

**PASTEURIZED MILK CONTAINERS, CLOSURES AND PACKAGING
IMS #22**

[Unless otherwise stated all tolerances are $\pm 5\%$]

1. Laboratory Requirements _____

- a. Record time and date when samples received _____
- b. Record time and date when samples examined _____

RINSE METHOD APPARATUS

2. See Cultural Procedures (CP) items 1-23 _____

3. To Add Rinse Solution to Containers _____

- a. Sterile hypodermic syringes (capacity 20 or 100 mL) and needles _____
- b. Or, sterile pipets _____
- c. Or, sterile automatic syringe _____
- d. Or, sterile graduated cylinder _____
- e. Or, pre-dispensed dilution bottles or tubes with rinse solution (see CP item 29.f); volumes checked _____

MATERIALS

4. See CP items 24-32 _____

5. Rinse Solutions _____

- a. Buffered Rinse Solution or Nutrient broth (see CP items 27.i-j) for Standard Plate Count (SPC) and Coliform Plate Count (CPC) agar based media _____
- b. Nutrient broth (see CP item 27.m) for 3M™ Petrifilm™ Aerobic Count (PAC), Coliform Count (PCC) and High Sensitivity Coliform Count (HSCC) plates, Charm® Peel Plate® Aerobic Count (PPAC), Coliform Count (PPEC) and High Volume Sensitivity Coliform Count (PPECHVS) _____

6. Ethyl Alcohol, 70% _____

7. Plastic Tape _____

PROCEDURE

8. Identify Plates (See SPC item 5, Petrifilm item 6 or Peel Plate item 5) _____

9. Controls (See SPC item 6, Petrifilm item 7 or Peel Plate item 6), in addition; _____

a. Transfer 1 mL of rinse solution to SPC, PAC or PPAC plate for sterility control _____

10. Rinse Solution Volumes for Collection of Surface Rinse Samples _____

a. 100 mL (± 2 mL) for gallons (3784 mL) or larger _____

b. 50 mL (± 1 mL) for $\frac{1}{2}$ gallons (1892 mL) _____

c. 20 mL (± 0.4 mL) for 100 mL to $\frac{1}{2}$ pints (236 mL), pints (473 mL), and quarts (946 mL) _____

d. For containers < 100 mL and closures use swab method, see items 18-32 _____

e. Irregular shaped containers of < 100 mL, use rinse method in item 10.c. Equally distribute the 20 mL among multiple units with the amount per unit no more than 20% of the volume _____

11. Collection of Surface Rinse Samples _____

a. Firm walled paper containers, sealed on line _____

1. Swab top of containers with 70% alcohol at the site of injection _____

2. Add required amount of rinse solution to each container by injection and seal puncture with plastic tape _____

3. Vigorously shake container length-wise on flat sides (or quadrants of round containers) 10 times, holding container horizontally _____

4. Each shake a complete back and forth movement of approximately 20 cm _____

5. Turn container 90° and repeat horizontal shaking treatment _____

6. Turn container 90° twice more and repeat horizontal shaking _____

7. Grasp container and swirl 20 times in a small flat circle while upright (top up) _____

8. Invert (top down) and repeat swirling of container 20 times _____

9. Stand upright and allow to drain for 1-3 min _____

b. Plastic capped containers (submitted with caps) _____

1. Swab top of container with 70% alcohol when appropriate _____

2. Add required amount of rinse solution by aseptically removing cap, pouring in solution without touching the top and replace cap _____

3. Complete rinse procedure as described in 11.a.3-9 above _____

c. Flexible-walled containers/bags _____

1. Add 100 mL aseptically by swabbing an area of tube adjacent to liner with 70% alcohol; introduce rinse by syringe and seal puncture with plastic tape _____
2. Place container/bag on smooth, clean, firm horizontal surface as flat as its construction permits _____
3. With hands or roller, move rinse solution back and forth 10 times, contacting all surfaces completely _____
4. Lift liner and hang with "fill tube" down to permit rinse solution to collect for 1-3 min _____
5. Transfer rinse solution to sterile container by cutting "fill tube" with sterile scissors _____

d. Irregular shaped containers of <100 mL _____

1. Swab top of container with 70% alcohol when appropriate e.g. at injection site _____
2. Aseptically add required amount of rinse solution to each container, seal with cap or appropriate sterile closure _____
3. Complete rinse procedure as described in 11.a.3-9 above _____
4. Transfer rinse solutions of the multiple containers in sequence by aseptically removing cap or sterile closure, pouring solution into a common sterile container without touching the tops and replacing cap or sterile closure on the sterile container _____

12. Sample Measurements _____

a. As described in SPC items 9 & 10, Petrifilm items 10 & 11 or Peel Plate items 9 & 10, except: _____

1. For Residual Bacterial Count (RBC), pipet 2 mL portion in a single SPC plate or pipet two 1 mL portions on 2 PAC or 2 PPAC plates _____
2. For Residual Coliform Count (RCC), pipet 10 mL of remaining rinse solution among 3 CPC plates, or pipet ten 1 mL portions of remaining rinse solution on 10 PCC or PPEC plates or two 5 mL portions on 2 HSCC or PPECHVS plates _____

13. Pouring Agar (See SPC item 13) _____

14. Incubating Plates (See SPC item 14, Petrifilm item 13 or Peel Plate item 13) _____

15. Confirmation Test for CPC (See SPC item 17.c)

**16. Counting and Recording Colonies
(See SPC items 15-17, Petrifilm items 14-16 or Peel Plate items 14-16)**

- a. Count obtained from RBC plate(s) recorded as colonies counted
- b. If no colonies on RBC plate(s), record as 0
- c. Count obtained from RCC plates recorded as colonies counted
- d. If no colonies on RCC plates, record as 0
- e. Values are recorded as number of colonies per container

REPORTS

17. Reporting Counts

- a. Report computed bacterial count as RBC/container
 - 1. Containers rinsed with 20 mL
 - a. 2 mL plated for RBC, multiply colony count by 10
 - 2. Containers rinsed with 50 mL
 - a. 2 mL plated for RBC, multiply colony count by 25
 - 3. Containers rinsed with 100 mL
 - a. 2 mL plated for RBC, multiply colony count by 50
- b. Report computed coliform count as RCC/container
 - 1. Containers rinsed with 20 mL
 - a. 10 mL plated for RCC, multiply colony count by 2
 - 2. Containers rinsed with 50 mL
 - a. 10 mL plated for RCC, multiply colony count by 5
 - 3. Containers rinsed with 100 mL
 - a. 10 mL plated for RCC, multiply colony count by 10
- c. If no colonies appear on plate(s), report as less than n/container, substituting for n the number that would be reported if 1 colony had been counted from the volume of rinse solution plated and multiplied by appropriate factor

SWAB METHOD

APPARATUS

18. See CP items 1-23

19. Screw-capped Containers

a. 7 to 10 cm long to contain:

1. 5 mL rinse solution for non-soluble swabs (see item 5)

2. 4.5 mL rinse solution for alginate swabs (see item 5, SPC & CPC only)

b. Sterile

20. Swabs

a. Cotton, non-absorbent (firmly twisted to about 5 mm diameter by 2 cm long over one end of applicator stick 12-15 cm long)

b. Or, calcium alginate fibers (SPC & CPC only)

c. Or, polyester or rayon fibers

d. Commercial source, sterile, non-toxic in protected containers

1. Supporting documentation from manufacturer

2. Maintain records

MATERIALS

21. See Items 4 & 5

22. Sodium Hexa-metaphosphate Solution, 10% (if calcium alginate swabs used, SPC & CPC only), sterile

23. Shaking Machine, optional (See SPC item 8.c, PAC item 9.c or PPAC item 8.c)

PROCEDURE

24. Identify Plates (See SPC item 5, Petrifilm item 6 or Peel Plate item 5)

25. Controls (See SPC item 6, or Petrifilm item 7 or Peel Plate item 6), in addition;

a. Pipet 1 mL of rinse solution to SPC, PAC or PPAC plate for sterility control

- b. For calcium alginate swab, break off swab head in container with 4.5 mL rinse solution plus 0.5 mL Na Hexa-metaphosphate solution and continue as described in 27.a.1, pipetting 1 mL rinse solution to plate for RBC sterility control of swab and bottle _____
- c. For all other fibers, break off swab head in container with 5 mL rinse solution and continue as described in item 27.a.2 & 27.b, pipetting 1 mL rinse solution to plate for RBC sterility control of swab and bottle _____

26. Collection of Swab Samples from Product Contact Surfaces _____

- a. 250 sq. cm of product contact surface must be swabbed or five 50 sq. cm for a total of 250 sq. cm (calculate or use template – must be sterile if swab will be in contact with template) _____
- b. Aseptically remove sterile swab from container _____
- c. Open vial of solution, wet swab and press out excess solution _____
- d. Holding swab at 30° angle to surface, rub over 50 sq. cm area three times, reversing direction between successive strokes _____
 - 1. For snap or screw cap closures, calculate number of closures required for product contact surface area of 50 sq. cm _____
 - 2. For cup shaped containers, determine 50 sq. cm for the product contact surface _____
- e. Rinse swab in solution and press out excess _____
- f. Swab four additional 50 sq. cm areas _____
- g. After fifth area has been swabbed, position swab head in vial and break stick, leaving swab head in vial _____

27. Sample Measurement _____

- a. As described in SPC items 9 & 10; _____
 - 1. For calcium alginate, add 0.5 mL of sterile Na Hexa-metaphosphate solution (see item 22) to 4.5 mL rinse solution in vial and shake until dissolved [**Not acceptable for use with 3M Petrifilm or Charm Peel Plate**] _____
 - 2. For all other fibers: _____
 - a. Shake swab container 50 times _____
 - b. Each shake a complete back and forth movement of approximately 15 cm _____

c. Strike palm of hand at end of each cycle _____

d. Complete shaking in approximately 10 sec _____

b. As described in Petrifilm items 10 & 11 or Peel Plate items 9 & 10; _____

1. Shake swab container 50 times _____

2. Each shake a complete back and forth movement of approximately 15 cm _____

3. Strike palm of hand at end of each cycle _____

4. Complete shaking in approximately 10 sec _____

c. For RBC, pipet 1 mL portion to a single SPC, PAC or PPAC plate _____

d. For RCC, pipet 3 mL to a single CPC plate or three 1 mL portions on three PCC or PPEC plates _____

28. Pouring Agar (See SPC item 13) _____

29. Incubation (See SPC item 14, Petrifilm item 13 or Peel Plate item 12) _____

30. Confirmation for CPC test (See SPC item 17.c) _____

**31. Counting and Recording Colonies
(See SPC items 15-17, Petrifilm items 14-16 or Peel Plate items 14-16)** _____

a. Count obtained from RBC plates, record as colonies counted _____

b. If no colonies on RBC plates, record as 0 _____

c. Count obtained from RCC plate(s) record as colonies counted _____

d. If no colonies on RCC plate(s), record as 0 _____

REPORTS

32. Reporting Counts _____

a. Report the count in 31.a as the RBC/50 sq. cm _____

b. If no colonies on RBC plate, report as < 1/50 sq. cm _____

c. Report the count in 31.c as the RCC _____

d. If no colonies on RCC plate(s), report as < 1 _____