DISINTEGRATION METHOD FOR PAPER, PAPERBOARD OR MOLDED PULP MATERIALS

[Unless otherwise stated all tolerances are ±5%]

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

APPARATUS AND MATERIALS

2. See Cultural Procedures, items 1-3, 10-21, 23-32 as appropriate .................................................................

3. Pipets, sterile ..............................................................................
   a. Capacity, 10 mL with 3 mm tip opening ..............................
   b. Or, 20 mL with large-bore tip opening ..............................

4. Pipet containers ........................................................................
   a. Used for sterilization, storage, non-toxic ..........................

5. Scalpel or scissors, and forceps sterile ......................................

6. Disintegrator blender ................................................................
   a. Sterile, high speed, electrically operated, corrosion-resistant cup .................................................................
   b. Capacity, 500 mL; optionally, 1000 mL ............................

7. 70% ethyl alcohol ......................................................................
   a. In a covered container to hold scalpels, scissors and forceps

8. Dilution buffer (Cultural Procedures, item 25a,c) .................
   a. In containers filled to contain 300±6 mL (or 500±10 mL) ...

9. Sterile kraft paper or envelopes ................................................

PROCEDURE

10. Not applicable when wax, plastic or metal is food contact surface .................................................................

11. Use sterile cutting device, cut 100g from butt roll and transfer to sterile wrapper or envelope ............................

12. With sterile cutting device, trim off 5 cm of the outer edge of the sample sheet .............................................

13. Handling with sterile forceps, cut into 0.5 cm pieces 3g of the center portion into a sterile petri dish ..................

14. Transfer this 3g into a sterile disintegrator cup containing 300 mL dilution water (5g in 500 mL) .................

15. Place cup on blender motor, run at high-speed 30 sec, check to insure no particles are on side of cup or trapped beneath blade .................................................................

16. Continue high speed blending for a total of 2 min, depending on paper type, checking at intervals for particles on side of cup .................................................................

17. Take great precautions to avoid dust, moisture, and other contaminants at all steps ................................

PLATING

18. With sterile pipet, divide 10 mL of disintegrated sample equally among 3 plates (optionally use 5 plates) ....

19. Pour agar (see SPC, item 13), thoroughly and evenly mix with test portion in plate ........................................

CONTROLS

20. See SPC, item 14 .....................................................................

INCUBATION

21. See SPC, item 15 .....................................................................
   a. Incubate at 32±1C for 48±3 hr ............................................

COUNTING COLONIES

22. See SPC, items 16, 17 .............................................................

REOUPS

23. Computing and Reporting Counts ........................................
   a. Multiply the sum of the colonies on 3 (5) plates by 10 .......
   b. Report as the number of colonies/g of stock .................