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Increased levels of polychlorobiphenyls in Italian women with endometriosis

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Abstract

Endometriosis has been hypothesised to be linked to persistent and toxic organochlorinated chemicals. Dioxins and dioxin-like compounds have in particular been associated with the disease, mainly on the basis of experimental studies. Data in women are conflicting. A case-control study on 80 Italian nulliparous women of reproductive age was carried out to assess whether there is a correlation between the presence of endometriosis and blood levels of polychlorobiphenyls (PCBs), a family of ubiquitary environmental pollutants which comprises congeners with dioxin-like activity. Higher levels of PCBs were found in women with endometriosis. A mean cumulative value of 410 ng g^{-1}, lipid base, was found in cases versus the value of 250 ng g^{-1} observed in the control group (odds ratio for upper tertile 4.0, CI 95% 1.3–13; $p = 0.0003$). PCB increase involved both dioxin-like (PCBs 105, 118, 156, and 167) and non-dioxin-like congeners (PCBs 101, 138, 153, 170, 180).

Keywords: Prevalent PCBs; Blood levels; Endometriosis; Nulliparous women

1. Introduction

Endometriosis is a common, estrogen dependent, gynaecologic disease, often associated with infertility and pelvic pain. Its prevalence in the general population is approximately 10% (Eskenazi and Warner, 1997), but this figure is probably underestimated, since it includes only women presenting symptoms and looking for medical care. Endometriosis has been found at laparoscopy in about 15% of asymptomatic women (Leibson et al., 2004). Direct visualisation and biopsy during laparoscopy or laparotomy is the gold standard diagnostic test for this condition and enables the gynaecologist to identify the location, extent and severity of the disease and to perform the treatment. Its etiology and pathogenesis are likely to be multifactorial: genetic susceptibility, menstrual and reproductive history, immunologic factors, and environmental factors may be involved in the disease etiology. The possible role as a risk factor of highly persistent and lipophilic organochlorinated environmental

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pollutants, which biomagnify along food chains and bioaccumulate in human body, has been recently driving much attention. Exposure to "dioxins"—the families of polychlorodibenzodioxins (PCDDs) and polychlorodibenzofurans (PCDFs) among which the 2,3,7,8-tetrachlorodibenzodioxin or "dioxin"—and to polychlorobiphenyls (PCBs) has been hypothesised to play a role in endometriosis pathogenesis via endocrine and/or immune dysregulations. This hypothesis was first proposed (Rier et al., 1993) when a dose-dependent increase in the incidence and severity of spontaneous endometriosis was observed in a colony of rhesus monkeys that had been chronically exposed to dioxin, and held up for over ten years after the termination of exposure. These findings, although "incidentally" obtained (endometriosis, which was not a prospectively defined primary endpoint of the experiment, was observed many years after dioxin treatment had ceased), were included by the World Health Organization (1998) and the Scientific Committee on Food SCF (2000) among the pivotal experimental information used to derive the tolerable daily intake of dioxin and related compounds for humans. A few years later, the analysis of blood levels of dioxins and PCBs in the same cohort of monkeys (Rier et al., 2001), revealed an association between endometriosis and some dioxin-like PCBs, whose source in the monkeys could not be clearly determined. In the light of these findings of difficult interpretation, the SCF decided not to include Rier's first study (Rier et al., 1993) among those used to carry out the updated risk assessment of dioxins (Scientific Committee on Food, 2001).

So far, the most solid indication (Guo, 2004; Scientific Committee on Food, 2001) on an association between dioxin and endometriosis on non-human primates comes from a study carried out on cynomolgus monkeys (Yang et al., 2000) showing a promotion by dioxin of the growth and survival of autotransplanted endometrial tissue.

In humans, an association of endometriosis with dioxins has been proposed by some Belgian researchers (Koninckx et al., 1994), who suggested that the higher prevalence of endometriosis in their country could be related to the relatively high dioxin concentration in the Belgian population (World Health Organization, 1989; Van Larebeke et al., 2001).

Indeed, in a pilot case-control study we carried out on a small number of Italian and Belgian women (De Felip et al., 2004a), a significant difference (2.5-fold) in dioxin-like compound blood levels was observed between women from the two countries, while no significant differences were found in dioxin-like compound body burdens between women with or without endometriosis. On the whole, results from the few human studies carried out to test the hypothesis of an association between endometriosis and dioxins and/or PCBs are contradictory. Some studies showed in fact a significant association between endometriosis and dioxin (Mayani et al., 1997), or some PCBs (Gerhard and Runnebaum, 1992; Heilier et al., 2004; Buck Louis et al., 2005), while a non-significative risk of the disease associated to exposure to dioxin-like compounds (Pauwels et al., 2001), dioxin (Eskenazi et al., 2002), and a number of PCBs (Lebel et al., 1998) was reported by others. A comparison among these studies is made difficult by major differences in study design and methodology.

Generally, exposure to dioxins and dioxin-like PCBs occurs at the same time as that of the much more abundant non-dioxin-like PCBs. These congeners, characterised by two or more chlorines in the *ortho* positions, are present in the environment, in food and hence in human tissues in concentrations of up to three orders of magnitude higher than dioxins and dioxin-like PCBs. In particular, congeners most abundant in human tissues (concentrations usually >1 ng g^{-1} fat), as a result of their biaccumulation in food and their poor metabolism, are the ortho-substituted PCBs 28, 52, 101, 118, 138, 153, 180, referred to as "indicators" (Appel, 2003), because currently used to estimate the PCB total content of a matrix, plus a few others as PCBs 105, 156, 167 (with weak dioxin-like activity), and 170. The three congeners 138, 153 and 180 dominate in all human tissues and account for at least 50% up to 80% of total PCB content in serum (Glynn et al., 2000) adipose tissue, breast milk (Noren and Meironyté, 2000; Wingfors et al., 2000), and follicular fluid (De Felip et al., 2004b).

Non-dioxin-like PCB toxicity has been raising growing concern for the adverse effects shown in vivo and in vitro models, including the ability to interfere with sexual hormone-regulated processes shown by PCBs 138, 153 and 180 (Bonefeld-Jorgensen et al., 2001).

Because of this potential of altering the steroid hormone signalling, and considering the abundance of these compounds in human tissues, we carried out the present study to explore the correlation between the blood levels of prevalent PCBs and the presence of endometriosis in nulliparous women of reproductive age from the area of Rome.

2. Materials and methods

2.1. Patients

One hundred and fifty-four women living in the Rome area, undergoing laparoscopy for suspected endometriosis or other benign gynaecological conditions from April 2000 to January 2004 at the Department of Gynaecological Sciences, Perinatology and Child Health, of the Policlinico Umberto I—University of Rome "La Sapienza", were considered eligible for the study. All women signed an informed consent to the study. A physician unaware of the indications to lapa-

roscopy administered a questionnaire before surgery which documented age, education, job, medical, gynecological and obstetrical history, height and weight, smoking and dietary habits. The questionnaire was designed to ascertain information on potential confounders, including metabolic diseases, gravidity, parity, and weight changes in the last years. For each woman, body mass index (BMI) was calculated. Only women undergoing laparoscopy were enrolled in the study in order to assess or rule out the presence of endometriosis, since its diagnosis cannot be achieved with other non-invasive diagnostic methods. Women with no visual and histological evidence of endometriosis were considered controls. Before laparoscopy, a blood specimen of approximately 30 ml was collected from cubital vein in vacutainer tubes containing heparin; specimens were stored at −20 °C until subjected to analytical processing. A 10-mm laparoscopy under general anesthesia was performed. The presence of endometriosis was confirmed by histological examination of lesions. Random peritoneal biopsies were performed in those women with no laparoscopic evidence of endometriosis; histology confirmed the absence of the disease. Seventy women had laparoscopic and histological confirmed endometriosis and 84 cases had other benign gynecological conditions with no visual or histological evidence of the disease. Only 80 patients participated to the study, meeting the inclusion criteria of selection: nulliparity, age (range: 20–40 years), absence of any other acute or chronic disease, and no evidence of relevant weight modifications (more than 10 kg) in the last years.

Forty women had histologically confirmed endometriosis (cases), whereas 40 had benign gynecologic conditions (ectopic pregnancies, uterine myomas, benign adnexal masses, fallopian tubes abnormalities), with no evidence of endometriosis (controls).

2.2. Analysis

Blood samples (10 ml) were added with a ^{13}C mixture of labelled PCBs (28, 52, 101, 118, 138, 153, 156, 180) and allowed to rest overnight. Samples were added with 15 ml of formic acid/2-propanol (4/1, v/v), sonicated and extracted by manual shaking with *n*-hexane. After centrifugation, the organic phase was removed and collected. This extraction process was repeated four times, and the *n*-hexane aliquots were pooled in a centrifuge tube and concentrated. After acidic treatment with concentrated H_2SO_4, the organic phase was eluted on a multilayer column (De Felip et al., 1990). Instrumental analysis was carried out by ion trap (IT) mass spectrometry (Thermofinnigan Polaris Q) coupled to high resolution gas chromatography and used in the MS–MS mode. The isotope dilution technique was applied throughout. Recovery ranges were 70–110% for ^{13}C-labelled congeners analysed.

Analytical reliability was specifically warranted by the analysis of frequent blind replicates and blanks. Accuracy was assessed by in-house reference matrices fortified with ^{13}C PCB congeners at concentrations close to background levels.

Lipid determination was carried out gravimetrically according to the method reported by Atuma and Aune (1999).

The laboratory has a long time experience in the analysis of microcontaminants. Over the years it has participated in several interlaboratory comparisons on the analysis of dioxin-like and indicator PCBs, PCDDs and PCDFs in food and maternal milk, among which the "Interlaboratory Comparison on Dioxins in Food 2005" organised by the Norvegian Institute of Public Health.

2.3. Statistical analysis

In order to assess if differences in PCB concentrations between cases and controls were statistically significant, a non-parametric test (Mann–Whitney's *U*-test) was performed (STATISTICA, version 6.0). We also divided the 80 women enrolled (cases and controls) into three groups on the basis of their PCB blood levels using tertiles. Odds ratios were then calculated to measure the risk of endometriosis for the two groups characterised by higher PCB blood levels using the group with PCB levels under the first tertile as the reference category. Adjusted odds ratios and their 95% CIs were calculated by logistic regression (STATA Statistical Software, version 8.0).

3. Results and discussion

Characteristics of the women enrolled in the study are shown in Table 1. Cases and controls are well matched regarding BMI ($p = 0.65$, Mann–Whitney's *U*-test), while the two groups' mean age differs by five years.

The results of the congener-specific PCB analysis carried out on the two groups of women (Table 2) show that PCB levels, as a sum of all congeners assessed, are higher in women with endometriosis, characterised by a mean cumulative value of 410 ng g^{-1}, lipid base, versus the value of 250 ng g^{-1} observed in the control group. PCB increase involves both dioxin-like (PCBs 105, 118, 156, and 167) and non-dioxin-like congeners (PCBs 101, 138, 153, 170, 180). The application of the two-sided Mann–Whitney's *U*-test shows a highly significant difference ($p < 0.001$) between cases and controls with respect to the sum of all PCBs, and for the individual congeners PCBs 138, 153, and 180 (Table 3). In particular, the lowest *p* values and the highest odds ratios are observed for PCBs 153 and 180, and for the sum of all

Table 1
Characteristics of the Italian women enrolled in the study

	Cases ($N = 40$)	Controls ($N = 40$)
Age		
Mean value (years)	29	34
Range (years)	22–40	20–40
Body mass index		
Mean value	22	23
Range	16–29	15–40
<20	14	16
⩾20	26	24
Smoking status		
Never	21	20
Former	5	3
Current	14	17
Average number of cigarettes per day		
Mean value (cigarettes/day)	14	12
<15	8	10
⩾15	6	7
Years of smoking		
Mean value (years)	14	9.8
<10	5	7
⩾10	9	10

Table 2
Selected PCB congener concentrations (ng g^{-1}, lipid base) in human blood samples of Italian women with (cases) and without (controls) endometriosis

PCB congener	Cases ($N = 40$)		Controls ($N = 40$)	
	Mean	SD	Mean	SD
PCB-28	5.5	9.5	5.6	8.3
PCB-52	2.5	2.7	3.0	5.5
PCB-101	3.6	3.0	3.1	3.2
PCB-105	10	10	8.0	9.6
PCB-118	51	91	24	24
PCB-138	85	50	53	26
PCB-153	150	81	95	55
PCB-156	22	28	10	10
PCB-167	11	21	5.2	5.0
PCB-170	13	8.7	10	11
PCB-180	65	25	45	28
∑PCBs	410	220	250	140

Values rounded off to two figures.

congeners: odds ratios for the patients in the mid and upper categories are 5.6 and 7.0 and 8.3 and 8.8 for PCBs 153 and 180, and 3.2 and 4.0 for the sum of all PCBs. When the potential confounding effects of age and smoking habits are taken into account, the differences between cases and controls remain significant and odds ratios increase for most of the congeners and for their sum (Table 3).

PCB congener-specific profiles in cases and controls appear to be substantially overlapping (Fig. 1), this suggesting homogeneity of exposure. Dietary habits of all the subjects, as characterised from administered questionnaires, were substantially representative of the mean Italian diet.

Our findings are consistent with two other works, although differences in study design, selection criteria (e.g. parity), and analytical methodology must be taken into account: an association between PCBs 138, 153 and 180 and endometriosis had been observed in a study carried out in German women (Gerhard and Runnebaum, 1992); in a recent study, higher levels of PCBs (expressed as a sum of selected congeners, among which PCBs 153 and 180) were assessed in Belgian women with rectovaginal nodules of adenomyosis (Heilier et al., 2004).

The present study was based on strict selection criteria, in order to exclude confounding factors, among which parity (breast-feeding is known to significantly reduce dioxin body burden), presence of any metabolic or other diseases that might influence the PCB levels, and substantial weight changes. Controls were not randomly selected from the local general population, because endometriosis cannot be excluded by other non-invasive diagnostic methods, therefore only patients with no laparoscopic and histological evidence of the disease were included. To our knowledge, no data are available on a possible influence of PCBs on the benign gynecological conditions that we observed in controls. As the analytical methodology used is a crucial parameter in providing univocal identification and reliable quantitation of different congeners, PCB analysis was carried out by a highly selective congener-specific technique; quality control and quality assurance measures were adopted to ensure analytical reliability.

The causes of PCB increase in women with endometriosis remain to be elucidated. Body burden is the result of an input–output balance. As for the input, dietary intake determines over 90% exposure: dietary habits documented by the questionnaires, did not account for any measurable difference in dietary exposure. PCB elimination is mostly determined by metabolism and faecal excretion; this latter is related to several factors, as BMI and a diet rich in non-absorbable lipophilic substances (Rohde et al., 1999), with respect to which we could not assess any difference. The difference in the observed levels might instead be due to variabilities in toxicokinetics, mainly regarding the biotransformation phase: different metabolic capabilities in this phase are usually due to both genetic polymorphism, or to exposure to environmental contaminants, resulting in induction/inhibition phenomena. PCBs are biotransformed by phase I cytochrome P450 isoforms (CYPs) into hydroxy metabolites (mainly monohydroxy metabolites, PCB-OHs) which are then conjugated for subsequent excretion with glucuronide, sulfate or glutathione, in

Table 3
Blood PCB concentrations (ng g^{-1}, lipid base) in Italian women with (cases) and without (controls) endometriosis

	Mean (SD)		p value[a]	Categories and tertiles	Odds ratios (CI 95%)[b]	
	Cases ($N = 40$)	Controls ($N = 40$)			Unadjusted	Adjusted[c]
PCB 118	51 (91)	24 (24)	0.0108	<15		
				15–28	3.2 (1.0–10)	3.2 (0.81–12)
				>28	4.0 (1.3–13)	4.6 (1.2–17)
PCB 138	85 (50)	53 (26)	0.0015	<47		
				47–76	2.8 (0.90–8.6)	4.5 (1.1–19)
				>76	4.8 (1.5–15)	6.6 (1.6–27)
PCB 153	150 (81)	95 (55)	0.0004	<93		
				93–130	5.6 (1.7–19)	10 (2.1–48)
				>130	7.0 (2.1–23)	9.1 (1.9–43)
PCB 180	65 (25)	45 (28)	0.0002	<37		
				37–64	8.3 (2.4–29)	5.8 (1.4–24)
				>64	8.8 (2.5–31)	4.0 (1.0–16)
∑PCBs	410 (220)	250 (140)	0.0003	<250		
				250–360	3.2 (1.0–10)	6.5 (1.5–28)
				>360	4.0 (1.3–13)	5.3 (1.3–23)

[a] Mann–Whitney U-test.
[b] The group with blood contamination level under the lower tertile is the reference category (odds ratio = 1.00).
[c] Adjusted by age and smoking habits.

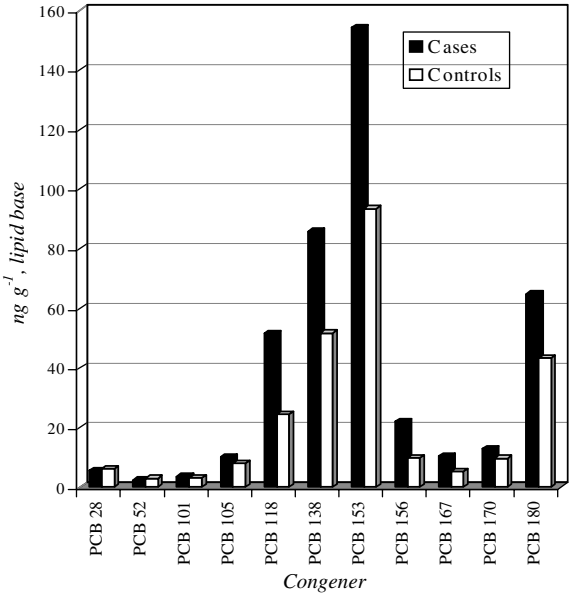

Fig. 1. Blood PCB congener-specific concentrations in Italian women with (cases) and without (controls) endometriosis.

reactions catalysed by phase II enzymes (James, 2001). CYP-mediated biotransformation is thus the first and critical event in the elimination of PCBs and CYP enzymes play an important role in determining the half-life and the extent of bioaccumulation of these compounds.

Also, genetic polymorphism of phase II enzymes, such as glutathione-S-transferases (GSTs), could be related to increased levels of PCBs and their metabolites. Indeed, the presence of GSTM1-null genotype has been hypothesised as a risk factor for the development of endometriosis (Baranova et al., 1999; Arvanitis et al., 2003): enzymes belonging to this family are important in the detoxification of active intermediates resulting from xenobiotic phase I metabolism, such as PCB oxygenated metabolites, which they conjugate for subsequent excretion. The absence of activity of this enzyme could result in an excess of oxygen species: chronic exposure of endometrium to these reactive intermediates may have an inflammatory effect in addition to the inflammatory activity of some PCB themselves. Some PCBs have in fact been shown to chronically stimulate the expression and activity of the pro-inflammatory cytokines and growth factors (IL1, IL-6, TNFα, and INFγ) involved in the cyclic regulation of endometrial remodeling, proliferation and cell death; these effects may promote ectopic tissue proliferation (Rier and Foster, 2002).

At present, the potential effects of PCBs in the pathogenesis of endometriosis are still to be elucidated. Many PCBs may interfere with the hormone receptor: in particular congeners found in higher concentrations (PCBs 138, 153, and 180) have been shown to have pleiotropic effects on the estrogen and androgen-receptor (Bonefeld-Jorgensen et al., 2001), and therefore to be able to interfere with sexual hormone-regulated processes, and to play a role in the disease etiology and/or progression.

The results obtained clearly show an increase of PCB congeners most abundant in human tissues in women with endometriosis. The interpretation of these results is made particularly difficult by the fact that the

observed increase involves congeners of both dioxin-like and non-dioxin-like type, this making necessary to considerate the broad range of different and not fully elucidated mechanisms of action pertinent to both groups of congeners. Further studies are ongoing aimed to understand the complex network of factors and the mechanisms that may be thought to be responsible of the association we observed, with a particular attention to polymorphism in genes encoding enzymes involved in PCB detoxification.

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References

Appel, K.E., 2003. Risk assessment of non-dioxin-like PCBs—report on a WHO-consultation. Fresenius Environ. Bull. 12, 268–275.

Arvanitis, D.A., Koumantakis, M.D., Goumenou, A.G., Matalliotakis, I.M., Koumantakis, E.E., Spandidos, D.A., 2003. CYP1A1, CYP19, and GSTM1 polymorphisms increase the risk of endometriosis. Fertil. Steril. 79, 702–709.

Atuma, S.S., Aune, M., 1999. Method for the determination of PCB congeners and chlorinated pesticides in human blood serum. Bull. Environ. Contam. Toxicol. 62, 8–15.

Baranova, H., Canis, M., Ivaschenko, T., Albuisson, E., Bothorishvilli, R., Baranov, V., Malet, P., Bruhat, M.A., 1999. Possibile involvement of arylamine *N*-acetyltransferases 2, glutathione *S*-transferase M1 and T1 genes in the development of endometriosis. Mol. Hum. Reprod. 5, 636–641.

Bonefeld-Jorgensen, E.C., Andersen, H.R., Rasmussen, T.H., Vinggaard, A.M., 2001. Effect of highly bioaccumulated polychlorinated biphenyl congeners on estrogen and androgen receptor activity. Toxicology 158, 141–153.

Buck Louis, G.M., Weiner, J.M., Whitcomb, B.W., Sperrazza, R., Schisterman, E.F., lobdell, D.T., Crickard, K., Kostyniak, P.J., 2005. Environmental PCB exposure and risk of endometriosis. Hum. Reprod. 20, 279–285.

De Felip, E., di Domenico, A., Grande, M., Pazzaglia, R., Falleni, M., 1990. Fast gas chromatographic assessment of polychlorobiphenyl levels in wall plaster coats. Inter. J. Environ. Anal. Chem. 38, 607–616.

De Felip, E., Porpora, M.G., di Domenico, A., Ingelido, A.M., Cardelli, M., Cosmi, E.V., Donnez, J., 2004a. Dioxin-like compounds and endometriosis: a study on Italian and Belgian women of reproductive age. Toxicol. Lett. 150, 203–209.

De Felip, E., di Domenico, A., Miniero, R., Silvestroni, L., 2004b. Polychlorobiphenyls and other organochlorine compounds in human follicular fluid. Chemosphere 54, 1445–1449.

Eskenazi, B., Warner, M.L., 1997. Epidemiology of endometriosis. Obstet. Gynecol. Clin. North Am. 24, 235–258.

Eskenazi, B., Mocarelli, P., Warner, M., Samuels, S., Verzellini, P., Olive, D., Needham, L.L., Patterson, D.G., Brambilla, P., Gavoni, N., Casalini, S., Panazza, S., Turner, W., Gerthoux, P.M., 2002. Serum dioxin concentrations and endometriosis: a cohort study in Seveso, Italy. Environ. Health Perspect. 110, 629–634.

Gerhard, I., Runnebaum, B., 1992. The limits of hormone substitution in pollutant exposure and fertility disorders. Zentralb. Gynakol. 114, 593–602.

Glynn, A.W., Wolk, A., Aune, M., Atuma, S., Zettermark, A., Mæle-Schmid, M., Darnerud, P.O., Becker, W., Vessby, B., Adami, H., 2000. Serum concentrations of organochlorines in men: a search for markers of exposure. Sci. Total Environ. 263, 197–208.

Guo, S.W., 2004. The link between exposure to dioxin and endometriosis: A critical Reappraisal of primate data. Gynecol. Obstet. Invest 57, 157–173.

Heilier, J.F., Thi Ha, A., Donnez, J., Tonglet, R., Nackers, F., 2004. Increased serum polychlorobiphenyl levels in Belgian women with adenomyotic nodules of the rectovaginal septum. Fertil. Steril. 81, 456–458.

James, M.O., 2001. Polychlorinated biphenyls: metabolism and metabolites. In: Robertson, Larry W., Hanse, Larry G. (Eds.), PCBs: Recent Advances in Environmental Toxicology and Health Effects. The University Press of Kentucky.

Koninckx, P.R., Braet, P., Kennedy, S.H., Barlow, D.H., 1994. Dioxin pollution and endometriosis in Belgium. Hum. Reprod. 9, 1001–1002.

Lebel, G., Dodin, S., Ayotte, P., Marcoux, S., Ferron, L.A., Dewailly, E., 1998. Organochlorine exposure and the risk of endometriosis. Fertil. Steril. 69, 221–227.

Leibson, C.L., Good, A.E., Hass, S.L., Ransom, J., Yawn, B.P., O'Fallon, W.M., Melton 3rd, L.J., 2004. Incidence and characterization of diagnosed endometriosis in geographically defined population. Fertil. Steril. 82, 314–321.

Mayani, A., Barel, S., Soback, S., Almagor, M., 1997. Dioxin concentrations in women with endometriosis. Hum. Reprod. 12, 373–375.

Noren, K., Meironyté, D., 2000. Certain organochlorine and organobromine contaminants in Swedish human milk in perspective of past 20–30 years. Chemosphere 40, 1111–1123.

Pauwels, A., Schepens, P.J., D'Hooghe, T., Delbeke, L., Dhont, M., Brouwer, A., Weyler, J., 2001. The risk of endometriosis and exposure to dioxins and polychlorinated biphenyls: a case-control study of infertile women. Hum. Reprod. 16, 2050–2055.

Rier, S.E., Martin, D.C., Bowman, R.E., Dmowski, W.P., Becker, J.L., 1993. Endometriosis in rhesus monkeys (*Macaca mulatta*) following chronic exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. Fundam. Appl. Toxicol. 21, 433–441.

Rier, S.E., Turner, W.E., Martin, D.C., Morris, R., Lucier, G.W., Clark, G.C., 2001. Serum levels of TCDD and dioxin-like chemicals in Rhesus monkeys chronically

exposed to dioxin: correlation of increased serum PCB levels with endometriosis. Toxicol. Sci. 59, 147–159.

Rier, S.E., Foster, W.G., 2002. Environmental dioxins and endometriosis. Toxicol. Sci. 70, 161–170.

Rohde, S., Moser, G.A., Päpke, O., McLachlan, M.S., 1999. Clearance of PCDD/Fs, via the gastrointestinal tract in occupationally exposed persons. Chemosphere 38, 3397–3410.

Scientific Committee on Food, 2000. Opinion of the SCF on the risk assessment of dioxins and dioxin-like PCBs in food. Adopted on 22 November 2000. European Commission, Brussels.

Scientific Committee on Food, 2001. Opinion of the SCF on the risk assessment of dioxins and dioxin-like PCBs in food. Update based on new scientific information available since the adoption of the SCF opinion of 22nd November 2000. Adopted on 30 May 2001. European Commission, Brussels.

Van Larebeke, N., Hens, L., Schepens, P., Covaci, A., Baeyens, J., Everaert, K., Bernheim, J.L., Vlietink, R., De Poorter, G., 2001. The Belgian PCB and dioxin incident of January–June 1999: exposure data and potential impact on health. Environ. Health Perspect. 109, 265–273.

Wingfors, H., Linström, G., van Bavel, A., Schuhmacher, M., Hardell, L., 2000. Multivariate data evaluation of PCB and dioxin profiles in the general population in Sweden and Spain. Chemosphere 40, 1083–1088.

World Health Organization, 1989. Levels of PCBs, PCDDs and PCDFs in breast milk: Result of WHO Coordinated Interlaboratory Quality Control Studies and Analytical fields Studies. In: WHO Environmental Health Series, 34, WHO, Geneva, Switzerland.

World Health Organization, 1998. Executive summary—Assessment of the health risk of dioxins: Re-evaluation of the tolerable daily intake (TDI). WHO Consultation (Geneva), European centre for environment and health, International Programme on Chemical Safety.

Yang, J.Z., Agarwal, S.K., Forster, W.G., 2000. Subchronic exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin modulates the pathophysiology of endometriosis in the cynomolgus monkey. Toxicol. Sci. 56, 374–381.