(3) Incubate the monolayer at 4 °C for 30 minutes, wash with phosphate buffered saline, and examine for hemadsorption.

(4) If no hemadsorption is apparent, repeat step (b)(2) of this section and incubate the monolayers at 20–25 °C for 30 minutes, wash with phosphate buffered saline, and examine again for hemadsorption. If desired, separate monolayers may be used for each incubation temperature.

(c) If specific cytopathology or hemadsorption attributable to an extraneous agent is found, the material under test is unsatisfactory and shall not be used to prepare biological products. If an extraneous agent is suspected because of cytopathology or hemadsorption and cannot be eliminated as a possibility by additional testing, the material under test is unsatisfactory.

[50 FR 441, Jan. 4, 1985, as amended at 58 FR 50252, Sept. 27, 1993]

§ 113.47 Detection of extraneous viruses by the fluorescent antibody technique.

The test for detection of extraneous viruses by the fluorescent antibody technique provided in this section shall be conducted when prescribed in an applicable Standard Requirement or in a filed Outline of Production for a product.

(a) Monolayer cultures of cells (monolayers), at least 7 days after the last subculturing, shall be processed and stained with the appropriate antiviral fluorochrome-conjugated antibody as specified in paragraph (b) of this section.

(i) Three groups of one or more monolayers shall be required for each specific virus prescribed in paragraph (b) of this section.

(ii) At the time of the last subculturing, one group of test monolayers shall be inoculated with approximately 100–300 FAID50 of the specific virus being tested for as positive controls.

(iii) One group of monolayers shall be the “material under test.”

(b) The antiviral fluorochrome-conjugated antibodies to be used shall depend on the type of cells required to be tested for extraneous viruses as specified in an applicable Standard Requirement or in a filed Outline of Production. Antiviral fluorochrome-conjugated antibodies specific for the extraneous viruses shall be applied to each respective type of cell in accordance with the following list. Under certain circumstances, additional tests may need to be conducted, as determined by the Administrator. When a specific antiviral fluorochrome-conjugated antibody is used in testing for the listed extraneous viruses specified in more than one cell type, it need only be applied to the most susceptible cell type.

(i) All cells shall be tested for:

(ii) Bovine virus diarrhea virus;

(iii) Reovirus;

(iv) Rabies virus.

(ii) Bovine, caprine, and ovine cells shall, in addition, be tested for:

(i) Bluetongue virus;

(ii) Bovine adenoviruses;

(iii) Bovine parvovirus; and

(iv) Bovine respiratory syncytial virus.

(iii) Canine cells shall, in addition, be tested for:

(i) Canine coronavirus;

(ii) Canine distemper virus; and

(iii) Canine parvovirus.

(iv) Equine cells shall, in addition, be tested for:
§ 113.50 Ingredients of biological products.

All ingredients used in a licensed biological product shall meet accepted standards of purity and quality; shall be sufficiently nontoxic so that the amount present in the recommended dose of the product shall not be toxic to the recipient; and in the combinations used shall not denature the specific substances in the product below the minimum acceptable potency within the dating period when stored at the recommended temperature.

[38 FR 29889, Oct. 30, 1973]

§ 113.51 Requirements for primary cells used for production of biologics.

Primary cells used to prepare biological products shall be derived from normal tissue of healthy animals. When prescribed in an applicable Standard Requirement or in the filed Outline of Production, each batch of primary cells used to prepare a biological product shall be tested as prescribed in this section. A batch of primary cells found unsatisfactory by any prescribed test shall not be used. A serial of biological product shall not be released if produced from primary cells that are found unsatisfactory by any prescribed test.

(a) Final container samples of completed product or samples of the final pool of harvested material or samples of each subculture of cells used to prepare the biological product shall be shown free of mycoplasma as prescribed in § 113.28. The sample for testing shall consist of at least 75 cm² of actively growing cells or the equivalent in harvest fluids; Provided, That, all sources of cells in the batch of primary cells are represented.

(b) Final container samples of completed product or samples of the final pool of harvested material or samples of each subculture of cells used to prepare the biological product shall be shown free of bacteria and fungi as prescribed in §113.26 or §113.27 (whichever is applicable).

(c) A monolayer at least 75 cm² from each batch of primary cells or each subculture of primary cells used to prepare a biological product shall be shown free of extraneous agents as prescribed in this paragraph.

(1) The test monolayer shall be maintained using the medium (with additives) and under conditions similar to those used to prepare biological products.