



Standard Test Method for Estimating Acute Oral Toxicity in Rats¹

This standard is issued under the fixed designation E 1163; 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 test method determines the lethality (LD50 value) and signs of acute toxicity from a material using a limited number of rats. The technique used in this test method is referred to as the “Up-and-Down Procedure.”² This test method is an alternative to the classical LD50 test and is applicable to both liquids and solids.

1.2 This test method is not recommended for test materials which typically produce deaths beyond 2 days postdosing.

1.3 *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 and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 ASTM Standards:

E 609 Terminology Relating to Pesticides³

IEEE/ASTM SI 10 Standard for Use of the International System of Units (SI) (the Modernized Metric System)⁴

3. Terminology

3.1 Definitions:

3.1.1 *delayed death*—an animal which does not die or appear moribund within 24 h but dies later during the observation period.

3.1.2 *gavage*—forced feeding, as by a tube that is passed down the throat to the stomach.

3.1.3 *LD50*—the statistically derived estimate of the dose of a test substance that would be expected to cause 50 % mortality to the test population under the specified test conditions.

3.1.4 *moribund*—at the point of death or extinction.

3.1.5 *suspension*—a mixture in which very small particles remain suspended without dissolving.

3.1.6 *toxicity*—poisonous quality.

3.1.7 *signs of toxicity*—objective, observable evidence of toxicity.

3.1.8 *pharmacotoxic*—gross physiological signs in response to a toxic material.

4. Summary of Test Method

4.1 Female rats are dosed one at a time starting the first animal at the best estimate of the LD50. If this animal survives, then the next animal receives a higher dose; but if the first animal dies, the next animal receives a lower dose. If possible, subsequent doses are adjusted by a constant multiplicative factor, for example 1.3. The dose for each animal is adjusted up or down depending upon the outcome for the previous animal.

4.2 The dosing is repeated as above until four animals have been dosed after reversal of the initial outcome.

4.3 The LD50 is calculated using the maximum likelihood method.⁵

5. Significance and Use

5.1 This test method is of principal value in minimizing the number of animals required to estimate the acute oral toxicity (LD50).

5.2 This test method is inappropriate for materials typically producing death 2 or more days after administration of the test compound unless the observation time between dosages is increased. This test method can be successfully applied, however, for materials producing only an occasional death 2 or more days after administration.

5.3 The LD50 is valuable as a measure of the relative acute toxicity of a material and can be used to make an estimate of potential hazard to humans when pesticides, other chemicals, or mixtures are ingested.

5.4 This test method allows for observation of signs of toxicity in addition to mortality. This information can be useful in planning additional toxicity testing.

6. Apparatus

6.1 *Syringe*,⁶ and an oral dosing needle or rubberized catheter to gavage the test compound are required.

¹ This test method is under the jurisdiction of ASTM Committee E35 on Pesticides and Alternative Control Agents and is the direct responsibility of Subcommittee E35.26 on Safety to Man.

Current edition approved Oct. 10, 2002. Published March 2003. Originally approved in 1987. Last previous edition approved in 1998 as E 1163 – 98.

² Bruce, R. D., “An Up-and-Down Procedure for Acute Toxicity Testing,” *Fundamental and Applied Toxicology*, Vol 5, 1985, pp. 151–157.

³ *Annual Book of ASTM Standards*, Vol 14.02.

⁴ *Annual Book of ASTM Standards*, Vol 11.05.

⁵ Finney, D. J., *Probit Analysis*, 3rd ed., Cambridge University Press, Cambridge, England, 1971, pp. 50–80.

⁶ Hamilton Milliliter syringes available from Hamilton Co., Reno, NV have been found suitable for this purpose.