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Toxicity reference values for mink exposed to 2,3,7,8-tetrachlodibenzo-*p*-dioxin (TCDD) equivalents (TEQs)

A.L. Blankenship[a,b,*], D.P. Kay[a], M.J. Zwiernik[c], R.R. Holem[a], J.L. Newsted[a], M. Hecker[a,c], J.P. Giesy[c,d,e]

[a]*Entrix, Inc., Okemos, 4295 Okemos Road, Suite 101, Okemos, MI 48864, USA*
[b]*University of Michigan Medical School, Ann Arbor, MI 48109, USA*
[c]*Department of Zoology, National Food Safety and Toxicology Center, Michigan State University, East Lansing, MI 48824, USA*
[d]*Department of Biomedical Veterinary Sciences and Toxicology Centre, University of Saskatchewan, Saskatoon, Saskatchewan, Canada*
[e]*Department of Biology and Chemistry, City University of Hong Kong, Kowloon, Hong Kong, SAR China*

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Abstract

Dietary and tissue residue-based toxicity reference values (TRVs) were derived for mink from the published results of studies in which mink were exposed to polychlorinated dibenzo-*p*-dioxins (PCDDs), dibenzofurans (PCDFs), biphenyls (PCBs), or related compounds. Because the primary mechanism of toxic action at the least concentration for these compounds is related to activation of the aryl hydrocarbon receptor (AhR), TRVs were described on the basis of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) equivalents (TEQ). Each published study was critically reviewed for its usefulness in deriving a TRV based on the following criteria: (1) close relatedness of the test species to the wildlife receptor of concern (only mink studies were reviewed in this paper); (2) chronic duration of exposure which included sensitive life stages to evaluate potential developmental and reproductive effects; (3) measurement of ecologically relevant endpoints; (4) availability of congener-specific data to calculate TEQ concentrations; and (5) minimal impact of co-contaminants. Dietary TRVs for mink exposed to TEQ ranged from 12.1 to 56.6 ng TEQ/kg feed (wet weight) for the no observable adverse effect level (NOAEL) and from 50.4 to 242 ng TEQ/kg feed (wet weight) for the lowest observable adverse effect level (LOAEL). TRVs based on tissue residue concentrations ranged from 50.2 to 77.8 ng TEQ/kg liver (wet weight) for the no observable adverse effect concentration (NOAEC) and the value was 189 ng TEQ/kg liver (wet weight) for the lowest observable adverse effect concentration (LOAEC). Selection of a TRV should be based on studies of compounds that are most similar to those at a site of interest. In particular, it was determined that the effects of PCDFs could not be accurately predicted from the use of TEQ-based TRVs developed from studies of PCDDs or PCBs. Risk assessors should be aware that exceedance of these TRVs would not necessarily be expected to lead to ecologically relevant adverse effects because of the inherently conservative assumptions made in the TRV derivation process.

Keywords: Ecological risk assessment; Dioxin; PCDDs; PCDFs; PCBs; Furans

1. Introduction

Mink (*Mustela vison*) are an important, albeit seldom seen, species that can be at risk in aquatic ecosystems contaminated with persistent, bioaccumulative, and toxic (PBT) pollutants (Platonow and Karstad, 1973; Aulerich et al., 1974; Hornshaw et al., 1983; Kihlstrom et al., 1992; Hochstein et al., 1998; Brunstrom et al., 2001). Mink have been found to bioaccumulate specific congeners of PBT pollutants such as polychlorinated dibenzo-*p*-dioxins (PCDDs), dibenzofurans (PCDFs), biphenyls (PCBs), and related compounds based on field studies of wild mink (Haffner et al., 1998; Millsap et al., 2004; Martin et al., 2006a, b) and laboratory exposures of ranch mink (Ringer et al., 1972; Tillitt et al., 1996; Halbrook et al., 1999; Bursian et al., 2006a–c). In addition, mink have been found

*Corresponding author. Entrix, Inc., Okemos, 4295 Okemos Road, Suite 101, Okemos, MI 48864, USA. Fax: +1 517 381 1435.
E-mail address: ablankenship@entrix.com (A.L. Blankenship).

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to be one of the most sensitive species to the toxic effects of these compounds (Heaton et al., 1995; Tillitt et al., 1996). It is for these reasons that mink are one of the most commonly selected receptors in ecological risk assessments (ERAs) for sites involving aquatic habitats with elevated concentrations of PCDDs, PCDFs, PCBs, and related compounds (USEPA, 1995, 2000, 2005a; Sample et al., 1996; GES/MDEQ, 2003). In order to effectively protect mink and have the best estimate of risk possible, it is important to reduce uncertainty regarding potential exposure by direct measurement of tissue residue concentrations in mink and their primary dietary items *and* to have a good understanding of toxicity thresholds.

From the early 1970s to the present, numerous toxicological studies have been conducted with mink (reviewed herein). However, many of these studies have been conducted with individual congeners, congener cocktails, various site-specific environmental mixtures (including some with potentially significant concentrations of co-contaminants), and conducted under different conditions, experimental designs, time periods, and with different endpoints. Because of these differences among studies, interpretation of individual studies and comparison among studies and derivation of appropriate toxicity reference values (TRVs) can be difficult. In addition, selection of the most appropriate study to form the basis of a TRV to assess risk at specific sites is not always straightforward. This is particularly true when using the 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) equivalent (TEQ) approach to describe threshold concentrations for one class of compounds from another. Thus, the overall objective of this paper is to critically review and summarize available toxicological studies and provide guidance on the selection of the most appropriate and scientifically defensible TRVs for mink exposed to PCDDs, PCDFs, PCBs, and related compounds expressed as TEQ. While it is beyond the scope of this paper to provide a comprehensive review of all mink toxicity data for these compounds, the focus will be on those data that provide toxicological information for ecologically relevant endpoints in response to chronic, dietary exposures. The limitations of the various studies are also discussed. Finally, the predictive capacity of the calculated TRV values will be assessed by comparing them to those derived in other laboratory and field studies of mink exposed to aryl hydrocarbon receptor (AhR)-active compounds, either singly to the same compound or in mixtures.

1.1. PCDDS, PCDFs, PCBs, TEFs, TEQ, and relative potencies

Theoretically, there are 75, 135, and 209 possible congeners of PCDDs, PCDFs and PCBs, respectively (Erickson, 1997). These congeners vary in the number and position of chlorine substitutions. Despite their structural relatedness, each of these congeners has different physical–chemical properties that affect their fate, transport, and bioavailability in the environment (Erickson, 1997). In the environment, PCDD, PCDF, and PCB congeners are predominantly associated with particulate material, such as sediments, suspended material, and soils (Erickson, 1997). Of the PCDD, PCDF, and PCB congeners, 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (2,3,7,8-TCDD), also referred to as TCDD, is considered to be the most potent and is the one most studied (Van den Berg et al., 1998, 2006). Observed effects of TCDD and related chemicals in wildlife and laboratory animals include biochemical adaptive changes such as enzyme induction, developmental deformities, reproductive failure, liver damage, wasting syndrome, and death (Giesy et al., 1994a; Blankenship and Giesy, 2002; Hilscherova et al., 2003). While there are a number of other structurally related polychlorinated, diaromatic compounds, in most situations the above-listed compounds account for most of the toxic potency of environmental mixtures (Giesy et al., 1994b; Blankenship et al., 2000).

Toxicity equivalency factors (TEFs) are used to allow assessment of the additive toxicity of PCDDs, PCDFs and similar compounds that act through a common mechanism of action when they occur in mixtures (Giesy et al., 1994b; Van den Berg et al., 1998, 2006; Blankenship et al., 2000). The critical mechanism of action which results in the least allowable exposure to a mixture of TCDD and related compounds at the cellular level is primarily mediated via the AhR (Giesy and Kannan, 1998; Kannan et al., 2000; Blankenship and Giesy, 2002). Because of this assumed similarity in the mechanism of action, concentrations of 17 PCDD and PCDF congeners substituted with chlorines at positions 2, 3, 7, and 8 (and a structurally related set of 12 PCB congeners) are often converted to TEQ using the 2005 World Health Organization (WHO) TEFs (Table 1, Van den Berg et al., 2006) (Eq. (1)). TEQ values reported herein were calculated using these 2005 TEF values. These calculated values may differ from those in the original studies because of different TEFs (such as 1998 TEFs; Van den Berg et al., 1998) formerly used in the calculation of TEQ. TEF values, such as those proposed by the WHO are not precise measures of relative potencies for PCDD, PCDF, and PCB congeners. Rather, they are consensus values that purposely overestimate the relative potency of congeners across a taxonomic class for the express purpose of risk assessment. TEF values are designed to be protective, rather than predictive of thresholds of effects. As such, they are uncertain and may vary among species, measurement endpoints, and relative proportions of chemicals in complex mixtures. Thus, relative potency factors (RPFs) from the scientific literature may be used in place of WHO TEFs in instances where related or same species data are available in order to reduce uncertainty (USEPA, 2003a). The TEFs are consensus values developed for use in risk assessments and are thus, intentional overestimates that provide a level of conservatism and safety, by resulting in overestimates of the relative potency of individual constituents in mixtures (Van den Berg et al.,

Table 1
Mammal-specific toxic equivalency factors (TEFs) from the World Health Organization (WHO) for the 2,3,7,8-chlorine substituted PCDD and PCDF congeners and 12 PCB congeners[a]

	Mammals/humans	
	WHO 1998 TEF values	WHO 2005 TEF values
Polychlorinated dibenzo-p-dioxins		
2,3,7,8-TCDD	1	1
1,2,3,7,8-PeCDD	1	1
1,2,3,4,7,8-HxCDD	0.1	0.1
1,2,3,6,7,8-HxCDD	0.1	0.1
1,2,3,7,8,9-HxCDD	0.1	0.1
1,2,3,4,6,7,8-HpCDD	0.01	0.01
OCDD	0.0001	0.0003
Polychlorinated dibenzofurans		
2,3,7,8-TCDF	0.1	0.1
1,2,3,7,8-PeCDF	0.05	0.03
2,3,4,7,8-PeCDF	0.5	0.3
1,2,3,4,7,8-HxCDF	0.1	0.1
1,2,3,6,7,8-HxCDF	0.1	0.1
1,2,3,7,8,9-HxCDF	0.1	0.1
2,3,4,6,7,8-HxCDF	0.1	0.1
1,2,3,4,6,7,8-HpCDF	0.01	0.01
1,2,3,4,7,8,9-HpCDF	0.01	0.01
OCDF	0.0001	0.0003
Non-ortho PCBs		
3,3′,4,4′-TCB (77)	0.0001	0.0001
3,4,4′,5-TCB (81)	0.0001	0.0003
3,3′,4,4′,5-PeCB (126)	0.1	0.1
3,3′,4,4′,5,5′-HxCB (169)	0.01	0.03
Mono-ortho PCBs		
2,3,3′4,4′-PeCB (105)	0.0001	0.00003
2,3,4,4′,5-PeCB (114)	0.0005	0.00003
2,3′,4,4′,5-PeCB (118)	0.0001	0.00003
2′,3,4,4′,5-PeCB (123)	0.0001	0.00003
2,3,3′,4,4′,5-HxCB (156)	0.0005	0.00003
2,3,3′,4,4′,5′-HxCB (157)	0.0005	0.00003
2,3′,4,4′,5′-HxCB (167)	0.00001	0.00003
2,3,3′,4,4′,5,5′-HpCB (189)	0.0001	0.00003

[a]Unless otherwise noted, 2005 WHO TEFs were used to calculate TEQ reported in this paper (Van den Berg et al., 2006).

2006). Some of the TEFs are based on *in vitro* studies and thus do not take into account the potential differences in accumulation, disposition and metabolism in animals. Also, for some compounds, there was so little information available that the TEFs were inferred by use of quantitative structure–activity relationships (QSAR) that are based on structural analogies among compounds. This too leads to some uncertainty in the derived TEFs and subsequent calculation of TEQ. Thus, at this point in time there is considerable uncertainty in assessments relying on the use of TEF values to calculate TEQ. This is particularly true of the PCDFs

$$\mathrm{TEQ} = \sum_{i \to n}[(\mathrm{Congener}_i \times \mathrm{TEF}_i) + \ldots\ldots.(\mathrm{Congener}_n \times \mathrm{TEF}_n)]. \quad (1)$$

Other terms that are sometimes used are concentrations of mixtures expressing equivalent toxicity or other responses, either *in vivo* or *in vitro* by use of bioassays to measure the responses to mixtures directly. In these cases, the equivalently toxic dose is referred to as TCDD-EQs (Blankenship et al., 2000). These terms refer to a single, integrated measure of TCDD equivalents that is determined in a bioassay such as with H4IIE rat hepatoma cells that respond specifically to compound mixtures that bind to and activate the AhR.

1.2. Toxicity reference values

There are several possible approaches to derive TRV values (Sample et al., 1996; Blankenship and Giesy, 2002; USEPA, 2003b). Most commonly, a single study is selected that is the most definitive and defensible among all of the available studies (Sample et al., 1996; USEPA, 1995). Such a study should have doses that bracket a clear threshold for effect with some doses above and some doses below the threshold for the endpoints of interest. Another approach to develop toxicity benchmarks is meta-data analysis, which can be powerful when there is a substantial database of studies (Reiss and Gaylor, 2005), especially those conducted in a nearly identical manner such as with a standard toxicity bioassay protocol (USEPA, 1994). However, it would be inappropriate to combine results of studies that were conducted with substantially different methodology, exposure routes, exposure duration, strains, diets, amount of co-contaminants, etc. In addition, meta-analyses often necessitate normalization to controls, which can produce misleading results. In this paper, the approach followed was selection of the most defensible and definitive study from which a toxicity threshold was bracketed by the experimental doses.

Because a given exposure in some studies may result in no adverse effects, occasionally, the NOAEL from one study may be greater than the LOAEL from a different study. Such a finding should lead to a careful review of potential causative factors for such differences. It is essential to perform a critical evaluation of the applicability of the toxicological data to the site-specific receptors of concern and exposure pathways. TRVs derived in the same species are generally not available for the majority of wildlife receptors; therefore it is often necessary to derive TRVs using toxicological data for surrogate species in combination with uncertainty factors (UFs). In this instance, however, we have restricted our literature review to studies on mink only. In addition, this paper addresses a number of other areas of uncertainty such as exposure duration and measurement endpoints that are ecologically relevant toxic effects versus those that are more suitable as biochemical markers of exposure.

1.2.1. Ideal TRV characteristics

A TRV is the concentration of a chemical in water, food, or tissues of a receptor below which toxicological effects in

receptors of concern are not expected. Ideally, TRVs are derived from chronic toxicity studies in which a dose-response relationship has been observed for ecologically relevant endpoint(s) in the species of concern, or a closely related species (Sample et al., 1996; USEPA, 1997). Specifically, some of the ideal characteristics of high-quality toxicity studies that can be used to derive TRVs include:

(1) relatedness of the test species to the receptor of concern;
(2) chronic duration of exposure including sensitive life stages to evaluate potential developmental and reproductive effects;
(3) measurement of ecologically relevant endpoints;
(4) minimal impact of co-contaminants.

There is a wide range of sensitivities of species to PCDDs, PCDFs and other AhR-active chemicals (Gasiewicz et al., 1991). Thus, the less related the test species is compared to the receptor of concern, the more uncertainty is associated with the TRV. Mustelids, such as mink and otter, have been observed to be among the most sensitive mammalian species for which information on the effects of PCDDs, PCDFs and related compounds is available (Tillitt et al., 1996; Kannan et al., 2000). Several studies have been conducted with mink, which makes a species UF unnecessary. As for exposure duration, acute studies are of little use when trying to establish no observable adverse effect levels (NOAELs) and lowest observable adverse effect levels (LOAELs) for chronic effects of PCDDs, PCDFs and related compounds on mink. Similarly, subtle biochemical effects may have little or no relevance to the long-term reproductive success of mink. In fact, some of these responses may be adaptive in nature and actually increase tolerance and/or resistance to the effects of PCDDs, PCDFs and related compounds (Santostefano et al., 1996). As for co-contaminants, their presence in test diets can substantially confound the toxicity results relative to a single chemical or class of chemicals. In particular, assignment of causality, which is important in risk assessment, can be problematic when toxicologically relevant concentrations of co-contaminants are present. Thus, such studies should be evaluated carefully to determine the potential impact of co-contaminants. Such studies can be useful and are most appropriate to answer site-specific questions but may not be applicable to other sites. Since few studies have been designed to fulfill all of the ideal characteristics of a high quality study that match the needs of an ERA, it is sometimes necessary to apply UFs (discussed later) or to reject a study from further consideration. In either case, the rationale should be clearly documented for applying UFs or for rejecting a study. In this paper, details are provided that explain the rationale for inclusion or exclusion of each study for further consideration. For example, ecologically relevant endpoints such as effects on reproductive and developmental toxicity and reduced survival were evaluated and used whenever possible.

Sources of toxicological data that were reviewed to develop TRVs included primary, peer-reviewed scientific literature, pertinent reviews of PCDDs, PCDFs and related chemicals, the Oak Ridge National Laboratory Report on benchmarks for wildlife, miscellaneous USEPA reports, and other relevant sources of information.

1.2.2. Negative result studies

Exposure in some studies may result in no adverse effects. While this "negative" result has limited utility for some purposes such as the direct determination of LOAEL or NOAEL values, depending on the dosing ranges employed, the results of such studies can be useful in placing bounds on the potential range of values for the NOAEL and/or as supportive line of evidence in establishing a NOAEL that is derived from a separate study. Occasionally, the NOAEL from one study may be greater than the LOAEL from a different study. Such a finding should lead to a careful review of potential causative factors for such differences.

1.2.3. Dietary-based TRVs

While mink can be exposed to PCDDs, PCDFs, PCBs and related compounds through ingestion, dermal, and inhalation exposure pathways, due to the physical–chemical characteristics of the compounds and the types and rate of food consumptions of mink, the predominant environmental exposure pathway is through ingestion (USEPA, 1993; Giesy et al., 1994a; Giesy and Kannan, 1998; Blankenship and Giesy, 2002). Thus, a literature search was conducted to identify studies from which dietary TRVs could be derived. Typically, studies of toxicity of compounds in the diet have been conducted by adding known concentrations of PCDDs, PCDFs and related compounds to the diet. If the body weights and ingestion rates of the test animals are known or can be estimated, then the dietary concentrations can be converted to a daily dose in the units of mg (test agent)/kg body weight/d (USEPA, 1993). The resulting TRVs can be compared to site-specific estimates of exposure through the calculation of an average potential daily dose (APDD). All concentrations are expressed on a wet weight basis unless specified otherwise. In addition, lipid-normalized data are presented where available.

1.2.4. Tissue residue-based TRVs

In addition to dietary TRVs, TRVs based on concentrations of TEQ in specific tissues of the receptor of concern are increasingly being used to evaluate the potential for adverse effects due to PCDDs, PCDFs and related compounds. For the purposes of this paper, the term "tissue residue-based TRV" is synonymous with "maximum allowable tissue concentration (MATC)", a term that is sometimes used by agencies and reported in the literature. In this paper, tissue residue-based TRVs were compiled for mink.

Tissue residue-based TRVs can be compared to site-specific, measurements of tissue concentrations in receptors

of concern. When food chain models are used to estimate concentrations of residues in receptor tissues, caution should be exercised due to the inherent uncertainty associated with such predictive models. Some of the uncertainty relating to food chain modeling includes factors such as site foraging frequency, dietary composition, concentrations of PCDDs, PCDFs and related compounds in dietary items. Tissue residue effect level data are gaining increasing regulatory acceptance as evidenced in the "Canadian Tissue Residue Guidelines (TRG) for the Protection of Wildlife Consumers of Aquatic Biota" (Canadian Environment Quality Guidelines, 2001).

1.2.5. Uncertainty

It is essential to perform a critical evaluation of the applicability of the toxicological data to the site-specific receptors of concern and exposure pathways. TRVs derived in the same species are generally not available for the majority of wildlife receptors and, therefore, it is necessary to derive TRVs using toxicological data for surrogate species in combination with UFs. Uncertainty concerning interpretation of the toxicity test information among different species, different laboratory endpoints, and differences in experimental design, age of test animals, duration of test, etc., are often addressed by applying UFs to the toxicology data to derive the final TRV (Sample et al., 1996; USEPA, 1995). However, for this paper, UFs were not used because TRVs were available directly from the published studies for chronically exposed mink from feeding studies in which ecologically relevant endpoints were evaluated as discussed below. In addition, there is uncertainty concerning the use of WHO TEFs since these are order of magnitude, conservative estimates of relative potency for all mammals and may differ for mink, which is discussed in more detail later in the paper.

2. Toxicity studies with mink exposed to dioxins and related AhR-agonists

2.1. Literature search and sorting strategy

To identify all relevant literature with regard to the development of TRVs for mink, a thorough literature search was conducted using the ISI/Medline search function of Reference Manager 11 (Thomson ResearchSoft, Carlsbad, CA). A keyword elimination system was applied to conduct a logical search by the stepwise inclusion of additional keywords. In brief, keyword combinations including different descriptors for the receptor, such as mink and *Mustela,* and chemical of interest, such as dioxin, TCDD, PCDD, dibenzofuran, TCDF, and PCDF, were used in the search schemes. To avoid duplication of already retrieved references, keywords that were previously searched in the same context were excluded in subsequent searches for the same receptor.

References retrieved after conducting the above described literature search were sorted based on their relevance with respect to deriving dietary TRVs for mink. A list of "relevant sort criteria" was established that allows one to distinguish between relevant and non-relevant studies (Fig. 1). An endpoint was considered relevant if it is predictive of a biological effect that can be associated with an impact on:

1. survival
2. reproduction
3. growth

A detailed list of acceptable predictive endpoints is presented (Fig. 1). *In vitro* and/or *ex vivo* studies were not considered relevant because to date there exist no reliable models to predict biological *in vivo* effects using cellular or isolated tissue systems. Furthermore, a study was eliminated in the sorting process when the only endpoints measured were either of a biochemical or molecular nature such as enzymatic or transcriptional assays. The reason for this was that most of these effects are likely to be reversible and are not reliable predictors of pathological alterations when not measured in combination with behavioral, morphological or histological endpoints. To avoid elimination of potentially relevant studies, all reference files that could not be clearly assigned to the "relevant reference" or "eliminated reference" data base were copied into an "uncertain reference" file folder for additional, detailed review.

2.2. Individual toxicity studies with mink exposed to PCDDs, PCDFs, PCBs, and related compounds

There is considerable toxicological information available for the effects of PCDDs and PCDFs and related chemicals, such as PCBs, both individually and as mixtures on mink and other mammals for both dietary and tissue residue-based effect levels. However, few studies were designed in such a way that TRVs could be determined reliably. Some of the more common limitations of available studies that were reviewed can be generally described as either:

- Duration was too short to be representative of long-term or chronic effects
- Exposure route was not dietary and is thus not appropriate to derive a dietary TRV
- Endpoints are not ecologically relevant (e.g. enzyme induction and other subtle biochemical effects with no clear relevance to adverse population-level effects)
- Doses were too great and therefore responses were too severe (adult mortality) to be used to derive a TRV
- Concentrations of TEQ could not be calculated from the available data
- Substantial influence of co-contaminants

Other limitations of the studies included limited information on the dose-response relationships. Specifically, in

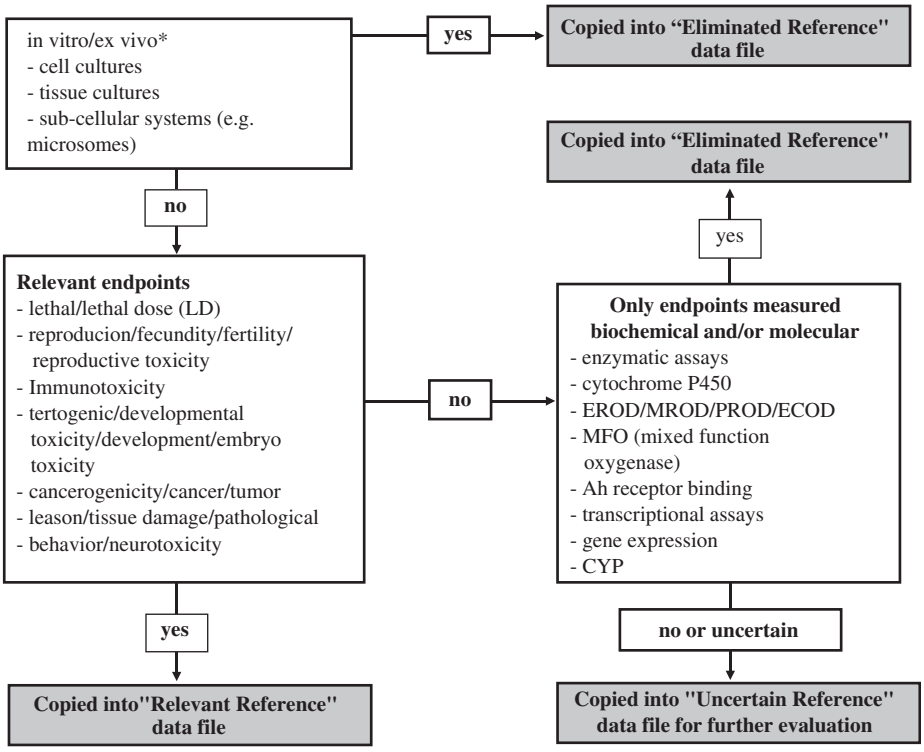

Fig. 1. Sort criteria for evaluation of ISI/Medline generated mink TRV reference search results.

many studies the dosing scheme did not bracket the NOAEL (e.g., the lowest dose tested was the LOAEL). In these studies, the NOAEL is usually determined by either applying UFs or using the control diet as the NOAEL. These studies are problematic because the LOAEL is somewhere between the lowest dose tested and the control dose. Thus, from such studies there is considerable uncertainty surrounding the LOAEL and NOAEL. The preferred approach in order to minimize uncertainty is to find studies from which doses less than the LOAEL were tested.

Additional limitations of the studies reviewed related to differences in congener-specific toxicokinetics and species-specific TEFs among PCDDs and related compounds. As discussed in the previous section, the various PCDD, PCDF, and PCB congeners differ in their *in vivo* absorption, distribution, metabolism, and elimination. These congeners also differ in their ability to bind to and activate the AhR, with toxicity proportional to the avidity of binding to the AhR (Safe et al., 1990). In addition, many of these parameters are species-specific and very few of these parameters have been directly measured in mink. Thus for the purposes of this paper, mink-specific data were used wherever possible. Where such data were not available, assumptions were based upon data for other species or consensus data such as the 1998 and 2006 WHO TEFs. In cases where a mink toxicity study used TEFs other than WHO TEFs, the reported TEQ were recalculated using the 2006 WHO TEFs when possible.

Finally, the dietary-based TRVs presented in this review utilize units based on concentrations in feed (unless noted otherwise). For the purposes of conducting an ERA, it may be necessary to convert such data to account for ingestion rate and body weight in order to compare to estimated exposures. Readers are encouraged to use study-specific data when available or otherwise utilize default assumptions (USEPA, 1995). In the rest of this section, toxicity studies are critically reviewed for their usefulness in deriving TRVs for mink.

2.2.1. *Aulerich et al. (1971)*

In this study, mink were fed diets of ocean perch, Great Lakes Coho salmon, other Great Lakes fish, or West Coast Coho salmon to investigate reproductive problems attributed to the feeding of Great Lakes Coho salmon to mink. Three experiments were conducted between 1968 and 1970. However, concentrations of PCB congeners and other co-contaminants, except DDT and its metabolites and dieldrin, were not measured in the diets. Thus, the results of this study could not be used to develop TRVs based on TEQ.

2.2.2. *Aulerich et al. (1973)*

This study continued the work of Aulerich et al., (1971) with two new experiments to investigate the possible involvement of PCBs (Table 2). Data on reproductive performance was reported for only one dose (30 mg/kg). At this dose, adult mortality and complete reproductive failure was reported. Thus, since the effects at this dose are relatively severe and congener-specific data were not available, the results of this study could not be used to develop TRVs.

Table 2
Average PCB residues in tissues from control mink (*Mustela vison*) and mink that died while receiving diets that contained Coho salmon or supplemental PCBs (Table 8, Aulerich et al., 1973)

Treatment group	No. of mink	Mean PCB concentration (mg PCB/kg) ± SE						
		Brain	Liver	Kidney	Spleen	Lung	Muscle	Heart
30% ocean fish mix control	4	<0.01	<0.01	<0.01	<0.01	<0.01	ND	ND
30% Lake Michigan Coho salmon	3	11.1 ± 0.78	5.21 ± 1.66	6.37 ± 0.25	6.19 ± 0.09	5.15 ± 0.22	4.73 ± 1.80	2.84 ± 1.21
30% ocean fish mix plus 30 mg PCB/kg; ppm)[a]	12	11.0 ± 1.43	4.18 ± 0.58	4.47 ± 0.42	4.79 ± 0.39	4.78 ± 0.53	4.88 ± 0.54	3.26 ± 0.49

ND = None detected.
[a]PCBs consisted of 10 mg/kg each of Aroclors 1242, 1248 and 1254.

2.2.3. *Aulerich and Ringer (1977)*

This study continued the work of Aulerich et al. (1971, 1973) with additional experiments to investigate the dose-response relationship of Aroclor 1254 and to investigate the relative potencies of different Aroclors with reproductive endpoints. In this study, mink were exposed by diet to several doses of Aroclor 1254 (0, 1, 5, 15 mg/kg in feed) for up to 130 d and a dose of 2 mg/kg for up to 298 d through a critical reproductive life stage. Conversions of concentrations in the diet to a daily dose were based on a normalized ingestion rate of 0.15 kg/kg/d (based on assumptions of a food consumption rate of 0.15 kg/d and a body weight of 1.0 kg; USEPA, 1995). No adverse effects were observed on the number of kits per female at a dose level of 1 mg/kg in feed (or 0.15 mg PCB/kg/d; see Table 13 of Aulerich and Ringer, 1977). At this dose, the number of kits per female was 4.3, which was not a statistically significant difference compared to three different sets of controls in which the number of kits ranged from 4.1 to 6.0. Furthermore, a litter average of 4.0 kits per mated female is considered normal for the Michigan State University mink studies (Ringer and Aulerich, 1980). At a dose of 2 mg/kg of Aroclor 1254 in feed (or 0.3 mg PCB/kg/d), adverse effects were observed including a reduction in the number of kits per female. However, when Aroclors 1221, 1242, and 1016 (41 percent chlorine) were tested at dietary concentrations of 2 mg/kg (or 0.3 mg PCB/kg/d), no effects were observed on reproduction. Thus, since Aroclor 1254 was found to be more toxic than other Aroclors tested, the NOAEL and LOAEL values of 1 and 2 mg PCB/kg (or 0.15 and 0.3 mg PCB/kg/d) of Aroclor 1254 in feed should be considered a conservative estimate of the NOAEL and LOAEL, respectively. Since the study considered dietary exposure during the sensitive and ecologically relevant time period of reproduction, the 0.15 and 0.30 mg PCB/kg/d doses were considered to be chronic dietary-based NOAELs and LOAELs, respectively. Since concentrations of specific PCB congeners were not measured, it was not possible to calculate concentrations of TEQ from this study.

2.2.4. *Aulerich and Ringer (1980)*

In this study, mink were exposed by diet to three doses of Aroclor 1016 (2, 10, and 25 mg/kg in feed) for up to either 349 or 539 d through a critical reproductive life stage. Conversions of concentrations in the diet to a daily dose were based on a normalized ingestion rate of 0.15 kg/kg/d (based on assumptions of a food consumption rate of 0.15 kg/d and a body weight of 1.0 kg; USEPA, 1995). No adverse effects were observed on the numbers of kits per female at concentrations up to 25 mg PCB/kg in feed. However, the authors state that growth and survival of kits was suboptimum. The only statistically significant difference was decreased kit weight at 4 week at 25 mg PCB/kg (or 3.8 mg PCB/kg/d). Thus, since the study considered dietary exposure during the sensitive and ecologically relevant time period of reproduction, the 1.5 and 3.8 mg PCB/kg/d doses were considered to be chronic dietary-based NOAELs and LOAELs, respectively. Concentrations of total PCBs were recorded for various organs (Table 3). Since concentrations of specific PCB congeners were not measured, it was not possible to calculate concentrations of TEQ from this study.

2.2.5. *Aulerich et al. (1985)*

In this study, mink were exposed to either 2.5 mg PCB/kg of Aroclor 1254 or lower doses of individual PCB congeners for approximately 100 d through a critical reproductive life stage. Aroclor 1254 at a dose of 2.5 mg PCB/kg, ww in feed resulted in only one kit (stillborn). Thus, since the effects at this dose are relatively severe, and congener-specific data were not available, the results of this study could not be used to develop TRVs.

2.2.6. *Backlin and Bergman (1992)*

This paper is a companion paper to the Kihlstrom et al. (1992) study discussed later in this section. In this paper, morphological endpoints are discussed which are not relevant endpoints to derive TRVs for reproductive and developmental endpoints in mink.

2.2.7. *Beckett et al. (2007, in press)*

The objective of this study was to evaluate the effects of PCB 126 on the reproductive performance of mink and survival of their offspring. Mink were provided diets containing either 0 (control), 0.24, 2.4, or 24 μg PCB 126/kg feed (0, 24, 240, or 2400 ng TEQ/kg) from 3 week prior to breeding until weaning of their kits at 6 week of age (average duration of exposure was 114 d). Although the

Table 3
Average PCB residues (mg/kg) in tissues from control mink and mink fed diets supplemented with Aroclor 1016 (Table 11, Aulerich and Ringer, 1980)

Treatment	Mink per pooled treatment	Sex	Brain	Liver	Heart	Kidney	Skeletal muscle	Adipose tissue	Kits (pooled sample of 4 kits/treatment)
Basic diet	3	F	ND	0.018	0.017	ND	ND	ND	0.0006
(control)	3	F	ND	ND	ND	ND	0.007	ND	
	3	F	ND	0.031	ND	ND	ND	ND	
	3	F	0.011	ND	ND	ND	ND	0.138	
	3	M	ND	ND	ND	ND	ND	0.072	
Basic	diet + 2 mg PCB/kg, ww	2	F	0.032	0.365	0.033	0.059	0.026	1.938
	0.040								
	2	F	0.032	0.137	0.066	0.060	0.033	2.272	
	2	F	0.044	0.306	0.048	0.047	0.063	2.675	
	2	F	0.047	0.175	0.051	0.089	0.056	0.949	
2	M	0.049	0.070	0.041	0.052	0.040	1.474		
Basic	2	F	0.056	0.214	0.055	0.218	0.201	4.513	0.149
diet + 10 mg	2	F	0.136	0.626	0.096	0.352	0.212	7.621	
PCB/kg, ww	2	F	0.832	0.296	0.020	0.172	0.190	5.606	
	2	F	0.089	0.393	0.122	0.116	0.177	4.129	
	3	M	0.170	0.529	0.097	0.399	0.320	4.109	
Basic	diet + 25 mg PCB/kg, ww	3	F	0.231	0.903	0.053	0.201	0.443	7.805
	0.254								
	3	F	0.177	0.517	0.145	0.287	0.318	7.308	
	3	F	0.203	0.882	0.245	0.417	0.253	6.757	
	2	F	0.147	0.714	0.042	0.216	0.315	9.651	
3	M	0.139	0.610	0.348	0.419	0.378	8.517		

ND = None detected; Note: these data are for mink exposed from 1/6/1976–6/28/1977 and in newborn kits whelped by these females.

number of matings across treatment groups was not statistically different ($p = 0.05$), complete reproductive failure (i.e., no whelping females) was observed in the 2.4 and 24 μg PCB 126/kg feed treatment groups. All females with confirmed matings in the control and 0.24 μg PCB 126/kg feed groups had kits while no kits were produced in the 2.4 and 24 μg PCB 126/kg feed groups. No statistically significant differences were observed in the gestation length, average number of live and stillborn kits whelped per litter, kit survivability, and kit body weights at 3 and 6 week of age in the control and 0.24 μg PCB 126/kg feed groups. Because complete reproductive failure occurred at the two greatest dose levels (2.4 and 24 μg PCB 126/kg feed), the results of this study could not be used to develop TRVs.

2.2.8. Bergman et al. (1992a)

This paper is a companion paper to the Kihlstrom et al. (1992) study discussed later in this section. In this paper, tissue residues are presented for PCBs and its metabolites. However, the endpoints are not relevant to derive TRVs for reproductive and developmental endpoints in mink.

2.2.9. Bergman et al. (1992b)

This paper is a companion paper to the Kihlstrom et al. (1992) study discussed later in this section. In this paper, liver histological endpoints discussed were not relevant endpoints from which to derive TRVs for reproductive and developmental endpoints in mink.

2.2.10. Bleavins et al. (1980)

A survival and reproduction study was reviewed in which mink were exposed to nominal concentrations of 0, 5, 10, 20 or 40 mg PCB/kg, ww of Aroclor 1242 in feed for up to 247 d. This study evaluated survival in adult mink as well as important reproductive endpoints and life stages. Conversions of PCB concentrations in diet to a daily dose were made by assuming a daily 150 g/mink food consumption rate and mean body weight of 800 g (provided in paper) that would result in an average food consumption rate of 0.19 kg/kg/d. Adverse effects on survival in both adult males and females were observed at 20 mg PCB/kg (3.8 mg/kg/d). At 5 mg/kg Aroclor 1242 in feed (0.94 mg PCB/kg/d), there was complete reproductive failure in that no females mated at this dose whelped. Male reproductive parameters were not evaluated in this study. Thus, since the effects at this dose are relatively severe and congener-specific data were not available, the results of this study could not be used to develop TRVs.

2.2.11. Brunstrom (1992)

This paper is a companion paper to the Kihlstrom et al. (1992) study discussed later in this section. In this paper, enzyme biomarker endpoints are discussed, which are not

relevant endpoints from which to derive TRVs based on reproductive and developmental endpoints in mink.

2.2.12. Brunstrom et al. (2001)

In this study, female mink were exposed by diet to a technical mixture of Clophen A50 (0 [<1.5 μg PCB], 0.1, and 0.3 mg PCB/mink/d) for up to 18 month (540 d). The study included two reproductive seasons and evaluated both developmental and reproductive parameters. Conversions of concentrations in diet to daily dose were based on information provided in the study (e.g., the mean body weight for females was 1.231 and 1.123 kg for the low dose and high dose treatments, respectively). At the greatest dose of 0.267 mg PCB/kg/d, in the second reproductive season adverse effects included a reduction in whelping frequency, and a decrease in litter size and birth weight. In addition, all kits died within 24 h of birth. At the least PCB concentration, 0.081 mg PCB/kg/d, adverse effects observed at the end of the second reproduction season included reduced kit production and survival as well as a reduction in kit birth weight. Since the route of exposure in the study was dietary and the study included two reproductive seasons, the 0.081 mg PCB/kg/d dose was considered to be a chronic LOAEL. On a TEQ basis, this LOAEL was estimated to be 2.4 ng TEQ/kg/d (22 ng TEQ/kg feed, based on information provided in the paper). A NOAEL was not identified in this study. Clophen formulations have been shown to contain parts per million concentrations of PCDFs (ATSDR, 2000). However, no studies were found that quantified specific congeners and thus, it is not possible to determine concentrations of TEQ in Clophen mixtures. In addition, attempts were made in this study to separate specific PCB congeners but it is unclear how these separation techniques affected concentrations of anything except some PCB congeners. Moreover, only dietary concentrations of these PCB congeners were measured and tissue residue concentrations were not measured as part of this study. Thus, the weaknesses of this study are that few congeners were evaluated in diet and none in mink tissues. Taken together, these concerns preclude the use of the results of this study to develop TRVs for TEQ.

2.2.13. Bursian et al. (2006a, b)

This was a chronic mink feeding study that was conducted as part of USEPA's investigations of PCB risks at the Housatonic River site in Massachusetts. Basically, it was a well-designed and rigorous study in which multiple doses were evaluated. Concentrations of PCBs, PCDDs, and PCDFs were measured in the diet and mink liver with concentrations of TEQ calculated. The Bursian et al. (2006a, b) study tested doses of 1.0 (control group), 2.7, 4.3, 6.8, 12.1 and 50.4 ng TEQ/kg (ww feed) over a period of approximately 147–164 d, although twelve kits per group were maintained on their treatment dosing for approximately 160 more d until kits were approximately 30–31 week old. This study reported a LOAEL of 50.4 ng TEQ/kg (ww feed) based on a statistically significant 46% lesser survival of kits at 6 week of age and a 24% less kit body weight at 3 week of age. At the next lower dose, 12.1 ng TEQ/kg (in diet; NOAEL), there were no statistically significant differences in kit survival or kit body weight. The TRVs derived from the study conducted by Bursian et al. (2006a, b) are strengthened by the fact that the dose intervals were very close together and there were four doses (plus a control) at which there were no observable adverse effects. Although co-contaminants were potentially present in this study, the influence of co-contaminants was likely minimal because they seemed to have no effect on the lowest 4 doses or the control group.

The NOAEL and LOAEL dietary concentrations were associated with concentrations of TEQ measured in livers of female adult mink of 50.2 and 189 ng TEQ/kg (ww in liver), respectively.

2.2.14. Bursian et al. (2006c)

This was a chronic study in which mink were fed fish collected in 2000 from the Saginaw River, Michigan. Basically, it was a well-designed study in which multiple doses were evaluated. Concentrations of PCBs, PCDDs, and PCDFs were measured in the diet and mink liver with concentrations of TEQ calculated. This study tested doses of 2.1 (control group), 22.4, 36.5, and 56.6 ng TEQ/kg (ww feed) over a period of approximately 120 d, although eight kits per group were maintained on their treatment dosing for approximately 147 more days until kits were approximately 27 week old. No adverse effects on reproductive or developmental endpoints were observed at any of the doses tested, with the exception of some mild effects of maxillary and mandibular squamous epithelial proliferation in the 36.5 ng TEQ/kg dose group and mild-to-moderate maxillary and mandibular squamous epithelial proliferation in the 56.6 ng TEQ/kg dose group. It is not clear if this endpoint is ecologically relevant and whether it would affect health and survival. A separate study determined that this endpoint did not have any negative population level effects in mink (Beckett et al., 2005). Therefore, since there were no statistically significant, adverse effects on ecologically relevant endpoints (e.g., breeding success, whelping success, gestation length, litter size, kit survivability), the greatest dose in this study (56.6 ng TEQ/kg, ww diet) is the NOAEL. While not included in this paper, it should be noted that there were no statistically significant changes in kit body weight from this study (Bursian and Yamini, 2003, unpublished data). The TRVs derived from the study conducted by Bursian et al. (2006c) are strengthened by the fact that the dose intervals were very close together, there were four doses (plus a control) at which there were no observable adverse effects, and thus, the effect of co-contaminants were minimal.

The NOAEL dietary concentrations was associated with a concentration of TEQ measured in livers of 27-week old juvenile mink of 77.8 ng TEQ/kg (ww in liver), respectively. It is important to note that the body weights of these

27 week old juvenile mink were essentially the same as adult mink in this study (mean 27 week old weight range of females [1180–1390 g] and males [1710–2190 g] as reported in Martin et al., 2006a, b).

2.2.15. Edqvist et al. (1992)

The results of the study conducted by Kihlstrom et al. (1992) are discussed later in this section. In this paper, biochemical endpoints are discussed which are not relevant to derive TRVs for reproductive and developmental endpoints in mink.

2.2.16. Hakansson et al. (1992)

This paper is a companion paper to that of Kihlstrom et al. (1992) and is discussed later in this section. In this paper, concentrations of vitamin A are discussed which are not relevant to derive TRVs for reproductive and developmental endpoints in mink.

2.2.17. Halbrook et al. (1999)

In this study, mink were exposed to several doses of an environmentally weathered PCB mixture in fish from Poplar Creek (located on the Oak Ridge Reservation in Tennessee) in their diet for 198 d, through a critical reproductive life stage. Conversions of concentrations in the diet to a daily dose were made based on a food consumption rate of 0.15 kg/mink/d (provided in paper) and mean, measured body weights in the study for each diet (e.g., 1.25, 1.14, and 1.08 kg for diets C, D, and E, respectively). At a dose of 1.86 mg PCB/kg of total PCBs in feed (or 0.26 mg PCB/kg/d), adverse effects were observed including a decrease in 6-week-old male kit weights. Furthermore, at this dose, the number of kits per female was 4.3, which was a reduction compared to controls, but not a statistically significant difference. A litter average of 4.0 kits per mated female is considered normal for the Michigan State University mink studies (Aulerich and Ringer, 1980). This study is strengthened by the absence of adverse effects at two lesser doses, including 0.52 and 1.01 mg PCB/kg in feed (0.06 and 0.13 mg PCB/kg/d), which provides confidence that this study bracketed the true threshold with the doses tested. While mercury was measured in the diet, the authors state that the concentrations of mercury were less than a threshold for effects. In addition, for all other measured endpoints such as female kit weights, litter size, number born alive, and number alive at 6 week, there were no statistically significant differences compared to the controls (Halbrook et al., 1999).

Corresponding tissue residue-based NOAELs and LOAELs in liver (ww basis) can be also derived from this study (Table 4). The LOAEL in diet of 0.26 mg PCB/kg/d, corresponds to a mean PCB concentration in liver of 7.25 mg PCB/kg. A tissue residue-based NOAEL in liver is not available from the study but can be estimated from the relationship between PCB concentrations in adipose and liver from diet E (e.g., basis of the LOAEL; refer to Table 1 in the original paper). This adipose to liver relationship was then applied to the concentration of PCBs in adipose from diet D (e.g., the basis for the NOAEL) in order to estimate the PCB concentration in liver. The resulting tissue residue-based NOAEL for PCBs in liver is estimated to be 5.97 mg PCB/kg (ww). Since the study considered dietary exposure during the sensitive and ecologically relevant time period of reproduction, the 0.13 and 0.26 mg PCB/kg/d doses were considered to be chronic dietary-based NOAELs and LOAELs, respectively. However, since concentrations of specific PCB congeners were not measured, it was not possible to calculate concentrations of TEQ from this study.

2.2.18. Heaton et al. (1995)

In this study, adult ranch mink were exposed to carp collected from Saginaw Bay, Michigan at levels of 0, 10, 20 or 40% in the diet. The diets contained 0.015, 0.72, 1.53, and 2.56 mg PCB/kg diet (0.7, 16.8, 32.8, and 65.7 ng TEQ/kg diet) and were fed to mink prior to and throughout the reproductive period for a total of 85 d. Survival and reproductive performance was evaluated in both adults and kits produced during the study. Adverse effects were observed at the 0.72 mg PCB/kg (ww in feed) or 16.8 ng TEQ/kg (ww in feed) dose and included reduced kit body weight and survival. Since this study considered the dietary route of exposure as well as examining significant and sensitive reproductive life stages, the

Table 4
Mean ± SE Aroclor 1260 concentrations (mg/kg, ww) in diets and in mink tissues of adult female mink and 6-week-old-kit tissues (as modified from Table 1, Halbrook et al., 1999)

	Diet A	Diet B	Diet C	Diet D	Diet E	*P*-value
Diets ($n = 10$)	<0.005	0.94 ± 0.02	0.52 ± 0.01	1.01 ± 0.03	1.86 ± 0.06	<0.001
Adult female ($n = 8$)						
Liver	<0.005	<0.005	<0.005	<0.005	7.25 ± 0.87	–
Fat	3.17 ± 1.85	61.3 ± 12.6	NQ	106 ± 11.3	129 ± 7.73	0.003
6-week-old kits ($n = 9$)	<0.005	0.018 ± 2	<0.005	<0.005	0.154 ± 0.024	0.001
Liver	0.099 ± 0.016	0.79 ± 0.13	NQ	NQ	5.40 ± 1.97	0.002
Whole body (minus liver)	0.082 ± 0.005	1.79 ± 0.28	NQ	NQ	6.26 ± 0.89	<0.001

NQ = not quantified; note for Diet E, the published table erroneously stated the dietary concentration as 1.36 mg PCBs/kg; the correct value is 1.86 (text of published study; Halbrook et al., 1999; S. Bursian, pers. comm.).

LOAEL was determined to be 0.72 mg PCB/kg (ww in feed) or 16.8 ng TEQ/kg (ww in feed). However, while the study was well designed, and concentrations were determined in both diet and tissues, the use of carp in the diet that were collected from a contaminated area also exposed the mink to co-contaminants. For instance, when total TEQ calculated from the identified AhR-active compounds were compared to total TEQ determined by use of the H4IIE bioassay, only 20% of the total TEQ determined to be present by use of the bioassay was accounted for by the presence of PCDDs, PCDFs and PCBs. Thus, 80% of the total TEQ was the result of unknown agents (Giesy et al., 1997). As a result, the assumption that the PCDDs, PCDFs, and PCBs were the sole source of toxic equivalent that contributed to the adverse effects observed in the study likely overestimates the contribution of these compounds. In addition, it has been reported that perfluorooctanesulfonate (PFOS) was also present in the carp tissue at concentrations of 240–300 ng PFOS/g, wet wt, which would result in final dietary concentrations of PFOS of 105, 124, and 160 ng PFOS/g, wet wt, respectively (Kannan et al., 2002). Thus, since the fish in the mink diets were field collected from contaminated areas with other co-contaminants likely present at toxicologically significant levels, the results of this study are not appropriate to derive TRVs for TEQ because of potentially confounding impacts of other co-contaminants on mink in this study. Thus, the TRV values determined for the TEQ calculated from the PCDD/DFs and PCBs is likely an underestimate of the true value that would have result in the same level effect in the absence of the co-occurring contaminants.

2.2.19. Hochstein et al. (1988)

In this study, adult male mink were administered single oral doses of 2,3,7,8-TCDD at concentrations of 0 (control group), 2.5, 5.0, and 7.5 μg TCDD/kg and observed for 28 d to characterize the acute toxicity of TCDD to mink. Survival, food consumption, and multiple histological and morphological endpoints were assessed. Seventy-five and one hundred percent mortality was observed in the 5.0 and 7.5 μg/kg groups (no deaths occurred in the 0 and 2.5 μg/kg groups). A 28 d LD_{50} value of 4.2 μg TCDD/kg body weight was reported. Food consumption (g/mink/d) was reduced in the 2.5, 5.0 and 7.5 μg TCDD/kg groups and body weight decreased in a dose-dependent fashion. Mink in the 5.0 and 7.5 μg TCDD/kg groups exhibited depleted adipose tissue stores, abdominal ascites, ulceration of the stomach, mottled and discolored livers, and bloody stools. When expressed as a percentage of body weight, mean brain and kidney weights were significantly greater than controls in the 5.0 and 7.5 μg TCDD/kg groups, and heart weight was increased in the 5.0 and 7.5 μg TCDD/kg groups. Adrenal gland weights were significantly increased in a dose-dependent fashion and thyroid gland weights were significantly higher in the 7.5 μg TCDD/kg group. Red blood cell counts, total white blood cell counts, leukocyte differential counts and hematocrit and hemoglobin concentrations were elevated in surviving TCDD-exposed animals, but not significantly. Although the calculation of an LD_{50} for TCDD is of importance, several factors including small sample size ($n = 4$/group), the use of male mink only, high incidence of mortality and acute exposure regime limit the use of this study for derivation of TRVs.

2.2.20. Hochstein et al. (1998)

In this study, adult female mink were fed diets supplemented with 0 (control group), 0.001, 0.01, 0.1, 1.0, 10, or 100 μg TCDD/kg for up to 125 d to characterize the toxicity of dietary TCDD to female mink. Survival, food consumption, and multiple histological and morphological endpoints were assessed. Limited whelping and kit morphological observations are also reported in this study. By day 125, mortality had reached 62.5%, 100%, and 100% in the 1.0, 10, and 100 μg TCDD/kg groups. Reported LC_{50} values for 28 and 125 d of dietary TCDD exposure were 4.8 and 0.85 μg TCDD/kg, ww, or 0.264 and 0.047 μg TCDD/kg body weight/d (conversion to daily dose calculated based on feed consumption of control mink −5.5 g/100 g body weight/d). Mink in all groups, including the control, exhibited reduced body weight during the course of the study with significant reductions observed in the 1.0, 10, and 100 μg TCDD/kg groups at various points throughout the study. Other observed effects included a dose-dependent decrease in food consumption, increased adrenal gland weights, and increased percentage of band neutrophils in the TCDD-treated groups. Mink that died during the study exhibited depleted adipose tissue stores, abdominal ascites, ulcerations of the stomach and upper gastrointestinal tract, and bloody stools. High incidence of mortality and endpoints not relevant to TRVs for reproductive and developmental endpoints in mink limit the use of this study.

2.2.21. Hochstein et al. (2001)

In this study, mink were fed diets supplemented with 0.00006 (control group), 0.016, 0.053, 0.180, or 1.04 μg TCDD/kg for up to 132 d to characterize the chronic toxic effects of TCDD in mink, including reproduction. There was a general dose-dependent loss in mink body weights and an increase in organ weights during the study. Seventeen percent mortality (2/11) was observed in the 1.4 μg TCDD/kg group and all mink at this dose were visibly lethargic after 4–5 week of exposure. Other effects observed in the 1.4 μg TCDD/kg group were gastric ulcers, intestinal hemorrhages, decreased adipose tissue level, mottled and/or discolored livers, spleens, and kidneys, as well as deformed and elongated toenails. Mating and reproduction in all treatment groups was substandard by commercial mink farms. In part, this was due to indoor breeding of males which the authors acknowledge is not optimal but was a university requirement given the need to contain and dispose of TCDD wastes. No females in the 0.016 or 1.4 μg TCDD/kg groups

whelped and there was a dose-dependent decrease in kit body weights in the groups where kits were whelped. Three-week survival rates of 83%, 47%, and 11% were recorded for the kits in the control, 0.053, and 0.18 μg TCDD/kg groups, respectively. Given the authors' concern that the mating and reproduction was substandard for this study, this study does not provided a sound basis for developing TRVs.

2.2.22. Hornshaw et al. (1983)

In this study, mink were exposed by diet to five different fish or fish products to evaluate the impact of PCBs on survival and reproduction. The diets included carp and white suckers collected from Saginaw Bay, MI., yellow perch from northern Lake Erie, lake white fish from Big Bay de Noc, Lake Michigan, and alewife fishmeal from Green Bay, Lake Michigan. There were two separate studies that were conducted: (1) five different fish diets (e.g., PCB concentrations ranged from 0.09 to 1.5 mg/kg in diets) were evaluated in year 1 with 13–15 week old females and an exposure period of 250 d and (2) a perch/sucker diet (PCB concentration was 0.66 mg/kg) was evaluated in the second year with 8–10 week old females for a 290 d exposure. This review only focuses on the second year's study results in which juvenile female mink were exposed to either a standard mink diet (0.04 mg PCB/kg feed) or to a perch/sucker diet (0.66 mg PCB/kg feed). Concentrations of PCB residues in adipose tissue were reported (Table 5). Conversion of dietary concentrations to a daily dose was based on an ingestion rate of 0.15 kg/mink/d and assumed a mean body weight of 1 kg (USEPA, 1995). Adverse effects were observed at 0.66 mg PCB/kg in feed (or 0.099 mg PCB/kg/d) and included a reduction in number of young born live per female, a reduction in the average number of live kits whelped and in percent survival of kits from birth to 4 week. Since this study considered dietary exposure during reproduction, the LOAEL was considered to be 0.66 mg PCB/kg. However, since the fish in the mink diets were field collected from contaminated areas with other co-contaminants likely present at toxicologically significant levels, the results of this study are not appropriate to derive TRVs for TEQ because of potentially confounding impacts of other co-contaminants on mink in this study. In addition, since concentrations of specific PCB congeners were not measured, it was not possible to calculate concentrations of TEQ from this study.

Table 5
PCB residues in adipose tissue of mink fed diets that contained various fish or fish products (as modified from Table 6, Hornshaw et al., 1983)

Mean PCB residues in adipose[a] (mg/kg, lipid)					
Dietary group	*N*	Total	Peak A	Peak D	Peak I
Control	7[b]	1.7	0.13	0.64	0.98
Carp	4	24.8	3.53	8.58	12.73
Sucker	4	13.5	0.36	1.70	11.46
Perch scraps	4	7.6	0.42	2.71	4.42
Whitefish racks	4	6.0	0.36	3.22	2.47
Alewife fishmeal	4	4.0	0.49	2.04	1.47
Control	8	2.9	0.02	1.14	1.76
Carp	2	42.8	3.70	9.84	29.32
Carp	4[c]	36.8	2.96	9.98	23.88
Sucker	2	10.8	1.23	1.70	7.88
Sucker	4[c]	9.5	1.20	1.81	6.39
Perch scraps	4	13.3	0.45	3.12	9.77
Whitefish racks	4	13.3	0.50	4.60	8.15
Alewife fishmeal	4	8.1	0.29	2.62	5.15

[a]Based on Aroclor 1254.
[b]One sample contaminated.
[c]Includes 2 mink added in January 1980 to replace mortalities.

2.2.23. Hornshaw et al. (1986)

In this study, mink were exposed by diet to five different doses of Aroclor 1254 (e.g., 10, 18, 32.4, 58.3, and 105 mg PCB/kg in diet) plus a control diet. Endpoints were mortality, organ weights, body weights, body weight changes, and feed consumption. Since this study was not chronic and did not focus on sensitive reproductive and developmental endpoints, it was not evaluated further for deriving TRVs. In addition, since concentrations of specific PCB congeners were not measured, it was not possible to calculate concentrations of TEQ from the results of this study.

2.2.24. Jensen et al. (1977)

In this study, female mink were exposed by diet to 3.3 mg PCB/kg (technical mixture was not stated) plus 3.3 mg DDT/kg, 11 mg PCB/kg alone, or 0.05 mg PCB/kg (as a "control") for 66 d and were mated with untreated males. Experimental details are insufficient to evaluate dosing methodology. Liver weights were greater in treated groups compared to controls. The number of implantation sites in breeding females was not different among groups, but the number of delivering females and the number of kits born per female significantly decreased in the 3.3 mg PCB/kg group. No kits were delivered in the 11 mg PCB/kg group. From this study, a LOAEL can be estimated to be 3.3 mg PCB/kg. However, since there were not sufficient data in this paper to review the methodology and quality of the study, this study was not evaluated further. In addition, since concentrations of specific PCB congeners were not measured in this study, it was not possible to calculate concentrations of TEQ.

2.2.25. Kakela et al. (2002)

In this study, two month old female mink were fed diets based on either Baltic herring or freshwater smelt for 147 d. One group of mink was fed smelt that contained Aroclor 1242 (approximately 1 mg PCB/d, or 3.5 mg/kg in feed, or 0.078 mg PCB/kg/d). Conversion of daily doses on a "per mink basis" to a "body weight normalized basis" was based on body weights provided in the paper for the PCB treatment group. The exposure to PCBs was from July to December (prior to mating) and then exposure to PCBs was terminated prior to breeding. From this study, a

LOAEL can be determined to be approximately 3.5 mg PCB/kg in feed (or 0.078 mg PCB/kg/d) based on decreased kit weight at 10 d for both male and female kits. However, this study has limited utility for deriving a TRV for mink due to a lack of dose-response characterization and a rather unusual exposure methodology. In addition, since concentrations of specific PCB congeners were not measured, it was not possible to calculate concentrations of TEQ from this study.

2.2.26. Kihlstrom et al. (1992)

In this study, female mink were exposed by diet to Clophen A50, Aroclor 1254 and fractions of these technical mixtures for 78–95 d. Conversion of daily doses on a "per mink basis" to a "body weight normalized basis" was based on body weights provided in the paper for each treatment group. Complete reproductive failure occurred with administration of either Clophen A50 or Aroclor 1254, in daily doses of 2 and 1.64 mg PCB/mink/d (or 1.99 and 1.28 mg PCB/kg/d), respectively. However, since the effects at these doses were relatively severe, the results of this study could not be used to develop TRVs.

2.2.27. Platonow and Karstad (1973)

In this study, mink were exposed by diet to PCBs at two different doses (0.64 and 3.57 mg PCB/kg, ww in feed) plus a control that also contained PCBs (0.3 mg PCB/kg) for up to 162 d. The source of PCBs in the diet was meat from cows that had been fed Aroclor 1254 for 24 h. Concentrations of total PCBs in mink tissues after exposure to different diets were reported (Table 6). Conversion of dietary concentrations to a daily dose was based on an ingestion rate of 0.15 kg food/mink/d and assumed a mean body weight of 1.0 kg. At a dose of 3.57 mg PCB/kg in diet (or 0.535 mg PCB/kg/d), there was 100% mortality of adult mink. At a dose of 0.64 mg/kg in feed (or 0.096 mg PCB/kg/d), only 1 of 12 female mink produced kits. All three of the kits died during the first day after birth. Based on these findings, the LOAEL was estimated to be 0.096 mg PCB/kg/d based on a food consumption rate of 0.15 kg food/d and a mink body weight of 1.0 kg (USEPA, 1995). This study reported poor reproduction of the experimental control mink, which makes interpretation of the results difficult. Thus, due to the lack of a true experimental control, lack of congener data, and lack of TEQ data, the results of this study were deemed to be inappropriate for development of TRVs.

2.2.28. Render et al. (2000)

In this study, 12-week-old-male mink were provided diets containing 0 (control) or 0.024 mg PCB 126/kg feed (2400 ng TEQ/kg feed) for up to 69 d to evaluate the effects of PCB 126 on the development of the baculum of growing mink. Researchers noted the development of lesions in the maxilla and mandible of all mink in the PCB 126 treatment group ($n = 20$). The presence of the lesion(s) also corresponded with the observation of one or more of the following symptoms: swelling of the upper and lower jaws, loose teeth, difficulty chewing, and marked porosity of maxillary and mandibular bone. Because the study was designed to examine baculum development and was not chronic in duration, the results could not be used for development of TRVs.

2.2.29. Render et al. (2001)

In a follow-up study to the PCB 126 experiment summarized above, 6- and 12-week-old mink were provided diets containing untreated feed (Control), 0.0024 mg TCDD/kg feed (2400 ng TEQ/kg feed), or 0.024 mg PCB 126/kg feed (2400 ng TEQ/kg feed) for up to 36 d to evaluate whether there was an age-related sensitivity to induction of mandibular and maxillary squamous epithelial proliferation in mink. Fifty (4/8) and 38 (3/8) percent mortality was observed in kits exposed to TCDD or PCB 126, respectively. Additional effects observed in mink exposed to TCDD or PCB 126 included, but were not limited to lethargy, a marked decrease in body weight gain, gross displacement of teeth, loss of alveolar bone, and mandibular osteolysis. Due to the short duration of exposure the results could not be used for development of TRVs.

Table 6
Mean PCB concentrations (±SD; mg/kg) in various tissues of mink fed rations containing ingredient(s) contaminated with PCBs (Table 2, Platonow and Karstad, 1973)

mg PCB/kg, ww, diet	*N*	Period	Blood	Brain	Kidney	Liver	Muscle	Heart
			Concentration (mg PCB/kg, ww)					
3.57	16	–	1.80±1.42	4.72±3.31	7.12±4.59	11.99±11.0	3.31±0.98	8.31±7.21
0.64	2[a]	[a]	0.71±0.01	0.52±0.01	1.20±0.28	1.10±0.08	0.62±0.12	1.10±0.28
0.64	4	0	0.12±0.02	1.36±0.45	1.74±0.66	1.23±0.10	0.97±0.51	1.12±0.45
0.64	4	1	0.10±0.05	0.60±0.26	1.12±0.87	0.87±0.15	0.83±0.43	1.60±0.71
0.64	4	2	0.24±0.03	0.90±0.13	1.86±0.43	1.21±0.05	0.77±0.19	1.25±0.26
0.64	2	3	0.06±0.07	0.33±0.01	1.09±0.04	1.33±0.16	0.64±0.09	1.11±0.08
Control (0.30±0.08)	8	–	0.12±0.08	0.32±0.09	0.29±0.07	0.39±0.14	0.23±0.15	0.35±0.14

Period (in months) after cessation of feeding PCB.
[a]Died during feeding the ration containing 0.64 mg PCB/kg, ww (ppm).

2.2.30. Restum et al. (1998)

In this study, adult ranch mink were exposed to carp collected from Saginaw Bay, Michigan at levels of 0%, 1%, 5%, or 13% in the diet with varying proportions of ocean fish trimmings. These diets were designed to result in nominal dietary concentrations of 0, 0.25, 0.50, and 1.0 mg PCB/kg diet, respectively. Mink were fed prior to and throughout the reproductive period for up to 557 d, although the experimental design had multiple exposure durations and dosing regimens to evaluate potential multigenerational effects and residual effects after being placed on clean feed. Conversion of dietary concentrations to a daily dose was based on an ingestion rate of 0.150 kg food/mink/d and assumed a mean body weight of 1.0 kg. Concentrations of PCBs were measured in liver (Table 7). Survival and reproductive performance were evaluated in both adults and kits produced during the study. Adverse effects were observed at the continuous dietary exposure to PCBs at a concentration of 0.25 mg/kg in feed (or 0.038 mg PCB/kg/d) including reduced kit body weight of the F_1-1 kits at 3 and 6 week. At this dose, no statistically significant adverse effects (relative to the control) were observed on kit body weight in the F_1-2 or F_2 kits, survivability of F_1-1, F_1-2, or F_2-generation kits, or litter size from parent groups P_1 (1992), P_1 (1993), or F_1-1. Since effects were seen at the lowest dose tested, a NOAEL was not identified in the study but likely lies somewhere between the control diet and a PCB concentration of 0.25 mg/kg in the diet. However, as discussed above for Heaton et al. (1995), the use of carp in the diet that were collected from a contaminated area also exposed the mink to co-contaminants. As a result, the assumption that the PCDDs, PCDFs, and PCBs were the only source of toxicity that contributed to the adverse effects observed in the study likely overestimates the contribution of these compounds. Thus, since the fish in the mink diets were field collected from contaminated areas with other co-contaminants likely present at toxicologically significant levels (see section reviewing Heaton et al., 1995), the results of this study are not appropriate to derive TRVs for TEQ because of potentially confounding impacts of other co-contaminants on mink in this study.

Table 7
PCB concentrations in pooled samples of liver from mink fed Saginaw Bay carp (modified from Table 12, Restum et al., 1998)

Generation	Dietary treatment (mg PCB/kg, ww)	Total PCB concentration (ng/g, ww)[a]			
		Number		Liver	
		Males	Females	Males	Females
P_1	0–0 (control)	4	8	68.4	71.9
	0.25–0	4	8	133	104
	0.5–0	4	7	220	226
	1.0–0	4	7	305	302
	0.25–0.25	4	8	622	980
	0.5–0.5	3	7	999	891
	1.0–1.0	3	8	1601	1572
F_1-1	0–0 (control)	4	7	24.0	117
	0.25–0	4	7	25.8	30.3
	0.5–0	4	6	32.0	37.2
	1.0–0	–	4	–	46.9
	0.25–0.25	4	5	637	634
	0.5–0.5	4	6	1458	961
	1.0–1.0	–	5	–	1474
F_2 and F_1-2[b]	0–0 (control)	4	8	20.5	15.0
	0.25–0	4	8	29.5	28.7
	0.5–0	4	–	39.5	–
	1.0–0	4	6	44.0	61.6
	0.25–0.25	4	8	640	92.4
	0.5–0.5	1 (F_1-2)	2 (F_2)	190	464
	1.0–1.0	3 (F_1-2)	1 (F_1-2)	171	181

Notation for generation and dietary treatment are as follows: F_1-1 refers to first generation kits whelped in the first year of the study, etc. Dietary treatments have two numbers which designate the PCB dose for the 1st and 2nd year of the study, respectively. For example, 0.25–0 refers to animals fed a diet containing 0.25 PCBs/kg feed during the first year of the study and uncontaminated feed during the second year of the study.

[a]Values in parentheses are less than the method quantitation limit of 10 ng/g.

[b]Each pooled sample consists of half F_2 and half F_1-2 kits unless noted otherwise in parentheses.

2.2.31. Shipp et al. (1998a, b)

These papers are companion papers to the Restum et al. (1998) study discussed previously. Concentrations of PCBs were measured in livers from male and female mink from each exposure group (Tables 8 and 9). In these papers, biochemical endpoints are discussed which are not relevant endpoints to derive TRVs for reproductive and developmental endpoints in mink.

2.2.32. Tillitt et al. (1996)

This paper is a companion paper to the Heaton et al. (1995) study discussed previously. In this paper, tissue residue data for Saginaw Bay carp, mink diets, and mink livers were presented (Table 10) and discussed. However, this paper does not represent a primary source of reproductive and developmental data that could be used to derive TRVs for mink.

2.2.33. Wren et al. (1987a, b)

In this study, mink were exposed by diet to PCBs (Aroclor 1254) and methylmercury (MeHg) singly and in combination for up to 182 d through a critical reproductive life stage. Conversions of concentrations in the diet to a daily dose were estimated based on a food consumption rate of 0.15 kg food/mink/d and a body weight of 1 kg (USEPA, 1995). At a dose of 1.0 mg PCB/kg of total PCBs in feed (or 0.15 mg PCB/kg/d), adverse effects were observed including a decrease in 6-week-old-male kit weights. Furthermore, at this dose, the number of kits per female, percentage of females whelped, and fertility of male mink were not affected. Concentrations of PCB in mink liver were reported from each treatment group and at multiple time points (Table 11). This study is strengthened

Table 8
Total PCB concentrations (ug PCB/kg, ww) in liver tissues of male mink exposed to PCBs *in utero*, via lactation, or through consumption of diets containing Saginaw Bay carp for up to 18 month (modified from Table 1, Shipp et al., 1998a)

Diet group	*N*	μg PCB/kg
P_1		
0.0–0.0	4	68.4
0.25–0.0	4	132.8
0.5–0.0	4	220.4
1.0–0.0	4	304.5
0.25–0.25	4	622.0
0.5–0.5	3	999.0
1.0–1.0	3	1600
F_1-1		
0.0–0.0	4	24.0
0.25–0.0	4	25.8
0.5–0.0	4	32.0
1.0–0.0	0	–
0.25–0.25	4	636.7
0.5–0.5	4	1458
1.0–1.0	0	–
F_1-2		
0.0–0.0	4	20.5[a]
0.25–0.0	4	29.5
0.5–0.0	4	ND[b]
1.0–0.0	4	44.0
0.25–0.25	4	639.9
0.5–0.5	1	190.1
1.0–1.0	3	170.6

Note: For PCB analysis, samples were taken from the indicated number of mink and pooled prior to analysis (Restum et al., 1998). Refer to footnotes for Table 7 for a description of diet group.

[a]These data were obtained by pooling tissues from both F_1-2 kits and the F_2 kits.

[b]ND, no data; although there were animals in these groups, total PCB concentrations were not determined.

Table 9
Total PCB concentrations (ug PCB/kg, ww) in liver tissues of female mink exposed *to PCBs in utero*, via lactation, or through consumption of diets containing Saginaw Bay carp for up to 18 month (modified from Table 2, Shipp et al., 1998a)

Diet group	*N*	μg PCB/kg, ww
P_1		
0.0–0.0	8	71.9
0.25–0.0	8	104.4
0.5–0.0	7	225.7
1.0–0.0	7	301.5
0.25–0.25	8	979.6
0.5–0.5	7	891.0
1.0–1.0	8	1572
F_1-1		
0.0–0.0	7	116.6
0.25–0.0	7	30.3
0.5–0.0	6	37.2
1.0–0.0	4	46.9
0.25–0.25	5	633.5
0.5–0.5	6	960.6
1.0–1.0	4	1474
F_1-2		
0.0–0.0	8	15.0[a]
0.25–0.0	8	28.7
0.5–0.0	4	39.5
1.0–0.0	6	61.6
0.25–0.25	8	15.0
0.5–0.5	2	463.6
1.0–1.0	1	180.7

Note: For PCB analysis, samples were taken from the indicated number of mink and pooled prior to analysis (Restum et al., 1998). Refer to footnotes for Table 7 for a description of diet group.

[a]These data were obtained by pooling tissues from both F_1-2 kits and the F_2 kits.

by the absence of adverse effects on 6-week-old-male kit weights at a lower dose of 0.5 mg PCB/kg, ww in feed (0.075 mg PCB/kg/d) in combination with 0.5 mg MeHg/kg, ww providing confidence that this study bracketed the true threshold with the doses tested. The only effect of this combined treatment of 0.5 mg PCB/kg, ww and 0.5 mg MeHg/kg, ww was a statistically significant reduction in the number of kits per female at 5 week. However, there was no PCB dose dependence for this endpoint since greater concentrations of PCBs (e.g., 1.0 mg PCB/kg in feed) did not affect this endpoint. Since the study considered dietary exposure during the sensitive and ecologically relevant time period of reproduction, the 0.075 and 0.15 mg PCB/kg/d doses were considered to be chronic dietary-based NOAELs and LOAELs, respectively. However, since concentrations of specific PCB congeners were not measured, it was not possible to calculate concentrations of TEQ from this study.

2.2.34. *Zwiernik et al. (2007a)*

In this study, female mink were exposed by diet to 0.0 (control), 0.24, and 2.4 μg/kg (ww) 2,3,7,8-tetrachlorodibenzofuran (TCDF) for up to 217 d. The authors analyzed the basal diet for AhR-active compounds and identified trace levels of contaminants resulting in a control diet concentration of 2 ng TEQ/kg (ww). Thus, the calculated dietary concentrations of TEQ incorporate this background concentration in the basal diet which results in concentrations of TEQ in these diets of 2.0 (control), 26, and 242 ng TEQ/kg (ww), respectively.

The study evaluated developmental and reproductive parameters including kit body weights. Kits were exposed *in utero* and throughout lactation. In addition, kits were maintained on their respective treatment diets until the conclusion of the study.

Among the more sensitive measurement endpoints, including kit weights, organ weights, relative organ weights, and organ and jaw histology, the only measurement endpoints that were significantly different from controls were kit body weights, when grouped by dose, sex, and time post birth (Table 12). In male kits, statistically significant weight reductions (17% and 26% decreases) were observed at 3 week in the 26 and 242 ng TEQ/kg (ww) diet treatment groups, respectively.

Table 10
Mammalian TEQ concentrations (ng/kg, wet weight) in diets and livers of mink fed diets containing carp from Saginaw Bay, Lake Huron, MI (Tables 2 and 4, Tillitt et al., 1996-data modified using 2005 WHO TEFs)

Compound	TEF	Mammalian TEQ (ng/kg, wet weight) in diet				Mammalian TEQ (ng/kg, wet weight) in livers			
		Control	10% carp	20% carp	40% carp	Control	10% carp	20% carp	40% carp
2,3,7,8-TCDD	1	0.0	2.0	3.0	7.0	1.0	21.0	34.0	50.0
1,2,3,7,8-PECDD	1	0.0	2.0	2.0	4.0	0.0	7.0	10.0	17.0
1,2,3,7,8-HXCDD	0.1	0.0	0.2	0.1	0.3	0.0	0.6	1.0	1.5
1,2,3,6,7,8-HXCDD	0.1	0.0	0.1	0.3	0.6	0.8	5.4	7.7	13.0
1,2,3,7,8,9-HXCDD	0.1	0.0	0.1	0.1	0.1	0.3	0.8	0.8	1.0
1,2,3,4,6,7,9-HPCDD	0.01	0.1	0.1	0.1	0.1	1.2	3.3	2.9	3.8
OCDD	0.0003	0.0	0.0	0.0	0.0	0.1	0.7	0.6	0.7
PCDD total		0.1	4.5	5.6	12.1	3.4	38.8	57.0	87.0
2,3,7,8-TCDF	0.1	0.0	0.2	0.4	1.2	0.0	0.2	0.2	0.3
1,2,3,7,8-PECDF	0.03	0.0	0.0	0.1	0.1	0.0	0.0	0.1	0.1
2,3,4,7,8-PECDF	0.3	0.0	1.2	1.8	4.2	0.6	51.0	96.0	147.0
1,2,3,4,7,8-HXCDF	0.1	0.0	0.1	0.2	0.3	0.0	3.3	7.3	13.0
1,2,3,6,7,8-HXCDF	0.1	0.0	0.1	0.1	0.2	0.0	2.5	4.9	7.1
1,2,3,7,8,9-HXCDF	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1
2,3,4,6,7,8-HXCDF	0.1	0.0	0.1	0.2	0.2	0.0	3.7	6.0	9.8
1,2,3,4,6,7,8-HPCDF	0.01	0.0	0.0	0.0	0.1	0.0	0.3	0.6	0.9
1,2,3,4,7,8,9-HPDCF	0.01	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
OCDF	0.0003	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
PCDF total		0.0	1.8	2.8	6.3	0.6	61.1	115.2	178.3
3,4,4′,5-TCB (81)	0.0003	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
3,3′,4,4′-TCB (77)	0.0001	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0
3,3′,4,4′,5-PECB (126)	0.1	0.5	8.8	21.0	41.0	11.5	121.0	170.0	328.0
3,3′,4,4′,5,5′-HXCB (169)	0.03	0.1	0.2	0.3	0.6	2.0	2.0	3.6	6.2
Non-*ortho*-PCB total		0.6	9.0	21.4	41.8	13.5	123.0	173.6	334.2
2′,3,4,4′,5-PECB (123)	0.00003	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.1
2,3′,4,4′,5-PECB (118)	0.00003	0.0	1.1	2.0	3.8	0.3	0.6	8.5	14.3
2,3,4,4′,5-PECB (114)	0.00003	0.0	0.0	0.1	0.1	0.0	0.1	0.3	0.4
2,3,3′,4,4′-PECB (105)	0.00003	0.0	0.4	0.7	1.2	0.1	1.6	3.2	5.4
2,3′,4,4′,5,5′-HXCB (167)	0.00003	0.0	0.0	0.1	0.1	0.0	0.2	0.3	0.6
2,3,3′,4,4′,5-HXCB (156)	0.00003	0.0	0.0	0.1	0.2	0.0	0.4	0.7	1.1
2,3,3′,4,4′,5′-HXCB (157)	0.00003	0.0	0.0	0.0	0.0	0.0	0.1	0.2	0.3
2,3,3′,4,4′,5,5′-HXCB (189)	0.00003	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1
Mono-*ortho*-PCB total		0.0	1.5	3.1	5.5	0.4	1.9	13.3	22.3
Grand total TEQ		0.7	16.8	32.8	65.7	17.9	225.9	359.0	621.8

Table 11
Summary of PCB concentrations (mean ± SE) in mink liver (Table 2, Wren et al., 1987a)

Day	Sex	N	PCB concentration (mg/kg)			
			Control	PCB	PCB/Hg	1/2PCB/Hg
0	M	2	0.04	NR	NR	NR
	F	2	<0.01	NR	NR	NR
78	F	1	NR	NR	2.3[a]	NR
118	M	2	0.08 ± 0.02	1.98 ± 0.03	1.21 ± 0.06	1.22 ± 0.13
161	F	2	0.13 ± 0.08	3.1 (1)	1.90 ± 0.28	1.10 ± 0.00
183	M	2	0.09 ± 0.04	2.80 ± 0.71	2.35 ± 0.64	1.50 ± 0.57
	M (kits)	2[b]	0.16 ± 0.09	1.75 ± 0.07	1.85 ± 0.21	2.00 ± 0.42

NR = Not reported.
[a]Died, all other mink euthanized.
[b]Each sample a composite of 2 pooled livers from 35 day old mink kits.
PCB concentrations in feed were 1 mg/kg for the PCB and PCB/Hg groups and 0.5 mg/kg in the $\frac{1}{2}$ PCB/Hg group.

In female kits, significant weight reductions were observed at week 24 in the 26 ng TEQ/kg treatment group (17% decrease) and at 6, 12, 24, and 36 week in the 242 ng TEQ/kg treatment group (15–20% decrease). Dietary exposure to 2,3,7,8-TCDF at 26 or 242 ng TEQ/kg (ww) diet did not significantly influence ($p < 0.05$) ecologically relevant measurement endpoints such as adult and kit survival, adult body weights, number of adult females bred per treatment, number of adult females that whelped per treatment, gestation length, number of kits whelped per female, number of kits live at birth, percent kits alive at birth, and kit survival to weaning.

The ecological relevance of kit weight reductions of this magnitude is unclear, and no statistically significant differences in any body weights were observed at adulthood, and overall, both dams and offspring appeared normal and healthy regardless of treatment (pers. comm..,

Table 12
Mean body weights (g) and sample sizes at seven time points for mink exposed to 2,3,7,8-TCDF (data from Zwiernik et al., 2007a)

		F_1 control	n	F_1 0.24 μg TCDF/kg	n	F_1 2.4 μg TCDF/kg	n
Female	Birth	8±1	(22)	7±2	(19)	8±1	(17)
	Week 3	115±18	(13)	101±13	(9)	98±25	(14)
	Week 6	284±65	(13)	256±47	(9)	227*±30	(13)
	Week 12	840±48	(10	810±192	(7)	700*±53	(11)
	Week 24	1170±130	(6)	970*±115	(5)	940*±90	(6)
	Week 36	1160±107	(6)	990±107	(4)	970*±123	(5)
	Adult	1000±196	(6)	760±29	(3)	850±39	(5)
Male	Birth	9±2	(24)	8±2	(25)	8±2	(18)
	Week 3	126±14	(16)	104*±10	(15)	94*±20	(13)
	Week 6	320±53	(16)	302±57	(15)	273±93	(13)
	Week 12	1190±134	(13)	1220±207	(12)	1040±154	(7)
	Week 24	1890±205	(10)	1840±422	(8)	1510±364	(5)
	Week 36	2160±229	(7)	2240±358	(8)	2080±156	(3)
	Adult	1660±142	(6)	1810±203	(7)	1650±123	(3)

*Significantly different ($p \leqslant 0.05$) from control; treatment exposures were 0 (control), 0.24, and 2.4 μg/kg 2,3,7,8-tetrachlorodibenzofuran (TCDF), or 2 (control), 26, and 242 ng TEQ/kg (ww) diet (2005 WHO TEFs); "Adult" refers to kits weighed at the termination of the experiment (age range 48–64 week); Note also that the adult weights are lower than 36-week-old kits due to decrease in feed ration prior to breeding season (pers. comm., Dr. S. Bursian).

Dr. S. Bursian). Furthermore, as kit survival was unaffected, it is likely that this transient decrease in kit body weight may not translate into population-level effects. Thus, for the purposes of this paper, the higher dose from this experiment, 242 ng TEQ/kg (ww in feed), is termed a conservative LOAEL that may be useful to provide an estimate of potential risk. However, as discussed in a later section for many TRVs, risk assessors should be aware that exceedance of this LOAEL may not lead to ecologically relevant adverse effects.

Data are not available for corresponding liver residue concentrations from this study.

3. Approaches to select or develop a recommended TRV

Taken together, there are over thirty individual mink feeding studies that were reviewed for their potential utility in deriving TRVs. A summary table is provided that indicates the rationale for inclusion of a particular study for further consideration or the rationale for exclusion of a particular study (Table 13). When faced with multiple toxicological studies, there are several possible approaches to derive TRV values. Either a single most probable value can be reported or a range of values can be presented. There are several methods for deriving a point estimate of the TRV, ranging from selection of a single best study to meta-analyses, interpolation, or averaging methods.

3.1. Selection of a single best study

Most commonly, a single study is selected that is the most definitive and defensible among all of the available studies. Such a study usually has doses that bracket a clear threshold for effect with some doses above and some doses below the threshold for the endpoints of interest. With the selection of a single study, other studies may provide additional information to supplement the primary study. Such an approach is commonly employed in the development of TRVs for ecological risk (USEPA, 2005b) and benchmarks for human health assessments as in the USEPA Integrated Risk Information System (IRIS; USEPA, 2006). In this paper, this is the approach that is followed in which the most defensible and definitive studies are selected from which a toxicity threshold was bracketed by the experimental doses.

3.2. Use of averaging methods or meta-analyses

There are several approaches that combine information from more than one study in determining the TRV. One such approach is to calculate an average (e.g., usually an arithmetic mean or geometric mean, as appropriate) of the most representative, highest quality studies. Such an approach assumes that the studies are of equal quality and should thus be weighted equally. Another approach that has been selectively employed to derive TRVs is meta-data analysis. This approach can be powerful when there is a substantial database of studies conducted in a nearly identical manner such as with a standard toxicity bioassay protocol. However, it would be inappropriate to combine results of studies that were conducted with substantially different methodology, exposure routes, exposure duration, strains, diets, amount of co-contaminants, etc., as is the case with most mink studies. In addition, meta-analyses often necessitate normalization to controls which can produce misleading results. For example, in a study evaluating mink exposure to 2 mg PCB/kg expressed as Aroclor 1242 (a commercial PCB mixture), mean litter size of exposed mink was 5.0 kits per mated female compared to a control value of 3.5 (Aulerich et al., 1977). In a second

Table 13
Studies reviewed for development of dietary toxicity reference values (TRVs) for mink exposed to PCDDs, PCDFs, and TEQ

	Data critical for TRV derivation[a]	Endpoints not relevant[b]	Doses too great[c]	TEQ data not available[d]	Exposure not chronic[e]	Co-contaminants[f]
Aulerich et al. (1971)				√		√
Aulerich et al. (1973)			√	√		√
Aulerich and Ringer (1977)				√		√
Aulerich and Ringer (1980)				√		
Aulerich et al. (1985)			√	√		
Backlin and Bergman (1992)		√		√	√	
Beckett et al. (2007, unpublished)			√			
Bergman et al. (1992a)		√		√	√	
Bergman et al. (1992b)		√		√		
Bleavins et al. (1980)			√	√		
Brunstrom (1992)		√		√	√	
Brunstrom et al. (2001)			√	√		
Bursian et al. (2006a, b)	√					√
Bursian et al. (2006c)	√					√
Edqvist et al. (1992)		√		√	√	
Hakansson et al. (1992)		√		√	√	
Halbrook et al. (1999)				√		
Heaton et al. (1995)					√	√
Hochstein et al. (1998)			√		√	
Hochstein et al. (1998)			√			
Hochstein et al. (2001)			√			
Hornshaw et al. (1983)				√		√
Hornshaw et al. (1986)				√	√	√
Jensen et al. (1977)				√	√	
Kakela et al. (2002)				√	√	
Kihlstrom et al. (1992)			√	√	√	
Platonow and Karstad (1973)			√	√		√
Render et al. (2000, 2001)					√	
Restum et al. (1998)			√			√
Shipp et al. (1998a, b)		√		√		√
Tillitt et al. (1996)						√
Wren et al. (1987a, b)		√		√		
Zwiernik et al. (2007a)	√					

Studies not applicable to mink dietary TRV derivation (e.g., non-relevant study species) not included in table.

[a]Data used in selection of mink dietary TRV for PCDDs/PCDFs.

[b]Endpoints are not ecologically relevant (e.g. enzyme induction and other subtle biochemical effects with no clear relevance to adverse population-level effects).

[c]Doses were too great and therefore responses were too severe (adult mortality) to be used to derive a TRV.

[d]Concentrations of TEQ could not be calculated from the available data.

[e]Duration was too short (defined as $\leqslant$100 days) to be representative of long-term or chronic effects.

[f]Potential or substantial influence of co-contaminants.

study that examined the effects of Aroclor 1242, mink exposed to 2.86 mg PCB/kg had a mean litter size of 5.0 kits per mated female compared to a control value of 6.6 kits per mated female (Kakela et al., 2002). Thus, in the first study, exposure to PCBs did not reduce mean litter size; in fact, it would appear that exposure to PCBs led to an increase of 43% in litter size. In the latter study, PCB exposure was associated with a 24% decrease in mean litter size. However, the normal mean litter size for control mink is approximately 4.0 kits per mated female for mink feeding studies conducted at Michigan State University (Aulerich and Ringer, 1980). Thus, while it is tempting to normalize to control values across multiple studies, the common sense comparison to a normal range of values is lost. Such a range may not always be available for a given lab and/or for a given endpoint, but should be considered when available. The result is that some effects may be improperly attributed to a contaminant-related effect. In the examples given above, both the 2 and 2.86 mg PCB/kg exposures would be associated with a mean litter size that is considered to be normal even though there were deviations from the control values.

There are several additional concerns with the use of meta-analysis to derive TRVs. These include selection criteria to screen studies in or out of the meta-analyses determination of confidence intervals and/or some measure of variability needs to be done with a meta-analysis. If data points are selected from several studies, there should be

Table 14
Dietary and tissue residue-based toxicity reference values (TRVs) from selected toxicity studies of mink exposed to TEQ (TRVs calculated using 2006 WHO TEFs unless otherwise noted)

Study	Dietary TRVs ng TEQ/kg feed (wet wt)		Tissue residue-based TRVs (mink livers) ng TEQ/kg liver (wet wt)		TEQ source
	NOAEL	LOAEL	NOAEC	LOAEC	
Bursian et al. (2006a)	12.1	50.4	–	–	PCBs, PCDD/Fs[a]
Bursian et al. (2006b)	–	–	50.2	189	PCBs, PCDD/Fs[a]
Bursian et al. (2006c)	56.6	–	77.8	–	PCBs, PCDD/Fs[b]
Heaton et al. (1995)	–	16.8	–	–	PCBs, PCDD/Fs[c,d]
Zwiernik et al. (2007a)	26	242	–	–	2,3,7,8-TCDF[e]

– Not available.
[a]Fish collected from Housatonic River, MA.
[b]Fish collected from Saginaw River, MI.
[c]Carp collected from Saginaw Bay, MI (some of the analytical data reported in Tillitt et al., 1996).
[d]Presence of co-contaminants likely.
[e]Lab diet supplemented with 2,3,7,8-TCDF.

enough overlap in exposure and results such that the selective inclusion or exclusion of one or a few data points would not result in drastically different TRV values. Said another way, the data from the studies should be internally consistent and not driven by one extreme value from one study. If meta-analyses are conducted, all of the data from all of the studies included should be used. The results of the selective meta-analysis can not be used to demonstrate internal consistency.

4. Recommended TRVs (dietary and tissue-residue based) for ERAs

As discussed in previous sections, individual mink studies were evaluated carefully to select the most relevant study or studies based on the following criteria: (1) close relatedness of the test species to the wildlife receptor of concern (in this case, only mink studies were reviewed); (2) chronic duration of exposure which included sensitive life stages to evaluate potential developmental and reproductive effects; (3) measurement of ecologically relevant endpoints; and (4) minimal impact of co-contaminants.

4.1. Recommended dietary TRV

For mink, the most relevant studies for TRV development were those of Bursian et al. (2006a–c) in which mink were exposed to an environmentally weathered mixture of PCBs, PCDDs, and PCDFs and Zwiernik et al. (2007a) in which mink were exposed only to 2,3,7,8-TCDF. Since these studies meet all of the stated criteria they were selected for use in deriving TRV values for TEQ. The strengths of these studies are that: (1) the test species is a wildlife species rather than a domesticated, laboratory species (note that this is a specific preference stated in the Great Lakes Water Quality Criteria documents; USEPA, 1995); (2) the duration of exposure was relatively long (greater than 120 d) and the exposure period included sensitive life stages to evaluate potential developmental and reproductive effects; (3) ecologically relevant endpoints were measured; and (4) there was no substantial impact of co-contaminants. Since the mink in the Bursian et al. (2006a–c) studies were fed field-collected fish, there is a possibility that potentially confounding effects may have resulted from the presence of co-contaminants. However, as there were no adverse effects on mink reproduction or kit survivability in the Saginaw River study (Bursian et al., 2006c), the impact of any co-contaminants from this study was negligible. In the Housatonic River study (Bursian et al., 2006a, b), effects were observed on kit survival at the greatest dose only. Thus, there is the possibility that co-contaminants were present and that the actual TRV might be greater in this study. Nevertheless, it is assumed for the purposes of this paper that all of the effects from this study (Bursian et al., 2006a, b) were due to concentrations of TEQ calculated from measured congeners. Mink TRVs from the reviewed toxicity studies are presented in Table 14 and Fig. 2 The TRVs listed from Heaton et al. (1995) are presented for comparison purposes only as these TRVs have been utilized extensively in the past. However, due to the presence of co-contaminants at toxicologically significant levels, it is not recommended to use this study as the basis of TRVs.

In summary, based on the most relevant studies (Bursian et al., 2006a–c; Zwiernik et al., 2007a), recommended TRVs range from 12.1 to 56.6 ng TEQ/kg (ww feed) for the NOAEL and from 50.4 to 242 ng TEQ/kg (ww feed) for the LOAEL. However, better resolution of the actual threshold level for effects and a better understanding of the differences in toxic potencies of specific congeners to mink may require additional studies that focus on congeners and relevant concentrations that match a particular site of interest as described below.

4.2. Recommended tissue residue TRV

The only studies that match the aforementioned criteria for TRVs and that have tissue residue levels in mink liver

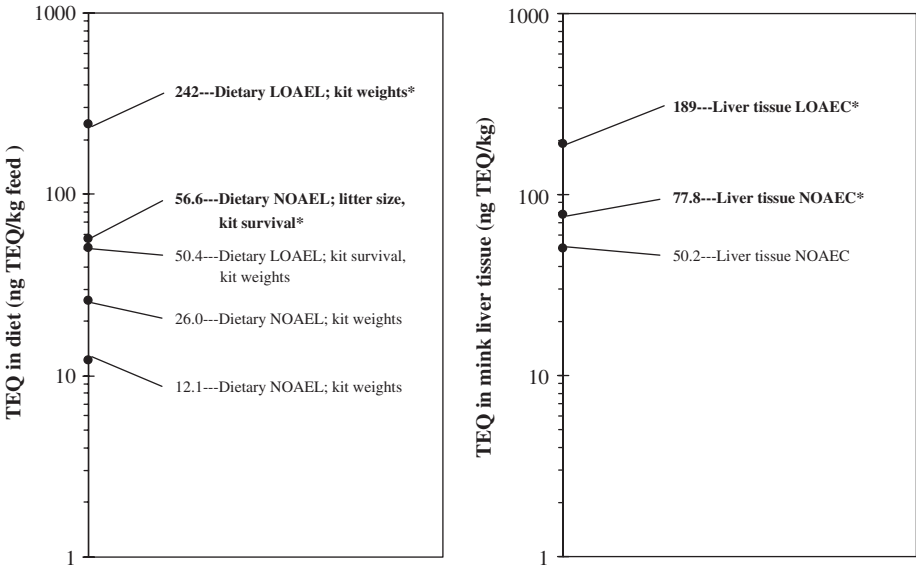

Fig. 2. Dietary and tissue residue-based toxicity reference values (TRVs) from reviewed toxicity studies for mink exposed to TEQ. *Recommended general and Tittabawassee River site-specific dietary and tissue residue-based TRVs for mink exposed to TEQ.

are those of Bursian et al. (2006a–c) in which mink were fed fish from the Housatonic River and the Saginaw River, respectively. Because residue concentrations in mink tissues were not reported in Zwiernik et al. (2007a), tissue residue-based TRVs could not be derived from this 2,3,7,8-TCDF exposure study. From the Housatonic River study (Bursian et al., 2006a, b), the NOAEC and LOAEC values are 50.2 and 189 ng TEQ/kg, ww liver, respectively. From the Saginaw River study (Bursian et al., 2006c), there were no adverse effects observed on reproductive and developmental endpoints at any of the doses. Therefore, the NOAEC from this study is from mink livers exposed to the greatest dose tested with a concentration in livers of 77.8 ng TEQ/kg, ww. While this was the mean concentration reported for 27-week-old kits, the body weight of the kits by this age is essentially the same as for adults (pers. comm., M.J. Zwiernik). Taken together, the NOAEC and LOAEC TRVs for TEQ in mink liver are 77.8 and 189 ng TEQ/kg, ww, respectively.

4.3. Example for the development of site-specific TRVs

Several of the studies reviewed in this paper evaluated the effects of environmentally weathered mixtures of PCBs, PCDDs, and PCDFs to mink. Each of these mixtures has a varying composition of congeners and thus, a variable potency. At each environmental site, different sources of PCDDs, PCDFs, and PCBs lead to different congener patterns (Wenning et al., 1992; Kannan et al., 1998; Hilscherova et al., 2003). While the concept of TEQ is an attempt to simplify the potency of a complex mixture that acts through an AhR-mediated mechanism of action, complex interactions are possible among measured mixture components and co-contaminants that may be present in an environmental sample (Tillitt et al., 1996; Giesy et al., 1997). Some of these complex interactions have the potential to increase or decrease the toxic potency of the mixture compared to that predicted by the calculation of TEQ (Aarts et al., 1995). In addition, it is well known that there are species-differences in the relative potencies of individual congeners (USEPA, 2003a). It is important to note that mammalian-based WHO TEFs are consensus values that may over- or under-predict the relative potency of a given congener for a given species.

An illustrative example of such a concept is the development of mink TRVs for the Tittabawassee River near Midland, MI. The river sediments, associated floodplains, and food web have been found to have elevated concentrations of PCDDs and PCDFs (Gale et al., 1997; Hilscherova et al., 2003). In dietary items for the mink from the Tittabawassee River, the congener groups that constitute the greatest proportion of TEQ are in the following order: PCDFs > PCDDs > non-*ortho*-substituted PCBs > mono-*ortho*-substituted PCBs (Fig. 3). Similarly, mink liver residue concentrations of congeners that constitute the greatest proportion of TEQ from this site are in the following order: PCDFs > non-*ortho*-substituted PCBs > PCDDs > mono-*ortho*-substituted PCBs (Fig. 4).

Among the considerations for which TRVs would be most appropriate, one might compare mink feeding studies that tested environmental mixtures from Saginaw River, MI (Bursian et al., 2006c) or from Housatonic River, MA (Bursian et al., 2006a, b). Of these studies, Bursian et al. (2006c) is particularly relevant to the Tittabawassee River because that study evaluated fish from the Saginaw River, which is immediately downstream of the Tittabawassee River. Fish from both the Saginaw and Tittabawassee Rivers have a generally similar composition of PCDDs and PCDFs. For example, PCDF congeners comprise an equal or greater proportion of the concentrations of TEQ

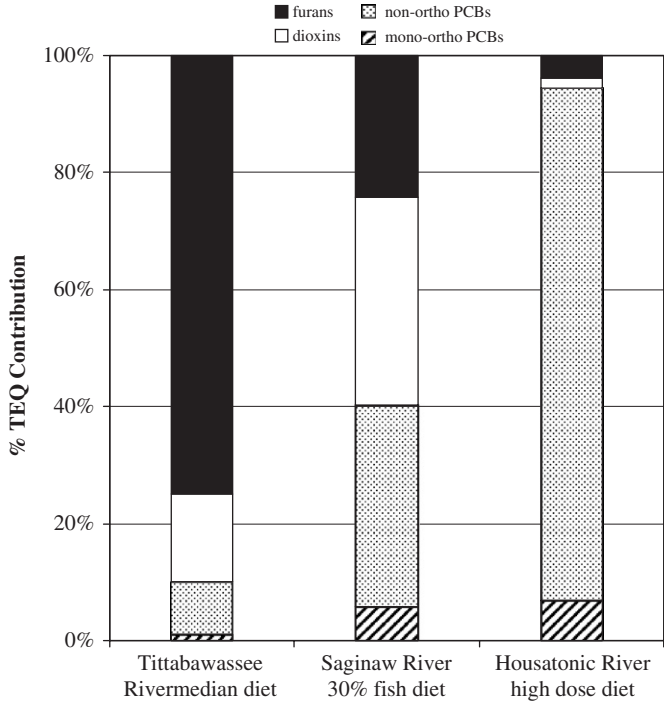

Fig. 3. Comparison of percent TEQ contribution in Tittabawassee River forage fish (Zwiernik et al., 2007b) and diets reported in Bursian et al. (2006a–c).

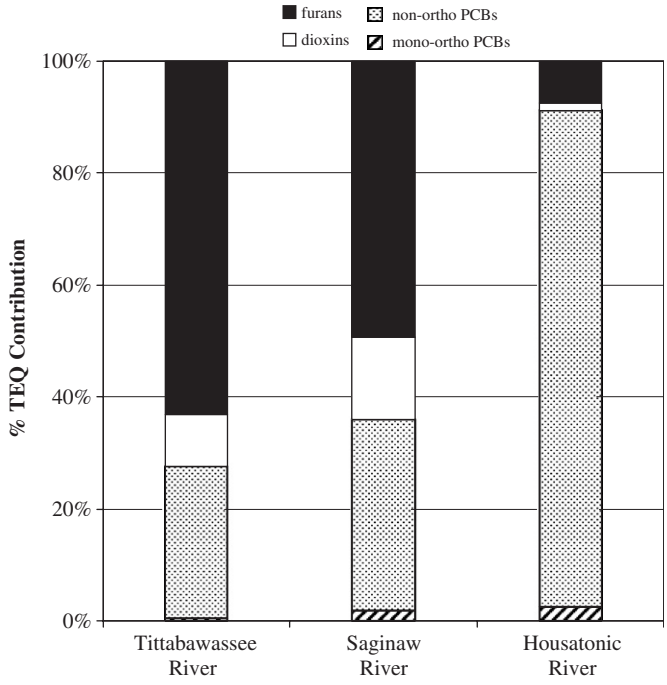

Fig. 4. Comparison of percent TEQ contribution in livers of Tittabawassee River mink (Zwiernik et al., 2007b) and ranch mink fed diets containing fish from Saginaw or Housatonic Rivers (Bursian et al., 2006a–c). Saginaw River diets composed of 30% Saginaw River carp and Housatonic River diets contained 50.4 ng TEQ/kg feed, ww.

in fish collected from the Tittabawassee River (GES/MDEQ, 2003) and from the Saginaw River (Bursian et al., 2006c). In addition, this study evaluated developmental and reproductive endpoints that are ecologically relevant. While a LOAEL was not determined from this study, the greatest dose tested (56.6 ng TEQ/kg, ww in feed) was clearly a NOAEL for reproductive and developmental endpoints. The only adverse effect noted was mild to moderate maxillary and mandibular squamous epithelial proliferation at the greatest dose tested. Currently, it is not thought that this endpoint by itself has any negative population level effects in mink (Beckett et al., 2005).

In comparison, the Bursian et al., study (2006a, b), using fish from the Housatonic River, established a NOAEL and a LOAEL of 12.1 and 50.4 ng TEQ/kg (ww feed), respectively. While these TRVs are both less than the NOAEL of Bursian et al. (2006c), the values are in a similar range. It should be noted that the differences between the Bursian et al. (2006a–c) studies may reflect differences in congener composition between studies. In the Housatonic River study, PCDFs comprised less than 5% of the total TEQ in the only diet from which toxicity was observed whereas PCDFs comprised approximately 24% of the total TEQ in the Saginaw River study (Fig. 3). Results from the study using only 2,3,7,8-TCDF (Zwiernik et al., 2007a) are consistent with the results of the Housatonic River and Saginaw River studies. The NOAEL is an intermediate concentration compared to the other studies and the LOAEL is approximately 4-fold greater than the LOAEL from the study conducted with mink fed fish from the Housatonic River in which most of the TEQ were due to PCBs.

The differences among these two studies likely reflect differences in congener potency and effects within a given mixture. Therefore, it is important to match the congener composition in dietary items from the site of interest (e.g., fish, crayfish, and small mammals along the Tittabawassee River and associated floodplains) as closely as possible to the congener composition in diets of a particular laboratory mink feeding study. Ideally, one could design a study in which mink would be fed dietary items collected from the Tittabawassee River or a congener mixture spiked into clean food that matches that observed in dietary items from the site. Based on existing data, the congener composition in the mink diet along the Tittabawassee River most closely matches the congener composition in the Bursian et al. (2006c) study with fish from the Saginaw River (Fig. 3). In addition, the congener composition in the liver of mink from the Tittabawassee River is very similar to the mink from the Saginaw River feeding study (Fig. 4). Thus, the best available TRVs would be from this study (Bursian et al., 2006c). However, since no adverse effects were observed on reproductive and developmental endpoints at any of the doses, only a NOAEL (56.6 ng TEQ/kg, ww, diet) could be determined from this study. For a LOAEL, the TRV derived for 2,3,7,8-TCDF (242 ng TEQ/kg, ww, diet; Zwiernik et al., 2007a) would be the best match as PCDF congeners represent the majority of TEQ in dietary items of mink foraging along the Tittabawassee River and,

in some samples, the 2,3,7,8-TCDF congener accounts for greater than 50% of the total TEQ.

4.4. TRVs used in recent ERAs and policy documents

Recently applied mink TEQ TRVs have been summarized from various environmental risk assessments (ERAs) and policy documents (Table 15). Each of these TRVs reflects site-specific considerations and the authors' best scientific judgment of the merits of the available toxicity studies at the time the TRV was derived. Careful examination of the references cited for each of these TRVs makes it apparent why these TRVs may not be the most appropriate for other sites (e.g., the Tittabawassee River watershed). The mink TEQ TRV selected by the Great Lakes Water Quality Initiative was based on a study by Murray et al. (1979) where the species studied was rat (USEPA, 1995). However, as discussed earlier, there are ample available toxicity studies with mink that negate the need to rely upon toxicity studies done with other species. The mink TEQ TRV developed for the Housatonic River ERA is based on a report to USEPA (Bursian et al., 2003, unpublished), which is the same study that was published as Bursian et al. (2006a, b) and described in section 5.12 of this paper. In this study, mink were fed a diet based on Housatonic River fish. This offers an appropriate site specific TRV for the Housatonic River but differences in congener profiles and specific congener potencies make this study much less applicable to other sites. The merits of the studies relied upon for mink TEQ TRV derivation in ERAs for sites including Hudson River, Sheboygan River and Bloomington have been discussed earlier in this paper (USEPA, 1998, 2000, 2005a). In summary, co-contaminants were likely present at toxicologically significant levels in each of these studies (see section reviewing Heaton et al., 1995). As a result, each of the studies has site-specific applications but is very limited in their application to other sites.

The mink TEQ TRVs of GES/MDEQ (2003) were developed specifically for the Tittabawassee River watershed. However, the site-specific dietary TRVs recommended in this paper are substantially different from the TRV recommended by GES/MDEQ (2003) in which they recommend a NOAEL TRV of 1.0 ng TEQ/kg ww, diet. Evaluation of more recent studies that were not available at the time that the GES/MDEQ (2003) TRV was developed suggests that more appropriate TRVs are now available. Specifically, the Bursian et al. (2006a–c), and Zwiernik et al. (2007a) studies tested doses greater than the GES/MDEQ TRV even in the control treatments with no reported adverse effects. These data suggest that the GES/MDEQ (2003) TRVs overestimate risks to mink exposed to TEQ from the Tittabawassee River. Furthermore, since there is uncertainty in the TEF values used to calculate the TEQ in mink due to PDCFs, it is likely that the actual TRV for the PCDF-dominated mixture in the mink diet from the Tittabawassee River is more similar to the TRVs reported by Zwiernik et al. (2007a).

5. Conclusions

Published toxicological studies involving mink exposed to PCDDs, PCDFs, PCBs, and related compounds were critically reviewed for their usefulness in deriving a TRV based on the following criteria: (1) close relatedness of the test species to the wildlife receptor of concern (only mink studies were reviewed in this paper); (2) chronic duration of exposure which included sensitive life stages to evaluate potential developmental and reproductive effects; (3) measurement of ecologically relevant endpoints; (4) availability of congener-specific data to calculate TEQ concentrations; and (5) minimal impact of co-contaminants.

Table 15
Dietary toxicity reference values in recent ecological risk assessments for mink exposed to TEQ (TRVs calculated using 1998 WHO TEFs unless otherwise noted)

Site or application	Dietary TRVs ng TEQ/kg feed (wet wt)		
	NOAEL	LOAEL	Primary literature cited
Great Lakes water quality initiative	0.5	4.5	Murray et al. (1979)
Housatonic River, MA, CT	16.1	68.5	Bursian et al. (2003, unpublished)[a]
Hudson River, NY	0.3	12.6	Tillitt et al. (1996)
Sheboygan River and Harbor, WI, aquatic ERA	0.9	22.0	Heaton et al. (1995)
Bloomington, IN ERA	4.6	18.0	Restum et al. (1998); Brunstrom et al. (2001)
GES/MDEQ Tittabawassee River ERA	1.0	–	Hochstein et al. (1998); Brunstrom et al. (2001); Heaton et al. (1995)

– Not reported.
[a]TRVs calculated using 2006 WHO TEFs.

Table 16
Recommended general and Tittabawassee River site-specific mink TEQ TRVs

Location	Dietary TRVs ng TEQ/kg feed (wet wt)		Tissue residue-based TRVs ng TEQ/kg liver (wet wt)	
	NOAEL	LOAEL	NOAEC	LOAEC
Tittabawassee River Watershed, MI	56.6	242	50.2–77.8	189
General (Non-Site-Specific)	12.1–56.6	50.4–242	50.2–77.8	189

Based on a review of over 30 such studies, very few of these studies meet all of the criteria to apply the results of these studies to derive ecologically relevant TRVs. This is understandable when one considers that each study was designed to answer specific questions and not necessarily to be used to develop TRVs that can be applied to other sites. However, many ERAs and public policy documents use the results of such studies. Awareness of the limitations of each study as it applies to a particular site is critical in order to make sound assessments of risk and ultimately, sound remedial decisions.

Recommended mink TRVs are summarized (Table 16). Recommended dietary TRVs for mink exposed to TEQ ranged from 12.1 to 56.6 ng TEQ/kg feed (wet weight) for the NOAEL and from 50.4 to 242 ng TEQ/kg feed (wet weight) for the LOAEL. Values of tissue residue-based TRVs ranged from 50.2 to 77.8 ng TEQ/kg liver (wet weight) based on the no observable adverse effect concentration (NOAEC) and 189 ng TEQ/kg liver (wet weight) based on the lowest observable adverse effect concentration (LOAEC). Selection of a TRV should be based on the closest match to the site-specific congener composition of PCDDs, PCDFs, PCBs, and related compounds. It should be noted that the basis of some of these TRVs are conservative endpoints such as a transient decrease in kit body weight, an endpoint for which the ecological relevance is unclear and may not translate into population-level effects. In addition, a few of these TRVs from field-exposed diets may include effects from co-contaminants. In these instances, the effect of co-contaminants was considered to be minor but nonetheless would tend to make the TRVs more conservative because of the assumption that 100% of the effects were due to TEQs. Therefore, as discussed earlier, risk assessors should be aware that exceedance of these conservative TRVs may not lead to ecologically relevant adverse effects. Additional future studies with congeners at ecologically relevant concentrations may be necessary to improve the resolution of site-specific TRVs.

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