Designation: E1366 – 23

Standard Practice for Standardized Aquatic Microcosms: Fresh Water[1]

This standard is issued under the fixed designation E1366; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This practice covers procedures for obtaining data concerning toxicity and other effects of a test material to a multi-trophic level freshwater community, independent of the location of the test.

1.2 These procedures also might be useful for studying the fate of test materials and transformation products, although modifications and additional analytical procedures might be necessary.

1.3 Modification of these procedures might be justified by special needs or circumstances. Although using appropriate procedures is more important than following prescribed procedures, results of tests conducted using unusual procedures are not likely to be comparable to results of many other tests. Comparison of results obtained using modified and unmodified versions of these procedures might provide useful information concerning new concepts and procedures for conducting multi-trophic level tests.

1.4 This practice is arranged as follows:

1.5 The values stated in SI units are to be regarded as the standard. The values given in parentheses are for information only.

1.6 *This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety, health, and environmental practices and determine the applicability of regulatory limitations prior to use.*Specific hazard statements are given in Section 7.

1.7 *This international standard was developed in accordance with internationally recognized principles on standardization established in the Decision on Principles for the Development of International Standards, Guides and Recommendations issued by the World Trade Organization Technical Barriers to Trade (TBT) Committee.*

2. Referenced Documents

2.1 *ASTM Standards:*[2]

D1193 Specification for Reagent Water

D3978 Practice for Algal Growth Potential Testing with

[1] This practice is under the jurisdiction of ASTM Committee E50 on Environmental Assessment, Risk Management and Corrective Action and is the direct responsibility of Subcommittee E50.47 on Biological Effects and Environmental Fate.

Current edition approved June 1, 2023. Published July 2023. Originally approved in 1990. Last previous edition approved in 2016 as E1366 – 11(2016). DOI: 10.1520/E1366-23.

[2] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

Pseudokirchneriella subcapitata
E729 Guide for Conducting Acute Toxicity Tests on Test Materials with Fishes, Macroinvertebrates, and Amphibians
E943 Terminology Relating to Biological Effects and Environmental Fate
E1023 Guide for Assessing the Hazard of a Material to Aquatic Organisms and Their Uses
E1192 Guide for Conducting Acute Toxicity Tests on Aqueous Ambient Samples and Effluents with Fishes, Macroinvertebrates, and Amphibians
E1193 Guide for Conducting *Daphnia magna* Life-Cycle Toxicity Tests
E1733 Guide for Use of Lighting in Laboratory Testing
IEEE/SI 10 American National Standard for Use of the International System of Units (SI): The Modern Metric System

3. Terminology

3.1 The words "must," "should," "may," "can," and "might" have very specific meanings in this practice. "Must" is used to express an absolute requirement, that is, to state that the test ought to be designed to satisfy the specific condition, unless the purpose of the test requires a different design. "Must" is only used in connection with factors that directly relate to the acceptability of the test (see Section 17). "Should" is used to state that the specified condition is recommended and ought to be met in most tests. Although a violation of one "should" is rarely a serious matter, violation of several will often render the results questionable. Terms such as "is desirable," "is often desirable," and "might be desirable" are used in connection with less important factors. "May" is used to mean "is (are) allowed to," "can" is used to mean "is (are) able to," and "might" is used to mean "could possibly." Thus, the classic distinction between "may" and "can" is preserved, and "might" is never used as a synonym for either "may" or "can."

3.2 For definitions of other terms used in this practice, refer to Guide E729, Terminology E943, and Guide E1023. For an explanation of units and symbols, refer to IEEE/SI 10.

3.3 *Definitions of Terms Specific to This Standard:*

3.3.1 *algal biovolume, n*—an estimate of the total volume of algal cells ($\times 10^4$ μ^3/mL) (see 14.1.10).

3.3.2 *available algae, n*—an estimate of the volume of algae ($\times 10^4$ μ^3/mL) presumed available to the *Daphnia* (see 14.1.10).

3.3.2.1 *Discussion*—The estimate is calculated from the numerical abundance of each species of algae, its nominal volume, and an availability factor based on its size and growth characteristics (see 14.1.10). Small algal cells are presumed 100 % available and large, filamentous forms are presumed 1 to 20 % available. Species that attach to sediment or walls are presumed to be less available than planktonic forms.

3.3.3 *axenic, adj*—a culture of organisms growing without neighbors, that is, pure culture free from contaminant organisms (see gnotobiotic (**1–2**)[3]).

[3] Boldface numbers in parentheses refer to the list of references at the end of this practice.

3.3.4 *community metabolism, n*—the oxygen or carbon dioxide balance of the entire community.

3.3.4.1 *Discussion*—In this microcosm, community metabolism is estimated by the gain in oxygen during the lighted period (an estimate of net photosynthesis—*P*) and the loss of oxygen during the dark period (an estimate of respiration—*R*). When expressed as a *P/R* ratio, a value of >1 indicates that autotrophic processes are dominant; a value of <1 indicates that heterotrophic processes are dominant. If the difference of *P* and *R* are considered (*P-R*), a positive number indicates autotrophic processes are dominant, and a negative number indicates heterotrophic processes are dominant. Because P and R often change in the same direction and magnitude, P/R maybe less sensitive than P or R considered separately.

3.3.5 *detritivore, n*—an organism that feeds on detritus, dead organic material.

3.3.6 *ecosystem, n*—a system made up of a community of animals, plants, and bacteria and its interrelated physical and chemical environment **(3).**

3.3.7 *gnotobiotic, adj*—a culture which the exact composition of the organisms is known, down to the presence or absence of bacteria.

3.3.7.1 *Discussion*—Such cultures are developed from axenic cultures. The word implies know biota (**2**). The microcosms described here are not gnotobiotic because of the bacteria and other microbes are not known. An organism growing "without neighbors" is axenic (that is, free of all contaminants); growing with one organism is monoxenic (that is, the rotifers growing with one species of food bacteria); growing with two organisms is dixenic; growing with many organisms (provided the organisms are known) is gnotobiotic. A culture or community with many undefined organisms can be termed "xenic." The aquatic microcosms used in this practice are xenic because the bacterial component is undefined and contaminating organisms can enter. (Definitions are in accordance with (**1, 2**)).

3.3.8 *grazer, n*—an animal that grazes or feeds on growing plants; in these aquatic communities, organisms that feed on algae.

3.3.9 *herbivore, n*—an animal that feeds on plants, synonymous with grazer.

3.3.10 *medium, n*—the chemical solution (for example, T82MV) used in the microcosms.

3.3.11 *microcosm, n*—a small ecosystem that is regarded as miniature or epitome of a large world.

3.3.12 *primary producer, adj, n*—an organism capable of converting inorganic chemicals and energy into organic compounds.

3.3.12.1 *Discussion*—Primary producers are synonymous with autotrophs; in these microcosms they are the algae (including Cyanobacteria).

3.3.13 *secondary producer, adj, n*—an organism that requires organic chemicals for its energy source.

3.3.13.1 *Discussion*—Secondary producers are synonymous with heterotrophs; some researchers define grazers as secondary producers, and carnivores as tertiary producers. In these