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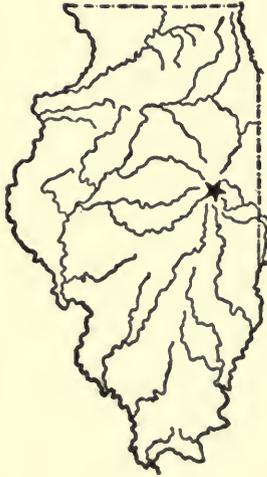
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BULLETIN No. 348

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EFFECT OF CERTAIN HYDROCARBON OILS ON  
RESPIRATION OF FOLIAGE AND DOR-  
MANT TWIGS OF THE APPLE

BY VICTOR W. KELLEY



URBANA, ILLINOIS, JUNE, 1930

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# EFFECT OF CERTAIN HYDROCARBON OILS ON RESPIRATION OF FOLIAGE AND DOR- MANT TWIGS OF THE APPLE<sup>1</sup>

BY VICTOR W. KELLEY, Associate in Pomology

Unsaturated (chemically active) oil emulsions have long been recognized as effective dormant insecticides, especially for the control of scale insects. But because of persistent reports of injury to the host plants from their application, their popularity has been somewhat cyclical. The last six or eight years have witnessed another revival of their use, due primarily to the failure of lime sulfur to give satisfactory control of San Jose scale under environmental conditions particularly favorable to its development. Even more recently the use of saturated (highly refined and chemically inert) oils as insecticides during the growing season has made the physiological reaction of deciduous fruits to oil sprays a subject of still greater interest.

Most of the experimental work with oils has been directed toward the perfecting of the sprays thru studies of the properties of the various oils, the methods of preparing the emulsions, and the insecticidal efficiency of the different oils. Very little research designed primarily to determine the physiological effect of oil sprays upon plants has been reported. It was to study this effect that this work was started in 1927. Respiration was chosen as the line of attack because the respiratory response of an organism is a good index of its reaction to environmental conditions.

## REVIEW OF LITERATURE

### Orchard Observations

In the horticultural and entomological literature various manifestations of abnormal behavior are attributed to oil sprays, some of which were thought to be beneficial and others which were considered harmful.

Ballard and Volck (1914), in their experiments on the control

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of apple powdery mildew in the Pajaro Valley, reported that the dormant application of an oil spray prepared from crude oil, lye, and fish-oil soap stimulated a vigorous early growth of foliage which was helpful in controlling the disease. According to Burroughs (1923), Essig stated that dormant applications of oil sprays hastened the blooming of deciduous trees in California from one to three weeks. Essig suggested that a darker color, which he seems to have observed, induced a higher temperature in the sprayed twigs, thus hastening blooming. Burroughs (1923) observed three to six days earlier blossoming of Montmorency cherries after an application of 20 percent Diamond Paraffine oil. According to deOng (1927), spraying with oils in the fall before complete dormancy, or in the spring after the buds began to open, caused a slight retardation in bloom, while applications in December and January usually induced a slightly earlier blooming. DeOng also reported that the practice of applying oil sprays for their effect on the tree and the changing of the blooming date was increasing rapidly among growers of both deciduous and citrus trees in California.

There is considerable further evidence that under some conditions oil sprays cause a retardation of growth. Yothers (1913) reported injury from the use of oil sprays on citrus trees, which manifested itself in a temporary yellowing of the foliage, stunting of leaf growth, dead twigs, and small, sour, late-maturing fruit. Pickering (1906) stated that dormant applications of oil sprays sometimes caused a marked retardation in bud opening. Burroughs (1923) reported observations on the killing of fruit buds and entire limbs following the dormant application of oil in high concentrations, and yellowing and abscission of foliage and even killing of portions of leaves by summer applications. He suggested that abscission might be due to a reduction in transpiration, and retardation in growth to an accumulation of carbon dioxide or a deficiency of oxygen. Later deOng, Knight, and Chamberlain (1927) observed yellowing and defoliation of citrus trees and stunting and killing of twigs and even larger limbs following the application of oils with a high boiling point which left a persistent oil film. Very young leaves were found to be more susceptible to injury from unrefined oils than mature, but not senile, leaves. Fully matured leaves, especially if senile, were more easily injured by saturated oils than younger ones which were still in the growing stage. Observed effects on the fruit of the saturated, so-called neutral, highly refined, white oils were the dropping of tree-ripe fruits and a marked delay in the coloring of fruit later subjected to ethylene gas treatment. Newcomer (1927) reported that the white oils caused less burning than oils less highly refined; however, the repeated applications of even white oils as ovicidal sprays

for codling moth frequently resulted in dwarfing, scalding, or spotting the fruit. The use of 60 percent unsulfonatable oils for dormant spraying and the more highly refined 95 percent unsulfonatable for summer applications was recommended by deOng. He also reported that even white oils applied in the summer may create functional disturbances, such as excessive vegetative growth and non-coloring of fruit. Woglum (1926) stated that applications of white oils on citrus trees at a concentration of  $1\frac{1}{2}$  percent produced, in many instances, undesirable effects on the trees. Among these he enumerated retarded coloration of fruit, reduced quality, lessened crop, and dropping of immature and tree-ripe fruits. Overlay and Spuler (1928) found that a 65 to 70 percent unsulfonatable oil applied on apples at a concentration of 4 percent during the dormant period from January 19 to March 15 had no harmful effect, but a 3.3 percent application on March 28, after the bud scales had separated, not only delayed the growth of flower and leaf buds but appeared to weaken them. A large number of fruit buds were killed outright and many leaf buds on one-year twigs failed to start growth. English (1928) reported that the lower the volatility and the higher the viscosity of an oil, the more likely it was to cause injury, whether it was saturated or unsaturated. When the residue persisted, which was necessary for insect control, the degree of unsaturation became the most important consideration in plant injury.

Published observations on injury by oil sprays reveal a lack of unanimity in regard to its occurrence and the factors involved in its production. Presumably oils do not always produce visible harmful effects upon the plants which they were designed to protect, but apparently one can never be sure that injury will not occur. Observers not infrequently associate injury with weather conditions. Felt (1913) recognized that certain kinds of weather promoted injury, altho he did not say what they were. He thought that favorable conditions, by producing vigorous, healthy plants, increased their resistance to penetration and injury by oils. On the other hand Yothers (1913) stated, "It is well known that neither hot sun nor shade, neither rain nor dry weather existing either during or following the application is influential in causing damage." The opinion is quite general that oil sprays should not be applied just previous to or during freezing weather; the reason advanced is that freezing may destroy the emulsion. However, it is more probable that a low temperature may be a contributing factor to oil injury because of its physiological effect upon the plant, such as the shriveling of twigs because of desiccation, or the change in the nature of the carbohydrates. As a means of lessening the danger of injury, deOng (1927) recommended that dormant sprays should not be applied when the trees are suffer-

ing from drouth due either to a dry soil or a dessicating wind, or when the temperature is as high as 75° F.

### Present Status of Oil Sprays

It is interesting to note the status of oil sprays in different parts of the country.

Regan (1926), in discussing "The Present Status of Oil Spraying in the Northwest" following two seasons of "disastrous" results in that section, said, "At present, oil sprays occupy a position of suspicion or total disfavor among the majority of the fruit growers of the Yakima and Wenatchee fruit districts of the Northwest." He attributed the poor results chiefly to the wholesale application of homemade emulsions prepared by inexperienced persons and to the use of dormant-strength sprays after the trees were in full foliage. However, deOng predicted an increasing use of oil sprays in California, based presumably on the use of more highly refined oils for both dormant and summer spraying.

Parrott (1927) was perhaps reflecting the eastern opinion when he stated that dormant and semidormant applications were regarded as safe, but advised the grower to "go slow" with foliage sprays.

Newcomer (1927) voiced the opinion that there was reason to believe that the use of oil during the summer would be greatly extended as further research was completed and the grower became more familiar with its limitations and possibilities.

### Experimental Work on Physiological Effect of Oil Sprays

Very little exact experimental work has been done to study oil-spray injury. Burroughs (1923) reported a preliminary test which seemed to show that apple leaves which had burned areas as a result of spraying with a 2-percent oil, generally had no starch in the portions which were still alive. Also, leaves which seemed to be stunted because of the oil spray contained no starch. Volck (1903) observed that injury to orange leaves was most pronounced when the spray was applied to the underside of the leaf, and he associated this injury with the presence of the stomata on the underside of the leaf. Kelley (1926) found that a number of oils, both saturated and unsaturated, markedly retarded the transpiration rate of the apple, pear, peach, plum, and sour cherry when the spray was applied to the lower surface of the leaf. In many cases the reduction was more than 50 percent, and in a few instances as high as 75 percent. Spraying the upper surface had little or no effect on the transpiration rate. The reduction of transpiration was greater with older than with younger leaves, more pronounced during the day than at night, and greater on clear than on cloudy days. This effect upon transpiration became evident soon after application, differences being noted within thirty minutes

after spraying. Little if any difference was noted in the effect between saturated and unsaturated oils. The stomata on leaves sprayed with oil emulsions on the undersurface did not open so widely as those on unsprayed foliage. Because of this fact and also because the greatest retardation occurred during the period when the stomata were normally open widest, this effect of the oil was thought to be associated primarily with stomatal activity. Since stomata open in response to the turgidity of the guard cells, it was concluded that the oil penetrating the leaf might have lowered the water content by reducing the supply of soluble carbohydrates. This hypothesis is supported by the work of Burroughs (1923), which seemed to show that photosynthesis is very much reduced in leaves injured by oil.

Magness and Diehl (1924) found that coating the surface of apples with oil or paraffin reduced the respiration rate at temperatures ranging from 32° F. to 80° F. Heavier oils caused a greater decrease than light oils. An excess of oil seemed to induce anaerobic respiration which injured the flavor. At all the temperatures there seemed to be an increase in the concentration of carbon dioxide within the tissues of the oil-coated fruits, and at the higher temperatures there appeared to be an actual limitation of the oxygen supply which retarded ripening and induced anaerobic respiration. Magness and Burroughs (1923) reported that Winesap apples dipped in a nondrying oil had a lower oxygen content as compared to control lots. Burroughs (1921-22) found that the respiration of ripe Wealthy, Baldwin, and Wagener was reduced as much as 50 percent after dipping in "crystal-ronite," but that the evolution of carbon dioxide was increased with immature fruits. He suggested that in the immature fruits anaerobic respiration must have occurred. Neller (1928), working with Winesap, found that untreated fruits respired about 40 percent faster than dry-brushed, oil-coated specimens after four months in storage, and 17 percent faster after an eight-months' storage period. At a temperature of 20° C. and a relative humidity of 30 percent, Delicious and Winesap untreated fruits lost weight more rapidly than oil-coated fruits. The moisture content of the oil-coated fruits was slightly higher.

Thus all experimenters who have attempted to measure the direct effect of oil sprays appear to agree that the application of oil affects the physiological behavior of the plant.

## METHODS OF EXPERIMENTATION

*Spray Materials.* The oils used in these experiments were supplied by the Standard Oil Company of Indiana. Data for Table 1 were furnished by Mr. H. J. Saladin, Assistant Manager of the Technical Division, Lubricating Department.

A saturated oil is more highly refined than the ordinary lubricating oil. It has been treated with concentrated sulfuric acid to remove the unsaturated hydrocarbons which the so-called unsaturated oils contain in addition to the saturated hydrocarbons. It is lighter in color than an unsaturated oil and is inert chemically. An unsaturated oil is not so highly refined and is chemically active. The reaction with sulfuric acid, termed " $H_2SO_4$  absorption," is an approximate measure of the amount of unsaturated hydrocarbons; the greater the reaction, the higher the percentage.

Diamond Paraffine is a light-colored, unsaturated lubricating oil. Formula 4389 is a darker lubricating oil which is slightly heavier than Diamond Paraffine and contains considerably more unsaturated hydrocarbons. Formula 3393 is a saturated oil which has approximately the same body or viscosity as Diamond Paraffine, but is much more highly refined. It is very nearly a medicinal oil. Perfection

TABLE 1.—TECHNICAL DESCRIPTION OF OILS USED IN EXPERIMENTS WITH OIL SPRAYS

Test	Formula 4389	Formula 3393	Diamond Paraffine	Perfection Kerosene	No. 9 Refined
$H_2SO_4$ absorption (%).....	27	0	6	3	0
Viscosity at 100° F.....	95 (max.)	95-100	100-105	....	....
Viscosity (Saybolt-Thermo) (seconds).....	.....	.....	.....	45	33
Specific gravity.....	.9071	.8576-.8550	.8899-.8844	.8299	.7972
Color.....	.....	Water-white	2½ to 3	22	25
Flash (° F.).....	305	365	350	120	140
Evaporation (%).....	.....	.....	.19	54.28	35.10
Acidity (mg. KOH) (grams)..	.....	.....	.023	.017	.015

Kerosene is a standard grade of kerosene. It is an unsaturated oil and has a much lower viscosity than Diamond Paraffine, Formula 4389, and Formula 3393. No. 9 Refined is a saturated oil approximately comparable to Perfection Kerosene in other characteristics.

Boiled emulsions were made according to the following formula:

Oil.....	500 cc.
Distilled water.....	125 cc.
Potash fish-oil soap (40%).....	115 grams

These materials were thoroly mixed, brought to a boiling point over a gas flame, and then pumped while hot, using one hundred strokes of a bucket pump equipped with a bordeaux nozzle. When only a part of the surface of the plant was to be covered, the diluted spray was applied with an atomizer. In all other cases the plants were dipped in the emulsion in order to insure a good coverage. The diluted sprays varied from 1.5- to 7-percent oil concentration. Tap water was used for diluting.

*Plant Materials.* For the dormant-period work, Jonathan, Grimes, and Delicious trees, twelve, eleven, and ten years old respectively,

were used. The trees were vigorous and had received uniform cultural, pruning, and spraying treatments since planting. The regular dormant applications of lime sulfur were not applied during the experiments. Twigs of the previous season's growth of moderate vigor were selected from the outside of the tree. Approximately the terminal 8 inches was used for the respiration determinations. The twigs were placed in water in small bottles in the respiration chambers; the basal ends were cut under water. During the first season's work all the twigs for any series of comparisons were selected from a single large tree, but in the second year, twigs were chosen from two to four smaller trees; the group after mixing together was considered as one lot. The same number of twigs, ranging from four to six, was placed in each respiration chamber. Each treatment was replicated three to four times in each series of comparisons. Rates of respiration were calculated on the basis of the volume of the twigs, which was determined by displacement of water.

For the foliage studies a uniform block of vigorous five-year-old Grimes was used. The trees were purposely left unsprayed during the season of the experiments. Terminal shoots of moderate vigor with uniform foliage characteristics, free from visible insect and disease injury, were selected. Shoots from two or three trees were used for a series, one shoot from each tree being placed in each respiration chamber. In about one-half the experiments three leaves from near the middle of each shoot were chosen and the terminal portion was then cut off, as was also the basal section, with the exception of enough of the stem to reach the water in the bottle. In the remainder of the experiments the leaves were selected nearer the distal end. The attempt was made in this way to select leaves uniform in age and size. Each treatment was usually repeated six times in each series. Respiration rates were calculated on the basis of leaf area; the measurements were made with a planimeter.

*Environmental Conditions.* The work with dormant twigs was done in a first-floor laboratory subject to the temperature fluctuations common to such places. In order to prevent photosynthesis, the respiration determinations with foliage were made in a dark basement room originally built for a photographic laboratory. In addition, the plant chambers were kept in a case, the front of which was covered with black oilcloth to exclude the dim light from an incandescent lamp, which was necessary for analytical work. A Wratten safety lamp provided with a screen which absorbed orange and red rays of wave lengths between 575 and 680, and a considerable portion of the light of shorter wave-length furnished sufficient light for the analytical work except for the reading of the burette. A small white light was turned on momentarily for making burette readings. Thermographic records were kept for most of the series and humidity

TABLE 2.—EFFECT OF OILS OF DIFFERENT VISCOSITIES AND DEGREES OF UNSATURATION ON RESPIRATION RATE OF APPLE TWIGS BEFORE THE OPENING OF THE BUD SCALES, IN AN ATMOSPHERE APPROXIMATELY SATURATED WITH MOISTURE

Series	Date started	Variety	Concentration of oil	Interval after dipping	Milligrams of CO <sub>2</sub> evolved per hour per cc. of plant material <sup>1</sup>						
					Light oils			Heavy oils			Water
					Perfection Kerosene (unsaturated)	No. 9 Refined (saturated)	4389 (unsaturated)	3393 (saturated)			
1.....	3-23-28	Jonathan	$\frac{7}{\%}$	hours 0-45 45-87 87-134 134-198	.1145 .1072 .0902 .1262	.1004 .0910 .0756 .1185	.1004 .0865 .0698 .1105	.0958 .1055 .1052 .1450	.0840 .0852 .0728 .0997		
2.....	3-27-28	Grimes	7	0-42 42-90	.0720 .0780	.0623 .0772	.0626 .0815	.0552 .0690	.0351 .0563		
3.....	4-3-28	Grimes	2	0-36 36-94	.1015 .1007	.0926 .0884	.0965 .0984	.0905 .0730	.0764 .0748		
4.....	4-3-28	Jonathan	2	0-34 34-86	.1505 .1048	.1440 .1147	.1325 .0950	.1492 .1070	.1055 .1037		
Average.....	.....	.....	.....	.....	.1045	.0965	.0934	.0995	.0793		

Mean difference  
-.0031  
+.0112

Odds  
2½ to 1  
1110 to 1

Viscosity comparison  
(1) Light saturated compared with heavy saturated.....  
(2) Light unsaturated compared with heavy unsaturated.....

Unsaturation comparison  
(1) Light saturated compared with light unsaturated.....  
(2) Heavy saturated compared with heavy unsaturated.....

<sup>1</sup>Average of 3 replications.

determinations at the time of spraying were made for practically all the foliage studies. The length of time required for the drying of the spray on the foliage was also recorded.

The respiration chambers necessarily subjected the plant material to abnormal atmospheric conditions. Calcium chlorid was used in many of the series to reduce the humidity. Determinations showed that the relative humidity in the jars containing calcium chlorid was 40 to 50 percent with dormant twigs and about 70 percent with foliage. Without calcium chlorid the air in the respiration chambers was approximately saturated with moisture, water being condensed on the sides of the jars most of the time.

*Respiration Apparatus.* The volumetric method, using the Haldane apparatus as modified by Newcomer (1921), was employed in the carbon dioxid determinations. A 17-percent solution of potassium hydroxid was the absorptive agent. One-half gallon Mason jars selected for uniformity in volume were used in all the dormant work and in part for the foliage studies. Jars of one-gallon capacity were employed for most of the work with foliage. Three-fourths-inch lengths of  $\frac{1}{4}$ -inch galvanized iron tubing soldered to the lids served as connections for sampling. Connections were made with the analyzing apparatus by 3-inch lengths of flexible rubber tubing provided with Hoffman screw clamps.

## EXPERIMENTAL RESULTS

As the experimental work includes the effect of oils on dormant twigs and also on foliage, it is convenient to present the data dealing with the two periods separately. In addition to the respiration studies observations were made on the subsequent growth of the dormant cuttings. This is presented in a separate division following the respiration data.

### Effect of Oils on Respiration of Dormant Apple Twigs

In spraying, the dormant season is divided into two periods: the "dormant" period, before the bud scales separate, and the "delayed-dormant" period, just after this stage but before the first leaves have fully unfolded. The respiration studies were made in the late winter and spring during both these periods, at the time when dormant and delayed-dormant sprays are applied in commercial orchards. Table 2 presents data showing the effect of oils on the respiration rate preceding the separation of the bud scales.

All the oils used, whether light or heavy, saturated or unsaturated, accelerated the respiration rate during the period immediately preceding the separation of the bud scales (Table 2). The respiration rate of the same twigs at successive intervals after a single spraying indicates that this initial acceleration was not maintained.

TABLE 3.—EFFECT OF SATURATED AND UNSATURATED OILS ON RESPIRATION RATE OF APPLE TWIGS AT HIGH AND LOW RELATIVE HUMIDITY, BEFORE THE BURSTING OF THE BUDS, GRIMES VARIETY  
(Concentration of oil, 2 percent)

Series	Date started	Interval after dipping	Milligrams of CO <sub>2</sub> per hour per cubic centimeter of plant material <sup>1</sup>					
			High relative humidity (100%)		Water		Low relative humidity (40-50%)	
			4389 (unsaturated)	3393 (saturated)	4389 (unsaturated)	Water	4389 (unsaturated)	3393 (saturated)
10. ....	3-12-29	hours 26-72 72-144	.1542 .1237	.1550 .1332	.1365 .1382	.1318 .1026	.1300 .1040	.1194 .1077
11. ....	3-19-29	0-42 42-67 67-138	.1058 .1600 .1492	. . . . .	.0965 .1415 .1432	.0988 .1400 .1316	. . . . .	.0842 .1215 .1168

<sup>1</sup>Average of 3 replications.

In some instances with the heavier oils the decline in acceleration reached the point where an actual retardation in rate occurred in comparison to the checks. In the single series which continued for a period of eight days, a second maximum increase was reached. The net result over the whole period was an accelerated respiration, as shown by the averages. Light oils as a group increased the respiration in the successive intervals with very little reduction in degree and with no retardation. The saturated light oil did not seem to maintain the accelerated rate so well as the unsaturated oil of similar viscosity.

*Effect of Viscosity of Oil.* Viscosity is an important factor in the efficiency of oils that are used as insecticides, but in these experiments on respiration it was not always important. A light unsaturated oil accelerated the respiration rate more than a heavy unsaturated oil; the odds were 1,110 to 1. On the other hand, with the saturated oils viscosity did not appear to be a factor since there was no significant difference between a light saturated oil and a heavy saturated oil in their effect upon respiration.

*Effect of Degree of Unsaturation.* Highly refined oils for spraying have received much attention during the past few years. The comparisons of the effects of saturated and unsaturated oils in Tables 2 and 3 indicate that unsaturation is not a factor which affects the respiration of dormant twigs with the heavier oils which are in general use for spraying. With oils of low viscosity (kerosene type) an unsaturated oil accelerated the rate of respiration more than a saturated one.

*Differences in Varietal Response to Oils.* Grimes, Jonathan, and Delicious, the varieties used in the dormant studies, varied considerably in the time of bud opening, and therefore were not in the same stage of development. Delicious bloomed earliest, followed by Jonathan and then by Grimes. Series 3 and 4 in Table 2 were run concurrently under identical environmental conditions; hence in these series Jonathan and Grimes may be compared directly. The data show that the Jonathan twigs respired considerably faster than the Grimes. In two preliminary tests three weeks earlier, when there was no perceptible difference in the activity of the buds, the respiration rate of Jonathan was higher than Grimes.

*Relative Humidity as a Factor.* In all the series shown in Table 2, the relative humidity in the respiration chambers was very high. In fact the atmosphere was saturated most of the time, as was evidenced by the condensation of water. Table 3 gives data for the comparison of the effect of oils at low and high humidity before the separation of the bud scales.

Without exception the respiration rate of the unsprayed twigs (Table 3) was less at low than at high relative humidity. With equal



consistency the sprayed twigs evolved more carbon dioxide per unit volume of plant material in a saturated atmosphere than in an atmosphere with a much lower relative humidity. There did not seem to be any consistent difference in the amount of acceleration produced by the oils at the two humidities. Series 10 and 11 both indicate an initial acceleration of the respiration rate followed by a reduction which in some cases becomes a retardation.

#### Effect of Oils on Respiration Rate at Delayed Dormancy

Experiments extending thru the late-dormant and delayed-dormant periods for three successive seasons indicate that at a definite time, which is closely associated with the separation of the bud scales, the effect of oil on the rate of respiration changes. Owing to variations in the time of opening of the buds on different twigs, the variation in rate among the individual twigs was evidently quite large, so it seemed advisable to ascertain the rate of respiration of each group of shoots, including the checks, preliminary to treatment, and to make the comparisons on the basis of the behavior before and after spraying.

Table 4 contains the data obtained in the comparisons of oils of different viscosities and degrees of unsaturation in their effect upon respiration during the delayed-dormant period of 1928. The most striking fact shown by these data is that all the oils retarded the rate of respiration, whereas during the period before the separation of the bud scales, these same oils had accelerated respiration.

*Effect of Viscosity.* Viscosity was a factor with both saturated and unsaturated oils; the heavier oils in both cases retarded respiration more than the light oils. This is in contrast to the results secured during the dormant period (Table 2), when viscosity was found to be important only with unsaturated oils. At that time a light unsaturated oil increased respiration more than a heavy saturated oil.

*Effect of Degree of Unsaturation.* The light saturated oil appeared to accelerate respiration very slightly at first, while the light unsaturated oil either caused no change or had a slightly retarding effect. With both of these oils, however, the initial effect was followed by a considerable retardation; the unsaturated oil checked respiration the more. As was found during the dormant period (Table 3), unsaturation was not a factor with the heavier oils ordinarily used in dormant spraying.

*Relative Humidity as a Factor.* In Series 8 and 9 (Table 4) the effect of a saturated and unsaturated heavy oil was compared at low and high humidity. These data indicate that the retardation was considerably greater at low humidity than at high. There does not seem to be any difference between the two oils in the amount of retardation. The respiration rate of unsprayed twigs is also higher

at low humidity. So far as evolution of carbon dioxide is concerned, these results are in contrast to the results secured at low and high humidity before the bursting of the buds, when the greatest acceleration in rate of both sprayed and unsprayed twigs occurred at high humidity. Since the relative humidity in the low-humidity chambers was 40 to 50 percent, this particular environmental factor was more nearly normal than that of the high-humidity chambers with the atmosphere saturated with moisture. It may therefore be concluded that high humidity in itself greatly reduced the respiration rate.

### **Effect of Oils on Respiration Rate of Foliage**

The experimental work with foliage was done during July and August of 1928 with the Grimes variety. Studies were made of the responses, at low and high humidity, of young leaves compared to old, of light versus heavy oils, of saturated oils compared with unsaturated, of the effect of spraying the upper or lower surface only, and of humidity alone as a factor in respiration. The data for nine series showing the effect of four oils upon the respiration of apple leaves from near the tip of the shoot at low and high relative humidity is shown in Table 5.

*Effect of Oils at Low and High Humidity.* Series 12 indicates that the respiration rate of leaves sprayed with a heavy saturated oil is less at low humidity than at high. Since the relative humidity in the "low-humidity" chambers was approximately 70 percent, this may be regarded as more nearly the normal atmosphere, and the greater respiration at high humidity may be considered an acceleration. Comparisons of Series 13 and 14 with 15 and 16 show that the heavy oils, both saturated and unsaturated, accelerated respiration at high humidity, but affected it very little, if any, at low humidity. On the other hand, the light oils (Series 17, 18, 19, and 20), both saturated and unsaturated, increased the respiration at both low and high humidity. No direct comparisons were made between light and heavy oils, but there is a suggestion that the light oils accelerated respiration more than the heavy oils. The effect of the oils was evident within four or five hours after dipping, which indicates that the effect began soon after application. It was also evident that the respiration rate of cut shoots kept in darkness became less as the time interval of separation from the plant increased. This would be expected since, in the absence of photosynthesis, the supply of respirable material is being gradually diminished.

Differences in type and degree of injury were striking. At high humidity there were "oil-soaked" translucent areas on the underside of the oil-sprayed leaves (Fig. 1). The injured areas were not located at any particular place on the leaf blade but appeared to be associated with the time required for the spray to dry. The

TABLE 5.—EFFECT OF OIL SPRAYS OF VARIOUS KINDS ON RESPIRATION OF APPLE FOLIAGE AT LOW AND HIGH HUMIDITY, GRIMES VARIETY (Low humidity, approximately 70 percent; high, 100 percent. Concentration of oil, 2 percent; emulsions applied by dipping. Two shoots of 3 leaves each selected from immediately below the youngest leaves at tip of shoot were placed in each respiration chamber; all shoots in a series from a single tree five years old.)

Series	Date started	Interval after dipping	Relative humidity in respiration chamber	Milligrams CO <sub>2</sub> per hour per square inch of leaf surface <sup>1</sup>						Percent of check	Observations <sup>2</sup> when removed from respiration chamber	
				4389 (heavy unsaturated)	3393 (heavy saturated)	Perfection Kerosene (light unsaturated)	No. 9 Refined (light saturated)	Water	Sprayed		Check	
				.....	.....	.....	.....	.....				
12.....	7-7-28	hours 18	High Low	..... .2096 .1692	..... .....	..... .....	..... .....	..... .....	..... .....	++ 0	- -	
13.....	7-10-28	0-5 5-17	High High	..... .....	..... .2088 .2000	..... .....	..... .....	.2700 .1768	107 113	++ ++	0	
14.....	7-11-28	0-5 5-22	High High	.2715 .1950	..... .....	..... .....	..... .....	.2312 .1638	117 119	-	-	
15.....	7-13-28	0-4 4-17	Low Low	..... .....	.2715 .1690	..... .....	..... .....	.2645 .1715	103 99	+	+	
16.....	7-13-28	0-5 5-17	Low Low	.2604 .1800	..... .....	..... .....	..... .....	.2590 .1810	101 99	+	+	
17.....	7-17-28	23	High	.....	.....	.2340	.....	.1832	128	++	0	
18.....	7-17-28	24	High	.....	.....	.....	.....	.1732	135	++	0	
19.....	7-26-28	17	Low	.....	.....	.....	.....	.1892	123	+	+	
20.....	7-26-28	17	Low	.....	.....	.....	.....	.1842	118	+	+	

<sup>1</sup>Average of 6 replications. 0 = no visible injury. + = slight marginal browning on lower surface. ++ = injured translucent areas on underside of leaf  
- = No record.

TABLE 6.—COMPARISON OF EMULSIONS MADE FROM SATURATED OILS AND FROM UNSATURATED OILS IN THEIR EFFECT ON THE RESPIRATION OF APPLE FOLIAGE, GRIMES VARIETY

(Low humidity, approximately 70 percent; high, 100 percent. Concentration of oil, 1.4 percent; emulsions applied by dipping; drying time of spray, about 30 minutes. Leaves were selected from near the center of the shoots.)

Series	Date started	Interval after dipping	Relative humidity in respiration chamber	Milligrams CO <sub>2</sub> per hour per square inch of leaf surface <sup>1</sup>						Observations <sup>2</sup> when removed from respiration chamber	
				4389 (heavy unsaturated)	3393 (heavy saturated)	Perfection Kerosene (light unsaturated)	No. 9 Refined (light saturated)	Water	Sprayed	Check	
25.....	8-9-28	18	Low	.1044 (98) <sup>3</sup>	.1074 (101)	.....	.....	.1066	0	0	
26.....	8-10-28	19	Low	.....	.....	.1404 (123)	.1200 (105)	.1142	0	0	
27.....	8-13-28	22	High	.1156 (112)	.1066 (103)	.....	.....	.1036	++ <sup>4</sup>	0	
28.....	8-14-28	22	High	.....	.....	.1154 (99)	.1192 (102)	.1164	++ <sup>4</sup>	0	

<sup>1</sup>Average of 6 replications. <sup>2</sup>0 = no visible injury. + = slight marginal browning on lower surface. ++ = injured, translucent areas on undersurface. <sup>3</sup>Ratio compared to check (percent). <sup>4</sup>Injury very slight.

TABLE 7.—COMPARISON OF EMULSIONS MADE FROM LIGHT OILS AND FROM HEAVY OILS IN THEIR EFFECT ON THE RESPIRATION OF FOLIAGE OF GRIMES VARIETY

(Humidity high, approximately 100 percent. Concentration of oil, 2 percent; emulsions applied by dipping; drying time of spray, 20 to 25 minutes. Leaves were selected from the middle of terminal shoots on which terminal buds had not formed.)

Series	Date started	Interval after dipping	Milligrams CO <sub>2</sub> evolved per hour per square inch of leaf surface <sup>1</sup>					Observations when removed from respiration chamber <sup>2</sup>	
			Light oils		Heavy oils		Water	Sprayed	Check
			Perfection Kerosene (unsaturated)	No. 9 Refined (saturated)	4380 (unsaturated)	3303 (saturated)			
30.....	8-21-28	hours 42	.0783 (91) <sup>3</sup>	.....	.0850 (96)	.....	.0857	++ <sup>4</sup>	0
32.....	8-23-28	25	.....	.1268 (108)	.....	.1342 (120)	.1170	++ <sup>4</sup>	0

<sup>1</sup>Average of 6 replications. <sup>2</sup>0 = no visible injury. + = slight marginal browning on lower surface. ++ = injury; translucent areas on undersurface. <sup>3</sup>Ratio compared to check (percent). <sup>4</sup>No injury with No. 9 or with Perfection Kerosene.

severity of the injury depended upon the age of the leaf and the length of the exposure to the saturated atmosphere. Exposures of a few hours produced translucent spots which, except in the case of very old leaves, gradually disappeared after the shoots were removed from the high humidity chamber. With longer exposures the translucent areas extended thru the mesophyll to the upper epidermis and became permanent; they later turned brown and eventually died.



FIG. 1.—OIL-INJURED AREAS ON LOWER SIDE OF LEAVES

The darker areas are the injured spots. They were not located at any particular place on the leaf blade, but appeared to be associated with the time required for the spray to dry. Such injury occurred only at high relative humidity.

At low humidity a different type of injury occurred. It consisted of a continuous narrow band of light-brown color which extended along the margin of the leaf on the underside. Such light-brown areas occurred both on oil-sprayed leaves and on checks which were sprayed with water. No injury occurred on the checks at high humidity with leaves of this age.

*Comparison of Saturated and Unsaturated Oils.* Young leaves were used in the respiration tests in the preceding Series 12 to 20 inclusive; they had been selected from immediately below the small leaves just back of the growing point. Beginning with Series 21, a change was made in the age of the leaves for the respiration tests. Three leaves were selected from the middle of the shoot instead of from near the tip. Terminal buds had not been formed on any of the shoots used. Saturated and unsaturated oils were compared in their effect upon the respiration rate (Table 6).

As was found with younger leaves, the heavy oils accelerated respiration only at high humidity, but the light oils did not seem to increase respiration at either low or high humidity so much as the work with the younger leaves indicated (Table 5). Neither was the injury at high humidity so great, only an occasional leaf being injured. In those instances where acceleration occurred, the unsaturated oils increased the respiration more.

As will be shown later, probably the greatest factor in the reduced effect of the oils was the difference in the age of the leaves used. There were other factors, however, which should be considered. The concentration of the oil was slightly less in the series reported in Table 6 than in those in Table 5. Also, Series 12 to 20 were sprayed in the respiration room in the basement, where the relative humidity was high and the spray dried very slowly, while the later series were sprayed in a room where the spray dried in 20 to 30 minutes. Then, too, the earlier series were run in July, while the others were run about one month later. The different weather conditions prevailing in August as compared with July presumably might bring about a change in leaf structure and contribute toward differences in the response of the leaves to oil sprays.

*Comparison of Light and Heavy Oils.* Series 30 and 32 (Table 7) provide a comparison of the effect of light and heavy oils at 2 percent concentration in an atmosphere practically saturated with water vapor.

Neither of the unsaturated oils increased respiration, and Perfection Kerosene retarded it. Both saturated oils increased the rate of respiration, the heavier oil accelerating it the more. Since in the work with dormant twigs it was found that the initial acceleration of respiration was followed by a retardation, it is possible that the longer interval of confinement in the respiration chamber may explain the retardation in Series 30. Wide differences occurred in the degree of injury. Nearly every leaf was injured by the heavy unsaturated oil, and a considerable number by the heavy saturated, while neither of the light oils had caused any visible injury at the time of removal from the respiration chamber although they had increased the respiration rate.

*Effect of Oil Spray on Upper and Lower Leaf Surfaces.* Since all the emulsions were ordinarily applied by dipping, which covered both surfaces of the leaf, Series 33 (Table 8) was planned to test the effect of an unsaturated heavy oil when applied to the upper and lower surfaces separately.

The results show that the oil increased the respiration rate when applied to either surface, but caused the greater acceleration when applied to the underside. Injury occurred only on those leaves sprayed on the undersurface.

TABLE 8.—EFFECT ON RESPIRATION OF A HEAVY UNSATURATED OIL APPLIED AT A HIGH RELATIVE HUMIDITY TO THE UPPER AND LOWER SURFACES OF FOLIAGE OF GRIMES VARIETY

(Concentration of oil, 2 percent; sprays applied with atomizer. Leaves were selected from near the middle of the shoots.)

Series	Date started	Interval after spraying	Milligrams CO <sub>2</sub> evolved per hour per square inch of leaf surface <sup>1</sup>					
			Upper surface sprayed	Lower surface sprayed	Check	Observations on removal from respiration chamber <sup>2</sup>		
						Upper	Lower	Check
33.....	8-24-28	hours 17	.1476 (108) <sup>3</sup>	.1550 (113)	.1368	0	++	0

<sup>1</sup>Average of 6 replications. <sup>2</sup>0 = no injury. + = slight marginal browning on lower surface. ++ = definite injury: translucent areas on undersurface. <sup>3</sup>Ratio compared to check (percent).

*Effect of Oil Sprays on Leaves of Different Ages.* A comparison of the effect of oil sprays on young and old leaves is supplied by the data from Series 22, 23, and 24 (Table 9). The young leaves consisted of the terminal portion of a growing shoot with the exception of the very small leaves near the growing point, which were removed. A portion of the shoot was then cut off and three leaves selected for the test with older leaves.

A comparison of the checks in the several series shows that the younger leaves of an elongating shoot respired very much more rapidly per unit area than the older ones farther removed from the growing point. This agrees with the work of Nicolas (1910), who compared the respiration rates of young and adult leaves of 20 species.

The respiratory response of leaves of different ages to oil sprays also was very different. Young leaves were affected more than older foliage. A heavy saturated oil retarded the respiration rate of young leaves but did not seem to have any significant effect upon the older leaves. The light oils, both saturated and unsaturated, accelerated the respiration of young leaves, but had little, if any, effect upon the older foliage on the same shoots. Differences were also observed among leaves of different ages in their resistance to injury by oil sprays. The young leaves were seriously injured by all the oils, while only heavy oil caused injury to the old leaves.

#### Extent to Which Humidity Alone Is a Factor in Respiration Rate

In the dormant-period studies (Table 3) it was found that the respiration rate of unsprayed twigs was higher in a saturated atmosphere than in an atmosphere with a relative humidity of 40 to 50 percent. Since humidity also seemed to be important in the effect of oils on the respiration of foliage, Series 29 and 31 were run to determine to what extent humidity alone was a factor in the respiration rate (Table 10).



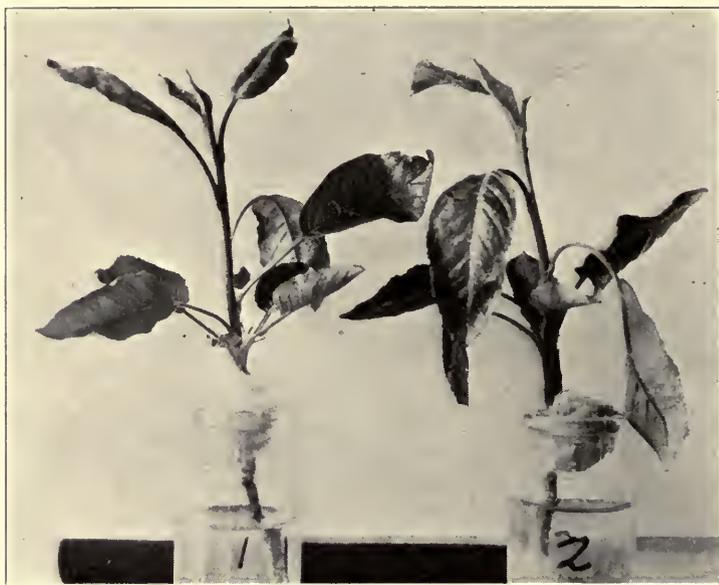


FIG. 2.—SERIOUS INJURY OF YOUNG UNSPRAYED LEAVES EXPOSED TO A SATURATED ATMOSPHERE

The length of the exposure was 3 hours. The relative humidity in 1 was 100 percent, and in 2, 70 percent. Older leaves were not visibly injured after an exposure of 19 hours.

The data from series 29 and 31 indicate that an atmosphere approximately saturated with moisture markedly increases the rate of respiration of unsprayed foliage. No visible injury was apparent on leaves of this age taken from the middle of growing shoots 12 to 16 inches in length at the conclusion of the 19-hour test. However, exposure of younger leaves to a saturated atmosphere for a much

TABLE 10.—HUMIDITY ALONE AS A FACTOR IN THE RESPIRATION OF APPLE FOLIAGE

(Leaves were selected from near the middle of the shoots; 2 shoots of 3 leaves each in each respiration chamber, 1 from each of 2 Grimes trees, five years old.)

Series	Date started	Length of test	Milligrams of CO <sub>2</sub> evolved per hour per square inch of leaf surface <sup>1</sup>			
			Low relative humidity (70%)	High relative humidity (100%)	Observations on removal from respiration chamber <sup>2</sup>	
					Low humidity	High humidity
29. . . . .	8-14-28	hours 19	.1116	.1320 (118%)	0	0
31. . . . .	8-22-28	19	.1208	.1640 (136%)	0	0

<sup>1</sup>Average of 6 replications. <sup>2</sup>0 = no injury.

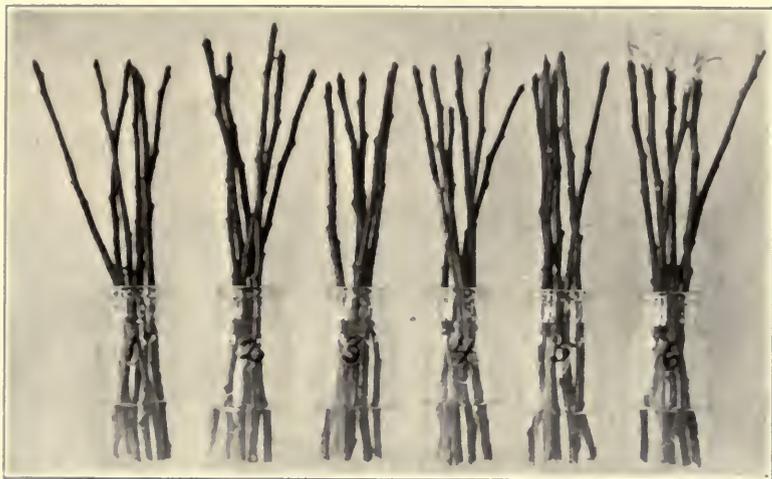


FIG. 3.—EFFECT OF FIVE OILS ON GROWTH OF GRIMES TWIGS AT HIGH RELATIVE HUMIDITY SPRAYED BEFORE THE SEPARATION OF THE BUD SCALES

All the oils accelerated the respiration of these twigs, but they retarded growth so much that none of the buds on the oil-sprayed twigs were open at the close of the growth observations about one month after they were sprayed. Photograph was taken 28 days after spraying. On 1, 4389 (heavy unsaturated) was used; on 2, 3393 (heavy saturated); on 3, Diamond Paraffine (heavy unsaturated); on 5, No. 9 Refined (light saturated); on 6, water. Compare with Figs. 4 and 5.

longer period caused serious injury. The injury at high humidity (100 percent) in comparison to low humidity (70 percent) after a three-day interval is illustrated in Fig. 2.

#### Effect of Oil Sprays on Growth of Twigs

In the analysis of the effect of oils on the respiration of dormant twigs it was stated that before the separation of the terminal bud scales the oils accelerated respiration, but after the bud scales opened, the effect of the oil was to retard it. At the conclusion of the respiration tests the twigs were placed in water in the laboratory for growth observations. Very striking differences were noted. These differences were associated with the development of the buds, the kind of oil from which the emulsion was made, and the relative humidity of the respiration chamber.

The effect of five oils upon the growth of Grimes twigs sprayed on March 27 before the separation of the bud scales is illustrated in Fig. 3. During the respiration tests the twigs were held at a high relative humidity. Photographs were taken on April 24. All the oils accelerated the respiration of these twigs, but they retarded

growth so much that none of the buds on the oil-sprayed twigs were open at the close of the growth observations about one month after they were sprayed.

Fig. 4 shows the effect of the same five oils upon the growth of the Jonathan twigs sprayed on April 3, which was about a week later than the application upon Grimes illustrated in Fig. 3. The photo-

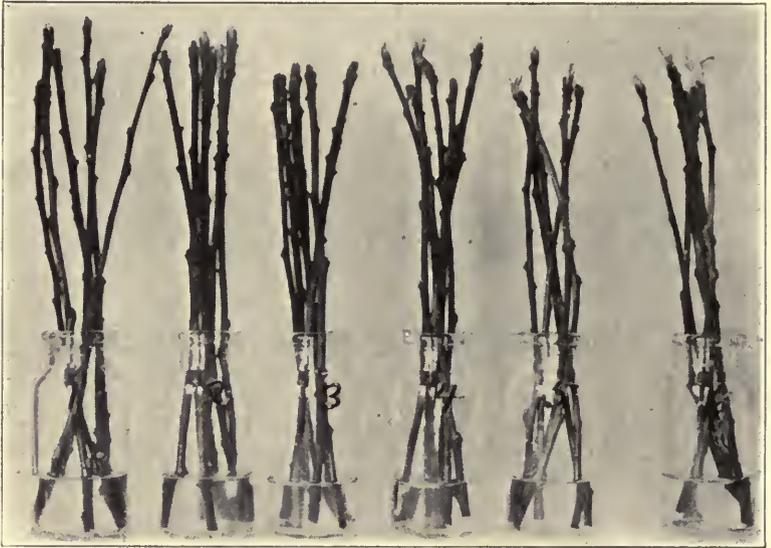


FIG. 4.—EFFECT OF THE FIVE OILS ON GROWTH OF JONATHAN TWIGS SPRAYED BEFORE THE SEPARATION OF THE BUD SCALES

The relative humidity was high during the respiration test. Photograph was taken about two weeks after spraying. On 1, 4389 (heavy unsaturated) was used; on 2, 3393 (heavy saturated); on 3, Diamond Paraffine (heavy unsaturated); on 4, Perfection Kerosene (light unsaturated); on 5, No. 9 Refined (light saturated); on 6, water. Compare with Figs. 3 and 5.

graph was taken on April 16. Jonathan was considerably earlier in bud opening in 1928 than Grimes. The oils greatly retarded bud opening, but not to so great an extent as when applied to Grimes one week earlier. The two light oils did not check growth so much as the heavier oils. There seemed to be no difference between the effect of saturated and unsaturated oils. When the growth observations were terminated two weeks later, the oil-sprayed twigs had not developed any more than they show in the photograph.

The buds of Delicious were much more advanced than those of Grimes and Jonathan. Fig. 5 shows the effect of two heavy oils, one saturated and the other unsaturated, upon the growth of Delicious twigs which were sprayed on April 3, the same day as the Jonathan

in Fig. 3. The photograph was taken on April 16. The retardation in growth was less with Delicious than with Jonathan. The unsaturated oil checked growth more than the saturated one. The effect of the oils in this test was to retard respiration.

The effect of the oils applied April 11 on the growth of Jonathan twigs is shown in Fig. 6. The photograph was taken on April 30.



FIG. 5.—EFFECT OF TWO HEAVY OILS ON GROWTH OF DELICIOUS TWIGS SPRAYED AFTER THE SEPARATION OF THE BUD SCALES

The unsaturated oil checked growth more than the saturated oil; the relative humidity was high during the respiration test. On 1, 4, and 6, 4389 (heavy unsaturated) was used; on 2, 5, and 7, 3393 (heavy saturated); on 3, water. Compare with Figs. 3 and 4.

There is a striking contrast between the effect of this application and the one a week earlier on the same variety shown in Fig. 4. The three heavy oils checked growth nearly as much as they did a week earlier, but the terminal buds on the twigs sprayed with the light oils grew as well as the checks. The lateral buds on the checks developed more, however.

Fig. 7 illustrates the effect on the growth of Grimes twigs of an application of oil sprays made April 17, about three weeks later than the one shown in Fig. 3. The twigs were held at high humidity during the respiration test. The photograph was taken on May 1. The oils checked growth much less than the previous application. The light oils retarded growth less than the heavier ones.

In the section dealing with the effect of oils upon the respiration of apple twigs, it was shown that the relative humidity of the at-

mosphere in the respiration chamber was an important factor. Figs. 3 to 7 inclusive show the effect upon growth of the application of oil sprays at a high relative humidity during the dormant and delayed-dormant periods. Fig. 8 illustrates the effect of a light and heavy unsaturated oil at low humidity before the separation of the bud scales. The photograph was taken three weeks after spraying. With



FIG. 6.—EFFECT OF OILS APPLIED TO JONATHÁN TWIGS AFTER THE SEPARATION OF THE BUD SCALES

There is a striking contrast between the effect of this application and the one made a week earlier on the same variety, shown in Fig. 4. All the twigs were held at high humidity during the respiration test. The three heavy oils checked growth almost as much as they did a week earlier, but the terminal buds on the twigs sprayed with the light oils grew as well as the checks. On 1, 4389 (heavy unsaturated) was used; on 2, 3393 (heavy saturated); on 3, Diamond Paraffine (heavy unsaturated); on 4, Perfection Kerosene (light unsaturated); on 5, No. 9 Refined (light saturated); on 6, water.

Figs. 9, 10, and 11 it is possible to compare the effect of a saturated and an unsaturated heavy oil with the effect of water upon the growth of Grimes twigs at low and high humidity. The twigs were sprayed on April 20, when the terminal bud scales were separating, and when the effect of the oils was to retard respiration. The photographs were taken April 24, at time of removal from the respiration chambers.

Fig. 9 shows that a heavy unsaturated oil checked growth more at high humidity than at low. Fig. 10 illustrates the same effect with a heavy saturated oil. There was no difference between these oils in



FIG. 7.—EFFECT OF THE FIVE OILS APPLIED TO GRIMES TWIGS THREE WEEKS LATER THAN TO TWIGS IN FIG. 3

The oils checked growth much less than the previous application, and the light oils retarded growth less than the heavier ones. On 1, 4389 (heavy unsaturated) was used; on 2, 3393 (heavy saturated); on 3, Diamond Paraffine (heavy unsaturated); on 4, Perfection Kerosene (light unsaturated); on 5, No. 9 Refined (light saturated); on 6, water.

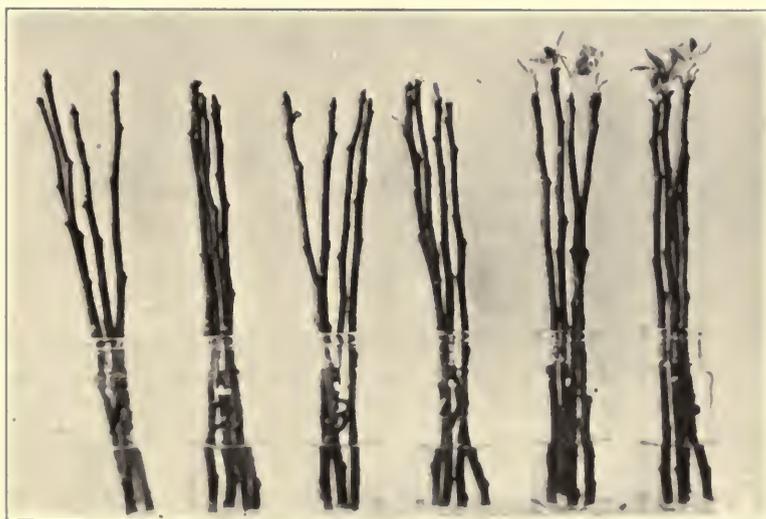


FIG. 8.—EFFECT OF LIGHT AND HEAVY UNSATURATED OIL AT LOW HUMIDITY BEFORE SEPARATION OF BUD SCALES

The relative humidity was 40 to 50 percent. Photograph was taken three weeks after spraying. On 1 and 2, 4389 (heavy unsaturated) was used; on 3 and 4, Perfection Kerosene (light unsaturated); on 5 and 6, water.

their effect upon growth at either low or high humidity. Fig. 11 illustrates the effect of different humidities on the growth of twigs

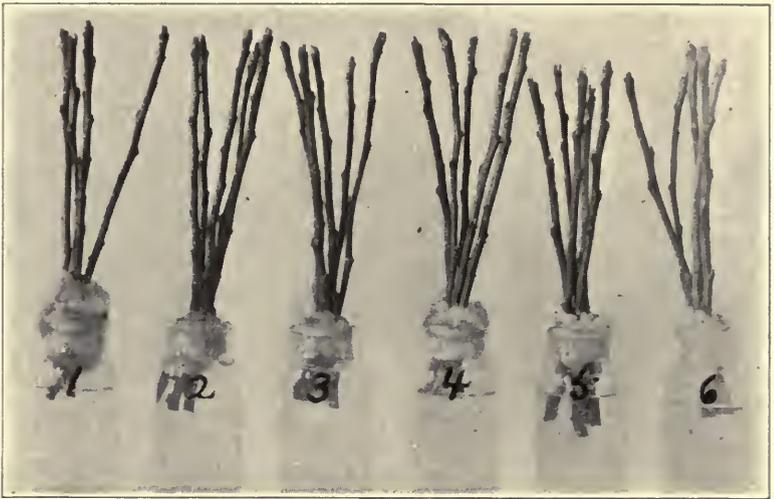


FIG. 9.—EFFECT OF A HEAVY UNSATURATED OIL ON GROWTH OF GRIMES TWIGS AT LOW AND HIGH HUMIDITY SPRAYED AFTER THE SEPARATION OF THE BUD SCALES

Growth was checked more at high humidity than at low. Photograph was taken 4 days after spraying. To 1, 2, and 3, 4389 (heavy unsaturated) was applied at high humidity; to 4, 5, and 6, 4389 (heavy unsaturated) was applied at low humidity.

sprayed with water. Those twigs which were held in the respiration chamber for 46 hours at a relative humidity of 40 to 50 percent made a greater growth than those held in a saturated atmosphere for the same length of time.

## DISCUSSION

### Penetration of Oil Into Plant Tissues

In these experiments the effect of oil sprays on the respiration of apple foliage and twigs, on the growth of twigs, and on leaf injury indicate penetration of the oil into the plant tissues. A number of investigators, including Volck (1903), have thought that oils penetrate plant tissue, but direct proof was lacking until Knight (1928), after perfecting a suitable technic, observed oil which had penetrated citrus leaves and the vascular system into the stem. According to Knight, the oil was finally deposited in storage cells in the trunk.

That oils penetrate the leaf was determined in these experiments by microscopic examination. Free-hand sections of dry, sprayed leaves, thoroly washed with soap and water to remove any emulsion

from the surface, revealed an abundance of oil emulsion in the intercellular spaces. Penetration of the emulsion, as such, must have

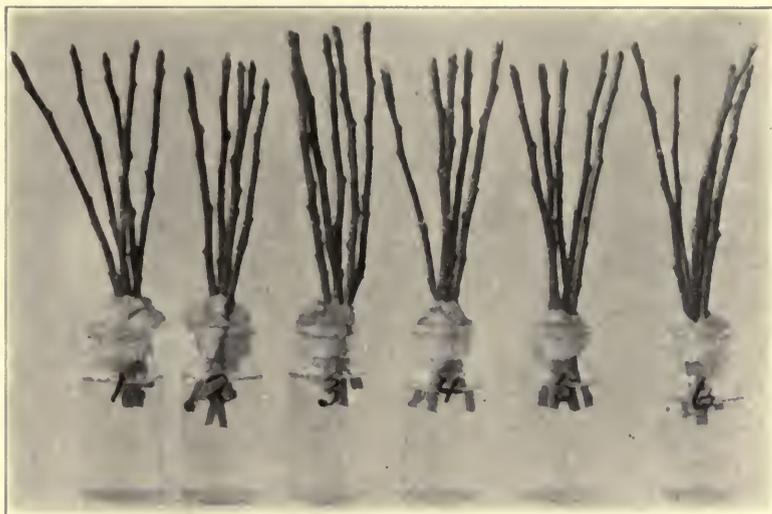


FIG. 10.—HEAVY SATURATED OIL PRODUCED SAME EFFECT AS HEAVY UNSATURATED OIL SHOWN IN FIG. 9

Spray was applied after the separation of the bud scales. There was no difference between these oils in their effect upon growth at either low or high humidity. Photograph was taken 4 days after spraying. To 1, 2, and 3, 3393 (heavy saturated) was applied at high humidity; to 4, 5, and 6, 3393 (heavy saturated) at low humidity.

taken place. That penetration of the spray as oil after the emulsion had broken, and recombination in the leaf after penetration could not have occurred, was evident. Very little water was required to keep the emulsions used from breaking, for emulsified oil was found in water washed from recently sprayed leaves that appeared to be dry. That the emulsion left on the surface broke eventually, however, was determined by the examination of water washed from leaves which had dried for a longer period.

Additional evidence that penetration is rapid is shown by the fact that the physiological effects of the oils are evident soon after application. Kelley (1926) found that oils applied to the underside of apple leaves retarded transpiration to a measurable degree within thirty minutes. In these experiments an effect upon the respiration of foliage was evident four hours after application. Young leaves dipped in oil emulsion and held at high humidity, where the water did not evaporate from the pubescent underside for three hours, showed injured areas before the leaves were dry.

It is evident that oil as an emulsion could not enter the leaf except thru an opening in the epidermis. There is considerable evi-

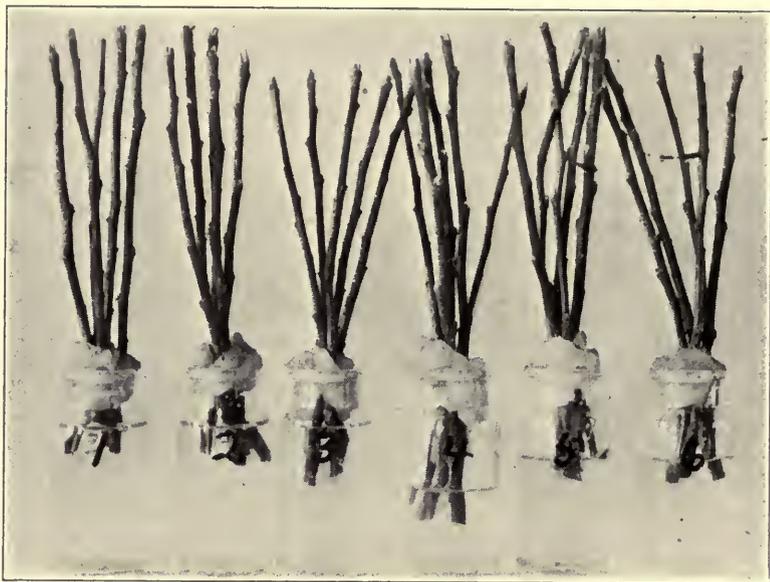


FIG. 11.—EFFECT OF DIFFERENT HUMIDITIES ON GROWTH OF TWIGS SPRAYED WITH WATER

Twigs which were held in the respiration chamber for 46 hours at a relative humidity of 40 to 50 percent made a greater growth than those held in a saturated atmosphere for the same length of time. To 1, 2, and 3 water was applied at high humidity; to 4, 5, and 6 water was applied at low humidity.

dence in these experiments that oil does not penetrate the leaf except thru the lower epidermis, where the stomata of the apple are located. In numerous microscopic examinations no emulsion was found inside the leaf when the spray had been applied to the upper surface only. Likewise, no oil injury occurred on leaves held at high humidity after spraying only the upper surface, while at the same humidity severe injury was produced when the spray was applied to the lower surface. Also, examination of a large number of leaves, so severely injured by dipping in oil emulsion that injured areas occurred on the upper as well as on the lower side, showed that the oil had entered below and extended thru the mesophyll to the upper surface. Every injury on the upper surface had a corresponding injured area on the lower epidermis. If the leaf is penetrated by oil spray applied to the upper side, it must enter as oil thru the cuticle. That such penetration may occur is indicated by the fact that a heavy oil ap-

plied to the upper surface accelerated respiration. However, it is also possible that a film of oil might cause a change in respiration without actually entering the leaf.

### Effect of Oils on Respiration and Growth During Dormant and Delayed-Dormant Periods

The effect of spraying with an oil emulsion depends largely upon the developmental stage of the plant or plant part when the application is made. Respiration was accelerated before the separation of the bud scales, but was retarded during the delayed-dormant stage; when sprayed during the dormant period, the buds failed to grow, but when sprayed during delayed dormancy, growth was only retarded.

The results on growth agree with the observations of Felt (1913), who reported that commercial applications of oil emulsion should not be made until shortly before growth started, as earlier applications seemed to cause greater injury. He regarded fall applications as particularly hazardous. Also, deOng (1927) observed that oil applied in the fall before complete dormancy, or in the spring after the buds were opening, retarded bloom; but that applications in December and January induced earlier blooming. It is possible that the earlier growth attributed to oil applied at certain periods by Felt and deOng as well as by Ballard and Volek (1914) may have been induced by an effect of the oil in breaking the rest period as suggested by Dutton (1924). In view of this hypothesis the greater oil injury observed by Felt and deOng, following an early-dormant application, may have been winter injury due to the breaking of the rest period by the oil.

A number of factors may contribute toward an understanding of the opposite results secured during the dormant and delayed-dormant seasons. These experiments indicate that the effect of oil upon respiration and growth is due primarily to penetration. Richards (1896) and others have reported that wounding increased respiration. In view of this, the increased respiration during the dormant period may indicate a greater injury than the retarded respiration at delayed dormancy. This hypothesis is further strengthened by the fact that oil stopped growth during the period when respiration was increased, but only retarded it during the delayed-dormant period, when the rate of respiration was decreased. It is probable that the slowly developing growing point of the unopened bud is especially susceptible to oil injury, and that the more rapidly growing leaves of the opening bud, because of the formation of new tissue, may not be so severely injured. Also, the closely laid bud scales so effectually check evaporation that oil emulsion, which has penetrated the unopened bud, may likely remain there for a considerable period, while evaporation may be rapid from the interior of the opening bud.

During the dormant period viscosity and unsaturation did not appear to be factors in the effect upon growth, for all the oils almost completely stopped the growth of the cuttings even at low humidity. At delayed dormancy, however, when growth was only retarded, the greater retardation was produced by the heavier oils. There seemed to be little or no difference between saturated and unsaturated oils in their effect upon growth, but differences were found in their effect upon respiration. During the dormant period, light unsaturated oils accelerated respiration more than heavy unsaturated oils. There was no difference, however, between light and heavy saturated oils. At delayed dormancy, the heavier oils retarded respiration more than the light oils. There was no difference between heavy saturated and unsaturated oils, but the light unsaturated retarded respiration more than the light saturated.

That there should be no difference between saturated and unsaturated heavy oils in their effect upon respiration or growth at either period is significant, in view of the importance placed, in recent years, upon the chemical nature of spraying oils. Theoretically, saturated oils, because they are less active chemically, should produce less injury. This theory suggests that the effect of the heavy oils is more physical than chemical. That a light unsaturated oil affected respiration more than a light saturated one during both periods may indicate a greater chemical effect with light oils, but it can also be explained on a physical basis. According to English (1928), unsaturated emulsions are more stable than saturated. The greater effect of unsaturated oils may be due to their longer persistence in the emulsified condition, in which case their removal by evaporation would be delayed. However, if oil penetrates the cell, the quicker breaking saturated emulsion should penetrate first. Difference in volatility was not always an important factor since there was no difference between light and heavy saturated oils in their effect upon respiration and growth before the separation of the bud scales. During delayed dormancy, however, when penetrated oil could more easily evaporate from the opening bud, oils of low viscosity affected respiration and growth less than heavier oils.

#### **Effect of Oils on Respiration of Foliage and Visible Leaf Injury**

Age of leaf, relative humidity, the surface (upper or lower) covered by the spray, and viscosity were the important factors in visible injury and respiration changes. The degree of unsaturation was not uniformly important. Neither was the effect of any oil on the foliage so constant nor so great as was found during the dormant and delayed-dormant seasons on twigs. The most frequent and also the most pronounced changes in rate, however, were in the direction of acceleration.

Younger leaves were more easily injured and their respiration rate was affected to a greater degree than older leaves. This observation agrees with those of deOng, Knight, and Chamberlain (1927). A number of factors may contribute toward these differences. More spray is retained by the young leaf because of its pubescent surfaces, and a much longer time is required for the leaf to dry. Since oil probably continues to enter as an emulsion until the surface of the leaf dries, a greater penetration occurs.

The viscosity of the oils when applied to foliage was under some conditions very important; it had more significance in oil injury than in respiration. When applied to young leaves, light oils apparently resulted in as much injury as the heavier oils, but on older leaves the light oils produced less injury. Viscosity did not always seem to be a factor in the effect upon respiration. Heavy oils are less volatile, therefore their persistency on the outside and inside of the leaf is greater than that of lighter oils. Greater persistency would probably mean increased injury, especially if penetration as oil occurs either thru the epidermis or cell wall.

#### Relative Humidity as a Factor in Respiration and Oil Injury

The most important single factor to be considered in avoiding injury from oil sprays appears from these experiments to be the relative humidity during and following the application of the spray. The greatest injury to twigs and leaves always occurred at high humidity; in fact, visible oil injury to foliage was not produced except at high humidity. In some instances the saturated atmosphere of the respiration chamber checked the growth of unsprayed twigs almost as much as oil. Relative humidity was also important in the respiration tests. An atmosphere approximately saturated with water vapor always caused a change in respiration in comparison with an atmosphere with a relative humidity of 40 to 50 percent for twigs and approximately 70 percent for foliage.

Few writers have associated high relative humidity with plant injury, but Ravaz (1927) observed that the common injury to grape foliage, which he describes as "scorching," was associated with fogs following drouth. A browning of young apple leaves similar to the injury observed on unsprayed leaves in the high-humidity respiration chambers has occurred in various sections of Illinois during the rainy season of April, 1929.

The reason for the effect of a saturated atmosphere upon respiration and plant injury was not determined experimentally. That it was not due to the excess of carbon dioxid in the respiration chamber was shown by the fact that removing the carbon dioxid as it was formed with soda-lime did not reduce the injury, and increasing the

carbon dioxid content to 10 percent did not increase the injury at high nor induce it at low humidity. It is probable, however, that exposure to a saturated atmosphere will cause a significant water-logging of the tissues. It is thought that greater oil injury occurred at high humidity not only because drying was retarded but because such an atmosphere actually facilitated the entrance of oil emulsion thru the stomata and lenticels. This view is in contrast to the opinions of Volck (1903), who thought that a dry atmosphere facilitated oil penetration, and Yothers (1913), who said that weather conditions had nothing to do with oil injury. It is certain that under conditions of high humidity, with the stable emulsions used in these experiments, oil remained on the surface longer as an emulsion, and that at least a part of the emulsion persisted in the intercellular spaces so long as the high humidity prevailed. Microscopic examination of sprayed leaves held at a humidity of 40 percent for several hours failed to reveal any oil emulsion in the intercellular spaces, while at high humidity the emulsion was found inside the leaf three days after application. These facts support the view expressed above that a saturated atmosphere produces a significant water-logging of the tissues.

### SUMMARY AND CONCLUSIONS

1. Oils of different viscosities and degrees of unsaturation were tested in their effect upon the respiration of dormant twigs and foliage of the apple.

2. The effect of oil emulsions on dormant twigs depended largely upon the developmental stage of the buds when the applications were made.

3. Before the separation of the bud scales, termed in spraying the "dormant season," all the oils accelerated respiration; when applied immediately after this stage but before the first leaves had unfolded, termed the "delayed-dormant season," they retarded respiration.

4. The buds of the cuttings, treated with any of the oils during the dormant season, failed to grow; treated at delayed dormancy, growth was only retarded; the light oils, however, had very little effect.

5. Viscosity was important in respiration, during the dormant period, only with unsaturated oils; at delayed dormancy it was a factor with both saturated and unsaturated oils. On foliage, the heavier oils affected respiration only at high humidity; the greater effect of the light oils was produced at this humidity.

6. The use of highly refined, saturated oils is not a guarantee against injury by either dormant or foliage sprays. Saturation of the heavier oils, comparable to those used in commercial spraying,

was not important in either the dormant or delayed-dormant periods. It was relatively unimportant in foliage applications.

7. Younger unsprayed leaves respired more rapidly than older leaves; their respiration was more affected, and they were more easily injured by oil sprays.

8. The relative humidity during and following the application of oil sprays is the most important single factor to be considered in avoiding injury. The greatest injury to twigs was always produced at high humidity; visible oil injury to foliage was produced only at high humidity. In some cases the saturated atmosphere of the respiration chamber checked the growth of unsprayed twigs almost as much as the oil. A saturated atmosphere is thought to produce a significant water-logging of the tissues.

9. Oil injury was produced on foliage only when the spray was applied to the lower surface of the leaves at a high relative humidity. It occurred as translucent areas on the lower sides of the leaves three to four hours after spraying. The severity of the injury depended upon age of leaf and length of exposure to high humidity. When the leaves were removed after short exposure, the translucent areas gradually disappeared; under longer exposure, they extended thru the mesophyll to the upper epidermis and caused the tissue to turn brown.

10. Microscopic examination showed that oils penetrated the leaf as an emulsion. The emulsion apparently starts to enter thru the stomata soon after application. High humidity is thought to facilitate the entrance of the emulsion.

11. The practice of spraying with oil for its physiological effect upon the tree, in view of these experiments, should not be encouraged.

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