Department for Environment, Food and Rural Affairs and the Environment Agency

CONTAMINANTS IN SOIL: COLLATION OF TOXICOLOGICAL DATA AND INTAKE VALUES FOR HUMANS. DIOXINS, FURANS AND DIOXIN-LIKE PCBs

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Statement of Use
This publication details the derivation of tolerable daily soil intakes or Index Doses for dioxins and dioxin-like PCBs. The report has been written for technical professionals who are familiar with the risks posed by land contamination to human health but who are not necessarily experts in risk assessment. It is expected to be of use to all parties involved with or interested in contamination, but in particular to those concerned with the assessment of land contamination.

Keywords
Tolerable daily soil intake, Index Dose, land contamination, risk assessment, human health, dioxins and furans, polychlorinated biphenyls (PCBs), dioxin-like PCBs, polychlorinated dibenzo-*para*-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs)

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1 Introduction

1.1 This report, one of a number on the assessment of risks to human health from contaminants in soil, presents key data and expert opinions on the toxicology and intake of dioxins and related compounds. It may be necessary to update this document in the future and incorporate new toxicological data as science advances.

1.2 The aim is to derive tolerable daily intakes (TDIs), which in turn are needed to derive Soil Guideline Values (SGVs) for dioxins, furans and dioxin-like polychlorinated biphenyls (PCBs), that is, concentrations that will pose no significant threat to human health.

1.3 There is a general discussion of TDIs in CLR9 *Contaminants in Soils: Collation of Toxicological Data and Intake Values for Humans. Consolidated Main Report* (Department for Environment, Food and Rural Affairs (Defra) and Environment Agency, 2002a). Reference to CLR9 is necessary to understand the concepts and terms used in this report.

1.4 The computer model used for deriving Soil Guideline Values is described in CLR10 *The Contaminated Land Exposure Assessment (CLEA) Model: Technical Basis and Algorithms* (Defra and Environment Agency, 2002b). The derivation of the Soil Guideline Values for dioxins, furans and dioxin-like PCBs will be published in SGV 11 *Guideline Values for Dioxin, Furan and Dioxin-like PCB Contamination* (Defra and Environment Agency, in preparation).

1.5 The literature up to June 2002 has been reviewed for this report.

2 Identity

2.1 The term “dioxins” originally referred to a family of organic compounds, the polychlorinated dibenzo-*para*-dioxins (PCDDs). It has recently broadened its scope and also now encompasses the structurally related family of polychlorinated dibenzofurans (PCDFs). Strictly speaking, the term “dioxins” should be reserved for the chlorinated dibenzo-*p*-dioxins and the term “furans” used for the chlorinated dibenzofurans. All are chlorinated tricyclic aromatics with similar chemical properties. Each compound is characterised by the number (up to eight) and the substitution position (1 to 4, and 6 to 9) of its chlorine atoms (Figure 2.1). There are 75 different PCDDs and 135 different PCDFs (WHO, 1989). The names of the individual compounds are usually abbreviated; for example, 2,3,7,8-tetrachlorodibenzo-*p*-dioxin is normally written as 2,3,7,8-TCDD. For the sake of brevity, 2,3,7,8-TCDD will subsequently be referred to in this review as “TCDD” and the dioxins/furans in general as “dioxins”. The coplanar polychlorinated biphenyls (PCBs) have demonstrated a close toxicological similarity to dioxins and are thought to operate by the same general mechanism (van den Berg *et al*, 1998, 2000). Expert Group assessments of dioxins are also applicable to these so-called dioxin-like PCBs (see paragraph 3.6).

2.2 Dioxins are solids with high melting and boiling points. There is some variation, however, between the volatility of individual congeners; for example, TCDD is considered to be significantly more volatile than octachlorodibenzo-*p*-dioxin (OCDD). Dioxins are almost insoluble in water (the solubility decreases with increasing chlorine content), but are quite soluble in organic solvents and fats. The solubility of TCDD is about 0.02 μg L^{-1} in water and about 0.5 g L^{-1} in benzene (ATSDR, 1998; WHO, 1989).

2.3 Dioxins have no known technical value, and are not produced intentionally. While there are some natural sources, such as forest fires and volcanic eruptions, these are not significant contributors in industrialised countries. Two main categories of anthropogenic sources are recognised: (i) the production and use of organochlorine chemicals that are contaminated with dioxins, and (ii) the combustion of chemical, clinical and household waste, as well as fuels such as coal, wood, natural gas and oil. Combustion is probably the main source of dioxins in the UK environment (Dyke *et al*, 1997; Harrad and Jones, 1992).

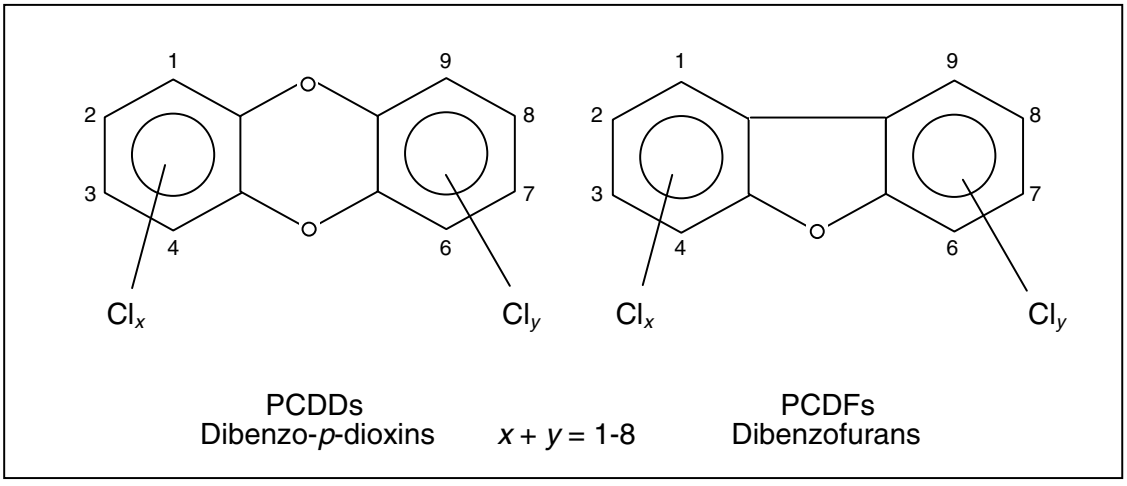

Figure 2.1 Structures of the chlorinated dibenzo-*p*-dioxins and dibenzofurans

2.4 Atmospheric dispersion, deposition and subsequent accumulation in the food chain are the major pathways of exposure, and subsequently uptake, for the general population. Dioxins adsorb strongly to soil and organic matter, where they persist for many years as a consequence of their chemical stability and their resistance to biodegradation.

3 Toxicity

3.1 Human exposure to TCDD has been associated with a wide variety of toxic effects, including cancer. Many of the published reports concern occupationally exposed groups, such as chemical production workers and pesticide users, and individuals who handled or were exposed to materials treated with TCDD-contaminated pesticides. There is also an extensive literature describing the health of those who had either consumed accidentally contaminated food products, or who were in areas contaminated with waste oil and industrial effluent (IARC, 1997; WHO, 1989).

3.2 There are numerous reviews of the health effects of dioxins, including those by the Agency for Toxic Substances and Disease Registry (ATSDR, 1998), the UK Government's independent advisory Committees on the Toxicity (COT), Mutagenicity (COM) and Carcinogenicity (COC) of Chemicals in Food, Consumer Products and the Environment (DoE, 1989; MAFF, 1992; DH, 1994, 1996, 1997, 1999a,b; FSA, 2001a,b), the International Agency for Research on Cancer (IARC, 1997), the United States Environmental Protection Agency (USEPA, 1994, 1995a,b, 2000), the EC's Scientific Committee for Food (EC, 2000, 2001) and the World Health Organization (van Leeuwen and Younes, 2000; WHO, 1989, 1991, 1998, 2001). This section is based largely on these reviews. More weight has been given to the more recent publications, and in general the primary publications have not been consulted.

3.3 **Toxic equivalency factors.** Only those 17 dioxins that contain chlorine atoms at any of four specific positions (denoted by 2, 3, 7 and 8) on the molecule are thought to be of toxicological significance. The most toxic is considered to be TCDD and, consequently, most of the toxicological studies have been carried out on this molecule. Although there are far fewer data on the other sixteen 2,3,7,8-substituted compounds, dioxins are found in the environment as complex mixtures, and it is necessary to have some way of assessing the toxicity of these mixtures. Various systems of "toxicity equivalents" have been developed for this purpose.

3.4 All such systems use the available toxicity data, together with a knowledge of the structural similarities among the PCDDs and PCDFs, to estimate weighting factors, each of which expresses the toxicity of a particular PCDD or PCDF relative to an equivalent amount of TCDD. Multiplication of the concentration of the particular compound by this factor gives a TCDD Toxic Equivalent (TEQ). The toxicity of any mixture relative to TCDD is then the sum of the individual TEQs. The method allocates Toxic Equivalency Factors (TEFs) only to the compounds of toxicological concern, that is, the seventeen 2,3,7,8-substituted compounds, with all other PCDD and PCDF compounds being given a weighting factor of zero.

3.5 Until recently, the most widely accepted system of TEQs was that proposed by the North Atlantic Treaty Organisation (NATO) Committee on Challenges to Modern Society, known as the International Toxicity Equivalency Factor (I-TEF) system (Kutz *et al*, 1990). In 1997, a World Health Organization (WHO) Expert Group reassessed TEFs using a tiered approach, in which the results of animal toxicity studies, especially those involving chronic or sub-chronic exposure, were given more weight than the results of *in*

vitro or biochemical studies (van den Berg *et al*, 1998). The UK Government's independent advisory Committee on the Toxicity of Chemicals in Food, Consumer Products and the Environment (COT) (DH, 1997, 1999a) has endorsed the use of the WHO-TEQs.

3.6 The close toxicological similarity of coplanar polychlorinated biphenyls (PCBs) to dioxins has led to the extension of the TEF system to some of the coplanar PCB compounds, the dioxin-like PCBs, and for the TEQ of an environmental sample to include contributions from both dioxins and dioxin-like PCBs. The WHO system of TEFs includes TEF values for 12 non-*ortho*- and mono-*ortho*-PCBs that have demonstrated dioxin-like toxicity (Table 3.1).

Table 3.1 TEF values according to the International System (I-TEFs; Kutz *et al*, 1990) and the WHO System (WHO-TEQs; van den Berg *et al*, 1998, 2000)

Compound [a]	TEF value		Compound	TEF value	
	I-TEF	WHO-TEQ		I-TEF	WHO-TEQ
PCDDs			**PCDFs**		
2,3,7,8-TCDD	1	1	2,3,7,8-TCDF	0.1	0.1
1,2,3,7,8-PeCDD	0.5	1	1,2,3,7,8-PeCDF	0.05	0.05
			2,3,4,7,8-PeCDF	0.5	0.5
1,2,3,4,7,8-HxCDD	0.1	0.1	1,2,3,4,7,8-HxCDF	0.1	0.1
1,2,3,6,7,8-HxCDD	0.1	0.1	1,2,3,7,8,9-HxCDF	0.1	0.1
1,2,3,7,8,9-HxCDD	0.1	0.1	1,2,3,6,7,8-HxCDF	0.1	0.1
			2,3,4,6,7,8-HxCDF	0.1	0.1
1,2,3,4,6,7,8-HpCDD	0.01	0.01	1,2,3,4,6,7,8-HpCDF	0.01	0.01
			1,2,3,4,7,8,9-HpCDF	0.01	0.01
OCDD	0.001	0.0001	OCDF	0.001	0.0001
Dioxins-like PCBs					
non-*ortho*			**mono-*ortho***		
3,3′,4,4′-TCB (PCB 77)	0.0005	0.0001	2,3,3′,4,4′-PeCB (PCB 105)	0.0001	0.0001
3,4,4′,5-TCB (PCB 81)	–	0.0001	2,3,4,4′,5-PeCB (PCB 114)	0.0005	0.0005
3,3′,4,4′,5-PeCB (PCB 126)	0.1	0.1	2,3′,4,4′,5-PeCB (PCB 118)	0.0001	0.0001
3,3′,4,4′,5,5′-HxCB (PCB 169)	0.01	0.01	2,3,4,4′5-PeCB (PCB 123)	0.0001	0.0001
			2,3,3′,4,4′,5-HxCB (PCB 156)	0.0005	0.0005
			2,3,3′,4,4′,5′-HxCB (PCB 157)	0.0005	0.0005
			2,3′,4,4′,5,5′-HxCB (PCB 167)	0.00001	0.00001
			2,3,3′,4,4′,5,5′-HpCB (PCB 189)	0.0001	0.0001

[a] T = Tetra, Pe = Penta, Hx = Hexa, He = Hepta

3.7 **Absorption.** In a human volunteer study, more than 87% of a single oral dose of 1.14 ng kg^{-1} bw (nanograms per kilogram body weight) of TCDD in corn oil was absorbed, with 90% of the body burden sequestered in fat (Poiger and Schlatter, 1986). A lower absorption would be expected from other matrices, and the Expert Groups

generally assume a 50% absorption of TCDD from the gastrointestinal tract of humans (van Leeuwen and Younes, 2000).

3.8 Rodent studies indicate that oral exposure to TCDD in the diet or in an oil carrier results in the absorption of between 50% and in excess of 90% of the administered dose (WHO, 1989). Absorption by the oral route appears to decrease as the size of the molecule increases and the solubility decreases. For example, the large and extremely insoluble OCDD is poorly absorbed, with values in the range 2–15% being reported. In some studies, absorption has been found to be dose-dependent, with greater proportional absorption occurring at lower doses.

3.9 Rodent feeding studies have shown that the absorption of TCDD is highly dependent upon the formulation in which it is administered. Absorption is greatest from oil vehicles (<80%) and lowest from aqueous solutions (<30%). In rodent studies, absorption of TCDD from contaminated soil is rather variable, but is lower (typically only about 50%) than from an oil carrier (ATSDR, 1998; IARC, 1997; Pollitt, 1999).

3.10 Although there are no human data from which to make a quantitative estimate of absorption following inhalation, the systemic effects observed in animals after pulmonary exposure provide evidence of efficient uptake via the lung. In a rat study in which a single intra-tracheal dose of TCDD was administered, 95% of the dose was absorbed (ATSDR, 1998).

3.11 No data are available from which to estimate dermal absorption for humans directly. Data on levels of dioxins in the blood of populations with above-background exposures (occupational and accidental settings) suggest that dermal absorption does occur in humans. Animal and *in vitro* studies (human cadaver skin) suggest that dermal absorption of these compounds is very slow. In one rat study, for example, absorption followed first-order kinetics with a rate constant of about 0.005 h^{-1} (at a concentration of 1 nmol kg^{-1}). Dermal absorption was found to be related to age in rats, with younger animals absorbing more than older ones (ATSDR, 1998; IARC, 1997).

3.12 **Distribution.** Once absorbed into the blood, dioxins are readily distributed to all organs. The predominant TCDD carriers in human plasma are serum lipids and lipoproteins. There do not appear to be any major species differences in the tissue distribution of these compounds in mammals (although the human data are very limited), with liver and adipose tissue becoming the primary storage sites within several hours of absorption. The distribution pattern is dose-dependent. At dose levels above 1 ng kg^{-1} bw day^{-1} (nanograms per kilogram body weight per day), TCDD is preferentially deposited in the liver; but at lower doses, steady-state levels in the liver and adipose tissue are similar on a lipid basis (ATSDR, 1998; Pollitt, 1999). Lipophilicity increases with chlorination (van Leeuwen and Younes, 2000).

3.13 **Metabolism.** Metabolites of dioxins have not been directly identified in humans. There is evidence that a wide range of mammalian species are capable of slow metabolism of TCDD by the microsomal monooxygenase system (cytochrome P450 1A1). The resultant polar metabolites can undergo conjugation with glucuronic acid and glutathione to form metabolites that are readily excreted (ATSDR, 1998; Hu and Bunce, 1999;

IARC, 1997; van Leeuwen and Younes, 2000). Metabolism is the rate-limiting step for elimination (van Leeuwen and Younes, 2000).

3.14 **Excretion.** Various estimates of the elimination half-life of TCDD in the body have been made using data derived from studies on exposed workers and others. The Expert Groups assume a value of 7.5 years (FSA, 2001a; EC, 2001) or 7.6 years (WHO, 2001). However, values ranging between 3 and 16 years have been estimated, depending on the congener. In the rat, the reported TCDD half-life is 20 days. The equivalent values in the mouse and guinea pig are 12 and 90 days respectively.

3.15 The major route of excretion in all species is through bile and faeces, with smaller quantities excreted in urine. Lactation also decreases the body burden of these compounds in mammals. In rats, lactation reduces the half-life of TCDD in the liver by 50% (ATSDR, 1998; IARC, 1997). In the UK in 1992, the mean level of dioxins in human milk was found to be 20 ng TEQ per kilogram of milk fat. Concentrations are highest during the first few weeks of feeding, and are 20–30% less for a second breast-fed child compared with the first. Although the pattern of chlorination is important, levels of dioxins in human milk generally reduce with decreasing degrees of chlorination (ATSDR, 1998).

3.16 **Dose metric[1].** In their discussion of the toxicokinetics of TCDD, the WHO (1998) considered what might be the most appropriate dose metric to use when equating effects across species. The ideal measure would be the concentration in the target tissue, but the use of this is generally impracticable. The body burden[2] is readily estimated in both people and rodents, and is highly correlated with tissue and serum concentration. It takes into account differences in toxicokinetics, including half-lives, between species.

3.17 For rodents and humans to achieve the same body burdens, much higher daily intakes are required by rodents (WHO, 1998). The WHO concluded that, in order to compare effects between animals and humans, body burden is the metric of choice. In 2001, the COT agreed that “despite some limitations, the body burden provides the appropriate dose metric, and that there is sufficient scientific evidence to support the use of body burden”. The Committee noted that, in humans exposed to dioxins over a period of 15–30 years during which time the environmental levels would have been decreasing, the body is not truly in steady state. In these circumstances it was estimated that the simple steady-state assumption would over-estimate daily intakes by about 20% when calculated on a lipid content basis (FSA, 2001a).

[1] A *dose metric* is a measure of dose that can be extrapolated across species, and can take several forms, including daily intake (ng kg^{-1} bw day^{-1}), body burden (ng kg^{-1} bw) and area under the curve (AUC) (USEPA, 2000).

[2] The *body burden* is defined as the concentration of TCDD and related chemicals in the body, and is typically expressed as ng kg^{-1} bw. In animals, from studies at or approaching steady state, these values are either calculated based on knowledge of the species-specific half-life and the exposure, or estimated based on the TCDD concentration, the size of the tissues and the weight of the animal. In humans, the values are typically presented as a steady-state body burden, and are estimated based on an intake rate and the half-life of TCDD in humans. Alternatively, body burden in humans is estimated from lipid-adjusted serum or adipose tissue TCDD or TEQ concentrations (USEPA, 2000).

3.18 **Mechanisms of toxicity.** In spite of the large number of papers on dioxins published in the scientific literature, the mechanism(s) of toxicity is(are) still not fully characterised. There is a consensus that many of the toxic effects of dioxins result from their binding to the Ah (aryl hydrocarbon) receptor. The Ah receptor is a member of a family of gene regulatory proteins. The precise chain of molecular events by which the ligand-activated receptor elicits the observed effects is not completely understood. It has been shown that the dioxins–protein complex binds to DNA and influences gene regulation, which in turn may lead to a variety of biochemical responses (van Leeuwen and Younes, 2000). In 2001, in accordance with the advice of the COC (FSA, 2001c), the COT "considered it appropriate to take a threshold approach to establishing a tolerable intake. This is based upon the negative genotoxicity in standard assays and evidence from studies of mechanisms" (FSA, 2001a).

3.19 It seems likely that the parent compound, rather than any metabolite, is the active species, and it is generally agreed that the toxicological consequences are the same irrespective of the route of uptake. There is little evidence of a "first-pass" effect in the liver or any other tissue. TCDD distributes and partitions throughout the body based on the lipid content of the tissue. The liver is an exception. In the liver, CYP1A2 acts as an inducible binding protein for TCDD and related compounds. Based on these principles, non-hepatic tissues with a high lipid content, such as fat and skin, will contain higher concentrations of TCDD and related compounds than tissues with low lipid content, such as blood. It should be noted that most data are presented as lipid-adjusted values. When tissue concentrations are expressed on a gram lipid basis, tissue concentrations are equivalent across tissues. Another key observation is that many of the effects of dioxins observed in experimental animal studies are the same whether or not the exposure is short or long term (ATSDR, 1998; van Leeuwen and Younes, 2000; WHO, 1989).

3.20 **Acute toxicity.** None of the studies that have examined humans acutely exposed to high concentrations of TCDD or other dioxin congeners reported death as an acute outcome. The LD_{50} for a single oral dose varies between other species and strains by a factor of more than 8000 (ATSDR, 1998; IARC, 1997; WHO, 1989), from the highly sensitive guinea pig (oral LD_{50} of 0.6 μg kg^{-1} bw) to the much more resilient hamster (oral LD_{50} of 5000 μg kg^{-1} bw). No single site of toxicity as the cause of death has been identified; each species has a different profile of organ toxicity, with a wasting syndrome and liver toxicity featuring most commonly (FSA, 2001a).

3.21 There have been many reports of human exposure to dioxins, and a number of epidemiological studies have been carried out on groups that have experienced accidental and relatively high exposures. The most consistent result of these high-level single exposures has been the occurrence of chloracne, a severe, acne-like, skin condition that develops within months of first exposure to high levels of TCDD. For many people the condition disappears after the cessation of exposure; for others it may remain for protracted periods. One of the shortcomings of all the epidemiological studies is a lack of exposure data (where exposure is taken to mean direct exposure through all environmental media). However, measurements of the concentrations of TCDD in lipids (serum and/or adipose tissue) have been reported in many of the studies and are considered to be a good indicator of uptake.

3.22 **Repeated toxicity.** There is evidence of toxicity in humans following repeated or long-term exposure to dioxins. Effects that have been reported include chloracne, elevated levels of liver enzymes and other disturbances in liver function, an increased death rate from non-malignant liver disease, a slightly greater risk of developing diabetes, changes in thyroid function, impaired immunological function, effects on the cardiovascular system (including an increased mortality from cardiovascular disease), mild neuropathies, influences on reproductive hormones and reproductive outcomes, and, in children, some neurobehavioural effects including neurodevelopmental delays (FSA, 2001a; van Leeuwen and Younes, 2000; WHO, 1989).

3.23 Chloracne has been reported in occupationally exposed groups. Positive associations between serum or adipose tissue levels of TCDD and increased risk of chloracne among individuals exposed at a young age amongst chemical production workers have been found in some studies. A threshold level below which chloracne does not occur has not been established (ATSDR, 1998; IARC, 1997; van Leeuwen and Younes, 2000; WHO, 1989).

3.24 Most of the other health effects listed in paragraph 3.22 are also the result of high exposure in the workplace or from accidental releases. Generally the body burdens of dioxins in the people affected are at least 10-fold higher than those occurring in the general population. The possible exception are the neurobehavioural effects reported in Dutch and US children (paragraph 3.30).

3.25 In all mammalian species tested, potent dioxin congeners have a number of characteristic toxic effects in common. Among these are progressive loss of body weight, reduced food intake, atrophy of the thymus, gastrointestinal haemorrhage and delayed lethality (several days or weeks). Other characteristic signs of toxicity are frequently found in the liver, skin and organs of the endocrine system (IARC, 1997).

3.26 There is agreement amongst Expert Groups in recent years that the toxicological findings in laboratory animals of most relevance to humans are effects on the immune system and on reproduction and development, including a possible hormonal action (endometriosis) (WHO, 1998). In addition, at even lower doses (producing body burdens in the tested animals in the range of 3–10 ng kg^{-1} bw), TCDD has been found to possess a biochemical or functional action (inducing liver enzyme activities, affecting EGF-receptor down-regulation and oxidative stress in rodents, and changing the proportion of lymphocyte sets in monkeys). These are currently classed as early expressions of events that may or may not result in adverse effects (van Leeuwen and Younes, 2000) and are not considered in the determination of TDIs.

3.27 **Immunotoxicity.** Several epidemiology studies and case reports have assessed immunological function in human populations exposed to TCDD, but they provide no overall confident insights of the dose–response (IARC, 1997). There is extensive evidence, from numerous studies in various animal species, that TCDD affects the immune system (IARC, 1997). Guinea pigs and mice appear to be among the most sensitive species; primates (and presumably humans) are among the least.

3.28 The main target for the immunotoxicity of TCDD is the thymus, where there is cellular depletion, with consequent reduced production of T lymphocytes and depression of cell-mediated immunity. Experimental animals exposed to TCDD during the perinatal period of development are more sensitive than are adults to its immunotoxic effects (the key studies are described in paragraph 3.32). Although damage to the thymus during adult life is less important, it must be assumed that such damage could have significant effects on immune function. It should also be noted that both cell-mediated and humoral immune responses are suppressed following TCDD uptake, which suggests that the thymus is not the only target within the immune system. There is also some evidence that the immune system is indirectly affected by TCDD-induced changes in non-lymphoid tissues (WHO, 1989, 2001).

3.29 **Reproductive and developmental toxicity.** Severe developmental effects have been reported in children born to mothers with high exposure to dioxin-like mixtures arising from the ingestion of rice oil contaminated with heated PCBs and PCDFs. This has occurred in two separate populations, in Taiwan and in Japan (the Yucheng and Yusho cohorts). The lowest intake of TEQs estimated to result in minimal Yusho symptoms was 28 ng kg^{-1} bw day^{-1} for 135 days, producing maternal body burdens of 2–3 μg TEQ kg^{-1} bw (Feeley and Brouwer, 2000).

3.30 A number of reports have suggested that environmental exposures to background levels of dioxins, dioxin-like and non-dioxin-like PCBs are also capable of producing subtle developmental delays (US and Dutch) and thyroid alterations (Dutch) in children exposed *in utero* (Feeley and Brouwer, 2000). The COT has concluded that “it was not possible to determine whether any reported differences were real effects on development, and if they were real, whether they were due to dioxins or some other cause” (FSA, 2001a).

3.31 In general, developmental toxicity is observed at lower TCDD exposure levels in experimental animals than are reproductive effects. The developmental actions range from the induction of stillbirth and abortion, through abnormal organ systems (including the reproductive organs), to changes in behavioural and cognitive functions. In considering prenatal or perinatal exposure to dioxins, the development of learning behaviour and the reproductive system appear to be the most sensitive end-points (IARC, 1997).

3.32 Perinatal exposure of rodents to TCDD has been shown to result in thymic atrophy and cell-mediated immune suppression at lower doses than are required to produce such effects following adult exposure. A suppressed immune response was seen in the offspring of rats treated by gavage on day 14 of pregnancy. The effects were present even at the lowest tested dose of 0.1 μg kg^{-1} bw (Gehrs and Smialowicz, 1998, 1999; Gehrs *et al*, 1997a,b). In rats given a single oral dose of 12.5, 50, 200 or 800 ng TCDD kg^{-1} bw on gestation day 15, there were dose-dependent biochemical changes in the thymus of the male offspring and some inconsistent effects on the numbers of splenocytes in the top dose group (Nohara *et al*, 2000). The SCF noted that “these studies demonstrate that the effects on the immune system of the male offspring of pregnant rats exposed to 2,3,7,8-TCDD occur only at higher doses than the effects seen on the reproductive organs and their function” (EC, 2001).

3.33 The effects of TCDD on the developing reproductive system of male rat foetuses (Faqi *et al*, 1998; Gray *et al*, 1995, 1997a,b; Mably *et al*, 1992a,b,c; Ohsako *et al*, 2001) were considered by the SCF, JECFA and the UK COT to be the most sensitive indicators of TCDD toxicity. The oral studies that reported reduced sperm levels in the offspring involved single gavage doses of 50 ng kg^{-1} bw to the mothers on day 15 of pregnancy (Gray *et al*, 1997b) or 64 ng kg^{-1} bw on day 16 (Mably *et al*, 1992b). In the study of Faqi *et al* (1998), the females were treated subcutaneously prior to mating and throughout mating, pregnancy and lactation. There were reduced sperm counts and an increased number of abnormal sperm in the male offspring at the lowest tested dose, an initial injection of 25 ng kg^{-1} bw, followed by weekly maintenance doses of 5 ng kg^{-1} bw. In the study of Ohsako *et al* (2001), the rats were treated by a single oral dose on day 15 of pregnancy. At 50 ng kg^{-1} bw, there was a decrease in anogenital distance in the male offspring (but no effects on sperm count). No adverse effects occurred at 12.5 ng kg^{-1} bw.

3.34 In an earlier three-generation reproduction study in rats, chronic dietary administration of 10 ng TCDD kg^{-1} bw day^{-1} produced significantly decreased fertility in the F_1 and F_2 generations, but not in the F_0 generation (Murray *et al*, 1979**)**. There were also decreases in litter size at birth, gestation survival (proportion of pups born alive), and neonatal survival and growth. No significant or consistent effects were reported at the lowest dose of 1 ng kg^{-1} bw day^{-1}.

3.35 Foetal malformations have also been observed in rodents following uptake of dioxins (Mukerjee, 1998). In mice, cleft palate and kidney anomalies occur at doses of TCDD below those required to induce foetotoxicity or maternal toxicity. For example, cleft palate and hydronephrosis have been observed in the offspring of pregnant mice treated with a subcutaneous dose of 3 μg TCDD kg^{-1} bw, a dose producing no overt signs of maternal toxicity. Selective developmental toxicity has not been demonstrated in other species; for example, cleft palate is induced only at foetotoxic and maternally toxic doses in rats (IARC, 1997).

3.36 Primates have been shown to be particularly sensitive species to the reproductive and developmental effects of TCDD (WHO, 1998, 2001). The types of reproductive and developmental toxicity that have been reported are reduced fertility in both males and females, decreased litter size or neurobehavioural effects in the offspring, decreased testosterone synthesis, a decreased production of oestrogen, and an increased incidence of endometriosis (IARC, 1997; van Leeuwen and Younes, 2000).

3.37 TCDD given to monkeys at dietary levels of 5 ng kg^{-1} food for around 3–4 years was claimed to induce endometriosis in the mothers 10 years after the end of the TCDD exposure (Rier *et al*, 1993), and to adversely affect the development of the offspring (Schantz and Bowman, 1989). These studies were amongst those used by the SCF in 2000 to determine a tolerable intake for TCDD. A number of reservations were expressed about the data on endometriosis; and in an assessment in 2001, additional studies from the same research team (Rier *et al*, 2001a,b) were said only to generate new questions. Because of “the uncertainties raised by the new findings, the Committee had less confidence in the quantitative relationship between exposure to 2,3,7,8-TCDD

and the incidence of endometriosis in monkeys …" and "decided not to include Rier *et al* (1993) as a pivotal study in the updated assessment" (EC, 2001).

3.38 **Conclusions.** The main non-cancer effects demonstrated in either human or experimental animal studies are chloracne, immune system suppression (primarily through thymic atrophy), and reproductive and developmental effects. The adverse effects reported at the lowest exposures are developmental and reproductive effects in rats and monkeys (Table 3.2) and it is these end-points that have been most influential in the derivation of the TDI.

Table 3.2 Studies favoured by the Expert Committees in the derivation of a TDI

Study	Response	Maternal body burden (ng kg^{-1} bw) at NOAEL [a]	Maternal body burden (ng kg^{-1} bw) at LOAEL [b]
RATS			
Mably *et al* (1992b,c)	Developmental: decreased sperm count in offspring		31 (WHO, 2001) 38 (EC, 2000) 100 (EC, 2001)
Gray *et al* (1997a)	Developmental: increased genital malformation in offspring		73 (WHO, 1998)
Gehrs *et al* (1997b), Gehrs and Smialowicz (1998)	Immunological: immune suppression in offspring		50 (WHO, 1998) 53 (WHO, 2001) 60 (EC, 2000)
Gray *et al* (1997b)	Developmental: decreased sperm count in offspring		28 (WHO, 1998) 31 (WHO, 2001) 30 (EC, 2000) 80 (EC, 2001)
Faqi *et al* (1998)	Developmental: decreased sperm production and altered sexual behaviour in offspring		33 (FSA, 2001a) 28–42 (WHO, 2001) 40 (EC, 2001)
Ohsako *et al* (2001)	Developmental: decreased prostate weight, and decreased anogenital distance in male offspring	16–22 (WHO, 2001) 20 (EC, 2001)	
MONKEYS			
Schantz and Bowman (1989)	Developmental: neuro-behavioural effects in offspring		42 (WHO, 1998) 25–37 (EC, 2000)
Rier *et al* (1993)	Reproductive: endometriosis		69 (WHO, 1998) 39 (EC, 2000).

[a] "No observed adverse effect" level.
[b] "Lowest observed adverse effect" level.

4 Carcinogenicity and Genotoxicity

4.1 An IARC Working Group in 1997 assigned TCDD to the highest IARC cancer classification (Group 1 – "carcinogenic to humans") based on "sufficient evidence" of its carcinogenicity in experimental animals, and some supporting data in humans (IARC, 1997). The paucity of carcinogenicity data on the other PCDDs and PCDFs meant a Group 3 ("not classifiable as to their carcinogenicity in humans") verdict. On the basis of a 2001 assessment, the COC concluded that TCDD should be regarded as a probable human carcinogen (FSA, 2001c). COC noted that TCDD was not genotoxic and that mechanistic data were consistent with a complex multi-step process involving receptor binding, and thus a threshold interpretation of TCDD-induced carcinogenicity. The Committee commented that "any increased risk of cancer at background levels of exposure is likely to be extremely small and not detectable by current epidemiological methods".

4.2 The most informative studies for the evaluation of the carcinogenicity of TCDD are those with the highest exposures, as these would be expected to produce the highest cancer risks. The IARC Working Group in 1997 abstracted from the published reports data on the most highly exposed groups, and focused on those with an adequate latent period (the time between exposure and the final date of the follow-up period) (Becher *et al*, 1996; Fingerhut *et al*, 1991; Hooiveld *et al*, 1996; Kogevinas *et al*, 1997; Ott and Zober, 1996). The blood TCDD levels at the time of exposure in a number of the workforces were in the same range as the estimated blood levels in the two-year rat study of Kociba *et al* (1978) described in paragraph 4.5 (van Leeuwen and Younes, 2000).

4.3 There was an overall increase in mortality from all cancers combined in all the studies. For the high-exposure sub-cohorts, the overall standardized mortality ratio (SMR)[3] for all cancers combined calculated by the Working Group was 1.4. The Working Group commented that, although the overall SMR is low, this finding is unlikely to be due to chance, is not the result of confounding by cigarette smoking, and is consistent across the high-exposure studies. The overall estimates for non-Hodgkin's lymphoma and soft-tissue sarcoma (SMRs of 2.6 and 4.7, respectively) were significantly elevated. There was a weak association for lung cancer (SMR of 1.4), not considered to be confounded by smoking. The association for gastrointestinal cancer was not statistically significant (SMR of 1.2, 95% confidence interval of 0.9–1.5) (IARC, 1997).

4.4 The IARC Working Group stated: "In summary, the epidemiological evidence from the most highly 2,3,7,8-TCDD-exposed populations studied produces strong evidence of increased risks for all cancers combined, along with less strong evidence of increased risks for cancers of particular sites. This situation appears to be unique, compared with established human carcinogens. The overall findings are unlikely to be due to chance. There is no obvious basis to infer that the findings are due to confounding with smoking, nor with occupational exposures to other chemicals, but such confounding cannot be

[3] SMR is a relative measure of the difference in risk between the exposed and unexposed populations in a cohort study. The SMR is similar to the relative risk in both definition and interpretation. The measure is usually standardized to control for any differences in age, sex and/or race between exposed and reference populations. The ratio is frequently converted to a percentage by multiplying it by 100 (USEPA, 2000).

ruled out. There is evidence in some studies of dose–response relationships, although dose–response data are not available for some of the largest studies. The relative risk estimates for all cancers, lung cancer and gastrointestinal cancer involve relatively low strengths of association. Higher relative risk estimates are present in some studies concerning non-Hodgkin lymphoma and soft-tissue sarcoma, but the total numbers of cancer are small, in particular for soft-tissue sarcoma" (IARC, 1997).

4.5 In a comprehensive two-year feeding study in male and female rats, the main finding was the development of liver tumours in the females (Kociba *et al*, 1978). An incidence of pre-cancerous liver pathology (hyperplastic liver nodules) of 8/86 in the untreated females was increased to 18/50 and 23/50 in the groups given 10 or 100 ng TCDD kg^{-1} bw day^{-1} respectively. The corresponding incidence of malignant liver tumours (hepatocellular carcinomas) was 1/86, 2/50 and 11/50 respectively. The top dose was therefore clearly carcinogenic in the liver of the females. For liver nodules, the LOAEL was 10 ng kg^{-1} bw day^{-1}, with the lowest dose of 1 ng kg^{-1} bw day^{-1} being the NOAEL, which corresponded to a body burden of 60 ng TCDD kg^{-1} bw (van Leeuwen and Younes, 2000).

4.6 Long-term studies conducted under the National Toxicology Program in the USA involved the administration of TCDD by gavage to rats and male mice at 0.01, 0.05 and 0.5 μg kg^{-1} bw $week^{-1}$, and to female mice at 0.04, 0.2 and 2 μg kg^{-1} bw $week^{-1}$ (NTP, 1982). In the high-dose rats, malignant liver tumours were seen in two females, and liver nodules developed in both sexes. Benign tumours of the thyroid were found in the male rats (1/69 controls; 5/48, 6/50 and 10/50 in the treated males). There was a treatment-related increase in malignant liver tumours in the high-dose male mice. In the females, there was an increase in the incidence of subcutaneous fibrosarcoma and benign thyroid tumours (follicular adenomas). A hexachlorodibenzo-*p*-dioxin mixture (1,2,3,7,8,9- and 1,2,3,6,7,8-) also produced malignant liver tumours in rats and mice treated twice weekly for 104 weeks by gavage (NTP, 1980). There was a dose-related increase in combined benign and malignant liver tumours in the male mice (from an incidence of 15/73 in the vehicle control, up to 24/48 of those receiving 5 μg kg^{-1} bw $week^{-1}$). In the female rats, the control incidence of 2/75 was increased to 18/50 at 5 μg kg^{-1} bw $week^{-1}$.

4.7 Skin painting studies in mice do not indicate that TCDD has any initiating (directly genotoxic) action. The promotion of the growth of a previously initiated lesion has been demonstrated, and the Ah receptor is thought to be involved. Extensive studies of liver tumour production in the female rat liver also point to a non-genotoxic route to tumour (van Leeuwen and Younes, 2000).

4.8 In their evaluations of the genotoxicity data on dioxins, the Expert Committees are essentially in agreement. The IARC Working Group in 1997 concluded that the "Experimental data indicate that 2,3,7,8-TCDD and probably other PCDDs and PCDFs are not direct-acting genotoxic agents*"* (IARC, 1997). In 1999, the COM considered that "the weight of the available experimental data continue to indicate that TCDD is not a genotoxic agent" (DH, 1999b). JECFA in 2001 noted that TCDD gave primarily negative results in the short-term assays for genotoxicity, and did not bind covalently to DNA from

the liver of mice. “The Committee concluded that TCDD is not an initiator of carcinogenesis” (WHO, 2001).

4.9 The conclusion of the COC in 2001 was that “although a precise mechanism for carcinogenesis in laboratory animals or humans could not be elucidated from the available information, the data (i.e. negative genotoxicity in standard assays, and evidence from studies of mechanisms) suggested that a threshold approach to risk assessment was likely to be appropriate” (FSA, 2001c).

5 Derivation of Tolerable Daily Intakes

The recommendations of the WHO consultations

5.1 In 1990, a Consultation of the WHO Regional Office for Europe carried out a toxicological evaluation of TCDD (WHO, 1991, 1992; Kello and Yrjänheikki, 1992). It was concluded that TCDD was carcinogenic in animals, probably a promoter carcinogen, and as such a TDI could be based on general toxicological effects. Studies of liver toxicity and reproductive and immunotoxicology in the various laboratory animal species identified a no-effect level of 100 pg kg^{-1} bw day^{-1}. Pharmacokinetic data indicated that this was equivalent to a dose of 100 pg kg^{-1} bw day^{-1} in humans. Because of the inadequate data based on reproductive effects in humans, an uncertainty factor of 10 was employed by the Consultation and therefore a TDI of 10 pg kg^{-1} bw was recommended.

5.2 In May 1998, a Consultation of the WHO European Centre for Environment and Health and the International Programme on Chemical Safety reviewed the evidence that had emerged since the 1990 overview and recommended a revised TDI value (WHO, 1998; van Leeuwen and Younes, 2000). The human data were again inadequate for establishing a TDI, and use had to be made of results of studies on experimental animals. The possibility of using toxicokinetic and dose–effect modelling in order to proceed with a "benchmark" approach was considered, but it was concluded that there were too many uncertainties involved, and therefore the traditional NOAEL/LOAEL approach was maintained.

5.3 The immunological, developmental and reproductive effects of TCDD were agreed as its critical (and most sensitive) toxicological end-points, in particular the rat studies of decreased sperm counts (Gray *et al*, 1997b; Mably *et al*, 1992c) and genital malformation (Gray *et al*, 1997a) and immune suppression (Gehrs *et al*, 1997b; Gehrs and Smialowicz, 1998) in the male offspring of treated females, and studies in female monkeys showing neurobehavioural effects in the offspring (Schantz and Bowman, 1989) and endometriosis in the adults (Rier *et al*, 1993). The WHO's preferred dose metric for the comparison of toxic effects across species was body burden rather than daily intake (paragraph 3.16). It was estimated that the LOAELs for the most sensitive adverse responses reported in the key studies were occurring at maternal body burdens in the experimental animals in the range of 28–73 ng kg^{-1} bw. The long-term human daily intakes that would have produced these same body burdens were estimated to be 14–37 pg kg^{-1} bw day^{-1} (assuming a fractional absorption from the gut of 50% for humans, and a half-life of 7.5 years). Because it was impossible to select a NOAEL from a single critical study, the WHO concluded that the range of estimated human intakes (i.e. 14–37 pg kg^{-1} bw day^{-1}) corresponding to the LOAELs from the group of key studies would provide a reasonable basis for the derivation of a TDI.

5.4 In deriving a TDI, various uncertainty factors had to be considered in order to allow for the use of LOAELs instead of NOAELs, as well as all other inter- and intra-species differences in toxicokinetics and toxicodynamics.

5.5 Since body burdens had been used to scale doses across species, the use of an uncertainty factor to account for species differences in toxicokinetics was not required. On toxicodynamics, it was noted that, for some end-points, humans might be as sensitive as experimental animals, whereas for other end-points they seem to be less sensitive. As the LOAELs seen in the key studies were considered to be within a factor of 2–3 of the NOAELs, and the differences in half-lives between TCDD and other dioxins, including dioxin-like PCBs, were also small (and partly accounted for in the establishment of the TEF values), the WHO was of the opinion that a composite uncertainty factor of 10 would be adequate to take account of the use of LOAELs (instead of NOAELs) and all other aspects related to inter- and intra-species variation.

5.6 Applying an uncertainty factor of 10 to the range of LOAELs of 14–37 pg TCDD kg^{-1} bw day^{-1} generated a TDI (rounded, and expressed as a range) of 1–4 pg WHO-TEQ kg^{-1} bw, which was said to be applicable to dioxins and dioxin-like compounds. The WHO emphasised that the upper limit of the range (4 pg WHO-TEQ kg^{-1} bw) should be considered a maximum tolerable daily intake on a provisional basis, and the ultimate goal should be to reduce intakes to levels less than 1 pg WHO-TEQ kg^{-1} bw day^{-1}.

The recommendations of the Scientific Committee on Food (SCF)

5.7 In November 2000, the SCF offered its view on the risk assessment of dioxins and PCB-like dioxins in food (EC, 2000). The animal studies that the Committee considered critical in the derivation of a tolerable intake were those of Mably *et al* (1992a,b,c), Gray *et al* (1997b) and Gehrs and Smialowicz (1998, 1999) in rats, and Rier *et al* (1993) in monkeys. The report of Schantz and Bowman (1989) of subtle non-persistent changes in the offspring of TCDD-treated female monkeys was described as being of “doubtful significance”.

5.8 It was estimated that LOAELs were occurring at body burdens in the experimental animals in the range of 25–60 ng kg^{-1} bw. Daily intakes in the range of 12.5–30 pg TCDD kg^{-1} bw were estimated to produce a body burden of this magnitude in humans (based on a gastrointestinal tract absorption in humans of 50% and a half-life in the body of 7.5 years). Applying a 10-fold uncertainty factor to this estimated human daily intake (EHDI) generated a tolerable intake in the range of 1–3 pg TCDD kg^{-1} bw day^{-1}. Because of the acknowledged uncertainties, the Committee concluded that the lower end of the range, 1 pg kg^{-1} bw day^{-1}, should be considered as a “temporary” tolerable intake. The Committee concluded that this safety limit for TCDD could be extended to include all 2,3,7,8-substituted PCDDs and PCDFs, and the dioxin-like PCBs, and established a group temporary tolerable weekly intake (t-TWI) of 7 pg WHO-TEQ kg^{-1} bw. A weekly rather than a daily limit was thought to be more appropriate owing to the long half-lives in humans of the compounds in question (EC, 2000).

5.9 New information that might have clarified some of the uncertainties in the 2000 opinion encouraged the SCF to review dioxins again in 2001 (EC, 2001). Additional data on endometriosis in the monkey (Rier *et al*, 2001a,b) only added to the difficulties of interpretation. It was decided therefore to base the updated opinion on the TWI solely on the toxicity studies of TCDD in the rat. There were two new reports of effects on the male offspring of treated females: Faqi *et al* (1998) identified effects at the lowest tested

dose (i.e. a LOAEL), and Ohsako *et al* (2001) generated a NOAEL. These were combined with Gray *et al* (1997b) and Mably *et al* (1992c) to derive a revised TWI. The reported LOAELs (Faqi *et al*, 1998; Gray *et al*, 1997b; Mably *et al*, 1992c) were seen at steady-state body burdens between 40 and 100 ng TCDD kg^{-1} bw and the EHDI lay in the range of 20–50 pg TCDD kg^{-1} bw. The NOAEL observed in the study of Ohsako *et al* (2001) occurred at an estimated maternal steady-state body burden of 20 ng kg^{-1} bw in the rats, and the equivalent EHDI was 10 pg TCDD kg^{-1} bw.

5.10 Uncertainty factors were applied to the NOAEL and the LOAELs to produce estimates of the tolerable intake. For the NOAEL, this needed to account for differences between humans and rats in susceptibility to TCDD (a product of toxicokinetics and toxicodynamics) and the potential inter-individual variation in susceptibility (toxicokinetics and toxicodynamics) to TCDD within the human population. The use of an uncertainty factor to account for differences between experimental animals and humans in toxicokinetics was not required since the default toxicokinetic factor was replaced by actual data in calculating the body burdens used to scale doses across species. On differences in toxicodynamics between rats and humans and within the human population, it was decided that the uncertainty factor was 1. Although rats in general are known to be more susceptible than are humans to toxicity based on Ah receptor binding, the Committee could not exclude the possibility that for some end-points the most sensitive human might be as sensitive as the experimental rats. A default uncertainty factor of 3.2 was considered acceptable to account for inter-individual variations in toxicokinetics in humans.

5.11 Applying the overall uncertainty factor of 3.2 to the EHDI of 10 pg kg^{-1} bw derived from the NOAEL produced a TDI of 3 pg kg^{-1} bw day^{-1}. Basing the TDI estimates on the LOAELs (or the equivalent EHDIs of 20–50 pg kg^{-1} bw) required an additional uncertainty factor. As the LOAELs reported for the sensitive end-points were considered to be close to the NOAELs and were representing marginal effects, a factor of 3 was judged to be sufficient. This produced an overall uncertainty factor of 9.6 (3 × 3.2) and tolerable intakes of 2 pg kg^{-1} bw day^{-1} from Faqi *et al*, 4 pg kg^{-1} bw day^{-1} from Gray *et al*, and 5 pg kg^{-1} bw day^{-1} from Mably *et al*. The lowest of these three figures was chosen on the premise that Faqi *et al* were using the most sensitive rat strain. Because of the long half-lives of TCDD and related compounds in the human body, the Committee considered that a weekly rather than a daily limit was appropriate. The tolerable weekly intake (TWI) of 14 pg TCDD kg^{-1} bw was extended to include all 2,3,7,8-substituted PCDDs and PCDFs, and the dioxin-like PCBs, expressed as a WHO-TEQ value. Because the new studies provided a firmer basis for the evaluation of the pivotal rat studies, the previous temporary designation for the Group TWI was removed.

The recommendations of JECFA

5.12 PCDDs, PCDFs and coplanar PCBs were evaluated at a June 2001 meeting of JECFA (WHO, 2001). The rat studies considered to be critical in the derivation of the TDI were those of Faqi *et al* (1998), Gray *et al* (1997b), Mably *et al* (1992c) and Ohsako *et al* (2001) reporting an impaired development of the male offspring of female rats treated with TCDD, and those of Gehrs *et al* (1997b) and Gehrs and Smialowicz (1998) on immunological deficits in the offspring.

5.13 Toxicokinetic data were applied to the lowest LOAEL reported by Faqi *et al* and the NOAEL of Ohsako *et al* to estimate maternal body burden LOAELs and NOAELs for effects on the male offspring of 28–42 and 16–22 ng kg^{-1} bw respectively. These values took some account of TCDD exposure from background contamination considered to be present in feed provided to laboratory animals. The body burdens were estimated to be equivalent to human monthly intakes (EHMI) of 423–630 and 237–330 pg kg^{-1} bw respectively (assuming a 50% absorption from food and a half-life of 7.6 years).

5.14 A default safety factor of 3.2 was thought to be suitable to account for inter-individual differences in toxicokinetics in humans (because of the limited data on TCDD itself), and an additional factor of 3 was proposed for the use of a LOAEL for a mild toxic effect (instead of a NOAEL) from the study of Faqi *et al* (1998). This second factor was not needed for the Ohsako *et al* (2001) study, as a NOAEL was identified. The use of body burdens to scale doses from animals to humans removed the need for a safety factor for inter-species differences in toxicokinetics. In addition, as it was assumed that the most sensitive human might be of a similar sensitivity to the rats in the critical studies, it was judged there was no need of an additional safety factor to take account of either intra-species or inter-individual differences in toxicodynamics. Application of the total safety factor of 3.2 or 9.6 to the appropriate EHMIs generated a range of provisional tolerable monthly intakes (PTMI) of between 40 and 100 pg kg^{-1} bw (after rounding), and the mid-point of this range, 70 pg kg^{-1} bw, was chosen as the recommended PTMI. On the basis of the 1998 WHO consultation, the Committee concluded that this tolerable intake should be applied to intakes of PCDDs, PCDFs and the coplanar PCBs expressed as TEFs.

5.15 The Committee emphasised that "substantial uncertainties remained which should be considered in applying the risk assessment and interpreting the estimates of intake of PCDDs, PCDFs, and coplanar PCBs … The safety assessment includes adjustment for a number of uncertainties, including estimates of TEF values within orders of magnitude to relate the potency of 28 relatively poorly studied compounds to that of one well-studied compound, 2,3,7,8-TCDD" (WHO, 2001).

The recommendations of COT

5.16 The 1991 recommendation of the first WHO Consultation (a TDI of 10 pg kg^{-1} bw) was endorsed by the COT (DH, 1996; MAFF, 1992). The COT recommended that, when considering mixtures of PCDDs and PCDFs, the TDI be regarded as 10 pg I-TEF kg^{-1} bw. This was revised in 1997 to include the dioxin-like PCBs, and in 1998 the WHO-TEQs for dioxins and dioxin-like PCBs were endorsed (DH, 1999a).

5.17 In 2001 the COT considered the dioxin risk assessments of all of the other Expert Groups and, because it could not determine which, if any, was the most scientifically justified, decided to conduct its own review of the available data (FSA, 2001a). The Committee concluded that the available human data did not provide a sufficiently rigorous basis for establishment of a tolerable intake (as the epidemiological studies did not reflect the most sensitive population identified by animal studies, there were

uncertainties in the exposure assessments and inadequate allowance for confounding factors, and the exposures did not reflect exposures experienced in the general UK population).

5.18 Effects on sperm production and morphology in the offspring of dosed animals represented the most sensitive indicator of TCDD toxicity. A detailed consideration of the results of the studies in rats of Faqi *et al* (1998), Gray *et al* (1995, 1997b), Mably *et al* (1992c) and Ohsako *et al* (2001) provided, therefore, the basis of the recommendation on TDI. The critical data point in the derivation of the TDI was generated by the study of Faqi *et al*. According to the COT estimates, the reduced sperm count in the male offspring was occurring at a TCDD body burden in the mothers on day 16 of pregnancy of 33 ng kg^{-1} bw.

5.19 The conversion of the steady-state maternal body burden in the female rat to the corresponding measure in humans required the use of uncertainty factors to allow for the use of a LOAEL and to take account of species differences and inter-individual variations. The values chosen for these uncertainty factors and their explanation were practically identical to those of the SCF and JECFA. As there may be inter-individual variations in the way different humans accumulate dioxin-like compounds, a factor of 3.2 was considered appropriate to take account of the potential increased body burden of these susceptible individuals. A factor of 3 was used to take account of the fact that the key rat study only identified a LOAEL rather than an NOAEL. The use of maternal body burdens as the dose metric and the presumption that the most sensitive human may have the same sensitivity as the tested rats meant that the remaining uncertainty factors were all 1 (to account for inter-species differences in pharmacokinetics, inter-species differences in pharmacodynamics and inter-human variations in pharmacodynamics).

5.20 An overall uncertainty factor of 9.6 was therefore applied to the LOAEL in rats to produce a tolerable maternal body burden in humans of 3.4 ng kg^{-1} bw. Based on an assumption of a TCDD bioavailability in humans of 0.5 and a half-life of 7.5 years, it was estimated that a daily intake of 1.7 pg kg^{-1} bw day^{-1} would lead to this body burden. Rounding this to a single figure (given the imprecision and assumptions in these calculations) generated the recommended TDI of 2 pg kg^{-1} bw day^{-1}. The COT emphasised that, because of the long half-life of TCDD, the body burden at steady state was about 2000-fold higher than the average daily intake. This meant that short-term variation in intake would not significantly alter the body burden, and therefore "occasional exceedance of the TDI would not be expected to result in harmful effects, provided that intake averaged over a prolonged period is within the TDI".

The recommendations of the USEPA

5.21 The risk assessment of TCDD has been on the agenda of the USEPA for a number of years. According to a Federal Register item of January 2002, TCDD is included in a list of compounds on which "assessments are underway or generally complete". Entry onto the Integrated Risk Information System (IRIS) databank is planned for 2002 or 2003 (USEPA, 2002).

5.22 A USEPA draft report circulated in 2000 noted that the derivation of RfD/RfC values would be uninformative for safety assessment purposes. [An RfD, an oral reference dose, or an RfC, an inhalation reference concentration, are estimates with uncertainty spanning perhaps an order of magnitude of a daily exposure to the human population that is likely to be without appreciable risk of deleterious non-cancer effects during a lifetime]. In the view of the USEPA, the average exposure of the US population to dioxins of around 1 pg TEQ kg^{-1} bw day^{-1} was clearly in excess of any RfD and RfC likely to be generated by the current database on the toxicology of dioxin and related compounds. Their ability to produce developmental, reproductive, immunological and endocrinological effects in multiple animal species was the key toxicology.

5.23 The use of steady-state body burdens was said to provide a reasonable description of dose for the purposes of species extrapolation and risk assessment. Margins-of-exposure, the ratio of the human body burden to the effect level in the comparison species, were calculated “to inform risk management decisions”. The margin was less than 1 for enzyme induction, and less than 3 for developmental effects.

5.24 As regards cancer, the epidemiological literature was described as being generally consistent with what was seen in laboratory animals, where dioxin-like compounds are both multi-site carcinogens and tumour promotors. The information available on dose-responses did not provide “consistent or compelling” support for a dose threshold and therefore the default of linear extrapolation of risk down to zero exposure was adopted. Based on both the epidemiological and laboratory animal data (and again using body burden as the measure of dose), the upper bound life-time cancer risk was estimated to be about 1×10^{-3} per pg TCDD kg^{-1} bw day^{-1}.

The recommendations of the ATSDR

5.25 The ATSDR has recommended a chronic minimal risk level (MRL) for oral exposure, based on a 16-month dietary study in which female rhesus monkeys were given TCDD and their offspring observed for behavioural and cognitive anomalies (Schantz *et al*, 1992). A LOAEL for altered social behaviour of 0.12 ng TCDD kg^{-1} bw day^{-1} was observed. The ATSDR used an uncertainty factor of 90 (3 for the use of a minimal LOAEL, 3 for inter-species variations, and 10 for intra-species variations) to arrive at an MRL of 1 pg kg^{-1} bw day^{-1} (applicable to TCDD or total TEQ). It should be noted that the ATSDR followed the traditional approach of using intake (rather than body burden) as the metric for the transfer of toxic effects across species (ATSDR, 1998).

Conclusions

5.26 The various Expert Committees that have reviewed risk assessments of dioxins (from the May 1998 WHO Consultation through to the 2001 verdicts of the COT, SCF and JECFA), indicate a high degree of agreement on the major issues. There is consensus that the most suitable dose measure is the body burden, the dose–response of the critical toxicology is likely to exhibit a threshold and the identification of NOAEL/LOAELs and the use of uncertainty factors is the preferred method to derive a TDI. These committees also agree that the resulting TDI, although exclusively based on TCDD data,

is applicable to other polychlorinated dibenzo-*p*-dioxins and dibenzofurans and the coplanar PCBs using TEF methods.

5.27 There is also good agreement on what are the key studies, the LOAELs or NOAELs they generate, and the uncertainty factors that are needed to determine the TDI. Subsequent Expert Committee assessments consider that the low-dose effects of TCDD on the male offspring of treated female rats are critical in the derivation of a TDI. The WHO Consultation considered that the reproductive and developmental toxicity studies in monkeys were also critical. A general lack of confidence in the validity of the monkey data has meant that subsequent Expert Committee assessments are based solely on the rat studies. Nevertheless the estimated body burdens producing the effects in monkeys were of the same order as those that were affecting the rat offspring, and so the exclusion of the monkey data had no impact on the derived TDI values.

5.28 There are some differences in the Expert Committee estimates of the rat body burdens associated with the reported LOAELs or NOAEL. The differences, though, are of no major importance in that they are lost in the rounding processes that are an inherent part of the derivation of a TDI or its weekly or monthly equivalent. The COT, JECFA and later SCF conversions of dose to body burden benefited by the availability of data from a new detailed study of the distribution of orally administered TCDD in the pregnant rat (Hurst *et al*, 2000a,b). The recommended tolerable intakes of the COT, SCF and JECFA of 2 pg kg^{-1} bw day^{-1}, 14 pg kg^{-1} bw $week^{-1}$ and 70 pg kg^{-1} bw $month^{-1}$, respectively, are essentially the same. The daily limit of the COT of 2 pg WHO-TEQ kg^{-1} bw applicable to the dioxins and dioxin-like PCBs is recommended here.

5.29 It is recognised that TCDD is carcinogenic in animals and that this is supported by some evidence of carcinogenicity in those occupationally exposed. TCDD should be regarded as a probable human carcinogen. It is not genotoxic and the mechanistic data are consistent with a complex multi-step process involving receptor binding and thus a threshold interpretation of TCDD-induced carcinogenicity. The TDI is based on the most sensitive toxicity end-point (reproductive toxicity) and would be expected to protect also against any carcinogenic effect of dioxins.

5.30 No Expert Group has derived an inhalation or dermal TDI for TCDD. The WHO did not recommend an air quality guideline on dioxins “because direct inhalation exposures constitute only a small proportion of the total [human] exposure” (WHO, 2000). There is no evidence to suggest that the toxicity of dioxins is route-specific, and therefore any contribution to body burden from all routes of exposure (including inhalation and dermal as well as oral) should be compared with the oral tolerable daily soil intake (see Section 8 below).

6 Intake of Dioxins, Furans and Dioxin-like PCBs from Food, Water and Air

6.1 Dioxins and dioxin-like PCBs have very low solubility in water, and are lipophilic, persistent and likely to bioaccumulate in the environment. They are therefore more likely to accumulate in fatty foods such as milk and milk products, and in certain meats and fish, than in other foodstuffs (FSA, 2000). In the UK, cereals, fats and oils are major components of our diet and therefore these foods contribute a significant proportion of our total dioxin and PCB intake (FSA, 2000). Although less likely to be present in vegetables, dioxins and dioxin-like PCBs may be present in soil adhered to vegetables if not thoroughly washed or peeled (ATSDR, 1998). Direct uptake from the soil via the roots is thought to be low (ATSDR, 1998; WHO, 1989).

6.2 The Food Standards Agency (FSA) and Defra – and its predecessor body, the Ministry of Agriculture, Fisheries and Food (MAFF) – have reported a number of surveys of dioxins and dioxin-like PCBs in food in the UK. The most recent reports on dietary exposure to dioxins and dioxin-like PCBs have been largely based on food surveys from 1982, 1992, 1995/1996 and 1997 (FSA, 2000; MAFF, 1997, 1999). In these surveys, chemical concentrations below the detection limit have been assumed to be at the detection limit for the purpose of estimating an upper-bound dietary intake (FSA, 2000; MAFF, 1997, 1999).

6.3 The most recent dietary data for dioxins and dioxin-like PCBs in the UK population are presented by FSA for 1997 (FSA, 2000), as summarised in Table 6.1. This table presents intakes for both the average and high-level (97.5th percentile) consumers. There has been a general decline in dioxin and dioxin-like concentrations in food over the past two decades, this decline being more significant for dioxins (FSA, 2000; MAFF, 1997, 1999). For both adults and toddlers, dioxin and dioxin-like PCBs contribute equally to the overall TEQ intake (i.e. 50% of the total dietary intake is due to dioxin and 50% is due to dioxin-like PCBs). It should be noted that the MDIs reported in Table 6.1 have been calculated using the TEFs recently recommended by the WHO (van den Berg *et al*, 1998, 2000).

Table 6.1 Dietary intakes of dioxins and dioxin-like PCBs for adults and 1.5–4.5 year olds in 1997

Age group	Dietary intake (pg WHO-TEQ day^{-1})[a,b,c]	
	Average consumers	High-level consumers
1.5–2.5 year old (12 kg)	55 (4.6)	86 (7.2)
3.5–4.5 year old (17 kg)	68 (4.0)	121 (7.1[d])
Adult (60 kg)	108 (1.8)	186 (3.1)

[a] The figures in parentheses are per unit bw, i.e. pg WHO-TEQ kg^{-1} bw day^{-1}.
[b] Dietary intake for toddlers estimated using food consumption data.
[c] Data presented in this table do not include intake via breast-feeding.
[d] Dietary intake is the average of values reported for boys and girls (6.9 and 7.2 pg WHO-TEQ kg^{-1} bw day^{-1}, respectively).

6.4 In the Total Dietary Survey (TDS), dietary intakes were calculated on the basis of an adult body weight of 60 kg (FSA, 2000). However, using updated Department of Health data, a mean body weight of approximately 70 kg is assumed in CLEA (Defra and Environment Agency, 2002a). Advice from the FSA suggests that the MDI should be scaled accordingly. This means that the adult average dietary intake of 108 pg day^{-1} (in Table 6.1) would increase to an intake of 126 pg day^{-1} (for a 70 kg person).

6.5 Individual PCB congeners in food were not reported by FSA in their 1997 report, but were reported by MAFF in the 1992 survey (MAFF, 1997; FSA, 2000). PCB congener patterns varied between individual food products, and there was no dominant PCB congener across all food types (MAFF, 1997). PCBs 77, 105 and 118, and 118 (see Table 3.1) were the greatest dioxin-like contributors to the overall TEQ in fish, cereal products and milk, respectively. These food groups (i.e. fish, cereals and milk) make up about 10%, 17% and 22% of total dietary exposure to total PCBs, respectively.

6.6 Few measurements have been made of dioxins and dioxin-like PCBs in the UK aquatic environment or elsewhere because of the difficulties of measuring these compounds in water and the low concentrations at which they are found. Rose *et al* (1994), for example, measured dioxin and furan concentrations of less than 6 ng L^{-1} in 40 UK surface fresh waters (concentrations here were expressed as the sum of total tetra- through octa-chlorinated dioxins and furans and not as TEQ values). TCDD levels in water have also been reported for various countries around the world, ranging from less than 0.2 pg L^{-1} for fresh water (River Elbe, Germany) to 167 pg L^{-1} for a drinking-water sample in the Russian Federation (IARC, 1997). Based on these reported levels, and in view of the low solubility of these compounds in water, it seems unlikely that human intakes from drinking water are significant when compared with those from food.

6.7 In the UK, the Toxic Organic Micropollutants survey is the principal source of data on the measured concentrations of dioxins and PCBs in ambient air at four urban sites and a semi-rural location. In 1996/97, the annual average concentration of dioxins in urban areas in London was less than 0.1 pg TEQ m^{-3} (DETR, 1998). Although a suite of individual PCB concentrations were also reported, only PCB 118 was measured at two of the sites (London and Stevenage) and PCBs 105 and 118 at the other three sites (Cardiff, Manchester and Hazelrigg). Mean concentrations in some urban areas for the first two quarters of 1999 are less than about 0.03 pg WHO-TEQ m^{-3} (AEAT, 1999). Currado and Harrad (1997) have also measured the air concentrations of individual PCB congeners in the ambient air in Birmingham, although not all the congeners detected were reported. Using these data (i.e. reported concentrations for PCBs 118, 105 and 156), the estimated air concentration of dioxin-like PCBs was 0.0024 pg WHO-TEQ m^{-3}. The mean combined air concentration of dioxins and dioxin-like PCBs in urban areas in the UK is therefore no more than about 0.1 pg TEQ m^{-3}.

6.8 An air concentration of 0.1 pg TEQ m^{-3} would give rise to daily intakes via inhalation of about 2 pg WHO-TEQ day^{-1} for adults (assuming respiration rates of 20 m^3 day^{-1}; Defra and Environment Agency, 2002a). These, again, are much lower than the estimated dietary intake from food.

6.9 In summary, the mean daily intakes of dioxins and dioxin-like PCBs from food, water and air combined are likely to be virtually the same as those for food. This is in agreement with WHO (1998), who concluded that over 90% of human background exposure to PCDDs, PCDFs and dioxin-like PCBs is estimated to occur through the diet, with food from animal origin being the predominant source. Thus, for a 70 kg adult, the mean daily intake is about 126 pg WHO-TEQ day^{-1}.

7 Other Sources

7.1 MAFF Food Surveillance Sheet 105 (MAFF, 1997) provides estimates of dietary intakes of dioxins and PCBs by breast-fed infants in 1993–94. MAFF calculated these intakes to be 170 pg TEQ kg^{-1} bw day^{-1} at two months, dropping to 39 pg TEQ kg^{-1} bw day^{-1} at 10 months. Despite the high intakes of dioxins experienced by nursing infants (about 100-fold those of an adult), the impact of breast feeding on infant body burden of dioxin is markedly less dramatic. Peak infant body burdens are only around twice those of an adult, a consequence of the infant's rapidly expanding body weight and lipid volume, as well as a possibly faster elimination rate (USEPA, 2000). When considering infant daily intakes, the FSA confirmed its previous advice that, although dioxin intakes are higher than desirable for breast-fed babies, encouragement of breast-feeding should continue on the basis of overwhelming evidence of the benefit of human milk to the overall health and development of the baby (FSA, 2001d).

8 Conclusions

8.1 The oral tolerable daily intake (TDI) and the oral mean daily intake (MDI) of dioxins and dioxin-like PCBs are given in Table 8.1.

Table 8.1 TDI_{oral}, oral MDI and TDSI for an adult and six-year-old child (expressed as WHO-TEQs)

TDI_{oral} (pg kg^{-1} bw day^{-1})	Oral MDI (pg day^{-1})	TDSI for an adult (pg kg^{-1} bw day^{-1})	TDSI for a six-year-old child (pg kg^{-1} bw day^{-1})
2	126	0.4	0.4

8.2 An oral tolerable daily soil intake (TDSI) is defined as the difference between the TDI_{oral} and the oral MDI (TDI – MDI), unless the MDI is close to, or exceeds, the TDI, in which case the TDSI is set at 20% of the TDI. Close to is defined as greater than or equal to 80% of the TDI (Defra and Environment Agency, 2002a).

8.3 For dioxins, the oral MDI for a 70 kg adult is equivalent to 1.8 pg WHO-TEQ kg^{-1} bw day^{-1}; as this value is close to the TDI, the TDSI would correspond to 0.4 pg WHO-TEQ kg^{-1} bw day^{-1}. Similarly, for a 20 kg child (aged six) who ingests 62% of the adult dietary intake, the TDSI would also be 0.4 pg WHO-TEQ kg^{-1} bw day^{-1} (Table 8.1).

8.4 No authoritative assessments of the health risks posed by inhalation or dermal exposures to dioxins or dioxin-like PCBs were identified.

8.5 These conclusions are for dioxins, furans and dioxin-like PCBs only (see Table 3.1 for further detail). A review is currently under way to derive health criteria for other toxicologically relevant PCBs, and Soil Guideline Values will be derived for these in the future.

References

AEAT (1999) *The UK National Air Quality Information Archive*. National Environmental Technology Centre – AEA Technology Environment.
Available on-line at http://www.aeat.co.uk/netcen/airqual/welcome.html
The site is now located at www.airquality.co.uk

ATSDR (1998) *Toxicological Profile for Chlorinated Dibenzo-p-dioxins*, Agency for Toxic Substances and Disease Registry, US Department of Health and Human Services, Atlanta.

Becher H, Flesch-Janys D, Kauppinen T, Kogevinas M, Steindorf K, Manz A and Wahrendorf J (1996) Cancer mortality in German male workers exposed to phenoxy herbicides and dioxins. *Cancer Causes and Control* **7** 312–321.

Currado GM and Harrad S (1997) The significance of indoor air inhalation as a pathway of human exposure to PCBs. *Organohalogen Compounds* **33** 377–381.

Defra and Environment Agency (2002a) *Contaminants in Soil: Collation of Toxicological Data and Intake Values for Humans. Consolidated Main Report. CLR9.* Available from the R&D Dissemination Centre, WRc plc, Swindon, Wilts.

Defra and Environment Agency (2002b) *The Contaminated Land Exposure Assessment (CLEA) Model: Technical Basis and Algorithms, Report CLR10*. Available from the R&D Dissemination Centre, WRc plc, Swindon, Wilts.

Defra and Environment Agency (in preparation). *Soil Guideline Values for Dioxin, Furan and Dioxin-like PCB Contamination,* R&D Publication SGV 11. Available from the R&D Dissemination Centre, WRc plc, Swindon, Wilts.

DETR (1998) *Digest of Environmental Statistics*, No 20, The Stationery Office, London.

DH (1994) *1993 Annual Report of the Committees on Toxicity, Mutagenicity and Carcinogenicity of Chemicals in Food, Consumer Products and the Environment*, The Stationery Office, London.

DH (1996) *1995 Annual Report of the Committees on Toxicity, Mutagenicity and Carcinogenicity of Chemicals in Food, Consumer Products and the Environment*, The Stationery Office, London.

DH (1997) *Statement by the Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment on the Health Hazards of Polychlorinated Biphenyls*.
Available on-line at http://www.doh.gov.uk/hef/pcb.htm

DH (1999a) *Statement on Surveillance for PCDDs, PCDFs and PCBs in Marine Fish and Fish Products*, COT/1999/02, Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment.
Available on-line at http://www.doh.gov.uk/COT/pcddfish.htm

EC (2000) *Opinion of the SCF on the Risk Assessment of Dioxins and Dioxin-like PCBs in Food – Adopted on 22 November 2000*, SCF/CS/CNTM/DIOXIN/8 Final, European Commission, Scientific Committee on Food, Brussels.
Available on-line at http://europa.eu.int/comm./food/fs/sc/scf

EC (2001) *Opinion of the Scientific Committee on Food on the Risk Assessment of Dioxins and Dioxin-like PCBs in Food. Updated opinion based on new scientific information available since the adoption of the SCF opinion of 22 November 2000. Adopted on 30 May 2001*, CS/CNTM/DIOXIN/20 Final, European Commission, Scientific Committee on Food, Brussels.
Available on-line at http://europa.eu.int/comm/food/fs/sc/scf

DH (1999b) *COM Statement on TCDD (2,3,7,8-tetrachlorodibenzo-p-dioxin)*, COM/99/S2, Committee on Mutagenicity of Chemicals in Food, Consumer Products and the Environment.
Available on-line at http://www.doh.gov.uk/com/tcdd.htm

DoE (1989) *Dioxins in the Environment*, Pollution Paper No 27, The Stationery Office, London.

Dyke PH, Foan C, Wenborn M and Coleman PJ (1997) A review of dioxin releases to land and water in the UK. *Science of the Total Environment* **207** 119–131.

Faqi AS, Dalsenter PR, Merker HJ and Chahoud I (1998) Reproductive toxicity and tissue concentrations of low doses of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin in male offspring rats exposed throughout pregnancy and lactation. *Toxicology and Applied Pharmacology* **150**(2) 383–392.

Feeley M and Brouwer A (2000) Health risks to infants from exposure to PCBs, PCDDs and PCDFs. *Food Additives and Contaminants* **17** 325–333.

Fingerhut MA, Halperin WE, Marlow DA, Piacitelli LA, Honchar PA, Sweeney MH, Greife AL, Dill PA, Steenland K and Suruda AJ (1991) Cancer mortality in workers exposed to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. *New England Journal of Medicine* **324** 212–218.

FSA (2000) *Dioxins and PCBs in the UK Diet: 1997 Total Diet Study Samples*, Food Standards Agency, Food Surveillance Information Sheet No 00/04, September 2000, Food Standards Agency, London.

FSA (2001a) *Statement on the Tolerable Daily Intake for Dioxins and Dioxin-like Polychlorinated Biphenyls*, Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment, Food Standards Agency, London.
Available on-line at http://www.foodstandards.gov.uk

FSA (2001b) *Lay Summary of the COT Statement on the Tolerable Daily Intake for Dioxins and Dioxin-like Polychlorinated Biphenyls*, Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment, Food Standards Agency, London.
Available on-line at http://www.foodstandards.gov.uk

FSA (2001c) *Carcinogenicity of 2,3,7,8-Tetrachlorodibenzo-p-dioxin*, Committee on Carcinogenicity of Chemicals in Food, Consumer Products and the Environment, Food Standards Agency, London.
Available on-line at http://www.doh.gov.uk/tetrachl.htm

FSA (2001d) *PCBs and Dioxins*, Statement, Food Standards Agency, London, 16 November 2001.

Gehrs BC and Smialowicz RJ (1998) Persistent suppression of delayed-type hypersensitivity in F344 rats perinatally exposed to TCDD. *Toxicologist* **42** 1501.

Gehrs BC and Smialowicz RJ (1999) Persistent suppression of delayed-type hypersensitivity in adult F344 rats after perinatal exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. *Toxicology* **134** 79–88.

Gehrs BC, Riddle MM, Williams WC and Smialowicz RJ (1997a) Alterations in the developing immune system of the F344 rat after perinatal exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin: I. Effects on the foetus and the neonate. *Toxicology* **122** 219–228 [erratum: 1998, *Toxicology* **130** 69].

Gehrs BC, Riddle MM, Williams WC and Smialowicz RJ (1997b) Alterations in the developing immune system of the F344 rat after perinatal exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin: II. Effects on the pup and the adult. *Toxicology* **122** 229–240 [erratum: 1998, *Toxicology* **130** 71].

Gray LE Jr, Kelce WR, Monosson E, Ostby JS and Birnbaum LS (1995). Exposure to TCDD during development permanently alters reproductive function in male Long Evans rats and hamsters: reduced ejaculated and epididymal sperm numbers and sex accessory gland weights in offspring with normal androgenic status. *Toxicology and Applied Pharmacology* **131** 108–118.

Gray LE, Wolf C, Mann P and Ostby JS (1997a) *In utero* exposure to low doses of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) alters reproductive development of female Long Evans hooded rat offspring. *Toxicology and Applied Pharmacology* **146** 237–244.

Gray LE, Ostby JS and Kelce WR (1997b) A dose–response analysis of the reproductive effects of a single gestational dose of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) in male Long Evans hooded rat offspring. *Toxicology and Applied Pharmacology* **146** 11–20.

Harrad SJ and Jones KC (1992) A source inventory and budget for chlorinated dioxins and furans in the United Kingdom environment. *Science of the Total Environment* **126** 89–107.

Hooiveld M, Heederik D and Bueno de Mesquita HB (1996) Preliminary results of the second follow-up of a Dutch cohort of workers occupationally exposed to phenoxy herbicides, chlorophenols and contaminants. *Organohalogen Compounds* **30** 185–189.

Hu K and Bunce NJ (1999) Metabolism of polychlorinated dibenzo-*p*-dioxins and related dioxin-like compounds. *Journal of Toxicology and Environmental Health* **2B** 183–210.

Hurst CH, DeVito MJ and Birnbaum LS (2000a) Tissue disposition of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) in maternal and developing Long Evans rats following subchronic exposure. *Toxicological Sciences* **57** 275–283.

Hurst CH, DeVito MJ, Setzer RW and Birnbaum LS (2000b) Acute administration of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) in pregnant Long Evans rats: association of measured tissue concentrations with developmental effects. *Toxicological Sciences* **53**(2) 411–420.

IARC (1997) *Polychlorinated Dibenzo-para-dioxins and Polychlorinated Dibenzofurans*, IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, vol. 69, International Agency for Research on Cancer, Lyon.

Kello D and Yrjänheikki E (1992) Assessment of health hazards associated with exposure to dioxins. *Chemosphere* **25** 1067–1070.

Kociba RJ, Keyes DG, Beyer JE, Carreon RM, Wade CE, Dittenber A, Kalnins RP, Frauson LE, Park CN, Barnard SD, Hummel RA and Humiston CG (1978) Results of a two-year chronic toxicity and oncogenicity study of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin in rats. *Toxicology and Applied Pharmacology* **46** 279–303.

Kogevinas M, Becher H, Benn T, Bertazzi PA, Boffetta P, Bueno-de-Mesquita HB, Coggon D, Colin D, Flesch-Janys D, Fingerhut M, Green L, Kauppinen T, Littorin M, Lynge E, Mathews JD, Neuberger M, Pearce N and Saracci R (1997) Cancer mortality in workers exposed to phenoxy herbicides, chlorophenyls, and dioxins: an expanded and updated international cohort study. *American Journal of Epidemiology* **145** 1061–1075.

Kutz FW, Barnes DG, Bottimore DP, Greim H and Bretthauer EW (1990) The International Toxicity Weighting Factor (I-TEF) method of risk assessment for complex mixtures of dioxins and related compounds. *Chemosphere* **20** 751–758.

Mably TA, Moore RW and Peterson RE (1992a) *In utero* and lactational exposure of male rats to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. 1. Effects on androgenic status. *Toxicology and Applied Pharmacology* **114** 97–107.

Mably TA, Moore RW, Goy RW, *et al* (1992b) *In utero* and lactational exposure of male rats to 2,3,7,8- tetrachlorodibenzo-*p*-dioxin. 2. Effects on sexual behaviour and the regulation of luteinizing hormone secretion in adulthood. *Toxicology and Applied Pharmacology* **114** 108–117.

Mably TA, Bjerke DL, Moore RW, *et al* (1992c) *In utero* and lactational exposure of male rats to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. 3. Effects on spermatogenesis and reproductive capability. *Toxicology and Applied Pharmacology* **114** 118–126.

MAFF (1992) *Dioxins in Food*, The 31st Report of the Steering Group on Food Surveillance, The Stationery Office, London.

MAFF (1997) *Dioxins and Polychlorinated Biphenyls in Foods and Human Milk*, Food Surveillance Information Sheet No 105, Ministry of Agriculture, Fisheries and Food, Food Safety Directorate, London.

MAFF (1999) *Dioxins and PCBs in UK and Imported Marine Fish*, Food Surveillance Information Sheet No 184, Ministry of Agriculture, Fisheries and Food, Joint Food Safety and Standards Group, London.

Manz A, Berger J, Dwyer JH, Flesch-Janys D, Nagel S and Waltsgott H (1991) Cancer mortality among workers in chemical plant contaminated with dioxin. *Lancet* **338** 959–964.

Mukerjee D (1998) Health impact of polychlorinated dibenzo-*p*-dioxins: a critical review. *Journal of the Air and Waste Management Association* **48** 157–165.

Murray FJ, Smith FA, Nitschke KD, Humiston CG, Kociba RJ and Schwetz BA (1979) Three-generation reproduction study of rats given 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) in the diet. *Toxicology and Applied Pharmacology* **50** 241–252.

Nohara K, Fujimaki H, Tsukumo S, Ushio H, Miyabara Y, Kijama M, Tohyama C and Yonemoto J (2000) The effects of perinatal exposure to low doses of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin on immune organs in rats. *Toxicology* **154**(1–3) 123–133.

NTP (1980) *Bioassay of 1,2,3,6,7,8- and 1,2,3,7,8,9-Hexachlorodibenzo-p-dioxin (Gavage) for Possible Carcinogenicity*, National Toxicology Program, DHHS Publ. No (NIH) 80-1754, US Department of Health and Human Services.

NTP (1982) *Carcinogenicity Bioassay of 2,3,7,8-Tetrachlorodibenzo-p-dioxin in Osborne-Mendel Rats and B6F3C1 Mice (Gavage Study)*, National Toxicology Program, NTP Report TR-209, Department of Health and Human Services, Research Triangle Park.

Ohsako S, Miyabara Y, Nishimura N, Kurosawa S, Sakaue M, Ishimura R, Sato M, Takeda K, Aoki Y, Soni H, Tohyama C and Yonemoto J (2001) Maternal exposure to a low dose of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) suppressed the development of reproductive organs of male rats: dose-dependent increase of mRNA levels of 5-alpha-reductase type 2 in contrast to decrease of androgen receptor in the pubertal ventral prostate. *Toxicological Sciences* **60** 132–143.

Ott MG and Zober A (1996) Cause specific mortality and cancer incidence among employees exposed to 2,3,7,8-TCDD after a 1953 reactor accident. *Occupational and Environmental Medicine* **53** 606–612.

Poiger H and Schlatter Ch (1986) Pharmacokinetics of 2,3,7,8-TCDD in man. *Chemosphere* **15** 1489-1494.

Pollitt F (1999) Polychlorinated dibenzodioxins and polychlorinated dibenzofurans. *Regulatory Toxicology and Pharmacology* **30** S63–S68.

Rier SE, Martin DC, Bowman RE, Dmowski WP and Becker JL (1993) Endometriosis in Rhesus Monkeys (*Macaca mulatta*) following chronic exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD). *Fundamental and Applied Toxicology* **21** 433–441.

Rier SE, Turner WE, Martin DC, Morris R, Lucier GW and Clark GC (2001a) Serum levels of TCDD and dioxin-like chemicals in rhesus monkeys chronically exposed to dioxin: correlation of increased serum PCB levels with endometriosis. *Toxicological Sciences* **59** 147–159.

Rier SE, Coe CL, Lemieux AM, Martin DC, Morris R, Lucier GW and Clark GC (2001b) Increased tumor necrosis factor-a production by peripheral blood leukocytes from TCDD-exposed rhesus monkeys. *Toxicological Sciences* **60** 327–337.

Rose CL, McKay WA and Ambidge PF (1994) PCDD and PCDF levels in river systems in England and Wales, UK. *Chemosphere* **29** 1279–1292.

Schantz S and Bowman RE (1989) Learning in monkeys exposed perinatally to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD). *Neurotoxicology and Teratology* **11** 13–19.

Schantz S, Ferguson SA and Bowman RE (1992) Effects of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin on behaviour of monkeys in peer groups. *Neurotoxicology and Teratology* **14** 433–446.

USEPA (1994) *Health Assessment Document for 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) and Related Compounds*, vols I to III, EPA/600/BP-92/001(a,b,c), External Review Draft, US Environmental Protection Agency, Office of Health and Environmental Assessment, and Office of Research and Development, Washington, DC.

USEPA (1995a) *Summary of the Public Comments Received on the External Review Draft of 'Estimating Exposure to Dioxin-Like Compounds'*, EPA/600/6-88/005C(a,b,c), Report NCEA-95-W-001, US Environmental Protection Agency, National Center for Environmental Assessment, Office of Research and Development, Washington, DC.

USEPA (1995b) *Review of the Office of Research and Development's Reassessment of Dioxin and Dioxin-like Compounds by the Dioxin Reassessment Review Committee. An EPA SAB Report: A Second Look at Dioxins*, US Environmental Protection Agency, Science Advisory Board, Washington, DC.

USEPA (2000) *Exposure and Human Health Reassessment of 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) and Related Compounds. Draft Report*, US Environmental Protection Agency, National Center for Environmental Assessment, Office of Research and Development, Washington, DC.

USEPA (2002) Integrated Risk Information System (IRIS); announcement of 2002 program; request for information. *Federal Register* **67** 1212.

van den Berg M, Birnbaum L, Bosveld ATC, Brunström B, Cook P, Feeley M, Giesy JP, Hanberg A, Hasagawa R, Kennedy SW, Kubiak T, Larsen JC, van Leeuwen FXR, Liem AKD, Nolt C, Peterson RE, Poellinger L, Safe S, Schrenk D, Tillitt D, Tysklind M, Younes M, Waern F and Zacharewski T (1998) Toxic equivalency factors (TEFs) for PCBs, PCDDs, PCDFs for humans and wildlife. *Environmental Health Perspectives* **106** 775–792.

van den Berg M, Peterson RE and Schrenk D (2000) Human risk assessment and TEFs. *Food Additives and Contaminants* **17** 347–358.

van Leeuwen FXR and Younes M, eds (2000) *Food Additives and Contaminants* **17**(4), Assessment of the health risk of dioxins: re-evaluation of the tolerable daily intake (TDI), Taylor & Francis, London.

WHO (1989) *Polychlorinated-para-dioxins and Dibenzofurans*, International Programme on Chemical Safety, Environmental Health Criteria 88, World Health Organization, Geneva.

WHO (1991) *Summary Report – Consultation on Tolerable Daily Intake from Food of PCDDs and PCDFs*, Bilthoven, the Netherlands, December 1990, EUR/ICP/PCS 030(S) 0369n, World Health Organization Regional Office for Europe, Copenhagen.

WHO (1992) Tolerable daily intake of PCDDs and PCDFs. *Toxic Substances Journal* **12** 101–128.

WHO (1998) *Assessment of the Health Risk of Dioxins: A Re-evaluation of the Tolerable Daily Intake (TDI)*, Consultation, May 1998, World Health Organization, Geneva.
Available on-line at http://www.who.int/pcs/docs/dioxin-exec-sum/exe-sum-final.html

WHO (2000) *Air Quality Guidelines for Europe*, 2nd edn, WHO Regional Publications, European Series No 91, World Health Organization, Geneva.

WHO (2001) *Summary and Conclusions*, Joint FAO/WHO Expert Committee on Food Additives Fifty-Seventh Meeting, Rome, June 2001.
Available on-line at http://www.fao.org/waicent/faoinfo/economic/esn/jecfa

Zober A, Messerer P and Huber P (1990) Thirty-four-year mortality follow-up of BASF employees exposed to 2,3,7,8-TCDD after the 1953 incident. *International Archives of Occupational and Environmental Health* **62** 139–157.