

STANDARDS ASSOCIATION OF AUSTRALIA

Australian Standard
METHODS FOR THE MICROBIOLOGICAL EXAMINATION OF FOOD

PART 2—EXAMINATION FOR SPECIFIC ORGANISMS

AS 1766.2.12
ESCHERICHIA COLI—
DIRECT PLATE METHOD

1 SCOPE. This standard sets out procedures for enumerating *Escherichia coli* in foods other than dairy products using a direct plate method.

NOTES:

1. Other methods for examination of foods for coliforms and *E. coli* are given in AS 1766.2.1.3.
2. Procedures for dairy products are given in AS 1095*.

2 REFERENCED DOCUMENTS. The following standards are referred to in this standard:

AS 1766 Methods for the Microbiological Examination of Food
1766.1 General Procedures and Techniques
1766.2 Examination for Specific Organisms
1766.3 Examination of Specific Products
1766.5 Preparation of Media, Diluents and Reagents.

3 DEFINITION. For the purpose of this standard, *E. coli* are defined as bacteria which grow at 44.0° C to 44.5° C to form characteristic indole positive colonies on cellulose ester membranes overlaid on tryptone bile agar, under the operational conditions described. The method will detect both typical *E. coli* (biotype 1) and lactose non-fermenting or anaerogenic variants.

4 CULTURE MEDIA, DILUENTS, REAGENTS AND MATERIALS. (See Appendix A).

0.1 percent peptone solution or other diluent if specified for a particular product in AS 1766.3.

Tryptone bile agar (TBA)

Minerals modified glutamate agar (MMGA)

Indole reagent (Vracko and Sherris)

Cellulose ester membranes (85 mm dia., 0.45 µm pore size)

NOTE: Some types of membranes are unsuitable for this method and a laboratory should test the membranes to ensure they are satisfactory.

Ultraviolet lamp, wavelength 365 nm.

5 REFERENCE CULTURES.

Escherichia coli NCTC 9001 or ATCC 11775.

Enterobacter aerogenes NCTC 10006 or ATCC 13048.

6 PROCEDURE.

6.1 General. The resuscitation procedure described in Clause 6.3 should be used when examining products in which *E. coli* may be damaged by freezing, cold storage, drying or by heat or chemical processes. This procedure should also be used in the examination of products containing high concentrations of carbohydrates which may have an inhibitory effect on indole production. This method permits the diffusion of carbohydrates present in the test sample during the resuscitation step.

6.2 Direct plate method.

- (a) Prepare Petri dishes of TBA using the method described in AS 1766.1 for preparing and drying plates for determining colony count by the surface spread method.

*AS 1095, Microbiological Methods for the Dairy Industry.