Vol. 65 SEPTEMBER 15, 1942 No. 6

JOURNAL OF AGRICULTURAL RESEARCH

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Published on the 1st and 15th of each month. This volume will consist of 12 numbers and the contents and index.

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Entire Journal: Domestic, $3.25 a year (2 volumes)
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JOURNAL OF AGRICULTURAL RESEARCH

VOL. 65 WASHINGTON, D. C., SEPT. 15, 1942 No. 6

THE EFFECTS OF LEAFHOPPER FEEDING INJURY ON APPARENT PHOTOSYNTHESIS AND TRANSPIRATION OF APPLE LEAVES [1]

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INTRODUCTION

The leafhoppers that attack apples have received relatively little attention from investigators and growers, probably because they do not cause immediate injury to the fruit itself as do codling moths, curculios, and similar chewing insects, and probably, too, because their greatest injury to the leaves appears late in the season when the fruit is almost mature.

Two types of leafhopper injury occur on apple leaves: (1) A yellowish-green stippling on the upper leaf surface caused by species of *Typhlocyba*, *Erythroneura*, and some *Empoasca*—the mesophyll feeders (*1, 4, 9, 10, 11*)[3]; and (2) a yellowing, puckering, and wilting of the leaf caused by one or more species of *Empoasca*—the vein feeders (*11, 12, 13*).[4] There appear to be no published data which show the effects of vein- or mesophyll-feeding leafhoppers on the metabolic processes of apple leaves. The literature deals largely with descriptions of visible leaf injury (*3, 6, 8*).

It is the purpose in this paper to point out the effect of various degrees of leafhopper feeding injury on apparent photosynthesis and transpiration of apple leaves. Seasonal trends in leafhopper population and the effect of feeding injury on leaf anatomy also are discussed.

MATERIALS AND METHODS

The leafhoppers used in this study were collected from a large McIntosh apple tree growing in an experimental orchard in southern Indiana and were shipped to Ohio State University by special delivery in hardware-cloth cages. The time in transit was less than 24 hours. The cages were ellipse-shaped, about 10 inches in height by 4 inches in diameter, and were filled with damp excelsior and wrapped with cheesecloth. This technique was employed in all seasons of the year; fresh material was used in the cages in summer. The population counts were made at 8 evenly distributed locations on the tree (fig. 1) during the summer of 1940. One thousand leaves were inspected weekly for the tree as a whole, 5 groups of 25 leaves each at each of the 8 locations.

When the leafhoppers arrived at the laboratory, they were released into special cages which covered the shoots of potted Stayman Winesap

[1] Received for publication October 8, 1941.

[2] The authors are indebted to the Tobacco By-Products & Chemical Corporation, Louisville, Ky., for a fellowship in connection with this study. Helpful advice from Prof. J. J. Davis, head, Department of Entomology, Purdue University Agricultural Experiment Station, is gratefully acknowledged.

[3] Italic numbers in parentheses refer to Literature Cited, p. 280.

[4] REED, T. W. THE APPLE LEAFHOPPERS OF WESTERN NEW YORK. 1933. [Unpublished thesis, Master of Science, Ohio State University, Columbus, Ohio.]

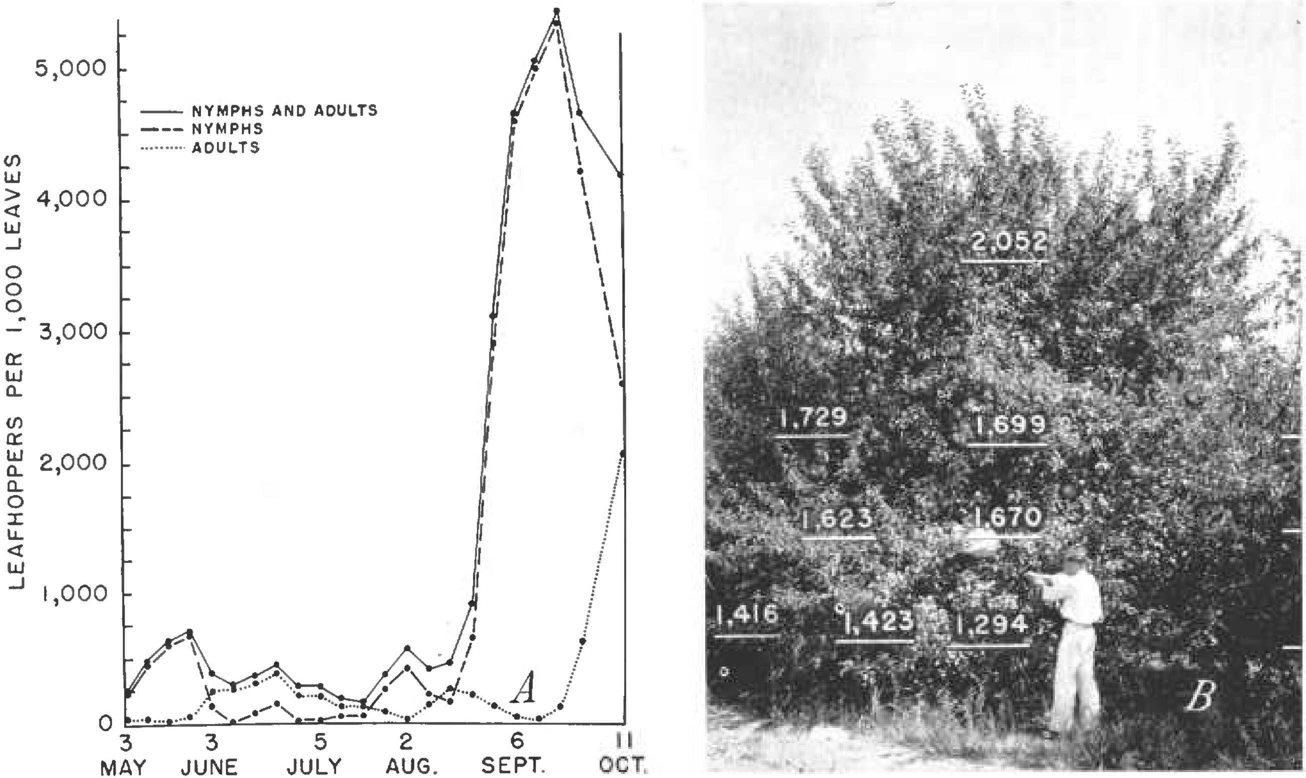

FIGURE 1.—*A*, Weekly population trend of leafhoppers on a McIntosh apple tree in southern Indiana during the 1940 season; *B*, the tree from which the leafhoppers were collected. The figures superimposed on the photograph are average numbers of insects per 1,000 leaves found each week in counts at 8 distributed locations at 3-, 7-, 12-, and 22-foot levels.

apple trees. The cages were made of ¼-inch-mesh hardware cloth covered with cheesecloth, and had small cellulose acetate windows through which the insects could be observed.

Photosynthesis and transpiration measurements were made at Ohio State University by the method of Heinicke and Hoffman (*7*), both outdoors and in the environment-control chamber (*2*). The metabolic activity of 12 leaves was studied in each experiment; 6 of the leaves were treated and 6 remained as checks. After a ratio between the rates of photosynthesis and transpiration of the 2 groups of leaves had been determined, a sleeve cage of insects was placed over one of the shoots as shown in figure 2. The insects were permitted to feed on the foliage for a given period, after which they were removed and the metabolism studies were resumed. Any change in the ratio after the feeding period as compared with the ratio before feeding was interpreted as the result of insect injury. The insects were permitted to feed at several successive periods in a given experiment.

Apple leaves upon which leafhoppers had been permitted to feed were examined microscopically, in cross section. The leaf material was mounted in paraffin, cut on a rotary microtome, and stained with Heidenhain's iron-alum haematoxylin.

Analysis of variance and significance of data were determined according to Fisher's tables for F and t.[5]

EXPERIMENTAL RESULTS

POPULATION STUDIES

Counts of leafhoppers in the experimental orchard were made each week, beginning May 3, 1940. After the overwintering population of *Erythroneura* species had died about the middle of May, *Typhlocyba pomaria* McA., the white apple leafhopper, was the only species present in significant numbers until late in the summer. The graph in figure 1 shows the weekly population of all leafhoppers, adults and nymphs, on the mature McIntosh tree. The first peak in the curve represents almost entirely the species *T. pomaria;* the second, several species of *Erythroneura;* the third, *T. pomaria* and some *Erythroneura;* and the last, *T. pomaria* and various species of *Erythroneura* in the ratio of 2.83 *T. pomaria* to 1 of Erythroneura. The figures on the photograph in figure 1, *B*, show the average number of leafhoppers per 1,000 leaves found each week in counts at the respective locations up to August 4, 1940. It will be noted that the insects were more abundant near the top of the tree.

Eggs of *Typhlocyba pomaria* were laid in the bark of 1- to 4-year wood in the fall and these overwintered before hatching; *Erythroneura* overwintered in the adult stage under leaves and other protective plant debris. Population counts of *Erythroneura* during the winter of 1939–40 revealed an average of 160 adults per square yard in fallen leaves and grass in the orchard. The leafhoppers were more numerous near the drip of the branches where ground cover was heavier.

EFFECTS ON PHOTOSYNTHESIS AND TRANSPIRATION OF APPLE LEAVES CAUSED BY FEEDING OF APPLE LEAFHOPPERS

One hundred leafhoppers per leaf were released in a sleeve cage over apple shoots after the pretreatment ratio for photosynthesis and transpiration had been established over a 3-day period. The insects were

[5] The statistical analyses were carried out under the direction of Mrs. Z. E. Alberts, Department of Zoology and Entomology, Ohio State University.

permitted to feed for 3 days, after which leaf activity was measured again. The data in table 1 and figure 3, *A*, show that the rate of photosynthesis was reduced 25 percent and transpiration 26.7 percent by the feeding of leafhoppers at the rate of 100 per leaf for 3 days, or 7,200 "leafhopper hours" per leaf (see fig. 4, *A*, *a*, for apparent leaf injury).

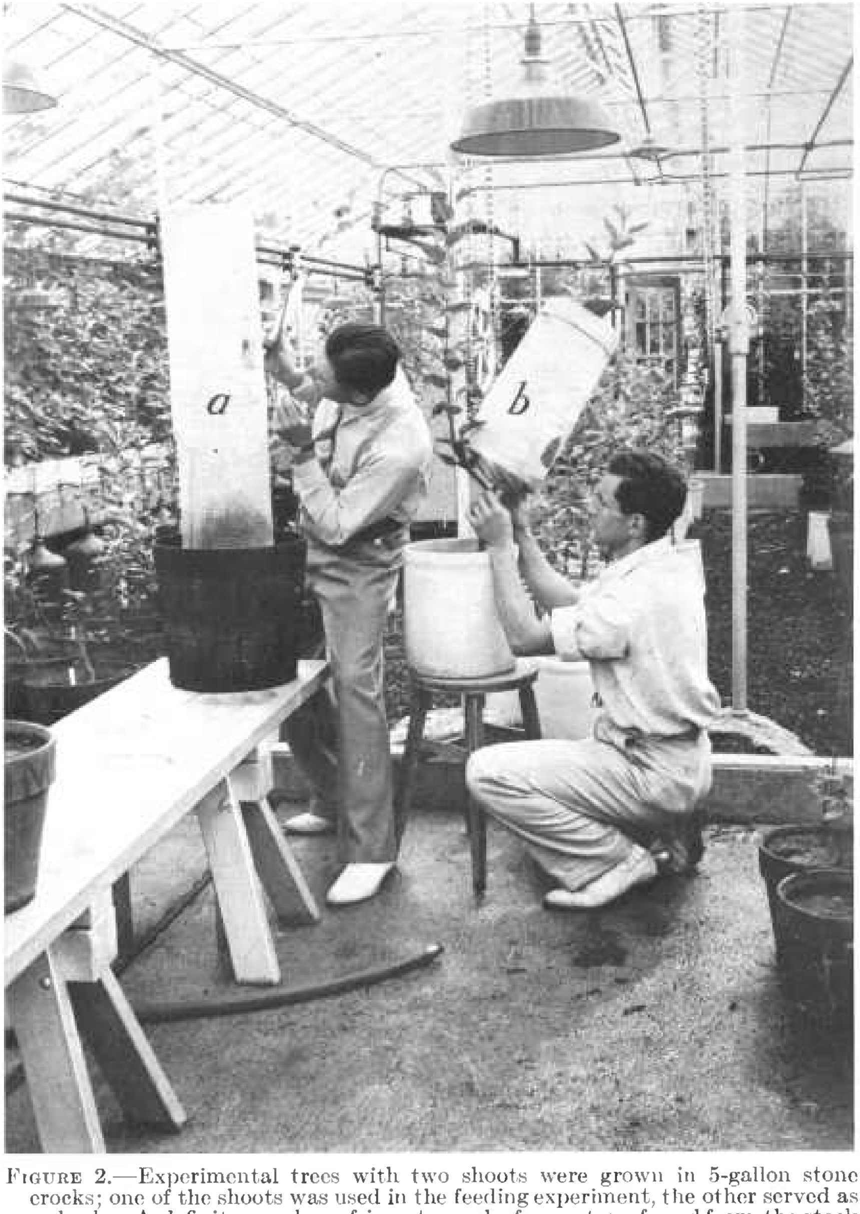

FIGURE 2.—Experimental trees with two shoots were grown in 5-gallon stone crocks; one of the shoots was used in the feeding experiment, the other served as a check. A definite number of insects per leaf were transferred from the stock cage (*a*) to the test cage (*b*) for each successive feeding period between photosynthesis-transpiration determinations.

TABLE 1.—*The effect of apple leafhopper injury on the rates of photosynthesis and transpiration of Stayman Winesap apple leaves, experiment A*

Date of leaf activity measurement [1]	CO_2 per cu. ft. of air	Photosynthesis (CO_2 per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}}\times 100$	Expected rate	H_2O per cu. ft. of air	Transpiration (H_2O per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}}\times 100$	Expected rate [2]
		Check	Test				Check	Test		
	Milligrams	*Milligrams*	*Milligrams*		*Percent* [2]	*Gram*	*Grams*	*Grams*		*Percent*
Mar. 3	19.9	10.5	14.0	102.9		0.302	0.56	0.83	148.2	
Mar. 4	19.0	7.0	7.8	97.1		.305	.94	1.18	125.5	
Mar. 5	18.0	17.5	20.8	118.3		.292	1.10	1.50	136.4	
Average				106.1					136.7	
Mar. 9	19.2	15.1	15.9	105.3	99.3	.251	1.47	1.55	105.4	77.1
Mar. 10	17.4	13.9	9.1	65.5	61.8	.267	1.62	1.48	91.3	66.8
Mar. 11	17.7	12.9	11.7	90.7	85.5	.230	1.63	1.65	101.9	74.6
Mar. 12	17.1	13.1	7.5	57.3	54.0	.261	1.56	1.63	104.5	76.5
Average				79.7	75.1				100.7	73.8
Mar. 16						.255	1.58	1.77	112.0	82.0
Mar. 17	17.7	20.3	16.6	81.8	77.1	.257	1.60	1.62	101.3	74.1
Mar. 18						.249	1.62	1.68	103.7	75.9
Mar. 19	16.8	18.6	15.4	82.8	78.1	.231	1.97	1.43	72.6	53.1
Average				82.3	77.6				97.4	71.3
Apr. 1	17.3	12.3	10.0	81.3	76.6	.264	1.75	1.75	100.0	73.2
Apr. 2						.261	2.06	1.65	80.1	58.6
Apr. 3	18.2	14.1	11.9	84.4	79.6	.249	2.13	1.76	82.6	60.4
Apr. 4	16.5	15.0	13.9	92.7	87.4	.255	2.10	1.25	59.5	43.5
Apr. 5	18.2	15.8	11.3	71.1	67.0	.265	1.44	1.56	108.3	79.3
Apr. 6	18.3	15.8	12.7	80.4	75.8	.250	1.97	1.41	71.6	52.5
Average				82.0	77.3				83.7	61.2
Apr. 12	17.8	17.1	12.8	74.9	70.6	.262	1.80	1.77	98.3	71.9
Apr. 13	17.4	14.1	10.6	75.2	70.9	.257	1.83	1.82	99.5	72.8
Apr. 14	17.7	14.2	11.7	82.4	77.7	.253	1.83	1.54	84.2	61.6
Apr. 15	19.4	16.3	16.7	102.5	97.6	.261	1.47	1.20	81.6	59.7
Apr. 16	18.0	17.5	17.3	98.9	93.3	.254	1.13	.96	85.0	62.2
Apr. 19	17.7	14.0	9.9	70.9	66.9					
Average				84.3	74.3				89.7	65.6
Apr. 26	15.9	12.3	11.0	89.4	84.3	.250	1.64	1.55	94.5	69.1
Apr. 28						.256	1.83	1.33	72.1	52.7
Apr. 29	16.8	15.5	6.3	40.6	38.3	.248	1.86	1.64	88.2	64.5
Apr. 30	16.5	15.1	8.8	58.3	55.0	.251	1.92	1.53	79.7	58.3
May 1	17.3	12.0	7.3	60.8	57.3	.250	1.90	1.35	71.1	52.0
May 2	17.7	14.5	6.6	45.5	42.9	.228	2.32	1.78	76.7	56.1
Average				58.9	55.8				80.4	59.6
May 21	16.5	4.4	.9	20.5	19.3	.216	1.32	.93	70.5	51.5
May 22	15.6	6.3	0	0	0	.269	1.18	.76	64.4	47.0
May 23	16.3	8.3	3.6	43.4	40.9	.275	1.14	.75	65.8	48.2
Average				21.3	20.1				66.9	48.9

ANALYSIS OF VARIANCE OF $\frac{\text{TEST}}{\text{CHECK}} \times 100$

Item	Degrees of freedom	Sum of squares	Mean of squares	*F*	Remarks
Photosynthesis	6	14,158.62	2,359.8	3.27	Significant.
	21	15,174.66	722.6		
Transpiration	6	9,553.04	1,592.2	10.54	Highly significant.
	24	3,626.39	151.1		

[1] Between each series of measurements the leafhoppers were permitted to feed.

[2] $$\frac{\dfrac{\text{Test after treatment}}{\text{Check after treatment}}}{\dfrac{\text{Test before treatment}}{\text{Check before treatment}}}\times 100 = \text{Percent expected rate}$$

From these data it could be inferred that 300 leafhoppers per leaf (assuming a constant area of about 50 cm.[2] for all leaves) may cause a reduction in photosynthesis and transpiration of about 1 percent in 1 hour.

Further injury (fig. 4, *A*, *b*) on the same leaves did not cause a significant reduction in either process. It was not until 4 weeks of

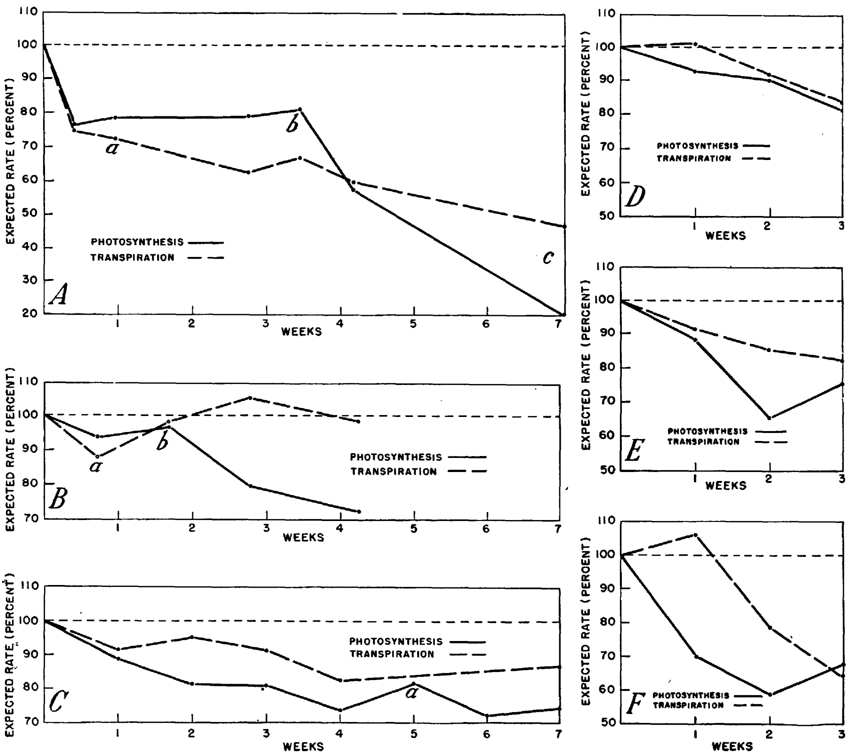

FIGURE 3.—*A*, *B*, *C*, *D*, and *E*, based on data from experiments of similar designation, show the effects of mesophyll-feeding by apple leafhoppers on apparent photosynthesis and transpiration of Stayman Winesap apple leaves. *F* shows the effect of the vein-feeding potato leafhopper, *Empoasca fabae*, on apple leaf metabolism. The value 100 represents photosynthesis and transpiration of the check leaves for every determination.

feeding with a slightly reduced insect population that further measurable reductions occurred. Seven weeks of feeding reduced the rate of photosynthesis 80 percent and transpiration 56.2 percent. At this time the leaves appeared severely injured; the surfaces were yellowish green, the spots had coalesced over the entire area, and there was a slight curling and burning at the margins (fig. 4, *A*, *c*). The reduction in photosynthesis was significant and in transpiration highly significant.

EXPERIMENT B

In similar experiments but with a smaller population of insects (50 to 60 per leaf), there was a reduction in photosynthesis but the

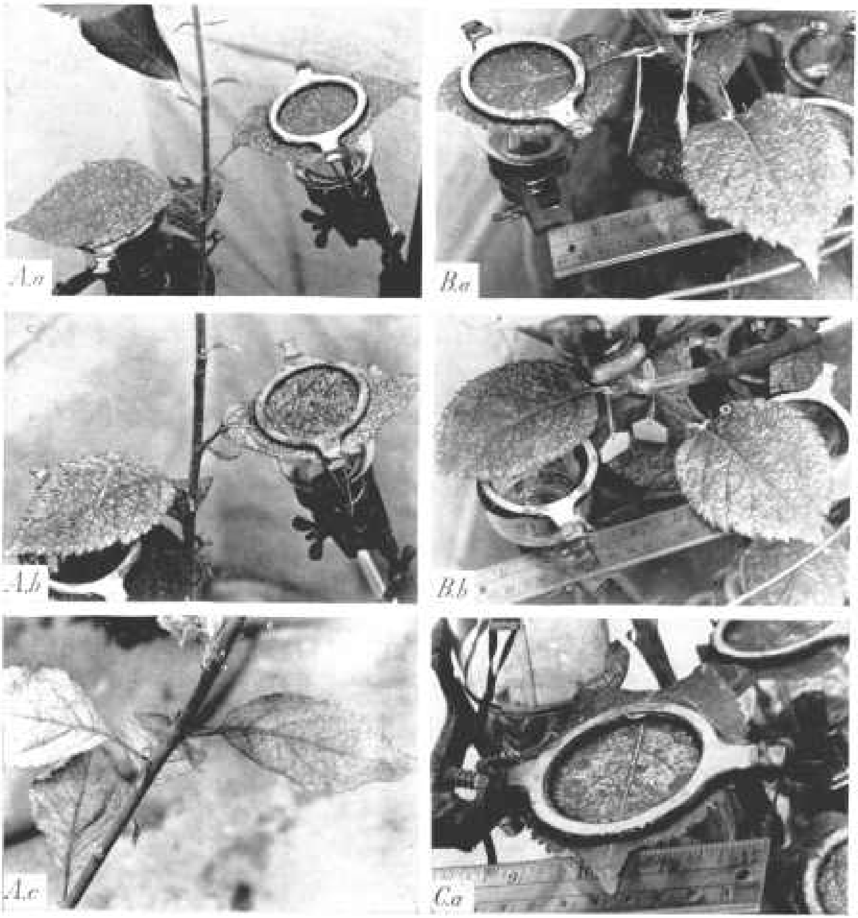

FIGURE 4.—Stages of feeding injury at positions marked on the photosynthesis-transpiration curves in figure 3; for example, *A*, *a*, shows apparent feeding injury when photosynthesis was reduced approximately 25 percent and transpiration 27 percent as indicated at point *a* in figure 3, *A*.

rate of transpiration was not significantly affected (table 2, fig. 3, *B*). Four weeks of feeding was accompanied by a significant reduction in photosynthesis of 26.4 percent. (See fig. 4, *B*, *a*, and *B*, *b*, for apparent injury at stages specified in fig. 3, *B*.)

TABLE 2.—*The effects of apple leafhopper injury on the rates of photosynthesis and transpiration of Stayman Winesap apple leaves, experiment B*

Date of leaf activity measurement [1]	CO_2 per cu. ft. of air	Photosynthesis (CO_2 per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}}\times 100$	Expected rate	H_2O per cu. ft. of air	Transpiration (H_2O per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}}\times 100$	Expected rate [2]
		Check	Test				Check	Test		
	Milligrams	*Milligrams*	*Milligrams*		*Per cent* [2]	*Gram*	*Grams*	*Grams*		*Per cent*
Mar. 16	19.4	19.4	19.3	99.5	-------	-------	-------	-------	-------	-------
Mar. 17	16.4	22.1	22.2	100.5	-------	0.248	1.78	1.46	82.0	-------
Mar. 18	16.4	16.5	15.5	93.9	-------	.240	1.81	1.69	93.4	-------
Mar. 19	17.6	18.3	17.9	97.8	-------	.237	1.87	1.66	88.8	-------
Average	-------	-------	-------	97.9	-------	-------	-------	-------	88.1	-------
Mar. 26	18.0	19.7	20.1	102.0	104.0	.244	1.33	1.06	80.0	90.8
Mar. 27	20.7	29.0	28.1	97.0	99.1	.252	1.31	.92	70.2	80.0
Mar. 28	22.4	24.6	18.0	73.2	74.7	.269	1.47	1.48	100.7	114.3
Mar. 29	17.7	16.9	17.6	104.1	106.3	.260	1.69	1.25	74.0	84.0
Mar. 30	17.2	17.0	13.7	80.6	82.3	.279	1.59	1.38	86.8	98.5
Mar. 31	16.9	12.2	12.7	104.1	106.3	.251	1.12	.84	75.3	85.5
Average	-------	-------	-------	93.5	95.5	-------	-------	-------	81.2	92.2
Apr. 7	18.6	15.1	14.4	95.3	97.4	.258	1.44	1.33	92 4	105.0
Apr. 8	17.0	14.6	15.7	107.5	109.8	.241	1.85	1.74	94.1	106.8
Apr. 9	16.8	15.3	13.4	87.8	89.7	.260	1.58	1.55	98.1	111.4
Apr. 10	18.2	10.4	10.1	97.1	99.2	.246	1.22	1.02	83.6	94.9
Apr. 11	-------	-------	-------	-------	-------	.259	1.46	1.27	87.0	98.8
Average	-------	-------	-------	96.9	99.8	-------	-------	-------	91.0	103.4
Apr. 20	30.6	23.5	16.7	71.1	72.6	.250	.86	.88	102.3	116.1
Apr. 21	25.6	22.2	18.5	83.3	85.1	.256	1.21	1.26	104.1	118.2
Apr. 22	17.5	14.7	11.9	81.0	82.7	.252	1.39	1.29	92.8	105.3
Apr. 23	17.9	13.4	9.5	70.9	72.4	.253	1.61	1.52	94.4	107.2
Apr. 24	15.9	14.7	11.6	78.9	80.6	.248	1.47	1.55	105.4	119.6
Apr. 25	18.1	24.0	21.1	87.9	89.8	.262	1.63	1.48	90.8	103.1
Average	-------	-------	-------	78.9	80.5	-------	-------	-------	98.3	111.6
May 5	16.3	14.3	10.0	69.9	71.4	.259	1.66	1.61	92.0	110.1
May 6	15.8	11.2	10.6	94.6	96.6	.259	1.98	1.59	80.3	91.1
May 7	17.7	14.1	7.3	51.8	52.9	.277	1.81	1.74	96.1	109.1
Average	-------	-------	-------	72.1	73.6	-------	-------	-------	92.5	103.4

ANALYSIS OF VARIANCE OF $\frac{\text{TEST}}{\text{CHECK}} \times 100$

Item	Degrees of freedom	Sum of squares	Mean of squares	F	Remarks
Photosynthesis	4	2153.45	538.36	4.28	Significant.
	18	2263.59	125.76		
Transpiration	4	901.98	225.5	3.40	Significant variance but not a reduction.
	18	1197.42	66.37		

[1] Between each series of measurements the leafhoppers were permitted to feed.
[2] See footnote 2, table 1, for method of calculation.

Experiments C and D

Experiments were run on experimental leaves in the laboratory to duplicate the population of insects in the field. At the time weekly population counts were made in the field, leafhoppers (adults and nymphs) were collected and shipped from the Indiana orchard to the environment-control chamber. The concentration of insects, species ratio, and interval between changes in insect population on the test leaves were adjusted to make the duration of the experiment in the laboratory shorter than an entire summer season. The data can be interpreted, however, as typical of the results of a season of leafhopper activity.

The technique employed in handling the leafhoppers in these experiments differed from that in the previous experiments. The leafhoppers were released in the leaf cups (*7*) on the lower surfaces of the apple leaves instead of in cheesecloth cages. According to Wigglesworth (*14, p. 198*), insects respire at the rate of approximately 3 mg. of carbon dioxide per gram of insect weight per hour. A leafhopper, then, would respire about 0.015 mg. of carbon dioxide per hour. At this rate the maximum number of leafhoppers per leaf at any time in the course of the 2½-hour determination would not respire enough carbon dioxide to interfere with the photosynthesis measurements. Thus, the insects were permitted to feed continuously even during the determinations and were confined to the part of the leaf surface included by the leaf cup rather than to all the surface included in a sleeve cage.

The data in table 3 and figure 3, *C*, show that at a point in the experiment corresponding to May 17 in the field, photosynthesis was reduced 16.5 percent and transpiration 4.9 percent. At a point corresponding to June 25 the reductions were, respectively, 19.2 and 13.8 percent, and by July 26 they were 27.1 and 12.9 percent. It would appear from these data that before mid-July, when the insect population is still rather low, marked reductions may occur in the rate of photosynthesis, especially in the early part of the period. The gradual reduction in photosynthesis recorded from week to week in experiment C was not significant, but the final reduction at a point corresponding to July 26 was highly significant (see fig. 4, *C*, *a*, for apparent injury toward end of experiment). The reduction in transpiration was not significant. In experiment D (table 4 and fig. 3, *D*), which was carried only to a point corresponding to June 5 in the field, reductions in transpiration and photosynthesis, although not significant, were recorded.

Experiment E

Table 5 and figure 3, *E*, show the results of a fifth experiment with the various species of apple leafhoppers. This experiment yielded data similar to those in the experiments described above. Three feeding periods with a population of approximately 25 leafhoppers per leaf brought about a reduction in the rate of photosynthesis of 23.8 percent and in transpiration of 16.5 percent. The reduction in transpiration over the entire period of the experiment was significant; the reduction in photosynthesis was significant from one feeding period to the next and highly significant over the 3-week period.

TABLE 3.—*The effects of apple leafhopper injury on the rates of photosynthesis and transpiration of Stayman Winesap apple leaves, experiment C*

Date of leaf activity measurement [1]	CO_2 per cu. ft. of air	Photosynthesis (CO_2 per 100 cm.² per hour)		$\frac{\text{Test}}{\text{Check}}\times 100$	Expected rate	H_2O per cu. ft. of air	Transpiration (H_2O per 100 cm.² per hour)		$\frac{\text{Test}}{\text{Check}}\times 100$	Expected rate [2]
		Check	Test				Check	Test		
	Milligrams	*Milligrams*	*Milligrams*		*Percent* [2]	*Grams*	*Grams*	*Grams*		*Percent*
May 25	17.0	14.3	24.6	172.0		0.212	1.49	1.97	132.2	
May 26	16.3	14.5	20.1	138.6		.200	1.56	2.06	132.1	
May 27	17.4	14.8	24.6	166.2		.224	1.35	1.86	137.8	
May 28	16.8	15.3	23.9	156.2		.215	1.51	2.07	137.1	
May 29	14.8	11.0	18.0	163.6		.217	1.39	1.78	134.3	
Average				159.3					133.5	
May 30	15.2	19.0	28.5	150.0	94.2	.212	1.69	1.99	117.8	88.2
May 31	15.5	20.3	26.0	128.1	80.4	.191	1.40	1.82	130.0	97.8
June 1	14.8	17.3	23.0	133.0	83.5	.185	1.56	1.90	121.8	91.2
June 2	14.9	18.8	25.7	136.7	85.8	.175	1.64	2.31	140.9	105.5
June 3	16.4	18.4	24.6	133.7	84.0	.182	1.69	1.96	116.0	86.9
June 4	17.3	16.3	25.6	157.1	98.7	.189	1.63	1.85	113.5	85.0
June 5	15.5	13.4	21.2	158.2	99.3	.233	1.44	1.66	115.3	86.4
June 7	17.6	18.7	25.8	138.0	86.7	.215	1.35	1.50	111.1	83.2
June 8	16.2	14.4	20.6	143.1	89.9	.573	1.12	1.79	159.8	119.7
June 9	17.7	21.0	29.1	138.6	87.0	.192	1.53	2.18	142.5	106.7
June 10	17.8	17.1	26.1	152.6	95.8	.181	1.55	2.03	131.0	98.3
June 11	17.9	34.9	43.4	124.4	78.1	.185	1.64	2.18	132.9	99.5
June 15	16.6	11.5	15.6	135.7	85.2					
June 16	16.6	15.2	15.7	103.3	64.9					
June 17	17.7	16.6	22.9	138.0	86.7					
June 18	17.1	13.2	22.1	167.4	105.1					
June 19	16.5	10.2	13.4	131.4	82.5	.385	1.38	1.70	123.2	92.3
June 20	16.5	11.2	14.9	133.0	83.5	.279	3.83	4.10	107.0	80.1
June 21	18.4	19.9	23.4	117.6	73.9	.405	1.23	1.53	119.5	89.5
June 22	15.7	13.7	14.3	104.4	65.6	.406	1.54	2.02	131.2	98.3
June 23	15.2	10.7	12.3	115.0	72.2	.430	1.31	1.68	128.2	96.0
June 24	15.1	5.9	8.5	144.1	90.5	.418	1.09	1.35	123.9	92.8
June 25	15.0	9.5	11.5	121.1	76.1	.409	1.21	1.42	117.4	87.9
June 27	15.9	16.0	15.6	97.5	66.8	.414	1.23	1.44	117.1	87.1
June 28	15.1	8.3	11.6	139.8	95.8	.414	1.28	1.26	98.4	79.4
June 29	15.3	12.4	13.3	107.3	73.5	.415	1.27	1.27	109.4	88.3
June 30	15.9	7.0	8.8	125.7	86.1	.415	1.16	1.32	113.8	91.8
July 1	17.1	10.3	9.7	96.1	65.8	.504	1.18	1.19	100.8	81.4
July 2	14.4	11.0	11.0	100.0	68.5	.316	1.03	.95	92.2	82.7
July 3	17.0	13.7	18.8	137.2	87.7	.403	2.08	2.35	113.0	88.4
July 8	16.1	7.2	11.6	161.1	102.9	.459	1.25	1.66	132.8	104.1
July 9	16.7	10.6	12.9	121.7	77.8	.464	1.25	1.50	122.0	95.6
July 10	18.3	12.5	16.0	128.0	81.8					
July 11	16.5	10.0	14.1	141.0	90.1					
July 13	16.8	11.7	11.6	99.1	63.3					
July 14	17.4	13.4	17.3	129.1	82.5					
July 15	21.1	15.2	22.7	149.3	95.4					
July 16	15.0	7.4	8.8	118.9	76.0					
July 17	15.8	9.5	8.6	90.5	57.8					
July 18	18.2	11.2	11.5	102.7	65.6					
July 19	21.7	14.2	12.4	87.3	55.8					
July 20	18.4	10.5	15.6	148.6	95.0	.404	1.21	1.37	113.2	88.7
July 23	16.4	10.7	12.1	113.0	80.8					
Average				127.9	81.8				120.5	92.2

ANALYSIS OF VARIANCE OF $\frac{\text{TEST}}{\text{CHECK}}\times 100$

Item	Degrees of freedom	Sum of squares	Mean of squares	*F*	Remarks
Photosynthesis	1 46	4,407.5 18,548.8	4,407.5 403.2	10.93	Highly significant.
Transpiration	1 31	870.1 44,461.7	870.1 1,434.3	1.65	Not significant.

[1] Leafhoppers were permitted to feed (field concentration) continuously even during the determinations.
[2] See footnote 2, table 1, for method of calculation.

TABLE 4.—*The effects of apple leafhopper injury on the rates of photosynthesis and transpiration of Stayman Winesap apple leaves, experiment D*

Date of leaf activity measurement [1]	CO_2 per cu. ft. of air	Photosynthesis (CO_2 per 100 cm.[2] per hour)		$\frac{Test}{Check}\times 100$	Expected rate	H_2O per cu. ft. of air	Transpiration (H_2O per 100 cm.[2] per hour)		$\frac{Test}{Check}\times 100$	Expected rate [2]
		Check	Test				Check	Test		
	Milligrams	*Milligrams*	*Milligrams*		*Percent* [2]	*Gram*	*Gram*	*Gram*		*Percent*
June 15	16.1	17.3	22.3	128.9		0.177	1.77	2.20	124.3	
June 16	15.6	15.0	19.2	128.0		.207	1.67	1.79	107.2	
June 17	17.7	19.3	23.2	120.2		.234	1.72	1.82	105.8	
June 18	16.1	17.7	22.7	128.2		.201	1.63	1.76	108.0	
June 20	16.5	16.7	21.5	128.7		.389	2.40	2.50	104.2	
June 21						.357	2.61	2.80	107.3	
June 23						.371	2.34	2.61	111.5	
June 24	14.5	13.8	17.3	125.4		.450	1.72	1.80	104.7	
June 25	14.7	11.3	13.7	120.2		.374	2.04	2.30	112.7	
Average				125.7					109.3	
June 28	13.9	17.8	17.2	96.6	76.8	.377	2.16	2.48	114.8	105.6
June 29						.401	2.07	2.35	113.5	104.4
June 30	14.9	20.7	24.9	120.3	95.7	.385	1.88	2.15	114.4	105.1
July 1						.324	1.59	1.82	114.5	105.2
July 2	15.2	12.1	15.6	128.9	102.6	.379	1.96	1.93	98.5	90.7
July 3	14.3	15.9	17.1	107.6	85.6	.371	1.81	2.21	122.1	112.2
July 8	15.8	9.7	13.4	138.1	109.9	.404	1.92	2.35	122.5	112.5
July 9	14.9	10.3	9.6	93.2	74.1	.408	1.92	1.82	94.8	87.1
July 11	14.8	10.6	13.6	128.3	102.1					
July 12						.367	1.87	1.96	104.8	96.3
July 13	16.2	10.1	12.2	120.8	96.2	.335	2.48	2.54	102.4	94.1
July 14	16.1	12.8	15.5	121.1	96.4	.313	2.09	2.18	104.3	95.9
July 15	14.4	9.6	10.1	105.2	87.7	.342	1.82	1.99	109.3	100.4
July 16	14.6	10.9	13.6	124.2	103.8	.334	2.14	2.12	99.1	84.5
July 17	14.9	11.9	9.9	83.2	65.1					
July 18	15.0	12.8	16.0	125.0	97.8	.236	2.01	2.03	101.0	97.0
July 20	14.0	12.6	13.3	105.6	82.7	.350	1.90	1.79	94.2	90.4
July 21	14.0	8.0	8.9	111.3	87.1	.348	1.67	1.70	101.8	97.7
July 22	14.2	13.3	10.0	75.2	58.9	.332	1.86	1.70	91.4	87.7
July 23	15.4	10.4	13.1	126.0	98.7	.312	1.96	1.49	76.0	73.0
Average				112.8	89.5				99.1	96.7

ANALYSIS OF VARIANCE OF $\frac{TEST}{CHECK}\times 100$

Item	Degrees of freedom	Sum of squares	Mean of squares	*F*	Remarks
Photosynthesis	1	868.3	868.3	3.85	Reduction, but not significant.
	22	4,960.1	225.5		
Transpiration	1	208.2	208.2	2.36	Reduction, but not significant.
	26	2,292.6	88.2		

[1] Leafhoppers were permitted to feed (field concentration) continuously even during the determinations.
[2] See footnote 2, table 1, for method of calculation.

TABLE 5.—*The effects of apple leafhopper injury on the rates of photosynthesis and transpiration of Stayman Winesap apple leaves, experiment E*

Date of leaf activity measurement [1]	CO_2 per cu. ft. of air	Photosynthesis (CO_2 per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}} \times 100$	Expected rate	H_2O per cu. ft. of air	Transpiration (H_2O per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}} \times 100$	Expected rate [2]
		Check	Test				Check	Test		
	Milligrams	*Milligrams*	*Milligrams*		*Per cent* [2]	*Grams*	*Grams*	*Grams*		*Per cent*
Aug. 2	16.9	30.1	32.3	107.3	-------	0.416	3.11	2.90	93.2	-------
Aug. 3	15.5	18.6	19.8	106.5	-------	.384	2.43	2.42	99.6	-------
Aug. 4	15.9	24.9	26.2	105.2	-------	.405	2.15	1.88	87.8	-------
Average	-------	-------	-------	106.3	-------	-------	-------	-------	93.4	-------
Aug. 14	16.4	25.2	23.4	92.9	87.4	.416	2.82	2.24	79.4	85.0
Aug. 15	15.9	25.1	24.8	98.8	93.0	.452	2.97	2.58	86.9	93.0
Aug. 18	16.1	18.4	17.4	94.6	89.0	.386	3.28	3.15	96.0	102.7
Average	-------	-------	-------	95.4	89.8	-------	-------	-------	87.4	93.6
Sept. 1	18.1	15.5	10.6	68.4	64.4	.340	2.84	2.61	91.9	98.3
Sept. 2	16.8	19.6	11.5	58.7	55.2	.332	2.46	1.88	76.4	81.7
Sept. 3	21.1	18.5	15.5	83.8	78.9	.410	2.45	1.87	76.3	81.6
Average	-------	-------	-------	70.3	66.2	-------	-------	-------	81.5	87.2
Sept. 9	-------	-------	-------	-------	-------	.436	1.81	1.30	71.8	76.8
Sept. 10	16.5	12.3	12.2	99.2	93.3	.427	2.54	2.06	81.1	86.8
Sept. 11	15.9	15.6	9.8	62.8	49.5	.434	2.23	1.69	75.8	81.1
Average	-------	-------	-------	81.0	71.3	-------	-------	-------	77.9	81.6

ANALYSIS OF VARIANCE OF $\frac{\text{TEST}}{\text{CHECK}} \times 100$

Item	Degrees of freedom	Sum of squares	Mean of squares	*F*	Remarks
Photosynthesis	1	1,249.8	1,249.8	11.76	Highly significant.
	9	956.4	106.3		
Transpiration	1	277.8	277.8	5.26	Significant.
	10	528.6	52.9		

[1] Between each series of measurements the leafhoppers were permitted to feed.
[2] See footnote 2, table 1, for method of calculation.

EFFECTS ON PHOTOSYNTHESIS AND TRANSPIRATION OF APPLE LEAVES CAUSED BY FEEDING OF THE POTATO LEAFHOPPER

EXPERIMENT F

The potato leafhopper (*Empoasca fabae* (Harr.)) is among the least common of the species that occur in the orchard, but the injury that is causes is easily identified. The typical feeding puncture is made in the vascular system of the leaf or stem and this usually results in blocking the conducting tissues (*11*). The leaf area distal to the point of injury becomes pale green in a triangular area bounded by veins and the leaf margin. Only mature leaves were used in this experiment to avoid the typical curling that results from the feeding of *E. fabae* on leaves that have not reached full size.

In a typical experiment about 10 to 15 leafhoppers per leaf were released in the cheesecloth cage over the apple shoot, after a relationship in metabolism had been established between the two groups of leaves. The insects were permitted to feed for a week. Apparent photosysthesis was reduced 29.1 percent, but there was no significant effect on transpiration (table 6 and fig. 3, *F*). Two additional feeding periods were accompanied by a reduction in the rate of photosynthesis of 31.1 percent and in transpiration of 33.6 percent. At this time the leaves exhibited the characteristic pale-green areas as the only external symptom of injury. The data for transpiration and photosynthesis were found to be significant and highly significant, respectively.

TABLE 6.—*The effects of potato leafhopper ((Empoasca fabae H.) injury on the rates of photosynthesis and transpiration of Stayman Winesap apple leaves, experiment F*

Date of leaf activity measurement [1]	CO_2 per cu. ft. of air	Photosynthesis (CO_2 per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}} \times 100$	Expected rate	H_2O per cu. ft. of air	Transpiration (H_2O per 100 cm.[2] per hour)		$\frac{\text{Test}}{\text{Check}} \times 100$	Expected rate [2]
		Check	Test				Check	Test		
	Milligrams	*Milligrams*	*Milligrams*		*Percent* [2]	*Grams*	*Grams*	*Grams*		*Percent*
July 29	13.5	16.1	20.1	124.8	-------	0.533	3.85	3.83	99.5	-------
July 30	15.5	19.0	23.3	122.6	-------	-------	-------	-------	-------	-------
Aug. 1	15.6	16.4	16.6	101.2	-------	.405	2.18	2.48	113.8	-------
Aug. 2	15.7	12.0	14.6	121.7	-------	.393	1.22	1.71	140.2	-------
Aug. 5	13.8	19.4	20.2	104.1	-------	.248	5.31	6.64	125.0	-------
Average	-------	-------	-------	114.9	-------	-------	-------	-------	119.6	-------
Aug. 14	15.9	13.0	9.6	73.8	64.2	.501	2.47	2.80	113.1	94.8
Aug. 15	14.8	13.9	11.1	79.9	69.5	.463	1.90	2.83	148.9	124.5
Aug. 18	15.8	15.0	13.6	90.7	78.9	.575	2.48	3.02	121.8	101.8
Average	-------	-------	-------	81.5	70.9	-------	-------	-------	128.0	107.0
Aug. 31	14.6	15.3	8.4	54.9	47.8	.335	3.60	3.22	89.4	74.7
Sept. 1	13.2	15.9	10.9	68.6	59.7	-------	-------	-------	-------	-------
Sept. 2	-------	-------	-------	-------	-------	.361	2.54	2.79	109.4	91.5
Sept. 3	16.2	15.9	12.5	78.6	68.3	.370	3.54	3.04	85.9	71.3
Average	-------	-------	-------	79.2	58.6	-------	-------	-------	78.4	87.4

ANALYSIS OF VARIANCE OF $\frac{\text{TEST}}{\text{CHECK}} \times 100$

Item	Degree of freedom	Sum of squares	Mean of squares	*F*	Remarks
Photosynthesis	1 12	4,856.1 1,331.0	4,856.1 110.9	43.8	Highly significant.
Transpiration	1 11	1,020.0 6,684.2	1,020.0 607.7	1.68	Not significant.

[1] Between each series of measurements leafhoppers were permitted to feed.
[2] See footnote 2, table 1, for method of calculation.

EFFECTS ON INTERNAL STRUCTURE OF APPLE LEAVES CAUSED BY THE FEEDING OF APPLE LEAFHOPPERS

Microscopic examination of moderately injured leaves revealed small groups of empty palisade cells, often with one or two uninjured cells in the center of each group. The cell walls appeared intact, although minute punctures through which cell contents were removed probably were present. Figure 5 shows cross sections of two apple leaves from which the cell contents of the upper palisade layer have been removed. Figure 5, *B*, shows an apple leafhopper killed, embedded, and sectioned in situ. The proboscis was lost in sectioning, but empty cells in the palisade layer show where the insect was feeding. During the progress

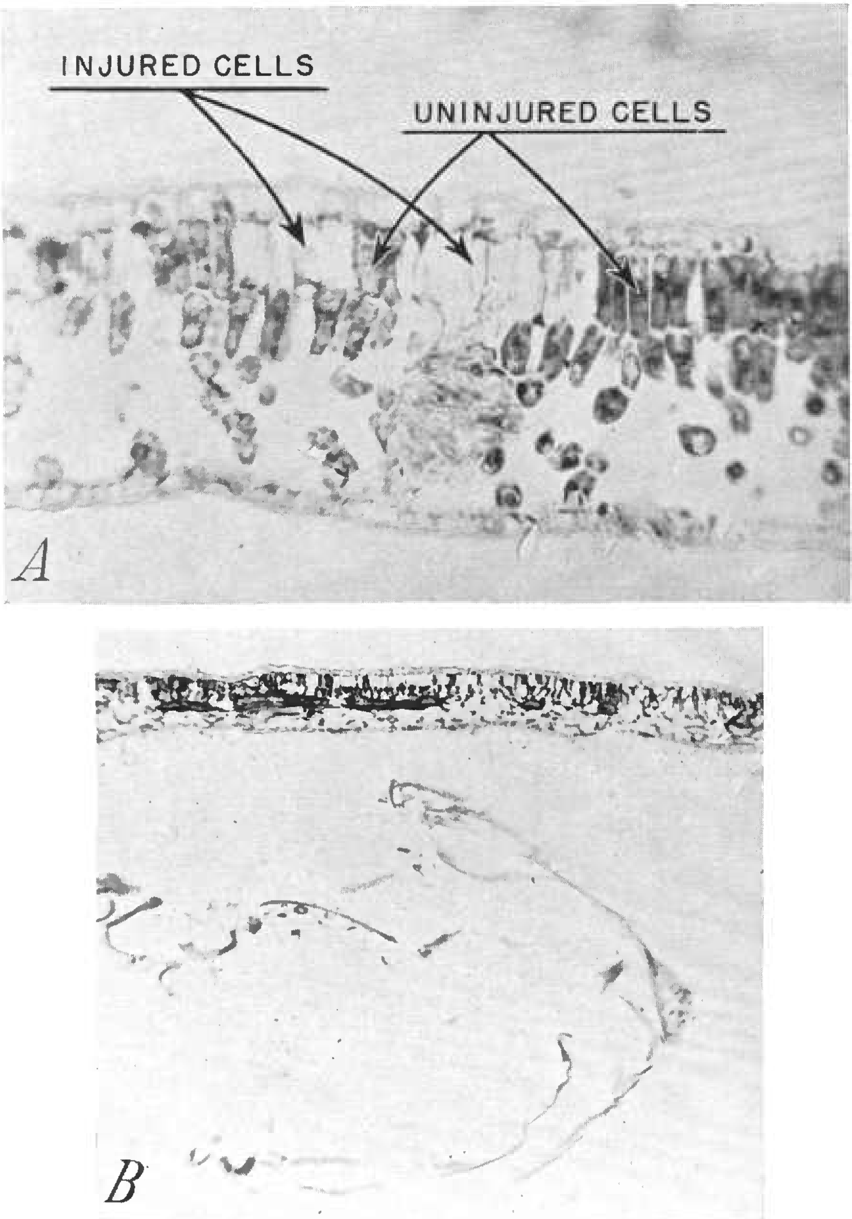

FIGURE 5.—Cross sections of Stayman Winesap apple leaves showing white apple leafhopper (*Typhlocyba pomaria*) feeding injury: *A*, Injured and uninjured cells of the palisade layer; *B*, cross section of leafhopper head in feeding position on under side of leaf, showing relative size of the insect and the cells upon which it has fed.

of these investigations a paper was published *(11)* in which somewhat similar results with apple leaves were reported. Smith and Poos *(12)* have pointed out that a small group of uninjured cells is often found in the center of a group of several injured ones where a leafhopper has been feeding.

Leafhopper injury caused a reduction in the thickness and fresh weight of apple leaves. Several hundred leaves were weighed and examined under the microscope in the field. Severely injured Black Twig leaves weighed approximately one-half as much (0.063 gm.) per unit area as uninjured leaves (0.123 gm.); they were from 0.097 to 0.120 mm. thick as compared with a thickness of 0.149 to 0.193 mm. in uninjured leaves.

DISCUSSION

In most instances considerable stippling was evident on the upper surfaces of the leafhopper-injured leaves before appreciable reductions in apparent photosynthesis and transpiration were recorded. A study of cross sections of injured leaves (fig. 5) shows that the spongy mesophyll tissue, in which probably a large part of the photosynthetic activity is carried on, is not affected until the palisade layers are markedly injured. When injured leaves are viewed from beneath by transmitted light, it is apparent that more light passes through the small injured spots than through the uninjured areas. It can be assumed, then, that that part of a leaf (spongy mesophyll cells) immediately under an injured area (palisade cells) receives more light after injury than it did before and conditions may be more favorable for photosynthesis in this region.

A given number of potato leafhoppers had a more detrimental effect on the metabolic activity than an equal number of leafhoppers of other species; this is in agreement with the data of Smith and Poos *(12)*. In almost every case the leafhopper injury, whether of the vein or mesophyll type, affected the rate of photosynthesis more than the rate of transpiration, and usually sooner. An examination of the injured leaves showed that the moderately injured areas were low in active chlorophyll, although in other respects they appeared normal, at least morphologically. Thus, while photosynthesis might be reduced in these areas because of relatively low chlorophyll content, the loss of water vapor could go on at a rate more nearly normal. Severe injury by the potato leafhopper (*Empoasca fabae*) or by apple or grape leafhoppers (*Typhlocyba* spp. and *Erythroneura* spp.) ultimately resulted in a reduction in transpiration, associated with a blocking of the vascular tissues in the case of *Empoasca* (*11, 12, 13*,) [6] and a breakdown of the mesophyll cells in the case of the other two (*5, 11*, and others).[6]

The decrease in leaf weight and thickness associated with leafhopper injury evidently results from the removal of the cell contents and the eventual drying and partial collapse of the injured cells. The total number of cells is probably not affected by leafhopper injury on a given leaf, as the leaf is a primary body; however, it is assumed that reduced vigor of the infested plant brought about by continued high populations of leafhoppers will eventually result in smaller leaves and other plant parts, especially the fruits.

[6] See footnote 4.

The question may arise as to which is more important to the welfare of the leaf—reduction in photosynthesis and transpiration caused by spray materials used to control the leafhopper, or injury from the insect itself. In practically all of the writers' experiments with nicotine sprays (data unpublished) there has been a reduction in photosynthesis and transpiration for a day or so after spray applications, and then recovery. However, the data on leafhopper injury presented here show that a leaf does not recover its original status in respect to apparent photosynthesis once it has been even moderately injured. This fact is of special importance where early season leafhopper injury occurs on leaves. Cell structure and the capacity of the leaf to function properly are adversely affected for the remainder of the season. This emphasizes the importance of early control of leafhoppers, both from the standpoint of reducing later populations of the insects and of obviating the permanent effects that they may have on leaf metabolism.

SUMMARY AND CONCLUSIONS

Injury to Stayman Winesap apple leaves caused by several species of apple leafhoppers (*Typhlocyba* spp.), grape leafhoppers (*Erythroneura* spp.), and the potato leafhopper (*Empoasca fabae*) was accompanied by a more or less marked reduction in apparent photosynthesis and transpiration. Photosynthesis usually was affected sooner and to a greater extent than transpiration.

A given number of potato leafhoppers had a more detrimental effect on leaf metabolism than an equal number of apple or grape leafhoppers.

Cross sections of injured leaves showed that the mesophyll-feeding types of leafhopper (*Typhlocyba* spp. and *Erythroneura* spp.) removed the contents of the cells in the palisade layers, while the spongy mesophyll cells were not significantly affected unless the leaf had been severely injured.

The data show that apparent photosynthesis and transpiration of apple leaves may be reduced early in the growing season when the leafhopper population is moderately low, and when this occurs the capacity of the injured leaves to function normally is permanently impaired. Early control of these insects is therefore important.

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