16.3	13.7	4.6	28.9	9.6	19.7	15.5	4.2	34.2	9.9
5 (tip)	17.4	12.6	5.4	31.5	9.8	21.2	17.3	3.9	34.2	11.1
Young (E) 1 (base)	11.4	8.3	3.1	19.8	2.8	9.8	7.7	2.1	16.5	2.3
2	18.3	13.9	4.4	32.0	7.0	18.5	15.6	2.9	30.1	6.9
3	18.8	14.7	4.1	29.8	8.5	19.1	15.2	3.9	33.4	8.8
4+5 (tip)	18.7	15.6	3.1	31.8	10.0	22.3	19.1	3.2	37.1	10.7
Stem										
Base	37.2	26.0	11.2	37.2	18.0	32.9	24.3	8.6	34.4	13.1
Middle	21.5	13.5	8.0	21.0	5.0	21.4	15.1	6.3	21.4	4.9
Apex	9.3	4.8	4.5	11.4	1.0	10.9	7.7	3.2	13.5	1.3
Roots	33.4	25.0	8.4	43.5	28.8	33.5	26.8	6.7	50.1	30.9

TABLE VI

MILLIGRAMS OF TOTAL HEMICELLULOSES AND THEIR FRACTIONS OF PENTOSANS AND HEXOSANS, CELLULOSES, AND LIGNIN PER GRAM OF FRESH TISSUE IN DIFFERENT SECTIONS OF THE LEAVES, STEM, AND ROOTS OF ONE-YEAR-OLD PLANTS OF *A. comosus* GROWN IN PLUS- AND MINUS-IRON SOLUTION CULTURES AND SUPPLIED WITH AMMONIUM SALTS AS SOURCES OF NITROGEN

PLANT SECTIONS	MINUS IRON					PLUS IRON				
	HEMICELLULOSES			CELLULOSES	LIGNIN	HEMICELLULOSES			CELLULOSES	LIGNIN
	TOTAL	PENTOSANS	HEXOSANS			TOTAL	PENTOSANS	HEXOSANS		
	mg.	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*	*mg.*
Leaves										
Old (B) 1+2 (base)	21.6	16.1	5.5	29.6	14.6	21.5	19.5	2.0	33.6	12.1
3	16.4	13.0	3.4	31.6	9.2	17.7	16.2	1.5	30.2	9.4
4	16.7	13.4	3.3	28.8	10.1	18.1	15.6	2.5	28.7	11.3
5 (tip)	16.4	13.0	3.4	30.4	11.3	19.4	16.2	3.2	31.2	10.9
Mature (C) 1 (base)	14.3	10.1	4.2	25.8	4.5	17.4	13.5	3.9	27.2	5.0
2	16.3	12.9	3.4	30.2	7.1	15.8	14.0	1.8	35.4	7.3
3	15.4	11.8	3.6	27.6	8.3	14.8	13.7	1.1	28.2	8.3
4	15.5	12.3	3.2	28.4	8.8	15.3	13.7	1.6	28.4	7.9
5 (tip)	18.8	14.7	4.1	32.0	10.1	17.9	16.3	1.6	31.0	9.8
Active (D) 1 (base)	10.1	8.3	1.8	23.6	4.2	12.5	10.5	2.0	24.8	3.4
2	14.7	12.8	1.9	33.7	7.0	17.7	13.9	3.8	33.2	6.8
3	13.5	12.4	1.1	30.4	8.4	16.4	13.9	2.5	28.6	7.5
4	14.0	12.6	1.4	27.1	8.1	16.9	14.4	2.5	27.8	8.9
5 (tip)	14.6	13.9	0.7	32.5	9.9	18.9	15.9	3.0	29.2	9.4
Young (E) 1 (base)	5.7	5.2	0.5	16.1	2.4	9.7	7.7	2.0	14.2	2.0
2	13.9	12.5	1.4	29.5	7.1	19.5	16.4	3.1	29.1	7.0
3	15.0	13.4	1.6	30.7	9.5	18.6	15.6	3.0	29.6	8.5
4+5 (tip)	15.8	15.3	0.5	31.9	10.7	21.8	18.6	3.0	33.2	9.7
Stem										
Base	26.2	21.1	5.1	27.1	18.2	27.8	21.2	6.6	29.2	10.4
Middle	15.2	12.1	3.1	13.9	9.7	18.3	13.4	4.9	19.5	4.2
Apex	5.2	4.4	0.8	10.4	2.1	9.5	5.7	3.8	11.2	1.1
Roots	27.4	19.0	8.4	31.6	19.9	30.1	21.6	8.5	32.5	18.8

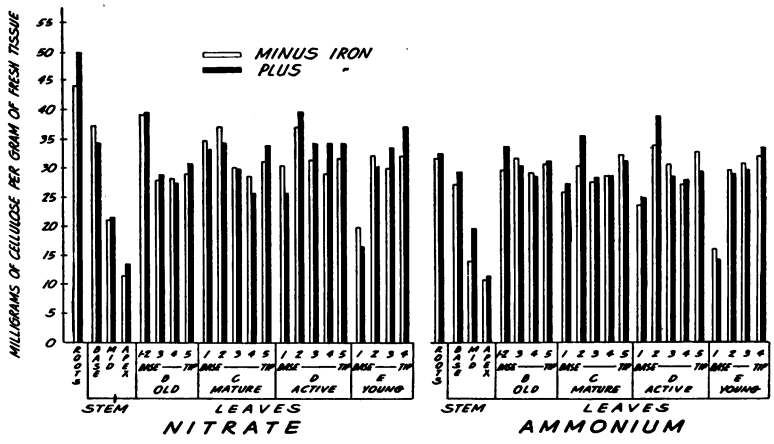

FIG. 15. Cellulose in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

were reversed. The results indicate that hexosans were more susceptible to variations by the difference in the amounts of iron and sources of inorganic nitrogen than pentosans.

CELLULOSE

The amounts of cellulose, reported in tables V and VI, and in figure 15, were affected very little by iron concentrations. The effects of the source of nitrogen in the solution culture on the amounts of cellulose in the plants of

TABLE VII

CALCULATED STATISTICAL SIGNIFICANCE* OF THE DIFFERENCE OF THE MEANS OF THE SUBSTANCES LISTED BELOW OF COMPARABLE SECTIONS OF THE LEAVES AND STEM OF PLANTS GROWN EITHER IN PLUS-IRON OR IN MINUS-IRON CULTURES AND SUPPLIED WITH NITRATE OR AMMONIUM SALTS AS SOURCES OF NITROGEN

SUBSTANCES	NITRATE-N				AMMONIUM-N			
	LEAVES		STEM		LEAVES		STEM	
	SIGNIFICANCE	IN FAVOR OF CULTURE	SIGNIFICANCE	IN FAVOR OF CULTURE	SIGNIFICANCE	IN FAVOR OF CULTURE	SIGNIFICANCE	IN FAVOR OF CULTURE
Chlorophyll	9999 : 1	+ Fe	.....	.....	9999 : 1	+ Fe	.....	.....
Carotenoids	9999 : 1	+ Fe	.....	.....	9999 : 1	+ Fe	.....	.....
Acidity	None	+ Fe	.....	.....	None	.....	.....	.....
Ascorbic acid	9999 : 1	− Fe	.....	.....	None	.....	.....	.....
Total sugars	9999 : 1	+ Fe	None	.....	9999 : 1	+ Fe	None	.....
Reducing "	None	.....	None	.....	9999 : 1	+ Fe	None	.....
Sucrose	9999 : 1	+ Fe	None	.....	None	.....	None	.....
Starch	4000 : 1	+ Fe	9999 : 1	+ Fe	9999 : 1	+ Fe	29 : 1	+ Fe
Pentosans	9999 : 1	+ Fe	None	.....	9999 : 1	+ Fe	None	.....
Hexosans	9999 : 1	− Fe	45 : 1	− Fe	None	.....	43 : 1	+ Fe
Celluloses	None	.....	None	.....	None	.....	None	.....
Lignin	None	.....	None	.....	None	.....	None	.....

* By CONRAD'S formula and from LOVE'S table.

the different treatments were small and lacked statistical significance. These small differences were in favor of the plants of the N-n series. It is suspected that they resulted from differences in the water content of the tissues, as shown in former publications (**10, 11**). The high cellulose content of the transitional subchlorophyllous (2) sections of the leaves of the "mature" (C) and "active" (D) groups is of great interest because it conforms with their structural requirements. The leaves of these groups require, on account of their great length and weight and their outwardly inclined position on the stem, greater structural reinforcement than do the leaves of the other groups in order to prevent their downward collapse.

Cellulose, being a structural and not a food or energy-yielding polysaccharide, did not undergo appreciable changes as a result of changes in the

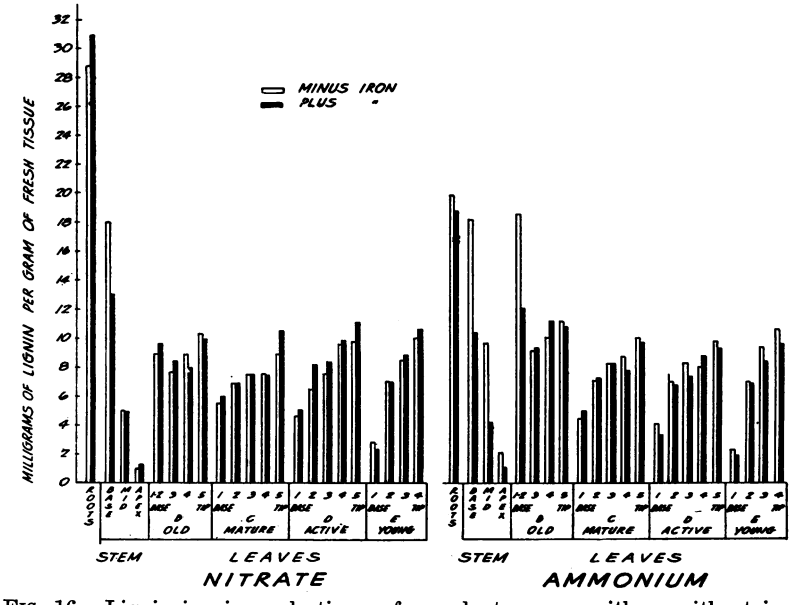

FIG. 16. Lignin in pineapple tissues from plants grown with or without iron, and with nitrate or ammonium nitrogen.

carbohydrate metabolism of the plants induced by the respective amounts of iron of the F and 0 cultures. Except for the smaller weights of the plants of the 0 than F cultures of the N-n series, no other changes could be observed appreciably affecting the values of cellulose.

LIGNIN

The lignin content of the plants of the different treatments recorded in tables V and VI, and in figure 16, ranged from one-half to one-tenth and averaged about one-fourth that of cellulose. Lignin, like cellulose, being a structural and not a food or energy-yielding substance was affected very little by the different amounts of iron in the 0 and F cultures. The data gathered so far indicate that the amounts of lignin, celluloses, and hemicellu-

loses were directly related to the quantities of the fibrovascular tissues of the respective plant sections.

Discussion

The traces of iron contained as impurities in the C. P. nutrient salts were not sufficient to promote good growth in the plants of the 0 cultures of the N-n series where the initial pH of the solution culture changed in a two-week period from 4.4 to 6.8. Such traces of iron were very effective, however, for promoting plant growth in the 0 cultures of the A-n series because the order of pH changes in the solution culture was reversed, shifting in the course of two weeks from an initial pH value of 6.6 to a final one of 4.3. Such changes affected the degree of iron solubility and its availability to plants (**12**). Where iron was available, as in the F cultures, chlorophyll was produced in ample amounts.

The minimum amounts of iron required by *A. comosus* for optimum chlorophyll development and growth are relatively small. Amounts ranging from 0.3 to 0.5 micrograms of iron per gram of fresh tissue have been found in many green tissues and may be considered adequate provided the rate of flow which maintains such iron concentrations within the plant tissues does not diminish appreciably (**12**).

The most obvious physiological function of iron is its influence on the formation of chlorophyll or more probably of protochlorophyll (**4**) and other related metalloporphyrins such as cytochrome-*c*, (**2**). Following the formation of chlorophyll the entire metabolic tempo of the plant is accelerated with respect to carbohydrate synthesis, nitrogen assimilation, and nutrient mineral requirements. This indicates that the physiological effects of iron on the general metabolism of the plant are very intimately related to the chlorophyll status of the plant. Hence it will be utterly misleading to place too much emphasis on the amounts of iron found in plant tissues or on those supplied externally to plants through solution cultures, and too little emphasis on the chlorophyll content of the leaves inasmuch as experience has shown that amounts of chlorophyll in the leaves do not always correlate with those of iron.

Certain interesting phenomena associated with the distribution of ascorbic acid are the gradients of concentrations of this substance in different sections of the leaves of different groups which in most cases were highest in the terminal and lowest in the basal sections. These gradients might have resulted from differences in the physiological functions of the different sections. The data also show that ascorbic acid had reached maximum values in the leaves of the "active" (D) and minimum values either in the "old" (B) or "young" (E) groups. The differences in the amounts of ascorbic acid between groups of leaves may be explained by the assumption that ascorbic acid, being a by-product of metabolic activity (possibly resulting, like many other acids, from the incomplete oxidation of sugars during respiration), is produced in greater amounts in leaves of great vigor and maximum synthetic activity such as the "active" (D) group than in immature

leaves such as the "young" (E) group. The relatively small amounts of ascorbic acid in the leaves of the "old" (B), and to a lesser extent of the "mature" (C) group, resulted possibly from catabolic processes caused by tissue senility. The lower values of ascorbic acid in the F than 0 cultures suggest possible oxidation of ascorbic acid by the iron of F cultures in all the groups of leaves of the N-n series [where average total Fe values, reported elsewhere (**12**), were 19.4 and 47.3 γ per gram of fresh leaf tissue for the 0 and F cultures, respectively]. In the "old" (B) and "mature" (C) leaf groups of the A-n series reduction in ascorbic acid values might have also been caused by oxidation since iron was 3.14 times more abundant in the F than 0 plants (F = 98.5, and 0 = 31.4 γ per gram of tissue). The only exceptions were certain sections of the leaves of the "active" (D) and "young" (E) groups of the A-n series, where the amounts of both ascorbic acid and iron were higher in the F than 0 cultures.

The values of titrable acidity, pH, and ascorbic acid indicate that the conditions in the chlorophyllous sections of the leaves were highly favorable for the solubility and reduction of iron, whereas in the basal sections of the leaves and in the stem they were not as satisfactory. However, chemical analyses for iron, reported previously (**12**), have shown no accumulations of iron either in the stem or in the basal sections of the leaves.

The plants of the N-n series, although including smaller amounts of chlorophyll than those of the A-n series of comparable iron cultures, contained greater amounts of sugars, suggesting that the conditions which had affected the metabolism of nitrogen rather than the amounts of chlorophyll were responsible for these differences.

The distribution of sugars varied considerably in the different groups of leaves. Such variations were caused by differences in the physiological functions of the sections and by the relative degree of vigor of the different groups of leaves. An analysis of such functions shows that the basal sections (1), lacking in chlorophyll, play no part in the photosynthetic processes of the plant in contrast to the chlorophyllous sections (2, 3, 4, 5) which are intimately associated with this mechanism. Also, the terminal sections (5) become senile and lose a great deal of their vigor and photosynthetic efficiency sooner than the immediately adjoining sections (2, 3, 4). Losses of vigor and photosynthetic activity are greater in the leaves of the "old" (B) and "mature" (C) than of the "active" (D) and "young" (E) groups. Therefore, accumulations of sugars in any section of the leaves should be associated either with a decreased rate of utilization as in cases of retarded growth, with highly favorable conditions for photosynthetic activity, or with a decreased rate of translocation from the leaves to the other organs. Low sugar values should be associated with conditions quite opposite those favoring accumulation. Thus, the lower values of sugars in the basal (1) sections of the leaves of the "young" (E) than of the "active" (D) or "mature" (C) groups suggest, on the basis of the above postulations, that a greater rate of sugar utilization for the building of new tissues was in

effect in the "young" (E) than in the "active" (D) or "mature" (C) and "old" (B) groups of leaves. Also, the accumulation of sugars in the terminal sections (5) of the leaves of all such groups may be attributed, on the basis of the same postulations, to a retarded rate of translocation or to the synthesis of much greater amounts than required for the energy-consuming processes of the cells and for the synthesis of various substances. However, certain exceptions to the above postulations as, for example, the slightly greater amounts of sugars in the basal sections of the leaves of the "old" (B) group of the 0 than F cultures of the N-n series cannot be satisfactorily interpreted. It is possible, however, that the higher values of sugars in the basal sections may be attributed either to cessation of the enzymic processes responsible for starch synthesis or to opposite effects such as hydrolysis of reserve starches resulting from tissue senility. The low sugar values in the medial section of the stem may be attributed to the rapid conversion of sugars into starch by very active enzymatic processes. The preponderance of dextrose and the relative absence of levulose in the stem might have resulted from hydrolyzed maltose which probably could have been released from starch by hydrolysis.

The amounts of sucrose increased gradually from the basal to the apical sections of the leaves and stem except in the transitional subchlorophyllous sections (2) of the leaves of the "young" (E) group where they decreased because of a high rate of metabolic activity tending to prevent the accumulation of this or other sugars in appreciable amounts.

Starch, although a reserve product like sucrose, accumulated in greater quantities in the stem than in the leaves. The stems of the N-n series contained almost twice as much starch as those of the A-n series. It is possible that this condition resulted either from differences in the rate of the photosynthetic activity of the leaves of the two series, or from the energy relations involved in the assimilation of the two forms of nutrient nitrogen. There is at present some suspicion, based on evidence from studies on the toxicity of chlorides to *A. comosus* (**13**), that chlorides supplied as $CaCl_2$ to nutrient solutions of the A-n series interfered with the polymerization of sugars into starches in the stem.

The small differences between the amounts of hemicelluloses in the plants of the F and 0 cultures indicate that they play a relatively minor rôle as energy-yielding substances in the carbohydrate economy of *A. comosus*. The data show that the hemicellulose content of the tissues was increased more by nitrate-nitrogen than by the amounts of iron in the F and 0 cultures.

The association of high values of hemicellulose sugars with plant organs containing great amounts of fibrovascular materials [such as the roots and to some extent the basal and terminal sections of the leaves of the "old" (B) and "mature" (C) groups] suggests the possible participation of these substances in the structural formation or cementation processes of fibers.

The amounts of celluloses and lignins were affected very little by the differences in the amounts of iron and by the forms of nitrogen. Both cellu-

lose and lignin, representing structural units in the plant body, remained in a static condition and were not susceptible to changes which had affected such energy-yielding carbohydrates as sugars and starches. The effects of the different treatments on cellulose and lignin were about the same as those on hemicelluloses, but differed considerably from those on starches or sugars.

Summary

The effects of plus- or minus-iron cultures in association with nitrate- or ammonium-nitrogen were studied in connection with the distribution of chlorophyll, carotenoids, ascorbic acid, sugars, starches, hemicelluloses, celluloses, and lignin in different sections of the leaves, stem, and roots of *A. comosus* and are summarized as follows.

1. Chlorophyll and carotenoid pigments were higher in the leaves of the plus-iron than minus-iron cultures. The plants of the ammonium-nitrogen series contained more pigments than the nitrate-nitrogen series for comparable iron cultures which possibly resulted from a greater availability of iron in the former than in the latter series.

2. Titrable acidity gradients reported as citric acid, increased from the basal to the terminal sections of the leaves of the plants of all cultures. The pH values of the sap were approximately inversely proportional to those of titrable acidity. The stem and roots had low values of titrable acidity and showed either little or no changes in the different treatments. The high acidity values of the sap from the chlorophyllous sections of the leaves and the exceedingly small amounts of iron present in the same tissues preclude the possibility of iron precipitation in these tissues of this organ.

3. Ascorbic acid was almost limited to the chlorophyllous sections of the leaves although the amounts present were not directly proportional to those of chlorophyll in the plants of the different cultures. The non-chlorophyllous sections of the plant, including the basal leaf, the stem, and the roots, contained either none or only mere traces of ascorbic acid. The gradients of distribution of ascorbic acid in the chlorophyllous sections of the leaves suggest that certain phases of metabolism rather than amounts of chlorophyll in these tissues were responsible for their ascorbic acid content. Reduction of the ascorbic acid values in old and senile leaves of plus-iron cultures suggests its possible oxidation by iron.

4. Sugars were found in greater amounts in the leaves of the "old," "mature," and "active" groups of the plus-iron than minus-iron cultures. They were higher in the plants of the nitrate- than of the ammonium-nitrogen series. The source of nitrogen in the nutrient solutions had influenced the amounts of sugars more than the amounts of chlorophyll in the leaves. Accumulations of sugars were greater in the basal sections of leaves of the "mature" than "young" groups because the rate of their utilization for new tissue-building processes had decreased in the former. Sugar values in the basal, medial, and apical sections of the stem were in most cases indirectly proportional to those of starches, suggesting that the rate of withdrawal of sugars from the stem is directly proportional to that of starch synthesis.

5. The amounts of levulose were slightly higher in the leaves of the minus-iron than plus-iron cultures of the nitrate-nitrogen series. Levulose was almost absent in the stem. Dextrose, occurring in amounts twice as great or more than levulose, was distributed in all organs of the plant. Sucrose was higher in the leaves of the plus-iron than minus-iron plants of the nitrate series, but in the ammonium series it was as high or slightly higher in the minus-iron than in the plus-iron cultures.

6. Starch was consistently higher in the stem of the plus- than minus-iron cultures, the former containing 101.3 per cent. more starch for the ammonium series and 33.2 per cent. more for the nitrate series. The plants of the nitrate-nitrogen series contained 161.6 per cent. more starch for the plus-iron and 295.0 per cent. for the minus-iron cultures than of the ammonium-nitrogen series.

7. The differences in total hemicellulose sugars between the plants of the plus- and minus-iron cultures were slight, with the plants of the former cultures containing, in most sections, greater amounts than the latter. The plants of the nitrate-nitrogen series contained greater amounts of hemicellulose sugars than those of the ammonium-nitrogen series. The differences between the amounts of hemicelluloses in the plants of the nitrate- versus the ammonium-nitrogen series were greater than those between the plus- versus the minus-iron cultures. The pentosan fractions of hemicelluloses of the nitrate series were higher in the plus- than in the minus-iron cultures, while the hexosan fractions were reversed. In the ammonium series the distribution of hexosans throughout the plant was irregular.

8. The amounts of cellulose were higher in the transitional sections which bear the strain for the erect position of the young leaves and reclined position of the mature leaves.

9. Lignin was generally high in those parts of the plant containing great amounts of fibrovascular tissues such as the roots, basal sections of the stem, and terminal sections of the leaves.

10. The amounts of both celluloses and lignin were affected only slightly by the different treatments and behaved in this respect like the hemicelluloses.

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LITERATURE CITED

1. ASSOCIATION OF OFFICIAL AGRICULTURAL CHEMISTS. Methods of analysis. 1940.
2. BALLS, ERIC G. Biological oxidations and reductions. Ann. Rev. Biochem. **11**: 1–25. 1942. Stanford University Press, California.
3. BESSEY, O. A., and KING, C. G. The distribution of vitamin C in plant and animal tissues and its determination. Jour. Biol. Chem. **103**: 687–698. 1933.

4. EYSTER, W. H. Protochlorophyll. Science n.s. **68**: 569–570. 1928.
5. PETERSON, C. J., WALDE, A. W., and HIXON, R. M. Effect of temperature on sulfuric acid method of lignin. Ind. & Eng. Chem. Anal. Ed. **4**: 216–217. 1932.
6. PUCHER, C. W., and VICKERY, H. B. Determination of starch in plant tissues. Ind. & Eng. Chem. Anal. Ed. **8**: 92–97. 1936.
7. QUISUMBING, F. A., and THOMAS, A. W. Conditions affecting the quantitative determination of reducing sugars by Fehling solution. Elimination of certain errors involved current in methods. Jour. Amer. Chem. Soc. **43**: 1503–1526. 1921.
8. SCHERTZ, F. M. The extraction and separation of chlorophyll (a + b) carotin and xanthophyll in fresh leaves preliminary to their quantitative determination. Plant Physiol. **3**: 211–216. 1928.
9. ————. The quantitative determination of chlorophyll. Plant Physiol. **3**: 323–334. 1928.
10. SIDERIS, C. P., KRAUSS, B. H., and YOUNG, H. Y. Assimilation of ammonium and nitrate by pineapple plants grown in nutrient solutions and its effects on nitrogenous and carbohydrate constituents. Plant Physiol. **13**: 489–527. 1938.
11. ————, ————, and ————. Distribution of different nitrogen fractions, sugars and other substances in various sections of the pineapple plant grown in soil cultures and receiving either ammonium or nitrate salts. Plant Physiol. **14**: 227–254. 1939.
12. ————, YOUNG, H. Y., and KRAUSS, B. H. Effects of iron on certain physiochemical properties and ash constituents of *Ananas comosus* (L.) Merr., grown in solution cultures supplied either with nitrate or ammonium salts as sources for nitrogen. Plant Physiol. **18**: 608–632. 1943.
13. ————, and YOUNG, H. Y. Influence of potassium on chloride toxicity in *Ananas comosus* (L.) Merr. Amer. Jour. Bot. **27**: 21s. 1941.
14. WIDDOWSON, ELSIE MAY. A method for the determination of small quantities of mixed reducing sugars and its application to the estimation of the products of hydrolysis of starch by taka diastase. Biochem. Jour. **25**: 863–879. 1931.