

Culture Media

VRBL AGAR (ISO) CM0968 AND VRBG AGAR (ISO) CM1082





Violet Red Bile Lactose (VRBL) Agar (ISO) is used for the detection and enumeration of coliforms in food, animal feed and environmental samples and conforms to ISO 4823:2006¹.

Violet Red Bile Glucose (VRBG) Agar (ISO) is used for the detection and enumeration of Enterobacteriaceae in food, animal feed and environmental samples and conforms to the formulation described in ISO 21528-2:2004².

- Manufactured in ISO 9001 certified production facilities
- Performance tested to ISO/TS 11133-2:2003³
- Available as Prepared Media
- Quantitative Quality Certificates available for every batch
- Part of an extensive range of ISO conforming media for food and environmental testing

SUMMARY

Various groups of bacteria or individual species are used to provide evidence of poor hygiene, inadequate processing or post-processing contamination of foods. Some bacteria, such as *E. coli*, are present in the gastrointestinal tract of many animals including humans so their presence can be used to indicate potential faecal contamination.

Two of the most common groups of bacteria employed as indicator organisms by the food industry are Enterobacteriaceae and coliforms.

Coliforms are commonly defined by their ability to ferment lactose rapidly, producing acid and gas, typically within 24 hours. The genera that would commonly be expected to belong to this group include *Enterobacter, Klebsiella, Citrobacter* and *Escherichia*, particularly *E. coli.* However, species belonging to other genera, for example *Erwinia* and *Serratia*, can

PRINCIPLES

Neutral red is included in both media to act as a pH indicator. As Enterobacteriaceae ferment glucose, or coliforms ferment lactose, the pH of the medium drops and the colonies appear red to purple; colonies are usually surrounded with a purple halo of precipitated bile sales. Crystal violet and bile salts inhibit the growth of Gram-positive flora.

According to ISO/TS 11133-2:2003³, both media should deliver a productivity ratio of \geq 0.5 for *E. coli* when compared to the same inoculum grown on a reference medium (Tryptone Soya Agar). Both Oxoid media are tested in line with the requirements of ISO/TS 11133-2:2003³ and Quality Certificates, indicating organisms tested and productivity, are available for every batch.

FORMULATION

	Grams per litre
Enzymatic digest of animal tissues	7.0
Yeast extract	3.0
Bile salts No. 3	1.5
Sodium chloride	5.0
Neutral red	0.03
Crystal violet	0.002
Agar	12.0
Glucose (in VRBG agar)	10.0
Lactose (in VRBL agar)	10.0

Final pH 7.4±0.2 @ 25°C

500g of dehydrated powder will make approximately 13 litres of medium.

DIRECTIONS

Suspend 38.5 g of dehydrated medium in 1 litre of distilled water. Bring to the boil while mixing. Continue to boil for 2 minutes or for the minimum time necessary to dissolve completely. DO NOT AUTOCLAVE. Cool to 44-47°C and use within 4 hours. Mix well before pouring.

REFERENCES 1. ISO 4823:2006 Horizontal Method for the detection and enumeration of coliforms - Colony count method. 2. ISO 21528-22004 Horizontal Methods for Detection and enumeration of Enterobacteriaceae Part 2. Colony-count method. 3. ISO(75 11133-22030 Microbiology of food and animal feeding stuffs - Guidelines on preparation and production of culture media - Part-2: Practical guidelines on performance testing of culture media. A Mossel, Eelderink, Koopmans and van Rossem. (1978). Lab Practice 27.1049. 5. Mossel, Eelderink, Koopmans and van Rossem. (1979). J. Food Protect 42-470. 6. Cowell, N.D. and Morisetti, M.D. (1969) J. Sci. Fd. Agric. 20:573. 7. ISO 6887 (all parts) Microbiology of food and animal feeding stuffs - Preparation of test samples, initial suspension and decimal dilutions for microbiological examination. ISO 8261 Milk and milk products - General guidance for the preparation of test samples, initial suspensions and decimal dilution for microbiological examination. also ferment lactose, albeit slowly, and some strains of *Citrobacter* and *Klebsiella*, as well as *Salmonella arizonae* and *Hafnia alvei*, show delayed or variable lactose fermentation ability.

Recently, there has been a gradual move towards testing for total Enterobacteriaceae rather than limiting testing for coliforms. In processed foods, particularly foods subjected to heat treatment, these bacteria can provide a reliable indication of process failure, under-processing or post process contamination.

Mossel *et al*^{*} modified Violet Red Bile Lactose Agar by adding glucose to improve recovery of all Enterobacteriaceae. Later work by Mossel and Cowell⁵⁶ demonstrated that lactose could be omitted, resulting in the formulation known as VRBG Agar.



OXOID PRODUCTS THAT MAY BE USEFUL FOR IDENTIFICATION

Brilliant Green (Lactose) Bile Broth	CM0031	
Nutrient Agar	CM0003	
(Prepared Media also available - please check with your local supplier)		
Oxidase Sticks	BR0064A	
Microbact GNB 12A	MB1132A/MB1076A	
Microbact GNB 12E	MB1130A/MB1073A	

QUALITY CONTROL TESTING

VRBG Agar (ISO)

Organism	Culti-loop® order code	Typical colony appearance
Escherichia coli ATCC® 25922™	C7050L	Red-purple colonies with or without halos
<i>Salmonella</i> Typhimurium ATCC [®] 14028 ^{™t}	C6000L	Red-purple colonies with or without halos
Enterococcus faecalis ATCC® 29212™t	C7030L	No growth

VRBL Agar (ISO) Organism	Culti-loop® order code	Typical colony appearance
Escherichia coli ATCC® 25922™	C7050L	Red-purple colonies with or without halos
<i>Pseudomonas aeruginosa</i> ATCC® 27853™t	C7060L	Colourless to straw coloured colonies
Enterococcus faecalis ATCC® 29212™†	C7030L	No growth



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