hard waters with a hardness of exactly 1000 p.p.m. expressed as calcium carbonate. A striking difference is apparent. The one on the right is in water which contains only calcium chloride and shows a heavy cream layer on the bottom. The emulsion on the left is in water containing both calcium chloride and magnesium sulfate in a molar ratio of 3 to 2, and shows far superior stability.

This also is an exaggerated case; no natural water would contain only a single mineral constituent. However, it does show the limitations to be expected from reliance solely on a series of synthetic waters. For this reason it has been found necessary to collect a large number of natural water samples, from all over the country, which are stored in polyethylene lined bottles. Obviously, a collection such as this cannot be considered representative of the over-all water supply, as some of these samples are from single wells or swamp waters which were supplied because they gave rise to some difficulty in spraying. These water samples are analyzed and the effect of their composition on emulsification is carefully studied. In time, sufficient data should be accumulated to permit the compensation in formulation for most of the unusual or difficult water conditions that may be encountered.

Aging Qualities

The final step in the formulation of concentrates, in so far as laboratory testing is concerned, is a study of aging qualities. Unsatisfactory aging may manifest itself in a variety of forms, from minor defects like discoloration and loss of clarity, which merely affect the appearance of the concentrates, to the more serious functional deficiencies, such as separation in layers, crystallization of the toxicant, or even total loss of emulsification. In addition, there is always the possibility of a reduction in biological potency of the toxicant, due to a decomposition which may not be apparent in an examination of the concentrate's physical properties. In many cases, when there is insufficient time to observe the effect of normal aging of a concentrate, it is necessary to use accelerated aging tests at elevated temperatures. Unfortunately, the results obtained under these conditions are not too reliable, and at best are only approximations. Because the effect of temperature on stability not only is different for each toxicant but also depends on the concentration of the toxicant in each formulation, it becomes almost impossible to find any exact correlation between the results of accelerated aging tests and the aging qualities under normal storage conditions. Some toxicants (such as toxaphene, *6*), when subjected to elevated temperatures even for only a very short time, seem to suffer a slight though autocatalytic decomposition which will greatly shorten stability on subsequent storage, even at ordinary temperatures.

When all the actual formulating work is done, there still remains a number of problems in which the laboratory can supply information or guidance on the proper use and handling of concentrates. These will include, for instance, corrosion studies on various metal surfaces and testing of resistant coating materials for their suitability as container linings. Special attention should be given to formulations which will be marketed in small containers such as 1-gallon cans, where the use of liners is not economically feasible.

Furthermore, there is the problem of the compatibility of the concentrate with other common agricultural materials, in either liquid or powder form, with which it may come in contact. For instance, liquid insecticide concentrates are often mixed with fungicides which are usually in the form of wettable powders or dusts. Some of these materials, especially certain types of Bordeaux mixtures, may well interfere with the normal emulsification of the insecticide concentrate and cause rapid settling or even flocculation. Knowledge of these possible limitations beforehand will avoid trouble and poor results in the field.

Conclusions

Although the problems are numerous and often complex, the art of formulation has now sufficiently advanced so that the new synthetic pesticides can be applied with a high degree of efficiency, safety, and economy.

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PESTICIDES FORMULATION

Influence of Formulation on Effectiveness

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PROMINENT AMONG THE FACTORS affecting the performance of pesticides is the manner in which they are formulated for commercial use. The principal proprietary pesticide formulations are the wettable powders, emulsifiable concentrates, emulsions, solutions, and dusts. The present investigation deals only with the first two, primarily in relation to four acaricides: Aramite, Geigy-338, Sulphenone, and parathion. Many other pesticides will be investigated in the course of a research project, of which the present paper should be considered as no more than a progress report. It is expected that all the aforementioned formulations will eventually be investigated with regard to the physical factors that influence their pesticidal effectiveness.

A wettable powder is a dry mixture of

Per unit of toxicant deposited, emulsions have usually been more effective in topical applications than have suspensions, but have been less effective as residues. This tendency has been demonstrated again with a number of acaricides used against the two-spotted mite. The mortality of the mites is influenced to a large degree by the portion of the toxicant that penetrates through the leaf cuticle, to be ingested by the feeding mites. The superiority of the "residue application" to the "topical application," when used against the two-spotted mite, is believed to be due, at least in part, to the additive effect of the surface and the subcuticular residues of the former. With surface residues removed, the subcuticular residues of some acaricides may be effective against mites for as long as a month. Likewise, an acaricide may result in the death of mites on the leaf surface opposite to the one that is treated. Bioassays indicate that, in the initial 24-hour period, toxicants deposited by emulsions penetrate in greater quantity than those deposited by suspensions.

a toxicant and an inert diluent, which usually contains 25 or 50% of toxicant and a small quantity of wetting agent. An emulsifiable concentrate is a single-phase liquid system containing a toxicant and one or more surface active agents. Usually the toxicant comprises 25% by weight of the solution. Both powders and concentrates are ordinarily formulated to contain the maximum possible amount of toxicant, for reasons of economy. The recent trend to low-volume application of pesticides has caused a great increase in the production of the emulsifiable concentrate. This formulation emulsifies spontaneously or with very little agitation and is therefore particularly suitable for the type of application equipment ordinarily used for low-volume spraying.

Per unit of toxicant deposited, the emulsions formed by the emulsifiable concentrates have usually been more effective in topical application than the suspensions formed by the wettable powders. It is believed that when emulsions are used, the toxicant is more uniformly and more completely in contact with the body of the insect. Likewise, the oil or solvent in which the toxicant is dissolved may aid in its penetration through the insect cuticle. On the other hand, the suspensions formed by the wettable powders have usually resulted in longer residual effectiveness. This is believed to be due to the fact that the particles deposited by a suspension are more readily "picked up" by the tarsi of the insects that crawl over them (*1*, *2*). Further information on this subject is presented in this paper with reference to acaricides used against the two-spotted mite, *Tetranychus bimaculatus* Harvey.

In the course of the present investigation it became evident that the mortality of two-spotted mites was influenced to a large degree by quantities of acaricide that penetrated through the leaf cuticle, to be ingested by the feeding mites. Preliminary data on the influence of subcuticular residues on acaricidal action are included in this paper as essential to further studies on the influence of formulation on the effectiveness of acaricide treatments.

Equipment

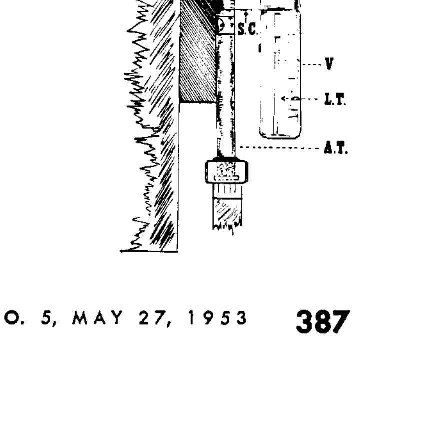
Settling Tower In the laboratory, treatments were made by means of either a settling tower or spray equipment, both based on the Venturi principle. Figure 1 is a diagram of the Venturi used for the settling tower. Air passing through the tube, *A.T.*, draws 5 ml. of liquid from the vial, *V*, in about 6 seconds through the liquid tube, *L.T.*, which has an inside diameter of approximately 1 mm., except that at its outlet the tube is compressed to an extremely small orifice. The liquid in the vial is shaken before it is inserted in the clamp, and this suffices to keep the suspended material uniformly distributed for the short period required for evacuating the 5-ml. charge.

Figure 2 shows the entire settling-tower assembly. Above the turntable is a glass cylinder 12 inches in diameter and 28 inches high. The greater part of the mist will settle down on the turntable in 3 minutes, the period selected for the experiments made in this laboratory. At the expiration of the 3-minute period, the lever shown in Figure 2 is pulled down. This simultaneously lifts the metal lid of the glass cylinder and elevates the right side of the sliding platform upon which the tower rests, so that the tower can be pushed to one side while the material to be treated in the following test is placed on the turntable. It is then slid back into its original position.

The settling tower has the merit of depositing at a given concentration a constant quantity of toxicant per unit of treated surface, whether the acaricides are applied as emulsions or suspensions. The amount of toxicant deposited, at a given concentration, depends entirely on the quantity of liquid falling on the treated surface. This quantity was determined by weighing, in an atmosphere of 90% relative humidity, the liquid settling on waxed paper disks cut into squares of an area of 9 square inches. The waxed papers were placed in Petri dish uppers resting on the revolving turntable of the settling tower so as to simulate the treatment of foliage in the experiments described later. For an average of five tests, water alone deposited almost exactly the same amount of liquid as a 50% DDT wettable powder suspension containing 0.5% (weight/volume) of toxicant. A 25% DDT emulsifiable concentrate at 1.0% of toxicant deposited only 5.2% less liquid than the wettable powder suspension. The smaller droplets in the mist formed by the dilute emulsion settled somewhat more slowly than those of the suspension, and, although a 3-minute period was

Figure 1. Equipment for generating aerosols or mists

A.T. Air tube
B. Bearing
L.T. Liquid tube
O. Orifice of air tube
O′. Orifice of liquid tube
P. Pulley and belt
S.C. Spring clamp for holding vial
T. Base of turntable
V. Vial



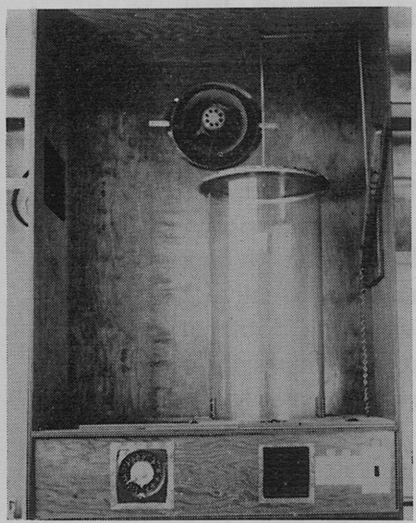


Figure 2. Settling tower

allowed for the settling of the mist, a greater quantity remained suspended in the air at the end of this period when emulsion was used. Kerosene deposited 29.4% less than the aqueous suspension.

The kerosene droplets were very small compared to those of either of the aqueous formulations and consequently had less completely settled to the bottom of the tower at the expiration of the 3-minute exposure period. From the weights of liquid deposited at the bottom of the settling tower from a 5-ml. charge, the quantity of toxicant deposited from suspensions, emulsions, or solutions of 1% concentration was calculated (Table I). The kerosene solutions deposited much less toxicant than the aqueous preparations, but kerosene was not used in the experiments reported in this paper.

Table I. Calculated Deposit on Turntable in Settling Tower from Three Formulations of 1% of Toxicant

Formulation	Quantity of Charge, Ml.	Deposit of Toxicant, γ/Sq. Cm.
Suspension (wettable powder)	5	14.59
Emulsion (emulsifiable)	5	14.10
Kerosene solution	5	10.30

Laboratory Spray Equipment The Venturi apparatus shown in Figure 1 was duplicated in essential detail in the construction of the spray equipment, Figure 3, except that the discharge orifice was larger. Consequently a greater volume of liquid is discharged and the spray is of a greater droplet size. Because a much longer period is required to empty the container, solids cannot be uniformly suspended merely by shaking the suspension before using, and an agitator (Figure 3, *A.B.*) is provided. The spray is discharged upward against a baffle plate in a hood made from an inverted 5-gallon Army ammunition can. Through a slit in the side of the can a leaf is inserted and held in place against the baffle for 3 seconds. Mist and vapors are exhausted from the hood by means of an exhaust fan, and liquid that drops to the bottom is drained out below.

For the spraying of individual leaves on cuttings in the laboratory, or the upper or lower surfaces of the leaves, a McCarl fog maker was used. This is an 18-ounce capacity sprayer, with positive trigger action pump, that can be operated with one hand, leaving the other free to hold the leaves in place while spraying.

Field Spray Equipment For the spraying of orchard trees a 200-gallon power sprayer was used. The spray was applied with a pressure of 600 pounds and with a 9/64-inch orifice in the disk of the spray nozzle, thus simulating commercial spray application. Sometimes, when individual branches were treated, a 2-gallon-capacity knapsack sprayer was used.

Technique

Rearing Of Mites Two-spotted mites were reared on Henderson lima beans. This variety of beans was selected because of the practically complete absence of plant hairs. The beans were grown in flats in a coarse vermiculite that could be easily shaken from the roots of the bean plants and reused after steam sterilization. The mites were reared in a greenhouse chamber at a temperature of 80° to 90° F., and a relative humidity of 70 to 80%.

Treatment The mites were treated by one of three methods: topical, topical-residue, and residue. Adult females were transferred by means of a No. 000 spotting (sable's hair) brush, and, with the aid of the low power of a stereoscopic microscope, from the bean leaves on which they were reared. In the topical method, they were placed on the lower surface of an avocado leaf section or on the lower surface of a bean leaf. For each dosage two leaves were used, with 25 mites per leaf.

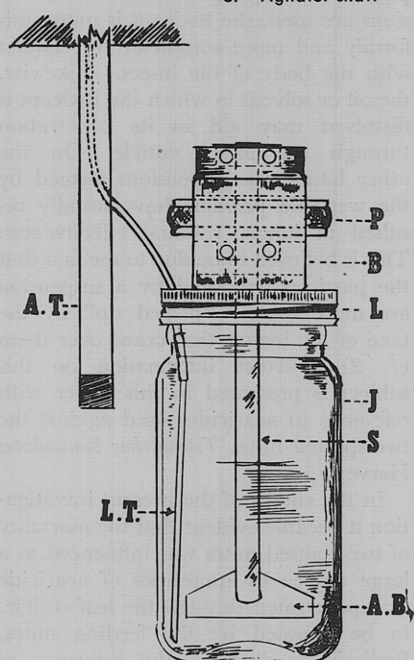
Sticky banding material was smeared around the margins of the avocado leaves to prevent the escape of the majority of the mites, but this was not necessary when bean leaves were used. The tendency for mites to become mired in the sticky banding material of the avocado leaves or the wet cotton surrounding the bean leaves increased with increasing concentration of acaricide up to the point of 100% mortality. The percentage of mites lost in this manner ranged from practically none with the lowest concentrations of toxicant to 50% or more with the highest concentrations. Only the mites remaining on the leaf and not mired in the sticky barrier were counted.

In the topical applications the avocado leaf sections were held in place on the turntable by placing them in petri dish covers. The bean leaves were prepared for treatment by placing them on circular disks of wet paper toweling covering a 0.5-inch layer of wet absorbent cotton, also in Petri dish covers (Figure 4, upper). After treatment, the mites from the treated avocado or bean leaves were placed on the lower surfaces of untreated bean leaves lying on pads of wet cotton in Petri dish covers. The mites were allowed to remain in the laboratory room for 48 hours before mortality counts were made. The temperature in the laboratory ranged from 75° to 85° F., with a mean of about 80° F.

In the topical-residue tests, the mites were left on the leaf surfaces on which they were treated. The bean leaves were already lying on wet paper toweling on cotton when they were treated. A strip of sticky banding material was smeared around the edges of the avocado leaf sections before treatment, for there was a greater tendency for mites to crawl off the avocado leaf than the bean leaf. After treatment, the leaf sections were placed on wet cotton in Petri dish covers (Figure 4, lower). Counts were made 48 hours after treatment. In these tests the mites were affected by both the acaricide settling on their bodies and the residue on or in the leaf.

Figure 3. Equipment for generating sprays

A.B. Agitator blade
A.T. Air tube
B. Bearing
J. Jar
L. Screw-type lid of jar
L.T. Liquid tube
P. Pulley
S. Agitator shaft



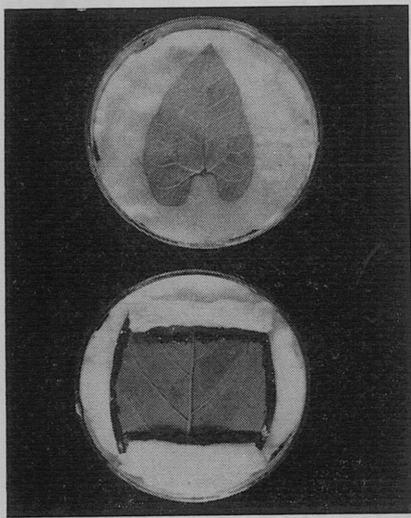


Figure 4. Method of confining mites after treatment and keeping leaf substrate fresh

Above. Lima bean leaf lying with upper surface on paper toweling resting on wet cotton
Below. Avocado leaf section bordered by Tanglefoot and lying with upper surface on wet cotton in Petri dish cover. Wet cotton roll covers each cut end

In the residue tests, the leaves were first treated, then the mites were placed on them. As in the other tests, 25 mites were placed on each of two leaves for each dosage tested. The mites were affected only by the residues over which they crawled on the leaf surfaces, and by toxicants within the leaf, which they ingested.

Criteria Used in Distinguishing Dead From Live Mites Mites killed by different acaricides may have a decidedly different appearance. For example, mites killed by Aramite or G-338 (recently given the name chlorobenzilate) retain a remarkably lifelike appearance and posture, whereas mites killed by the organic phosphates rapidly become dark and shriveled. Even before they die, and while they are still crawling about, they may be very shrunken and dark in appearance. Many mites, after they have been exposed to acaricide residues for 48 hours, lift their forelegs vertically above their bodies and stand or crawl about on three pairs of legs.

For the purpose of the experiments, mites were considered as "dead" if they no longer responded to prodding by crawling forward. Thus mites that were too moribund to crawl were considered dead even though they still showed unmistakable signs of life.

Acaricides Used in Experiments

Acaricide	*Chemical Name*
Aramite	2-(*p-tert*-Butylphenoxy) isopropyl-2′-chloroethyl sulfite
Arathane	Dinitrocapryl phenyl crotonate
876	Bis(*p*-chloropheyl) ethynylcarbinol
Sulphenone	*p*-Chlorophenylphenylsulfone and related materials
Ovotran	*p*-Chlorophenyl *p*-chlorobenzenesulfonate
G-338[a]	4,4′-Dichlorobenzilic acid ethyl ester
Dimite	4,4′-Dichloro-α-methylbenzhydrol
Parathion	*O,O*-Diethyl *O-p*-nitrophenyl thiophosphate
EPN	*O*-Ethyl *O-p*-nitrophenyl benzenethiophosphonate
Malathon	*S*-(1,2-Dicarbethoxyethyl) *O,O*-dimethyl dithiophosphate
Systox	*O*-[2-(Ethylmercapto)ethyl] *O,O*-diethyl thiophosphate

[a] Recently given the name chlorobenzilate.

Experiments with the Settling Tower

Effect of Leaf Substrate A sufficient number of tests were made to determine a dosage for each acaricide that would give between 0 and 100% mortality. Then seven of these acaricides were selected for application to bean and avocado leaves. Only the residual action of the acaricides was tested. Three avocado leaves and three bean leaves were treated in the settling tower simultaneously, so that an accurate comparison could be made between the two types of foliage. Twenty-five mites were placed on each leaf as described under the section on technique. The results of the experiment are shown in Table II. Aramite and Ovotran were used only as emulsions. (It was later discovered that the Aramite suspension was ineffective because of decomposition. For this reason, in this and some of the following experiments, data are presented for Aramite emulsion but not the suspension.) The remaining acaricides were used as emulsions and suspensions. With all the acaricides, in both formulations, there was always a higher mortality on bean leaves than on avocado leaves at a given concentration. The average mortality was 72.5% on bean leaves and only 13.5% on avocado leaves. The average mortality on avocado leaves was only 18.6% of the average on bean leaves.

The low "natural mortality" on the untreated leaves resulted primarily from molting mites. These do not respond to prodding and are consequently considered as dead according to the criterion established for the present series of experiments.

As the preliminary trials indicated such a great difference in the mortality from acaricide residues on avocado and bean leaves, in subsequent experiments all tests made with avocado leaves were repeated with bean leaves.

Dosage-Mortality Regressions Four acaricides (Sulphenone, Aramite, G-338, and parathion) were tested to determine the dosage-mortality regressions for each in two of its formulations, the emulsifiable concentrate and the wettable powder.

Table II. Effectiveness of Acaricide Residues on Avocado and Bean Leaves[a]

Acaricide	*Formulation*	*Concentration*	*Leaf*	*Mortality, %*
Aramite	Emulsion	0.01	Avocado	15.2
		0.01	Bean	95.5
Ovotran	Emulsion	3.0	Avocado	2.4
		3.0	Bean	75.2
Sulphenone	Emulsion	0.2	Avocado	2.6
		0.2	Bean	65.7
	Suspension	1.0	Avocado	22.8
		1.0	Bean	83.3
G-338	Emulsion	0.015	Avocado	18.9
		0.015	Bean	97.5
	Suspension	0.015	Avocado	7.8
		0.015	Bean	30.5
876	Emulsion	0.15	Avocado	14.6
		0.15	Bean	100.0
	Suspension	0.10	Avocado	14.9
		0.10	Bean	100.0
Arathane	Emulsion	0.05	Avocado	25.0
		0.05	Bean	43.1
	Suspension	0.10	Avocaco	2.7
		0.10	Bean	63.0
EPN	Emulsion	0.005	Avocado	20.0
		0.005	Bean	38.5
	Suspension	0.01	Avocado	14.3
		0.01	Bean	77.8
		Average	Avocado	13.5
			Bean	72.5
		Check	Avocado	3.3
			Bean	2.9

[a] Treated with settling tower under conditions in which a concentration of 1% of toxicant would result in a deposit of 14.34 γ/sq. cm. on treated leaves. Twenty-five adult female mites were placed on each of three treated leaves and counts were made 48 hours later.

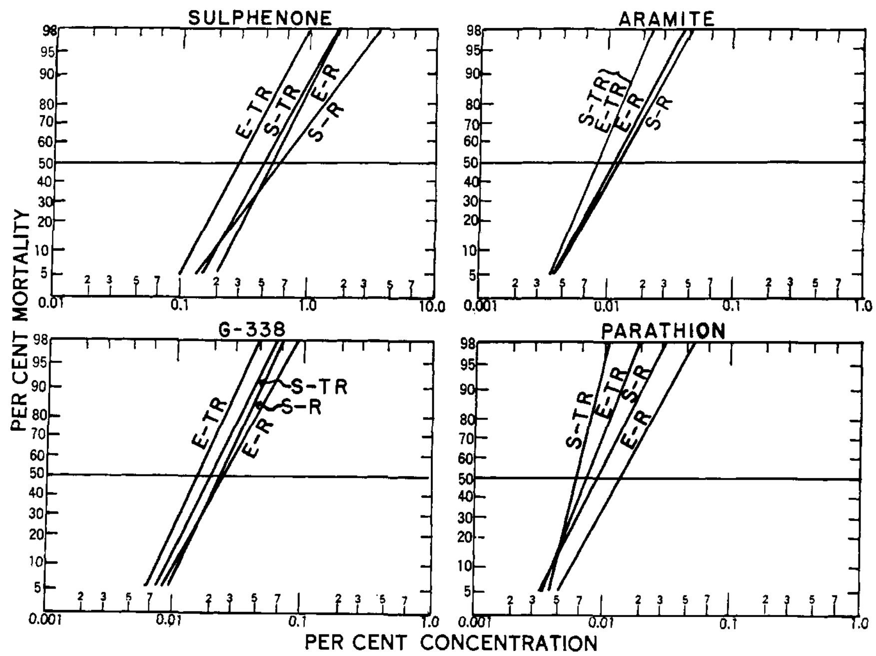


Figure 5. Dosage mortality regressions for adult two-spotted mites on avocado leaves treated with four acaricides, using settling tower

E. Emulsion
S. Suspension

T. Topical
R. Residue
TR. Topical-residue

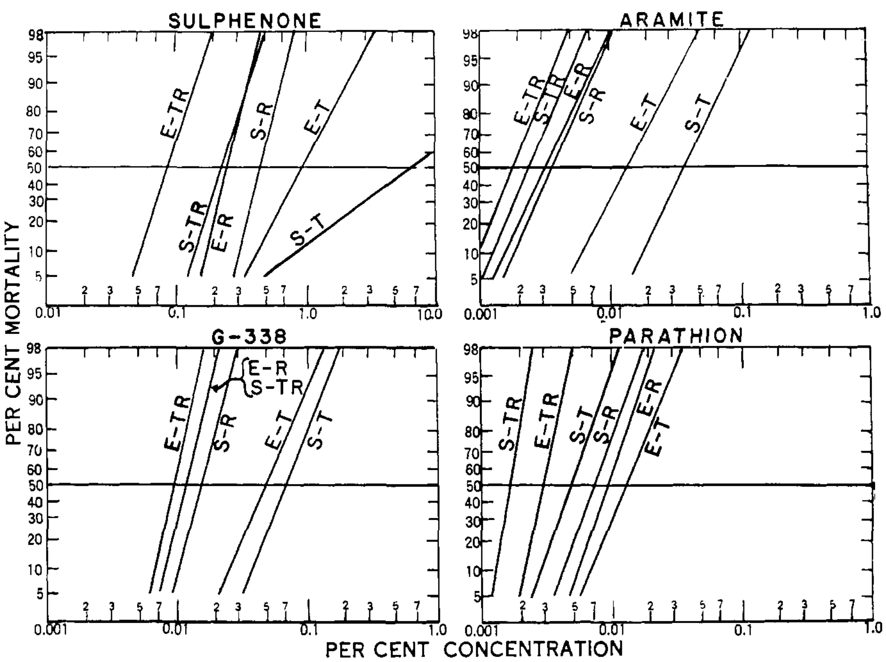


Figure 6. Dosage mortality regressions for adult two-spotted mites on bean leaves treated with four acaricides, using settling tower

E. Emulsion
S. Suspension

T. Topical
R. Residue
TR. Topical-residue

Well-known proprietary brands of each acaricide were used, the two formulations being supplied by the same manufacturer.

The results of the experiments are presented in Figures 5, 6, and 7.

It did not appear practical to select mites of a uniform adult age. Nevertheless the steepness of the regression lines indicates a remarkable degree of homogeneity among the mites with regard to their response to the various acaricides. In fact, the slopes of the regression lines are greater than those obtained by Ebeling (*2*) when testing insecticides against adult fruit flies (Tephritidae) all of the same adult age.

The LD_{50}'s for each of the treatments indicated in Figures 5, 6, and 7 are tabulated in Table III. Parathion differed widely from the other acaricides with respect to the relative LD_{50}'s from the settling tower and the sprayer. With parathion, when the sprayer was used, the LD_{50}'s with the emulsions and suspensions were, respectively, only 9 and 5% as high as when the settling tower was used. In other words, parathion, as a residue treatment, was improved far more by spraying, as compared to using a settling mist, than were the other acaricides included in the tests.

In order to compare the acaricide-depositing properties of field sprays with deposits obtained with the present laboratory equipment, Aramite and parathion in the two formulations were applied to orange trees by means of a power sprayer, using a pressure of 600 pounds and 9/64-inch disk orifices in the spray guns. Chemical residue analyses showed that Aramite suspension deposited 4.6 times more toxicant and the emulsion 2.8 times more toxicant than in the settling tower at equivalent concentrations of toxicant (analyses made by Francis A. Gunther, University of California Citrus Experiment Station, Riverside, Calif.). The parathion suspension residues from the field spraying were 3.5 times greater in quantity than those from the settling tower and the parathion emulsion residues were 2.4 times greater.

Comparison of Formulations It can be seen from Table III that with Aramite, Sulphenone, and G-338, the suspensions were generally less effective, at a given concentration, than the emulsions, regardless of the type of treatment (topical, residue, or topical-residue). Using the data obtained with the bean leaves, when the average LD_{50}'s for emulsions were divided by the average LD_{50}'s for suspensions, the following figures were obtained for the tests with the settling tower with the three types of treatment: *T*, 0.45; *R*, 0.73; and *TR*, 0.63. The corresponding figures for the tests with the sprayer were: *T*, 0.53; *R*, 0.55; and *TR*, 0.38. Thus, with both settling tower and sprayer, and with all three types of treatment, the emulsions were superior to the suspensions. On the other hand, with parathion the suspensions were consistently superior to the emulsions, regardless of the type of treatment.

Comparison of Leaf Substrate The residue and topical-residue treatments were consistently more effective on bean leaves than on avocado leaves. With the residue treatments, on the average only 47.8% as much toxicant was required for 50% mortality of adult mites on the bean leaves with emulsions and 62.8% as much with suspensions (Table IV). It is believed that the greater effectiveness of the acaricides when applied to bean leaves is due to the fact that they penetrate the leaf cuticle more readily. There they act as stomach poisons and contribute to some extent to the mortality. In addition, on bean leaves the mites feed more and consequently consume more of the subcuticular residues.

Comparison of Types Of Treatment With a given formulation the residue treatments were consistently superior to the topical treatments, a rather anomalous situation in view of the fact that the mites do not move about much after the initial excitement incident to transfer from the source leaves. Much of the time they are off the leaf surface on their extensive webbing. In a previous investigation, Ebeling (*2*) showed that the effectiveness of insecticide residue is proportional to the degree of activity of the insects that crawl about on it. With the adult oriental fruit fly, *Dacus dorsalis* Hendel, an insect that crawls about actively, the residue treatments with the same settling tower as used in the present experiments required on the average about five times as much toxicant as the topical applications to effect a 50% kill. Yet in the present experiments the reverse was true; 2.8 times as much toxicant was required in the topical as in the residue treatments when emulsions were used and 1.9 times as much was required when suspensions were used (Table V).

Long-Term Effectiveness of Spray Residues in the Field

On January 12, 1953, avocado trees were sprayed with 7 acaricides at a concentration of 0.12% of toxicant (1 pound to 100 gallons). Each acaricide was used as a suspension and as an emulsion. Two leaves were picked from the south sides of the sprayed trees immediately after spraying, and every day thereafter. Twenty-five adult two-spotted mites were placed on the lower surfaces of leaf sections placed on wet cotton in Petri dishes and allowed to remain in the laboratory for 48 hours, before mortality counts were made. This procedure was continued until the residues no longer resulted in a 50% kill of the mites.

From a hygrothermograph set up in a shelter in the treated block of trees the following data on the temperature and relative humidity during the period of the experiment were obtained.

Period in Experiment[a]	Mean Max. Temp., ° F.	Mean Min. Temp., ° F.	Mean Max. Rel. Humidity	Mean Min. Rel. Humidity
First week	61.8	50.3	62.4	37.5
Second week	70.0	50.8	65.8	35.4
Third week	77.4	56.6	32.6	10.2
Fourth week	72.0	53.7	57.5	22.0

[a] Experiment begun January 12, 1953.

The results of the experiment are shown in Figure 8. There was a great variation in the performance of the acaricides, ranging from practically no effect with Sulphenone emulsion to a residual effectiveness of 32 days with the EPN suspension.

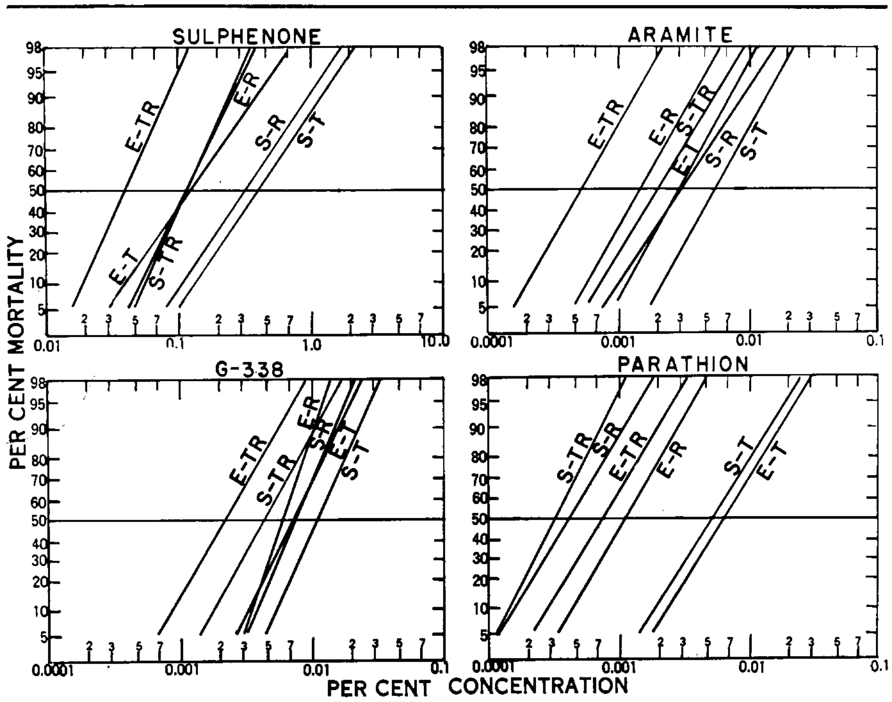
If a population of mites were existing on the treated trees, they would be in contact with the residues for the remainder of their lives rather than for only 48 hours as in these tests. Consequently, the residual effectiveness of the acaricides against adult mites is probably longer than indicated in Figure 8.

The suspension resulted in a more prolonged residual effectiveness than the emulsion with every acaricide. With G-338, however, there was little difference between the two formulations and in a subsequent experiment, with foliage taken from the north side of the tree, the emulsion remained effective longer. Not including Sulphenone, with which the emulsion resulted in less than 100% initial mortality, the suspensions remained effective, on the average, 52.7% longer than the emulsions.

The above experiment was made only for the purpose of comparing the emulsifiables and wettable powders of a representative group of acaricides with regard to their long-term residual effectiveness against adult mites. The ovicidal properties of the acaricides, as well as their effect against active stages other than the adult, are important factors concerning which no tests have yet been made, but for which experiments are being planned. An earlier test showed that Dimite and Systox were among the most effective of the acaricides from the standpoint of long-term residual effectiveness, but they were not included in the above test because they are not available as wettable powders.

Figure 7. Dosage mortality regressions for adult two-spotted mites on avocado leaves treated with four acaricides, using laboratory sprayer

E. Emulsion
S. Suspension
T. Topical
R. Residue
TR. Topical-residue



Subcuticular Acaricide Residues As Stomach Poisons

Mortality of Adult Mites after Removal Of Surface Residues In the experiment shown in Figure 8, 9 days after spraying two leaves were picked from the south

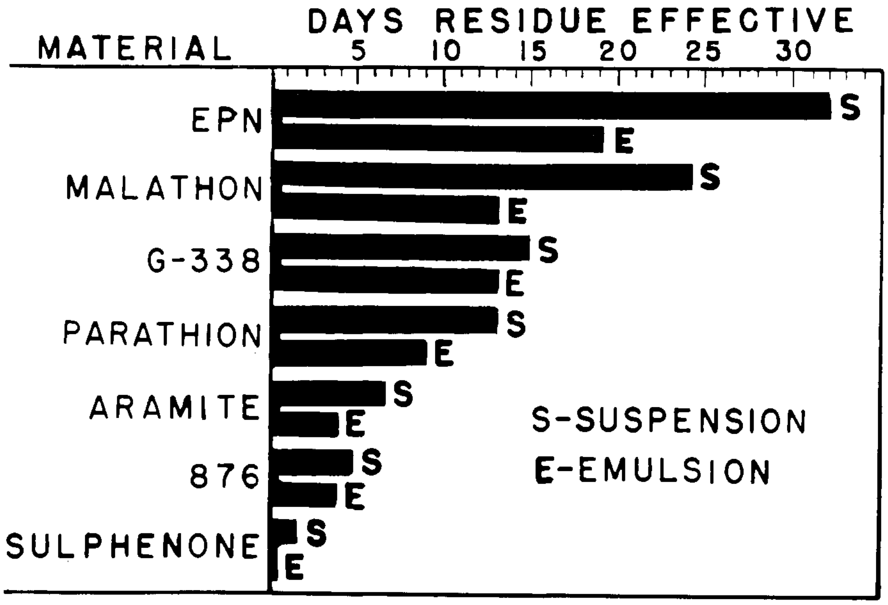


Figure 8. Differences in periods residues of seven acaricides remained effective against adult two-spotted mites

E. Emulsions
S. Suspensions

sides of trees sprayed with emulsions and suspensions of EPN, malathon, parathion, and G-338. The leaves were washed with a detergent solution and a rubber sponge, then rinsed and rubbed with an uncontaminated sponge under running water. This washing treatment was far in excess of that which was required to remove insecticide residues from foliage so as to render it completely ineffective against fruit flies (Tephritidae). The latter are much more sensitive to certain toxicants used in the present experiment, such as parathion, malathon, and EPN, than are the two-spotted mites.

After the washed leaves were dry, they were cut into sections that would fit into Petri dish uppers and mites were placed on the lower surfaces of the leaves and confined with sticky banding material as described before. Counts were made in 48 hours. A 100% kill was obtained on all leaves except those sprayed with malathon emulsion (56.3%) and G-338 suspension (95.8%). With the latter acaricides, however, the unwashed leaves also no longer resulted in 50% mortality 4 and 6 days later, respectively (see Figure 8). Unwashed leaves of parathion emulsion no longer resulted in 50% kill even 2 days after the washed leaves became ineffective. Subsequent tests showed that in general the unwashed leaves resulted in better kills than the washed leaves during the last few days of the life of the residue, showing that the surface residues were still active. However, during the greater part of the period of residual effectiveness, adult mites were killed in 48 hours of contact with the leaves, even when the residues were completely removed from the surface. This indicated that the toxicant that had penetrated through the leaf cuticle was present in sufficient quantity to act effectively as a stomach poison for prolonged periods under field conditions.

It was realized that the toxicant might be continuously penetrating into the leaf as long as surface residues remained in their original chemical composition. The question arose as to whether the sprayed leaves would remain toxic to mites for prolonged periods even if the residues were removed soon after spraying. In another experiment individual limbs on the north sides of avocado trees were sprayed with 0.12% of actual toxicant, but using a knapsack sprayer for application. G-338 and parathion as emulsions and suspensions were the only acaricides applied in this experiment. Both surfaces of all the leaves were thoroughly sprayed. In 24 hours all the leaves on certain branches were washed with detergent solution and rub-

Figure 9. Differences in periods washed and unwashed avocado leaves remained effective in killing adult two-spotted mite after leaves sprayed with 0.12% of toxicant

Washing 24 hours after treatment
Counts made separately on upper, *U*, and lower, *L*, surfaces of leaves

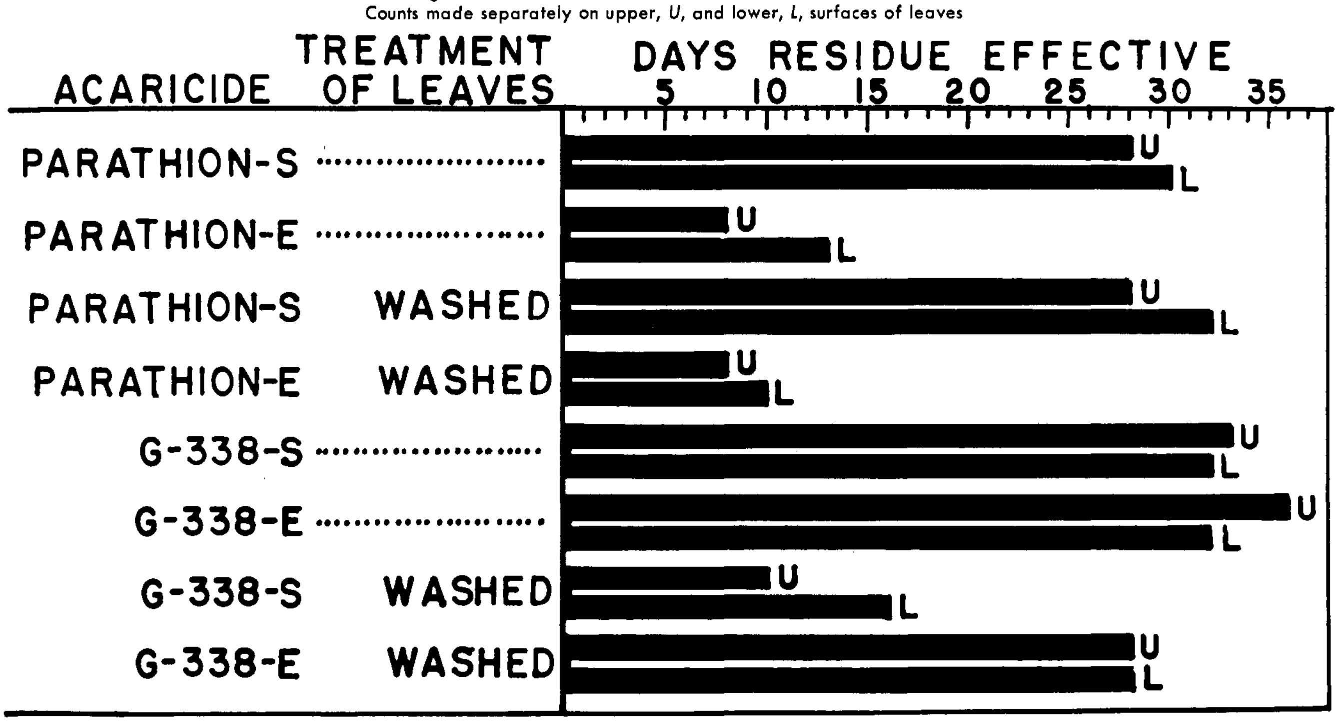


Table III. LD_{50}'s for Four Acarides Applied as Emulsions and Suspensions by Settling Tower and Laboratory Spray Equipment

Equipment	Substrate leaf	Formulation	Type treatment[a]	LD_{50}[b] (Sulphenone)	Equipment	Substrate leaf	Formulation	Type treatment	LD_{50} (Aramite)
Settling tower	Bean	Emulsion	T	0.93	Settling tower	Bean	Emulsion	T	0.014
			R	0.25				R	0.0031
			TR	0.085				TR	0.0018
		Suspension	T	5.40			Suspension	T	0.0380
			R	0.45				R	0.0035
			TR	0.26				TR	0.0023
	Avocado	Emulsion	R	0.54		Avocado	Emulsion	R	0.0120
			TR	0.29				TR	0.0089
		Suspension	R	0.60			Suspension	R	0.0140
			TR	0.48				TR	0.0088
Sprayer	Avocado	Emulsion	T	0.12	Sprayer	Avocado	Emulsion	T	0.0031
			R	0.11				R	0.0015
			TR	0.037				TR	0.0006
		Suspension	T	0.32			Suspension	T	0.0056
			R	0.28				R	0.0033
			TR	0.11				TR	0.0020

Equipment	Substrate leaf	Formulation	Type treatment	LD_{50} (G-338)	Equipment	Substrate leaf	Formulation	Type treatment	LD_{50} (Parathion)
Settling tower	Bean	Emulsion	T	0.059	Settling tower	Bean	Emulsion	T	0.013
			R	0.012				R	0.0095
			TR	0.0097				TR	0.0030
		Suspension	T	0.072			Suspension	T	0.0048
			R	0.016				R	0.0072
			TR	0.012				TR	0.0017
	Avocado	Emulsion	R	0.026		Avocado	Emulsion	R	0.013
			TR	0.017				TR	0.0075
		Suspension	R	0.024			Suspension	R	0.0081
			TR	0.020				TR	0.0061
Sprayer	Avocado	Emulsion	T	0.0070	Sprayer	Avocado	Emulsion	T	0.0068
			R	0.0060				R	0.0012
			TR	0.0021				TR	0.00076
		Suspension	T	0.0112			Suspension	T	0.0051
			R	0.0074				R	0.00040
			TR	0.0042				TR	0.00031

[a] For topical application, *T*, mites were transferred to untreated leaves after treatment; for residue treatment, *R*, they were placed on treated leaves; and for topical-residue treatment, *TR*, mites were allowed to remain on treated leaves.
[b] Grams of toxicant per 100 ml. of spray mixture.

ber sponge, then thoroughly rinsed several times with clean water and an uncontaminated sponge. Other leaves were left unwashed.

Every day four leaves were picked from each washed and each unwashed branch. Twenty-five mites were confined on the upper surfaces of two leaves and on the lower surfaces of the others. The leaves were then kept in the laboratory. The results of the experiment are shown in Figure 9. It will be noted that G-338 in both formulations and parathion as a suspension remained effective much longer than in the previous field experiment (Figure 8). However, the leaves in the present experiment were taken from the north sides of the trees where they were in shade throughout the day.

With G-338 there was no significant difference in residual effectiveness between the suspension and the emulsion as far as the unwashed leaves were concerned, although on the upper leaf surface there was a numerical difference of 3 days in favor of the emulsion. With the emulsion, the washed leaves remained effective 28 days, 82.4% as long as the unwashed leaves. With the suspension, the washed leaves remained effective 13 days (average of upper and lower leaf surfaces), only 40.0% as long as the unwashed leaves. This indicates that the residue deposited by the suspension did not penetrate the leaf cuticle, in the 24 hours prior to washing, in as great a quantity as did the residue deposited by the emulsion.

With parathion, when the data for the upper and lower leaf surfaces were averaged, the leaves sprayed with emulsion remained effective only 34.4% as long as those sprayed with suspension. There was no significant difference in the residual effectiveness of the washed and unwashed leaves on the branches sprayed with the suspension, although there was a numerical difference of 2 days in favor of the unwashed leaves on the lower leaf surfaces. This difference is ascribed to chance variation. This would seem to indicate, at least in this particular test, that the surface residues lost their effectiveness as soon as, if not sooner, than the subcuticular residues. With the parathion emulsion there was no difference in the washed and unwashed leaves when the upper surfaces were compared, but with the lower surfaces there was a difference of 3 days in favor of the unwashed leaves.

The data depicted in Figure 9 show that subcuticular acaricide residues have a prolonged residual effectiveness. With G-338 emulsion, they resulted in at least a 50% kill of adult mites for 28 days on both the upper and lower leaf surfaces. With parathion suspension they resulted in at least a 50% kill for 28 days on the upper leaf surfaces and 32 days on the lower leaf surfaces.

An experiment was made with the settling tower, so as to ensure a uniform deposit of toxicant with different acaricides, using parathion, G-338, and Aramite as emulsions and suspensions. Two upper surfaces and two lower surfaces were treated with each dosage. After the residues had remained on the leaves for an hour, they were washed off with detergent solution as described before. Mites were then confined on the leaves for 48 hours. Another group of leaves was treated as before, but as soon as they were dry they were stuck into wet sand so that about an inch of the basal cut edge was buried in the sand. The leaves were kept fresh in this manner for 24 hours, then the basal inch was cut off and the leaves were washed with detergent solution and rinsed. Mites were confined on the leaves for 48 hours as in the previous test. The results of the experiment are shown in Table VI.

Table VI shows that even in 1 hour sufficient toxicant had penetrated through the leaf cuticle to result in considerable toxicity to mites confined on the foliage after the residues were washed off. Taking into consideration the fact that the 5-ml. charge of spray, as used in the settling tower, deposited only about a third as much toxicant as that which is deposited in ordinary field spraying with a power sprayer, the amount of toxicant required to result in an effective subcuticular residue can be seen to be well within the limits of general commercial usage. The subcuticular residues of parathion killed adult greenhouse thrips, *Heliothrips haemorrhoidalis* Bouché, as well as the mites.

It can be calculated from the data presented in Table VI that the average mortality of mites confined with leaves washed 1 hour after treatment was 73.3% for the upper surfaces and 67.4% for the lower surfaces, an insignificant difference. When the leaves were not washed until 24 hours after treatment, the upper surfaces of the leaves were more toxic, at the same concentrations of toxicant, and the lower surfaces were less toxic than for the leaves washed 1 hour after treatment. It is difficult to conceive how the lower surfaces could become less toxic when the residues are allowed to remain longer before removal. This phenomenon seemed to be influenced by the fact that the leaves were picked off the tree. When the leaves were sprayed on the tree, washed in 24 hours, and allowed to remain on the tree, the lower leaf surfaces usually remained toxic longer than the upper surfaces (see Figure 9).

A striking feature regarding the data shown in Table VI is the outstanding toxicity of the upper surfaces of the leaves sprayed with Aramite emulsion and washed 24 hours after treatment. A 100% mortality was obtained at a 0.1% concentration of toxicant, equivalent to only 0.83 pound of toxicant to 100 gallons. Only about a third as much toxicant is deposited in the settling tower as would be deposited in field spraying with a power sprayer. On the other hand, the Aramite suspension was the least effective of the three suspensions tested. Possibly the large particle size of the wettable powder causes it to be less effective than the emulsion in penetrating the cuticle. The great proportion of diluent (85%) may have a "masking effect" and may prevent the maximum contact of toxicant with the leaf surface.

Mortality of Adult Mites on Surface Of Leaf Opposite One Treated

In the treatment of certain crops, such as beans and cotton, the spray is directed downward, striking only the tops of the leaves. Nevertheless, a satisfactory mortality of spider mites is obtained on the lower as well as the upper surfaces. Various opinions have been offered as to the reason for this phenomenon: that a spray directed downward has a "boiling effect," resulting in the deposit of considerable amounts of toxicant on the lower surfaces of the leaves; that the mites on the lower surfaces move about and eventually crawl over the upper surface and come in contact with the main acaricide deposits; and that with some acaricides the mites on the lower surfaces succumb to toxic vapors.

The late W. E. Blauvelt informed the writers in October 1952 that when he covered the upper surfaces of bean leaves and directed a spray downward on the plants, the two-spotted mites on the lower surfaces were not killed by various acaricides that normally result in a complete kill on both surfaces. Luminous substance was added to the spray and none could be detected on the lower surface. He concluded that acaricides usually penetrate from the upper to the lower surface of the leaf and exert their toxic effect in that way. The larvae of various leaf-mining Diptera were killed by the penetration of parathion from dilute emulsions through the leaf tissue (*5*). Likewise the larvae of the agromyzid leaf miner, *Liriomyza langei* Frick, in aster leaves were killed by penetration of parathion and EPN applied as dusts. A 1% parathion dust was found to be superior to a 2% EPN dust for this purpose. Parathion wettable powder was, of course, also effective (*4*).

To test the translocation of acaricides in bean leaves, the leaves were placed with either their upper or their lower surfaces on damp cotton pads. They adhered firmly to the pads, with no intervening space. The leaves were treated with emulsions and suspensions in the settling tower, using 0.12% of actual toxicant. Adult two-spotted mites were placed on surfaces opposite to those treated and were allowed to remain 48 hours before they were examined. Parathion emulsion and suspension resulted in 100% mortality when applied to the lower sides of the leaves but only 77.8 and 73.9%, respectively, when applied to the upper surface. Aramite emulsion resulted in a 100% kill when applied to the lower surface, but only 4.2% when applied to the upper surface.

As the settling tower deposits only about a third as much toxicant as the average spray application, the experiment was repeated with the McCarl fog maker, applying the spray directly against the surface to be treated. Six successive blasts of spray were directed against each leaf. The opposite surface received no visible trace of acaricide. Again a 0.12% concentration of toxicant was used, except that only 0.03% of parathion (actual toxicant) was used on bean leaves, because preliminary tests showed parathion to be much more effective than the other acaricides in penetrating into the bean leaf.

Three varieties of leaves were used: bean, grapefruit, and avocado. With the bean and grapefruit, two leaves were left on stems that were kept in water for 24 hours after treatment. Then adult mites were placed on the surfaces of the leaves opposite to those that were sprayed. With the bean cuttings one leaf was allowed to remain on the stem and the other was placed

Table IV. Residual Effectiveness of Four Acaricides Applied to Bean and Avocado Leaves in Settling Tower

Acaricide	Formulation	LD_{50} on Bean	LD_{50} on Avocado	Bean / Avocado
Sulphenone	Emulsion	0.25	0.54	46.3
	Suspension	0.45	0.60	75.0
Aramite	Emulsion	0.0031	0.012	25.8
	Suspension	0.0035	0.014	25.0
G-338	Emulsion	0.012	0.026	46.2
	Suspension	0.016	0.024	62.5
Parathion	Emulsion	0.0095	0.013	73.1
	Suspension	0.0072	0.0081	88.9
			Average, emulsion	47.8
			Average, suspension	62.8

on wet cotton, although this appeared to cause no significant difference in results. With the grapefruit cuttings the leaves were cut into sections of appropriate size and placed on wet cotton, with wet cotton rolls on the cut ends. The avocado leaf sections were placed in wet sand, in the manner previously described, for 24 hours. Then they were placed on wet cotton and the test mites were transferred to them. The margins of the grapefruit and avocado leaves were bordered with sticky band material to prevent escape of the mites, and the margins of the bean leaves left on the cuttings were also bordered with sticky banding material to prevent mites from crawling from one surface to the other. The mites were examined in 48 hours and again in 96 hours. The results of the experiment are shown in Table VII.

Table VII shows that with bean leaves 100% mortalities of adult mites were obtained in either 48 or 96 hours with all three acaricides when the spray was applied to the lower surface and the mites were confined on the upper surface. When the spray was applied to the upper surface, its effectiveness on the opposite surface was not so great. Nevertheless in 96 hours the mortalities ranged from 95.4 to 98.2%. As there was a great increase in mortality from the 48-hour to the 96-hour interval, it is likely that if the mites were allowed to remain for an even greater period they would all succumb. With grapefruit leaves the toxicant penetrating through the leaf was not so effective as with bean leaves, but in 96 hours 100% kills were obtained on both surfaces of the leaf with parathion and on one surface with G-338. With avocado leaves only parathion, applied to the lower surfaces of avocado leaves, resulted in 100% mortality of mites on the opposite surfaces.

Table VII shows that Aramite was the least effective of the three acaricides used in killing mites placed on the surfaces opposite to those that were sprayed. This is in sharp contrast to its superiority as a subcuticular poison when it is applied to a leaf surface and washed off in 24 hours (see Table VI).

The effectiveness of the acaricides in causing a mortality of mites on the surfaces of the leaves opposite to those that were sprayed was greatest on bean and least on avocado. This is what would be expected on the basis of the relative thickness of the three varieties of foliage.

The question may arise as to whether the death of the mites on the leaf surface opposite to that which is sprayed is caused by contact with residues that may have issued to the surface from inside the leaf or to subcuticular residues ingested by the mites in the course of their feeding. The writers are inclined to the latter view, for 100% mortalities can be obtained even after both the sprayed surface and the one opposite the sprayed surface are thoroughly washed, and even after repeated washings of the same leaf on successive days.

In one instance, after both surfaces were washed, polyvinyl alcohol was applied to the surface opposite to the one sprayed. This results in a thin artificial "cuticle" of high polarity that would be expected to be nonpermeable to the lipoid-soluble acaricides and yet prevent contact of the tarsi of the mites with any toxicant that might be present on the plant cuticle. On a suitable untreated host, mites can feed actively and survive and reproduce indefinitely in a normal way when the leaf is covered with a membrane of polyvinyl alcohol. Mites were placed on this artificial cuticle and all succumbed to the acaricide. Likewise, polyvinyl alcohol membranes on sprayed leaf surfaces from which the residues were removed did not prevent the death of mites feeding through the membranes.

Discussion

The findings reported in this paper are particularly significant in relation to a previous investigation of similar nature made by Ebeling (*2*) with regard to three species of fruit flies (Tephritidae). These insects obtained no food on or in the substrate on which the insecticides were deposited. Food and water were supplied by means of a cotton wick, saturated with sugar solution, which extended into the cages containing the residues. With the melon flies, contact of the tarsi with the surface residues of insecticide is the sole means of their obtaining any toxicant on or in their bodies. The factors that influence the effectiveness of insecticide residues against insects that do not feed on their substrate are those that affect (1) the period of contact of the insect with the residue and (2) the activity of the insect while in contact. The longer the period of contact, and the greater the activity of the insect while on the residue, the greater the mortality.

Table V. Average Quantity of Toxicant Required for 50% Mortality of Adult Two-Spotted Mites

Application Equipment	Formulation	Type of Treatment	Relative LD_{50}'s (Topical = 100)
Settling tower	Emulsion	Topical	100.0
		Residue	35.6
		Topical-residue	15.4
	Suspension	Topical	100.0
		Residue	53.5
		Topical-residue	19.9
Sprayer	Emulsion	Topical	100.0
		Residue	47.3
		Topical-residue	15.8
	Suspension	Topical	100.0
		Residue	64.7
		Topical-residue	34.0

Table VI. Effectiveness of Acaricides as Emulsions in Killing Adult Mites Placed on Surface of Leaf Opposite That Sprayed[a]

Acaricide	% Toxicant	Leaf Substrate	Leaf Surface Sprayed	Net % Mortality In 48 hours	Net % Mortality In 96 hours
Parathion	0.03	Bean	Upper	91.5	95.4
	0.03	Bean	Lower	100.0	100.0
G-338	0.12	Bean	Upper	76.0	95.9
	0.12	Bean	Lower	100.0	100.0
Aramite	0.12	Bean	Upper	49.4	98.2
	0.12	Bean	Lower	87.5	100.0
Parathion	0.12	Grapefruit	Upper	77.0	100.0
	0.12	Grapefruit	Lower	96.7	100.0
G-338	0.12	Grapefruit	Upper	24.8	45.0
	0.12	Grapefruit	Lower	96.4	100.0
Aramite	0.12	Grapefruit	Upper	13.1	34.2
	0.12	Grapefruit	Lower	60.8	96.1
Parathion	0.12	Avocado	Upper	52.8	82.5
	0.12	Avocado	Lower	85.3	100.0
G-338	0.12	Avocado	Upper	0.0	60.8
	0.12	Avocado	Lower	5.4	70.7
Aramite	0.12	Avocado	Upper	0.0	26.0
	0.12	Avocado	Lower	0.0	55.3

[a] Leaves sprayed in uniform manner with McCarl fog maker. When only one side of leaf was treated, opposite side was left completely unwet by spray.

The factors mentioned above as affecting the effectiveness of insecticide residues against insects that do not feed on the treated substrate no doubt also apply to insects that do. However, the sucking phytophagous species, in addition to making contact with the surface residues, also are able to obtain the toxicant by sucking up the contaminated sap beneath the plant cuticle. That is probably the reason why less toxicant is required to kill mites, over a 48-hour period, by residual action than by topical application. This is in direct contrast to the experience with adult melon flies, insects that do not feed on the treated substrate. This increases the complexity of any systematic investigation of the factors involved in the residual action of acaricides.

Two-spotted mites were less active on bean leaves than on avocado leaves, for on the latter they spent more of their time searching for a new source of food. With fruit flies any such increase in activity would result in increased mortality, but with the mites the mortality was actually much lower on the avocado leaves. This is ascribed to the greater ease of penetration of residues into the bean leaf (as shown by the penetration tests by bioassay), resulting in a greater stomach-poison effect from the subcuticular residues. In addition, observation of the mites shows that they do more feeding on bean leaves than on avocado leaves, which would result in the ingestion of a greater quantity of toxicant.

Two-spotted mites can be kept alive for 48 hours on thin waxed paper, such as used for wrapping lunches, if the paper is floated on water. Apparently the mites are able to obtain water by piercing the waxed paper with their mouth parts. One might expect that residues on waxed paper would be especially effective, for the mites move about actively in search of a more suitable substrate. The mites cannot get their webbing off the surface of their substrate, for no midrib, veins, or irregularities, such as exist on a leaf, are present. Consequently they cannot crawl about on webbing that is separated from the surface residues and are thus in continuous contact with them. Nevertheless the LD_{50}'s on floating waxed paper disks averaged 3.3-fold greater than on bean leaves with the three formulations (Aramite emulsion and G-338 emulsion and suspension) that were tested when the two substrata were treated simultaneously in the settling tower and adult mites were placed on the residues. This difference in mortality is ascribed to the poisoning effect of ingested sap when the mites were placed on the bean leaves. These mites were subjected to both a contact and a poison effect of the acaricides, while those on waxed paper were subjected to contact action only.

Since it requires a much higher concentration of toxicant to result in a mortality of mites when the residues are removed, the question may arise as to how the subcuticular residues could account for the difference in the mortality of the mites on bean and avocado foliage at the low concentration of toxicant indicated in Table III. However, when the residues are left on the leaf surface they act as a continuous reservoir of toxicant for the tissues just below the cuticle. A low level of acaricide might keep these subcuticular tissues continuously supplied with a lethal dose of toxicant, whereas if the surface residues were removed, this minute quantity would become dispersed away from the area that can be reached by the very short stylets of the mites, and could not be replenished. It so happens that the concentration of toxicant required for adequate long-term control of mites under field conditions also results in a sufficiently high level of subcuticular toxicant to ensure a prolonged residual acaricidal effectiveness of foliage from which the surface residues have been removed. It is also sufficient to cause the mortality of mites on the leaf surface opposite to that which is sprayed on some plants on which ordinarily only one surface is treated.

The comparatively high degree of effectiveness of parathion in killing mites located on the surface of the leaf opposite to that which is treated is to be expected in view of the fact that parathion can be carried with moisture ascending the stems of bean cuttings to some extent. Cuttings of Henderson lima beans in the two-leaf stage, and with the petioles of the leaves banded with sticky banding material, were placed in 8-ounce bottles containing parathion and malathon suspensions with 0.12% of toxicant. Presumably the quantity of toxicant available to the cuttings was limited by the solubility of the toxicant in the water (20 p.p.m. for parathion and 145 p.p.m. for malathon).

In each bottle the stem of the cutting was packed with cotton in the mouth of the bottle and the opening was sealed with masking tape. Two-spotted mites were placed on the leaves and examined in 48 hours. Parathion resulted in only 30.2% mortality of mites, but malathon, being over seven times as water-soluble, resulted in 99.5% mortality. The experiment was repeated with emulsifiable concentrate with approximately the same results. EPN, Aramite, and G-338 were ineffective. It is important to keep the mouth of the bottle completely sealed, for the vapors of parathion and malathon result in

Table VII. Mortality of Mites Placed on Upper and Lower Surfaces of Avocado Leaves from Which Residues Were Removed after Treatment in Settling Tower

Acaricide	% Toxicant	Interval Between Treatment and Washing, Hours	Surface of Leaf	% Mortality
Parathion	0.2	1	Upper	69.8
(emulsion)	0.2	1	Lower	97.2
	0.2	24	Upper	100.0
	0.2	24	Lower	98.0
Parathion	0.2	1	Upper	98.3
(suspension)	0.2	1	Lower	100.0
	0.2	24	Upper	100.0
	0.2	24	Lower	95.5
G-338	0.3	1	Upper	88.7
(emulsion)	0.3	1	Lower	61.6
	0.3	24	Upper	100.0
	0.3	24	Lower	23.4
G-338	0.3	1	Upper	83.3
(suspension)	0.3	1	Lower	45.6
	0.2	24	Upper	100.0
	0.2	24	Lower	17.5
Aramite	0.2	1	Upper	100.0
(emulsion)	0.2	1	Lower	100.0
	0.1	24	Upper	100.0
	0.1	24	Lower	0.0
Aramite	0.2	1	Upper	0.0
(suspension)	0.2	1	Lower	0.0
	0.2	24	Upper	0.0
	0.2	24	Lower	0.0

complete kills in 48 hours at extremely low concentrations, under the conditions of the above experiment, if the bottles are not sealed.

The writers consider that the ability of acaricides to kill mites by penetrating the leaf cuticle and acting as stomach poisons is of considerable significance in relation to practical control measures. It is likely that translocation of the subcuticular residues in large measure accounts for the control of mites on the lower surfaces of the leaves of truck crops and such crops as cotton when the spray is directed downward against the upper surfaces of the leaves and little, if any, spray material gets on the lower surfaces. If this were not the case, control programs would be greatly complicated with these types of crops.

Subcuticular residues should also be of benefit in controlling sucking insects or mites in areas of frequent rain. Here again a comparison with the situation that obtains with insects that do not feed on their substrate is of interest. Insecticide residues on corn foliage, used as a trap crop, were rendered ineffective against the melon fly, *Dacus cucurbitae* (Coquilett), by a single shower of rain. A rainfall of 0.15 inch destroyed the residual effectiveness of a heavy deposit of parathion by washing off the residue (*3*). Yet a 100% mortality of mites may be obtained after a complete removal of residues from treated foliage. Parathion was the most affected by rain of all the insecticides used in fruit fly control, yet in the present experiments it was the least affected by washing, owing to its ability to penetrate the cuticle in greater quantity than the other acaricides. Although surface residues were easily removed, internal residues continued to kill the mites for extended periods.

Because mite control depends so greatly on prolonged residual effectiveness, it is probably fortunate that the subcuticular residues are present in sufficient quantity to be lethal. This would be particularly true under circumstances in which frequent rains or other weathering agencies result in rapid losses of surface residue, or in which it is not practicable to treat more than one surface of the leaves of the crop to be protected.

In the investigation of formulations, the comparison of the reactions of mites with those of other arthropods is also of interest. With 13 insecticides used against fruit flies, the LD_{50}'s averaged lower for emulsions than for suspensions in topical application, but the reverse was true when the flies were placed on freshly deposited residues for 24 hours. It was believed that the flies were able to pick up larger quantities of insecticide when crawling over the discrete particles deposited by suspensions than when crawling over the smooth film of residue deposited by emulsions (*2*). In contrast, if the emulsions were more effective against two-spotted mites in topical treatment, they were also more effective as freshly deposited residues.

Regardless of their relative merits, as compared to emulsions, in topical treatment or as freshly deposited residues, the suspensions were superior to the emulsions in long-term residual effectiveness against adult mites under orchard conditions, except with G-338. With the latter, the suspension and emulsion appeared to be approximately equal in long-term residual effectiveness.

With the exception of parathion, the suspensions were less effective than the emulsions, when they were removed 1 hour or 24 hours after treatment (Table VII and Figure 9). This is probably because the residue left by a suspension consists of larger particles and these particles are partly masked by inert diluents. Both factors would result in less rapid penetration of toxicant into the leaf tissue. However, when the surface residues were allowed to remain on the leaves, as in the experiment the results of which are shown in Figure 8, the subcuticular residues were effective for a longer period where suspensions were applied than where emulsions were applied.

Summary

In "topical treatments" adult female two-spotted mites reared on bean plants were placed on either uninfested bean leaves or avocado leaf sections and treated, after which they were removed to untreated bean leaves and examined 48 hours later to determine the per cent mortality. In "residue treatments" the bean leaves or avocado leaf sections were treated and the mites were placed on them as soon as the residues were dry. In "topical-residue treatments" the mites were on the leaves at the time they were treated and were allowed to remain on the treated leaves for 48 hours. These mites were subjected to the combined effects of topical and residue treatments.

The three types of laboratory application of acaricides against adult two-spotted mites varied in their effectiveness in the order topical-residue > residue > topical. The superiority of the residue to the topical treatment was believed to be due, at least in part, to the additive effect of the surface and the subcuticular residues in the former.

With the exception of parathion, emulsions were more effective than suspensions when applied directly against the mites (topical treatment) or as freshly deposited residues. Suspensions were usually the more effective of the two formulations in long-term residual effectiveness in the field. With the acaricide G-338, suspensions and emulsions were about equal in long-term residual effectiveness.

The acaricides were more effective on bean leaves than on avocado leaves. This is believed to be due to the greater effect of subcuticular residues of the toxicants in bean leaves, which appear to exert an effect on the mites as stomach poisons.

In laboratory tests, avocado leaves from which acaricide residues were removed 1 and 24 hours after treatment remained toxic to mites placed on either surface of the leaf. In field treatments, with a concentration of 1 pound of toxicant to 100 gallons of spray, the subcuticular residues remained effective against adult mites almost as long as the subcuticular and surface residues combined. When leaves were washed 24 hours after treatment to remove the surface residues, they remained toxic to mites about as long as unwashed leaves when parathion suspension or emulsion was used, 82.4% as long when G-338 emulsion was used, and only 40.0% as long when G-338 suspension was used.

Acaricides applied to either the upper or lower surfaces of bean, grapefruit, and avocado leaves resulted in the death of mites confined to the leaf surface opposite to the one that was treated. Parathion was particularly effective in this respect.

The ability of a toxicant to kill mites on the leaf surface opposite to that which was treated may account for the control of mites on such crops as beans and cotton when only the upper surfaces of the leaves are wet by the spray.

The effectiveness of a spray directed against phytophagous insects and mites is less apt to be adversely affected by rain and other weathering agencies than is that of a spray directed against insects that do not feed on the treated substrate, for the subcuticular residues of the former continue to act as stomach poisons after the surface residues are removed.

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