DoD/DOE QSM 6.0 Module 7 Toxicity Testing Checklist

Checklists used for this assessment activity:

- ☐ **M1/M2 PT/QMS**
- ☐ **M3 Asbestos Testing**
- ☐ **M4 Chemical Testing**
- ☐ **M5 Microbiological Testing**
- ☐ **M6 Radiochemical Testing**
- ☐ **M7 Toxicity Testing**
- ☐ **M8 Industrial Hygiene Testing**

This checklist is only a tool, and not considered as the requirements of the standard(s)!

If there is a disagreement between this checklist and the standard(s), the standard(s) shall prevail.

Identify conformity for each requirement along with comments/objective evidence for each clause assessed.

A *clarifying statement* provides additional information to help understand a requirement.

A *permission* is an approach that a conformity assessment body can use to achieve compliance.

Assessment Number:

CAB Name:

Physical Address:

Assessment Date(s):

Assessors(s):

DoD/DOE QSM 6.0 Clause	Requirement	Conformity C / NC / NA	Comments/Objective Evidence
M7	Quality Systems for Toxicity Testing		
M7: 4.0	Method Selection		
M7: 4.0	The requirements in Module 2 Section on “Selection, Verification and Validation of Methods” apply.		Clarifying Statement
M7: 4.0	When it is necessary to use testing methods not covered by a reference method, is there agreement with the data user and is there a clear specification of the data user’s requirements and the purpose of the environmental test?		
M7: 4.0	Is the developed method validated appropriately before use?		

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M7: 4.0	Is the characteristics of validated methods (e.g., the uncertainty of the results, limit of repeatability and/or reproducibility, robustness against external influences and/or cross-sensitivity against interference from the matrix of the sample/test object), as assessed for the intended use, relevant to the customer's needs?		
M7: 5.0	Method Validation		
M7: 5.0	Validation is the confirmation by examination and the objective evidence that the requirements for a specific intended use are fulfilled.		Clarifying Statement
M7: 6.0	Demonstration of Capability (DOC)		
M7: 6.1	General		
M7: 6.1	Prior to acceptance and institution of any method for data reporting, is satisfactory initial DOC performed?		
M7: 6.1	Thereafter, ongoing DOC, as per the QC requirements in Section 7.1.2, is required.		Clarifying Statement
M7: 6.1	In cases where a laboratory analyzes samples using a method that has been in use by the laboratory for at least one year prior to applying for accreditation, and there have been no significant changes in personnel or method, the ongoing DOC shall be acceptable as an initial DOC. Are records retained to demonstrate that an initial DOC is not required?		
M7: 6.1	For the initial DOC, are appropriate records as discussed in Section 6.2.1 completed?		
M7: 6.1	Is an initial DOC completed each time there is a change in personnel, or method and before any results are reported?		
M7: 6.1	In general, this demonstration does not test the performance of the method in real world samples. However, before any results are reported, the initial DOC shall be performed.		Clarifying Statement
M7: 6.1	An initial DOC may be completed by a group of analysts and is for situations in which several individuals perform part of a set of activities that would produce a testing result.		Permission
M7: 6.1	Are all demonstrations recorded?		
M7: 6.1	Is all data applicable to the demonstration retained and readily available at the laboratory?		
M7: 6.2	Initial DOC		
M7: 6.2	Is an initial DOC made prior to using any method, and at any time there is a significant change in personnel or method or any time that a method has not been		

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	performed by the laboratory or individual in a 12-month period?		
M7: 6.2.1	Are records of each initial DOC maintained in a manner such that the following information is available for each affected employee?		
M7: 6.2.1.a	Individual(s) involved in preparation and/or analysis?		
M7: 6.2.1.b	Matrix?		
M7: 6.2.1.c	Species and endpoint(s)?		
M7: 6.2.1.d	Identification of method(s) performed?		
M7: 6.2.1.e	Identification of laboratory-specific procedures used for analysis, including revision number?		
M7: 6.2.1.f	Date(s) of analysis? and		
M7: 6.2.1.g	Summary of analyses, including information outlined in Section 6.2.2?		
M7: 6.2.2	If the reference method or regulation does not specify an initial DOC, the following procedure is acceptable.		Permission
M7: 6.2.2	Are other approaches to initial DOC documented to be adequate?		
M7: 6.2.2	Does each analyst meet the QC requirements as specified in Section 7.1.2?		
M7: 6.3	Ongoing DOC		
M7: 6.3	Is there a procedure describing ongoing DOC?		
M7: 6.3	Do the individual(s) demonstrate ongoing capability by meeting the QC requirements of the reference method, laboratory procedure, customer specifications, and/or this Standard?		
M7: 6.3	Are other approaches to initial DOC documented to be adequate?		
M7: 6.3	This ongoing demonstration may include performing another initial demonstration of capability as per 6.2 or a documented process of analyst review using QC samples may serve as the annual ongoing DOC.		Permission
M7: 6.3	Are QC samples reviewed to identify patterns for individuals or groups of analysts and determine if corrective action or retraining is necessary?		
M7: 7.0	Technical Requirements		
M7: 7.1	Quality Control		
M7: 7.1	Are there QC procedures for monitoring the validity of environmental tests undertaken?		
M7: 7.1	Is the resulting data recorded in such a way that trends are detectable and, where practicable, statistical techniques are applied to the reviewing of the results?		
M7: 7.1	Is this monitoring planned and reviewed?		

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M7: 7.1	This monitoring may include, but not be limited to, the following:		Permission
M7: 7.1.a	Regular use of certified reference materials and/or internal QC using secondary reference materials;		Permission
M7: 7.1.b	Participation in inter-laboratory comparison or proficiency-testing program;		Permission
M7: 7.1.c	Replicate tests using the same or different methods;		Permission
M7: 7.1.d	Retesting of retained samples; and		Permission
M7: 7.1.e	Correlation of results for different characteristics of a sample (for example, total phosphate should be greater than or equal to orthophosphate).		Permission
M7: 7.1.1	Essential Quality Control Procedures These general QC principles shall apply, where applicable, to all testing laboratories. The way they are implemented is dependent on the types of tests performed by the laboratory and are further described in this module. The standards for any given test type shall assure that the applicable principles are addressed:		Clarifying Statement
M7: 7.1.1.a	Are there detailed written protocols in place to monitor the following QCs?		
M7: 7.1.1.a.i	Positive and negative controls to monitor tests such as blanks, spikes, reference toxicants?		
M7: 7.1.1.a.ii	Tests to define the variability and/or repeatability of the laboratory results such as replicates?		
M7: 7.1.1.a.iii	Measures to evaluate method capability (e.g., percent minimum significant difference (PMSD))?		
M7: 7.1.1.a.iv	Selection of appropriate formulae to reduce raw data to final results such as regression and statistical analyses?		
M7: 7.1.1.a.v	Selection and use of reagents and standards of appropriate quality?		
M7: 7.1.1.a.vi	Measures to assure the selectivity of the test for its intended purpose? and		
M7: 7.1.1.a.vii	Measures to assure constant and consistent test conditions (both instrumental and environmental) where required by the reference method such as temperature, humidity, light, or specific equipment conditions?		
M7: 7.1.1.b	Are all QC measures assessed and evaluated on an ongoing basis, and QC acceptance criteria used to determine the usability of the data?		
M7: 7.1.1.c	Are there procedures for the development of acceptance/rejection criteria where no reference method or regulatory criteria exist?		

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M7: 7.1.1.d	Are the QC protocols specified by the laboratory's method manual followed?		
M7: 7.1.1.d	Are the essential standards outlined in this document or regulations (whichever are more stringent) incorporated into the method manuals?		
M7: 7.1.1.d	When it is not apparent which is more stringent, is the QC in the regulations followed?		
M7: 7.1.2	Positive and Negative Controls		
M7: 7.1.2.a	Positive Control		Clarifying Statement
M7: 7.1.2.a.i	Has the ability to obtain consistent results with standard reference toxicants (SRT) been demonstrated?		
M7: 7.1.2.a.ii	Has ongoing laboratory performance been demonstrated by performing routine SRT testing for each method, species, and endpoint in accordance with the minimum frequency requirements specified in Section 7.1.2.a.iii?		
M7: 7.1.2.a.iii	Does the frequency of ongoing laboratory reference toxicant testing conform to the following unless the reference method specifically requires less frequent SRT tests (e.g., sediment tests)?		
M7: 7.1.2.a.iii	For methods conducted at a frequency of monthly or greater, are SRT tests conducted monthly?		
M7: 7.1.2.a.iii	For methods and species commonly used in the laboratory, but which are tested at a frequency of less than monthly, are SRT tests conducted concurrently with the environmental test?		
M7: 7.1.2.a.iii	If the test organisms are obtained from an outside source, is the sensitivity of each batch of organisms received from a supplier determined via a concurrent SRT test unless the supplier can provide control chart data for the last five SRT tests using the same SRT and test conditions?		
M7: 7.1.2.a.iii	Is supplied SRT data not older than six months?		
M7: 7.1.2.a.iv	These standards do not currently specify a particular reference toxicant and dilution series. However, if the regulation identifies a reference toxicant or dilution series for a particular test, are the specified requirements followed?		
M7: 7.1.2.a.iv	Do all reference toxicant tests conducted for a given method and species use the same reference toxicant, test concentrations, dilution water and data analysis methods?		
M7: 7.1.2.a.iv	Is a dilution factor of 0.5x or greater used for both acute and chronic tests?		
M7: 7.1.2.a.v	Are the reference toxicant tests conducted following the procedures required in the reference method?		

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M7: 7.1.2.b	Negative Controls – Control, Brine Control, Control Sediment, Control Soil, and Dilution Water		
M7: 7.1.2.b.i	Are the standards for the use, type, and frequency of testing of negative controls specified by the reference methods and by permit or regulation and followed?		
M7: 7.1.2.b.i	Is a negative control is included with each test to evaluate test performance and the health and sensitivity of the specific batch of organisms?		
M7: 7.1.2.b.ii	Are appropriate additional negative controls included when sample adjustments (for example addition of thiosulfate for de-chlorination) or solvent carriers used in the test?		
M7: 7.1.3	Variability and/or Reproducibility Is intra-laboratory precision determined on an ongoing basis using further reference toxicant tests and related control charts as described above?		
M7: 7.1.4	Test Sensitivity		
M7: 7.1.4.a	Is the PMSD calculated according to the formula specified by the reference method and reported with the test results?		
M7: 7.1.4.b	Point estimates: (LCp, ICp, or ECp) Are confidence intervals reported as a measure of the precision around the point estimate value, when the calculation is possible?		
M7: 7.1.5	Selection and Use of Reagent and Standards		
M7: 7.1.5.a	The grade of all reagents used in toxicity tests is specified in the reference method except the reference standard.		Clarifying Statement
M7: 7.1.5.a	Are all reference standards prepared from chemicals that are analytical reagent grade or better?		
M7: 7.1.5.a	Are records maintained of the preparation of all standards and reference toxicants?		
M7: 7.1.5.b	Do all standards and reagents associated with chemical measurements, such as dissolved oxygen, pH, or specific conductance, comply with Module 4?		
M7: 7.1.5.c	Is only reagent-grade water collected from distillation or de-ionization units used to prepare reagents?		
M7: 7.1.6	Constant and Consistent Test Conditions		
M7: 7.1.6.a	If closed refrigerator-sized incubators are used, are culturing and testing of organisms separated to avoid cross-contamination?		
M7: 7.1.6.b	Is the laboratory space adequate for the types and numbers of tests performed?		
M7: 7.1.6.b	Does the building provide adequate cooling, heating, and illumination for conducting testing and culturing?		

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M7: 7.1.6.b	Are hot and cold running water available for cleaning equipment?		
M7: 7.1.6.c	Is the air used for aeration of test solutions, dilution waters and cultures free of oil and fumes?		
M7: 7.1.6.d	Does the laboratory or a contracted outside expert positively identify test organisms to species on an annual basis?		
M7: 7.1.6.d	Are the taxonomic reference (citation and page(s)) and the names(s) of the taxonomic expert(s) kept on file at the laboratory?		
M7: 7.1.6.d	When organisms are obtained from an outside source, does the supplier provide this same information?		
M7: 7.1.6.e	Is equipment used for routine support measurements of chemical and physical parameters such as pH, DO, conductivity, salinity, alkalinity, hardness, chlorine, ammonia, and weight calibrated, and/or standardized per manufacturer's instructions?		
M7: 7.1.6.e	Are records maintained of all measurements and calibrations?		
M7: 7.1.6.f	Is the test temperature maintained as specified by the reference method?		
M7: 7.1.6.f	Is the temperature control equipment adequate to maintain the required test temperature(s)?		
M7: 7.1.6.f	Is the average daily temperature of the test solutions maintained within method specified range?		
M7: 7.1.6.f	Is the minimum frequency of measurement once per 24-hour period?		
M7: 7.1.6.f	Is the test temperature for continuous-flow toxicity tests recorded and monitored continuously?		
M7: 7.1.6.f	Where electronic data loggers are used, is the temperature monitored at a frequency sufficient to capture temporal variations of the environmental control system?		
M7: 7.1.6.g	Does the reagent grade water, prepared by any combination of distillation, reverse osmosis, ion exchange, activated carbon and particle filtration, meet the reference method requirements?		
M7: 7.1.6.h	Is the quality of the standard dilution water used for testing or culturing sufficient to allow satisfactory survival, growth and reproduction of the test species as demonstrated by routine reference toxicant tests and negative control performance?		
M7: 7.1.6.h	Is the water used for culturing and testing analyzed for toxic metals and organics whenever the minimum acceptability criteria for control survival, growth or reproduction are not met and no other cause, such as contaminated glassware or poor stock, can be identified?		

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M7: 7.1.6.i	Is the quality of the food used for testing or culturing sufficient to allow satisfactory survival, growth and reproduction of the test species as demonstrated by routine reference toxicant tests and negative control performance?		
M7: 7.1.6.i	Are there procedures for the evaluation of food acceptance?		
M7: 7.1.6.j	Is a subset of organisms used in bioaccumulation tests analyzed at the start of the test (i.e., baseline) for the target compounds to be measured in the bioaccumulation test?		
M7: 7.1.6.k	Do the test chamber size and test solution volume meet specifications in the reference method?		
M7: 7.1.6.k	Are all test chambers used in a test shall be identical?		
M7: 7.1.6.l	Are test organisms fed the quantity and type of food or nutrients specified in the reference method?		
M7: 7.1.6.l	Are the organisms fed at the intervals specified in the reference method?		
M7: 7.1.6.m	Are all organisms in a test from the same source and lot?		
M7: 7.1.6.m	Where available, are certified seeds used for soil tests?		
M7: 7.1.6.n	Do all organisms used in tests or used as broodstock to produce neonate test organisms (e.g., cladocerans and larval fish), appear healthy, show no signs of stress or disease and exhibit acceptable survival (90% or greater) during the 24-hour period immediately preceding use in tests?		
M7: 7.1.6.o	Are all materials that contact test samples (e.g., test chambers, culture tanks, tubing, solutions, control water, sediment, soil, or food) non-toxic and cleaned as described in the reference method?		
M7: 7.1.6.o	Do materials not reduce or add to sample toxicity?		
M7: 7.1.6.o	Are appropriate materials for use in toxicity testing and culturing described in the reference methods?		
M7: 7.1.6.p	Is light intensity maintained as specified in the reference method?		
M7: 7.1.6.p	Are measurements made and recorded on a yearly basis?		
M7: 7.1.6.p	Is photoperiod maintained as specified in the reference method and checked and recorded at least quarterly?		
M7: 7.1.6.p	For algal and plant tests, is the light intensity measured and recorded at the start of each test?		
M7: 7.1.6.q	Are records maintained of the health and culturing conditions of all organisms used for testing?		
M7: 7.1.6.q	Do these records include culture conditions (e.g., salinity, hardness, temperature, pH) and observations of any stress, disease, or mortality?		

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M7: 7.1.6.q	When organisms are obtained from an outside source, are records obtained of these water quality parameters and biological observations for each lot of organism received?		
M7: 7.1.6.q	Do these observations adequately address the 24-hour time period referenced in item 7.1.6 n) above?		
M7: 7.1.6.q	Are each of these observations recorded and water quality parameters recorded upon the arrival of the organisms at the testing laboratory?		
M7: 7.1.6.r	Are the age and the age range of the test organisms as specified in the reference method?		
M7: 7.1.6.r	Are supporting information, such as hatch dates and times, times of brood releases and metrics (for example, chironomid head capsule width) recorded?		
M7: 7.1.6.s	Does the maximum holding time of effluents (elapsed time from sample collection to first use in a test) not exceed 36 hours?		
M7: 7.1.6.s	Samples may be used for renewal up to 72 hours after first use except as specified by the reference method and approved by the regulatory agency having authority for program oversight.		Permission
M7: 7.1.6.t	Do all tests have at least the minimum number of replicates per treatment as specified by the reference method?		
M7: 7.1.6.u	Do the control population of Ceriodaphnia in chronic effluent or receiving water tests contain no more than 20% males?		
M7: 7.1.6.v	Is the culturing of C. dubia adequate such that blocking by parentage can be established?		
M7: 7.1.6.w	Are dissolved oxygen and pH in aquatic tests within acceptable range at test initiation?		
M7: 7.1.6.w	Is minimal aeration provided to tests if acceptable dissolved oxygen concentrations cannot be otherwise maintained?		
M7: 7.1.6.x	Are test soils or sediments within the geochemical tolerance range of the test organism?		
M7: 7.1.6.y	An individual test may be conditionally acceptable if temperature, dissolved oxygen, pH, or other specified conditions fall outside specifications, depending on the degree of the departure and the objectives of the tests (see test conditions and test acceptability criteria specified for each reference method).		Permission
M7: 7.1.6.y	Does the acceptability of the test depend on the experience and professional judgment of the technical director and the permitting authority?		
M7: 7.2	Data Acceptance/Rejection Criteria		

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M7: 7.2.1	Positive Controls		
M7: 7.2.1	Are the control performance and statistical endpoints recorded for each method and species on control charts?		
M7: 7.2.1	Is precision (i.e., coefficient of variation, CV) evaluated for these tests against reference method or laboratory-derived criteria to determine validity of the testing result?		
M7: 7.2.1	For endpoints that are point estimates (e.g., lethal concentration (LCp), inhibition concentration (ICp), or effective concentration (ECp)), are control charts constructed by plotting the cumulative mean and the control limits, which consist of the upper and lower 95% confidence limits (± 2 standard deviations)?		
M7: 7.2.1	For endpoints from hypothesis tests (e.g., no observed effect concentration (NOEC) or no observed adverse effect concentration (NOAEC)), are the values plotted directly, and do the control limits consist of one concentration interval above and below the concentration representing the central tendency (i.e., the mode)?		
M7: 7.2.1	For endpoints that are point estimates, is the cumulative mean CV calculated?		
M7: 7.2.1	For endpoints from hypothesis tests, is the PMSD calculated?		
M7: 7.2.1	Are these values maintained on control charts?		
M7: 7.2.1	Control chart limits are expected to be exceeded occasionally regardless of how well a laboratory performs. Acceptance limits for point estimates that are based on 95% confidence limits should theoretically be exceeded for one in twenty tests. Depending on the dilution factor and test sensitivity, control charts based on hypothesis test values (NOEC, NOAEC) may be expected to be exceeded on a similar frequency. Test results that fall outside of control chart limits at a frequency of 5% or less, or which fall just outside control chart limits (especially in the case of highly proficient laboratories which may develop relatively narrow acceptance limits over time), are not rejected de facto. Such data are evaluated in comparison with control chart characteristics including the width of the acceptance limits and the degree of departure of the value from acceptance limits.		Clarifying Statement
M7: 7.2.1	Are acceptance/rejection policies developed, consistent with the reference methods, for SRT data which considers source of test organisms, the direction of the deviation, test dilution factor, test sensitivity (for		

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	hypothesis test values), testing frequency, out-of-control test frequency, relative width of acceptance limits, inter-test CV, and degree of difference between test results and acceptance limits?		
M7: 7.2.1	In the case of reference toxicant data which fail to meet control chart acceptance criteria, is the test data examined for defects, corrective action taken, and the test repeated if necessary, using a different batch of organisms or the data is qualified?		
M7: 7.2.1	Is intra-laboratory precision determined on an ongoing basis using control charts?		
M7: 7.2.1	Are the control charts plotted as point estimate values, such as EC25 for chronic tests and LC50 for acute tests, or as appropriate hypothesis test values, such as the NOEC or NOAEC, over time within a laboratory?		
M7: 7.2.2	Negative Controls		
M7: 7.2.2	Does the test acceptability criteria specified in the reference method achieved for both the reference toxicant and the effluent or environmental sample toxicity test?		
M7: 7.2.2	Are the criteria calculated and meet the reference method requirements for performing toxicity tests?		
M7: 7.2.3	Selection of Appropriate Statistical Analysis Methods		
M7: 7.2.3.a	Are methods of data analysis and reporting as specified by language in the regulation, permit, or the reference method followed?		
M7: 7.2.3.b	Is toxicity data plotted on semi-logarithmic graph paper, relating time, mortality, and effluent concentration to verify computational results?		
M7: 7.2.4	Sample Handling		
M7: 7.2.4	Are all samples chilled to 0-6 °C during or immediately after collection except as specified by the reference method and approved by the regulatory agency having authority for program oversight?		