

RESIDUES OF ORGANOCHLORINE PESTICIDES IN
SOME FOODSTUFFS OF ANIMAL ORIGIN COLLECTED
IN CROATIA, YUGOSLAVIA

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The organochlorine compounds p,p'-DDT (1,1,1-trichloro-2,2-di-(4-chlorophenyl)ethane), p,p'-DDD (1,1-dichloro-2,2-di-(4-chlorophenyl)ethane), p,p'-DDE (1,1-dichloro-2,2-di-(4-chlorophenyl)ethylene), alpha-HCH (alpha-hexachlorocyclohexane), beta-HCH (beta-hexachlorocyclohexane), gamma-HCH (lindane) (gamma-hexachlorocyclohexane) and HCB (hexachlorobenzene) were measured in cow's milk and butter, in pig's fat and fat tissue, and in hen's fat tissue. Samples of foodstuffs (10 from each) were collected during 1985/6 in a continental town of Croatia, Yugoslavia either at the market or from individual farmers. p,p'-DDE was present in all the 50 samples (conc. range 1.6—330 $\mu\text{g}/\text{kg}$ fat). The frequencies of gamma-HCH, HCB, alpha-HCH and p,p'-DDT were between 92 and 52% (conc. range 1.0—200 $\mu\text{g}/\text{kg}$ fat). p,p'-DDD was present only in five samples and beta-HCH only in six (conc. range 2.2—130 $\mu\text{g}/\text{kg}$ fat). The highest median concentrations of HCB, gamma-HCH and alpha-HCH (63, 38 and 25 $\mu\text{g}/\text{kg}$ fat resp.) were found in cow's milk and of p,p'-DDE (64 $\mu\text{g}/\text{kg}$ fat) in hen's fat tissue.

Organochlorine compounds are known contaminants of the environment and they are taken up by humans and animals alike. Owing to their lipophilic properties organochlorine pesticides are mostly stored in fat tissues. The aim of this work was to determine levels of the DDT-complex, HCH-group and of HCB in foodstuffs which are representative of the diet in the continental parts of Croatia. For the general population food is the main pathway of intake of organochlorine pesticides, and we have chosen to measure the pesticides and some of their metabolites in food specimens from the cow, pig and poultry. The restricted use of organochlorine pesticides in developed countries has brought about a gradual decrease of residues in food products. We have therefore undertaken this survey in order to compare the present residue levels in our food with the data published in previous years for Yugoslavia and with the levels reported for other countries.

EXPERIMENTAL.

Food samples (number of samples in brackets) were: cow's milk (10) and butter (10), pig's fat (10) and fat tissue (10), and hen's fat tissue (10). Pig's fat was obtained by melting fat tissue. Samples were collected during the years 1985 and 1986 in a continental town of Croatia, Yugoslavia, either at the market (21 samples) or from individual farmers (29 samples).

Extraction of compounds

Milk samples (2.5 g) were extracted with chloroform / methanol using a procedure for human milk (1).

Butter (1.0 g) and pig's fat (1.0 g) were dissolved in hexane (10 ml). Solutions were washed with conc. H_2SO_4 (18 ml), the mixture was carefully shaken (1.0 min) and centrifuged. The hexane extract was evaporated just to dryness (N_2 stream; water bath $< 40^\circ C$) and the residues were dissolved in 1.0 ml hexane before gas chromatographic analysis.

Fat tissue (1.0 g) was mixed with anh. Na_2SO_4 (2 g) in a mortar with a pestel. The mixture was extracted with hexane (30 ml) and filtered through filter paper into a preweighed tube. The mortar, pestel and filter paper were washed with $10 + 2$ ml hexane and the hexane extract was added to the tube. The hexane was evaporated just to dryness as described above, and the fat content was weighed. The fat was dissolved in hexane (5 ml) and transferred into 10 ml centrifuge tubes. The hexane solution was washed with 3×4 ml H_2SO_4 and after centrifugation the H_2SO_4 layer was discarded. The hexane was evaporated as described above and the residues were dissolved in 1.0 ml hexane before gas chromatographic analysis.

The procedures for butter, fat and fat tissue were based on those described in the literature (2). All reagents were from Merck (pro analysi or pesticide grade).

Gas chromatographic analysis

Instruments: Varian 1400 with a 3HSc electron-capture detector and Pye Unicam 204 with a ^{63}Ni electron-capture detector.

Columns: 1.3% SF-96 + 5.3 QF-1 on Supelcoport 100/120 mesh (1.8 m length) and 5% OV-101 on Chromosorb W DMCS /AW 80/100 mesh (2.0 m length). Column diameter: 6.349 mm. Carrier gas: N_2 (flow rate 30-40 ml/min).

All samples were chromatographed on both columns. Only the peaks observed on both columns were evaluated. The compounds were identified by their retention times as compared to known standards. Quantitation was done by comparing the peak heights in the sample with those in known standard mixtures.

Recoveries were determined for concentration levels similar to those found in the samples. For some compounds and some foodstuffs low recoveries were obtained which might be due to low concentration levels in the samples. However, the recoveries were reproducible. The recoveries obtained are given in Table 1 and all results were corrected accordingly.

Table 1.

Recoveries of compounds. The number of determinations is given in brackets.

| Compound | Recoveries (%) | | | | |
|-----------|----------------|---------|-------------|-----------|-------------------|
| | Cow: Butter | Milk | Pig: Fat | Fat tiss. | Hen: Fat tiss. |
| p,p'-DDT | 92 (2) | 103 (4) | 84 (3) | 94 (3) | 51 (2) |
| p,p'-DDD | 82 (4) | 89 (4) | 59 (3) | 67 (3) | 55 (3) |
| p,p'-DDE | 76 (3) | 80 (4) | 67 (3) | 69 (3) | 28 (3) |
| alpha-HCH | 65 (3) | 38 (4) | 49 (3) | 32 (2) | 36 (3) |
| beta-HCH | 76 (3) | 44 (3) | 51 (2) | 49 (2) | 30 (3) |
| gamma-HCH | 65 (4) | 48 (4) | 56 (2) | 24 (3) | 36 (3) |
| HCB | 67 (1) | 44 (3) | 21 (2) | 26 (2) | 28 (3) |

RESULTS AND DISCUSSION

The organochlorine compounds: p,p'-DDT, p,p'-DDD, p,p'-DDE, alpha-HCH, beta-HCH, gamma-HCH and HCB were measured in foodstuffs of animal origin (50 samples in all). Their concentrations were expressed in micrograms per kilogram fat. The concentrations of the compounds were not normally distributed and therefore median concentrations and concentration ranges are given in Table 2.

p,p'-DDE was found in all the 50 samples. The next highest frequencies were for gamma-HCH, HCB, alpha-HCH and p,p'-DDT.

Beta-HCH was found only in six samples and p,p'-DDD in five. The concentration ranges of the compounds were broad. The median concentrations of all compounds but p,p'-DDE were highest in cow's milk. On the average butter contained lower residue concentrations than milk. The highest median concentration of p,p'-DDE was found in hen's fat tissue. There was no evident difference in concentrations between the samples coming from the market and those collected from individual farmers. The concentrations of all compounds were below the limits recommended for foodstuffs by FAO and WHO (3).

The concentrations of the DDT-complex and the HCH-group have been measured in food products in Yugoslavia over the past 20 years (4, 5). Data for HCB seem to be available only for chicken meat and cow's milk (6, 7) and are about the same as those reported here. Comparing the data published for the DDT-complex and the values presented here the distribution of the compounds, particularly of p,p'-DDE, seems to be as wide as it was in the past. The incidence of p,p'-DDE was up to 80% in the year 1971 (8) and up to 90% in the year 1974 (9) depending on the type of foodstuff. Unlike the DDT complex the HCH group seems to show a slight increase in frequency. *Spirić and Višacki* (4) found a 22% incidence of the HCH compounds in the year 1976, but a 78%

Table 2.

Concentrations of compounds determined in 10 samples of each foodstuff. ND stands for values below the determination limits*. The number of positive samples is given in brackets; concentration ranges apply to positive samples

| Compound | Cow: Butter | Milk | Fat | Pig: Fat | Fat tiss. | Hen: Fat tiss. |
|--|----------------|---------|----------|-------------|-----------|-------------------|
| Median concentrations ($\mu\text{g}/\text{kg}$ fat) | | | | | | |
| p,p'-DDT | ND (4) | ND (0) | 10 (7) | 6.3 (6) | 14 (9) | |
| p,p'-DDD | ND (3) | ND (1) | ND (1) | ND (0) | ND (0) | |
| p,p'-DDE | 17 (10) | 43 (10) | 8.5 (10) | 8.1 (10) | 64 (10) | |
| alpha-HCH | 9.9 (10) | 25 (9) | 1.9 (7) | 0.9 (5) | 2.3 (7) | |
| beta-HCH | ND (0) | ND (1) | ND (0) | ND (2) | ND (3) | |
| gamma-HCH | 6.3 (10) | 38 (10) | 3.0 (7) | 12 (10) | 2.5 (9) | |
| HCB | 7.2 (7) | 63 (10) | 17 (10) | 8,7 (9) | 5.4 (8) | |
| Concentration ranges ($\mu\text{g}/\text{kg}$ fat) | | | | | | |
| p,p'-DDT | 2.9- 8.6 | — | 4.0-17 | 6.0-16 | 4.5-66 | |
| p,p'-DDD | 2.0-12 | 24 | 3.4 | — | — | |
| p,p'-DDE | 3.8-38 | 21-140 | 4.0-19 | 1.6-22 | 16 -330 | |
| alpha-HCH | 4.0-15 | 21-140 | 1.0- 9.2 | 1.7-26 | 1.7- 4.7 | |
| beta-HCH | — | 132 | — | 2.2- 3.9 | 3.1-15 | |
| gamma-HCH | 2.2-18 | 31-89 | 1.3-46 | 6.3-19 | 2.2- 9.4 | |
| HCB | 3.0-33 | 42-200 | 8.6-45 | 5.0-12 | 3.3-18 | |

* Determination limits ($\mu\text{g}/\text{kg}$ fat basis):
 0.5 for HCB, alpha-HCH and gamma-HCH
 1.0 for beta-HCH and p,p'-DDE
 2.0 for p,p'-DDD and p,p'-DDT

incidence in the year 1983. The published concentration ranges for all compounds however are close to the values reported in this paper (5, 8, 9, 10).

From our results it appears that at present the levels of organochlorine pesticides in Yugoslavia are similar to those found in developed countries in Europe and outside it (11, 12, 13, 14).

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Sažetak

OSTACI ORGANOKLORIRANIH PESTICIDA U NEKIM UZORCIMA HRANE ZIVOTINJSKOG PORIJEKLA

Organoklorirani spojevi p,p'-DDT /1,1,1-triklor-2,2-di-(4-klorfenil)etan/, pp',-DDD /1,1-diklor-2,2-di-(4-klorfenil)etan/, p,p'-DDE /1,1-diklor-2,2-di-(4-klorfenil)etilen/, alfa-HCH (alfa-heksaklorcikloheksan), beta-HCH (beta-heksaklorcikloheksan), gama-HCH (lindan) (gama-heksaklorcikloheksan) i HCB (heksaklorbenzen) određivani su u uzorcima kravljeg mlijeka i maslaca, svinjske masti i masnog tkiva te kokošjeg masnog tkiva. Uzorci hrane (10 od svake vrste) sakupljeni su tokom 1985/86. godine u kontinentalnom gradu Hrvatske bilo u trgovini ili od individualnih proizvođača. p,p'-DDE je nađen u svih 50 uzoraka (raspon koncentracija 1,6-330 µg/kg masti). Učestalosti gama-HCH, HCB, alfa-HCH i p,p'-DDT bile su između 92 i 52 % (raspon koncentracija 1,0-200 µg/kg masti). p,p'-DDD je nađen u samo pet uzoraka, dok je beta-HCH nađen u samo šest uzoraka (raspon koncentracija 2,2-130 µg/kg masti). Najviši medijani koncentracija za HCB, gama-HCH i alfa-HCH (63, 38 odnosno 25 µg/kg masti) nađeni su u kravljem mlijeku, a za p,p'-DDE (64 µg/kg masti) u kokošjem masnom tkivu.

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