



ATSDR Office of Community Health Hazard Assessment Toxic Equivalents Guidance for Dioxin and Dioxin-like Compounds

Citation:

[ATSDR] Agency for Toxic Substances and Disease Registry. 2019. Toxic Equivalents Guidance for Dioxin and Dioxin-like Compounds. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

Contents:

List of Tables	ii
List of Abbreviations	iii
Note	iii
1.0 Introduction	1
1.1 When to Use This Guidance	1
1.2 Topics Not Covered by This Guidance.....	2
1.3 Resources for Further Information	3
1.4 How to Use This Guidance	3
2.0 Background	3
2.1 What are TEQs and When to Use Them	4
3.0 Guidance Recommendation: Calculating TEQs for a Single Sample	6
3.1 General Approach for Calculating TEQs.....	6
3.1.1 <i>Environmental Samples without Non-detect Observations</i>	6
3.1.2 <i>Environmental Samples with Non-detect Observations</i>	7
3.2 Software Applications	8
3.3 Data Processing Steps	8
3.4 Special Considerations for Environmental Samples with Non-Detects	9
3.4.1 <i>Calculating TEQs for Environmental Samples with Fewer than Three Detected Congeners</i>	9

3.4.2	<i>Calculating TEQs for Environmental Samples with Three or More Detected Congeners....</i>	10
3.5	Calculating TEQs with ATSDR’s KMcalc Macro in Excel	11
3.6	Sensitivity Analyses	13
3.7	Quality Control Checks.....	15
3.8	Special Considerations	15
4.0	References.....	15
	Appendix A. Glossary	18
	Appendix B. Example TEQ Calculations in Microsoft Excel with KMcalc Macro	19
	Appendix C. Example Sensitivity Analyses	27

List of Tables

Table 1.	WHO 2005 Mammalian TEFs for Dioxin and DLCs	17
----------	---	----

List of Abbreviations

ADS	Associate Director for Science
AhR	aryl hydrocarbon receptor
ATSDR	Agency for Toxic Substances and Disease Registry
CAS	Chemical Abstracts Service
CV	comparison value
DCHI	Division of Community Health Investigations
DLC	dioxin-like compound
EDG	Exposure Dose Guidance
EMPC	estimated maximum possible concentration
EPC	exposure point concentration
ISM	incremental sampling methodology
KM	Kaplan-Meier
MRL	Minimal Risk Level
PAH	polycyclic aromatic hydrocarbon
PCB	polychlorinated biphenyl
PCDD	polychlorinated dibenzo-p-dioxin
PCDF	polychlorinated dibenzofuran
PHA	public health assessment
RfD	reference dose
RPD	relative percent difference
2,3,7,8-TCDD	2,3,7,8-tetrachlorodibenzo-p-dioxin
TEC	toxic equivalent concentration
TEF	toxic equivalent factor
TEQ	toxic equivalent
USEPA	U.S. Environmental Protection Agency
95UCL	95 percent upper confidence limit of the arithmetic mean
WHO	World Health Organization

Note

Since the time when this guidance was originally drafted, the Agency for Toxic Substances and Disease Registry (ATSDR) has developed an online tool that automates the calculations outlined in this document (i.e., ATSDR's EPC Tool). Health assessors are encouraged to use that tool when estimating EPCs for dioxin and dioxin like congeners. This guidance walks through steps to use a separate tool developed with an Excel macro. Health assessors may still choose to use this tool when they need to estimate a toxic equivalent (TEQ) value for a single sample.

This guidance mentions software applications by name (e.g., Microsoft Excel). Use of these trade names is for identification purposes only and does not constitute an endorsement of their use.

1.0 INTRODUCTION

When multiple chemicals in the same chemical class have sufficiently similar toxicological properties, toxic equivalents (TEQs) can be used to express the numerous chemicals' overall toxicity as a single value. This document presents the Agency for Toxic Substances and Disease Registry's (ATSDR's) guidance for calculating TEQs for dioxin and dioxin-like compounds (DLCs)—a class of related halogenated aromatic hydrocarbons that are specially handled in ATSDR public health evaluations.

This guidance presents ATSDR's preferred statistical procedures for calculating TEQs and specifically considers uncertainties associated with environmental sampling data that contain non-detect observations. Calculating TEQs with non-detect results is not a new concept, and many textbooks, articles, and other publications present different approaches for doing so. However, health assessors are expected to follow the computational approaches presented in this guidance, which address specific nuances of environmental data for dioxin and DLCs. Computational approaches other than those prescribed in this guidance should not be used, unless first approved by an Associate Director for Science (ADS) group.

Focus of This Guidance

This guidance walks health assessors through the process of calculating the TEQ for *a single sample* of dioxin and dioxin-like compounds.

Once TEQs have been calculated for all samples in an exposure unit following the procedures in this guidance, health assessors should then apply principles in other guidance documents (e.g., ATSDR, 2019a; 2020a) to determine what TEQ value to use as the exposure point concentration for the health evaluations.

This guidance supersedes ATSDR's 2012 guidance document on dioxins and DLCs (ATSDR, 2012). The current guidance incorporates more advanced statistical approaches to handling non-detect results and provides an Excel macro tool for easy computation.

1.1 When to Use This Guidance

During the public health assessment process, health assessors perform many activities, such as: developing a conceptual site model, evaluating exposure pathways, identifying exposure units, compiling and reviewing environmental data, and screening those data against health-based comparison values (CVs). ATSDR has developed other guidance to assist health assessors with these and many other steps in public health evaluations. Health assessors should follow the guidance in this document when working on sites with measured concentrations of dioxins and DLCs in any environmental media (e.g., soil, groundwater, surface water, fish tissue, outdoor air, and indoor air).

This guidance specifically outlines how to reduce data for an environmental sample analyzed for dioxin and DLCs into a single TEQ value, and should be applied prior to screening data against CVs and calculating exposure point concentrations (EPCs) for health evaluations. The guidance was not developed to address how to use and interpret the results for those evaluations. But briefly, health assessors should treat TEQs like concentrations of any other environmental contaminant. In this case, health assessors should evaluate TEQs using comparison values, health guidelines, and other resources specific to 2,3,7,8-tetrachlorodibenzo-p-dioxin (2,3,7,8-TCDD). Health assessors should also be mindful that several individually measured chemicals accounted for in the TEQ have their own CVs and/or health guidelines and should therefore be evaluated separately—meaning, these chemicals should both (1) be evaluated individually and (2) factored into the TEQ calculations. These chemicals currently include:

- 2,3,7,8-Tetrachloro dibenzo-p-dioxin (CAS No.: 1746-01-6)
- 1,2,3,6,7,8-Hexachloro dibenzo-p-dioxin (CAS No.: 57653-85-7)
- 1,2,3,7,8,9-Hexachloro dibenzo-p-dioxin (CAS No.: 19408-74-3)
- 2,3,4,7,8-Pentachloro dibenzofuran (CAS No: 57117-31-4)

Once health assessors have calculated a TEQ for each environmental sample with the methods described in this document, they should screen both the calculated TEQs and the measured concentrations of the four chemicals listed above against ATSDR recommended CVs for **both cancer and non-cancer** (e.g., environmental media evaluation guides [EMEGs] and cancer risk evaluation guides [CREGs]). If any of the individual sample TEQs or chemical measurements exceed applicable CVs in a potential or completed exposure pathway, health assessors must perform a more detailed risk evaluation.

Using Calculated TEQs

Health assessors can use the TEQs calculated following the procedures outlined in this guidance to evaluate both cancer and non-cancer effects. Evaluate the calculated TEQs like any other contaminant in the public health assessment process, by first screening the TEQ against the recommended cancer and non-cancer CV. If the calculated TEQ meets or exceeds a CV, health assessors should determine the EPC for that exposure unit and then estimate non-cancer hazards and cancer risks.

For these evaluations, health assessors estimate EPCs using the calculated TEQs for all environmental samples collected within an exposure unit. To do so, they should follow applicable ATSDR Exposure Dose Guidance (EDG) on how to define an exposure unit (ATSDR, 2020b) and on how to calculate EPCs (e.g., ATSDR, 2019a; 2020a). Health assessors would use these EPCs to calculate doses and then compare those doses to appropriate health guidelines (e.g., minimal risk levels [MRLs], reference doses [RfDs], cancer slope factors [CSFs]), like any other contaminant in the public health assessment process. For dioxin and DLCs, this means applying health guidelines for 2,3,7,8-TCDD to calculated TEQs, as well as applying available health guidelines for the four chemicals specified above.

1.2 Topics Not Covered by This Guidance

This guidance specifically addresses approaches for evaluating environmental sampling data for dioxin and dioxin-like compounds. This document is just one part of a larger series of ATSDR guidance documents that outline approaches for evaluating exposures to environmental contamination. Moreover, the guidance applies to the calculation of TEQs for dioxin and DLCs based on environmental sampling data collected with discrete, composite, and incremental methods. This guidance does not apply to:

- *TEQ calculations for Polycyclic Aromatic Hydrocarbons (PAHs).* ATSDR is currently developing separate guidance that applies specifically to PAHs, and that guidance will explain the toxicity weighting scheme health assessors should apply for this separate class of chemicals. Health assessors who need to address PAHs before the chemical-specific EDG is available should consult with their ADS group about preferred approaches.
- *Calculating EPCs from multiple environmental samples.* ATSDR has developed separate guidance (e.g., ATSDR, 2019a; 2020a) that describes the process of calculating EPCs with discrete sampling data and non-discrete sampling data. After using this guidance to determine TEQ values for individual samples in an exposure unit, health assessors should then apply the other guidance documents to determine EPCs on a TEQ basis for the entire exposure unit.

While health assessors can use this guidance to calculate TEQs for dioxin and DLC data collected via any sampling strategy, they must use sampling specific guidance when calculating EPCs. ATSDR has developed EDGs to calculate EPCs for data collected with 1) discrete sampling (ATSDR, 2019a) and 2) composite and incremental sampling methodology (ISM) (ATSDR, 2020a).

1.3 Resources for Further Information

This guidance was developed to make TEQ calculations for dioxin and DLCs a straightforward process. For additional background information on the general EPA approach followed in this guidance, health assessors are referred to the following sources:

- United States Environmental Protection Agency’s (USEPA’s) User Guide Uniform Federal Policy Quality Assurance Project Plan Template for Soils Assessment of Dioxin Sites (EPA, 2011).
- USEPA’s Recommended Toxicity Equivalence Factors (TEFs) for Human Health Risk Assessments of 2,3,7,8-Tetrachlorodibenzo-p-dioxin and Dioxin-like Compounds (USEPA, 2009).

Some health assessors may want to access additional resources for information on the advanced topics mentioned in this document. In those cases, health assessors should:

- Review Dr. Dennis Helsel’s paper on estimating TEQs for dioxin and DLCs. (Helsel DR. Summing nondetects: Incorporating Low-Level Contaminants in Risk Assessment. Integr Environ Assess Manag. 2010 Jul;6(3):361-366.)
- Consult with their ADS group for other resources. ATSDR recommends this option to ensure that all health assessors consistently rely on a common approach when calculating TEQs, rather than having health assessors individually seeking input from different (and perhaps inappropriate or conflicting) sources.

1.4 How to Use This Guidance

Health assessors will find all TEQ guidance in this document’s text. Appendix A includes a glossary of key terms, Appendix B presents example TEQ calculations, and Appendix C provides example sensitivity analyses. Additional information is provided in text boxes, as follows:

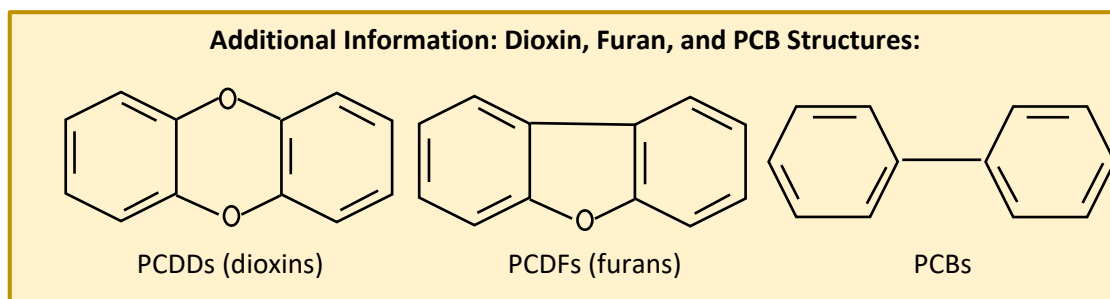
Key Point
Blue text boxes concisely summarize major elements of this TEQ guidance.

Additional Information
Yellow text boxes provide scientific background information on issues related to TEQ calculations.

2.0 BACKGROUND

The term “dioxin and DLCs” refers to a wide range of compounds in three chemical classes: polychlorinated dibenzo-p-dioxins (PCDD), polychlorinated dibenzofurans (PCDF), and coplanar polychlorinated biphenyls (PCBs). These three chemical classes represent structurally (e.g., “non-ortho-” and “mono-ortho-” substitutions) and toxicologically similar halogenated aromatic hydrocarbons (USEPA, 2011). The individual compounds within each class are referred to throughout the remainder of this document as “congeners.”

Congeners within these three chemical classes (e.g., PCDDs, PCDFs, and PCBs) have similar structures, but varying physical and chemical properties based on the position of chlorine atoms. PCDD and PCDF congeners have two benzene rings joined by a single or double oxygen bridge, while PCBs consist of two benzene rings joined by a carbon to carbon bond. Varying numbers of chlorines are attached to the carbons of the congeners, and congener names are assigned based on the position and number of those chlorine atoms. See the yellow box below for the basic structure of PCDDs, PCDFs, and PCBs.



While PCDDs, PCDFs, and PCBs comprise almost 420 individual congeners, only a small subset of these congeners are considered to have dioxin-like toxicity and are therefore included in TEQ calculations. Congeners with dioxin-like toxicity are those known to bind to the aryl hydrocarbon receptor (AhR) in humans and to elicit toxic or biochemical responses. Table 1 (found at the end of this guidance) lists these congeners—any PCDD, PCDF, or PCB congeners not listed in Table 1 should not factor into TEQ calculations.

Key Point: PCDDs, PCDFs, and PCBs with dioxin-like toxicity

Dioxin and DLC Groups	Total Number of Congeners	Number of Congeners with Known Dioxin-like Toxicity
PCDDs - Dioxins	75	7
PCDFs - Furans	135	10
PCBs	209	12

The term “dioxin and DLCs” refers to the seven PCDDs, ten PCDFs, and 12 PCBs with dioxin-like toxicity. Only these congeners are included in TEQ calculations.

2.1 What are TEQs and When to Use Them

TEQs provide a means for reducing measurements of numerous different congeners analyzed from one environmental sample to a single value that can be used for health assessment purposes. They are calculated to represent the overall toxicity of complex mixtures of PCDD, PCDF, and PBB congeners, relative to a single benchmark compound. In the case of dioxin and DLCs, the toxicity of each individual congener is weighted against that of 2,3,7,8-tetrachlorodibenzo-p-dioxin (2,3,7,8-TCDD), historically considered the most toxic member of these chemical classes. Most dioxins and DLCs are thought to have lower toxicity than TCDD, except for 1,2,3,7,8-pentachlorodibenzo-p-dioxin, which is considered approximately equal in toxicity to 2,3,7,8-TCDD. Measured concentrations and detection limits of each

congener and the congeners' corresponding weighting factors are used to characterize the toxicity of dioxin and DLCs.

To calculate TEQs, toxic equivalent factors (TEFs) have been assigned to each dioxin and DLC congener. TEFs are assigned by comparing the relative toxicity of individual congeners by “order-of-magnitude” to that of 2,3,7,8-TCDD, based on detailed scientific review of chemical structures and toxicological databases. Currently, values range from 0.00003 to 1.0. A TEF of 0.1 indicates that the congener is 1/10th as toxic as 2,3,7,8-TCDD, whereas a TEF of 1.0 indicates that the congener is equally as toxic as 2,3,7,8-TCDD.

TEF consensus values developed by the World Health Organization (WHO) and accepted by ATSDR are shown in Table 1, at the end of this guidance (Van den Berg et al., 2006). These are TEFs for mammalian species and should be used for all TEQ calculations by health assessors to support public health evaluations. The specific TEF values used represent order-of-magnitude relative toxicity increments, expressed on a logarithmic scale (USEPA, 2009). WHO periodically reviews and updates these values, and ADS groups will inform health assessors of any notable future updates. The most recent TEFs were developed in 2005. Note that EPA recommends that TEFs be used for all effects mediated through aryl hydrocarbon receptor binding by dioxin and DLCs, including cancer and noncancer effects (EPA, 2009).

To calculate the TEQ for an environmental sample, the concentrations of individual congeners are first multiplied by their respective TEFs to produce congener-specific toxic equivalent concentrations (TEC). The individual TECs are then summed to obtain a total TEQ for the sample. The two equations used to calculate TEQs are shown in the blue box below.

Key Point: How to calculate a TEQ

Equation 1:

$$TEC_i = x_i \times TEF_i$$

Equation 2:

$$TEQ = \sum_{i=1}^k (TEC_i)$$

Where:

- TEC_i is the toxic equivalent concentration of the i^{th} individual congener in the sample.
- x_i is the measured concentration of the i^{th} individual congener in the sample.
- TEF_i is the individual toxic equivalent factor.
- TEQ is the toxic equivalent for the environmental sample.
- k is the number of congeners that make up the TEQ.

While TEQs provide a useful technique to reduce measured concentrations of numerous congeners into a single value, health assessors should be familiar with the following underlying assumptions that went into developing TEFs, before applying them to public health evaluations (Safe, 1990).

1. The compounds exert toxicity through a common receptor-mediated mechanism.
2. The effects of the individual congeners within the mixtures are additive.
3. The individual congeners have similarly shaped dose-response curves.

Overall, the TEQ framework provides a scientifically justified, health protective, and widely accepted method for evaluating toxicity of mixtures of dioxin and dioxin-like compounds (NAS, 2006). However, the method has inherent uncertainties and may not capture the true health risks of all congener exposure scenarios.

3.0 GUIDANCE RECOMMENDATION: CALCULATING TEQS FOR A SINGLE SAMPLE

This section presents ATSDR's preferred approach to calculating TEQs for evaluating health implications of exposures to dioxin and DLCs. Since health assessors will frequently encounter data sets that include non-detect results, this section focuses largely on how to handle "censored data"—a term commonly used to describe data sets including non-detect observations. Health assessors should review this section and the example calculations shown in Appendix B and C before applying this guidance.

Key Point: General strategy for calculating TEQs with non-detect results

To calculate TEQs for an environmental sample, health assessors should:

- Review and prepare the data for TEQ calculations (Section 3.3).
- Calculate the TEQ using ATSDR's KMcalc macro function in Excel (Section 3.5).
- Complete sensitivity analyses to evaluate the influence of different methods to handle non-detects and rejected results (Section 3.6).

3.1 General Approach for Calculating TEQs

The general approach to calculate TEQs differs for environmental samples without non-detect observations (see Section 3.1.1) and with non-detect observations (Section 3.1.2). However, the same software applications (Section 3.2), data processing steps (Section 3.3), use of ATSDR's KMcalc Excel tool (Section 3.5), sensitivity analyses (Section 3.6), and quality control checks (Section 3.7) apply to both scenarios. The remainder of this section describes the general approach for data sets without and with non-detects.

3.1.1 Environmental Samples without Non-detect Observations

To illustrate the general TEQ computational approach shown in Section 2.1 and applicable to environmental samples without non-detect observations, this section presents a hypothetical soil sample in which seven DLCs were measured and all had detected concentrations. Refer to the text box later in this section for the specific values. To calculate the TEQ for this sample, the detected results for the seven dioxin congeners are first multiplied by applicable TEFs to calculate TECs, and then summed to calculate the TEQ for the sample.

In this example, the TEQ for the soil sample is 2.51 ng-TEQ/kg. The health assessor would first compare this TEQ concentration to the CV for 2,3,7,8-TCDD. If the TEQ concentration exceeds the CV, the TEQ would then be used in the health effects evaluation, following guidelines for 2,3,7,8-TCDD.

Note that the TEQ calculated this example is presented in units of ng-TEQ/kg. This is consistent with ATSDR's preferred approach for reporting the TEQ concentrations, as explained in the blue box below. Health assessors should report all calculated TEQs following this naming convention.

Additional Information: Example TEQ Calculations with Detected Results from a Single Soil Sample

Dioxin Congener	Analytical Result (ng/kg)	TEF ^a (unitless)	TEC ^b (ng/kg)
2,3,7,8-TCDD	1.7	1	1.7
1,2,3,7,8-PeCDD	0.18	1	0.18
1,2,3,4,7,8-HxCDD	0.26	0.1	0.026
1,2,3,6,7,8-HxCDD	2.1	0.1	0.21
1,2,3,4,6,7,8-HpCDD	25	0.01	0.25
1,2,3,7,8,9-HxCDD	0.77	0.1	0.077
OCDD	220	0.0003	0.066
2,3,7,8-TCDD TEQ^c = 2.51 ng-TEQ/kg			

Notes:

- a. 2005 WHO TEFs as presented in Table 1 at the end of this guidance.
- b. TEC for each congener = (Analytical Result) x (TEF).
- c. 2,3,7,8-TCDD TEQ for sample = Sum of TECs for the seven detected congeners.

With this simple illustration, health assessors should recognize that TEQs are weighted sums of the individual congeners, with weights based on each congeners' relative toxicity to 2,3,7,8-TCDD. Another way to consider the result from the example above is that exposure to the measured concentrations of the seven different DLC congeners is essentially equivalent to exposure to 2.51 ng/kg of 2,3,7,8-TCDD.

Key Point: Reporting Calculated TEQs

Chemical concentrations are typically reported in units of mass of the chemical per mass or volume of the sampled media (e.g., air, soil, sediment, groundwater, etc.). Health assessors should report all calculated TEQs in units of measurement that include "TEQ" in the unit's numerator. This is particularly important so that TEQ results are not confused with congener-specific results. Several examples of this TEQ naming convention are shown below.

- Soil and sediment: pg-TEQ/kg and ng-TEQ/kg
- Indoor and outdoor air: pg-TEQ/m³ and ng-TEQ/m³
- Groundwater and surface water: pg-TEQ/L and ng-TEQ/L

This hypothetical example is only shown to illustrate the TEQ computational approach. In most cases, health assessors will encounter samples in which some dioxin and DLC congeners are detected and others are not. The following section reviews general approaches for those scenarios.

3.1.2 Environmental Samples with Non-detect Observations

Non-detects are valid measurements in which the concentration of the contaminant of interest is too low to measure with confidence. Sampling reports typically present non-detects as being less than a specified limit (e.g., "<25 ng/kg"), with that limit being either a method detection

limit or a quantification limit. In these cases, health assessors can only conclude that the actual contaminant level is somewhere between zero and the specified level. Nonetheless, non-detects still provide useful information and should be included in TEQs.

ATSDR's preferred approach to calculate a TEQ from an environmental sample with non-detect observations follows Dr. Dennis Helsel's approach of Kaplan-Meier (KM) estimation (Helsel, 2012). This non-parametric approach uses an unbiased framework for handling non-detect observations in TEQ calculations. Specific considerations for environmental samples with non-detect results are summarized in Section 3.4.

3.2 Software Applications

Health assessors should use the Excel macro function (KMcalc) developed by ATSDR to calculate TEQs for environmental samples with dioxin and DLC data. This macro function will calculate TEQs for samples without non-detects following the process described in Section 3.1. For samples with non-detect results, the macro will impute congener concentrations for non-detects with the KM method (where appropriate) and process data qualifiers per ATSDR procedures. In one rare exception (i.e., for samples with fewer than three detected results), the KMcalc macro cannot be used to calculate TEQs and health assessors are directed to Section 3.4.1. Details regarding how the TEQs are calculated for non-detects are presented in Section 3.4, instructions on how to use the KMcalc macro are provided in Section 3.5, and the process is demonstrated with an example data set in Appendix B.

Health assessors may notice that USEPA provides publicly available Microsoft Excel spreadsheet tools that calculate TEQs for dioxin and DLCs with the KM method. These tools, however, were designed to accommodate incremental sampling data (e.g., with inputs for triplicate sampling results) and therefore do not apply to all sampling scenarios; and other aspects of the TEQ calculations do not align with ATSDR's guidance (e.g., how to process environmental samples when the highest result is a non-detect). Health assessors are therefore encouraged to use ATSDR's calculation tool for determining EPCs.

3.3 Data Processing Steps

Regardless of whether a given sample includes non-detect results, health assessors should always complete the following two data processing steps:

- *Perform a data quality review.* Before proceeding with TEQ calculations, health assessors should review their environmental sampling data to confirm the data are of a known and high quality and meet the data quality objectives for the health evaluation in question. Any R-qualified (e.g., rejected) results should be removed from the data set. TEQs should never be calculated with R-qualified data.

Health assessors should, however, take note of any R-qualified values that are removed from a data set to conduct subsequent sensitivity analyses. These analyses should be completed to ensure that the decision to exclude R-qualified data does not substantially impact the final TEQ. Sensitivity analyses are described in Section 3.6 and demonstrated in Appendix C, and should be completed whenever R-qualified results are removed.

- *Identify and process duplicate samples and replicate analyses.* Two commonly used approaches to characterize measurement precision in environmental samples are through analyzing

duplicate samples (i.e., two samples collected from the exact same place and time) or conducting replicate analyses of a single environmental sample (i.e., two separate laboratory analyses of the same sample). Health assessors should not use both measurements from duplicate samples or replicate analyses in health evaluations, because doing so artificially assigns greater weight to these samples or analyses. Instead, health assessors should reduce results from duplicate samples or replicate analyses into the result for a single sampling event.

For dioxin and DLCs, the preferred approach is to calculate TEQs for each duplicate sample or each replicate analysis, and then use the average of these TEQs to represent the single sampling event in CV comparisons and health evaluations. As an example, assume a laboratory received a single soil sample and analyzed that sample in replicate (i.e., the laboratory generated two different profiles of congener concentrations for the same sample). To determine the TEQ for this sample, health assessors should first calculate the TEQ for each replicate following the procedures outlined in this guidance; and then average the two TEQ values from the two replicates to determine the TEQ of the overall sampling event. This procedure should be applied for both duplicate samples and replicate analyses.

3.4 Special Considerations for Environmental Samples with Non-Detects

ATSDR's preferred approach to calculating TEQs for dioxin and DLCs for a single sample that includes non-detect results is to use the KM method. However, this method does not perform well for data sets with very few detected results (Helsel, 2010) and therefore should only be used to calculate TEQs for environmental samples with at least three detected congeners. For example, if a laboratory analyzed an environmental sample for 17 DLC congeners, the KM method should only be applied when at least three of those congeners have detected concentrations.

Section 3.4.1 describes the computational approaches for environmental samples with fewer than three detected congeners, and Section 3.4.2 describes the computational approaches for environmental samples with three or more detected congeners.

3.4.1 Calculating TEQs for Environmental Samples with Fewer than Three Detected Congeners

For an environmental sample with fewer than three detected congeners, the KM method should not be used. In this case, the total TEQ in the sample is uncertain, and will range between the sum of detected congeners and their respective TEFs and the sum of detected congeners times their respective TEFs, plus the detection limits of the non-detected congeners times their respective TEFs. The KMcalc spreadsheet and macro cannot be used to calculate a TEQ for such samples.

To calculate TEQs for samples with fewer than three detected congeners, health assessors should follow the calculations outlined in Section 2.1 (i.e., Equations 1 and 2). In these calculations, the maximal TEQ value is the detection limit and it is calculated from the sum of (1) the measured concentrations for the fewer than three congeners that were detected multiplied by their respective TEFs and (2) the full detection limit for the congeners that were not detected multiplied by their respective TEFs. Health assessors should find congener-specific detection

limits in the environmental sampling reports that present the measurements for dioxin and DLCs.

The resultant TEQ value should be U-qualified if TECs from non-detect records represent 80% or more of the sum of the TECs from all congeners (U-qualified data indicate that a sample was analyzed for a contaminant but the contaminant was not detected, and the concentration reported with the qualifier is the quantitation limit). For example, consider the sample TEQ calculated in Section 3.1.1, in which the resultant TEQ was 2.51 ng-TEQ/kg. For this example, suppose that only one of the congeners was detected rather than all of them. If only OCDD was detected, the resultant TEQ would be U-qualified because the OCDD TEC contributed only 2.6% of the sum of the TECs ($0.066 \text{ ng/kg} \div 2.51 \text{ ng/kg} = 2.6\%$) and the remaining non-detect congeners contributed 97.4% of the sum. If, however, only 2,3,7,8-TCDD was detected, the resultant TEQ would not be U-qualified because 2,3,7,8-TCDD contributed 68% of the resultant TEQ and the remaining congeners contributed only 32%.

Health assessors should also examine whether the one or two detected congeners were measured at concentrations of potential health concern. If the detections were for any of the congeners listed in Section 1.1, health assessors should compare the measured concentrations of these congeners to their corresponding CVs.

3.4.2 Calculating TEQs for Environmental Samples with Three or More Detected Congeners

For environmental samples with at least three detected congeners, TEQs should be calculated with the KM method. However, in cases where the lowest TEC or the highest TEC is a non-detect, special considerations apply to the TEQ calculation. The KMcalc macro will account for these considerations, described here:

An environmental sample with the lowest TEC reported as a non-detect: If the lowest TEC across all congeners is a non-detect, the non-detect observation will be replaced with a detected concentration equal to the value of the detection limit, before calculating TEQs. If multiple non-detect records have the lowest TEC, this substitution should be performed for only one of the congeners. If multiple records with the lowest TEC are a mixture of detects and non-detects, no changes are required.

An environmental sample with the highest TEC reported as a non-detect: In the unlikely event that the highest TEC across all congeners is a non-detect, the TEQ will be calculated while considering this value a detected concentration at the detection limit. If multiple non-detect records have the highest TEC, this substitution should be performed for only one of the congeners. If multiple records with the highest TEC are a mixture of detects and non-detects, no changes are required.

Similar to the case of one or two detected congeners, the resultant TEQ will be presented as a non-detect with a U-qualifier if TECs from non-detect records represent 80% or more of the sum of the TECs from all congeners. When calculating these percentages, health assessors should use the original detect or non-detect status of each congener record. Any changes made to records associated with the highest or lowest TECs for the KM method calculations should not be considered when evaluating whether the resultant TEQ is U-qualified.

3.5 Calculating TEQs with ATSDR's KMcalc Macro in Excel

As described in Section 3.1, health assessors can use ATSDR's KMcalc macro to calculate TEQs for environmental samples with and without non-detect results. This single spreadsheet macro function applies Equations 1 and 2 (as shown in Section 2.1) for samples without non-detects and the KM method for samples with non-detects. In one rare exception (i.e., for samples with fewer than three detected results), the KMcalc macro cannot be used to calculate TEQs and health assessors are referred to Section 3.4.1.

The macro can be used in two ways: If health assessors are working with an existing Excel workbook that already contains dioxin and DLC analytical results, they can simply load the KMcalc macro into the workbook for TEQ calculations; or health assessors can transcribe or copy their sampling data into an Excel file that already has the macro loaded. Instructions on how to load the macro to Excel are provided in Appendix B, with options for "general users" and "advanced users." Health assessors who have questions about applying the macro should contact their ADS group.

The macro function calculates a TEQ and assigns a qualifier based on three inputs: concentrations (which may include detected values and detection limits for non-detect results), qualifiers (i.e., U-flags and J-flags), and TEFs. Health assessors must confirm that their Excel spreadsheets include these necessary data inputs. Refer to the example shown in Appendix B for instructions on how these three main inputs must be formatted for use with the KMcalc macro. The following list describes important considerations for each input:

- Congener Concentrations:
 - All inputs for congener concentrations must be numeric values. Symbols, letters (e.g., "<" or lab qualifiers in the concentration field), and extraneous blank spaces will result in formula errors.
 - All congener concentrations must be in the same units (e.g., ng/kg or ng/m³), which is usually the case for environmental sampling data; meaning, laboratories typically will not use two different units of measurement when reporting concentrations for congeners within a single environmental sample. These units, however, should not be entered with congener concentrations in the spreadsheet macro; the KMcalc function will return an error when units are typed into the concentration field. The calculated TEQ will be in the same units as the individual congener concentrations.
 - Congener data do not need to be sorted.
- Concentration Qualifiers:
 - TEQ calculation spreadsheets should include any U, J, or UJ qualifiers assigned by the analytical laboratory, as the KMcalc macro has unique computational approaches for U-qualified and J-qualified data. These qualifiers must be entered into the spreadsheet as capital letters and any extraneous spaces in the qualifier

data will result in computational errors. (Note: Appendix B.1 explains an exception to this guidance for the “general user” calculations.)

- A U qualifier (entered as a capital letter) must be entered for all non-detect observations. This is because the computational algorithm recognizes the “U” as a non-detect observation. Use of any other letter (or letters) for non-detects will result in erroneous calculations. Therefore, in cases where a laboratory uses other letters or conventions to represent non-detects (e.g., some laboratories might report them as “ND” or “<MDL”), health assessors must replace those notations with U when using the KMcalc macro. Similarly, a J qualifier (entered as a capital letter) must be entered for all estimated observations. The KMcalc macro only recognizes “J” as an estimated value. Health assessors may encounter data with a wide range of data qualifying conventions, including use of multiple qualifiers for a single measurement. Any questions regarding data qualifiers in the context of KMcalc should be directed to ADS groups.
- Toxic Equivalent Factors – TEFs: Health assessors must enter the congener-specific TEFs from Table 1 (at the end of this guidance document) into the spreadsheet before calculating TEQs. (Note: Appendix B.1 explains an exception to this approach for the “general user” calculations.)

Once data are properly entered to Excel, health assessors can use the KMcalc function in just the same way as any other Excel function (e.g., average, min, max, etc.). The KMcalc function can be selected through Excel’s Insert Function button or by entering the formula function directly into an empty cell =KMcalc(Conc,Qualifiers,TEF,ResultType). Both processes are described in detail in Appendix B.2 and the required function inputs are presented in the blue box below.

For every environmental sample requiring a TEQ calculation¹, health assessors should use KMcalc for two purposes: (1) to calculate the TEQ and (2) to determine whether the TEQ result should have a qualifier. The KMcalc macro will output a TEQ value when the health assessor enters “Sum” in the “ResultType” field of the equation, and the KMcalc macro will output a qualifier when the health assessor enters “Qualifier” in the “ResultType” field of the equation. (Note: The “general user” example in Appendix B.1 presents a simplified way for obtaining the TEQ results.) There is only one scenario possible for the KMcalc-assigned qualifiers. If the contribution of TECs to the TEQ from congener results that are U-, J-, or UJ-qualified is greater than 50 percent, the KMcalc function will return a J-qualifier. The KM-calc function does not assign U-qualifiers.

¹ In one rare exception (i.e., for samples with fewer than three detected results), the KMcalc function cannot be used to calculate TEQS and health assessors are directed to Section 3.4.1.

Before using the outputted TEQ for public health evaluations, health assessors should first complete the sensitivity analyses described in Section 3.6.

Key Point: KMcalc Function

The KMcalc function is as follows:

=KMcalc(Conc,Qualifier,TEF,ResultType)

Where:

- Conc is the range of spreadsheet cells that have the measured concentrations of detected congeners or full detection limits of non-detect congeners. Remember to only consider congeners included in the TEQ.
- Qualifier is the range of spreadsheet cells with data qualifiers (U, J, UJ, or blank) for the corresponding congener concentrations.
- TEF is the range of spreadsheet cells with the congener-specific toxic equivalent factors.
- ResultType is the desired output:
 - If “Sum” is entered, the KMcalc function will output the TEQ.
 - If “Qualifier” is entered, the KMcalc will output the qualifier that should be assigned to the calculated TEQ.

As Appendix B.2 explains, advanced users must run the KMcalc function twice—one for each ResultType—to obtain the estimated TEQ and TEQ-qualifier. That means that the formula must be entered once with the ResultType set to “Sum,” and then again with the ResultType set to “Qualifier.” However, general users can follow the procedures in Appendix B.1, which are more straightforward.

3.6 Sensitivity Analyses

Health assessors should complete two sensitivity analyses to ensure that the initial TEQ estimate is not heavily influenced by the KM method used to handle non-detects or by exclusion of R-qualified results. These analyses are described below and demonstrated in Appendix C.

- *Sensitivity Analyses for Handling Non-Detects.* Health assessors should use the KM method for all dioxin and DLC TEQ calculations for data sets containing non-detect results. However, to confirm that the initial TEQ is not heavily influenced by the KM method, the initial estimate should be compared to lower- and upper-bound TEQ estimates with the following two substitution methods:

- Lower-bound TEQ estimate: Substitute all non-detect congeners with a detected value set to zero. This is done in the calculation spreadsheet by setting the concentrations for all non-detect observations to zero, removing the U flag in the qualifier field, and using KMcalc to determine the TEQ for this scenario.
- Upper-bound TEQ estimate: Substitute all non-detect congeners with a detected value set to the detection limit. This is done in the calculation spreadsheet by setting the concentrations for all non-detect observations to their detection limits, removing the U flag in the qualifier field, and using KMcalc to determine the TEQ for this scenario.

Additional Information:
Sensitivity Analysis

The sensitivity analyses described in Section 3.6 are steps the health assessor must take to determine whether the TEQ value calculated according to the guidance is acceptable to use for health assessment purposes. *The health assessor should not use the sensitivity analysis TEQs for their health evaluations.* The appropriate TEQ to use in health evaluations are calculated following procedures in Section 3.1.1 (for samples without non-detect observations) and using the KMcalc spreadsheet (for samples with non-detect observations).

For both the upper-bound and lower-bound TEQ estimates, health assessors should make substitutions based on whether each congener record was a non-detect in the original dataset. Changes to the detection status of records with the lowest or highest TEC made before completing the KM method calculations should be ignored in these sensitivity analyses.

Once these additional TEQs have been calculated, health assessors should compare the highest estimated TEQ to the lowest estimated TEQ by calculating the relative percent difference (RPD) between the two values. An RPD of greater than 50 percent suggests the method for replacing non-detects has an unacceptably large influence on the results. In this case, health assessors should consult with their ADS group on how to proceed. If the RPD is less than 50 percent, health assessors should proceed with CV comparisons and health effects evaluations using the initially calculated TEQ. The equation to calculate the RPD between two values is shown in Equation 3.

Equation 3:

$$RPD = \left(\frac{|Value_1 - Value_2|}{(Value_1 + Value_2) \div 2} \right) \times 100$$

Health assessors who are working in ATSDR's spreadsheet that already has the KMcalc macro loaded will be provided with the results of this sensitivity test along with the TEQ concentration and qualifier. Refer to Appendix B.1 and Appendix C for more detail.

- *Sensitivity Analyses for Handling R-qualified Data (e.g., rejected).* As described in Section 3.3, R-qualified data should never be included in initial TEQ estimates. However, to confirm that exclusion of R-qualified results does not have an unacceptably large influence on the TEQ, health assessors should compute the TEQ while including the rejected results and then estimate the RPD between this value and the initial TEQ estimate. This computation can only be performed if a concentration was included with the R-qualified results. If the RPD is less than 50

percent, health assessors should proceed with CV comparisons and health effects evaluations using the initially calculated TEQ.

If the RPD exceeds 50 percent and the TEQ including the R-qualified value is at a level of health concern, health assessors should still use the initial TEQ in the health evaluation but acknowledge the limitations associated with excluding the rejected value. The health assessor should also consider whether additional sampling is needed to reduce the uncertainties associated with the rejected values. Influence of rejected results will be greatest when congeners with relative high toxicity (e.g., a TEQ close to 1.0) are R-qualified.

3.7 Quality Control Checks

Health assessors are encouraged to have a colleague double-check their calculations. It is always good practice to have colleagues review calculations that support PHA conclusions, especially TEQ calculations used in health effects evaluations. To facilitate this review, health assessors should document their calculations—presumably in the KMcalc spreadsheet—and ask an experienced colleague to replicate the results.

3.8 Special Considerations

While ATSDR developed this guidance to apply to a broad range of site-specific scenarios, some environmental data sets will present unique challenges for calculating TEQs. In general, health assessors should consult with their ADS group when they encounter any site-specific scenarios or other circumstances not sufficiently covered by the general practice presented earlier in this section. Health assessors should also discuss with their ADSs group how to handle estimated maximum probable concentration (EMPC) data when calculating TEQs with their ADS group. These data are briefly described below.

Some dioxin results may contain EMPC values (e.g., when a congener peak is present at an acceptable signal-to-noise ratio, but ion abundance criteria are not met for definitive identification of that congener) (USEPA, 2011). These data are generally further qualified with a “J” flag for values that are estimated and a “U” flag for non-detect values. In general, “J” flagged EMPC values may be treated as qualified results, and U-qualified results may be considered non-detected values, and used in TEQ calculations following the process prescribed in Sections 3.3 to 3.5. However, given that EMPC values may overestimate the TEQ, a sensitivity analysis should be performed to assess the influence of EMPC results on calculated TEQs (USEPA 2011). Health assessors should perform a sensitivity analysis by calculating TEQs while excluding all EMPC values. When estimated TEQs are considerably different with and without these data, health assessors should contact their ADS group on how to proceed.

4.0 REFERENCES

[ATSDR] Agency for Toxic Substances and Disease Registry Division Community Health Investigations (DCHI). 2012. SOP: Toxic Equivalency Factors (TEFs) and Toxic Equivalent (TEQ) Concentration for Dioxin and Dioxin-Like Compounds in Environmental Media. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

[ATSDR] Agency for Toxic Substances and Disease Registry. 2019a. Exposure Point Concentration Guidance for Discrete Sampling. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

[ATSDR] Agency for Toxic Substances and Disease Registry. 2020a. Exposure Point Concentration Guidance for Non-Discrete Sampling. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

[ATSDR] Agency for Toxic Substances and Disease Registry. 2020b. Identifying Exposure Units for the Public Health Assessment Process. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

Helsel, DR. 2010. Summing nondetects: Incorporating Low-Level Contaminants in Risk Assessment. *Integr Environ Assess Manag*; 6(3):361-366.

Helsel, DR. 2012. *Statistics for Censored Environmental Data Using Minitab and R*. 2nd ed. Hoboken: John Wiley & Sons.

[NAS] National Academy of Science. 2006. Health Risks from Dioxin and Related Compounds: Evaluation of the EPA Reassessment. National Academies Press, Washington, DC. Available at: http://www.nap.edu/catalog.php?record_id=11688.

Safe, S. 1990. Polychlorinated biphenyls (PCBs), dibenzo-p-dioxins (PCDDs), dibenzofurans (PCDFs), and related compounds: environmental and mechanistic considerations which support the development of toxic equivalency factors. *Crit Rev Toxicol*; 21(1):51-88.

[USEPA] US Environmental Protection Agency. 2009. Recommended Toxicity Equivalency Factors (TEFs) for Human Health Risk Assessments of Dioxin and Dioxin-Like Compounds. External Review Draft. Available at: https://www.epa.gov/sites/production/files/2013-09/documents/hhtef_draft_090109.pdf.

[USEPA] US Environmental Protection Agency. 2011. User guide uniform federal policy quality assurance project plan template for soils assessment of dioxin sites.

Van den Berg M, Birnbaum LS, Denison M, De Vito M, Farland W, Feeley M, et al. 2006. The 2005 World Health Organization re-evaluation of human and mammalian toxicity equivalency factors for dioxins and dioxin-like compounds. *Toxicol Sci*; 93(2):223-241

Table 1. WHO 2005 Mammalian TEFs for Dioxin and DLCs

CAS Number	Full Congener Name	Shorthand Congener Name ¹	TEF ²
Dioxin Congeners			
1746-01-6	2,3,7,8-Tetrachloro dibenzo-p-dioxin	2,3,7,8-TCDD	1
40321-76-4	1,2,3,7,8-Pentachloro dibenzo-p-dioxin	1,2,3,7,8-PeCDD	1
39227-28-6	1,2,3,4,7,8-Hexachloro dibenzo-p-dioxin	1,2,3,4,7,8-HxCDD	0.1
57653-85-7	1,2,3,6,7,8-Hexachloro dibenzo-p-dioxin	1,2,3,6,7,8-HxCDD	0.1
19408-74-3	1,2,3,7,8,9-Hexachloro dibenzo-p-dioxin	1,2,3,7,8,9-HxCDD	0.1
35822-46-9	1,2,3,4,6,7,8-Heptachloro dibenzo-p-dioxin	1,2,3,4,6,7,8-HpCDD	0.01
3268-87-9	1,2,3,4,6,7,8,9-Octachloro dibenzo-p-dioxin	OCDD	0.0003
Furan Congeners			
51207-31-9	2,3,7,8-Tetrachloro dibenzofuran	2,3,7,8-TCDF	0.1
57117-41-6	1,2,3,7,8-Pentachloro dibenzofuran	1,2,3,7,8-PeCDF	0.03
57117-31-4	2,3,4,7,8-Pentachloro dibenzofuran	2,3,4,7,8-PeCDF	0.3
70648-26-9	1,2,3,4,7,8-Hexachloro dibenzofuran	1,2,3,4,7,8-HxCDF	0.1
57117-44-9	1,2,3,6,7,8-Hexachloro dibenzofuran	1,2,3,6,7,8-HxCDF	0.1
72918-21-9	1,2,3,7,8,9-Hexachloro dibenzofuran	1,2,3,7,8,9-HxCDF	0.1
60851-34-5	2,3,4,6,7,8-Hexachloro dibenzofuran	2,3,4,6,7,8-HxCDF	0.1
67562-39-4	1,2,3,4,6,7,8-Heptachloro dibenzofuran	1,2,3,4,6,7,8-HpCDF	0.01
55673-89-7	1,2,3,4,7,8,9-Heptachloro dibenzofuran	1,2,3,4,7,8,9-HpCDF	0.01
39001-02-0	1,2,3,4,6,7,8,9-Octachloro dibenzofuran	OCDF	0.0003
PCB Congeners			
32598-13-3	3,3',4,4'-Tetrachlorobiphenyl	3,3',4,4'-tetraCB (PCB 77)	0.0001
70362-50-4	3,4,4',5-Tetrachlorobiphenyl	3,4,4',5-tetraCB (PCB 81)	0.0003
32598-14-4	2,3,3',4,4'-Pentachlorobiphenyl	2,3,3',4,4'-pentaCB (PCB 105)	0.00003
74472-37-0	2,3,4,4',5-Pentachlorobiphenyl	2,3,4,4',5-pentaCB (PCB 114)	0.00003
31508-00-6	2,3',4,4',5-Pentachlorobiphenyl	2,3',4,4',5-pentaCB (PCB 118)	0.00003
65510-44-3	2,3',4,4',5'-Pentachlorobiphenyl	2',3,4,4',5-pentaCB (PCB 123)	0.00003
57465-28-8	3,3',4,4',5-Pentachlorobiphenyl	3,3',4,4',5-pentaCB (PCB 126)	0.1
38380-08-4	2,3,3',4,4',5-Hexachlorobiphenyl	2,3,3',4,4',5-hexaCB (PCB 156)	0.00003
69782-90-7	2,3,3',4,4',5'-Hexachlorobiphenyl	2,3,3',4,4',5'-hexaCB (PCB 157)	0.00003
52663-72-6	2,3',4,4',5,5'-Hexachlorobiphenyl	2,3',4,4',5,5'-hexaCB (PCB 167)	0.00003
32774-16-6	3,3',4,4',5,5'-Hexachlorobiphenyl	3,3',4,4',5,5'-hexaCB (PCB 169)	0.03
39635-31-9	2,3,3',4,4',5,5'-Heptachlorobiphenyl	2,3,3',4,4',5,5'-heptaCB (PCB 177)	0.00003

Notes:

- PCBs are named by the number and position of the chlorine atoms around the biphenyl ring. They are also sometimes referred to by their specific congener number, as shown in parentheses. For example, 3,3',4,4'-tetrachlorobiphenyl may be referred to by the shorthand name of 3,3',4,4'-TCB or PCB-77.
- Source: Mammalian TEFs as published in Van den Berg M, Birnbaum LS, Denison M, De Vito M, Farland W, Feeley M, et al. 2006. The 2005 World Health Organization re-evaluation of human and mammalian toxicity equivalency factors for dioxins and dioxin-like compounds. Toxicol Sci. 2006 Oct;93(2):223-241.

Appendix A. Glossary

Censored Data: A term commonly used to describe data sets including non-detect observations.

Detection Limit. For environmental sampling, detection limits (often referred to as method detection limits) are thresholds below which measured concentrations are not significantly different from a blank signal, at a specified level of probability. Measurements above detection limits are evidence of a nonzero signal at a given probability, confirming that the analyte of interest is present in the sample.

Environmental Sample. A collected quantity of air, water, soil, food, or other media in which contamination levels are measured, whether directly in the field or at a laboratory.

Estimated maximum probable concentration (EMPC). Some dioxin results may contain EMPC values. This designation is used when a congener peak is present during laboratory analysis at an acceptable signal-to-noise ratio, but ion abundance criteria are not met for definitive identification of that congener (USEPA, 2011). These data should still be used in the public health assessment process, and they are generally further qualified with a “J” flag for values that are estimated and a “U” flag for non-detect values.

Exposure Point Concentration (EPC). The representative contaminant concentration within an exposure unit or area in an exposure pathway to which receptors are exposed for acute, intermediate, or chronic durations during the past, present, or future.

Dioxin-like. Dioxin-like compounds are structurally related groups of chemicals from the family of halogenated aromatic hydrocarbons. In this group, we include chlorinated dibenzo-p-dioxins, chlorinated dibenzofurans, and certain PCBs that have dioxin-like structural characteristics (“non-ortho-” and “mono-ortho-” substitutions).

Kaplan-Meier (KM). Kaplan-Meier is a non-parametric standard method for calculating statistics for data sets included censored data. In the context of this guidance, Kaplan-Meier method should be applied when calculating TEQs for an environmental sample that has non-detect observations with different values of the detection limit.

Non-parametric methods. Non-parametric methods are statistical methods that do not assume data have a known shape or distribution. These methods can be applied with little information about the underlying distribution, including instances where details of the data distribution are not known. In this guidance, health assessors use non-parametric Kaplan-Meier methods to estimate toxicity equivalents for data sets with non-detect results.

Toxic Equivalent Concentration (TEC). The TEC is calculated as a congener’s concentration multiplied by its TEF. The TEC is calculated as an intermediate step of the TEQ estimation.

Toxic Equivalent Factor (TEF). The TEF is based on the relative potency of the compound to the benchmark compound. A TEF of 0.1 indicates that the compound is 1/10th as toxic as the benchmark compound, while 1 indicates that it is as toxic as the benchmark compound.

Toxic Equivalents (TEQ). Calculated values that represent the overall toxicity of a complex mixture of congeners, relative to a single benchmark compound (e.g., the most toxic compound in that family).

Appendix B. Example TEQ Calculations in Microsoft Excel with KMcalc Macro

This example demonstrates the preferred approach for calculating TEQs using ATSDR’s Microsoft Excel KMcalc macro function. The data set considered in this example includes 17 congeners measured in a single soil sample (Helsel, 2010), and are presented in the table below. Health assessors who are using the KMcalc macro for the first time are encouraged to replicate this example before using the macro to calculate TEQs for site-specific scenarios. The remainder of this example follows the process outlined in Section 3.0. All figures are screen shots from Excel in Microsoft Office 365™.

Congener Name	Concentration (ng/kg) ^a
1,2,3,4,6,7,8-HpCDD	25
1,2,3,4,6,7,8-HpCDF	1.8
1,2,3,4,7,8,9-HpCDF	<0.56
1,2,3,4,7,8-HxCDD	0.26
1,2,3,4,7,8-HxCDF	<0.6
1,2,3,6,7,8-HxCDD	2.1
1,2,3,6,7,8-HxCDF	0.33
1,2,3,7,8,9-HxCDD	0.77
1,2,3,7,8,9-HxCDF	0.37
1,2,3,7,8-PeCDD	0.18
1,2,3,7,8-PeCDF	0.24
2,3,4,6,7,8-HxCDF	<0.14
2,3,4,7,8-PeCDF	<0.8
2,3,7,8-TCDD	1.7
2,3,7,8-TCDF	5.1
OCDD	220
OCDF	44

Notes:

- a. data as presented in Helsel, D (2010).
“<” indicates that result was not detected. The value presented is the full detection limit.

ATSDR has developed the KMcalc macro and spreadsheet for two different categories of Excel users: a general user and an advanced user. The general user is encouraged to work with “Option 1” in the KMcalc spreadsheet to determine TEQs for a single sample. General users should refer to Appendix B.1 for instructions on how to do so. The advanced Excel user may elect to import the KMcalc macro into other spreadsheets and apply the macro accordingly. This user should refer to Appendix B.2 for instructions on how to apply the macro. Health assessors who have questions about applying the macro should contact their ADS group.

Note that the KMcalc macro can be used to calculate TEQs for datasets with and without non-detects. In one rare exception (i.e., for samples with fewer than three detected results), however, the spreadsheet cannot be used to calculate TEQs and health assessors are directed to Section 3.4.1.

Appendix B.1: Instructions for the General User

The general user is encouraged to use “Option 1” of the KMcalc spreadsheet. This option has a few key benefits. First, health assessors need not manually enter the TEF values in the spreadsheet. This option has the TEF values coded into the file. Provided the health assessor uses the correct spelling for the congeners, the spreadsheet will automatically populate the TEF values after the congener names are entered. Second, once the congener names, concentrations, and qualifiers are entered and the user clicks “Run TEQ Calculations,” the spreadsheet instantly generates all necessary outputs (which would otherwise have to be generated manually in “Option 2”). The one limitation of this option is that it may prove inefficient in cases where health assessors need to calculate TEQs for large numbers of samples. In such cases, the second KMcalc option (see Appendix B.2) may save time.

Three spreadsheets in this KMcalc file contain all information needed for TEQ calculations according to “Option 1.” The health assessor enters the relevant inputs for a single sample, and the spreadsheet outputs the TEQ value, the qualifier that should be applied to the TEQ value (if any), the units for the TEQ, and whether the TEQ calculation passed the non-detect sensitivity analysis test referred to in Section 3.6 of this guidance. The “Option 1” spreadsheets include:

- “Option 1 – Instructions”: This spreadsheet lists the three steps to conduct a TEQ calculation for a single sample. More details on the required data entry steps are listed in the next bulleted item. Upon opening the spreadsheet, users might be prompted to “enable macros.” Macros must be enabled to run the “KMcalc” function and perform the TEQ calculations.
- “Option 1 – Calculations”: After the health assessor enters the congener-specific environmental data into this spreadsheet and clicks “Run TEQ Calculations”, key outputs appear in the orange-shaded cells, provided the user entered the sampling data according to the following specifications:
 - Only enter data in the blue-shaded cells. The spreadsheet includes blue-shaded cells where the date, site name, sample media, notes, concentration units, congener names, congener concentrations, and data qualifiers should be entered. Do not insert new rows or delete existing rows. Some interim values for the calculations appear in hidden cells.
 - Enter a separate row for every dioxin or dioxin-like compound that was measured in the sample, including those with non-detect results. Do not include blank rows between entries.
 - The “shorthand congener name” in the first column must exactly match one of the congener names listed in the “Option 1 – TEF Values” spreadsheet, which lists the current universe of 29 congeners and corresponding TEF values that have been approved for these calculations. A pop-up error message will appear if a shorthand congener name is entered incorrectly. If this occurs, check to make sure congener names have been entered correctly, with no typographical errors or extra spaces. Shorthand congener names may also be selected from those provided in the drop-down lists found in each cell of the first column. Upon entering shorthand congener names in the first column, the congener’s category, CAS number, and full name will automatically populate the following three columns. Note that this spreadsheet only calculates TEQs

- for the congeners listed in “Option 1 – TEF Values.” Consult with you ADS group if a congener of interest does not appear on the list.
- In the “concentration” field, enter a numerical value for every congener that was evaluated. That numerical value should be the measured concentration for detected congeners and the method detection limit for non-detect observations. Every row with a congener name must have an entry in this column. Be sure to enter data in the same units of measurement for every congener. Congener concentrations entered as negative values or zero will result in a pop-up error message.
 - In the “qualifier” field, only four options are allowed for each congener: U, J, UJ, or nothing. Enter the corresponding qualifier for every measurement from this list. Note that “U” refers to non-detect observations, “J” refers to estimated values, and “UJ” refers to estimated values below the detection limit. These qualifiers must be entered as capital letters; any other entries will result in a pop-up error message. Qualifiers may also be selected from those provided in the drop-down lists of each cell in this column.
 - After completing all data entry steps, click “Run TEQ calculations” and save the spreadsheet. The entries in the orange-shaded cells in rows 4 through 7 present the final results. Those include the TEQ value for the sample (row 4), the units for the calculated TEQ (row 5), the qualifier to use for the TEQ value (row 6), and whether the TEQ calculation passed the non-detect sensitivity test outlined in Section 3.6 of this guidance (row 7). (Note: If no qualifier is needed for the calculated TEQ value, then “no qualifier” will appear in the orange-shaded cell in row 6.)
- “Option 1 – TEF Values”: This spreadsheet lists the TEFs that ATSDR has approved for TEQ calculations for dioxin and DLCs. It is included for reference purposes only, and entries should not be changed. The spreadsheet can be useful in cases where a “check congener spelling” error appears in column D.

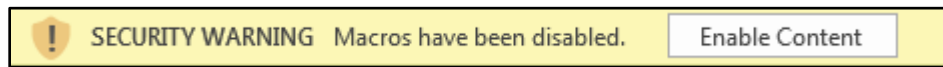
Appendix B.2: Instructions for the Advanced Excel User

The advanced Excel user may choose to follow this second option for calculating TEQ values. In this option, the user must enter the equations for the KM calculations. This allows for greater flexibility and can save time when calculating TEQs for numerous samples at once. However, in this option, health assessors also must enter TEF values on their own—and therefore must take great care in that process to ensure that correct values are used. Additionally, health assessors must run separate calculations for the TEQ values, the qualifiers, and the sensitivity analyses (and all three calculations are performed automatically for the first option). The process for using the second option for advanced users is outlined below in a series of four steps.

Step One: Loading KMcalc macro to Excel

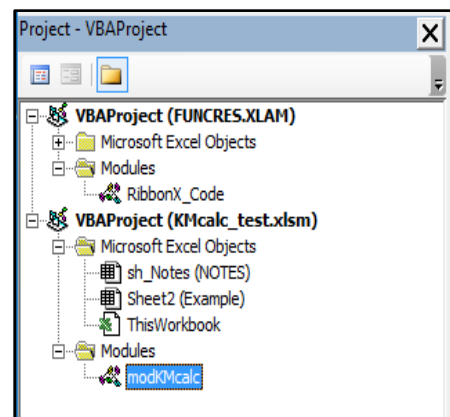
The KMcalc macro is written in Visual Basic for Applications (VBA) for Excel. Health assessors may access the KMcalc macro in one of two ways: (1) save a copy of the example spreadsheet that already has the macro loaded and enter new site-specific data or (2) import the VB module (modKMcalc.bas) to an existing file that contains the environmental data. Both options are described below:

Option 1: If you chose to use the example spreadsheet that already has the macro loaded, you must enable macros through Excel’s Trust Center Settings. To do so, go into Excel’s File menu and select → Options → Trust Center → Trust Center Settings → Macro Settings → Disable Macros with Notification. This will force Excel to present a notification each time you open the Excel file (as shown below). You must select “Enable Content” from this pop-up window to use any macros.



Option 2: If you chose to load the macro to an existing spreadsheet that already contains the analytical data, follow the directions below:

1. Open the existing spreadsheet that contains your analytical data and save the file as an Excel Macro-Enabled Workbook (.xlsm).
2. Open the Visual Basic (VB) Editor by simultaneously clicking the “Alt” key and “F11” key on your keyboard.
3. Import the macro “modKMcalc.bas” by first selecting the VBA Project with the name of your file in the left upper portion of the window. Then select File → Import file, and locate the KMcalc VB module (modKMcalc.bas).
4. “ModKMcalc” will now appear in the VBA Project window, as shown in the screenshot below. Once “modKMcalc” appears in this window, you can use the KMcalc function in any cell of your data spreadsheet.



Step Two: Initial Data Processing Steps

1. *Perform a data quality review.* In this example, it is assumed that the data are of a known and high quality and meet all data quality objectives for the health evaluation in question. There are no R-qualified (e.g., rejected) results and therefore no data must be removed from the spreadsheet before calculating the TEQ.
2. *Identify and process duplicate samples and replicate analyses.* The example data set is for a single sample without replicate or duplicate analysis, and no further data processing is required.

Step Three: Prepare data in Excel for TEQ Calculations

This example includes concentrations for 17 congeners, four of which are reported as non-detect. As described in Section 3.4, the KM method can be used to calculate TEQs for data sets with non-detect results, if there are at least three detected concentrations. The data for this environmental sample meets this criterion and the KMcalc macro will apply the KM method.

The data was entered to Excel, as shown in the screenshot below. Note that this spreadsheet includes the necessary KMcalc function inputs: congener concentrations, qualifiers, and TEFs. Any concentration shown in the original data table above with a “<” (e.g., indicating a non-detect result) is included in the Excel spreadsheet with a U-qualifier.

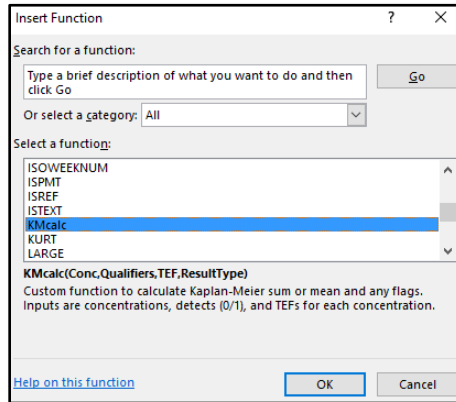
	A	B	C	D	E
	Congener	Concentration (ng/kg)	TEF	Qualifier	
1					
2	1,2,3,4,6,7,8-HpCDD	25	0.01		
3	1,2,3,4,6,7,8-HpCDF	1.8	0.01		
4	1,2,3,4,7,8,9-HpCDF	0.56	0.01	U	
5	1,2,3,4,7,8-HxCDD	0.26	0.1		
6	1,2,3,4,7,8-HxCDF	0.6	0.1	U	
7	1,2,3,6,7,8-HxCDD	2.1	0.1		
8	1,2,3,6,7,8-HxCDF	0.33	0.1		
9	1,2,3,7,8,9-HxCDD	0.77	0.1		
10	1,2,3,7,8,9-HxCDF	0.37	0.1		
11	1,2,3,7,8-PeCDD	0.18	1		
12	1,2,3,7,8-PeCDF	0.24	0.03		
13	2,3,4,6,7,8-HxCDF	0.14	0.1	U	
14	2,3,4,7,8-PeCDF	0.8	0.3	U	
15	2,3,7,8-TCDD	1.7	1		
16	2,3,7,8-TCDF	5.1	0.1		
17	OCDD	220	0.0003		
18	OCDF	44	0.0003		
19					

Step Four: Calculate the TEQ

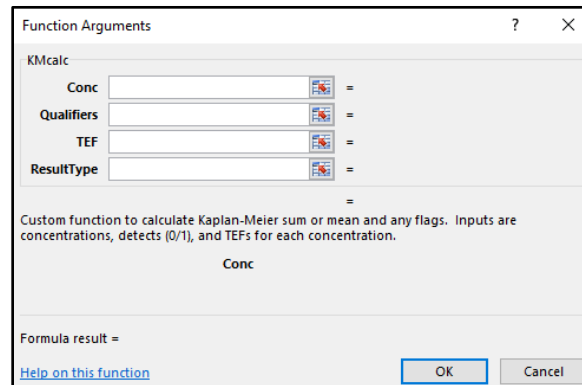
As described in Section 3.5, the KMcalc function can be used by either selecting Excel’s Insert Function button and highlighting the necessary data or entering the formula directly into an empty cell. Detailed instructions for both methods are provided below.

1. *Using Excel’s Insert Function button.* The KMcalc formula can be calculated in any cell of the spreadsheet by clicking on that cell and using Excel’s Insert Function option. Once the required inputs and outputs are selected, the calculated value will appear in the selected cell. To launch

the Insert Function dialog box, users can either click the “Insert Function” button in the “Formulas” tab of the Excel ribbon, or click the “fx” (function) button in the Formula Bar. Users must then find and select the KMcalc function and click “OK.” A screenshot of the search tool for Excel functions is shown below.



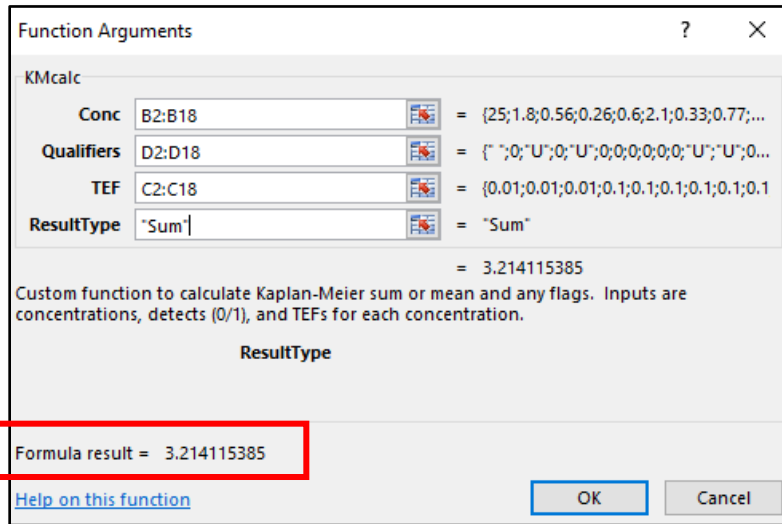
In the KMcalc Function box, the required arguments/inputs are defined by clicking on the small red, white, and blue button to the right of each argument and then highlighting the appropriate data in the Excel spreadsheet. A screenshot of the KMcalc function box is shown below.



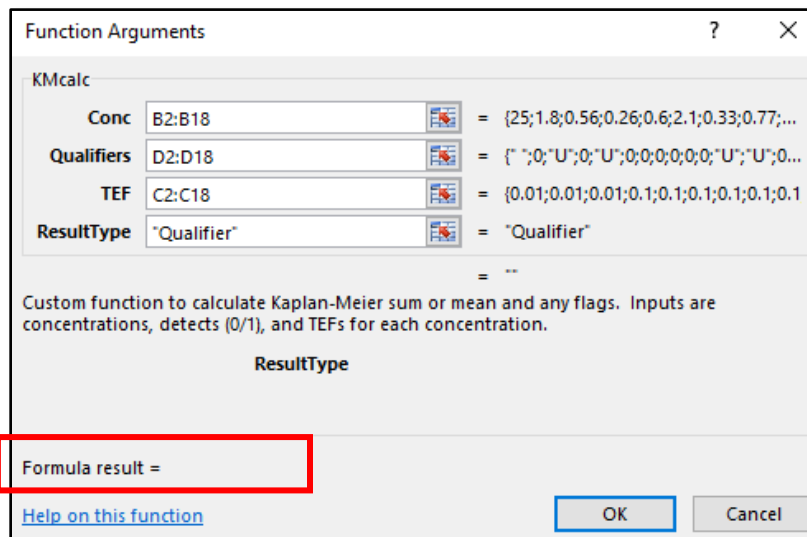
In this example, congener concentrations are located in cells B2 through B18, congener qualifiers are located in cells D2 through D18, and congener TEFs are located in cells C2 through C18. Health assessors would select the function arguments as shown in the Function Arguments dialog box screenshots below. Health assessors are reminded that the function must be run twice; once with the ResultType set to “Sum” (e.g., to calculate the TEQ) and then again with the ResultType set to “Qualifier” (e.g., to determine what, if any, qualifier should be assigned to the calculated TEQ).

After the inputs are entered and “OK” is selected, the macro will return the result (whether a calculated TEQ or the TEQ qualifier) to the empty cell. A preview of the requested result is presented in the bottom left-hand corner of the Function Arguments dialog box.

Function Arguments for the *TEQ*:



Function Arguments for the *TEQ-Qualifier*:



In the first screenshot, the function is requesting the ResultType “Sum” (e.g., the TEQ), and that value is shown at 3.21 ng-TEQ/kg. In the second screenshot, the function is requesting the ResultType “Qualifier”, and that value is shown as a blank result.

In this example, the 2,3,7,8-TCDD TEQ is calculated at 3.21 ng-TEQ/kg, with no qualifiers.

2. *Entering the function formula directly into an empty cell.* Alternatively, the KMcalc formula function can be entered to any empty cell in the spreadsheet. This requires typing the formula into an empty cell and selecting the appropriate range of cells for each required input. The KMcalc function formula is as follows:

$$= \text{KMcalc}(\text{Conc}, \text{Qualifier}, \text{TEF}, \text{ResultType})$$

When typing the “Conc” part of the formula, one must enter the range of the cells that contain the congener concentrations. Similarly, when typing the “Qualifier” and “TEF” parts of the formula, one must enter the range of cells where these data are found. The KMcalc formula will return the calculated ResultType to the cell where the formula was entered. Health assessors must run the function once with the ResultType set to “Sum” and then again with the ResultType set to “Qualifier.”

For this example, the inputs for the KMcalc formula require the following cell ranges:

- Conc = B2:B18 (The congener concentrations are found in cells B2 through B18).
- Qualifier = D2:D18 (The congener qualifiers are found in cells D2 through D18).
- TEF = C2:C18 (The congener TEFs are found in cells C2 through C18).
- ResultType = “Sum” and “Qualifier” (The two required outputs must be run separately).

The KMcalc functions would therefore be entered to separate empty cells as:

```
=kmcalc(B2:B18,D2:D18,C2:C18,"Sum")
```

```
=kmcalc(B2:B18,D2:D18,C2:C18,"Qualifier")
```

The KMcalc function will output the following results for this sample:

- Sum = 3.21
- Qualifier = *[blank cell]*

In this example, the TEQ is calculated at 3.21 ng-TEQ/kg with no qualifiers.

Appendix C. Example Sensitivity Analyses

As described in Section 3.6, health assessors should complete additional TEQ calculations when working with data sets containing non-detect (U-qualified) or R-qualified values. This appendix presents sensitivity analyses using the same data set from Appendix B for different approaches to both scenarios. Health assessors who are using the KMcalc Excel macro to calculate TEQs for the first time are encouraged to replicate this example before using the macro for site-specific scenarios. The remainder of this appendix follows the process outlined in Section 3.6. All figures are screen shots from Excel.

1. Sensitivity Analyses for Different Approaches to Handling Non-Detects:

Health assessors should compare estimated TEQs using two substitution methods:

- Substituting non-detect congeners with a detected value set to zero.
- Substituting non-detect congeners with a detected value set to the full detection limit.

Health assessors can calculate these additional TEQs with the KMcalc macro function by substituting non-detect results and removing all qualifiers (as shown in the image below).

	A	B	C	Data Sets for Sensitivity Analyses		
				D	F	G
1	Measured Dioxin/Furan Congener	TEF (unitless)	Original Data Set (ng/kg)	Substitute 0 for non-detects	Substitute full DL for non-detects	Qualifiers
2						
3	1,2,3,4,6,7,8-HpCDD	0.01	25	25	25	
4	1,2,3,4,6,7,8-HpCDF	0.01	1.8	1.8	1.8	
5	1,2,3,4,7,8,9-HpCDF	0.01	<0.56	0	0.56	
6	1,2,3,4,7,8-HxCDD	0.1	0.26	0.26	0.26	
7	1,2,3,4,7,8-HxCDF	0.1	<0.6	0	0.6	
8	1,2,3,6,7,8-HxCDD	0.1	2.1	2.1	2.1	
9	1,2,3,6,7,8-HxCDF	0.1	0.33	0.33	0.33	
10	1,2,3,7,8,9-HxCDD	0.1	0.77	0.77	0.77	
11	1,2,3,7,8,9-HxCDF	0.1	0.37	0.37	0.37	
12	1,2,3,7,8-PeCDD	1	0.18	0.18	0.18	
13	1,2,3,7,8-PeCDF	0.03	0.24	0.24	0.24	
14	2,3,4,6,7,8-HxCDF	0.1	<0.14	0	0.14	
15	2,3,4,7,8-PeCDF	0.3	<0.8	0	0.8	
16	2,3,7,8-TCDD	1	1.7	1.7	1.7	
17	2,3,7,8-TCDF	0.1	5.1	5.1	5.1	
18	OCDD	0.0003	220	220	220	
19	OCDF	0.0003	44	44	44	
21			sum:	3.13	3.45	
22			qualifiers:			

For this example, the following TEQs are estimating with the different approaches to handle non-detects. None of the TEQs are J- or U-qualified.

Approach to Handling Non-Detect Observations	TEQ (ng-TEQ/kg)
Initial TEQ based on the KM method	3.21
Lower Bound TEQ substituting zero for non-detects	3.13
Upper Bound TEQ substituting full detection limit for non-detects	3.45

After calculating TEQs with the various methods for handling non-detects, health assessors are instructed to compare the highest estimated TEQ to the lowest estimated TEQ by calculating the relative percent difference (RPD) between the two values. For this example, the highest TEQ was estimated at 3.45 ng-TEQ/kg (while substituting the full DL for non-detects) and the lowest TEQ was estimated at 3.13 ng-TEQ/kg (while substituting zero for all non-detects). The RPD between 3.45 ng-TEQ/kg and 3.13 ng-TEQ/kg is calculated as shown below.

$$RPD = \left(\frac{|Value_1 - Value_2|}{(Value_1 + Value_2) \div 2} \right) \times 100$$

$$RPD = \left(\frac{|3.45 - 3.13|}{(3.45 + 3.13) \div 2} \right) \times 100$$

$$RPD = 9.7 \text{ percent}$$

For this example, the RPD between the highest and lowest calculated TEQ is 9.7 percent. This value is below an RPD of 50 percent and therefore suggests that the TEQ is not particularly sensitive to how the non-detects are handled. No further evaluation is necessary and the health assessor should use the initial TEQ based on the KM method.

Health assessors who are calculating TEQs following “Option 1” (described above in Appendix B.1) will be provided with the results of this sensitivity test in the orange-shaded cells of the “OPTION 1 – Calculations” spreadsheet, along with the TEQ result, units, and qualifier outputs.

2. Sensitivity Analyses for Different Approaches to Handling R-qualified results

As described in Section 2, R-qualified results should not be included in TEQs. However, if health assessors excluded any R-qualified results from TEQ calculations, the initial TEQ estimate should be compared to that while including the R-qualified data. There were no R-qualified results in the example shown in Appendix B. However, for illustrative purposes, this section presents the sensitivity analysis for the Appendix B data set, except the detected result for OCDD (220 ng/kg) is assumed to be rejected. TEQ estimates excluding and including the R-qualified result (200 ng/kg for OCDD) are shown in the table below.

Approach to Handling R-Qualified Results	Estimated TEQ (ng/kg)
Initial TEQ based on the KM method excluding R-qualified result	3.13
TEQ based on the KM method including R-qualified result	3.21

The RPD between the initial TEQ and the TEQ included R-qualified results is 2.9 percent, suggesting that the exclusion of rejected data has minimal impact on the overall TEQ. In this case, health assessors would proceed with the initial TEQ.