Determination of Isopropyl *N*-(3-Chlorophenyl)carbamate Residues in Potatoes Treated for Sprout Inhibition

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Potatoes treated with isopropyl *N*-(3-chlorophenyl)carbamate (CIPC) to inhibit sprouting during storage have been tested for micro amounts of chemical residue. Potatoes treated by dipping or spraying with a 0.5% emulsion produced residues of about 0.4 p.p.m. Peeling the potatoes prior to analysis reduced the residue concentration. Potato chips prepared from treated potatoes gave a residue concentration level below 0.05 p.p.m. of CIPC, which is the lower limit of sensitivity of the analytical method.

THE ECONOMIC BENEFITS to be realized by treating potatoes to retard sprout formation and shrinkage during storage can be very important to the Quartermaster Corps of the Army, wholesalers of potatoes, and other large users of potatoes such as potato chip manufacturers.

Experiments on the ability of isopropyl *N*-(3-chlorophenyl)carbamate (CIPC) to inhibit the sprouting and shrinkage of potatoes during storage (*6*) and on the various modes of applying the chemical to potato tubers (*4*) have been reported.

As CIPC effectively inhibits potato sprouting, the amount of chemical residue remaining on the crop after storage and after washing, peeling, and cooking the potatoes into chips has become an important aspect of the treatment.

Analytical Methods

The analytical method described by Gard and Rudd (*3*) for determining micro amounts of CIPC in treated crops at concentration levels down to 0.05 p.p.m. has been applied successfully to many food crops (*1, 2*), including sweet potatoes and peanuts. As the problems anticipated in the residue analyses of potatoes and potato chips were similar to those already encountered and solved in the analysis of peanuts and sweet potatoes, the same methods were applied in the current investigation.

Potatoes. Before the residue analyses were started, considerable attention was given to appropriate sampling practices which would be applicable to the potato tubers.

Medium-sized potatoes were quartered; larger ones were divided into eighths. Segmented portions were selected at random from a sufficient number of different tubers to yield 200 grams of sample for each test.

The analytical method entailed the maceration of 200 grams of the sample at room temperature for approximately 3 minutes with methylene dichloride in a Waring Blendor. After blending, the extract was separated from the pulp by centrifugation. The extract phase was heated slightly and the solvent was evaporated at a reduced pressure to isolate the residue. The residue which remained was hydrolyzed by refluxing with dilute sulfuric acid to convert the CIPC to 3-chloroaniline and isopropyl alcohol. After the solution had been made alkaline with sodium hydroxide, the 3-chloroaniline was steam-distilled and collected in the distillate. The distillate was then treated with hypochlorite and the phenol–ammonium hydroxide solution to produce the blue color of the 3-chloroaniline complex. The resulting color of the solution was measured with a photoelectric colorimeter. The amount of CIPC residue found in the sample was determined from a previously prepared calibration curve for the photoelectric colorimeter.

Potato Chips. The same basic analytical method applied to potato tubers was also used in the analysis of potato chips. With the chips, however, the method (*3*) applicable to oily crops such as cotton seeds and peanuts was required, because of the comparatively large amounts of cooking oils (about 30% by weight) which were extracted by the methylene dichloride.

Cooking oils which were extracted along with any CIPC residue were concentrated in the flask by evaporating the methylene dichloride under reduced pressure with gentle heating. After the methylene dichloride had been expelled, the residual oil was shaken with small portions of acetonitrile to extract the CIPC residue into the nitrile phase, while leaving the cooking oils essentially undissolved. The use of acetonitrile as a supplementary extractant to separate similar organic residues from plant and animal tissue was used by Jones and Riddick (*5*) in connection with insecticide separations.

Following successive extractions with acetonitrile, the nitrile phases were combined in the hydrolysis flask and evaporated at reduced pressure and moderate heat to remove the solvent and concentrate any CIPC residue. Acidic hydrolysis, steam distillation of the 3-chloroaniline, and colorimetric measurements of the chloroaniline were conducted in the same manner as for the potato tubers.

A very careful evaluation of the reagent blank was necessary, because of the extremely low concentration levels of CIPC expected. Experiments showed that the transmittance readings for the reagent blank involving no crops, but with varying lots of reagents, ranged between 87 and 94% as compared with distilled water, and were dependent on the purity of the particular lots of reagents used. Each new lot of reagents, therefore, required careful evaluation prior to use, in order to establish the origin of the calibration curve. All analytical values listed in Table I, except transmittance results, were computed from transmittance curves based on the blank obtained for the reagents used. For ease of comparison, the transmittance results were corrected to the value they would have had, if the reagent blank had been 94% in each case. The precision of this blank for given lots of reagents was ±1% transmittance.

Treatment of Samples and Analytical Results

Samples of treated Kennebec variety potatoes, including some grown in Maine and some in North Dakota, were used. The Maine crop was treated with a 1% emulsion of CIPC and the North Dakota crop was treated with a 0.5% emulsion of the chemical. Treatment for both crops involved dipping in the particular emulsion, followed by storage at 45 and 55° F., respectively, during a 9- to 10-month period prior to being sent

to the Research Laboratory of the Columbia-Southern Chemical Corp. for residue analysis. The residue analyses for these potatoes obtained by the analytical method outlined are given in Table II.

Portions of the potatoes treated by dipping in the 0.5% emulsion of CIPC were soaked and superficially washed in tap water, scrubbed with a vegetable brush, and peeled, prior to analysis, in an effort to appraise the effects of these common operations in reducing the residue concentrations of the potatoes. The results of these different treatments on residue content are given in Table III.

A potato chip manufacturer (*8*) has also investigated the use of CIPC in the sprout inhibition of potatoes. In these experiments, samples of Kennebec and Russet Sebago potatoes were spray-treated with a 0.5% emulsion of CIPC and permitted to air-dry prior to their placement in mesh sacks for storage at 55° F. and 75 to 85% relative humidity for 3 months. The potatoes, with appropriate controls, were then placed in the curing environment which ranged from 72 to 78° F. for a 4-week period. Portions of the potatoes were processed into chips and samples of the potatoes and the potato chips were furnished to this laboratory for residue analyses. A composite, comprising both varieties of the potatoes and potato chips, was made prior to commencing the analyses. The analyses of these peeled potatoes and potato chips are also given in Table II.

In addition to the samples of potatoes and potato chips described above, the same potato chip manufacturer (*8*) had earlier supplied samples of other treated potatoes and potato chips to the Wisconsin Alumni Research Foundation Laboratories for residue analyses. These potatoes had been treated with a higher concentration of CIPC. A summary of these analyses is given in Table IV.

Table I. Recovery of Isopropyl *N*-(3-Chlorophenyl)carbamate from Potatoes and Potato Chips

CIPC Added		Red Light Transmittance,	CIPC Found			
			Total		Net	
Mg.	P.p.m.	%	Mg.	P.p.m.	P.p.m.	Recovery, %
			Potatoes			
0.000	0.000	88	0.0060	0.030	...	...
		89	0.0050	0.025	...	...
		88	0.0060	0.030	...	...
0.010	0.050	78	0.0166	0.083	0.055	110
		81	0.0137	0.069	0.041	82
		80	0.0148	0.074	0.046	92
0.020	0.100	70	0.0266	0.133	0.105	105
		70	0.0266	0.133	0.105	105
			Potato Chips			
0.000	0.000	91	0.0034	0.017	...	...
		94	0.0000	0.000	...	...
		93	0.0018	0.009	...	...
0.010	0.050	83	0.0114	0.057	0.048	96
		86	0.0080	0.040	0.031	62
		85	0.0096	0.048	0.039	78
0.020	0.100	77	0.0174	0.087	0.078	78
		75	0.0204	0.102	0.093	93

Table II. Isopropyl *N*-(3-Chlorophenyl)carbamate Residues in Potatoes and Potato Chips Receiving Treatment

Sample	Treatment, % CIPC in Emulsion	CIPC Found, P.P.M.						
		Replicate Tests						
		1	2	3	4	5	Av.	Net
Potatoes, unpeeled	None	0.03	0.03	0.02	0.03	0.03	0.028	...
	1.0[a]	0.80	0.80	0.75	0.75	0.80	0.780	0.752
	0.5[a]	0.43	0.43	0.42	0.42	0.42	0.424	0.396
Potatoes, peeled	None	0.01	0.03	0.01	0.02	0.01	0.016	...
	0.5[b]	0.09	0.10	0.08	0.09	0.08	0.088	0.072
Potato chips	None	0.02	0.00	0.01	0.01	0.00	0.008	...
	0.5[c]	0.02	0.01	0.02	0.00	0.01	0.012	0.004

[a] Treated by dipping, potatoes from P. C. Marth. [b] Treated by spraying, potatoes from Red Dot Foods. [c] Prepared from 0.5% spray-treated potatoes, from Red Dot Foods.

Table III. Effect of Method of Sample Preparation on Residue Content

Potato Sample Preparation[a]	CIPC Residue, P.P.M.[b]		
	Test 1	Test 2	Av.
Unpeeled	. .	. .	0.40[c]
Soaked and washed	0.27	0.28	0.28
Scrubbed with brush	0.35	0.33	0.34
Peeled	0.09	0.15	0.12

[a] Potatoes treated by dipping in a 0.5% emulsion of CIPC. [b] Net analyses corrected for crop interference. [c] From Table II.

Table IV. Residue Analyses of Treated Potatoes and Potato Chips

(Data furnished by Wisconsin Alumni Research Foundation)

Crop and Treatment	CIPC Residue, P.P.M.
Potatoes	
Unpeeled, 2% dip	4.2
Peeled, 1% spray	0.85
Potato chips	
1% spray	0.075
1% dip	0.073

Discussion

The analyses of potatoes treated for sprout inhibition with CIPC by either a dipping or spraying process show measurable concentrations of residue. This is exemplified by the analyses obtained both in this laboratory (Table II) and at the Wisconsin Alumni Research Foundation (Table IV) on samples of treated potatoes furnished by the United States Department of Agriculture and by the potato chip manufacturer (*8*). These findings were not unexpected, because the chemical was applied directly to the harvested crop. An entirely different situation prevailed when CIPC was added, as a herbicide, to the soil during pre-emergence and growth of the plant. In this latter case, application of the herbicide is made only to the soil in which the crop is grown, and many opportunities exist for evaporative and other losses of the herbicide (*7*).

The replicate analyses of potatoes and of potato chips utilizing 200-gram specimens are given in Table II. To obtain the apparent net amount of residue which remained with the treated potatoes and potato chips, the control analysis represented by the samples receiving no treatment was subtracted from the values obtained for the samples receiving the various concentration levels and modes of treatment.

The analyses of potatoes given in Tables II and IV show that as the treatment level of the tubers increases, higher concentrations of residue prevail after storage. Peeling of the potatoes prior to analysis resulted in a significant reduction in the residue content. It is likely that the thickness of the peel influences the amount of residue remaining with the potato. Soaking or scrubbing the potatoes without the peeling operation reduced the amount of residue somewhat, but not to an extent equal to peeling.

The residue analyses of potato chips prepared from treated potatoes which were spray-treated with a 0.5% emulsion of CIPC are also shown in Table II. The CIPC residue was either destroyed or removed from the potatoes during the deep-frying process necessary during the preparation of chips. The first studies

on potato chips were carried out with potatoes receiving treatment with a 1% emulsion of CIPC. Table IV shows that measurable amounts of residue were found in these chips. The effect of reducing the CIPC emulsion to 0.5% of active ingredient was then investigated with the results shown in Table II. In this case no measurable residues were found in the potato chips.

In interpreting the residue analyses of the potatoes and potato chips it is recognized that the CIPC may have undergone chemical change or was assimilated and metabolized during storage of the potatoes and their subsequent preparation into chips and therefore may not be detected as such by the analytical method.

Because the ability of CIPC to inhibit the sprouting of potatoes during storage has been effective on an experimental basis, and CIPC residues have always been found with the treated potatoes, the Procurement Section of the United States Army has arranged for chronic toxicity studies on CIPC to be conducted at the University of Virginia. These tests are now in progress.

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The potatoes analyzed were supplied by P. C. Marth, United States Department of Agriculture, Beltsville, Md., and E. D. Jones, Red Dot Foods, Inc., Madison, Wis. The potato chips were also furnished by E. D. Jones.

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PLANT GROWTH INHIBITORS

Factors Affecting the Performance of Maleic Hydrazide

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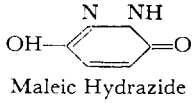
The plant growth inhibitor 6-hydroxy-3(2*H*)-pyridazinone (maleic hydrazide) must be absorbed and translocated to be effective. Studies using tracer, spectrophotometric, and chromatographic techniques showed that maleic hydrazide is stable and nonvolatile, and is efficiently translocated. Absorption is often slow. Experimental techniques made it possible to get quantitative data on variables influencing absorption rate. Light, temperature, and application rate were not critical within the usual range. Plant species and plant condition had significant effects. Relative humidity and formulation were more important. Maleic hydrazide was absorbed poorly from all formulations at low relative humidity. At moderate and high humidities formulation differences were evident. The diethanolamine salt was the most practical of the type absorbed efficiently.

The plant growth inhibitor 6-hydroxy-3(2*H*)-pyridazinone (maleic hydrazide or MH)



Maleic Hydrazide

is applied to plants as a foliage spray under commercial use conditions. To be effective, it must be absorbed and translocated by the plant. Other factors, such as chemical instability or washoff could also affect performance under field conditions.

A program designed to determine the factors which were important to maleic hydrazide performance, and to compare various formulations, was therefore planned. While field results are the final means of determining such effects, the obvious advantages of working under reproducible, controlled conditions indicated the need for laboratory studies.

Experimental

The following techniques were used to study the absorption and translocation of maleic hydrazide.

Absorption Study Technique. Tomato plants (var. Bonnie Best) were grown in individual plastic pots. Plants were selected for uniformity at the time of use, when the plants were about 4 inches tall and had five to six leaves. The watering schedule was such that soil moisture was about halfway between the field capacity (30%) and air-dried levels. The pots were wrapped in polyethylene film to minimize unequal moisture loss from the soil surface. The plants were then sprayed to runoff with a solution of the formulation containing 1500 p.p.m. of maleic hydrazide. This resulted in a deposit of about 1.5 mg. per plant.

For each treatment 19 plants were sprayed. Ten of these were held for 30 minutes to reduce unequal drip off during handling, then cut at the soil surface. The cut plants were washed, two plants per replicate, in 200 ml. of a 100 p.p.m. solution of sodium lauryl sulfate. (The washing technique used had been shown to give complete removal of applied maleic hydrazide in preliminary experiments with several species of plants.) Aliquots of the five wash waters were adjusted to pH 10. This required about one drop of a 40% sodium hydroxide solution. The effect on volume was negligible. The absorbance of the solutions was measured at wave lengths of 303, 328, and 353 mμ using 1-cm. cells with a Beckman DU spectrophotometer (maximum at 328 mμ for salts of maleic hydrazide in water). The readings of 303 and 353 mμ were used to eliminate the interference by using Equation 1.

$$OD_{328} - \left[\frac{OD_{303} + OD_{353}}{2}\right] = N \quad (1)$$