§ 113.115 Staphylococcus Aureus Bacterin-Toxoid.

Staphylococcus Aureus Bacterin-Toxoid shall be prepared from toxoided broth cultures of selected toxigenic strains of Staphylococcus aureus which has been inactivated and is nontoxic. Each serial of biological product containing Staphylococcus Aureus Bacterin-Toxoid shall meet the applicable requirements in §113.100 and shall be tested for purity, safety, and potency as prescribed in this section. A serial found unsatisfactory by any prescribed test shall not be released.

(a) Purity test. Final container samples of completed product from each serial shall be tested for viable bacteria and fungi as provided in §113.26.

(b) Safety test. Bulk or final container samples of completed product shall be tested for safety as provided in §113.33(b). Also, the rabbits used in the potency test provided in paragraph (c) of this section shall constitute an additional safety test. If unfavorable reactions attributable to the product occur in any of the rabbits during the observation period, the serial is unsatisfactory.

(c) Potency test. Rabbits, each weighing 2000–3000 grams, shall be used as test animals. Either a five rabbit individual serum test or an eight rabbit pooled serum test shall be conducted. At the start of the test, individual sera from the five rabbits or pooled sera from the eight rabbits shall contain less than 0.2 alpha antitoxin units per ml.

(1) Each rabbit shall be given a series of not more than three intramuscular injections at 7 day intervals (1.0 ml, 2.0 ml, 3.0 ml) and observed from 7–14 days following the third injection. At the end of the observation period, a blood sample shall be taken from each rabbit.

(2) The sample of serum from each rabbit, if the five rabbit individual test shall be conducted, shall be tested to determine the staphylococcus alpha antitoxin units per ml as provided in paragraphs (c)(3), (4), (5), (6), (7), and (8) of this section.

(3) Inactivate rabbit serum 56°C for 30 minutes.

(4) Make serial twofold dilutions of the serum samples and conduct the test, using 1 ml of the serial dilutions. Appropriate controls should be included for accurate interpretations.

(5) Add 1 ml of the standardized toxin containing the established “Lh” dose. The “Lh” dose is the amount of toxin which when mixed with one unit of standard antitoxin produces a 50 percent hemolysis of rabbit red blood cells.

(6) Incubate toxin-antitoxin mixture at room temperature for 30 minutes and add 1 ml of a 1.5 percent suspension of washed freshly drawn rabbit red blood cells suspended in normal saline to each tube. Mix and incubate the combined product in a 37°C water bath for 1 hour. Refrigerate at 5°C overnight.

(7) Read the hemolysis produced and establish the 50 percent end point. The 50 percent end point of hemolysis should be established by determining the size of the button produced by the unlysed red blood cells.

(8) Determine the units of antitoxin per 1 ml of serum.

(9) If the individual samples from four of the five rabbits in the individual serum test or the pooled samples from the eight rabbits in the pooled serum test do not contain three alpha antitoxin units per ml, the serial is unsatisfactory.