Table VII. Persistence of 2,4-Dichlorophenol in Lake Waters

Time, Days	100 μg. per Liter pH	Concn.	Oxid., %	500 μg. per Liter pH	Concn.	Oxid., %	1000 μg. per Liter pH	Concn.	Oxid., %
				AERATED AND BUFFERED					
0	7.4	100	0.0	7.4	500	0.0	7.4	1000	0.0
2	7.3	64	36.0	7.6	390	22.0	7.6	760	24.0
9	7.3	0	100.0	7.5	170	66.0	7.4	460	54.0
16	6.9	0	100.0	7.1	92	81.6	7.2	165	83.5
23	..	..	...	7.5	32	93.6	7.5	78	92.2
30	..	..	...	7.3	13	97.5	7.3	25	97.5
				UNAERATED AND UNBUFFERED					
0	7.3	100	0.0	7.3	500	0.0	7.3	1000	0.0
3	6.2	80	20.0	5.1	390	22.0	4.1	780	22.0
7	6.1	70	30.0	6.1	380	24.0	6.0	770	23.0
14	..	..	...	..	..	..	6.1	620	38.0
17	7.9	40	60.0	6.5	253	49.4	6.3	560	44.0
24	..	40	60.0	..	192	61.6	..	540	46.0
43	..	20	80.0	..	192	61.6	..	506	49.4

posed under these conditions. This finding is in agreement with the observations of other investigators that 2,4-D esters were hydrolyzed by microbial action in soils (*5*) or during absorption by plant leaves (*6*).

The lake bottom mud and manometric studies indicate that 2,4-D is biologically decomposed in a relatively short period of time with adapted microorganisms. However, 2,4-D is applied usually once every 1 or 2 years to a surface water for aquatic plant control. Consequently, the development of adapted microorganisms may take a relatively long time under such unfavorable anaerobic conditions that may develop when dead aquatic plants decompose. Since some 2,4-D–treated surface waters serve as continuous potable water supplies, this herbicide may persist for sufficient periods of time to warrant its removal at the treatment plant.

2,4-Dichlorophenol, on the other hand, was biologically broken down in the lake water indicating the presence of microorganisms capable of decomposing this compound in natural waters. Concentrations of the phenol up to 1000 μg. per liter did not affect the rate of oxidation since the two lines in Figure 5 were parallel and reached 50% removal level at the same time. Under unfavorable environmental conditions that may result from the decomposition of excessive amounts of organic matter in lake waters, the pH value will drop, anaerobic conditions prevail, and the phenol will persist for longer periods of time.

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PESTICIDE PURITY MEASUREMENT

Cryoscopic Analysis of Organic Phosphate Pesticides—Malathion, Dimethoate, *O, O*-Diethyl *O*-2-Pyrazinyl Phosphorothioate, and Phorate

R. J. BEST[1] and N. R. HERSCH
Central Research Division, American Cyanamid Co., Stamford, Conn.

It was necessary to establish the purity of specially purified samples of malathion, dimethoate, *O,O*-diethyl *O*-2-pyrazinyl phosphorothioate, and phorate. Organic phosphate compounds may be analyzed by vapor phase chromatography, ultraviolet spectroscopy, infrared spectroscopy, polarography, and various chemical methods such as bromination, hydrolysis, silver titration, etc. However, any of these methods requires a primary standard or comparison compound and in addition is frequently subject to interference from other compounds present. In the case of large molecules, these difficulties are more pronounced. Therefore the cryoscopic method, which is not subject to interference from other compounds and does not require a standard, was selected. The cryoscopic method consists of measuring the equilibrium temperature of a solid-liquid system as the compound being analyzed is slowly frozen or melted (*1, 2, 4–8*). The method is particularly useful in the analysis of a compound approaching 100% purity because it directly measures the amount of impurity present without requiring knowledge of the nature of any impurity.

[1] Present address: Building Products Division, American Cyanamid Co., Sanford, Me.

Experimental

Instrumentation and Apparatus. A Thwing-Albert constant temperature differential cryoscope with a controller

The determination of the absolute mole per cent purity of malathion, dimethoate, O,O-diethyl O-2-pyrazinyl phosphorothioate, and phorate pesticides of high purity was achieved by a cryoscopic method involving the measurement of the melting or freezing behavior of the compounds. The time-temperature melting curve of each was analyzed by the Taylor-Rossini method or the Witschonke method to determine the melting point of the pure compound. The cryoscopic constant, K, was established by deliberately adding impurities and measuring the new melting point. In no case was the formation of a solid solution indicated. The heat of fusion is calculated from the data.

recorder and a temperature recorder was used in conjunction with platinum resistance thermometers. The sample was stirred by a reciprocating helical stirrer. The thermometer recorder system was calibrated at the ice point, water triple point, benzoic acid point, and by direct comparison with a certified L. & N. platinum resistance thermometer and Mueller bridge.

Materials. The four compounds studied were malathion, dimethoate, *O,O*-diethyl *O*-2-pyrazinyl phosphorothioate, and phorate. These were prepared in the Cyanamid Agricultural Center. In addition, the following compounds were used in the determination of the cryoscopic constants: diethyl fumarate, *O,O-S*-trimethyl phosphorodithioate, *O,O*-diethyl *O*-phenyl phosphorothioate, and several common laboratory solvents.

Results and Discussion

Malathion. Malathion, $(CH_3O)_2P(S)SCH(COOC_2H_5)CH_2COOC_2H_5$, is very viscous at its freezing point and consequently does not crystallize rapidly; even under optimum conditions it takes about an hour to properly crystallize a sample of the material. Therefore, melting curves were run on the malathion samples instead of freezing curves to ensure phase equilibria during the runs. Table I summarizes the results of a number of melting curves of samples of malathion of different purities. The samples were better than 98 mole % pure with four exceptions.

The Witschonke (*8*) relationship was

$$T_o = T_m + (T_m - T_{1/2})$$

used to calculate T_o, the melting point of 100% material. T_m is the observed melting point of each sample, and $T_{1/2}$ is the temperature at which the sample is half liquid and half solid. The melting curves were so flat as to render the uncertainty in estimating $T_{1/2}$ very small. Any estimate of the percentage melted from 35 to 65% would give a value of $T_{1/2}$ not more than a few hundredths of a degree in error. Therefore, it was sufficiently accurate and quite reproducible to select that point during the melting experiment when the stirrer first became free to move up and down as corresponding to a 1:1 liquid-solid ratio.

Table I. Estimation of T_o from Various Analyses of Malathion

Test	T_m	$T_{1/2}$	$T_m - T_{1/2}$	T_o (Calcd.)	Purity, Mole %
1	3.44	3.04	0.40	3.84	98.8
2	3.32	3.06	0.26	3.58	98.3
3	3.14	2.65	0.49	3.63	97.7
4	3.46	3.24	0.22	3.68	98.9
5	3.50	3.30	0.20	3.70	99.08
6	3.56	3.47	0.09	3.65	99.36
7	3.54	3.30	0.24	3.78	99.26
8	3.40	3.03	0.37	3.77	98.6
9	3.48	3.27	0.21	3.69	99.0
10	3.07	2.34	0.73	3.80	97.1
11	2.49	1.42	1.07	3.56	94.4

The concordance of results for T_o in Table I also confirms the validity of the method for selecting $T_{1/2}$. The average of 11 runs is 3.70° C. + 0.09° std. dev. Runs 4, 5, 6, 7, 8, and 9, with the highest melting points, have the best "clustering" of shots around the average T_o of the whole 11 points.

The standard deviation of T_o is 0.09° C. Experience has shown that the precision of T_o determined by the above method is not better than 1/4 to 1/2 of the difference between T_m and $T_{1/2}$. This is apparent from examination of the $T_m - T_{1/2}$ values in Table I.

The cryoscopic constant K for malathion is easily determined by deliberately adding various impurities and measuring the new T_m. The compounds added were diethyl fumarate and *O,O,S*-trimethyl phosphorodithioate. These were selected because they are possible contaminants in malathion. Figure 1 shows a plot of mole % malathion versus melting temperature. The cryoscopic constant K is the reciprocal of the slope at 100 mole %. In subsequent analyses of malathion, it is sufficient only to measure T_m accurately and read the corresponding value from the graph.

The results in Table I were obtained by the Witschonke method (*8*). Compounds of lower purity may be analyzed by the Taylor-Rossini method (*7*), which is unreliable above 99.5 mole % purity if the temperature sensitivity is not better than 0.01° C. In the Taylor-Rossini derivation, there is a critical algebraic expression which is very sensitive to the precision of measurement of the temperature at high purities.

Dimethoate. Dimethoate, $(CH_3O)_2PS(SCH_2CONHCH_3)$, is a solid at room temperature, melts at about 50° C., and crystallizes readily. Table II summarizes the data obtained on a number of samples of dimethoate.

Until recently, the best sample of dimethoate available had a melting point corresponding to a purity of 99.2 mole %. At this level of purity, both freezing point and melting point agreed with each other—i.e., one could cool liquid dimethoate to the crystallization temperature or heat solid dimethoate to the melting point and get the same answer (see runs 1, 2, and 3 in Table II). Re-

Table II. Determination of Purity of Dimethoate

Run No.	T_f	T_m	T_o (Calcd.)	Purity, Mole %
1	49.10	49.13	50.30	97.0
2	49.13	49.18	50.56	96.4
3	49.96	49.91	50.25	99.20
4	50.32	50.54	...	99.87
5	50.19	50.48	50.59	99.67
6	...	50.64	50.69	99.87
7	...	50.62	50.69	99.82
8	...	50.58	50.72	99.64

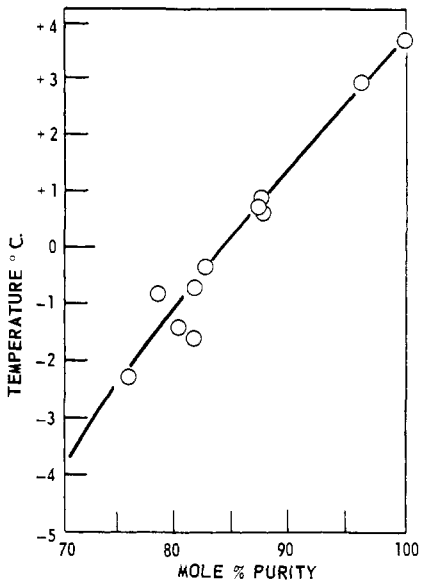

Figure 1. Malathion: temperature of melting vs. mole % purity

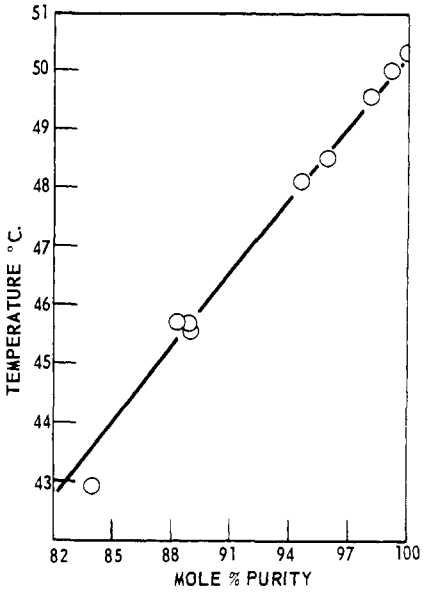

Figure 2. Dimethoate: temperature of freezing vs. mole % purity

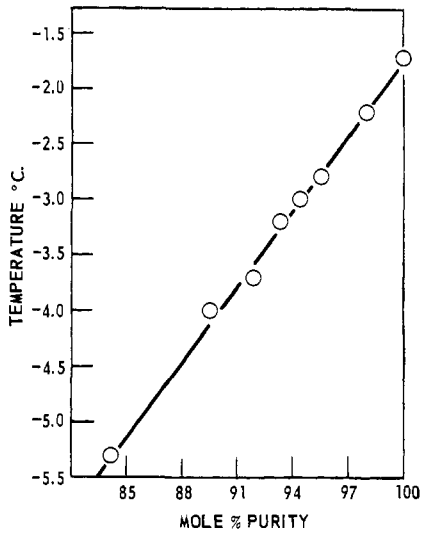

Figure 3. O,O-Diethyl O-2-pyrazinyl phosphorothioate: temperature of melting vs. mole % purity

cently, a specially purified sample of dimethoate, received from the Cyanamid Research Center, was analyzed by the cryoscopic technique and the Witschonke method and yielded the analysis 99.75 ± 0.13% as received. Pumping under vacuum overnight at room temperature increased the assay by 0.12%.

An interesting point developed with this sample. The cited analyses were obtained only with melting curves; attempts to get freezing curves for these samples ran into a supercooling problem. In contrast to lower purity samples, these 99.8% samples supercooled to the extent of one degree even with seeding and then recovered so slowly that they did not return to equilibrium condition within one-half hour. As a result, it was not possible to apply any calculation to the freezing curves. In runs 4 and 5 in Table II, the discrepancy between T_m and the apparent value of T_f is very large. The cause of this difference in behavior between moderately pure and highly pure samples is not known. Possibly the presence of impurity which does not crystallize with the major constituent may increase the mobility of the molecules at the solid-liquid interface, acting perhaps to reduce the local viscosity, especially if the impurity molecules are smaller than the major constituent molecules.

The cryoscopic constant was determined by adding toluene, heptane, and *O,O,S*-trimethylphosphorodithioate to dimethoate and observing the depression of the melting point (Figure 2). Criteria for selection of the added compounds will be described in the Discussion. The analyses of samples of dimethoate from 85 to 100 mole % pure can be determined from the measured melting point and Figure 2.

***O,O*-Diethyl *O*-2-Pyrazinyl Phosphorothioate.** This compound, useful as a nematocide, can be readily crystallized and melts slightly below 0° C. It has an ultraviolet absorption peak at 2700 A. and can be analyzed by this method provided no other compound interferes at this wavelength. The cryoscopic method is not affected by ultraviolet absorbing species, but it does give the answer in mole % rather than weight %.

The sample of *O,O*-diethyl *O*-2-pyrazinyl phosphorothioate was crystallized by cooling well below the freezing point and then a time-temperature melting curve was run on the constant temperature differential apparatus. The data were analyzed by the Taylor-Rossini method (*7*) and yielded a T_m of −2.21° C. and a T_0 of −1.67 ± 0.1° C. Following this, methyl ethyl ketone, *O,O*-diethyl *O*-phenyl phosphorothioate, and ethanol were added in known quantities and new melting points were measured. Figure 3 shows the graph of melting point versus mole % purity. The cryoscopic constant K has a value of 4.41 mole % per degree. The purity of the original purified material is thus calculated to be 97.6 mole % from the formula

$$P = 100 - K\Delta T$$
$$= 100 - 4.41\ (-1.67° + 2.21°)$$
$$= 97.6\%$$

Phorate. Phorate, $(C_2H_5O)_2PS(SCH_2SC_2H_5)$, is the lowest melting compound the authors analyzed by cryoscopy. The compound melts at about −44° C. It is difficult to get crystalline phorate, but this was accomplished by holding in dry ice for several days and then seeding with dimethoate (which is structurally similar). When the initial batch of phorate had been obtained, it was stored in dry ice and additional crystalline phorate could be obtained in minutes by seeding with this. Like the other organic phosphates, this material tends to supercool even with seeding, and it recovers so slowly that melting curves had to be performed. Since the temperature of melting was below the

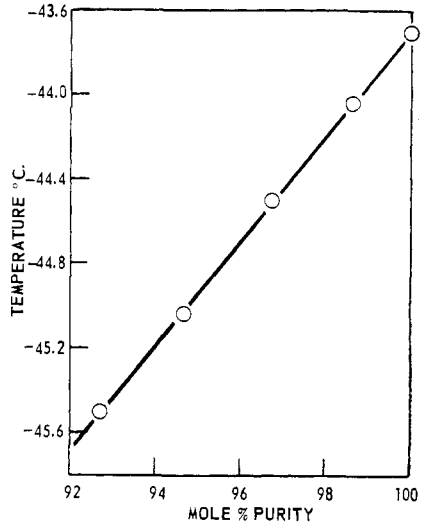

Figure 4. Phorate: temperature of melting vs. mole % purity

Table III. Summary of Cryoscopic Data

Compound	T_f, °C.	T_o, °C.	K, Mole % per °C.	Purity, Mole %	Heat of Fusion, Col. per Mole
Malathion	3.56	3.70	4.49	99.4	6850
Cygon[a] dimethoate	50.64	50.69	2.56	99.87	5340
Zinophos[a,b]	−2.21	−1.67	4.41	97.9	6470
Thimet[a] phorate	−44.04	−43.7	4.03	98.6	4220

[a] Trademark of American Cyanamid Co.
[b] Trademark for *O,O*-diethyl *O*-2-pyrazinyl phosphorothioate.

limit of the constant temperature differential instrument, a constant temperature bath a few degrees warmer than the melting point was employed. The method of analysis of the warming curve was similar to that described by Mair, Glasgow, and Rossini (*3*). The T_m of the sample was −44.04° C.; estimated T_o was −43.7° C. The cryoscopic constant K was determined in the usual manner, adding malathion, toluene, and methyl isobutyl ketone and measuring the depression. The results are illustrated in Figure 4. The sample is 98.6 mole % pure.

Discussion

Table III summarizes the results of this work. The heat of fusion has been calculated from the cryoscopic constant using the relationship

$$K = \frac{100\ \Delta H_f}{RT_o^2}$$

The entropy of fusion can also be calculated from the relationship $\Delta S_f = \Delta H_f / T_o$.

For the determination of the cryoscopic constant, the compounds to be added are selected with several views in mind. Probable or possible impurities are included. Different types of compounds such as polar and nonpolar, aromatic and paraffinic, isomeric or other related structures are included. Not every one of these classes has to be included for every analysis, but several types are recommended. The purpose of the selection is to establish whether a solid solution forms with any of these added compounds. If a solid solution forms, the normal treatment of the data is not valid. If the graph of melting point *vs.* mole % purity is a smooth curve, there is assurance that a solid solution is not formed with the compounds selected.

If successive cryoscopic runs on the same sample show a drift in the analysis toward higher purity, low molecular weight impurities may be suspected. In this case, a significant difference will exist between weight and mole percent purity. The effect of degassing on the cryoscopic result indicates the extent of this difference.

Once T_o and K are determined accurately by the analysis of the time-temperature curve of the pure sample, it is not necessary to obtain a complete melting curve for a subsequent sample, only for temperature of final melting. This melting temperature may be obtained with a simple Beckmann melting point apparatus.

The cryoscopic method is one which does not rely on a prior standard compound—its results depend on the care taken in ensuring thermal and phase equilibrium during the melting and the precision of the temperature measurement. It is primarily useful for high assay materials—i.e., 90 to 100% compounds—but in many instances may be used for analyses down to the eutectic composition. It is most precise as the purity approaches 100%.

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