Reference: TUWR3023 **A.B.E. - Technical Data Sheet**

Product: # Violet Red Bile Glucose Agar (VRBG)

Specification

Selective solid medium for the enumeration of enterobacteria, acccording to ISO standard 21528 and Pharmacopeial Harmonised Methods.

Presentation

	Packaging Details	Shelf Life	Storage
20 Tubes Tube 17 x 145 mm with: 15 ± 0.3 ml	17x145 mm glass tubes, ink labelled, metal-Non injectable cap. - 20 tubes per box .	12 months	8-25 ºC

Composition

Composition (g/l):

Peptone from gelatin	7.00
Yeast extract	3.00
Bile salts	1.50
D(+)Glucose	10.0
Sodium chloride	5.00
Neutral red	0.03
Crystal violet	0.002
Agar	13.0

Description /Technique

Description

This medium is a modification of the Violet Red Bile Agar and the MacConkey Agar as described by Mossel et al. The addition of glucose to the Violet Red Bile Agar enhances both the growth of the most fastidious enterobacteria and the recovery of those having suffered from adverse conditions. Mossel himself realized that by removing the lactose and keeping the glucose, the medium's efficiency remained stable.
This medium can be used as a presumptive medium for E. coli (by fluorescent reaction) if before sterilization MUG is added.

Technique

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.
Melt the medium contained in the tube in a water bath (100ºC) or in a microwave oven, avoiding overhating, before pouring into Petri dishes when cooled to room temperature.
Once solidified on a flat surface, Spread the plates by streaking methodology or by spiral method. Incubate the plates right side up aerobically at 35ºC± 2,0 for 24 h.
(Incubation times longer than those mentioned above, different inoculation methods or different incubation temperatures may be required depending on the sample, on the specifications,...
This medium can be inoculated directly or after any enrichment broth)
after incubation, enumerate all the reddish-violet colonies that have appeared onto the surface of the agar, with a red-violet halo due to bile salts precipitation.
Presumptive isolation of E.coli or coliforms must be confirmed by further microbiological and biochemical tests.
calculate total microbial count per ml of sample by multiplying the average number of colonies per plate by the inverse dilution factor if streaked a diluted sample. Report results as Colony Forming Unit (CFU's) per ml or g along with incubation time and temperature,that enables to differentiate total coliforms and faecal coliforms.

Note: The solid mediums can be melted in different ways: autoclave, bath and, if the customer considers appropriate, also the microwave. Whenever the microwave option is chosen, it is necessary to take certain safety measures to avoid breaking of the containers, such as loosening the screw cap and putting the bottle or tube in a water bath in the microwave. The fusion temperature and time will depend on the shape of the container, the volume of medium and the heat source. Avoid overheating as both the heating periods.

Quality control

Physical/Chemical control

Color : Violet-pink pH: 7.4 ± 0.2 at 25ºC

Microbiological control

Melting- pour plates- Inoculate: 10-100 CFU accord. to Eur. Pharm. & 100 ± 20 CFU; min. 50 CFU (productivity)/ 10^4-10^6 CFU (selectivity) acc. to ISO.
Microbiological control according to ISO 11133:2014/A1:2018.

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Aerobiosis. Incubation: 30-35 ºC. Reading at 24h (E.P.) / 37 ± 1 ºC. Reading at 24 h (ISO)

Note: results ATCC® 8739/6538/9027 (30-35 ºC) & ATCC® 8739/25922/19433/14028 (37 ºC).

Microorganism	Growth
Enterococcus faecalis ATCC® 19433, WDCM 00009	Inhibited
Staphylococcus aureus ATCC® 6538, WDCM 00032	Inhibited
Ps. aeruginosa ATCC® 9027, WDCM 00026	Good (50%) -Colourless colonies
Salmonella typhimurium ATCC® 14028, WDCM 00031	Good (50%)- Red purple colonies - Biliar precipitate
Escherichia coli ATCC® 25922, WDCM 00013	Good (50%)- Red purple colonies - Biliar precipitate
Escherichia coli ATCC® 8739, WDCM 00012 (32,5ºC)	Good (50%)- Red purple colonies - Biliar precipitate
Escherichia coli ATCC® 8739, WDCM 00012 (37ºC)	Good (50%)- Red purple colonies - Biliar precipitate

Sterility control

Incubation 48 h at 30-35 ºC and 48 h at 20-25 ºC: NO GROWTH.
Check at 7 days after incubation in same conditions.

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