RESEARCH BULLETIN 630 MAY, 1957

UNIVERSITY OF MISSOURI COLLEGE OF AGRICULTURE

AGRICULTURAL EXPERIMENT STATION

J. H. Longwell, *Director*

Lindane Residue in the Fermentation and Processing of Pickles

PART I
CHANGES IN LINDANE RESIDUE

MELVIN R. JOHNSTON

(Publication authorized May 14, 1957)

COLUMBIA, MISSOURI

TABLE OF CONTENTS

ACKNOWLEDGMENTS

The author gratefully acknowledges the counsel of Dr. R. A. Schroeder, the analysis of lindane made by Drs. C. W. Gehrke and E. E. Pickett and their staff in the Agricultural Chemistry Laboratories, and the handling of insect control schedules on the field plots by Dr. W. R. Enns, Department of Entomology. The bulletin reports on Department of Horticulture Research Project 197, "Elimination of Chemical Residues."

Lindane Residue in the Fermentation and Processing of Pickles

INTRODUCTION

The salting or brining of food is one of the oldest methods of food preservation practiced by man. Brining cucumbers, the initial step in processing pickles, is no exception to this statement. Started as an art in the home by the homemaker, the brine method of processing cucumbers into pickles has become an important segment of the commercial food industry. Through research, the transformation from an art to a science by the application of sound chemical and bacteriological principles has resulted in a stable commercial method of food preservation.

In commercial practice, large, unsheltered wooden vats are filled with green cucumbers. Generally the cucumbers are not washed. The filled vats are fitted with wooden false heads which retain the buoyant cucumbers in the cover brine. An initial brine concentration of 8 to 10 percent (30 to 40 degree Salometer) salt is used. Dry salt is added to the brine to compensate for the dilution by the water from the cucumber. An active fermentation becomes evident shortly after the cucumbers are brined and it persists for a period of 3 to 4 weeks. In most instances, after a period of time, the brine concentration is increased gradually to about 15 percent (60 degree Salometer). A curing process is completed under these conditions which converts the cucumber from a green, opaque, air-filled, buoyant fruit to an olive-colored, translucent, air-free salt stock. At this point they will remain preserved indefinitely, or they may be processed directly into one of the many possible styles of pickles.

The more the cucumber fermentation is studied the more apparent its complexity becomes. What at first seemed to be a simple lactic acid fermentation has many variations, depending upon a variety of conditions. After further consideration this is not altogether strange since a large number of micro-organisms from the soil are introduced into the vats with the cucumbers. The micro-flora of the soil is extremely varied, including species of molds, yeasts, actinomycetes, and many bacteria. Due to the selective action of the salt brine, yeasts and bacteria are the predominant groups in the micro-flora of the brine. For their nutritional requirements, species of yeasts and bacteria utilize the soluble constituents that diffuse into the brine due to the osmotic action of salt on the cucumber tissue. The persistence of any one species is dependent upon the organism's ability to tolerate the initial brine conditions as well as adapt itself to the conditions which develop during the predominantly lactic

acid fermentation. As studies of the process over the past 50 years have indicated, the micro-flora is quite sensitive to many environmental factors.

Within recent years many synthetic pesticides have been developed and used for the protection of food crops. These powerful pest control agents have received a great deal of attention from food investigators as to their toxicological nature and their ability to alter the flavor of various foods. Studies of the influence which these materials might have on food fermentations such as cucumber brining have not been reported in the literature.

Lindane, the gamma isomer of hexachlorocyclohexane, is a very effective insecticide which is used to control certain cucumber insects. The influence of lindane residue on cucumber fermentation, certain species of the micro-flora, and the flavor of sour, sweet and dill pickles constitutes the basis for this study.

Approach to the Problem

This study has as its objective the evaluation of the residues of the insecticide lindane on the pickle making process. That is, its effect on the brining of cucumbers (fermentation) and on the flavor of the finished pickles (sour, sweet and dill). Laboratory work on the problem was done during a four-year period.

The first year's work (1952-53) was designed to establish the effect, if any, of a maximum residue of lindane from a recommended spray concentration (1 pound of 25 percent wettable lindane per 100 gallons of water). The cucumbers (variety, Earliest of All) were obtained from a commercial grower in Newton County. By previous arrangement with the grower, the cucumber plants were not sprayed with lindane prior to harvest. The cucumbers were harvested in the morning and transported to Columbia in the afternoon. They were placed in 40 degree Fahrenheit storage for two days. The cucumbers were removed from storage and divided into two lots. One lot was sprayed with lindane (14 grams of 25 percent wettable per three gallons of water). To assimilate maximum residue from the spray, the cucumbers were spread one layer thick on a tarpaulin for application. A three-gallon manually operated pressure sprayer was used to apply the insecticide. The nozzle of the sprayer was held about two feet above the surface of the cucumbers. With maximum pressure on the sprayer by manual pumping, the nozzle was moved over the cucumbers so as to cover them with a spray from a single pass of the nozzle. The control lot was treated with tap water in a like manner. The prepared samples were moved directly to the brining kegs.

Each treatment (sprayed and control) was replicated three times in

the brining process. Each replication was composed of 75 pounds of cucumbers. The six containers were 21-gallon paraffin-lined white oak kegs. The heads were returned to the kegs after they had been filled with cucumbers. The kegs were placed on their sides with the open bung holes on the top side. Each keg was filled with a 40-degree Salometer brine (about 11 gallons) and salt was added to maintain it at this concentration during the fermentation (see Table 4). The temperature of the room was controlled automatically at 80 degrees Fahrenheit throughout the period of fermentation.

At the end of the fermentation period the salt concentration of the brine was increased five degrees Salometer each week until it was 60 degree Salometer. The salt concentration was maintained at 60 degrees Salometer until the salt stock was ready to process into sour, sweet and dill pickles. After the curing process had been completed the salt stock was transported to a commercial pickle processing plant in St. Louis where it was made into sour, sweet and dill pickles.

The processing steps used to produce the three styles of pickles were: Desalting—each of the three replications of a given treatment were lumped together in one large barrel. The hot water desalting method was used. A steam syphon was used to heat and circulate the leaching water. The temperature of the water was controlled at 150 degrees Fahrenheit. Alum and tumeric were added to the first desalting water. At the end of eight hours the water was removed and fresh water added. Three desalting waters were used and at the end of 24 hours the salt concentration in the pickle tissue was reduced to about 2 percent. At this point the stock was ready for processing into an edible product.

The stock for sour pickles was returned to the clean 21-gallon kegs. A sour liquor composed of vinegar and salt was put on them.

The dill pickle stock was returned to the clean 21-gallon kegs. A liquor composed of water, vinegar salt and dill oil was put on them.

Stock for sweet pickles was returned to the kegs and the sweetening process was begun. First the stock pickles were covered with a strong vinegar. After the vinegar had penetrated throughout the stock, sugar was added. The beginning sour-sweet liquor was about 15-degree Brix and this was increased gradually to 43-degree Brix over a 14-day period. When the stock had equalized with the high density liquor, it was removed. This was replaced with a fresh sweet liquor composed of water, sugar, vinegar and spice oil.

In this way it was possible to produce three styles of pickles from each of the lindane treated and control lots under commercial conditions of processing. The kegs of pickles were returned to Columbia where they

were put into jars for further testing (chemical and organoleptic analyses).

The second year's work (1953-54) was designed to confirm or reject the first year's findings on the effect of a maximum residue of lindane on the pickle making process. The cucumbers (variety Chicago Pickling) were obtained from a grower in Cole County (about 30 miles from Columbia). By arrangement with the grower, the cucumber plants were not sprayed with lindane prior to harvest. The cucumbers were harvested in the morning and transported to Columbia in the afternoon. They were placed in 40-degree Fahrenheit storage over night. The cucumbers were removed from storage and divided into two lots. One lot was sprayed with lindane (14 grams of 25 percent wettable per three gallons, equivalent to one pound per 100 gallons). To produce maximum residue from the spray, the cucumbers were spread one layer thick on a tarpaulin and the 1953 spray technique was repeated.

Each treatment (sprayed and control) was replicated three times in the brining process and each replication was composed of 100 pounds of field run cucumbers. The 21-gallon paraffin-lined white oak kegs were filled with the cucumbers. Keg heads were left off. The kegs were placed upright on stands and three different brining schedules were used with a sprayed and control keg making up each salting schedule. Kegs number 1 and 2 were filled with a 45-degree Salometer brine. Brine was maintained at that concentration throughout the fermentation. Kegs number 3 and 4 were filled with a 45-degree Salometer brine and the brine was permitted to equalize with the cucumbers. At weekly intervals during the fermentation period the concentration of the brine was increased by three degrees Salometer.

Kegs number 5 and 6 were filled with a 35-degree Salometer brine and the brine was permitted to equalize with the cucumbers. At weekly intervals during the fermentation period the concentration of the brine was increased three degrees Salometer. In all cases the initial volume of brine per keg was 10 gallons. The temperature of the room in which the fermentations were made was not controlled. Brine temperature during the fermentation was an average 80.6 degrees Fahrenheit and ranged from 75 to 84 degrees. Ultraviolet sun lamps were used four hours each day to reduce the growth of film-forming micro-organisms; one sun lamp was placed 18 inches above the brine surface of each keg.

At the end of the fermentation period the salt concentration of the brine was increased three degrees Salometer each week until it was 60 degrees Salometer. The salt concentration was maintained at 60 degrees Salometer until the salt stock was ready to process into sour, sweet and dill pickles. After the curing process had been completed, the salt stock

was processed into finished pickles in the University of Missouri horticultural products pilot plant.

The processing steps used to produce the sour, sweet and dill pickles were:

Desalting—Each 21-gallon replication of a given treatment was desalted separately and the cold water leaching method was used with a slight modification. The first water was left on the salt stock for 15 hours and circulated with an air pump. It was replaced with fresh water and circulated for nine hours. The third water was warmed to 130 degrees Fahrenheit and left for 15 hours without circulating and for each gallon of water used, nine grams of alum and six grams of tumeric were added. After the initial heating to 130 degrees Fahrenheit, the stream was turned off and the contents of the kettle permitted to cool during the 15 hours. The water cooled to about 90 degrees Fahrenheit during this time. The third water was drained off the stock and a spray of water was used to rinse the surface of the stock. At this point the stock was ready for processing into an edible product.

Each 21-gallon replication was size graded into large, medium and small stock. Manual grading was used and the size range of each grade was kept as uniform as possible from replication to replication. The large-size grade was made into sour pickles, the medium-size grade was made into dills and the small-size grade was made into sweets.

Sour Pickles—The stock for sour pickles was packed directly into one-gallon jars. A 60-grain vinegar containing a mixed spice formula was put on the stock. The spice formula was made of 2 grams cloveroyal, 2 grams maceroyal, 4 grams alspiceroyal, 2 grams red pepperoyal and 2 grams carnwaroyal per 23 pounds of vinegar. Jars were closed and stored for equalization of the sour liquor with the stock.

Dill Pickles—The stock for dill pickles was packed directly into one-gallon jars. A liquor composed of 30 grains of vinegar (acetic acid), 20-degree Salometer salt and five milliliters of dill oil emulsion per gallon was used to cover the medium size stock. The jars were closed and stored for equalization of the dill liquor with the stock.

Sweet Pickles— The small stock was sweetened in four-gallon stone jars. A liquor composed of 60 grains of vinegar and 15-degree Brix sugar syrup was used to just cover the stock. At the end of the first 24 hours, the concentration was increased to 20-degree Brix, the second 24 hours to 30 degree Brix, the third 24 hours to 35 degree Brix, the fourth 24 hours to 40 degree Brix, the fifth 24 hours to 45 degree Brix and after each of the next two 24-hour periods the liquor was corrected to 45-degree Brix. This sweet-sour liquor was replaced with a spiced sweet pickle liquor. The

composition of the final liquor was 43-degree Brix, 19 grain acetic acid, and 12 milliliters of a formulated sweet pickle spice oil emulsion per gallon of liquor. The jars were closed and stored for equalization of the sweet liquor with the stock.

In this way it was possible to produce three styles of pickles from each of the lindane treated and control lots under conditions which were practically the same as those used in commercial processing. Chemical and organoleptic analyses were made on the finished pickles.

The third year's work (1954-55) was designed to determine the effect, if any, of lindane residue from various recommended insect control schedules. A second phase was a study of the effect of lindane and i-inositol in graded combinations on the fermentation of cucumbers. In a third phase, pure cultures of yeast, mold and bacteria were cultured with lindane and i-inositol to determine the effect of these materials on cell production and growth.

Cucumbers (variety Model) were harvested from four insect control schedules and a control (no treatment) scheduled. Each schedule was replicated four times in one-fourth acre plots. The schedules were as follows:

I. Lindane, 1 percent dust and Captan, 7.5 percent dust.
II. Lindane spray, 1 pound 25 percent wettable per 100 gallons.
III. Lindane spray, 1 pound 25 percent wettable per 100 gallons and Captan spray, 2 pounds 50 percent wettable per 100 gallons.
IV. Lindane spray, 1 pound 25 percent wettable per 100 gallons and Methoxychlor spray, 2 pounds 50 percent wettable per 100 gallons.
V. Control—no treatment.

The plots were treated seven times prior to harvest. The cucumbers were grown on the same farm which grew the previous year's cucumbers.

Harvesting of the cucumbers was done in the morning with each treatment replication being kept separate. They were transported to Columbia in the afternoon and placed in 40-degree Fahrenheit storage over night. Each insect control schedule was replicated twice in the brining process. Each replication was composed of 100 pounds of field run cucumbers, 25 pounds from each of the four field replications. They were contained in 21-gallon paraffin-lined white oak kegs and the kegs without heads were placed upright on stands. The same brining schedule was used in all replications. Ten gallons of 40-degree Salometer brine were put in each keg and the brine was permitted to equalize with the cucumbers. The temperature of the room was not controlled and the brine temperature during the fermentation averaged 84 degrees Fahrenheit with a range of 79 to 91 degrees. Ultraviolet sun lamps were used four hours each afternoon to reduce the growth of film forming micro-organisms. One ultra-

violet sun lamp was placed 18 inches above the brine surface of each keg.

At the end of the fermentation period the salt concentration of the brine was increased three degrees Salometer each week until it was 60 degrees Salometer. The salt concentration was maintained at 60 degrees Salometer until the salt stock was ready to process into sour, sweet and dill pickles. After the curing process had been completed the salt stock was processed into finished pickles in the University of Missouri horticultural products pilot plant.

In processing the salt stock into sour, sweet and dill pickles, the same procedure was used as was described previously for the second year's work. The modified cold water leaching method was used to desalt the salt stock. The same method and materials were used in making the three types of pickles. Chemical and organoleptic analyses were made on the finished pickles.

In the second phase of the year's work, various combinations of lindane and i-inositol were used in the brine to determine their effect on the fermentation. The Model variety of cucumbers was used in the experiment. They were harvested from the number two treatment (lindane spray, 1 pound 25 percent wettable per 100 gallons). The bags of cucumbers were transported to Columbia and put directly into the brine. Each sample (17 pounds) of the experiment was placed in a four-gallon stone jar. Enough 40-degree Salometer brine (1.75 gallons) was added to the jars to cover the cucumbers. The amount and concentration of each chemical in each jar was as indicated in the following table.

Jar No.	Lindane		i-inositol	
	gm	ppm	gm	ppm
1	.1506	10	0	0
2	.1506	10	.0376	2.5
3	.1506	10	.0753	5
4	.1506	10	.1128	7.5
5	.1506	10	.1506	10
6	.1128	7.5	.1506	10
7	.0753	5	.1506	10
8	.0376	2.5	.1506	10
9	0	0	.1506	10
10	0	0	0	0

The concentrations indicated for each material are based on a four-gallon volume of water. An ultraviolet sun lamp was used four hours each day to inhibit the growth of film formers. This experimental design was replicated three times. The replications were at different times with a day between them to unload and reload the jars. In the first replication the fermentation was followed for 14 days, whereas the second and third replications were permitted to ferment 20 days. The fermented stock was

not processed into pickles.

A third phase of the year's work concerned pure culture work with these two materials: One mold, nine yeast and two bacteria were studied. The mold was *Neurospora crassa* (A-3507*), an inositolless mutant strain of *Neurospora. Candida krusei* (Y-301*), *Debaromyces globosus* (Y-2021*), *Hansenula fabianii* (Y-2240*), *Torulaspora rosei* (Y-1567*), *Saccharomyces carlsbergensis* (Y-1089*), Bakers yeast (*Saccharomyces,* Y-862*), *Saccharomyces fragilis* (Y-2007*), *Schizosaccharomyces pombe* (Y-659*), and *Zygosaccharomyces acidifaciens* (Y-1011*) were the yeast cultures. *Lactobacillus plantarum* (23-2**) and *Aerobacter aerogenes* (G-4**) were the bacterial cultures. Various combinations and concentrations of lindane and i-inositol were used in the invetro study with the pure cultures. Growth or the lack of growth was measured with a colorimeter. A wave length of 625 millimicrons was used. The aqueous cultures were incubated at 75 degrees Fahrenheit for a time period which varied with the growth rate of the organism. The range of time for the incubation period was from 11 to 93 hours.

For the fourth year's (1955-56) experiment, the amount of lindane in the field spray was varied and fungicides were combined with the recommended amount of lindane. To further evaluate the findings of the previous year's results, the number of spray applications prior to harvest was increased. Cucumbers (variety Ohio MR-17) were harvested from four insect-disease control schedules and a control (no treatment) schedule, each of which was replicated four times by one-fourth acre plots. The treatment schedules were:

I. Lindane spray, 1 pound 25 percent wettable per 100 gallons.
II. Lindane spray, 2 pounds 25 percent wettable per 100 gallons.
III. Lindane spray, 1 pound 25 percent wettable per 100 gallons and Zineb, 2 pounds per 100 gallons.
IV. Lindane, 1 pound 25 percent wettable per 100 gallons and Captan, 2 pounds 50 percent wettable per 100 gallons.
V. Control, no treatment.

The plots were sprayed 11 times prior to harvest, with the last spray being applied the day before harvest. They were grown on the farm which grew the previous year's crop.

Experimental samples were harvested in the morning with each schedule replication being kept separate. They were transported to Columbia and placed directly in the fermentation kegs. Three fermentation keg replications were prepared from each spray schedule. Each replication was composed of 100 pounds of field run cucumbers, 25 pounds from each field replication. The brining operation was the same as in 1955. Average

*Strain numbers from the Northern Utilization Research Laboratory, U.S.D.A.

**Strain numbers from the Dairy Department collection, Missouri Agricultural Experiment Station.

brine temperature during the fermentation period was 83 degrees Fahrenheit with a range of 75 to 89 degrees. The salting and curing after fermentation were handled as indicated for the previous year.

In processing the salt stock into sour, sweet and dill pickles, the procedure described for the second year's work was used. The methods and materials for making the three styles of pickles were the same. Chemical and organoleptic analyses were made on the finished pickles.

The study is reported in three parts. Part I, which follows, deals with changes in Lindane residues during fermentation and processing. Part II will report effects of Lindane on the microflora of the cucumber fermentation and Part III, the pickle flavor changes due to Lindance residue.

Part I
CHANGES IN LINDANE RESIDUE

The relation of insecticide residues to food products is becoming of increasing importance, as the amount of literature on the subject attests. However, the literature is not replete with quantitative amounts of insecticide residues on processed foods. Exact amounts of residues on raw produce (fruits and vegetables) have been reported to the greatest extent. Lamb (5) suggests that to cope fully with this problem there must be cooperation between insecticide manufacturers, universities, state experiment stations and various industrial laboratories. He points out that the problem is the responsibility of the entomologist, toxicologist, analytical chemist and food technologist. Some very constructive suggestions are made by Lamb (5) for carrying out residue studies on processed foods.

The importance of knowing the amounts of insecticide residue on and in our food is emphasized by McKay (6). His discussion of the application of the Federal Food, Drug and Cosmetic Act of 1938 to food raw materials bearing chemical residues is significant. He expresses the opinion of the Food and Drug Administration which has as its primary mission the protection of the consuming public. McKay relates the current data on the toxicity of new insecticides.

In a piece of work reported by Hornstein (4), no evidence was found to indicate that benzene hexachloride accumulated in the soil with repeated application of the insecticide. Peanuts that were grown on plots treated the previous year with the insecticide did not accumulate the compound. Reynolds (7) reported 1.8 ppm of benzene hexachloride in peanut butter which was made from peanuts grown in soil that was treated with the insecticide.

Residues of the insecticides parathion, DDT and TDE were studied by Ginsburg (2). He found that corn, peas, potatoes and turnips did not have detectable amounts of these materials on them after being treated with a 0.5 percent dust or spray at the rate of 0.125 to 0.25 pounds per acre. Pea vines had 0.1 ppm parathion at the time of harvest, 19 days after being sprayed with 0.5 pounds of parathion per acre. Corn which was dusted three times with a three percent DDT dust contained 13.1 ppm of DDT at harvest, 13 days after the last dusting. Corn which was dusted five times with a five percent TDE contained 20.3 ppm of TDE. Rainfall during the growing period of these crops was 0.99 inches.

Lamb (5) found that canned tomatoes had 0.5 to 1.0 ppm of DDT and DDD when they were dusted with these materials prior to processing. He does not see any immediate problem with these insecticides on tomatoes.

Gunther (3) discussed the analytical problems encountered in attempting to characterize and localize metabolized insecticide residues in food products. An example is parathion. It is not volatile, yet in 21 days two-thirds of the original material has been degraded.

The study reported here was made to determine quantitatively the amount of lindane on cucumbers from various insect control schedules with lindane, and to investigate the effect of fermentation and processing on this residue.

Materials and Methods

Samples for residue (gamma-hexachloro-cyclohexane) analyses were taken after each of the three major steps in the processing of cucumbers into pickles: (1) The fresh (raw) cucumbers were sampled after the spray treatments; (2) they were sampled again when the curing process was completed; and (3) the final sampling was from the finished sour, sweet and dill pickles. A five to six-pound aliquot was drawn from each treatment at each sampling. The aliquot was taken over a cross-section of each treatment.

Preparation of the samples for analysis was the same throughout the four years of the work. The samples in a liquid medium (salt stock, sour sweet and dill pickles) were drained on an eight-mesh screen for two minutes. They were cut into one-fourth inch cubes and placed in a Waring blendor. The blending time was five minutes. This period of time was found to be sufficient to reduce the samples to a uniformly fine puree. Each blendor load of an aliquot was transferred to a stainless steel bucket and the blended material thoroughly mixed. About two pounds of the blended material were placed in a plastic container and sealed. All the raw cucumber samples were stored in a 0 degree Fahrenheit storage room and the self preserved salt stock, sour, sweet and dill pickles were stored in a 40-degree Fahrenheit storage room prior to being analyzed.

The Agricultural Chemistry Laboratory of the Missouri Agricultural Experiment Station did the analysis of the samples for benzene hexachloride. During the four-year period, 136 different samples were analyzed by the Station Laboratory. The Schechter- Hornstein method (8) for the determination of benzene hexachloride was used by the Station Laboratory. Gehrke and Bevert (1) have reported on a study of the critical points in the Schechter-Hornstein colorimetric method for benzene hexachloride.

Results

Results of the analysis of the samples from the 1952 work are shown in Table 1. The raw sprayed cucumbers contained 10.4 ppm and the salt stock contained 10.0 ppm of gamma benzene hexachloride. In the finished pickles. the insecticide was found in the sour, sweet and dill style pickles at concentrations of 12.0, 2.75 and 4.0, respectively. This residue resulted from a spray solution which was composed of one pound of 25 percent wettable lindane per 100 gallons of water. The spraying technique was designed to leave a maximum residue of the spray material.

TABLE 1. AMOUNT OF LINDANE RESIDUE ON FRESH WEIGHT BASIS FROM 1952 SAMPLES

	Raw Cucumbers (ppm)	Salt Stock** (ppm)	Pickle Types		
			Sour (ppm)	Sweet (ppm)	Dill (ppm)
Spray*	10.40	10.00	12.00	2.75	4.00
Control	3.08	0.21	0.40	2.15	0.72

*1 pound of 25% wettable lindane per 100 gallons of water.
**covered with 40° Salometer brine which was maintained during fermentation.

Table 2 gives results of the analysis of samples from 1953 work. The raw sprayed cucumbers contained 11.72 ppm of gamma benzene hexachloride. This is approximately the same amount that was present on the 1952 raw cucumbers. The lindane sprayed salt stock samples contained less than 1 ppm and the sour, sweet and dill had 1 ppm or less of the spray material. These data are considerably lower than those on similar samples from the 1952 work, yet they were sprayed the same way.

TABLE 2. AMOUNT OF LINDANE RESIDUE ON FRESH WEIGHT BASIS FROM 1953 SAMPLES

	Raw Cucumbers (ppm)	Salt Stock (ppm)	Pickle Types		
			Sour (ppm)	Sweet (ppm)	Dill (ppm)
Spray*	11.72	0.05	0.85	1.12	0.88
Control	1.70	0.75	1.19	0.45	0.24
Spray*	11.72	0.50**	0.49	0.31	0.48
Control	1.70	1.65**	0.20	0.65	0.37

*1 pound of 25% wettable lindane per 100 gallons of water.
**Covered with 46° Salometer brine which was permitted to equalize.

Residue of lindane on the samples from the controlled spray schedules of 1954 was very low (see Table 3). The raw cucumbers (variety Model) from the spray plots contained about 1 ppm of lindane. The salt stock and processed sour, sweet and dill pickles contained from 0.54 ppm to no detectable residue of lindane. The field control schedules were: Plot 1, 1 percent lindane dust and 7.5 percent captan dust. Plot 2, one pound 25 percent lindane per 100 gallons. Plot 3, one pound 25 percent lindane

TABLE 3. CONCENTRATION OF LINDANE RESIDUE ON FRESH WEIGHT BASIS FROM 1954 SAMPLES

Spray Plot No.	Raw Cucumber (ppm)	Salt Stock (ppm)	Pickle Types		
			Sour (ppm)	Sweet (ppm)	Dill (ppm)
1	0.93	0.54	0.41	0.42	0.20
2	0.73	0.00	0.00	0.05	0.08
3	0.85	0.00	0.00	0.42	0.07
4	1.07	0.06	0.00	0.00	0.00
5	1.00	0.12	0.00	0.00	0.06
11	*	4.25	3.73	4.40	5.50
12	**	7.30	4.68	8.10	7.00

*1.13 gm of lindane and 1.13 gm i-inositol placed in brine and cucumbers (30 gallon volume). Equivalent to 10 ppm of each material.
**1.13 gm of lindane placed in brine and cucumbers (30 gallon volume). Equivalent to 10 ppm of material.

and two pounds 50 percent captan per 100 gallons. Plot 4, one pound 25 percent lindane and two pounds 50 percent methoxychlor per 100 gallons. Plot 5, control (no treatment). The plots were treated seven times prior to harvest with the last one being made the day before harvest.

Analysis for lindane on samples from two fermentations in which known amounts of lindane and i-inositol were added is of interest (see Table 3). Enough lindane (1.13 gm) was added to samples 11 and 12 to make a 10 ppm concentration in the cucumbers (125 pounds) and brine (15 gallons). In addition, sample 11 was treated with 1.13 grams of i-inositol which would yield a 10 ppm solution. Sample 11 salt stock had 4.25 ppm of lindane after curing and sample 12 salt stock had 7.30 ppm of lindane. The sour, sweet and dill pickles from sample 11 contained 3.73, 4.40 and 5.50 ppm of lindane, respectively; whereas, similar pickles from sample 12 had 4.68, 8.10 and 7.00 ppm of lindane, respectively.

As shown in Table 4 the concentration of lindane on samples (varie-

TABLE 4. CONCENTRATION* OF LINDANE RESIDUE ON FRESH WEIGHT BASIS FROM 1955 SAMPLES

Spray Plot	Raw Cucumbers (ppm)	Salt Stock (ppm)	Pickle Types		
			Sour (ppm)	Sweet (ppm)	Dill (ppm)
I	0.35	0.14	0.13	0.18	0.34
II	0.10	0.23	0.16	0.15	0.26
III	0.15	0.25	0.21	0.18	0.17
IV	0.25	0.28	0.14	0.27	0.18
V	0.00	0.23	0.06	0.14	0.17

*Each value is the average of three independent determinations.

ty, MR-17) from the 1955 field control schedules were very low. Treatments used on the plots were: (I). one pound 25 percent lindane per 100 gallons; (II.) two pounds 25 percent lindane per 100 gallons; (III.) one pound 25 percent lindane and two pounds zineb per 100 gallons; (IV.) one pound 25 percent lindane and two pounds 50 percent captan per 100 gallons; and (V.) control (no treatment). Eleven spray applications were made prior to harvest with the last one being made the day before harvest. The data from these plots agree very well with the 1954 results.

Discussion

Cucumbers that have been treated directly with a spray containing one pound of 25 percent wettable lindane per 100 gallons of water can be expected to contain about 11 ppm of benzene hexachloride residue. The raw cucumbers contained 10.4 ppm in 1952 and 11.72 ppm in 1953. They were treated with a spray of the same concentration. In actual practice, it should be very difficult to approach these amounts of lindane residue. Undoubtedly the protection afforded by the foliage of the plant would prevent accumulation of large amounts of insecticide residue on the cucumbers. Results of the analysis of raw cucumbers from field plots in 1954 and 1955 indicate that this is the situation. A concentration of 1.07 ppm of lindane was the highest residue found from the field trials. In nearly all cases the amount of residue on the raw cucumber was greatly reduced by subsequent processing.

Salt stock in 1952 was found to contain 10.0 ppm of benzene hexachloride, whereas the 1953 salt stock contained 0.05 ppm of benzene hexachloride. The amount of residue found in 1952 seemed high considering that the cucumbers remained in an approximately equal volume of brine for seven months. Dilution by the brine would be expected to reduce the 1952 salt stock residue to 5 or 6 ppm. The extent to which the kegs were closed was different; the brining schedule that was used for each of the two years was different. These might have been factors contributing to the difference. In the 1954 salt stock, the residue ranged from 0.05 to 0.0 ppm. These data were similar to the 1955 results.

Sour, sweet and dill pickles from the 1952 work were found to contain considerably more spray residue than the 1953 pickles. The value for sour pickles in 1952 was somewhat higher than the concentration of residue found on the raw cucumber. No logical explanation is possible, for the vigorous leaching of the salt stock would tend to remove the residue rather than increase it. In 1952 the hot water desalting method was used, whereas in 1953 a modified cold water desalting method was used. Due to the low amount of residue on the 1953 salt stock, it is not possible to compare the efficiency of the two desalting methods. The amount of residue found on the three styles of pickles in 1954 and 1955 was extremely low. In all cases less than 0.5 ppm was present in each of the three styles

of pickles. The desalting method used in 1954 and 1955 was the same as 1953.

Reduction in the amount of residue due to processing was of interest in the case of Samples 11 and 12 (Table 3). In Sample 11 the concentration of residue was 4.25 ppm and in sample 12 it was 7.3 ppm. Both fermentations were treated with 10 ppm lindane and sample 11 was treated with an additional 10 ppm of i-inositol. After the salt stock from the two fermentations was processed into pickles, there was little or no reduction in the amount of residue. The difference in the amount of residue in the salt stock of the two treatments was maintained in the finished pickles. Again, the sweet and dill pickles from both fermentations were found to contain a higher amount of residue than the salt stock.

In only three cases was it found that the observed data were higher than the theoretical solubility of benzene hexachloride in water. As reported by Slade (9) the maximum solubility of gamma-benzene hexachloride in water is 10 ppm.

Conclusion

The data indicate that residue from lindane spray on cucumbers in the field — after fermentation and processing into pickles is insignificant, but a concentration of 10 ppm could result if harvested cucumbers were sprayed directly.

Literature Cited

1. Gehrke, C. W. and J. L. Bevert. Critical points in the Schechter-Hornstein colorimetric method. Mo. Agr. Exp. Sta. Res. Bul. No. 606 (1956).
2. Ginsburg, J. M. How much newer insecticides parathion, DDT and TDE remain on treated crops at harvest? N.J. Agr. Exp. Sta. Ann. Rept. (1950).
3. Gunther, F. A. Analytical problems encountered in attempts to characterize and localize metabolized insecticide residues in food products. Food Technol. 8:36 (1954).
4. Hornstein, I. *et al.* Benzene hexachloride content and flavor of peanuts grown in rotation with cotton dusted with this insecticide. Agr. and Food Chem. 2:776 (1954).
5. Lamb, F. C. Relation of spray residue problem of food processing. Food Technol. 3:339 (1949).
6. McKay, McK. Jr. Application of the Federal Food, Drug and Cosmetic Act to food raw materials bearing chemical residues. Food Technol. 4:25 (1950).
7. Reynolds, H. *et al.* Palatability and chemical studies on peanuts grown in rotation with cotton dusted with insecticides containing benzene hexachloride. Ag. and Food Chem. 1:772 (1953).
8. Schechter, M. S. and I. Hornstein. Colorimetric determination of benzene hexachloride. Anal. Chem. 24:544 (1952).
9. Slade, R. The gamma isomer of hexachlorocyclohexane (Gammexane); an insecticide with outstanding properties. Chem. and Indus. 40:314 (1945).