A Rapid On-Column Extraction-Cleanup Method for Animal Fat[1]

by W. P. CAHILL, B. J. ESTESEN, and G. W. WARE
Department of Entomology, The University of Arizona, Tucson, Arizona

Several methods of extraction and cleanup are currently in use for ECGC analysis of chlorinated insecticide residues in animal fat. Generally they include grinding with sand, sodium sulfate, or with a tissue homogenizer; extraction with petroleum ether, ethyl ether, hexane, or a mixture of polar and apolar solvents; partitioning into acetonitrile or dimethyl formamide; redissolving in an apolar solvent followed by Florisil column cleanup. All of these require much time, involve many pieces of glassware and consequently suffer surface adsorption losses in transferring from one container to another.

We have found a very simple, rapid and efficient method for the extraction and cleanup of animal fat tissue directly on the Florisil column.

Up to 0.8 gram of the frozen fat tissue is shaved onto aluminum foil with a scalpel and weighed to the nearest milligram. Using a powder funnel the slices are transferred to a 4" activated Florisil column, capped with 0.5" sodium sulfate and prewetted with 50 ml of hexane. Our best results have been obtained by activating the Florisil at 120-130° C for 24 hours and storing at this temperature. The foil is then reweighed, subtracting the adhering fat weight from the original. The funnel and column walls are rinsed with 25 ml of hexane.

The sample is allowed to dissolve for 5 minutes before eluting the 25 ml hexane rinse from the column at 1 drop/3 seconds. When the hexane has entered the column bed the walls and column are rinsed 3 times with 5 ml volumes of hexane. The column is then eluted with 200 ml of 5% ethyl ether in hexane at 1 drop/second. Though the connective tissue remains intact and inflated with solvent after elution, all lipids have been completely dissolved from the sample.

---

[1]Contribution to Regional Project W-45, "Residues of Selected Pesticides--Their Nature, Distribution, and Persistence in Plants, Animals and the Physical Environment." University of Arizona Agricultural Experiment Station journal series #1565.

Bulletin of Environmental Contamination & Toxicology,
Vol. 5. No. 1. 1970. published by Springer-Verlag New York. Inc.

Results of this technique are presented in the table and compared with the previously-used method for DDE, DDD and p,p' DDT residues in the same beef fat sample. The latter involved grinding in a tissue homogenizer with 5 ml each of ethyl ether, hexane and methanol, transferring to a separatory funnel with ether rinses, washing with water, and transferring to a beaker with petroleum ether rinses. From this stage on the older method ends by cleanup on the Florisil column. It will be seen that the range of results was less varied and recoveries were notably higher from the on-column method.

Using the on-column extraction method for repeat samplings, the relative coefficient of variance for DDE values only was 5.92%. Repeat injections and analysis by ECGC of the same sample resulted in a 4.7% relative coefficient of variance.

We recommend this combination extraction-cleanup method for speed and economy of laboratory glassware.

TABLE

Results (PPM) of replicated beef fat extractions by two methods.

DDE	DDD	p,p' DDT	Total
	Tissue Grinder		
2.83	.074	----	----
2.77	.10	1.61	4.48
2.75	.10	1.47	4.32
2.80	.093	1.53	4.42
2.76	.11	1.48	4.35
			$\bar{X}$ = 4.39
	On-Column Extraction		
2.97	.11	1.51	4.59
2.93	.11	1.64	4.68
2.95	.11	1.48	4.54
2.79	.11	1.63	4.53
			$\bar{X}$ = 4.59