

pads used to collect the samples (i.e., one pad per 5 chick box papers) into an 18 oz. Whirl-Pak bag and add 1 to 2 tablespoons of double-strength skim milk.

(v) Promptly refrigerate the Whirl-Pak bags containing the samples and transport them, on ice or otherwise refrigerated, to a laboratory within 48 hours of collection. The samples may be frozen for longer storage if the Plan participant is unable to transport them to a laboratory within 48 hours.

(2) The Plan participant may send chick box papers directly to a laboratory, where samples may be collected as described in paragraph (c)(1) of this section. To send chick box papers directly to a laboratory, the Plan participant shall:

(i) Collect 1 chick box paper for each 10 boxes of chicks placed in a house and place the chick papers immediately into large plastic bags and seal the bags.

(ii) Place the plastic bags containing the chick box papers in a clean box and transport them within 48 hours to a laboratory. The plastic bags do not require refrigeration.

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[38 FR 13709, May 24, 1973. Redesignated at 44 FR 61586, Oct. 26, 1979, and amended at 57 FR 57342, Dec. 4, 1992; 59 FR 12805, Mar. 18, 1994; 59 FR 67617, Dec. 30, 1994; 61 FR 11524, Mar. 21, 1996; 62 FR 44070, Aug. 19, 1997; 63 FR 3, Jan. 2, 1998]

§147.13 Procedure for bacteriological culturing of eggshells for colon bacilli organisms.

Proper precautions to avoid environmental contamination of the samples during the collection and laboratory process, and proper handling of the samples following collection are essential. Each State Inspector involved in eggshell culture activities must receive instruction in the necessary sanitation procedures, sampling procedures, and sample handling by the authorized laboratory involved. The Official State Agency will maintain a record showing that the required instruction was given to each State Inspector.

(a) *Sample selection.* Forty (40) eggs in the top flats of each of three randomly selected cases of sanitized eggs from

each flock will be utilized for each sampling.

(b) *Swab procedure.* A 2.5 centimeter diameter circular area of the large end of each of the eggs will be rubbed with a sterile swab previously moistened with sterile lactose broth, or other suitable liquid media provided by the authorized laboratory. One swab will be used for five eggs, and four swabs will be pooled to each sterile, capped tube provided by the authorized laboratory.

(1) From the tube containing four swabs and lactose broth or other suitable media, 1 ml. will be transferred to 10 ml. lactose in a fermentation tube.

(2) Incubate at 37° C for 48 hours. The presence of acid, and gas in the amount of 10 percent or more after 24 and 48 hours of incubation, provides a presumptive conclusion of the presence of colon bacilli organisms.

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§147.14 Procedures to determine status and effectiveness of sanitation monitored program.

The following monitoring procedures¹³ may be applied at the discretion of the Official State Agency:

(a) Monitor effectiveness of sanitation program.

(1) Culture the surface of cased eggs periodically for fecal contaminating organisms as described in §147.13.

(2) Culture a sample of dead-in-shell eggs periodically from each breeding flock for coliforms. Such eggs should also be cultured for the dependable recovery of *salmonellae*. Culturing for the dependable recovery of *salmonellae* should include the use of:

¹³Laboratory procedures for monitoring operations proposed here are described in the following two publications: Isolation and Identification of Avian Pathogens, American Association of Avian Pathologists, University of Pennsylvania, New Bolton Center, Kennett Square, Pennsylvania 19348–1692, 1980, and Culture Methods for the Detection of Animal Salmonellosis and Arizonosis, Iowa State University Press, Ames, Iowa 50010, 1976.