

DEPARTMENT OF HEALTH AND HUMAN SERVICES PUBLIC HEALTH SERVICE FOOD AND DRUG ADMINISTRATION MILK LABORATORY EVALUATION FORM	LABORATORY	
	LOCATION	LAB #
	DATE	X = DEVIATION O = NOT USED U = UNDETERMINED NA = NOT APPLICABLE

FLAT LID METHODS
 [Unless otherwise stated all tolerances are ±5%]

- 1. Laboratory Requirements**
- a. Record time and date samples received
- b. Record time and date samples examined

POUR CONTACT METHOD
APPARATUS AND MATERIALS

- 2. CP, items 1-32 (as necessary)**
- 3. Forceps, sterile**
- a. 140 mm hemostatic type preferred
- 4. Tweezers, sterile**
- 5. Petri dishes, sterile**
- a. 15 x 100 mm for lids up to 8 cm diameter
- b. 15 x 150 mm for lids up to 13 cm diameter
- 6. Plate count agar, see CP item 27b**
- 7. 70% ethyl alcohol**
- a. In covered container large enough to hold forceps and tweezers
- 8. Incubator, 32±1C, see CP item 15**

PROCEDURE

- 9. Number of lids examined is the square root of the number of items in the package, to a maximum of 21**
- 10. Identify petri dishes (see SPC, item 4)**
- 11. Food contact surface extends beyond lip**
- a. Pour PCA (SPC, item 13) into dishes to a depth of 3 mm and allow to harden
- b. Using sterile forceps remove and discard end unit from package
- c. Carefully sliding stack from package, remove a lid periodically with sterile forceps
- d. Place lid on agar with food contact surface in contact with agar, pressing lid against agar to ensure contact
- e. Repeat until required number of lids have been selected
- f. Incubate plates for 24 hours at 32±1C
- g. Remove lid from each plate, incubate plate another 24 hours
- 12. Food Contact Surface Recessed**
- a. Select lids as in 11 b&c
- b. Place lid in dish with food contact surface up using sterile forceps
- c. Pour PCA (SPC item 13) into the lid to a depth of 3 mm if possible
- d. Incubate for 24 hours at 32±1C
- e. With sterile forceps, remove lid from dish and slip agar out of lid into dish with the lid contact side of agar up. Sterile tweezers may be used to loosen agar from lid
- f. Incubate dishes for another 24 hours at 32±1C
- 13. If lid diameter is >13 cm or constructed so that full agar contact is impossible, use swab test (1 lid/swab)**
- 14. Coliform test for flat lids (all sizes)**
- a. Use swab method (items 19-34)

- 15. Controls (see SPC item 14)**
- a. Check sterility of agar, petri dishes and forceps used for each group of samples
- b. Expose a poured plate to air for each group of samples
- 16. Counting colonies**
- a. See SPC, items 16-18
- b. Count after 48±3 hours incubation
- 17. Calculations**
- a. Determine food contact area in sq cm
- b. Divide number of colonies/lid by area
- 18. Report**
- a. Report the number of colonies/sq cm for each lid

SWAB METHOD
APPARATUS AND MATERIALS

- 19. See items 2-8**
- 20. Buffered rinse solution (see CP item 27i)**
- 21. Sodium hexametaphosphate or Na citrate, 7% solution**
- 22. Screw-capped vials**
- a. 7 to 10 cm long to contain 7 mL solution
- b. Containing 6 mL rinse solution
- c. Sterile
- 23. Swabs**
- a. Non-toxic tested using *B. stearothermophilus* type assay
1. Test each lot by swirling several swabs in 5 mL of sterile dilution buffer, test using *Bacillus stearothermophilus* type assay
2. Maintain records
- b. Calcium alginate fibers on wood applicator stick
- c. Packaged in convenient protective containers and sterilized
- 24. M-endo broth agar (see CP item 27m)**
- a. Dispense in membrane filter petri dishes, 4 - 5 mL/dish
- 25. Membrane filters**
- a. 0.45 µm pore size, 47 mm diameter
- b. Sterile
- 26. Incubator, 35±1C**

PROCEDURE

- 27. Sample size, 35 lids/unit package**
- a. See item 11 b&c for selection procedure
- 28. Identify plates (see SPC item 4)**
- 29. Collection of swab samples**
- a. Aseptically remove sterile swab from container
- b. Open vial of solution, wet swab and press out excess solution
- c. Holding swab at 30° angle to surface, rub over entire food contact surface
- d. Position swab head in vial and break stick, leaving swab head in vial
- e. Repeat a-d for remainder of lids (34 vials)

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FLAT LID METHODS
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- f. Repeat a-e with a second set of 35 lids for coliform determination (5 lids/vial)
- 30. Sample measurement - SPC**
- a. As described in SPC, item 9, except:
- b. Add 1 mL sterile Na hexa-metaphosphate or Na citrate solution to each vial
- c. Shake vigorously until swabs dissolve
- d. Transfer vial contents to 2 plates
- 31. Sample measurement - coliforms**
- a. Add 1 mL sterile Na hexa-metaphosphate or Na citrate solution to each vial
- b. Shake vigorously until swabs dissolve
- c. Add additional 1 mL phosphate or citrate solution and filter through membrane filter (item 25)
- d. Rinse filter and holder with sterile buffer (CP item 25)
- e. Transfer filter to M-endo broth agar plate
- 32. Plating (see SPC item 13)**
- 33. Controls (see SPC item 14)**
- a. Check sterility of agars, petri dishes, rinse solution and swabs for each group of samples
- b. Expose a poured plate to air for each group of samples
- 34. Incubation**
- a. See SPC, item 15
- b. Coliforms, 35±1C for 18-24 hours
- 35. Counting Colonies**
- a. See SPC items 17
- b. Coliforms, count typical dark red colonies and those with green metallic sheen
- 36. Calculations**
- a. Determine food contact area in sq cm
- b. Add the colonies for 2 plates and divide by area for SPC
- 37. Reporting**
- a. Report SPC as number of colonies/sq cm
- b. Report coliforms as number of colonies/lid

ALTERNATE SWAB METHOD
APPARATUS AND MATERIALS

- 38. See items 2-8, 20-21, 23**
- 39. Screw-capped Vials**
- a. 7 to 10 cm long to contain 10 mL of solution
- b. Containing 9 mL of rinse solution
- c. Sterile
- 40. Plate Count Agar (see CP item 27b)**
- 41. Violate Red Bile Agar (see CP item 27d)**

PROCEDURE

- 42. See items 27-28**
- 43. Collection of Swab Samples**
- a. Aseptically remove sterile swab from container
- b. Open vial of solution, wet swab and press out excess solution
- c. Holding swab at 30° angle to surface, rub over entire food contact area
- d. Repeat b - c for remainder of lids (34)
- e. Position swab head in vial and break stick, leaving swab head in vial
- 44. Sample Measurement - SPC and coliforms**
- a. As described in SPC item 9, except:
- b. Add 1 mL of sterile Na citrate solution to vial (see item 39)
- c. Shake vigorously until swab dissolves
- d. Transfer 2 mL of vial contents to each of two (2) petri dishes
- 45. Plating (see SPC item 13)**
- a. To one plate add SPC agar and to the other VRB agar
- 46. Controls (see SPC item 14)**
- 47. Incubation (see SPC item 15)**
- 48. Counting Colonies (see SPC items 16-18)**
- 49. Calculations**
- a. Determine food contact area in sq. cm
- b. Multiply number of colonies on each plate by dilution factor of 5, divide by area of lid determined in item a
- 50. Reporting**
- a. Report SPC and coliforms as number of colonies/sq. cm