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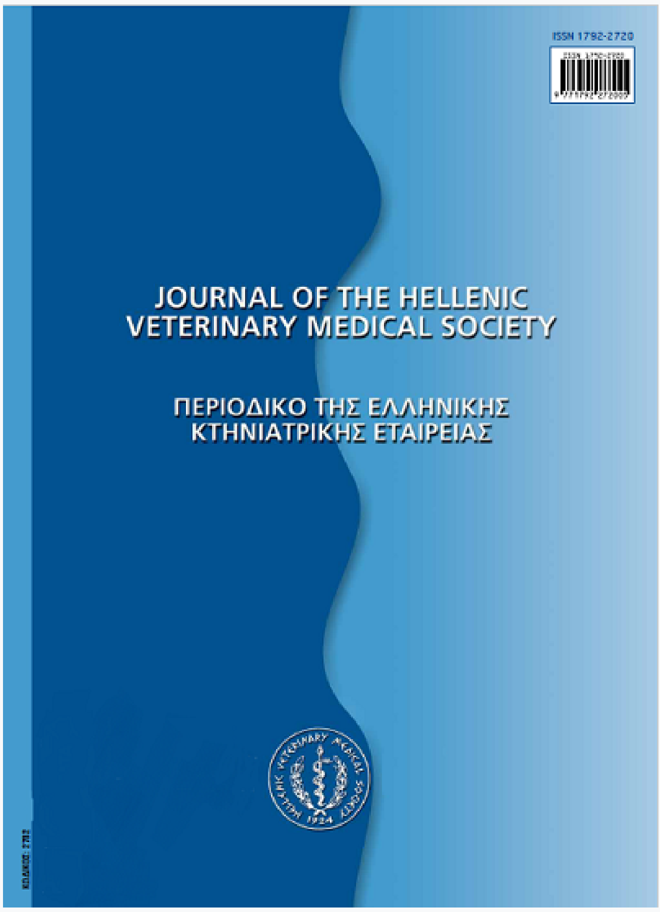

Monitoring of dioxins and related toxic compounds in food of animal origin in Greece

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Review article
Ανασκόπηση

Monitoring of dioxins and related toxic compounds in food of animal origin in Greece

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Έλεγχος διοξινών και παρόμοιων τοξικών ρύπων σε τρόφιμα ζωϊκής προέλευσης στην Ελλάδα

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ABSTRACT. Polychlorinated dibenzo-p-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs) and "dioxin-like" polychlorinated biphenyls (PCBs) are environmental pollutants belonging to the persistent organic pollutants (POPs). PCDDs and PCDFs have never been produced intentionally, but they are formed by incomplete combustion of organic material in the presence of chlorine, during industrial processes or intentional and unintentional burning of solid waste. PCBs have been produced industrially as mixtures and have been used in various applications, though their production has been banned since the 1970's. Their toxicity is mediated through the interaction with the aryl hydrocarbon (AH) receptor, causing severe health effects to humans after chronic exposure, which include developmental and immunological effects, carcinogenicity and chloracne. More than 90% of the daily human intake is due to food ingestion and especially, to the consumption of fat-containing animal food products, due to the lipophilic nature of these compounds. In order to control food contamination by these compounds, the European Union has set maximum limits for PCDD/Fs and PCBs in food products (EU Regulation 1881/2006/EC). The Mass Spectrometry and Dioxin Analysis Laboratory of "NCSR" Demokritos has been operating since 2002 and it performs analysis of PCDD/Fs and PCBs in food, feed, biological and environmental samples, applying international protocols of analysis (EPA 1613, EPA 1668), based on appropriate sample clean-up and determination by high resolution mass spectrometry (HRMS), using the isotope dilution method, as specified by the EU Regulation 1883/2006/EC. The Laboratory has been accredited according to ISO/IEC 17025 by ESYD and it is the National Reference Laboratory of the European Commission for Greece and Cyprus. Here we present here a comprehensive report of the activities of the Mass Spectrometry and Dioxin Analysis Laboratory during its operation, concerning the monitoring of dioxins and similar toxic compounds in food of animal origin in Greece. Through collaboration with the official bureaus of food, environment and health control organizations of Greece, the Laboratory has conducted analysis in a large number of food and animal feed products sampled randomly in the Greek market. The results are well below the EU maximum levels and, in general, lower than those monitored in other European and Mediterranean countries. This has been attributed to the fact that Greece is not highly industrialized and has never produced PCBs and does not perform systematic waste incineration. On the other hand, samples have been collected and analysed from areas suspected for dioxin contamination, such as areas affected by accidental

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fires, landfills or exposed to industrial pollution. In some of these cases, unusually high levels of dioxin contamination in food samples of animal origin have been observed, in some cases above EU limits, and appropriate measures have been taken by Greek authorities to prevent their consumption and to restore normal dioxin levels.

Keywords: dioxins, dioxin-like PCBs, Greek food, mass spectrometry

ΠΕΡΙΛΗΨΗ. Οι πολυχλωριωμένες διβενζο-παρα-διοξίνες (PCDDs) τα πολυχλωριωμένα διβενζοφουράνια (PDCFs) και τα «παρόμοια με διοξίνες» πολυχλωριωμένα διφαινύλια (PCBs) ανήκουν στους ανθεκτικούς οργανικούς ρύπους. Τα PCDDs και PCDFs δεν έχουν ποτέ παραχθεί σκοπίμως, αλλά παράγονται κατά την ατελή καύση οργανικών υλικών παρουσία χλωρίου. Τα PCBs, αντίθετα, έχουν βρει διάφορες βιομηχανικές εφαρμογές μέχρις ότου απαγορεύτηκε η παραγωγή τους τη δεκαετία του '70. Οι ενώσεις αυτές αλληλεπιδρούν με τον υποδοχέα αρυλο-υδρογονανθράκων (AH) προκαλώντας σοβαρή τοξική δράση στον ανθρώπινο οργανισμό μετά από χρόνια, που περιλαμβάνει επίδραση στην ανάπτυξη και το ανοσοποιητικό σύστημα, καρκινογένεση και χλωρακμή. Πάνω από το 90% της ημερήσιας ανθρώπινης πρόσληψης οφείλεται στη διατροφή και, μάλιστα, λόγω της λιποφιλικότητας των ενώσεων αυτών, οι τροφές που είναι πιο επιβαρυμένες στις ενώσεις αυτές είναι τα προϊόντα που περιέχουν λίπος, κυρίως τα λιπαρά τρόφιμα ζωϊκής προέλευσης (γαλακτοκομικά προϊόντα, κρέας, αβγά) και τα λιπαρά ψάρια. Η Ευρωπαϊκή Ένωση (ΕΕ) έχει θεσπίσει μέγιστα επιτρεπτά όρια για τα PCDD/Fs και τα PCBs στα τρόφιμα, με σκοπό τον έλεγχο την επιβάρυνσής τους. Το Εργαστήριο Φασματομετρίας Μάζας κι Ανάλυσης Διοξινών «ΕΦΑΜΑΔ» του ΕΚΕΚΕ «Δημόκριτος» λειτουργεί από το 2002, διεξάγοντας μεταξύ άλλων προσδιορισμό διοξινών σε τρόφιμα, ζωοτροφές, βιολογικά και περιβαλλοντικά δείγματα εφαρμόζοντας διεθνή πρωτοκόλλα (EPA 1613, EPA 1668), με κατάλληλη επεξεργασία του δείγματος και μέτρηση σε φασματογράφο μάζας υψηλής διακριτικής ικανότητας, όπως ορίζονται στον κανονισμό 1883/2006/ΕΚ. Το Εργαστήριο είναι διαπιστευμένο κατά ISO/IEC 17025 από το ΕΣΥΔ (Αρ. Πιστ. 321) και είναι το Εθνικό Εργαστήριο Αναφοράς για την Ελλάδα και την Κύπρο. Στην παρούσα ανασκόπηση παρουσιάζονται συνολικά οι δραστηριότητες του ΕΦΑΜΑΔ από την αρχή της λειτουργίας του, όσον αφορά στην παρακολούθηση των επιπέδων διοξινών και παρόμοιων με τις διοξίνες ενώσεων σε τρόφιμα ζωικής προέλευσης από την ελληνική αγορά. Το Εργαστήριο ανταποκρίνεται στις απαιτήσεις των κρατικών φορέων ελέγχου τροφίμων σε τακτική βάση, μέσω της ανάλυσης μεγάλου αριθμού δειγμάτων τροφίμων που λαμβάνονται με τυχαία δειγματοληψία, από τρόφιμα που διακινούνται και καταναλώνονται στην Ελλάδα. Τα αποτελέσματα των αναλύσεων αυτών δείχνουν ότι τα τρόφιμα που κυκλοφορούν στην ελληνική αγορά είναι αρκετά κάτω από τα μέγιστα αποδεκτά όρια που έχει ορίσει η ΕΕ με τον κανονισμό 1881/2006/ΕΚ και γενικά κάτω από αυτά που αναφέρονται για άλλες ευρωπαϊκές και μεσογειακές χώρες. Επίσης, το ΕΦΑΜΑΔ έχει εκπονήσει μελέτες σε περιοχές όπου υπάρχουν υπόνοιες για επιβάρυνση τροφίμων από διοξίνες και παρόμοιες με διοξίνες ενώσεις, π.χ. λόγω της έκθεσης σε μολυσμένη τέφρα μετά από πυρκαγιές, κοντά σε χωματερές ή από βιομηχανική ρύπανση. Μερικά από τα δείγματα που αναλύθηκαν στις μελέτες αυτές εμφάνισαν ασυνήθιστα υψηλά επίπεδα επιβάρυνσης, ακόμα και πάνω από τα όρια που έχει θεσπίσει η ΕΕ. Στις περιπτώσεις αυτές, η δραστηριότητα του ΕΦΑΜΑΔ ήταν σημαντική για την προστασία της δημόσιας υγείας. Συγκεκριμένα, λήφθηκαν κατάλληλα μέτρα ώστε τα αντίστοιχα τρόφιμα να απομακρυνθούν από την κατανάλωση και να επανέλθουν οι συγκεντρώσεις διοξινών στα παραγόμενα τρόφιμα στα φυσιολογικά τους επίπεδα.

Λέξεις ευρετηρίασης: διοξίνες, παρόμοια με τις διοξίνες PCB, ελληνικά τρόφιμα, φασματομετρία μάζας

1. Introduction

Dioxins and dioxin-like compounds are highly toxic environmental pollutants belonging to the persistent organic pollutants (POPs). They include polychlorinated dibenzo-p-dioxins (PCDDs), seven of which are especially toxic, polychlorinated dibenzofurans (PCDFs), ten of which have "dioxin-like" toxicity, and polychlorinated diphenyls (PCBs), twelve of which also, possess "dioxin-like" properties. All these compounds are often referred to collectively – though technically incorrectly – as "dioxins". The toxicity of this broad group of compounds varies significantly and, in order to facilitate risk assessment and regulatory control, each one of the above toxic congeners (7 PCDDs, 10 PCDFs and 12 PCBs) has been assigned a Toxic Equivalency Factor, assuming as reference that the most toxic of these compounds, 2,3,7,8-tetrachlorodibenzodioxin (TCDD) by definition has a TEF of one (Pohjanvirta et al. 1994, Van den Berg et al. 2006). The concentration of each compound multiplied by its TEF is the Toxic Equivalency (TEQ) and the sum of TEQs of compounds detected in a sample consists its overall toxicity.

The toxicity of dioxin and dioxin-like compounds is mediated through the interaction with a specific intracellular protein, the aryl hydrocarbon (AH) receptor, a transcriptional enhancer, affecting a number of other regulatory proteins (Dencker 1985, Poellinger 2000, Linden et al. 2010). Their lipophilicity and long biological half-life (7-8 years for TCDD) lead to their chronic accumulation in the animal and human body (Froescheis et al. 2000, Startin and Rose 2003). The suspected toxic action of chronic exposure of humans to dioxins includes developmental effects (disturbance of tooth and sexual development), thyroid dysfunction, hepatic damage, immunological effects, as well as carcinogenicity, while the best proven effect of exposure to high amounts of dioxin is chloracne (Van den Berg et al. 1998, Bleeker et al. 1999, Birnbaum and Tuomisto 2000, ATSDR Tox Profiles 2002, Bencko 2003).

PCDD/Fs have never been produced intentionally for industrial use. Their route of formation and their entry in the environment are by incomplete combustion of organic material in the presence of chlorine, either in industrial processes, such as metal smelting and refining, manufacturing of chlorinated chemicals and paper bleaching (Schramm et al. 1998, Ruokojarvi et al. 2000, McKay 2002), or in intentional or unintentional combustion of plastics that takes place during incineration of solid medical and household waste (Walker and Cooper 1992, Quass et al. 2004) and accidental fires occurring in forest areas and landfills. In Greece, the dominant source of PCDDs/Fs is considered to be the uncontrolled combustion of municipal solid waste in open landfills, since industrial emission sources are very limited (Martens et al. 1998, Vassiliadou et al. 2009).

In contrast to PCDD/Fs, PCBs have been produced industrially for use in various applications including electronic appliances, heat transfer systems and hydraulic fluids, as coolant fluids in transformers and dielectric fluids in capacitors. PCBs were produced as mixtures; individual congeners were seldom synthesized. PCBs production was banned in 1979 by the United States Congress, because of their reported health effects, and in 2001, they were included in the list of compounds of The Stockholm Convention on Persistent Organic Pollutants (http://chm.pops.int/Convention/ThePOPs/tabid/673/language/en-US/Default.aspx). Human exposure still occurs through leakage of old capacitors and transformers and disposal of contaminated materials (Porta and Zumeta 2002, Rudel et al. 2008).

Food is generally recognized as the main source for human intake of PCDDs/Fs and dioxin-like PCBs. Over 90% of human exposure to dioxin-like compounds has been attributed to food intake (Liem et al. 2000). In particular, fat-containing food of animal origin (such as meat, dairy products and eggs) are considered, together with fatty fish, the main source of human exposure to dioxins. This is due to the slowly metabolizing and lipophilic nature of these compounds, leading them to accumulate in the fat of animals with a high position in the food chain (Liem and Theelen 1997, Focant et al. 2002, Fernandes et al. 2010). In evaluating the risk for humans, surveys in various countries have determined the intake of PCDDs/Fs and dioxin-like PCBs from total diet study (TDS) samples and individual foodstuffs (Liem et al. 2000). The European Union has set maximum limits for PCDDs/Fs and dioxin like PCBs in food products, which are specified in Commission Regulation 1881/2006. Moreover, the World Health Organization (WHO) has established a tolerable daily intake (TDI) of 1-4 pg TEQs/kg body weight (van Leeuwen et al. 2000).

Dioxin analysis is especially difficult and demanding, since concentrations of ppt (parts per trillion) need to be determined with accuracy and reproducibility. The analytical procedure involves the selective clean-up of a lipid extract on silica gel, modified silica, alumina or Florisil and activated carbon columns.

The final determination is performed by gas chromatography-mass spectrometry, by the isotope dilution technique with ^{13}C labelled analogues as internal standards. The detection is achieved by high resolution mass spectrometry, HRMS, at 10.000 resolving power (10% valley definition), so as to minimize the detection limit and increase selectivity.

The Mass Spectrometry and Dioxin Analysis Laboratory of NCSR "Demokritos" is the only laboratory for analysis of dioxins and dioxin-like PCBs in Greece. It has been operating since 2002 and it performs analysis of dioxins and dioxin-like compounds in food, feed, biological and environmental samples, applying international protocols (EPA 1613, EPA 1668), using the isotope dilution method, as specified by the EU Regulation 1883/2006/EC and the Directive 2002/70/EC. It has been accredited according to ISO/IEC 17025 by ESYD and it is the National Reference Laboratory of the European Commission for the control of dioxins and dioxin-like compounds for Greece and Cyprus. It participates in the United

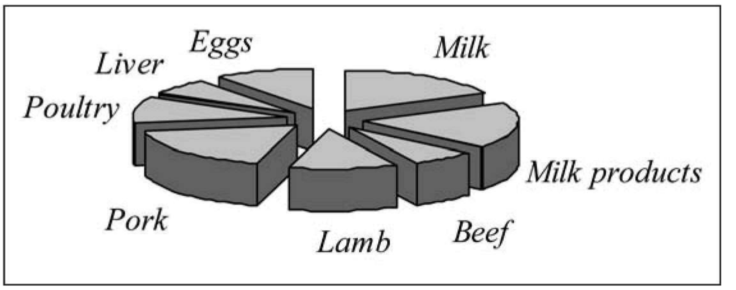

Figure 1. Circle graph representing the percentage of samples from each category of products of animal origin analyzed at the Dioxin Analysis and Mass Spectrometry Laboratory between 2002 and 2010 (total = 286 samples).

Nations laboratories network for pollution monitoring by toxic organic compounds.

Here, we present a comprehensive report of the activities of the Mass Spectrometry and Dioxin Analysis Laboratory during its operation, concerning the monitoring of dioxins and similar toxic compounds in food of animal origin in Greece.

Through collaboration with the official bureaus of food, environment and health control organizations of Greece, the Laboratory has conducted analysis in a large number of food products sampled randomly in the Greek market, as well as food products of animal origin sampled near areas suspected for dioxin contamination, such as areas affected by accidental fires, landfills or exposed to industrial pollution.

It should be added that during its operation, besides food samples, the Mass Spectrometry and Dioxin Analysis Laboratory has analysed more than 400 samples of animal feed collected by the Greek Ministry of Agriculture or private companies. This feed is either imported for domestic use or produced in Greece and exported to other countries.

2. Levels of dioxins and dioxin-like compounds in food samples of animal origin from the Greek market

Since its establishment in 2002, the Mass Spectrometry and Dioxin Analysis Laboratory of the National Centre for Scientific Research "Demokritos" has aimed at obtaining, for the first time in Greece, representative data on the levels of dioxins and dioxin-like compounds in food consumed by the general population of Greece. In collaboration with the Hellenic Food Authority, food samples are collected at production sites, as well as food retailers, in order to control their compliance to the accepted limits specified by the EEC law (Regulation 1881/2006). Food samples are collected according to the European Commission Regulation 1883/2006, which takes into account the production of each country. Special care is taken so that the food items selected are representative of the Greek diet. The samples are transported to the laboratory and stored at -20°C until their analysis. Analysis is conducted according to an accredited method that has already been described in detail (Papadopoulos et al. 2004, Costopoulou et al. 2006).

Here, we present the results collected in the eight years of operation of the laboratory for food products of animal origin, namely butter, cheese, yoghurt, milk, eggs and meat (286 samples in total). The percentage of samples from each category of food products is represented schematically in Figure 1. The average upperbound WHO-TEQ values for PCDD/Fs and PCBs are given in Table 1, along with minimun and maximum values for each sample category. The term "upperbound" refers to the value calculated, assuming that non-detected individual congener concentrations are equal to their corresponding limits of detection. The WHO-TEQ values are obtained by multiplying the concentrations with the corresponding WHO-TEF for each congener (Van den Berg et al. 2006). As required by the EC Regulation 1881/2006, concentrations and TEQ values of all compounds are reported on a fat basis (pg/g fat) for all food products, except fish.

Although, as expected, the levels of dioxin contamination vary among the different food categories, in all cases they are well below the EU maximum levels and, in general, lower than those monitored in other European and Mediterranean countries (Bascompta et al. 2002, Bocio et al. 2002, Bordajandi et al. 2002, Hosseinpour et al. 2002). In Table 2, we present the average WHO-TEQ values for TCDD/Fs and total dioxin like-PCBs, compared to the EU maximum levels and the normal levels in food across European countries, according to the Scientific report of EFSA, 2010, where the levels of dioxin contamination amongst European countries are compared. These results are in

Table 1. Food of animal origin average concentrations of PCDD/Fs, non-ortho PCBs and mono-ortho PCBs, along with minimum and maximum values for each sample category (n= No of samples)

	Concentration (pg WHO-TEQ / g lipid)					
Food category	PCDD/F	n	non-ortho PCB	n	mono-ortho PCB	n
Milk	0.39 (0.34-0.59)	51	0.17 (0.00-0.79)	51	0.07 (0.00-0.27)	45
Milk products	0.53 (0.29-1.88)	52	0.34 (0.01-2.48)	52	0.10 (0.03-0.45)	42
Beef	0.52 (0.34-0.94)	19	0.28 (0.01-0.57)	19	0.12 (0.02-0.37)	16
Lamb	0.49 (0.22-2.69)	33	0.20 (0.05-0.67)	33	0.09 (0.01-0.52)	27
Pork	0.36 (0.34-0.6)	52	0.07 (0.01-1.22)	52	0.05 (0.01-0.44)	49
Poultry	0.46 (0.23-1.48)	30	0.50 (0.01-11.00)	30	0.14 (0.01-1.18)	27
Liver	3.03 (1.17-5.9)	19	0.91 (0.01-2.79)	19	0.08 (0.02-0.27)	16
Eggs	0.38 (0.26-0.73)	30	0.08 (0.01-0.33)	30	0.03 (0.00-0.12)	26
Total n=		***286***		***286***		***248***

Table 2. Average WHO-TEQ values for TCDD/Fs and total dioxin like-PCBs, compared to the EU maximum levels and the normal levels in food across European countries.

Food category	Maximum levels (EC Regulation 1881/2006)		Normal levels in Greek food of animal origin		Normal levels in food across European countries (Scientific report of EFSA, 2010)	
	Concentration (pg WHO-TEQ / g lipid)		Concentration (pg WHO-TEQ / g lipid)		Concentration (pg WHO-TEQ / g lipid)	
	PCDD/F	Total	PCDD/F	Total	PCDD/F	Total
Milk	3.0	6.0	0.39	0.63	0.67	1.62
Milk products	3.0	6.0	0.53	0.97	0.67	1.62
Beef	3.0	4.5	0.52	0.92	2.39	3.32
Lamb	3.0	4.5	0.49	0.78	2.39	3.32
Pork	1.0	1.5	0.36	0.48	0.42	0.70
Poultry	2.0	4.0	0.46	1.1	0.64	1.00
Liver	6.0	12.0	3.03	4.02	2.63	4.93
Eggs	3.0	6.0	0.38	0.49	0.83	1.71

agreement with the fact that Greece is not highly industrialized and has never produced PCBs. Any possible dioxin contamination is attributed to exposure to fire-affected areas or to contamination of the feed used for animal rearing (Papadopoulos et al. 2004).

3. Dioxin contamination of food products after a fire in the municipal landfill of Tagarades

In July 2006, a fire broke out in the public landfill of Tagarades, which is situated southeast of Thessaloniki, and it has been used for the disposal of domestic waste (organic, plastic, paper, etc) from the wider area of Thessaloniki for the last 30 years. Because of the presence of chlorine-containing waste, the combustion of which leads to the generation of dioxins and dioxin-like compounds, contaminated ash was produced, which was inevitably transferred through air and water to the surrounding area, where various animal rearing and agricultural units are situated. It was, therefore, judged necessary to monitor the levels of contamination of food produced in the area, in order to protect public health. For this purpose, meat, milk, egg and vegetable samples, as well as soil samples, were collected from locations up to 7 km from the affected area, during a time period beginning immediately after the fire and ending one year later. Sampling was performed by the Department of the Veterinary Public

Table 3. Results of monitoring of dioxins in food samples of animal origin collected in three sampling sessions (July, September and December 2006) around the area of the landfill of Tagarades, from locations indicated in the Table. Values above the maximum EU levels are in bold.

Sample	Distance and orientation relative to landfill	Dioxins pg/g fat TEQ WHO 1997			Normal levels in Greek food	Max. levels EU
		25/7/06	15/9/06	4/12/06		
Goat milk	0.5 km N	**3.10**	**11.00**	**6.67**		3
Sheep milk	2 km NE		1.65			3
Sheep milk	2.5 km NW	**3.84**		0.97		3
Goat milk	4 km SE	**9.12**	**8.98**	**4.50**	0.3-0.6	3
Goat milk	4 km SW	**3.72**	§2.22			3
Goat milk	6 km N (control)		0.80			3
Sheep milk	6 km E (control)		0.45			3
Goat meat	0.5 km N	**8.82**		**15.03**		3
Sheep meat	0.5 km N		1.77		0.5-0.9	3
Goat meat	2.5 km NW		2.76			3
Goat meat	4 km SE			**3.30**		3
Goat meat	4 km S		1.74			3
Chicken	2 km NE		**2.20**		0.2-0.5	2
Eggs	2.5 km NW		0.63		0.3-0.7	3

Health of the Veterinary Directorate. In particular, the milk, egg and meat samples were collected from nearby pens/milking facilities. The locations of sampling points, as well as the levels of dioxins in the food samples, are presented in Table 3.

In general, the levels of dioxins in all food samples of animal origin are higher than those normally found in food from the Greek market and production and, in some cases, indicated in bold in the Table, above the maximum accepted dioxin levels according to the EU requirements (Regulation 1881/2006/EC). Food samples collected more than 5 km away from the affected area showed no sign of contamination. No PCB contamination was detected. Interestingly, goat milk and meat had higher dioxin levels than sheep products and this fact was attributed to the higher motility of goats and to the difficulty in controlling their feed.

As a result of this study, the consumption of food products from the contaminated area was prohibited until it was established that their dioxin levels were back to normal. More than 80,000 kg of milk and more than 1.000 sheep and goats were destroyed. Animals living in a pen 500 m away from the fire location showed elevated rates of genetic disorders. More specifically, 10 of the 400 kids (not bred to the same buck) of the spring kidding period (2007), following the fire case, were born with visible birth defects, including bloated appearance of the head, especially the jaws, hairless body with hair coat only in the upper part of the head (forehead to muzzle) and the lower part of the legs, and, in one case, a very oblong muzzle (Vassiliadou et al. 2009).

Besides food of animal origin, the study, also, included the measurement of olives and soil from the contaminated area (Vassiliadou et al. 2009).

4. Study of the levels of dioxins and dioxin-like PCBs in food produced near industrial areas

In 2007, the Prefecture of Volos assigned the Dioxin Analysis and Mass Spectrometry Laboratory to investigate the levels of pollution by dioxins and dioxin-like compounds in the industrial area of Volos. Volos is a coastal port city situated in Central Greece (Thessaly), the economy of which relies to a large extent on industrial production. After the Second World War, it became one of the main industrial centres in Greece, contributing significantly to the Greek economy, and, though in the years that followed its industrial development slowed down considerably, many important industrial units continue to operate there, including a major Greek cement company, units for steel production and processing and several food-processing factories. Since the wider area of Volos is, also, a significant producer of agricultural and animal food products, it was considered important to investigate the impact of industrial pollution on the quality of food produced near the industrial area. In a study coordinated by the Mass Spectrometry and Dioxin Analysis Laboratory, the local Veterinary Authorities collected samples of animal products (sheep and goat milk and meat, cheese, eggs and chicken), in two sampling sessions conducted according to the above-mentioned Commission Regulation in 2007. The samples were analysed in the Dioxin Analysis and Mass Spectrometry Laboratory according to methods already described (Papadopoulos et al. 2004, Costopoulou et al. 2006) and the results are summarised in Table 4.

The sites selected were two farm units located less than 500 m away from the industrial area. The samples of the first sampling session included nine milk samples (sheep and goat), one chicken sample, one egg sample, one cheese sample and two lamb meat samples. Most of the samples had unusually high levels of dioxins and PCBs, especially PCBs. More specifically, as it can be seen in Table 4, some samples were above the EU limits for dioxins, some were above the EU limits for the sum of dioxins and PCBs, particularly due to PCB contamination, while others were below the EU limit, but nevertheless higher than normal levels of Greek food. This is an indication of a high contamination of the area due to industrial waste and the operation of blast furnaces.

After the analysis of the first group of samples, free grazing was prohibited and animals were constricted to controlled areas. A month later, two more milk samples were collected from the same farm units. Though the levels of dioxins and PCBs of the second sampling group were considerably lower than the first ones, one of the samples was still well above the accepted limits.

Similar studies have since been conducted by the Laboratory near other industrial areas of Greece. During November and December of 2010, samples of food of animal origin were collected and analysed, from the area of Doxato, Drama, in northern Greece. All samples originated from farms near a production plant involved in wood processing and impregnation, a process known to cause the production of dioxins (Schramm et al. 1998, McKay 2002). However, the samples analysed (two samples of chicken meat, two egg samples and four samples of sheep milk) were not found to contain levels of dioxins and dioxin-like PCBs above the usual levels of Greek products.

Table 4. Results of monitoring of dioxins and dioxin-like PCBs in samples collected near the industrial area of Volos from two sampling sessions performed in the summer of 2007.

Food sample	Concentration pg (WHO-TEQ/g lipid), calculated as upper-bound concentration		
	PCDD/Fs	PCBs	Total TEQ
First sampling			
Sheep milk No 1	1,78	9,81	11,59
Sheep milk No 2	4,55	22,60	27,15
Sheep milk No 3	1,53	9,62	11,15
Sheep milk No 4	4,49	18,43	22,92
Sheep milk No 5	0,55	1,55	2,10
Sheep milk No 6	1,24	1,85	3,09
Sheep milk No 7	0,49	1,22	1,71
Goat milk No 1	0,92	1,26	2,18
Goat milk No 2	0,41	1,22	1,63
Cheese	3,54	13,16	16,70
Chicken	11,80	26,87	38,67
Eggs	5,28	12,87	18,15
Sheep meat No 1	2,89	13,92	16,81
Sheep meat No 2	0,80	3,42	4,22
Second Sampling			
Sheep milk No 1	0,75	4,64	5,39
Sheep milk No 2	2,15	11,74	13,89

Similarly, food product samples (ten samples of sheep milk, one egg sample and one sheep meat sample) collected from the area of Almyros, Central Greece, near a steel pipe manufacturing plant, between May and June 2010, were all within the usual levels of Greek food concerning dioxin and PCB contamination.

Conclusions

Since 2002, the Mass Spectrometry and Dioxin Analysis Laboratory has been monitoring the levels of dioxins and dioxin-like compounds in food products from the Greek market. In general, Greek food of animal origin have been found to contain levels of these compounds similar to or even lower than those of similar products from other European countries, which are well below the limits set by the European Union. This has been attributed to the fact that industrial development in Greece is limited and PCBs have never been produced. A few contaminated samples have been detected in areas affected by fire or industrial pollution. In each of these cases, measures were taken by the Greek authorities in order to constrict their distribution and consumption.

REFERENCES

ATSDR (2002). ATSDR Tox Profiles, Atlanta, 2002. http://www.atsdr.cdc.gov [accessed 6 June 2011]

Bascompta O, Montana MJ, Marti R, Broto-Puig F, Comellas L, Diaz-Ferrero J, Rodriguez-Larena MC (2002) Levels of persistent organic pollutants (PCDD/F and dioxin-like PCB) in food from the Mediterranean diet. Organohalogen Compounds 57: 149.

Bencko V (2003) Risk assessment and human exposure to endocrine disrupters. In: Jedrychovski WA, Petera FP, Maugeri U (Eds.), Molecular Epidemiology in Preventive Medicine. International Center for Studies and Research in Biomedicine in Luxembourg: pp. 315–327.

Birnbaum LS, Tuomisto J (2000) Non-carcinogenic effects of TCDD in animals. Food Addit Contam 17: 275–288.

Bleeker I, Fischer AB, Tilkes F, Eikmann T (1999) PCB Konzentrationen im menschlichen Blut. Umwelt Forsch Prax 4: 84–96.

Bocio A, Llobet JM, Domingo JL, Casas C, Teixido A, Muller L (2002) Levels of PCDD/PCDFs in food samples from Catalonia, Spain. Organohalogen Compounds 57: 105.

Bordajandi LR, Herrero L, Abad E, Rivera J, Gonzalez MJ (2002) Presence of PCDDs, PCDFs and PCBs in selected food samples from Huelva (Spain). Organohalogen Compounds 57: 69.

Costopoulou D, Vassiliadou I, Papadopoulos A, Makropoulos V, Leondiadis L. Chemosphere (2006) Levels of dioxins, furans and PCBs in human serum and milk of people living in Greece. Chemosphere 65:1462-9.

Dencker L (1985) The role of receptors in 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) toxicity. Arch Toxicol Suppl 8: 43-60.

EFSA Journal (2010) 8:1701-35.

Fernandes A, Mortimer D, Gem M, Rose M (2010) Dioxins and PCBs in Offal: occurrence and dietary exposure. Chemosphere 81: 536–540.

Focant J-F, Eppe G, Pirard C, Massart A-C, Andre J-E, De Pauw E (2002) Levels and congener distributions of PCDDs, PCDFs and non-ortho PCBs in Belgian foodstuffs. Assessment of dietary intake. Chemosphere 48: 167–179.

Froescheis O, Looser R, Cailliet GM, Jarman WM, Ballschmiter K (2000) The deepsea as a final global sink of semivolatile persistent organic pollutants? Part 1. PCBs in surface and deepsea dwelling fish of North and South Atlantic and the Monterey Bay Canyon (California). Chemosphere 40, 651–660.

Hosseinpour J, Rottler H, Joas R, Potrykus A, Schott R (2002) Dioxin-like PCB levels in feed and food from across the European Union. Organohalogen Compounds 57: 77.

Leondiadis L, Costopoulou D, Vassiliadou I, Papadopoulos A (2008) Monitoring of dioxins and dioxin-like PCBs in food, feed and biological samples in Greece. In: The Fate of Persistent Organic Pollutants in the Environment. Springer Ed: pp 83-98.

Liem AK, Furst P, Rappe C (2000) Exposure of populations to dioxins and related compounds. Food Additives and Contaminants 17: 241–259.

Liem AKD, Theelen RMC (1997) Dioxins: chemical analysis, exposure and risk assessment. Thesis, National Institute of Public Health and the Environment, Bilthoven, The Netherlands, ISBN 90-393-2012-8.

Lindén J, Lensu S, Tuomisto J, Pohjanvirta R (2010) Dioxins, the aryl hydrocarbon receptor and the central regulation of energy balance. A review. Frontiers in Neuroendocrinology 31: 452–478.

Martens D, Balta-Brouma K, Brotsack R, Michalke B, Schramel P, Klimm C, Henkelmann B, Oxynos K, Schramm K-W, Diamadopoulos E, Kettrup A (1998) Chemical impact of uncontrolled solid waste combustion to the vicinity of the Kouroupitos ravine, Crete, Greece. Chemosphere 36: 2855–2866.

McKay G (2002) Dioxin characterization, formation and minimization during municipal solid waste (MSW) incineration: review. Chemical Engineering Journal 86: 343–368.

Papadopoulos A, Vassiliadou I, Costopoulou D, Papanicolaou C, Leondiadis L (2004) Levels of dioxins and dioxin-like PCBs in food samples on the Greek market. Chemosphere 57:413-9.

Poellinger L (2000) Mechanistic aspects – the dioxin (aryl hydrocarbon) receptor. Food Additives and Contaminants 17 (4): 261–266.

Pohjanvirta R, Tuomisto J (1994) Short-term toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin in laboratory animals: effects, mechanisms and animal models. Pharmacol Rev 46: 483–549.

Porta M, Zumeta E (2002) Implementing the Stockholm treaty on POPs (Editorial). Occupational & Environmental Medicine 59: 651-652.

Quaß U, Fermann M, Broeker G (2004) The European dioxin air emission inventory project – Final results. Chemosphere 54: 1319–1327.

Rudel RA, Seryak LM, and Brody JG (2008) PCB-containing wood floor finish is a likely source of elevated PCBs in resident's blood, household air and dust: a case study of exposure. Environmental Health 7: 2-9.

Ruokojarvi P, Aatamila M, Ruushanen J (2000) Toxic chlorinated andpolyaromatic hydrocarbons in simulated house fires. Chemosphere 41: 825–828.

Schramm K-W, Kaune A, Lehnardt R, Hofmaier A, Henkelmann B, Kettrup A (1998) Isokinetic sampling of PCFF/F response in low and high volatile fractions of a wood incinerator. Organohalogen Compounds 36: 289–292.

Startin JR, Rose MD (2003) Dioxins and dioxin-like PCBs in food. In: Schecter A, Gasiewicz TA (Eds.), Dioxins and Health. WileyInterscience, New York, USA: pp. 89–136.

Van den Berg M, Birnbaum L, Bosveld AT, Brunstrom B, Cook P, Feeley M, Giesy JP, Hanberg A, Hasegawa R, Kennedy SW, Kubiak T, Larsen JC, van Leeuwen FX, Liem AK, Nolt C, Peterson RE, Poellinger L, Safe S, Schrenk D, Tillitt D, Tysklind M, Younes M, Waern F, Zacharewski T (1998) Environmental Health Perspectives 106: 775–792.

Van den Berg M, Birnbaum LS, Denison M, De Vito M, Farland W, Feeley M, Fiedler H, Hakansson H, Hanberg A, Haws L, Rose M, Safe S, Schrenk D, Tohyama C, Tritscher A, Tuomisto J, Tysklind M, Walker N, Peterson RE (2006) The 2005 World Health Organization re-evaluation of human and mammalian toxic equivalency factors for dioxins and dioxin-like compounds. Toxicol Sci 93: 223–241.

Van Leeuwen FX, Feeley M, Schrenk D, Larsen JC, Farland W, Younes M (2000) Dioxins: WHO's tolerable daily intake (TDI) revisited. Chemosphere 40: 1095-101.

Vassiliadou I, Papadopoulos A, Costopoulou D, Vasiliadou S, Christoforou S, Leondiadis L (2009) Dioxin contamination after an accidental fire in the municipal landfill of Tagarades, Thessaloniki, Greece. Chemosphere 74:879-84.

Walker B, Cooper C (1992). Air pollution emission factors for medical waste incinerators. Journal of the Air and Waste Management Association 42: 784–789.