In preparing the retinal antigen, a clear-cut separation of the retina from adjacent tissues, particularly the uvea at the ora serrata, is difficult if not impossible. The contamination of the retinal antigen could not be quantitated; variations in the admixture of nonretinal tissue may have influenced the ocular response.

A possibility other than the inadvertent contamination of retinal antigen with uveal antigen, or that of cross antigenicity, is the natural anatomic association of disparate tissue antigens as exemplified by the presence of myoid cells in thymic tissue (*6*). Bullington and Waksman (*7*) and Wacker and Lipton (*8*) reported the induction of uveitis by immunization with optic nerve and with retinal antigens, respectively. Although Bullington and Waksman (*7*) and Wacker and Lipton (*9*) failed to produce uveitis in rabbits by immunization with rabbit uvea, this has been done by Aronson (*10*). Thus, the suggestion by Wacker and Lipton (*9*) that the antigen responsible for the uveitis is a primarily retinal component remains to be confirmed.

After sensitization with retinal antigen, the eye was the only organ to show disease change; no involvement of the central nervous system or any other organ system was noted clinically in any of these animals.

The mechanism of the retinopathy (*1*), which is elicited by injection of central nervous system tissue, remains obscure. The possibility had to be considered that monkey retina might contain myelinated components as is the case in the rabbit retina (*7*). The absence of AE in monkeys injected with monkey retina and Freund's complete adjuvant is an argument against this possibility, although quantitative considerations prevent excluding it completely. To examine the possible role of vascular antigenicity in HR in producing eye lesions, heterologous kidney, which contains large amounts of vascular tissue and basement membrane, and homologous choroid plexus were used as controls. The choroid plexus was particularly suitable since it is a highly vascular tissue located within the brain itself. Although data are lacking for clarification of the pathogenesis of the HR, our results with the chorioretinitis suggest that the lesion induced in rhesus monkeys by injection of homologous retinal tissue in Freund's complete adjuvant is an autoimmune disease of organ- and species-specific nature.

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Pesticide Mobility: Determination by Soil Thin-Layer Chromatography

Abstract. *Pesticide movement was evaluated by the comparison of* R_F *values on thin layers of soils. Results from the new technique correlated well with existing information on pesticide movement, facilitating the grouping of pesticides into classes on the basis of mobility. Thin-layer chromatography may have broad applicability in soils research.*

Movement of a pesticide from its site of application is one cause of environmental contamination. Thus, mobility data are useful in evaluating the persistence of applied chemicals, as well as in defining conditions associated with pesticide use. Such information, however, is limited by the expense and difficulty of conducting leaching studies (*1*). Lack of a standard method makes comparisons among pesticides and soils difficult.

We report a new approach to the investigation of pesticide mobility—the use of untreated soil as an adsorbent in thin-layer chromatography (TLC). The new method is termed "soil thin-layer chromatography." Soil TLC is rapid, reproducible, and inexpensive; requirements for equipment, laboratory space, test chemicals, and soil are modest. A quantitative index of relative pesticide mobility, based on R_F values, correlated well with published observations on movement.

Three soils from Ap horizons (a layer 0–6 inches, cultivated soil) which differ substantially in texture and content of organic matter were studied: Lakeland sandy loam (12.0 percent clay, 0.9 percent organic matter), Chillum silt loam (26.3 percent clay, 3.1 percent organic matter), and Hagerstown silty clay loam (39.5 percent clay, 2.5 percent organic matter). Medium sand (> 250 μ thick) was removed by dry-sieving from Chillum and Hagerstown soils, and coarse sand (> 500 μ) by dry-sieving from Lakeland sandy loam prior to chromatography.

Conventional TLC apparatus was used to prepare most soil plates. Immediately before spreading, a slurry of soil and water was prepared. Chillum and Hagerstown layers (500 μ thick) were prepared with a variable-thickness spreader, and the Lakeland layer (750 μ) was prepared with a glass rod moved over masking tape along the plate edges (*2*). Six or seven ^{14}C-labeled pesticides (3 to 10 μg each) were aplied to a plate (20 by 20 cm) and developed 10 cm with water by ascending chromatography. The plates were visualized by autoradiography with "no-screen" medical x-ray film.

The relative mobilities of six herbicides are readily differentiated in Fig. 1. Little tailing was noted for the highly mobile dicamba (*3*), indicating that the adsorption isotherm is nearly linear. Compounds of slightly lower mobility (for example, amiben, fenac, and 2,4-D) exhibit increased tailing, resembling the movement of tritiated water in a clay soil (*4*). With monuron and other less mobile compounds, movement is seen as a continuous streaking or elution from the origin.

In the comparison of pesticide compounds, we measured R_F values as the front of a streak or spot. This value changed only slightly in the range of sample size from 0.5 to 200 μg. Streak-

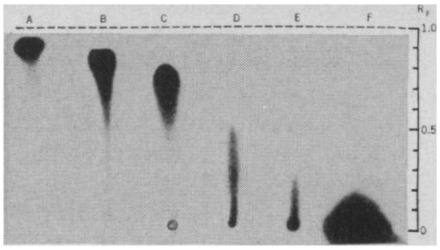

Fig. 1. Autoradiogram showing movement of six herbicides on a Hagerstown silty clay loam plate (19 cm by 500 μ) by soil TLC. (*A*) dicamba; (*B*) amiben; (*C*) fenac; (*D*) monuron; (*E*) diuron; (*F*) CIPC.

ing increased with sample size. The streaking is probably similar to movement within a soil profile, where dissolution and adsorption-desorption processes replenish the percolating soil water. The analysis in depth of many column and field experiments corroborates this soil TLC picture (*5*).

In addition to defining R_F, the autoradiograph also indicates the diffusion of pesticides in soil. This process may account for much of the observed movement of CIPC (both vertical and lateral), although it is not certain whether diffusion in the vapor or liquid phase predominates (*6*). Among other pesticides which appear to diffuse under conditions of soil TLC are diphenamid and atrazine. Using moist (field moisture capacity) soil plates (*7*), with no mass transfer of water, we observed substantial diffusion with dicamba and 2,4-D; on dry plates, only CIPC diffused.

Table 1. Mobility of pesticides determined by soil TLC and expressed as R_F values. The R_F values in the same column followed by the same letter designation are not significantly different at the 5 percent level; these values were derived by Kramer's method for unequal repetitions of Duncan's multiple range (*15*).

Pesticide	R_F value on		
	Chillum silt loam	Hagerstown silty clay loam	Lakeland sandy loam
Dicamba	0.96a	0.96a	1.00a
Amiben	.87b	.91b	1.00a
Fenac	.62c	.84c	1.00a
MCPA	.62c	.78d	1.00a
2,4-D	.50d	.69e	1.00a
Diphenamid	.39ef	.49f	0.94ab
Monuron	.44de	.48f	.89b
Atrazine	.35fg	.47f	.89b
Simazine	.31g	.45fg	.96ab
Propazine	.24h	.41g	.77c
Diuron	.23h	.24h	.60d
Prometryne	.08ij	.25h	.37e
CIPC	.13i	.18i	.59d
Diquat	.04j	.06j	.19f
Paraquat	.00j	.00k	.13f
Trifluralin	.00j	.00k	.00g

The mobilities of 16 herbicides are expressed in Table 1 as R_F values; the R_F value is a quantitative indication of the front of pesticide movement and a reproducible index of mobility. Pesticides differing by more than 0.06 R_F units were distinguishable because of the small errors involved.

Enough detail has been reported to permit an estimate of mobility as an R_F value. These column-leaching experiments, despite inherent differences within soil types, show striking correlation with the results from soil TLC: for dicamba on loam and sandy loam soils, $R_F = 0.95–1.0$ (*8*); fenac on silty clay loam, 0.7 (*9*); monuron on silty clay loam, 0.2–0.5 (*10*); silty clay loam, 0.4 (*9*); sand, 0.7 (*11*); diuron on silty clay loam, 0–0.3 (*10*); and CIPC on silty clay loam, 0–0.1 (*10*). Furthermore, the ranking of mobility in Table 1 corresponds well to a consensus of the mobility literature (*9, 12, 13*).

Differences in soil type were easily distinguished by the TLC method. In every case, the mobility of a pesticide increased from Chillum to Hagerstown to Lakeland soil, as Harris (*13, 14*) observed with columns of identical soils. Average R_F values for all pesticides used, which increase from 0.36 to 0.47 to 0.70, indicate the effect of the decreasing content of organic matter in soil. On Lakeland soil the most mobile pesticides migrated with the water front and were indistinguishable by R_F values; however, some could be differentiated by the amount of streaking on the autoradiograph.

A general classification of pesticide mobility would be of value in many aspects of pesticide behavior in soils. If a single, nationally recognized soil standard were used, the classification could be defined by R_F groupings with the soil TLC technique.

In lieu of a standard soil, a system of five classes of pesticides arranged on the basis of mobility is presented. The classification is based on R_F values from the Hagerstown and Chillum soils and is consistent with most literature on pesticide mobility. The mobility scheme comprises the following R_F ranges from Hagerstown: class 1, 0–0.09; class 2, 0.10–0.34; class 3, 0.35–0.64; class 4, 0.65–0.89; and class 5, 0.90–1.00. Pesticides, listed by and within classes of decreasing mobility, include: class 5, dicamba, dalapon, amiben; class 4, picloram, fenac, MCPA, 2,4-D; class 3, diphenamid, monuron, atrazine, simazine; class 2, diuron, prometryne, siduron, CIPC, azinphosmethyl; and class 1, diquat, paraquat, dieldrin, trifluralin, and heptachlor. As expected, acidic herbicides are among the most mobile; insecticides containing chlorinated hydrocarbons are among the least mobile.

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3. Common and chemical names of pesticides cited are: amiben: 3-amino-2,5-dichlorobenzoic acid; atrazine: 2-chloro-4-ethylamino-6-isopropylamino-*s*-triazine; azinphosmethyl: *O,O*-dimethyl *S*-[4-oxo-1,2,3-benzotriazin-3(4*H*)-ylmethyl] phosphorodithioate; CIPC: isopropyl *N*-(3-chlorophenyl) carbamate; 2,4-D: 2,4-dichlorophenoxyacetic acid; dalapon: 2,2-dichloropropionic acid; dicamba: 2-methoxy-3,6-dichlorobenzoic acid; dieldrin: 1,2,3,4,10,10-hexachloro - 6,7 - epoxy - 1,4,4a,5,6,7,8,8a - octahydro - 1,4-*endo,exo*-5,8-dimethanonaphthalene; diphenamid: *N,N*-dimethyl-2,2-diphenylacetamide; diquat: 6,7-dihydrodipyrido[1,2,-a: 2′,1′-*c*]pyrazidinium salt; diuron: 3-(3,4-dichlorophenyl)-1,1-dimethylurea; fenac: 2,3,6-trichlorophenylacetic acid; heptachlor: 1,4,5,6,7,8,8 - heptachloro-3a,4,7,7a-tetrahydro-4,7-methanoindene; MCPA: 4-chloro-2-methylphenoxyacetic acid; monuron: 3-(3-chlorophenyl)-1,1-dimethylurea; paraquat: 1,1′-dimethyl-4,4′-bipyridinium salt; picloram: 4-amino-3,5,6-trichloropicolinic acid; prometryne: 2-methylmercapto-4,6-bis(isopropylamino)-*s*-triazine;propazine: 2-chloro-4,6-bis(isopropylamino)-*s*-triazine; siduron: 1-(2-methylcyclohexyl)-3-phenylurea; simazine: 2-chloro-4,6-bis(ethylamino)-*s*-triazine: trifluralin: α,α,α,-trifluoro-2, 6-dinitro-*N,N*-dipropyl-*p*-toluidine.
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