Dichlorodiphenyltrichloroethane and Dichlorodiphenyldichloroethylene Content of Prepared Meals

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Small quantities of DDT and DDE have been found in the body fat of humans. DDT has had wide use on food and forage crops and for control of insect-borne diseases. DDT and DDE were found in all meals analyzed, but in small quantities, and are not considered a toxicological health hazard. In the case of foods intended for human consumption, the DDE content as well as the DDT content should be determined.

THE FINDING of small quantities of dichlorodiphenyltrichloroethane (DDT) and dichlorodiphenyldichloroethylene (DDE) in the body fat of humans (*16*, *18*) with no known direct exposure to DDT or DDE raises the question of the quantity of these compounds ingested with normal food intake.

The use of DDT for the control of insects affecting man and animals is well established and the compound has been applied to almost all food and forage crops. Though not currently recommended, DDT has been applied to dairy cattle and dairy barns for fly control and its secretion in butterfat has been established (*15*), as it has in dairy cattle fed vine silage or alfalfa hay containing DDT (*4*, *19*). Foodstuffs have been contaminated by the impregnation of sacking (*2*) and DDT has been added to drinking water for the control of mosquitoes (*12*). DDT spray residues have been reported on fruits (*3*), on bread and cake made from DDT-treated flours (*11*), and on milled products (*10*).

The results of experimental feeding of DDT to dogs, its accumulation in the body fat, and its appearance in the milk of dogs have been reported (*22*). The storage of DDT in the tissue of pigs (*5*) and sheep (*13*) and its transfer from feed to eggs and body tissue of chickens (*6*) under experimental conditions have also been reported.

The storage of DDT in human fat was reported in 1948 (*14*) and in human milk in 1951 (*9*). Pearce, Mattson, and Hayes (*18*) pointed out that much of the reported DDT in human fat was actually the degradation product, DDE.

Methods

Collection Of Meals

To determine the amounts of DDT and DDE ingested during normal food intake, 18 meals were obtained from restaurants and 7 were obtained from a correctional institution. Most of the food prepared by the restaurants was not locally grown. Much of the food consumed at the correctional institution was produced within the institution itself. Regional items, such as fresh sea food and unusual foods, were avoided, so that a representative cross section of food items consumed by the public could be obtained. Home-cooked meals also were avoided because of the difficulty of obtaining a representative cross section.

The individual items comprising the meals were collected in chemically clean glassware directly from the kitchen of the restaurant or the institution, in order to obtain the samples in containers known to be free of DDT and to prevent the contamination of one food with another. In some instances the portions obtained were much larger than those usually served. Where such discrepancies were noted, the samples were reduced to reasonable portions before proceeding with the analysis. Upon receipt of the samples at the laboratory, each item was weighed after the inedible portions had been removed.

Extraction And Analysis

The edible portions were extracted with acetone in a blender and filtered, and the filter cake was extracted with carbon tetrachloride in a large Soxhlet extractor. The two extracts were concentrated and combined for analysis. Prior to the final analysis, the combined extracts were chromatographed by a modification of the method of Davidow (*21*) for the removal of interfering fats and plant pigments. In certain individual portions the fat content was fairly high. Because it is not practical to remove more than 2 or 3 grams of fatty material with the modified Davidow column, an aliquot of the samples was chromatographed. If the amount of DDT and/or DDE in this aliquot was less than the lower limit of sensitivity of the method, such samples were reported on the basis of the aliquot taken and not on the basis of the entire sample; the entire samples may have contained determinable amounts of DDT or DDE. DDT and DDE were determined by a modified Schechter-Haller procedure (*16*, *21*).

Prior to analysis, several of the food items were divided into equal parts. One half of each sample was analyzed in the regular manner; the other half was analyzed after addition of known amounts of DDT and DDE. The recovery of known amounts of DDT averaged 73.7% with a range of recoveries from 40.0% for French toast to 110.0% melted butter. The recovery of known amounts of DDE averaged 82.2% with a range of from 66.1% for toast to 103.0% for melted butter.

Brittin and Faring (*7*) were able to recover 51.2% of the DDT they added to strained peaches, pears, peas, green beans, and vegetable and beef soups. Tressler (*20*) recovered 89% of the DDT added to strained peaches and 61% of the DDT added to applesauce. The formation of emulsions with organic solvents used in the extraction procedure and the difficulty of breaking these emulsions in order to obtain complete recovery of the DDT-bearing organic solvent layer probably account for most of the added DDT lost.

The analytical method employed enables estimation of 5 γ of either DDT or DDE with an accuracy of approximately 10%. Values below 5 γ for either compound may be subject to considerable error due to the effect of interfering colors from the sample. For this reason values below 5 γ can be considered only an approximation. One microgram of DDT and/or DDE can be detected in control samples where no interfering colors are present. In totaling the individual food items of the 25 meals, the value of 2.5 γ was used for food items that contained between 1 and 5 γ of DDT or DDE.

The results obtained on the individual portions were combined into the original meals, including eight morning, nine noon, and nine evening meals. Table I presents a summary of the analyses of these meals.

Results

As will be noted, portions of nine meals were lost during analysis. Analysis of subsequent samples indicated that the loss of samples of fried eggs from meal 3, and of cherry pie a la mode and sliced lemon from meal 18, may have affected the total DDT and or DDE content of these two meals. The loss of items from the other meals probably did not reduce the total DDT and/or DDE content of these meals by more than 5 γ per item. The effect of the loss of a sample of grapefruit from meal 16-IM could not be determined, as only one sample of grapefruit was obtained in all the meals analyzed.

One of the significant results of these analyses is the finding of relatively large portions of DDE in the meals. In individual meals the ratio of DDE to DDT ranged from 0.21 to 1 up to 1.55 to 1 with an average of 0.50 to 1. Three noon meals contained more DDE than DDT, but in each case a large amount of the DDE was confined to a few food items. The finding of DDE in food emphasizes the importance of separating DDE from DDT in studies of residues on food products ultimately intended for human consumption; not only does the DDE represent less potential health hazard while stored in the body fat, but a portion of the DDE found in the fat may have never existed as the more toxic DDT in the human body.

Every meal analyzed contained both DDT and DDE. Institutional meals in general contained about the same amounts of DDT and DDE as did restaurant meals, except in the cases of the morning meals. The institutional breakfasts consisted only of cereal, milk, fruit, breakfast rolls, and margarine and contained none of the fried foods which account for most of the DDT and DDE in restaurant meals.

Aside from this, the amount of DDT in the three types of meals was roughly proportional to the size of the meal. The noon and evening meals contained about the same amount of DDE regardless of size.

A total of 179 individual portions of food representing 86 different items was included in the 25 meals, and of these 40 contained insufficient DDT to be detected. Ten other samples showed no DDT, but these 10 samples had been so reduced in size in order to reduce the fat content that they cannot be included with assurance in the DDT-free group.

The individual portions of the 25 meals were divided into 21 classifications. The number of samples within a classification that contained 5 γ or more, 1 to 5 γ, and less than 1 γ of DDT and DDE are listed in Table II.

Generally those foodstuffs cooked in fat, and containing meat, meat combinations, or butter, were found to have a higher DDT content than foodstuffs not cooked with fat, and not containing meat, meat combinations, or butter. DDE content was roughly porportional to the DDT content. A few individual food classifications are worthy of note.

Discussion

Beverages. The three samples that contained trace quantities of DDT and the two samples that contained trace quantities of DDE were coffee with added cream. All other beverages contained no cream.

Potatoes, Mashed. The two samples that contained trace quantities of DDT and DDE also contained added butter. The other six DDT-free samples contained no added butter.

Breakfast Cereals. The one sample that contained a measurable quantity of DDT was cooked oatmeal. An additional sample of cooked oatmeal did not contain a measurable quantity of DDT.

Fruit and Fruit Juices. One sample of pitted prunes (dried fruit that had been cooked) contained a measurable quantity of DDT.

Vegetables. Of the 21 samples of vegetables (peas, beans, corn, lima beans, tomatoes, mixed vegetables, swiss chard, beets, carrots, cabbage, and soups), 8 samples contained 5 γ or more of DDT and 1 sample contained 5 γ or more of DDE. Two of the 8 samples, lima beans and carrots, contained 15 and 21 γ, respectively, of DDT. The lima beans had been cooked with ham and the carrots were prepared in a white cream sauce. The creamed carrots contained DDT and DDE, while the lima beans contained DDT only.

Sea Foods. The sea foods contained a greater quantity of DDE than DDT. Two of the three samples (oysters and halibut) had been fried in deep fat; the other sample was clam chowder. No DDT but an appreciable quantity of DDE was found in the chowder.

Bread, Buttered Bread, and Sweet Rolls. More DDT was found on the buttered bread and sweet rolls (having a

Table I. DDT and DDE Found in Typical Restaurant and Institutional Meals

Meal	Total Weight (Including Beverage), Grams	Total Found, γ DDT[a]	DDE[b]	Ratio of DDE/DDT
Morning				
1	527	27.5	12.5	0.46
2	443	62.5	30.5	0.49
3	522[c]	59.5	24.5	0.41
4	728[d]	48.0	26.5	0.55
5	536[e]	70.0	47.5	0.68
6	731	27.5	8.5	0.31
7-IM	805	8.5	5.0	0.59
8-IM	383[f]	10.0	5.0	0.50
Mean	584	39.2	20.0	0.51
Noon				
9	701[g]	40.0	23.5	0.59
10	848	163.5	44.5	0.27
11	687	16.5	21.0	1.27
12	987	17.0	17.5	1.03
13	805	65.5	50.0	0.76
14	689	37.5	58.0	1.55
15-IM	1508	34.5	15.0	0.44
16-IM	652[h]	79.0	25.5	0.32
17-IM	548	71.5	30.0	0.42
Mean	825	58.3	31.7	0.54
Evening				
18	666[i]	118.0	43.0	0.36
19	900	46.0	40.5	0.88
20	1017[j]	161.5	34.5	0.21
21	1196	51.0	31.5	0.62
22	1108	32.5	27.0	0.83
23	914	34.0	12.5	0.37
24-IM	1083[k]	50.0	30.5	0.61
25-IM	1288	39.5	27.0	0.68
Mean	1022	66.6	30.8	0.46
Mean of all meals	811	54.8	27.7	0.50

IM Institutional meals.
[a] Calculated as technical DDT.
[b] Calculated as recrystallized DDE.
Amounts lost on analysis and not included in totals. [c] 140 grams. [d] 49 grams. [e] 42 grams. [f] 192 grams. [g] 471 grams. [h] 75 grams. [i] 193 grams. [j] 268 grams. [k] 51 grams.

frosting or a jelly filling) than on the unbuttered bread.

Dairy Products. Though the correctional institution reported that margarine was used, all the restaurants reported their spreads as butter. Whether or not they actually were butter was not determined, but they are included with dairy products in Table II. Because of considerable doubt as to the validity of this assumption, butter was not included in Table III. Six of these samples contained measurable amounts of DDT, four contained between 1 and 5 γ, and four contained less than 1 γ. Seven servings of oleomargarine were obtained from the correctional institution, but as they were all from the same lot, only one sample was analyzed and reported separately in Table II. It contained less than 1 γ of DDT. A milkshake contained more DDE than DDT.

Seasonings. One sample of mayonnaise and one sample of barbecue sauce contained measurable quantities of DDT and DDE. The barbecue sauce contained a little more than twice as much DDT as DDE.

Bread, Fried. The term "bread, fried" was used to cover hot cakes, pancakes, and French toast. Three samples contained measurable amounts of DDT and two out of the three contained measurable amounts of DDE.

Desserts. Three out of four samples of pie contained DDT and two of these samples contained DDE. One sample of mince pie contained more DDT than any other food item except meat.

Potato, Baked. The one sample of baked potato had a very glossy appearance, indicating that it had been brushed with melted butter. The DDT and DDE content of the baked potato (entire potato including the skin) confirmed this observation.

Meats, Gravies, and Meat Combinations. Twenty-four of the 27 samples of meats, gravies, and meat combinations (meat loaf, chile con carne, spaghetti and meat balls) had measurable quantities of DDT. Twenty out of the 27 contained measurable quantities of DDE. In one sample of boiled beef tongue there was approximately four times as much DDE as DDT. Several of the samples (such as gravies) were reduced in size, owing to their fat content, and the aliquot taken for analysis did not contain measurable quantities of DDT and/or DDE.

Potatoes, Fried. All seven samples of fried potatoes, (fried, French-fried, and hashed-brown) contained measurable quantities of DDT. Three of the seven samples contained determinable amounts of DDE.

Eliminating duplication, 28 foods are listed in Table III, one or more portions of which did not contain detectable (1 γ) amounts of DDT. Some of these foods, however, were represented by other portions which did contain measurable (5 γ or more) quantities of DDT. Most of the remaining foods listed in the table represent foods that were analyzed only once.

To date there is no precise information as to the amount of DDT which can be consumed by humans over a long period of time without the possibility of adverse results. Basing his conclusion mainly on chronic toxicity studies conducted on laboratory animals, but taking a safety factor into account, Neal (*11*) estimated that 5 mg. of DDT can be ingested daily without untoward effects. Fitzhugh (*7*) and Heyroth (*8*) estimated that man can ingest 2.5 mg. of DDT daily over a long period of time without injury.

If an individual were to consume in one day the three meals found to contain the highest amounts of DDT (meals 2, 10, and 20), his total DDT intake would be 0.388 mg. or 15.5% of the smaller amount which it has been estimated that humans could consume regularly without adverse effects. The average daily intake based on all meals would be about 0.184 mg. of DDT per day. This is equivalent to a dosage of about 0.0026 mg. per kg. per day for a man of average size or a concentration of about 0.31 p.p.m. of DDT in the total dry diet. By comparison, a rat would have to eat a diet containing about 0.05 p.p.m. to obtain the same dosage, because the rat eats more food than man in proportion to body weight.

Summary and Conclusions

DDT was present in detectable but very small quantities in all meals analyzed.

DDE was present in all meals, though in somewhat lower quantities.

Few if any foods can be relied upon to be entirely free of DDT.

Fried foods, and those having a high fat content, either natural or added, contained more DDT than other types of foods.

In general, high DDE content of foods is associated with high DDT content, though it is possible to have measurable DDE in the absence of DDT and measurable DDT in the absence of DDE.

No meals contained enough DDT to be considered a toxicological health hazard on the basis of the estimated chronic oral toxicity of the compound.

DDT residues on foods ultimately intended for human consumption should be assayed in such a way that the DDE content as well as the DDT content would be determined.

Table II. Number of Samples Containing DDT and DDE

Classification	5 γ or More		1 to 5 γ (Trace Quantities)		Less than 1 γ	
	DDT	DDE	DDT	DDE	DDT	DDE
Oleomargarine	0	0	0	0	1	1
Beverages	0	0	3	2	14	15
Potatoes, mashed	0	0	2	2	6	6
Breakfast cereals	1	0	2	4	3	2
Sirup	0	0	2	1	1	2
Salads	0	0	2	1	1	2
Potatoes, escalloped	0	0	1	1	1	1
Fruits and fruit juices	1	0	5	4	2	4
Macaroni	0	0	1	1	0	0
Vegetables	8	1	7	11	6	9
Seafoods	1	3	1	0	2	1
Bread, buttered bread, and sweet rolls	6	2	7	10	3	4
Dairy products	7	3	4	3	4	9
Seasonings	2	2	1	1	2	2
Bread, fried	3	2	0	1	0	0
Dressing, bread	1	0	0	1	0	0
Dessert	6	3	5	7	1	2
Potato, baked	1	1	0	0	0	0
Meat, gravies, and meat combinations	24	20	2	5	3	4
Potatoes, fried	7	3	0	2	0	2
Eggs, fried	2	2	0	0	0	0

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Table III. Foods Without Detectable (1 γ) Amounts of DDT

Applesauce	Corn[a]	Potatoes, mashed[a]
Beans, string[a]	Corn bread	Rolls[a]
Bean soup	Dry cereal[a]	Root beer
Cake[a]	Grapefruit juice	Steak, pan broiled[b]
Catsup	Gravy[a]	Sirup[a]
Clam chowder	Jello salad	Tea[a]
Coffee, black	Oleomargarine	Toast[a]
Cola beverage	Peas[a]	Tuna fish salad
Combination salad	Potatoes, escalloped[a]	Vegetables, mixed
		Vinegar

[a] Other portions contained a measurable amount of DDT (5 γ or more).
[b] Calculation based on aliquot chromatographed.

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BIOCHEMISTRY OF MYOGLOBIN

Quantitative Determination in Beef and Pork Muscle

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Chemical Studies with Purified Metmyoglobin

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Studies have been initiated to determine the concentration and chemical reactivity of myoglobin, the major heme pigment present in lean meat. A procedure has been developed for the quantitative determination of myoglobin in beef and pork muscle and reactions of purified metmyoglobin (Fe^{+++}) prepared from beef muscle with ascorbate to form myoglobin (Fe^{++}). Reactions of the latter compound with nitrite to form nitric oxide myoglobin were also studied. This work on quantitating the concentration and reactivity of myoglobin is of importance in evaluating the uniformity and stability of myoglobin derivatives in fresh and cured meats during storage or various processing procedures. Chemical changes in the myoglobin of meat attributable to irradiation with gamma rays from a cobalt-60 source are now being studied in this laboratory.

THE HEME PIGMENTS constitute a class of very important biochemical compounds. Much work has been done with hemoglobin and heme catalysts in oxidation (catalase, peroxidase, cytochromes, etc.), but relatively little has been done to elucidate the functions and reactions of myoglobin, the oxygen-carrying pigment in muscle.

The importance of understanding the chemical changes associated with the color of lean meat during irradiation by gamma rays (cobalt-60) or other treatments prompted studies on myoglobin in muscle.

While visual observation of beef and pork muscle indicates that considerable variation exists in the heme pigment content, few quantitative data are available on the myoglobin content of beef and pork muscle. Shenk, Hall, and King (*10*) described a method for the determination of "muscle hemoglobin" (myoglobin) and presented data for beef muscle. Crandall and Drabkin (*1*) and Drabkin (*2*) reported data on the myoglobin content of rat, human, horse, dog, and beef muscle. Husaini (*6*), Hershberger (*5*), and Weiser (*12*), and their associates, have reported twofold variation in amount of total pigment (hemoglobin plus myoglobin) in beef muscle.

As the authors were interested in measuring the changes occurring during irradiation, a method was adopted for the determination of myoglobin in beef and pork muscle.

Work was also undertaken to gain information about the in vitro chemical