TECHNICAL DATA SHEET

CRONOBACTER SCREENING BROTH (CSB)

SELECTIVE ENRICHMENT FOR *CRONOBACTER* SPP.

1 INTENDED USE

Cronobacter Screening broth is a selective enrichment media used for the detection of *Cronobacter* spp. in food products and ingredients intended for human consumption and the feeding of animals. It is also used for the control of production environmental samples.

The typical composition responds to that defined in the standard NF EN ISO 22964.

2 PRINCIPLES

Saccharose fermentation by *Cronobacter,* resulting in an acid shift in the media, is revealed by the presence of bromocresol purple, where the normal blue-violet color turns yellow.

Vancoycin is an antibiotic that inhibits secondary, primarily Gram positive, flora.

3 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

- Meat peptone ..... 10,00 g
- Meat extract..... 3,00 g
- Sodium chloride ..... 5,00 g
- Bromocresol purple ..... 0,04 g
- Saccharose ..... 10,00 g
- Vancomycin..... 0,01 g

pH of the ready-to-used media at 25 °C : 7,4 ± 0,2.

4 INSTRUCTIONS FOR USE

Detection protocol for *Cronobacter* according to the standard ISO 22964 :

- Aseptically introduce 10 g or 10 mL of the product to test into 90 mL of Buffered Peptone Water.
- Homogenize or use a stomacher if needed.
- Incubate the broth at 36 ± 2 °C for **18 ± 2 hours**.
- Reinoculate 0,1 mL of the pre-enrichment into 10 mL of the *Cronobacter* Screening broth (BM155).
- Incubate at 41,5 ± 1,0 °C for 24 ± 2 hours.
- On the surface of *Cronobacter* Chromogenic agar (BM154) brought to room temperature, inoculate by streaking 0,1 mL of the enrichment broth.
- Incubate at 41,5 ± 1,0 °C for 24 ± 2 hours.

✓ **Pre-enrichment :**
1:10 dilution
18 h at 36 °C
✓ **Enrichment :**
0,1 mL
24 h at 41,5 °C
✓ **Detection :**
Reinoculation 1 loop
24 h at 41,5 °C

Notes :

For testing samples of greater volume, pre-heat the Buffered Peptone Water to 36 ± 2 °C.
Regrouping numerous samples together for testing can compromise the recuperation of stressed strains of *Cronobacter*, in the presence of an important secondary microflora. It is the user's responsibility to demonstrate the conformity of his testing protocol.

5 QUALITY CONTROL

Prepared media : violet solution, limpid.

Typical culture response after 24 hours of incubation at 41,5 °C, followed by subculture on CCI Agar or TSA :

Microorganisms		Growth
Cronobacter sakazakii + *Staphylococcus aureus*	WDCM 00214 WDCM 00034	Yellow broth > 10 characteristic colonies on CCI (blue-green)
Cronobacter muytjensii + *Staphylococcus aureus*	WDCM 00213 WDCM 00034	Yellow broth > 10 characteristic colonies on CCI (blue-green)
Staphylococcus aureus	WDCM 00034	Violet broth, inhibition on TSA

6 STORAGE / SHELF LIFE

Ready-to-use media in tubes : 2-8 °C.
The expiration date is indicated on the label.

7 PACKAGING

Ready-to-use media in tubes :
50 x 10 mL tubes BM15508

8 BIBLIOGRAPHY

Mallmann, W.L., and Darby, C.W.. 1941. Uses of a lauryl sulfate tryptose broth for the detection of coliform organisms. American Journal of Public Health and the Nations Health, **31** : 127-134.

NF EN ISO 22964. June 2017. Microbiologie de la chaine alimentaire. Méthode horizontale pour la recherche de *Cronobacter* spp.

9 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

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