

CHROM. 3498

**Thin-layer chromatography; relative migration data ( $R_{TDE}$ ) of chlorinated pesticides**

Thin-layer chromatography (TLC) is the technique most widely used to confirm the identity of pesticide residues found present by gas chromatography. The TLC systems described by Kovacs<sup>1-3</sup> are routinely employed in a number of laboratories and provide the basis of the U.S. Food and Drug Administration's application of this technique. For the most effective use of TLC in a multiple residue determination, migration values should be available for a large number of pesticide chemicals. Kovacs calculated the ratio of distance traveled by the pesticide to the distance traveled by a reference compound, *p,p'*-TDE, to indicate the migration distance ( $R_{TDE}$ ) of the respective pesticide. However, his data are limited to 12 compounds, with one solvent system and with two different adsorbents. Other workers<sup>4</sup> have determined migration relative to *p,p'*-TDE for 26 compounds with each of two solvent systems, but values were based on only one or two determinations per compound.

The objective of this work was to determine  $R_{TDE}$  values for a large number of chlorinated pesticide chemicals using three solvent systems reported by Kovacs<sup>1,3</sup> and a large number of replicates on several plates.

*Experimental*

Aluminum oxide adsorbent layers of 8 × 8 in. and of 250 μ thickness were prepared according to Kovacs<sup>1</sup>, except that air drying for 3 days was substituted for oven drying and the plates were not prewashed. Three solvent systems were used: (1) *n*-heptane, (2) 2% acetone in *n*-heptane, and (3) iso-octane with dimethylformamide (DMF) immobile phase. Merck Aluminum Oxide G with 10% CaSO<sub>4</sub> binder was the adsorbent with the first two solvent systems. Neutral Alumina AG-7 with 15% CaSO<sub>4</sub> binder (Bio-Rad) was used with the DMF immobile phase. The DMF immobile phase was applied by dipping the prepared plate into a solution of 25% DMF in ethyl ether. Before development with *n*-heptane or 2% acetone-*n*-heptane, the tank was pre-saturated for at least 30 min with the respective solvent. Plates developed with *n*-heptane or 2% acetone-*n*-heptane were air dried for 5 min before spraying. Plates with the DMF immobile phase were dried at 50° for 10 min before spraying. The AgNO<sub>3</sub> chromogenic reagent was as described by Kovacs<sup>3</sup> except that the water content was increased to 10 ml. After the plates were sprayed, they were air dried for 15 min before ultraviolet irradiation.

$R_{TDE}$  values were determined for 44 pesticide chemicals with the *n*-heptane and 2% acetone-*n*-heptane solvent systems, and for 40 compounds with the iso-octane solvent-DMF immobile phase system. *p,p'*-TDE was spotted either in admixture with or adjacent to the compound being measured. The quantity spotted was dependent on the compound and ranged from 80-200 ng. The  $R_{TDE}$  was calculated by dividing the distance from the origin to the center of the spot being determined by the comparable distance for *p,p'*-TDE. Distance of travel was measured to the nearest millimeter. The  $R_{TDE}$  for each compound was determined at least twice on a single plate and the determinations were repeated on at least two additional plates with the *n*-heptane and 2% acetone-*n*-heptane systems. The  $R_{TDE}$  values for 8 to 15 determinations for

TABLE I

MIGRATION OF CHLORINATED PESTICIDES RELATIVE TO *p,p'*-TDE BY THIN-LAYER CHROMATOGRAPHY

Plate size, 8 × 8 in.; developing tank, 9 × 9 × 3.5 in.; temperature, 24–26°; front travel, 10 cm; detection, AgNO<sub>3</sub>.

System I: Thin-layer, Aluminum Oxide G with 10% CaSO<sub>4</sub> (Merck), 250 μ thick, air dried 72 h at room temperature; amount spotted, 80–200 ng; tank, saturated; developing solvent, *n*-heptane; approximate travel of *p,p'*-TDE reference, 39 mm.

System II: Thin-layer, Aluminum Oxide G with 10% CaSO<sub>4</sub> (Merck), 250 μ thick, air dried 72 h at room temperature; amount spotted, 80–200 ng; tank, saturated; developing solvent, 2% acetone in *n*-heptane; approximate travel of *p,p'*-TDE reference, 57 mm.

System III: Thin-layer, aluminum oxide with 15% CaSO<sub>4</sub> (Bio-Rad Neutral, AG-7), 250 μ thick, air dried 72 h at room temperature; amount spotted, 200–400 ng; developing solvent, 25% DMF in ethyl ether for immobile phase, iso-octane for mobile phase; approximate travel of *p,p'*-TDE reference, 16 mm.

Pesticide	<i>R</i> <sub>TDE</sub>		
	System I	System II	System III
Hexachlorobenzene	2.7	1.7	— <sup>c</sup>
Aldrin	2.1	1.4	4.3
<i>p,p'</i> -DDE	2.0	1.4	3.4
Heptachlor	2.0	1.4	3.7
Chlordane	2.0, 1.8, 1.4, 1.2 <sup>a,b</sup>	1.4, 1.3, 1.2, 1.1 <sup>a,b</sup>	3.7, 3.4, 2.9, 1.6 <sup>a,b</sup>
<i>o,p'</i> -DDT	1.9	1.3	3.0
PCNB	1.8	1.4	3.7
Perthane olefin	1.8	1.4	4.4
<i>p,p'</i> -TDE olefin	1.8	1.4	3.2
TCNB	1.7	1.3	3.2
Telodrin®	1.7	1.4	3.5
Toxaphene	1.7, 1.2 <sup>b</sup>	1.3, 1.2 <sup>b</sup>	3.0, 2.3 <sup>b</sup>
Strobane®	1.7, 1.2 <sup>b</sup>	1.3, 1.2 <sup>b</sup>	3.0, 2.3 <sup>b</sup>
<i>p,p'</i> -DDT	1.6	1.2	2.2
<i>o,p'</i> -TDE olefin	1.6	1.3	2.9, 2.2
Chlorbenside	1.3 (grey)	1.2 (fuzzy grey)	2.0, 0.0
BHC	1.3, 1.1, 0.27, 0.10 <sup>a</sup>	1.1, 0.92, 0.72, 0.25 <sup>a</sup>	1.8, 1.3, 0.75, 0.30 <sup>a</sup>
α-BHC	1.3	1.1	— <sup>c</sup>
Perthane®	1.3	1.2	2.5
Lindane	1.1	0.92	1.3
<i>o,p'</i> -TDE	1.1	0.95	1.1
<i>p,p'</i> -TDE	1.0	1.0	1.00
Endosulfan	0.88, 0.07 <sup>a</sup>	0.92, 0.24 <sup>a</sup>	3.1, 0.0 <sup>a</sup>
Ronnel	0.85	1.1	2.2
Heptachlor epoxide	0.71	1.0	2.4
Endrin	0.71	1.0	2.9
Dieldrin	0.52	0.90	2.8
Carbophenothion	0.42 (yellow)	1.0 (fuzzy yellow)	1.9
Methoxychlor	0.33, 0.27	0.79	0.85
β-BHC	0.27	0.72	— <sup>c</sup>
Ovex	0.18	0.76	0.61
Dichlone	0.16	0.72, 0.00 <sup>a</sup>	0.00
Dyrene®	0.15 (grey)	0.51	0.20
Tetradifon	0.11	0.82	0.90
δ-BHC	0.10	0.25	— <sup>c</sup>
Endrin ketone			
(Delta-Keto 153)	0.09	0.23 (very small)	0.56
Kelthane®	0.06	0.28	0.00
Sulphenone®	0.00 (large and fuzzy)	0.31 (yellow)	0.00
Captan	0.00 (sharp edged grey)	0.09	0.25
Chlorobenzilate	0.00 (light)	0.05	0.25
Monuron	0.00 (large and dark)	0.00	0.00
Diuron	0.00	0.00 (dark spot)	0.00
Endrin aldehyde	0.00 (very small)	0.00 (very small)	0.23, 0.00 <sup>a</sup>
Endrin alcohol	0.00	0.00	0.14, 0.00 <sup>a</sup>

<sup>a</sup> Most intense spot underlined.

<sup>b</sup> Leaves a streak with these major spots.

<sup>c</sup> Not determined

each compound on 8 to 11 plates were averaged to obtain the  $R_{TDE}$  values for the *n*-heptane system. Six to eight determinations for each compound on three or four plates were averaged to obtain the  $R_{TDE}$  values for the 2% acetone-*n*-heptane system.  $R_{TDE}$  values for the DMF immobile phase-iso-octane system were determined by averaging the values obtained from single determinations on three separate plates; 200-400 ng of each compound was spotted.

### *Results and discussion*

Table I lists the  $R_{TDE}$  values for more than 40 chlorinated pesticide chemicals with *n*-heptane, 2% acetone-*n*-heptane and DMF immobile phase-iso-octane solvent systems, respectively. These values should be a helpful reference when utilizing TLC to obtain information to confirm the identity of chlorinated pesticide residues. Numerical values may not be exactly reproducible among all analysts or among replicate determinations; however, the sequence should remain unchanged. Since  $R_{TDE}$  values are listed for a larger number of compounds for these solvent systems than previously reported, possible alternate identity assignments may become apparent in some cases. In other situations, certain compounds may be eliminated from consideration. These data should also quickly indicate possible separations with the three solvent systems.

To obtain the maximum value from this tabulation, users should add  $R_{TDE}$  data for additional important pesticide chemicals.

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