SF 538 .5 P65 P94



PURDUE UNIVERSITY

Agricultural Experiment Station

BULLETIN No. 247 JULY, 1920



BEES AND THEIR RELATION TO ARSENICAL SPRAYS AT BLOSSOMING TIME

Published by the Station:
LAFAYETTE, INDIANA
/ U.S.A.



The original of this book is in the Cornell University Library.

There are no known copyright restrictions in the United States on the use of the text.

BEES AND THEIR RELATION TO ARSENICAL SPRAYS AT BLOSSOMING TIME

W. A. Price

INTRODUCTION

Fruit-growers who apply lead arsenate or other arsenical sprays to trees in full bloom have long been accused by beekeepers of causing a tremendous death rate of bees from poisoning. Beekeepers and entomologists have noticed the large number of dead bees around sprayed trees and about the hive. Even when they made allowance for death from old age, disease, and other causes, the mortality was abnormally high. growers have not been willing to assume responsibility for this excessive death rate, and up to the present time very little in the way of exact data¹ has been submitted to support either side of the question. The experiments recorded here are an attempt to contribute definite evidence on the subject.

The foregoing discussion may bring at least two questions to the mind of the reader. Why is a spray applied when apple trees are in full bloom? Could it not be applied later with just as good results? Pathologists tell us that spraying is necessary to control scab just before the blossoms open and again after the petals fall. The effective period for applying the first spray is only three days in extent, and for the second spray about five days.

An arsenical is also applied in the first spray to control curculio, and in the second spray for codling moth. The spray for codling moth must be applied before the calyx lobes close, which is from a week to ten days after the blossoms fall. If growers could only adhere strictly to the welldefined time limits mentioned for each of these sprays, the discussion regarding bee-poisoning would have to end, for it is a well known fact that bees do not visit trees to any large extent before the blossoms open, or after the petals have fallen. But fruit-growers have a practical difficulty to meet in this connection. They must buy enough high-priced spraying equipment to spray large acreages within the short spaces of time previously mentioned. None of the other sprays during the season is so limited in the effective time for application.

When rainy weather or lack of capital for equipment forces the orchardist to begin spraying while trees are in bloom, the beekeeper naturally becomes concerned about having his bees feed upon poisoned blossoms. And if we assume that such feeding causes the wholesale poisoning of bees, the orchard owner not only causes serious loss to a neighboring beekeeper but he kills the only agent which is effective in fertilizing his fruit blossoms and thus increasing the set and yield of fruit.

During the course of a clear, warm day in blossoming season a bee usually makes several trips to the blossoms to gather nectar. If spray material has previously been applied to the blossoms, the assumption is that because of the feeding habit of the bee, it can scarcely avoid gathering poison with the nectar.

¹ Detection of Arsenic in Bees, by E. B. Holland, Journal of Economic Entomology, Spraying with Arsenites vs. Bees, by F. M. Webster, Bul. No. 68, Ohio Agricultural Experiment Station.

It was the object of this experiment to determine definitely whether bees absorbed poison into their systems in this manner, and if so, whether such doses were sufficiently large to be fatal.

The work was carried on in cooperation with the Department of Horticulture of Purdue University, and the analysis and feeding phase of the work was generally supervised by Mr. H. A. Noyes, Research Chemist, then of that department.

FIELD OPERATIONS

Apple trees were caged with screen and cheese-cloth (See Fig. 1). When the trees were in full bloom lime-sulphur testing one degree Baumé

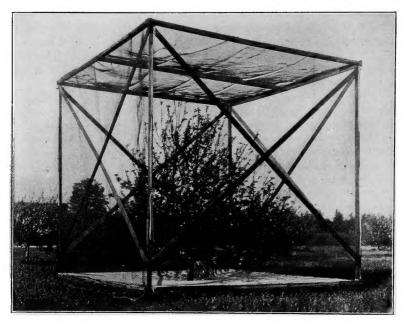


Fig. 1. Tree surrounded by a cage having window screen on the top and sides and cheese-cloth on the bottom. A colony of bees is shown near the trunk of the tree.

and one pound powdered arsenate of lead to 50 gallons of water was applied. Bee colonies were then moved into the cages and observations made on them. Also trees in full bloom in the open were sprayed, while others were left unsprayed. Observations were made on both. Dead bees were gathered from the cages, counted, and analyzed for arsenic.

LABORATORY WORK

These experiments naturally divided themselves into three divisions: (1) the ascertaining of the amount of soluble arsenic it took to kill a bee; (2) whether a bee working upon a mixture of insoluble arsenic and syrup would take up the arsenic particles; and (3) whether bees that were found dead near the experimental trees contained arsenic internally that accounted for their death.

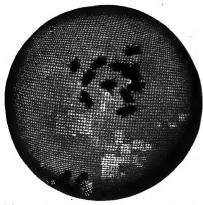


Fig. 2. Bee cage used in the laboratory.

In determining the fatal dose of arsenic we proceeded about as follows: Flying bees were taken in front of the hive by first drumming to cause them to fly densely at that place, then while so swarming, a wire cage (See Fig. 2) was dropped over them on to a smooth wide board, thus trapping them. The cages weré then moved into the laboratory and placed in a dark closet over night; the bees were fed the next morning. When treated in this way they are easily handled, and feed readily when given the

syrup.

The syrup was a 40 per cent solution of cane sugar. To this was added soluble arsenic, sufficient to give .000581 gram to each gram of syrup.¹ A drop of this was placed on a piece of wax foundation, and the whole weighed to one-tenth of a milligram. This was immediately taken from the balance and slipped under the cage containing the bees. Very soon (almost instantly) a bee would find it and begin feeding. The cage was then raised a bit from the board and placed to one side leaving the bee on the foundation in the open. A glass funnel was then placed over the individual (See Fig. 4) and

¹ Examination of dead bees to determine the presence or absence of arsenic was made by the Gutzeit method. Briefly, the laboratory procedure was as follows:

One bee that had been treated with sodium hydroxide and nitric acid for the removal of arsenic on the surface of the body

One dee that had been treated with soulain hydroxide and nitric acid for the removal of arsenic on the surface of the body was placed in a test-tube; a little water, an equal amount of hydrochloric acid, and some potassium chlorate were added, and the test-tube placed in a steam bath. Chlorine was evolved, which breaks down the tissue of the body, thus liberating the arsenic. The contents of the test-tube were then evaporated to near dryness to drive off the free chlorine. The residue was then examined in the apparatus shown in Fig. 3, which consists of an eight-ounce bottle (d) and three small tubes and stoppers. In the tube just above the bottle (c) a folded filter paper was placed which had been saturated with lead acetate and dried. The tube above this (b) contained a mass of glass wool saturated with lead acetate. The last tube (a) contained a small strip of filter paper which had been saturated with mercuric chloride and dried.

The contents of the test-tube were now placed in the eightounce bottle. To this were added hydrochloric acid, stannous chloride, ammonium ferric alum, water, and zinc. The zinc and acid cause an evolution of gas, hydrogen sulphid, arsine, etc. The sulphides were caught by the lead acetate, and the arsine was passed on to the sensitized mercuric chloride paper. When arsine is present, the paper turns yellow; if absent, no change is apparent. Figure 5 shows some tests of bees in the control feeding work when the amount of arsenic taken was known.

Considerable experience by the operator is necessary before reliable results can be obtained. For this work everything was standardized and unusual pains taken to secure accuracy. All materials used were frequently tested for arsenic, as this method properly handled is delicate enough to show positive results with much so-called chemically pure acid and zinc.

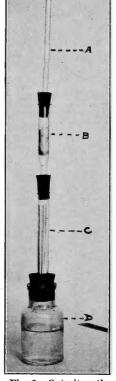


Fig. 3. Gutzeit method. D (8-oz. bottle) contains emulsified bee and reagents; C contains filler paper saturated with lead acetate then dried; B contains glass wool saturated with lead acetate; A contains sensitized mercuric chloride paper.

records kept of the time it lived after feeding and also of the post-mortem examination. As soon as it had finished feeding, the foundation and remaining syrup were weighed and the amount of syrup and arsenic computed. These records are shown in Table I.

Table I.—Showing Grams Syrup Taken, Grams Arsenic Taken, and the Number of Minutes Bees Remained Alive After Feeding,
Also the Post-Mortem Analysis for Arsenic.

Bee No. Gms. Syrup Gms. Arsenic taken Dead min. later		21150 the 1 00	Thortem I mary 31			
taken	Bee No	Gme Syrun	Gms Argenia	Dond	Arseni	c Anal.
2	Dec No.			min. later	*	0
2						
8 .0135 .0000078 375 * 9 .0105 .0000061 360 * 10 .0166 .0000096 300 * 11 .0072 .0000071 425 * 12 .0137 .0000079 410 * 13 .0232 .0000134 157 * 14 .0128 .0000074 455 * 15 .0122 .00000308 * * Arsenic in syrup reduced so that 1 gm. of syrup contains .000120 gms. arsenic 16 .0257 .00000308 500 * 17 .0215 .00000388 430 * 18 .0058 .00000258 430 * 19 .0303 .0000363 450 * 20 .0214 .00000256 430 * 21 .0157 .00000188 420 * 22 .0165 .00000188 420 * 23 .0263 .0000015 480 *	1					
8 .0135 .0000078 375 * 9 .0105 .0000061 360 * 10 .0166 .0000096 300 * 11 .0072 .0000071 425 * 12 .0137 .0000079 410 * 13 .0232 .0000134 157 * 14 .0128 .0000074 455 * 15 .0122 .00000308 * * Arsenic in syrup reduced so that 1 gm. of syrup contains .000120 gms. arsenic 16 .0257 .00000308 500 * 17 .0215 .00000388 430 * 18 .0058 .00000258 430 * 19 .0303 .0000363 450 * 20 .0214 .00000256 430 * 21 .0157 .00000188 420 * 22 .0165 .00000188 420 * 23 .0263 .0000015 480 *	2					
8 .0135 .0000078 375 * 9 .0105 .0000061 360 * 10 .0166 .0000096 300 * 11 .0072 .0000071 425 * 12 .0137 .0000079 410 * 13 .0232 .0000134 157 * 14 .0128 .0000074 455 * 15 .0122 .00000308 * * Arsenic in syrup reduced so that 1 gm. of syrup contains .000120 gms. arsenic 16 .0257 .00000308 500 * 17 .0215 .00000388 430 * 18 .0058 .00000258 430 * 19 .0303 .0000363 450 * 20 .0214 .00000256 430 * 21 .0157 .00000188 420 * 22 .0165 .00000188 420 * 23 .0263 .0000015 480 *	3					
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	4					
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	5					
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	6					
9	7				1	
10 .0166 .0000096 300 * 11 .0072 .0000041 425 * 12 .0137 .0000079 410 * 13 .0232 .0000134 157 * 14 .0128 .0000074 405 * 15 .0122 .0000074 450 * Arsenic in syrup reduced so that 1 gm. of syrup contains .000120 gms. arsenic 16 .0257 .00000388 500 * 17 .0215 .00000258 430 * 18 .0058 .00000363 450 * 20 .0214 .00000256 430 * 21 .0157 .00000188 420 * 22 .0165 .00000188 420 * 23 .0263 .00000188 420 * 24 .0046 .00000025 590 o 25 .0240 .0000015 590 o	8					
11		.0105	.0000061	360		
12			.0000096			
13						
14 .0128 .0000074 405 * Arsenic in syrup reduced so that 1 gm. of syrup contains .000120 gms. arsenic 16 .0257 .00000308 500 * 17 .0215 .00000258 430 * 18 .0058 .00000069 590 * 19 .0303 .00000363 450 * 20 .0214 .00000256 430 * 21 .0157 .00000188 420 * 22 .0165 .00000188 420 * 23 .0263 .00000188 420 * 24 .0046 .00000315 480 * 25 .0240 .0000055 590 o 25 .0240 .00000188 440 * 26 .0051 .00000161 610 * 27 .0205 .00000129 600 * 28 .0225 .00000130 600 *<						
15						
Arsenic in syrup reduced so that 1 gm. of syrup contains .000120 gms. arsenic 16						
Contains .000120 gms. arsenic .00000308 .500 *	15		.0000070	450		
Contains .000120 gms. arsenic .00000308 .500 *		Arsenic	in syrup reduced se	o that 1 gm	. of syru	р
$\begin{array}{cccccccccccccccccccccccccccccccccccc$			contains .000120 g	gms. arsenic		
18 .0058 .00000069 590 * 19 .0303 .00000363 450 * 20 .0214 .00000256 430 * 21 .0157 .00000188 420 * 22 .0165 .00000198 370- * 23 .0263 .00000315 480 * 24 .0046 .00000055 590 O 25 .0240 .00000288 440 * 26 .0051 .00000129 600 * 28 .0225 .00000142 540 * 29 .0131 .0000082 600 * 30 .0208 .0000131 610 * 31 .0207 .0000130 600 * 32 .0293 .0000184 480 * 33 .0160 .00000100 500 * 34 .0144 .00000090 630 0 35 .0120 .0000075 670 * <td></td> <td></td> <td></td> <td></td> <td></td> <td></td>						
19 .0303 .00000363 450 * 20 .0214 .00000256 430 * 21 .0157 .00000188 420 * 22 .0165 .00000198 370- * 23 .0263 .00000315 480 * 24 .0046 .00000055 590 o 25 .0240 .00000288 440 * 26 .0051 .00000061 610 * 27 .0205 .00000129 600 * 28 .0225 .00000142 540 * 29 .0131 .00000082 600 * 30 .0208 .0000131 610 * 31 .0207 .00000130 600 * 32 .0293 .00000184 480 * 33 .0160 .00000100 500 * 34 .0144 .00000090 630 <						
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$						
$\begin{array}{cccccccccccccccccccccccccccccccccccc$					*	
$\begin{array}{cccccccccccccccccccccccccccccccccccc$				430	*	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$						
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$				370-		
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$.00000315	480	*	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$						0
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$				440	*	
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$					*	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$					*	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$				540		
$\begin{array}{cccccccccccccccccccccccccccccccccccc$				600	*	
$\begin{array}{cccccccccccccccccccccccccccccccccccc$				610	*	
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$				600	*	
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	32			480	*	
35 .0120 .00000075 670 *	33			500	*	
35 .0120 .00000075 670 *				630		0
			.00000075	670	*	_
ı j	36	.0329	.00000207		*	
		I ,				

Numbers 16-26 inclusive were fed between 8:40 and 9:12 a.m. This gives some idea regarding the rapidity of the feeding process.

The sign * is used in the table to indicate that the bee gave a positive test for arsenic; the o sign indicates the opposite reaction.

It might have been possible to attribute the death of the bees fed with arsenic, considered in Table I, to other causes. For instance, death

might presumably occur as a result of old age, or through injuries received in colliding with the sides of the cage in a desperate attempt to escape.

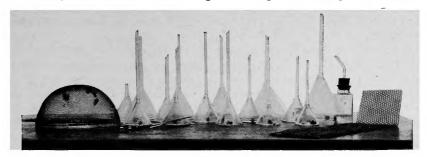


Fig. 4. Part of laboratory apparatus used in the feeding work.

In order to determine the actual death rate resulting from such causes, 48 bees were placed in a screen cage alongside the poisoned ones, and kept well supplied with unpoisoned syrup (See Fig. 4). None of the bees in this cage died during a period of nearly four days. This was fully three days after all the bees were dead in the cages containing poisoned syrup. At the end of this period, the cage was lifted, and all the bees fed on pure syrup flew away.

Blank	.0000010 9/NS.	.0000020 9me.	0000040 9ms.	0000060	.0000080	.0000120 gms.	0000150	0000200
					1		ı	1
Blank	A Positive. Field Bee.			.00000184			,0000/12 1ms.	.000037/
Blank	rositive	.0000005	00000/2	00000/84	0000025	0000010	2000/12	,000037.

Fig. 5. Mercuric chloride papers showing tests from known amounts of arsenic. Arsenic caused the papers to turn yellow. In the picture it shows dark.

The third syrup (I gm. sol. containing .000063 gm. arsenic—See Table I) was diluted with equal parts of sugar syrup and given to seven bees under the cage. A supply was kept near them continuously. This was given them on July 19, 1:30 p. m. At 8 a. m. on July 20 there were four dead and two on their backs; on July 20, at 2 p. m., there were five dead and two on their backs; at 10:30 p. m. on July 20 all were dead.

This same syrup (.000063 gms. arsenic per gm. syrup—See Table I) was diluted again. This time three parts of the poisoned syrup was diluted with one part clean syrup. Eleven bees were given the syrup in abundance on July 19, at 2:15 p. m. On July 20, at 8 a. m., there were eight dead and one on its back; at 2 p. m., nine were dead; and at 10:30 all were dead.

A third dilution of the .000063 gm. poisoned syrup, was made by adding three parts of clean syrup to one part of the arsenic syrup. This was supplied in the same way as the above to eight bees on July 19, at 2 p. m. On July 20, at 2 p. m., one was on its back, all others appeared healthy; at 10:30 p. m., on July 20, one was dead and two on their backs. On July 21, at 11:30 a. m., two were dead and three were on their backs; on July 22, at 8 a. m., all were dead.

Of the 27 bees used as a check on the last three groups, only two died during the period of observation.¹

In order to see what the effect of the ordinary commercial arsenate of lead would be, a 40 per cent sugar solution was made containing the equivalent of one pound of powdered arsenate of lead to 50 gallons. It was fed to 26 bees on July 23, at 10:15 a. m. All bees were in one cage, and no individual records were kept. Twenty-five of the 26 died during the night of July 23, and all were dead at 10 a. m. on July 24. A check of 31 carried alongside of this cage showed one dead bee.

It will be seen from the data given above that the bee is capable of taking into its system at one feeding .063 grams of syrup. This amount of syrup in our first dilution contained .0000371 grams of arsenic, which was the largest dose of poison taken in these experiments. The bee lived 150 minutes after feeding. Another bee with a smaller dose of poison (.0000142 grams) lived 110 minutes after feeding. The smallest dose administered was .00000055 grams of arsenic, and the bee lived 590 minutes after feeding. The experiments offer evidence that a very small amount of arsenic is fatal to the bee, and that a bee takes up lead arsenate particles in feeding on a sugar solution having them in suspension.

It was noticed in the control feeding work that the poisoned bee began to behave abnormally (sick) soon after feeding, usually within 20 to 30 minutes. It would drop from the top or side of the cage to the floor buzzing and spinning on its back like a decapitated fly. It seemed unable to right itself and continued to struggle in this way until it finally died.

¹ In all the above experiments arsenic in soluble form was used, because it lent itself to more accurate computation in the small doses given. The commercial arsenate of lead is insoluble in water and nectar, and therefore it was impracticable to use it in these tests. The small particles of arsenate being in suspension or grouped in the liquid prevents equal distribution. A bee feeding on a certain amount of the liquid might take a large amount of the poison particles, whereas another bee taking a like amount of liquid from the same receptacle might take only a small amount of the poison particles. There would thus be no reliable way of computing the amount of arsenic in the doses taken in the control feeding experiments.

It may appear from this that the possibility of bees bringing much poisoned honey into the hive is rather remote. Demuth¹ has shown that the bee makes on the average about four trips to the field in a day. If this is the case, and if sickness comes in 20 minutes after feeding to a degree that makes it impossible to fly to the hive, it is clear that those unfortunate enough to gather the poisoned material will in most cases be unable to return to the hive with their loads.

Dead and dying bees were found in abundance in the blossoms and flower clusters as well as under the trees in the vicinity of the trees sprayed in the open. This condition was not confined to the sprayed trees alone, but the trees in the immediate vicinity of those sprayed were well littered with dead. In the other end of the orchard, where there were no sprayed trees (See Fig. 8), no dead or dying bees could be found.²

Is it not possible that the bees found on and around the unsprayed trees in the vicinity of the sprayed ones secured poison from the sprayed tree, and in further quest for nectar flew to the adjacent trees which had no spray on them, and there became sick and unable to proceed farther?

FIELD WORK-First Year

The first year two trees were caged. Cheese-cloth was used for the four sides, top, and bottom. The trees were of the Jonathan variety and twelve years old.

On Sunday, May 5, the trees were in full bloom. The next morning, May 6, one of the trees was sprayed with lime sulphur (1° B. or one gallon lime sulphur to 50 gallons of water) and arsenate of lead, one pound to 50 gallons. A colony of bees was moved in and liberated. The other tree was dusted with flowers of sulphur and arsenate of lead (85 per cent sulphur and 15 per cent arsenate of lead). A colony of bees was put within its enclosure and liberated. Dead bees were gathered in the cages and data secured as shown in the following table. At the conclusion of this phase of the work (morning of May 10), escape boards were placed on the hives and the bees counted. In the hive in the sprayed lot there were found 3880 living bees, and in the dusted lot there were found 4960. This gives a mortality in the case of the sprayed tree of 13.5 per cent, and in the dusted one 18.6 per cent.

¹ The following unpublished data are from the writer's notes from one of Demuth's lectures: Experiments were carried on by Geo. E. Demuth, Bureau of Entomology, U. S. D. A., for another purpose which incidentally furnished data concerning the number of trips a bee makes per day in gathering nectar. Six colonies were weighed in the morning and evening each day. The difference in their weights represented the amount of nectar gathered that day. Bees with and without loads of nectar were weighed and it was found that when nectar was abundant they carried from one-half to two-thirds of their own weight in nectar (four to six m.g.). With this information, knowing the number of bees in the colony and the proportion that go to the fields, it was not difficult to compute the number of trips made each day by each bee. It was found that the average number of trips made was about four; old bees making 2½ trips and young ones more than five. These experiments were carried on in the midst of large commercial orchards when apple blossoms with much nectar were only four rods away.

 $^{^2}$ This observation was made by Prof. Laurenz Greene, Chief in Horticulture, Purdue Univ. Agr. Exp. Sta.

³ Twenty-four bees were analyzed that were not treated with sodium hydroxide and nitric acid; 21 of these were strongly positive in the arsenic tests. This indicates the importance of careful treatment with sodium hydroxide, acid, and washing.

TABLE II.—Showing Time of Gathering and the Number of Dead Bees Taken From the Sprayed and Dusted Trees

TN	No. bees					
Time of gathering	Sprayed	Dusted				
May 7—8:00 A. M	175 13 101 11	240 428 29 279 17 141				
Mortality	607	1134 18.6 per cent				

Table III.—Showing Positive and Negative Tests for Arsenic Obtained From the Examination of 210 Bees From the Sprayed Tree.

Each Bee Is Represented by * or o, the * Indicating a Positive Test and the o a Negative Test

Lo	t 1	Lo	t 2	Lo	t 3	Lo	ot 4	Lo	t 1	Lo	t 2	Lo	t 3	Lo	t 4
*	0	*	0	*	0	*	0	*	0	*	0	*	0	*	0
* * •	 0 0	* *	 0	* 	0 0 0		0 0 0	*	 o	*	0 0	*	 0 0	* *	
*		*	- -		0	F%		*			ő	*		*	
*	0	* *			0	*	0	*		*	0		0.	*	
	o		0		ő		o	*			o	*			0
	0	*		*		*			0		0	*		*	
	0		0		0		0	*		*		*		*	
	0	*	0	*	0		0	*			0	*			0
	0		0	*			0		0	*			0	*	
-	0	*	0		0		0	*		*			0	*	
	0	*		*			0		0	*	0	*		*	
*	0	*		*			0	*		*		*		*	
	o	*		*			0		0		0	*	0	*	
*		. *	- -	*			0		0	*		*		*	0
*			0		0		0			*				*	
	0	*			0	*							0	*	
*			0	*		*									0
*			O	*		*									0
*	0		0	*			0	25	25 50	29	24 53	30	22 52	29	25 54

This mortality cannot be attributed entirely to poisoning. It is reasonable to assume that part of these were killed mechanically or died of other causes. To determine which of the dead were poisoned, chemical analyses were made. After the bees were collected from the cages they were taken to the laboratory and treated with one-tenth normal sodium hydroxide, heated to boiling; then enough nitric acid was added to make the solution acid, and this boiled two minutes, washed five times with distilled water, and dried in the oven. This treatment was for the purpose of removing any arsenic that might be on the surface of the bee. Individuals were then taken from the various lots and analyzed according to the Gutzeit method for determining small amounts of arsenic. The records are shown in Tables III and IV.

Table IV.—Showing Positive and Negative Tests for Arsenic Obtained From Examination of 216 Bees From the Dusted Tree. Each Bee Is Represented by * or o, the * Indicating a Positive Test and the o a Negative Test

	Lo	t 2	Lo	t 3	Lo	t 4	Lo	t 1	Lo	t 2	Lo	t 3	Lo	t 4
* 0	_	0	*	0	*	0	*	0	*	0	*]	0	*	0
* *	* * * * * * * *	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	* * * * * * * * * * * * * * * * * * *		* * * * * * * * * * * * * * * * * * *	0 0 0 0 0 0 0								
	0	. O 	*			0 0		0 			* .			
*		0	* *			0 0 0	31	21 52	30	21 51	38	15 53	24	24 48

¹ See note 3, page 9.

FIELD WORK-Second Year

This year (1919) three trees, of the Jonathan variety and about 12 years old, were caged with wire screen on the sides and top and cheese-cloth on the bottom. One was used as a check; the other two were sprayed and dusted with sulphur and arsenate of lead when in full bloom, as described for the previous year. Colonies of bees having 18 pounds or more of capped honey were moved in soon after the spraying and dusting had been done.

The weather was cold and rainy during a portion of the period of observation (April 30-May 15). Dead bees were gathered in the cages at intervals as follows:

TABLE V.—Showing Date of Gathering and the Number of Dead Bees Taken From the Check, Sprayed, and Dusted Trees

Date	Check tree	Sprayed tree	Dusted tree
April 30	5	143	45
May 2	1716	1770	1556
May 7	768	4906	1593
May 10	486	2174	451
May 13	427	1675	735
May 15	287	959	327
Living bees at end of	3,689	11,627	4,716
observation	15,726	7,223	5,536
Per cent mortality	19 per cent	69 per cent	46 per cent



Fig. 6. Tree being examined was one of the several trees to be sprayed in the open.

These three colonies were normal, healthy colonies with plenty of stores at the beginning of the experiment. The bees were not compelled to work the blossoms for food, and their activities generally were such as would arise from choice. On coming out of the hive the morning after moving into the cages, a goodly number of the bees flew directly to the screen, where they tried to escape. A large number of these, it is reasonable to assume, died as a result of their frantic efforts to escape. It is probable that these were the individuals that showed no trace of poison in the arsenic test. While a large number went to the walls of the cage to escape, a goodly number went to work immediately on the blossoms.

It was interesting to note that individuals coming from the hive to the blossoms would, after feeding, return to the hive or go directly to the walls of the cage.

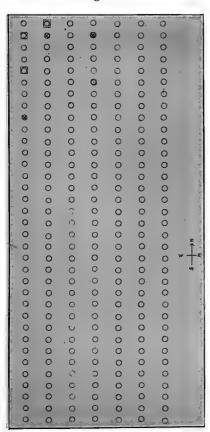


Fig. 7. Plan of orchard. Circles represent trees. Circles in squares indicate caged trees, and circles with x's represent trees sprayed in the open.

In front of the hives in the sprayed and dusted trees there were piles of dead bees. This may be explained on the basis of death occurring in the hive and the bodies being carried out by the other workers. There were no dead bees in the hives at the end of the experiment. The spot directly in front of the hive and the floor near the walls contained the large majority of the dead bodies, but the entire floor of the cage was always well littered with dead. Those found directly under the trees probably died on the blossoms and later fell to the floor.

As another part of the field work, trees in the open and in full bloom and in the same orchard (See Fig. 6) were sprayed and observed to determine whether bees visited them. thought there might be a possibility of a forced working of the sprayed blossoms by the bees in confinement (in the cages) as they were not free to choose the flowers that they worked. It was a case of visiting the sprayed blossoms or none at all. However, with trees sprayed in the open and with unsprayed trees on all sides the possibilities of forced feeding on these trees was removed, and it is reasonable to assume that bees found on these trees were there by virtue of their desire, not because of imposed conditions.

At intervals a survey was made of the sprayed trees and the trees on each side of them, which were essentially the same except that they had no spray on them. Bees actually on the blossoms were counted. The count was made while walking around each tree, and an equal amount of time was given for the count on each tree. The results of these surveys are shown in the following table. The rows of circles indicate three rows, and each circle represents a tree. The daggers show the location of the sprayed trees in the open. Asterisks indicate caged trees, and the figures represent the number of bees counted on the tree. (Fig. 7 shows plan of orchard).

TABLE VI.—Showing the Number of Bees Found on the Sprayed Trees in the Open and the Number of Bees Found on the Unsprayed Trees About Them.

April 30—11 a. m. O ⁴ *0 O ⁸ *0 †0 ⁷ O ¹⁰ O ⁵ O ¹³ O ⁷ O O O O O *0 O O O O O O *0 O O O O O O *0 O O O O O O O O *0 O O O O O O O O *0 O O O O O O O O O *0 O O O O O O O O O *0 O O O O O O O O O *0 O O O O O O O O O *0 O O O O O O O O O *0 O O O O O O O O O O *0 O O O O O O O O O O O *0 O O O O O O O O O O O O *0 O O O O O O O O O O O O *0 O O O O O O O O O O O O *0 O O O O O O O O O O O O O O O O *0 O O O O O O O O O O O O O O O O O O O				11				
Nay 6-11 a. m. Nay 6-11 a. m.	Ap	ril 30—11 a	ı. m.					
*0 †07 O10	04	*0	O ⁶			oi may	b, and 1	recoraea
05 013 07 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	_	•	•	as Tollow		C11_c	. m	
**O O O O O O O O O O O O O O O O O O O	=		-		_			0.5
0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	_	_	-		-	~	_	-
**O O O O **O O O O O O O O O O O O O O	О	О	0		-	_		
*O O O O O O O O O O O O O O O O O O O	0	О	0		_	-		_
April 30—2 p. m. O O O O O O O O O O O O O O O O O O O	*O	О	0					
April 30—2 p. m. O O O O O O O O O O O O O O O O O O O				-	-			
O	An	ril 30—2 p	m	0	Ō	Ō	Ō	
*0 †0° 0					O_6	О	0	0
*0 †0° 0° 04 0° 04 *0 *0° 04 0° 04 *0 *0° 04 0° 08 *0 *0 *0° 01° 00 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	Ω^7	*0	O^{12}					
O⁴ O⁵ O⁴ May 6-3 p. m. O *O O¹¹¹ O⁵ May 2-2 p. m. O *O O¹¹² O⁵ O⁵ *O O¹¹² O° O° <td>_</td> <td>_</td> <td>-</td> <td> O13</td> <td>O^2</td> <td>O</td> <td>0</td> <td>0</td>	_	_	-	O13	O^2	O	0	0
May 2—2 p. m. O *O O1 ¹⁷ O1 ¹² O8 *O O O1 ¹⁸ †O1 ¹⁴ O3 O O O O8 *O O O O O O *O O O O O O *O O O O O	=	•	_	\ \	Mav	6—3 p	. m.	
May 2—2 p. m. **O	O*	O°	O.	0		_		O8
May 2—2 p. m. 0 0 08 07 09 08 *0 010 0 <td></td> <td></td> <td></td> <td></td> <td>_</td> <td></td> <td></td> <td>-</td>					_			-
Os *O O10 *O †O6 O11 O2 O5 O8 Trees sprayed again May 3—2 p. m. Count made 30 minutes later. O12 *O O4 *O †O10 O12 O O O O O O *O O O O O O O O O O O O O O O O O O O O O O O O O O O O O O O *O O O O	, M	ay 2—2 p.	m.					
*O	08	*0	010	0	0	О	0	0
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	_	_	•					
Trees sprayed again May 3—2 p. m. Count made 30 minutes later. O12 *O O4 *O †O10 O12 O7 O14 O8 May 3—3 p. m. O4 O8 O O O †O4 O5 O O May 7—10 a. m. O *O O10 O15 O4 *O O O6 †O8 O1 O O O O12 O13 O2 O O O O O *O O O O O *O O O O O *O O O O		•	=				_	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	O^2	O _E	O_8		-		_	
Count made 30 minutes later. O1 O2 O O O May 7—10 a. m. O2 O O O May 7—10 a. m. O3 O O O O May 7—10 a. m. O4 O O O O O O O5 O O O O May 3—3 p. m. O1 O2 O O O O May 7—10 a. m. O2 O O O O May 7—10 a. m. O3 O O O O O O4 O O O O O O5 O O O O O6 O O O O O7 O O O O O8 O O O O O8 O O O O O8 O O O O					-	_		
May 7—10 a. m. O *O O¹° O¹° O¹° O¹° O¹° O¹° O¹° O° †O° O° †O° O° O								
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	Count ma	ade 30 min	utes later.	0-	_	_	-	U
*O †O¹¹ O¹²	019	**	0.4		May		. m.	
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	•	_	•					
May 3—3 p. m. O O O O O O O O O O O O O O O O O O O	- •		•					
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	O^{τ}	O14	O_8				_	
$\begin{array}{cccccccccccccccccccccccccccccccccccc$					_			
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	M	ay 3—3 p.	m.					
*0 $+0^{8}$ 0^{12} 0^{9} 0^{8} 0 0 0 0 0	0.15							
*0 $\dagger O^3$ O^{12} $\dagger O^7$ O^5 O O	_	_					_	
	*O	†O³	O12		_	-		
	O ₁	Q ⁴	Ο ⁷					

It is seen from these surveys, a part of which were made by disinterested and impartial persons who did not, at the time, know the purpose of the count, that the bee does not avoid sprayed trees. There were 245 trees in the orchard (Fig. 7), and all were available to the bee except the three that were caged. If there had been any discrimination against the

spray, there certainly would have been no bees found on the trees so treated.

As a matter of more definite record bees were taken while working the blossoms of the sprayed trees. This was done by pinching off the flower cluster and dropping it with the bee on it into an insect net. Seventeen bees were gathered in this manner by the writer from one of the sprayed trees in 25 minutes. Later in the day three of us gathered 36 in 30 minutes under the same conditions.

Soon after gathering these two lots, they were taken to the laboratory, placed under a wire cage, and given clean (containing no arsenic) syrup and observed.

At the same time 20 bees were taken from the check hive in the caged experiment and run alongside these as a check.

The results here are very significant. They show the 17 bees gathered at 11 a. m. to be dead at 1 p. m. of the same day, and also the 36 gathered between three and four o'clock were all dead at 8 p. m. of the same day. The check of twenty were all alive the next day at noon, and when liberated flew away.

Chemical analyses were made of all of the dead, and the records show that 48 of the 53 gave a strong reaction to the test for arsenic. (At the same time 20 worker bees and six drones from the check hive were analyzed and no arsenic was found in any of them.) Five gave negative results. This might be explained on the basis of the amount of arsenic being so small, yet fatal, that it escaped our test. See Table I.

Since the Gutzeit method, which is considered a delicate test, failed to show a reaction for arsenic with these five bees, it is obvious that an extremely small dose of arsenic may cause the death of a bee. It is not likely that the five bees showing the negative test could have died from natural causes, since there were no deaths from the check lot of 20 bees.

The general conclusion which may be drawn from this experiment is that a sufficient amount of poison is to be found in lead arsenate or other arsenical sprays applied to the blossoms to cause a tremendous death rate of bees.

SUMMARY

1. The Gutzeit method of determination of arsenic appears to be sensitive enough for most doses that kill bees; however, it is believed that some very small fatal doses may escape detection.

2. A very small amount of arsenic (less than .0000005 grams, as 203)

is a fatal dose for a bee.

- 3. The time required to kill the bee with arsenic depends upon the size of the dose. Some expire within one and one-half hours from the time of administration of the poison, others linger on for a period of five or six hours. Most of those observed to die from a dose gathered in the field did so within three hours.
- 4. Bees work freely on sprayed trees in the open, even when there are unsprayed trees all about.
- 5. The mortality in the check cage was 19 per cent, as compared with 69 per cent in the lime-sulphur-arsenate of lead sprayed cage, and 49 per cent in the sulphur-arsenate of lead dusted cage.
- 6. For the sake of the bee, fruit trees should not be sprayed while in full bloom.

	그 그는 그 그 그는 그는 그는 그는 그는 그는 그를 만든다. 경우를 받는다.
	기계 등에 가는 사람들이 되었다. 사람들은 사람들이 가지 않는 수 있 었다.
	그 얼마나 그 아이를 하는데 하는데 하는데 하는데 없었다.
	보다 하는 그 그는 것이 되는 사람들은 것이 되고 해결합니다. 생물
•	