



HEALTH PRODUCTS AND FOOD BRANCH

OTTAWA

ENUMERATION OF TOTAL AEROBIC BACTERIA IN FOOD PRODUCTS  
AND FOOD INGREDIENTS USING COMPACT DRY AEROBIC COUNT PLATES

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**1. APPLICATION**

Compact Dry is applicable for microbial testing. The plates can be used to test raw material as well as finished products like food, beverage, meat, cosmetics or others. The Compact Dry plates can also be used like contact plates for difficult area using the Compact Dry swab.

**2. PRINCIPLE**

Compact Dry TC is a ready to use, chromogenic plate for detection of total viable bacterial count developed by HyServe GmbH & Co. KG, Uffing, Germany. Compact Dry combines the features and benefits of the old traditional plate media with the modern features of dehydrated film media. Compact Dry TC is a medium for total viable bacterial count, which contains nutrient standard agar. One mL of sample is placed onto the plate. The liquid samples will then homogenously self diffuse over the whole plate. The plates are then incubated and counted. The colonies grown on Compact Dry TC are red due to a redox indicator, tetrazolium salt. Bacteria can easily be picked up for further investigation.

**3. DEFINITION OF TERMS**

Compact Dry TC is a medium for total viable bacterial count, which contains nutrient standard agar. The colonies grown on Compact Dry TC are red due to redox indicator tetrazolium salt.

**4. COLLECTION OF SAMPLES**

See Appendix B of Volume 2.

**5. MATERIALS AND SPECIAL EQUIPMENT**

- 1) Compact Dry TC kits, stored at room temperature.

- 2) Package insert, including instructions for use.
- 3) Incubator capable of maintaining 20 to 42°C\*.

Note: \*Please use the incubation temperature/time according to the legal specification of each country's food analysis regulations.

## 6. PROCEDURE

Carry out the test in accordance with the following instructions:

### 6.1 Sample Pre-treatment

- 6.1.1 Viable count in water or liquid foodstuff: Drop 1 mL of specimen (dilute if necessary) on the middle of the Compact Dry plate.
- 6.1.2 Viable count in solid foodstuff: Add buffer solution to the sample and homogenize by stomacherâ. Drop 1mL of specimen (dilute if necessary) on the middle of the dry sheet of the Compact Dry plate.
- 6.1.3 Viable count in swab test specimen: Use the swab to wipe the surface, put into the device with wiping solution. Drop 1 mL of wiping solution (dilute if necessary) on the middle of the Compact Dry plate. It is recommended to use "Swab for Compact Dry" offered by HyServe Id-No. 1 002 952/3 (40/400 pieces).

### 6.2 Test Instructions

- 6.2.1 Open the cap and drop 1 mL of specimen on the middle of the Compact Dry plate.
- 6.2.2 Specimen diffuses automatically and evenly into the sheet and transforms the dried sheet into a gel within seconds.
- 6.2.3 Put the cap again on the plate and write the information needed on the memorandum section.
- 6.2.4 Turn over the capped plated and put in the incubator for 48 hours.
- 6.2.5 After incubation, count the number of coloured colonies underneath the plate. White paper placed under the plate helps to count the colonies.

### 6.3 Reading Results

- 6.3.1 Promptly count plates after incubation period. If impossible to count at once, store plates in the freezer. This should be avoided as a routine practice.
- 6.3.2 Use a standard colony counter for counting purposes. A magnified-illuminator may also be used to facilitate counting.
- 6.3.3 The circular growth area is approximately 20 cm<sup>2</sup>. Estimates can be made on plates containing more than 250 colonies by counting the number of colonies in one or more representative squares and determining the average number per square. Multiply the average number by 20 to determine total count per plate.
- 6.3.4 Calculate the number of colonies per mL or g of sample from the number of colonies obtained in plates chosen at dilution levels that give a statistically significant result.
- 6.3.5 When counting colonies on duplicate plates of consecutive dilutions, compute mean number of colonies for each dilution before determining average bacterial count.

6.3.6 To isolate colonies for further identification, lift the cap and pick the colony from the plate.

#### 6.4 Interpretation of Results

6.4.1 Count all red dots regardless of size or intensity. Colonies grown are almost all red. Red and otherwise coloured colonies together are the total count. The presence of very high concentrations of colonies on the plates will cause the entire growth area to become red or pink; record results as "too numerous to count" (TNTC). Occasionally, on overcrowded plates, the center may lack visible colonies but small colonies will be seen on the edges. When this occurs, record results as TNTC; further dilution of the sample is required.

6.4.2 Some organisms can liquefy the gel, allowing them to spread out and obscure the presence of other colonies. If a liquefier interferes with counting, an estimated count should be made by counting the unaffected areas.

### 7. NOTES

7.1 Some colonies might not be clearly red coloured.

7.2 High concentrations on plates will cause the entire growth area to become red/pink. In this case, dilute the sample.

7.3 After use please follow the current disposal regulations.

7.4 The growth area is 20 cm<sup>2</sup>. The back of the plate has a grid carved of 1cm x 1cm to make colony counting easier. In case if any difficulties to count colonies due to large number of colonies grown, total viable count can be obtained by multiplying 20 by an average number of colonies per grid counted from several grids.

7.5 Compact Dry plates are produced at an ISO 9001 certified site.

7.6 AOAC approval No. 010404

### 8. REFERENCES

8.1 Nissui Pharmaceutical granted PTM status for Compact Dry TC, Inside Laboratory Management; AOAC, July 2004:19-22

8.2 Bachmann, B., Lüthi, M. 2003. Evaluation mikrobiologischer Methoden zur Prüfung von Trinkwasser im Feld für Katastropheneinsätze. Mitt. Lebensm. Hyg. 94: 579-593.

8.3 Ellis, P., Kirchhof, G., and Meldrum, R. 2003. Evaluation of the Compact Dry SL method for the detection of Salmonella in spiked food samples. Poster presentation at HPA 1<sup>st</sup> Scientific Conference, University of Warwick, September 2003.

8.4 Ellis, P. and Meldrum, R. 2002. Comparison of the Compact Dry TC and #M Petrifilm ACP dry sheet media methods with the spiral plate method for the examination of randomly selected foods for aerobic colony count. J. Food Prot. 65: 423-425.

8.5 Ellis, P. and Meldrum, R. 2001. Evaluation of dryfilm methods for aerobic colony counts. Poster presentation at PHLS 26<sup>th</sup> Scientific Conference, University of Warwick, September 2001.

8.6 Mizuochi, S. and Kodaka, H. 2000. Evaluation of dry sheet medium culture plate (Compact Dry TC) method for determining numbers of bacteria in food samples. J. Food Prot. 63: 665-667.

- 8.7 Mizuochi, S., Kamiya, H., Kodaka, H., Sengoku, H., and Horigome, K. 1999. Compact Dry for the Enumeration of Bacteria in Food. ASM 1999 General Meeting, Chicago.
- 8.8 Kodaka, H. and Ishikawa, M. 1995. Evaluation of new medium with chromogenic substrates for members of the family Enterobacteriaceae in urine samples. J. Clin. Microbiol. 33: 199-201.
- 8.9 Curiale, M.S. and Sons, T., *et. al.* 1991. Dry rehydratable film for enumeration of total coliforms and *Escherichia coli* in foods: Collaborative study. J. Assoc. Off. Anal. Chem. 74: 635-648.