ANALYTICAL DETERMINATION OF ORGANOCHLORINE
PESTICIDES IN SOILS AND FOODS

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Abstract

Owing to the widespread use of organochlorine pesticides in the past, significant amounts of them still occur in nature, mainly in the form of by-products, which, however, often present a greater risk than the primary product used. Danger comes mainly from their ability to accumulate along the food chain and because they can enter the human body in different ways – through inhalation, penetration through the skin, through ingestion, etc.

The current study on the content of this group of pesticides and their metabolites in soil and meat samples is an up-to-date development to analyse the potential hazards for environment and people. Eight soil samples and three types of meat foods were tested for the content of organochlorides and their metabolites. The results presented show that although these substances were found in some of the samples, their concentrations were below the regulatory permissible values.

Key words: organochlorine pesticides (OCPs), dichlorodiphenyl-trichloroethane (DDT), soil, meat foods, chromatography

Introduction. Some of the organic pesticides are halogenated aliphatic and aromatic hydrocarbons, part of the group of synthetic organic insecticides [1,2],

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which group includes dichlorodiphenyl-trichloroethane (DDT). The effect of pesticides in soils is expressed in violation of soil properties and suppression of soil microflora, which in turn affects soil fertility [2,3]. According to SOARES et al. [4], once adsorbed onto the soil particles, these compounds may be degraded or may be slowly released into the atmosphere, subterranean aquatic systems and living organisms. The lipophilic nature, hydrophobicity and low chemical and biological degradation rates of the organochlorine pesticides can lead to their accumulation and concentration in biological tissues [1,5,6]. The widespread use of pesticides in the past has led to contamination of the water, soil and air and their accumulation in products (e.g. fruits, vegetables, meat and fat), and along the food chains (Fig. 1). Pesticides may also be transported by rain and wind from the points of application to adjacent areas where their presence may be undesirable or harmful [7].

The pathways for the entry of pesticides into the human body are through the skin, lungs and through the digestive system, when consuming products containing pesticides. Pesticides are mainly accumulated in liver, kidneys and adipose tissues. Low concentrations are also found in the stomach wall, lungs, testes, brain and myocardium. In addition, exposure of the human body to OCPs can lead to

Fig. 1. Accumulation of pesticides in food chains

carcinogenicity and mutagenicity, disorders of the endocrine system, reproductive problems, etc. [5,8,9].

The monitoring of persistent organic pollutants (POPs) and in particular of pesticides distributed in the environment and contained in consumed foods, is an integral part of the European Union's policy of controlling the harmful effects on species and protecting human health. In this context, European Directive 2009/128/EC was issued with the aim of strengthening controls on the use of pesticides in the community [10]. Despite the ban on DDT, its residues and intermediates (metabolites) are still found in soils. Due to its persistence in the environment, its residues are also established in livestock feed and hence in food and products made from them [11].

The aim of current work is to present the results of the analysis of the content of organochlorine compounds in eight soil samples and three types of meat foods used for human consumption.

Materials and methods. Sampling and sample preparation. The soil samples were taken in 2019. The sampling points are located in areas overlapping in full or particularly with the sampling points at the soils monitoring system of the Republic of Bulgaria: sample 1 – fallow land in Pernik district; sample 2 – field in Kyustendil district; sample 3 – vineyard in Plovdiv district; sample 4 – field in Pleven district; sample 5 – meadow in Sofia province district; sample 6 – fallow land in Sofia district; sample 7 – vegetable garden in Burgas district; and sample 8 – field in Silistra district. They are taken according to standard BNS ISO 10381 [12]. A sampling depth for the non-arable soils is 0–10 cm and for the arable – 0–40 cm. The pre-treatment has been carried out in accordance with the requirements of BNS ISO 11464:2012 [13].

Procedure for treatment of the soil samples: 10 g soil (± 0.0001 g) were weighed on an analytical balance, placed in Teflon vessels and 30 cm³ mixture of hexane and acetone and 50 μ L standard containing 10 μ g/mL DDT, were added. The samples were microwave-degraded for 15 min at 200 °C, then cooled, filtered and 2 cm³ toluene were added. Then they were concentrated by vacuum evaporation and purified through chromatographic columns containing wadding, silica gel and sodium sulfate.

The meat samples were also taken in 2019. Different number of samples were taken from different types of meat from different market places: number of chicken samples – 14; number of pork samples – 18; and number of lamb samples – 9. The chicken meat samples were taken in accordance with the Bulgarian normative acts and standards [14,15]. For the other samples adapted similar sampling techniques were used.

Procedure for treatment of the meat samples: 50 g (± 0.0001 g) minced meat were weighed on an analytical balance, placed in a glass beaker, anhydrous sodium sulphate was added to these and the samples were repeatedly extracted with petroleum ether. The resulting extract was filtered and the solvent was removed

by vacuum evaporation at 40 °C, purified by dilution with hexane in a 1:9 ratio using a glass chromatographic column loaded with florisil, and re-purified from the solvent.

Reagents. To perform the experimental work were used analytical reagents classified as pure for analysis: hexane (Merck); acetone (Merck); toluene for chromatography (Merck); sodium sulphate anhydrate (Merck); petroleum ether (40–60 °C, Sigma-Aldrich); florisil (60–100 mesh, Sigma-Aldrich).

Instrumentation and measurement conditions. The determination of OCPs in the soil samples was carried out in accordance with the requirements of BNS ISO 10382: 2005 [16] by gas chromatographic separation with subsequent identification by means of a mass selective and electron capture detector. Blank samples are developed in parallel with the working samples. The analysis of soil samples were carried out on Thermo Trace DSQ (Thermo Scientific) gas chromatograph triple quadrupole mass spectrometer including Ion Max Housing with GC/MS. A chromatographic column TG-5 sil MS, 30 m × 0.25 mm × 0.25 µm, was used.

The determination of pesticide residues in the meat samples was carried out in accordance with the requirements of BNS EN 1528:2001 (its four parts) [17]. Residues of 4,4-DDE in fat samples extracted from the meat samples tested were determined. Gas chromatograph with electronic detector (GC-ECD) “VARIAN CP 3800” were used for the analysis, and chromatographic column DB-5, 30 m × 0.25 mm × 0.25 µm.

Results and discussion. The results for the content of organochlorine pesticides in the soil samples are presented in Table 1.

The admissible values for the concentration of organochlorine pesticides in the soil samples are regulated in the Bulgarian legislation [18]. In Bulgaria there are three types of admissible concentrations – precautionary concentrations (PC), maximum admissible concentrations (MAC), and interventional concentrations (IC). Their values are described in Table 2.

Absence of γ hexachlorocyclohexane; o,p and p,p DDD; p,p DDT; o,p DDE and hexachlorobenzene was established in the soil samples. The results obtained for the other OCPs available in the soil samples show that the established values are below the value for the maximum admissible concentrations introduced by Regulation 3 [18].

In all analysed soil samples, the presence of at least one of the analysed pesticides was found, which did not allow any of the studied geographical areas to be designated as “clean” of organochlorine pesticides. This, in turn, implies the presence of these pesticides and their metabolites in plant and animal products reared/harvested within the study areas.

The maximum residue levels (MRLs) of pesticides in food or feed, have been established by Regulation (EU) No 396/2005 [19]. In some of the tested working samples of the three different meat types, residues of DDE were found. The

T a b l e 1

Concentration of organochlorine pesticides in the soil samples

Pesticide, mg/kg	Sample No							
	1	2	3	4	5	6	7	8
α HCH	0.0025 ± 0.0008	< DL	< DL	< DL	< DL	< DL	0.00012 ± 0.0005	< DL
β HCH	0.0013 ± 0.0004	< DL	< DL	0.0091 ± 0.0027	0.0086 ± 0.0033	0.0023 ± 0.0007	< DL	0.0018 ± 0.0008
γ HCH	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
o,p DDD	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
p,p DDD	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
o,p DDT	0.0013 ± 0.0005	< DL	0.0044 ± 0.0017	< DL	< DL	< DL	< DL	< DL
p,p DDT	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
o,p DDE	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
p,p DDE	< DL	0.0047 ± 0.0017	< DL	< DL	< DL	0.0032 ± 0.00012	< DL	< DL
HCB	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL

< DL– Concentration below the detection limit

DDD – o,p DDD (2,4-dichlorodiphenyl-2,2-dichloroethane);

p,p DDD (4,4-dichlorodiphenyl-2,2-dichloroethane);

DDT and DDE – o,p DDT (2,4-dichlorodiphenyl-2,2,2-trichloroethane);

p,p DDT (4,4-dichlorodiphenyl-2,2,2-trichloroethane);

o,p DDE (2,4-dichlorodiphenyldichlorethylene);

p,p DDE (4,4-dichlorodiphenyldichlorethylene);

 α , β , γ HCH and HCB (α , β and γ hexachlorocyclohexane; hexachlorobenzene)

T a b l e 2

Admissible concentration values for soils in Bulgaria [18]

Pesticide	PC	MAC	IC
	Total mg/kg dry soil content		
HCB	0.025	0.25	10
α , β , γ HCH	0.001	0.01	2
DDX*	0.3	1.5	4
o,p and p,p DDE	0.1	0.5	—
o,p and p,p DDD	0.1	0.5	—
o,p and p,p DDT	0.1	0.5	—

*DDX represents the sum of concentrations of DDD, DDT and DDE

T a b l e 3

DDE values in the tested meat samples

Type of sample	Sample No	Result, mg/kg
Chicken	0102	0.020 ± 0.004
	0107	0.003 ± 0.001
	0111	0.008 ± 0.001
	0112	0.009 ± 0.002
Pork	0208	0.013 ± 0.002
	0217	0.009 ± 0.001
Lamb	0301	0.043 ± 0.004
	0302	0.052 ± 0.009
	0304	0.118 ± 0.022
	0305	0.053 ± 0.011
	0306	0.189 ± 0.025
	0308	0.056 ± 0.010
	0309	0.167 ± 0.024

values of each were lower than the maximum value (1 mg/kg fat) for the sum of all metabolic residues of DDT in organisms (Table 3).

It is clear from the results presented in Table 3 that despite the ban on the use of DDT, which has been in force for a relatively long time, this pesticide and its metabolites can still be detected in animal samples. Presence of the pesticide was found in 11% of the tested 14 chicken meat samples, in 11% of the 18 pork samples and in 78% of the 9 lamb meat samples tested, albeit within the acceptable limits. The results for the other samples analysed are outside the range of concentrations (from 0.002 to 0.05 mg/kg) for which the method is applicable.

Similar to the data reported in [11], in the present study, the presence of DDT residues was found in the largest number of lamb meat samples (in 7 of 9 samples). This is most likely due to the herbivorous farming conditions that allow for grazing and direct contact with environmental components. Another reason for the accumulation of DDT in their body is the longer sheep breeding period and the significant amounts of fat and muscle mass (e.g. greater than those of chickens) that they accumulate.

In the other two types of samples, residues of DDT were detected in 1/9 of the pork samples tested and in 2/7 of the chicken samples tested. The values found were significantly lower than those in lamb meat (0.04–0.189 mg/kg) – in chicken between 0.003 and 0.02 mg/kg and in pork 0.009 and 0.013 mg/kg. A probable reason for this difference is the way in which these two species are reared, namely under controlled conditions under which they are fed with controlled feed in which the presence of DDT is unacceptable.

Pigs also accumulate large amounts of fat, but they feed only on controlled feeds, and it is therefore assumed that the logically established DDT values in them are lower than those in lambs.

Conclusions. The experimental data obtained show that despite the ban on the use of DDT, which has been in force for a relatively long time, this pesticide and its products can still be detected in soil samples throughout the country as well as in samples of animal origin.

The values for the content of OCPs in all tested soil samples where they are found do not exceed the MAC values prescribed by national legislation.

Residues of DDE were detected in some of the meat samples tested, the values of each of which was lower than the maximum value of 1 mg/kg. When differentiating the obtained data according to the origin of the analysed meat sample, the results indicate lower concentrations of DDE in chicken and pork meat, which are grown under controlled conditions.

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