J. Int. Environmental Application & Science, Vol. 11(4): 401-408 (2016)

Organochlorine Pesticides and PCB in Meat and by-Products from Albanian Markets

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Received September 26, 2016; Accepted December 07, 2016

Abstract: In this study were determine concentrations of organochlorine pesticides and polychlorinated biphenyls in meat and by-products samples from different markets of Tirana, Albania. Meat and by-products with origin from Albanian farms were sampling in March 2016. Organochlorine pesticides, their residues and PCBs are stable compounds, lipophilic, with toxic properties. These pollutants could found in meat and by-products because of feed, bioaccumulation process and meat treatment processes. Ultrasonic bath extraction assisted with n-Hexane/Dichloromethane (3:1) as organic solvent and two clean-up steps have been used for analytical treatment of meat and by-products samples. The quantitative analysis of organochlorine pesticides and PCBs were performed by the gas chromatography method with electron capture detector. Analytic steps were based in EN 1528 Method. The profiles of detected pesticides were: Endosulphanes > HCHs > Aldrines > Mirex > Methoxychlor > DDTs. PCB 28, the volatile PCB marker was the main congener for all analyzed samples. In a large number of samples were identified organic pollutants as a result of their previous uses, degradation processes, atmospheric deposition etc. Their levels do not exceed the rates set for them, but should serve as alert for institutions to conduct ongoing controls in foods because it is directly related to the health of consumers.

***Key words**: Organochlorine pesticides, PCBs, Meat samples, Gas chromatography*

Introduction

Persistent organic pollutants (POPs) such as organochlorine pesticides (OCPs) and polychlorinated biphenyls (PCBs) are chemicals that are used for agricultural and industrial purposes or for the synthesis of other products. Characteristic of these pollutants is their high toxicity. These compounds are found in environments such as soil, water, air, biota so they can be found in food products, etc. It is important their assessment levels especially in food matrices where they can be found as result of their use. The main source of organic pollutant is from anthropogenic, water cycle or other factors such as atmospheric deposition. Food is the main source of human contamination by organic pollutants. They adversely affect human health and the environment across the world. Most of the organic pollutants can affect people and animals even far from the place where they are released because they can be transported by wind and water. POPs stand for a long time in the environment and can accumulate or pass from one species to another through the food chain. Although in many countries these chemicals have been banned, they remain stored. They can persist for many years because of their stability. Persistent organic pollutants resist chemical, biological and photolytic degradation, they degrade very slowly. They are characterized by low solubility in water and high solubility in fats leading their bioaccumulation in fatty tissues. Some of them can evaporate easily so they can move in long distances in the atmosphere before deposition occur. This characteristic has shown the presence of compounds such as PCBs throughout the world, even in regions where they are not used.

Today, unfortunately, facing a problem of the past, because many tones of POP compounds were used in large amounts for different purposes almost in all countries include Albania. Faster development of human society over the past decades has brought positive changes in terms of security, detection and management of these chemical compounds (Ahlborg *et al*., 1992; Papadopulos *et al*., 2000). Chemical pollutants in our food can come from different sources but the obligation of institutions is regular analyzes in food matrices. POP levels must be strictly lower than allowed levels decided from national governments or international norms such as Codex Alimentary Commission. Analyzes of organic pollutants and especially the chlorinated organic one is a legal obligation for all

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food products. Analysis of organochlorine pesticides and PCBs in fatty food matrices realized with the technique of gas chromatography with MS or electron capture detector. GC/ECD, GC/MS or GC/MS/MS can determine the chemical contaminants in food even in very low concentrations in ppm to ppb levels. These techniques are recommended in literature and based in EN 1528 Method. Therefore the relevant chemical pollutants analysis is an essential part of the program of food safety testing to provide customer security and regulation compliance (Di Muco, 1996; Erikson, 2001; ATSDR, 2000).

Several methods have been published for the determination of organochlorine pesticides and PCBs in meat samples. Most analytical methods based on the extraction of the chlorinated pollutants with organic solvent, clean-up with solvent-solvent partition and/or adsorption chromatography, followed by determination of the residues by gas chromatography with electron capture detection or mass spectrometry detection. The majority of methods are based on fat analysis because residues of organochlorine pollutants accumulate in this matrix. In order to remove the fatty interferences, extract must be purified through adsorption chromatography using Florisil, silica gel, alumina, etc.

The aim of this study was to having some preliminary data for concentrations of OCPs and PCBs in meat samples, which fulfils the requirements of European Regulations regarding the official controls of chemical residues in food of animal origin.

Materials and Methods

Sampling of meat and by-products samples

13 meat and by-products samples were collected in different markets of Tirana City, Albania. The sampling was realized in March 2016. The meat and by-products were sampled in random mode from the most known production industries of meat and by-products from Albanian farms. Meat and by-products samples were transported and storeat - 4^{o}C before analysis.

Preparation of meat and by-products samples for pesticide residues and PCB analysis

The extraction method used was based on EN 1258 for determination of organchlorine pesticides in fatty matrices. About 10 g of fresh meat or by-products was put into a flask and about 50 ml of n-hexane/dichloromethane (3/1) was added and the samples were extracted for 60 min in ultrasonic bath. The sample was spiked with recovery standard (PCB 29) before extraction. 40 g of silicagel with 45% acid sulphuric (m/m) were added for lipids hydrolyze. For a second clean-up procedure were used a florisil 5% water column. Extracts were concentrated under a kuderna Danish to approximately 2 ml and analyzed using gas chromatography-ECD (Bernard et al, 1999; Focant et al, 2004; Nuro et al 2007; Lazaro et al 1995). The following organchlorine pesticides: hexachlorocyclohexane (HCH) isomers, dieldrin, endrin, heptachlors, endosulphanes, methoxychlor, mirex and the DDT-related chemicals (*o,p*-DDE, *p,p*-DDE, *p,p*-DDD, *p,p*-DDT) were detected. PCB markers were studied simultaneous with above pesticides in meat and by-products samples.

Apparatus and chromatography

Gas chromatographic analyses were performed with a Varian 450 GC equipped with a ^{63}Ni electron-capture detector and PTV injector. The column used was Rtx-5[low/mid polarity, 5% (phenyl methyl siloxane)] (30 m x 33 mm I.D., x 25mm film). The split/splitless injector and detector temperatures were set at 280^{0}C and 300^{o}C, respectively. Carrier gas was He at 1 ml/min and make-up gas were nitrogen at 24ml/min. The initial oven temperature was kept at 60^{o}C for 4 min, which was increased, to 200^{o}C at 20^{o}C/min, held for 7 min, and then increased to 280^{o}C at 4^{o}C/min for 20 min. The temperature was finally increased to 300^{0o}C, at 10^{0}C/min, held for 7 min. Injection volume was 2 µl, when splitless injections were made. Pesticide quantification was performed by internal standard method (Santillo, 2004).

Results and Discussion

Levels of organochlorine pesticides, their metabolites and polychlorinated biphenyls were analyzed in meat and by-products samples from markets of Tirana, Albania. Meat and by-products samples were sampled randomly in March 2016. EN 1528/1/2/3/4 protocols were used for isolation and quantification of chlorinated compounds in meat samples with origin from Albanian farms. Organchlorine pollutants were detected using capillary gas chromatography with ECD technique.

Total of organochlorine pesticides in meat and by-products samples were shown in Figure 1. The totals were between 4.03 – 25.6 µµg/kg. The average of pesticides in meat samples were 13.5 µg/kg. The minimum was for pork samples while the maximum in chicken sausages, chicken meat and chicken fillet. This fact could be because of the main feeds for chicken farm are maize and other grains. This could affect directly in found levels. Cow, pigs and sheep's feed mostly in natural habitats. Figure 2 presented the distribution of organochlorine pesticides in samples of meat and its by-products. It was noticed the same distribution of analyzed pesticides because the origin of the samples is from our country. Figure 3 present profile organochlorine pesticides in samples of meat and by-products. Heptachlor > Aldines > HCHs > DDTs was their profiles on studied samples of meat from Albanian origin. The highest levels of HCHs, cyclopentadiene pesticides and DDTs were found in chicken, chicken thigh and chicken fillets samples. This is generally associated with the use of grain as feed for chickens. The profiles of HCHs in analyzed samples were: a-HCH> b-HCH> d-HCH> Lindane. Obviously, their presence is a result of previous uses of lindane for agricultural purposes. Profile of chlorinated cyclopentadiene pesticides in meat and by-products samples were: Heptachlors > Endosulphanes > Aldrines > Chlordanes. It was noticed that the main contribution were by their metabolites because of their previous uses. Profiles of DDTs were: DDD > DDE > DDT. DDTs presence is a result of previous uses of DDT for agricultural purposes. Mirex, HCB and Dicofol were found only for 26% of samples. The main levels for these pesticides were also in chicken meat. For all meat and by-product samples found levels were lower than allowed values for individual or total organochlorine pesticides in meat and by-product samples (Codex Alimentarius, 2010).

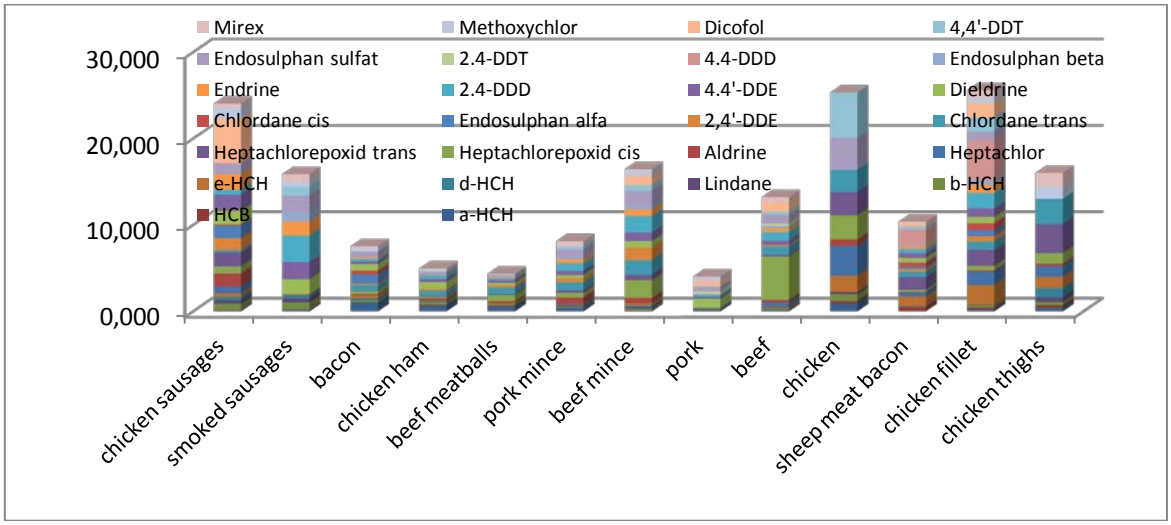

Figure1. Total of organochlorine pesticides(µg/L) in meat and by-products samples

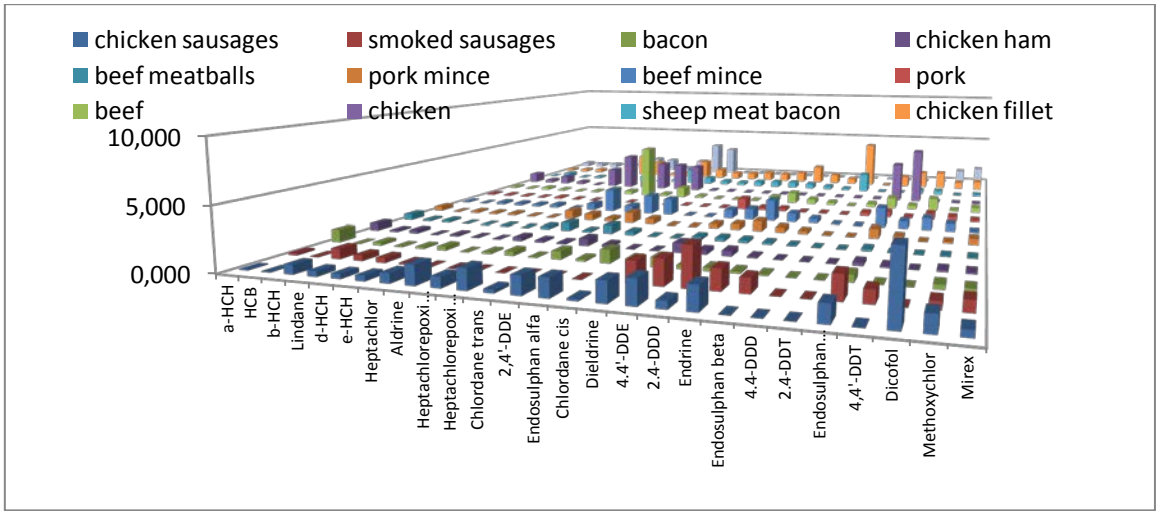

Figure 2. Distribution of organochlorine pesticides (µg/L) in meat and by-products samples

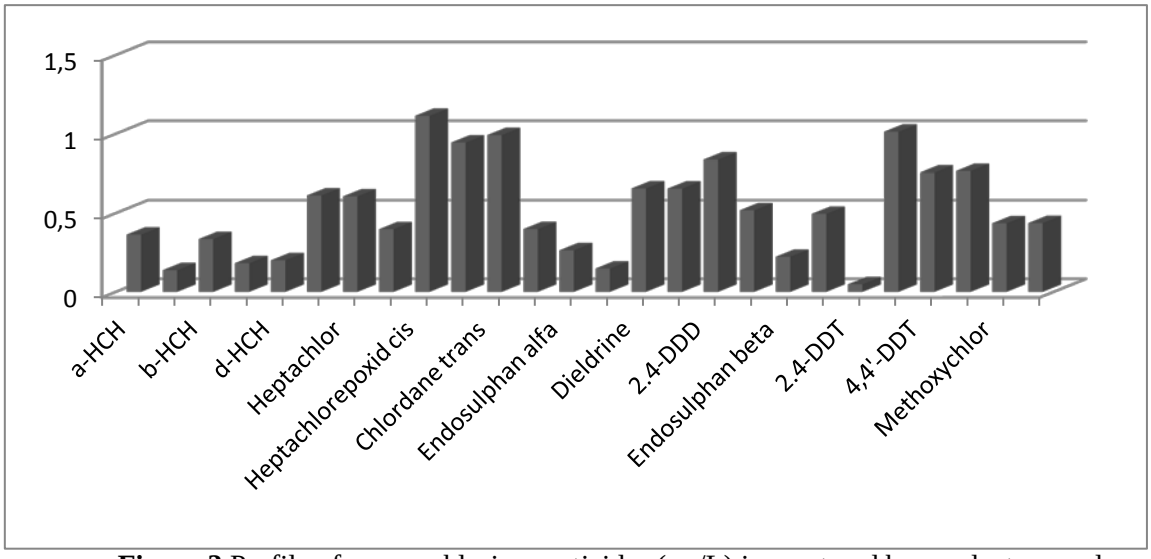

Figure 3.Profile of organochlorine pesticides (µg/L) in meat and by-products samples

Statistical processing of the data for the organochlorine pesticides concentrations in samples of meat and by-products were done using Cluster Analysis. Figure 4 shown dendrogram of organochlorine pesticides in samples of meat and its derivatives. There are several main groups who combine all three with a 90% similarity level. Lindane and its isomers have the highest similarity, then come Endosulphanes and DDTs. Similarities clusters formed by the level of organochlorine pesticides for all samples of meat and its derivatives obtained in the analysis. Figure 5 showed Dendrogram of meat and its by-products samples against concentrations of organochlorine pesticides found for each samples type. The main group with the highest similarity consists for by-product samples with level of similarity around 85%. This is associated with the same origin of these samples.

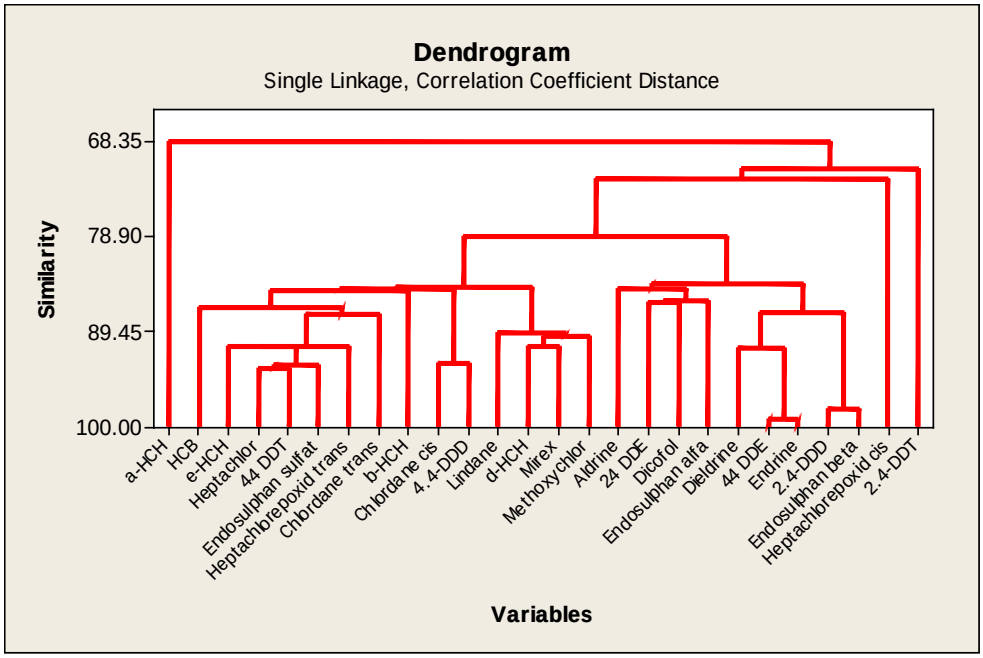

Figure 4. Dendrogram of organochlorine pesticides(µg/L) in meat and by-products samples

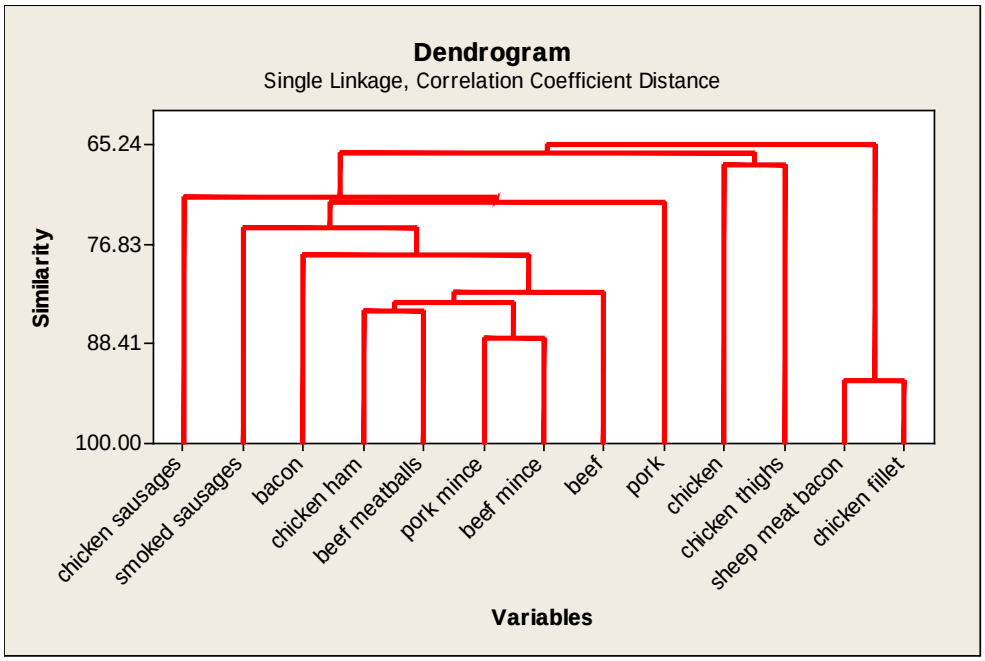

Figure 5. Dendrogram of organochlorine pesticides(µg/L) in meat and by-products samples

Figure 6 presented data for PCBs in meat and by-products samples. The minimum levels were in pork, cow meat and their by-products samples with 0.99 µg/kg while the maximum was 21.88 ug/kg in chicken samples. This fact could be because the nature of feeding these animals. Figure 7 shows the distribution of PCBs in samples of meat and its by-products. It were noted the presence of PCBs in some samples related to habitat and feed used for respective samples. Figure 8 provides profile for PCBs in analyzed samples. PCBs profiles were: PCB153 > PCB 52 > PCB 138 > PCB 180. PCB 153 and PCB 138 are justified by their higher solubility in fats. PCB 52 is a representative of the PCB volatile, which means that and these congeners have a significant impact on the found levels and their distribution in meat samples. The levels found were lower than their allowed value for total PCBs in meat samples (Codex Alimentarius, 2010).

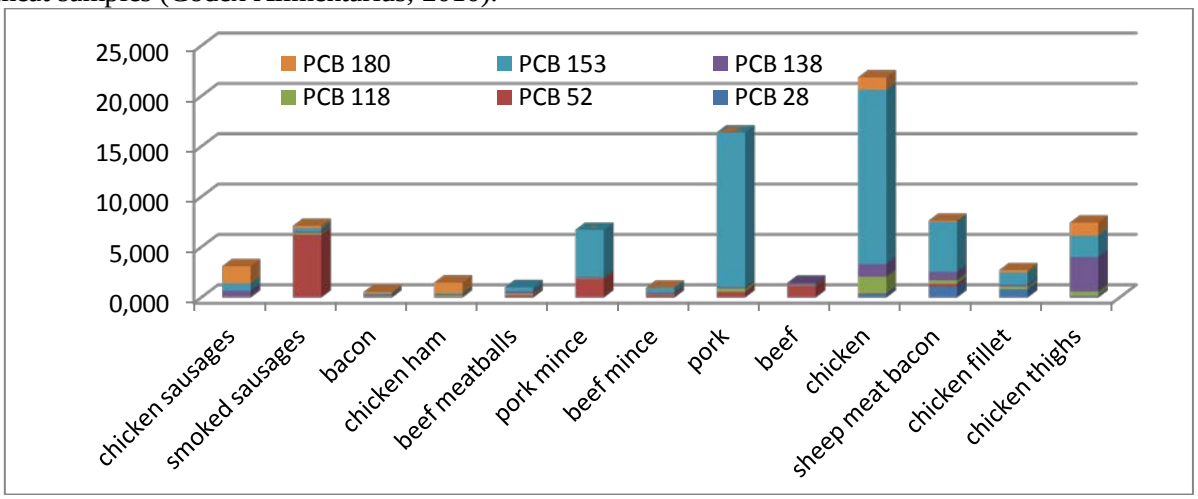

Figure 6. Total of PCB markers (µg/L) in meat and by-products samples

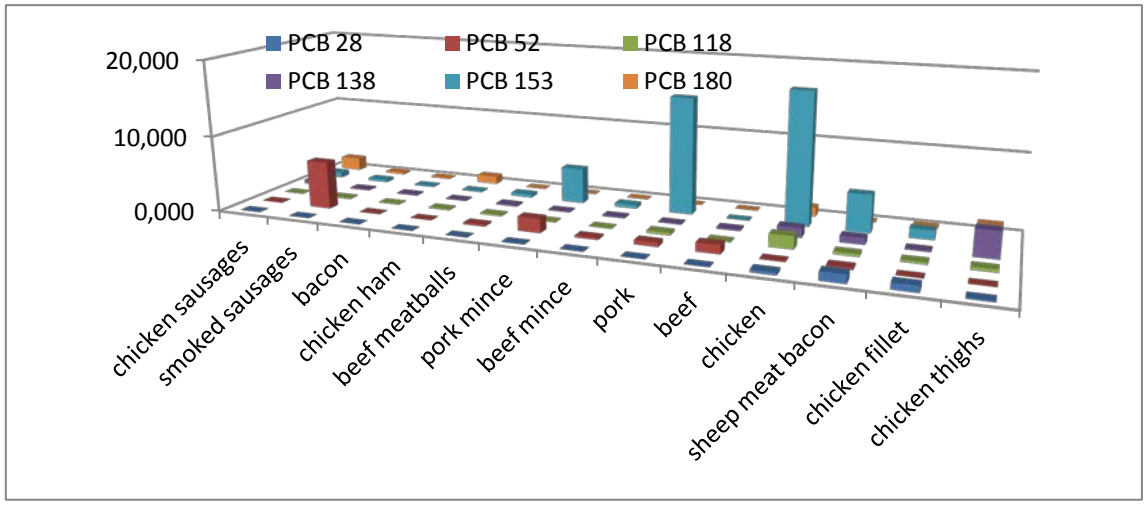

Figure 7. Distribution of PCB markers (µg/L) in meat and by-products samples

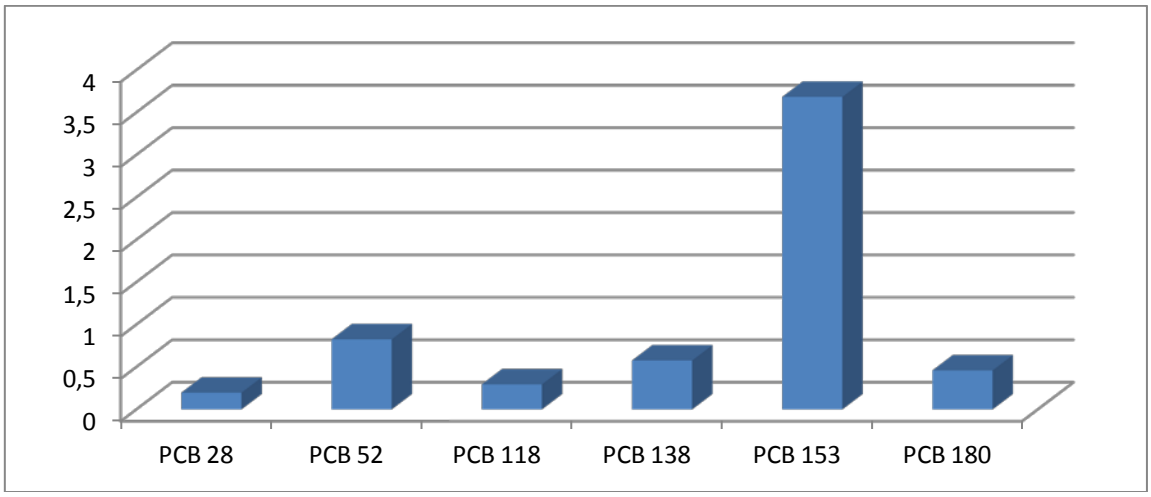

Figure 8. Profile of PCB markers (µg/L) in meat and by-products samples, January 2015

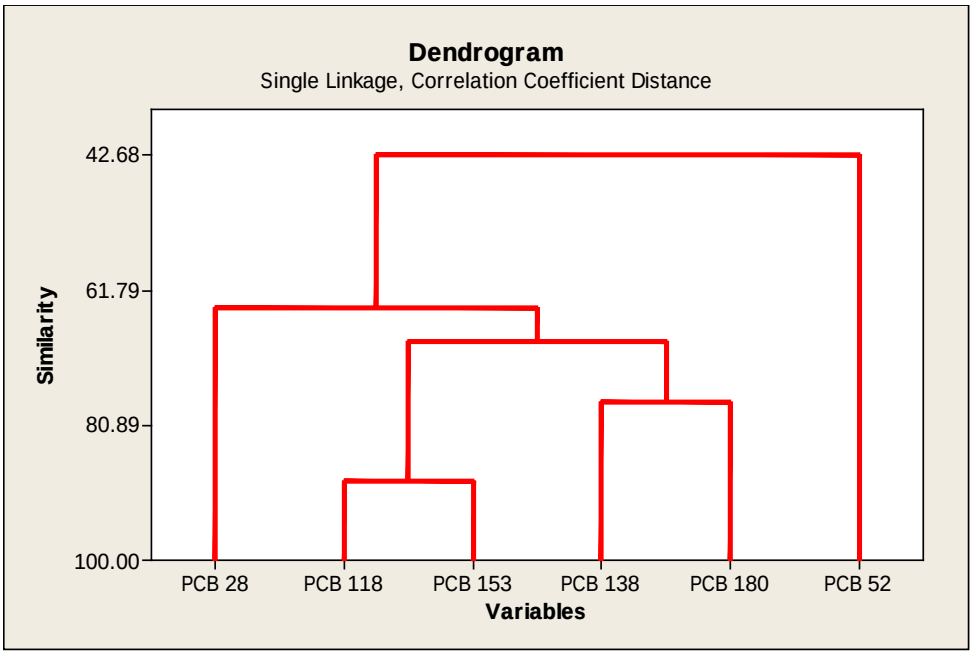

Figure 9. Dendrogram of PCBs (µg/L) in meat and by-products samples

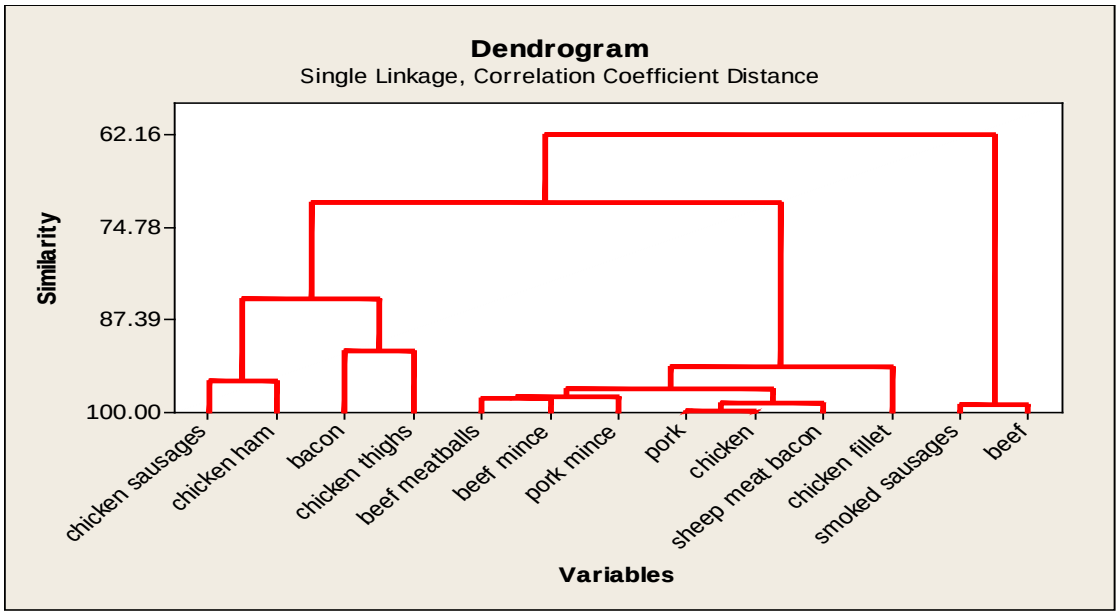

Figure 10. Dendrogram of PCBs (µg/L) in meat and by-products samples

For details explanation of PCBs in samples of meat and its derivatives were performed statistical processing using Cluster Analysis. Figure 9 shows dendrogram of individual PCBs in meat and by-products samples. The two main groups were PCB 118 with PCB 153 (87% similarity) and PCB 138 with PCB 180 (77% similarity), PCB 153 and PCB 138 make up the leading group with 70% similarity between them followed by PCB 28 and PCB 209 to the level of similarity greater than 57%. The clusters formed by PCBs were because of their origin in meat samples. The atmospheric deposition and their solubility in fats are the main factors. Figure 10 shows the Dendrogram for the meat and by-products samples against to the PCB concentration found for each of them. There is a core group with very high similarity consisting of samples of meat and its by-products. Their similarities were from 99 to 93%. The chicken and their by-product samples had lower similarity, around 85%. This could be because of the different parts of analyzed tissues.

Conclusions

Levels of organochlorine pesticides, their metabolites and polychlorinated biphenyls were analyzed in meat and by-products samples with origin from the main farms of Albania in March 2016. EN 1528/1/2/3/4 protocols were used for determination of chlorinated compounds in meet samples because they classified as fatty samples. Almost in all analyzed meat and by-products samples were found organochlorine pollutants concentrations. The average of pesticides in meat samples were 13.5 µg/kg while the average of PCBs were 6.0 µg/kg. The minimum was for pork samples while the maximum in chicken sausages, chicken meat and chicken fillet. This fact could be because of the main feeds for chicken farm are maize and other grains. This could affect directly in found levels. Cow, pigs and sheep's feed mostly in natural habitats. It was noticed the same distribution of analyzed pesticides because the origin of the samples is from our country. Heptachlor > Aldines > HCHs > DDTs was their profiles on studied samples of meat from Albanian origin. The highest levels of organochlorine pesticides were found in chicken, chicken thigh and chicken fillets samples. This is generally associated with the use of grain as feed for chickens. Diversification of samples and organic pollutants that were analyzed in this study make it a valuable guide for analytical laboratories performed these tests. For all meat and by-product samples found levels were lower than allowed values for individual or total organochlorine pesticides or PCBs in meat and by-product samples (Codex Alimentarius, 2010).

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