



Standard Practice for Guinea Pig: Split Adjuvant and Closed Patch Testing for Contact Allergens¹

This standard is issued under the fixed designation F 2147; 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 is intended to determine the potential for a substance, or material extract, to elicit contact dermal allergenicity.

1.2 This practice is intended as an alternative to the Guinea Pig Maximization Test (GPMT), given the limitations on dosage form and tendency for false positives associated with the latter test. See Rationale and References.

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:*

F 619 Practice for Extraction of Medical Plastics²

F 720 Practice for Testing Guinea Pigs for Contact Allergens: Guinea Pig Maximization Test²

2.2 *ISO Document:*

ISO 10993-10, 1995 Tests for Irritation and Sensitization³

3. Terminology

3.1 *Definitions:*

3.1.1 *2,4 dinitrochlorobenzene (DNCB)*—strong sensitizer, used as a positive control.

3.1.2 *Freund's Complete Adjuvant (FCA)*—a commercially-available mixture of oil and *Mycobacterium* that is known to elicit an immune response.

3.1.3 *Guinea Pig Maximization Test (GPMT)*—procedure described in Practice F 720 accepted as a “worst case” assay for allergenic potential.

4. Summary of Practice

4.1 The split adjuvant method is used when topical application is considered relevant, and the dosage form is a solid,

liquid, extract, paste, or gel. The method includes four induction doses applied over ten days to the same shaved or depilated site on guinea pigs, followed by occlusive patching. Freund's Complete Adjuvant (FCA) is injected near the dose site on the fourth day (second induction dose). Following a rest period, animals are challenged at a previously unexposed site, and the reaction evaluated at 24, 48, and 72 h.

4.2 The closed patch method is used when topical application is relevant, but the preferred dosage form does not permit injection under the skin or intradermally, and the discomfort involved with extended occlusive patching and adjuvant use is to be avoided. It involves repeated induction doses (3 to 6) over 14 days at the same shaved/depilated site, followed each time by 6 h of occlusive wrapping. After a rest period, animals are challenged at previously untreated sites, and their reactions evaluated at least 24 and 48 h later.

5. Significance and Use

5.1 In selecting a material for human contact in medical applications, it is important to ensure the material will not stimulate the immune system to produce an allergic reaction under relevant exposure conditions. Extractable chemicals produced by skin contact or during physiological exposures may cause allergic reactions. Therefore, this practice provides for evaluations of solid or semisolid dosage forms using material extracts or direct evaluation of the test article. The rationale for this animal model is based on the fact that the guinea pig has been shown to be an appropriate animal model for predicting human contact dermatitis; its tractable nature, its availability from reputable suppliers, the historical database of information already acquired using this species, and the correlation of such results to data on known human allergens, all contribute to its widespread use for allergenicity studies **(1-5)**.⁴

5.2 The need for sensitization procedures other than the maximization test (Practice F 720) is based on: **(1)** the need for a route of exposure more similar to use conditions, **(2)** concern over the use of adjuvant because of its recruitment of cell types to the test site which are not typically involved in immunologic reactions, and because of the discomfort this causes in the animals, **(3)** absence of a proper FCA-irritant control group in

¹ This practice is under the jurisdiction of ASTM Committee F04 on Medical and Surgical Materials and Devices and is the direct responsibility of Subcommittee F04.16 on Biocompatibility Test Methods.

Current edition approved Oct. 10, 2001. Published January 2002.

² *Annual Book of ASTM Standards*, Vol 13.01.

³ Available from American National Standards Institute, 25 W. 43rd St., 4th Floor, New York, NY 10036.

⁴ The boldface numbers in parentheses refer to the list of references at the end of this standard.