BRILLIANCE SALMONELLA AGAR BASE

Code: CM1092

Brilliance™ Salmonella Agar (formerly OSCM II) is used to produce a selective medium for the presumptive identification and differentiation of Salmonella species.

Typical Formula*	gm/ litre
Inhibigen™ mix	14
Chromogenic Mix	25
Agar	15
pH 7.3 [†] ± 0.1 @ 25ºC	

^{*} Adjusted as required to meet performance standards

SALMONELLA SELECTIVE SUPPLEMENT

Code: SR0194

Vial Contents (each vial is sufficient for 500ml of medium)	per vial	per litre
Novobiocin	2.5mg	5mg
Cefsulodin	6mg	12mg

Brilliance Salmonella Agar is the isolation medium used in the Oxoid Salmonella Precis rapid culture method. For information simply download the *Brilliance* Salmonella Agar data sheet (1MB) or the <u>Salmonella Precis data sheet</u> (1396KB) in PDF format. Please confirm prepared media codes with your local supplier.

Directions

Suspend 27g of *Brilliance* Salmonella Agar Base in 500ml of distilled water. Add the contents of 1 vial of Salmonella Selective Supplement, resuspended as directed. Mix well and sterilise by bringing to the boil with frequent agitation. Cool to around 50°C, mix well and pour into sterile Petri dishes.

NOTE: It is critical that the selective supplement is added prior to heating. When preparing volumes greater than 1 litre contact Oxoid Technical Support for directions.

Description

An Inhibigen™ compound is comprised of two components, combined together by a bond that can only be cleaved by a specific enzyme. When bound together, the inhibitor compound is not toxic and therefore can exist in a medium without harming micro-organisms. Once inside the cell, the bond will be cleaved if the target enzyme is present. When the bond is cleaved, the inhibitor molecule is released and disrupts cell wall synthesis, causing death of the organism. As cells die and lyse, free inhibitor is released but cannot be taken up by other cells, resulting in targeted inhibition. The Inhibigen in *Brilliance* Salmonella Agar targets *Escherichia coli*. Novobiocin and cefsulodin, presented as a freezedried supplement (SR0194), are added to the medium to inhibit the growth of other competing flora such as *Proteus* spp. and *Pseudomonas* spp.

[†] pH of supplemented medium

Differentiation of *Salmonella* from the other organisms that grow on *Brilliance* Salmonella Agar is achieved through the inclusion of two chromogens that also target specific enzymes: caprylate esterase and ß-glucosidase. Caprylate esterase is an enzyme present in all samonellae as well as some species of *Klebsiella*, *Enterobacter* and *Proteus*¹. Organisms possessing caprylate esterase cleave the chromogen to release an insoluble purple chromophore. As the cells grow, the chromophore builds up and produces a purple-coloured colony. Some Enterobacteriaceae, including *Klebsiella* and *Enterobacter* but not *Salmonella*, possess ß-glucosidase². If these organisms grow, they will form blue or dark blue colonies, even if they are esterase positive, which make them easy to differentiate from purple *Salmonella* colonies.

Technique

Testing food samples

Brilliance Salmonella Agar can be used for testing clinical (faecal) samples, either directly or following a suitable enrichment protocol. It can also be used for food and environmental samples following a variety of enrichment procedures e.g. ISO, NMKL, FDA, HPA, etc. Depending on the method, Brilliance Salmonella Agar can be streaked with sample or post-incubation enrichment broth. The plates are then incubated at 37°C for 24 ± 3h. Salmonella produce purple colonies.

The following method is a summary of ISO 6579:2002 +A1:2007³ illustrating how *Brilliance* Salmonella Agar can be used alongside XLD Medium, when following this method:

- 1. Add 25g of food sample to 225ml of Buffered Peptone Water and stomach for a minimum of 30 seconds to mix the sample
- 2. Incubate the broth at 37° C for $18 \pm 2h$
- 3. Gently agitate the bag then, using a pipette, inoculate 1ml into 10ml of MKTTn Broth and 0.1ml into 10ml of RVS Broth. Incubate MKTTn broth at 37°C for 24 ± 3h and RVS broth at 41.5°C for 24h ± 3h
- 4. Using a microbiological loop, inoculate each broth onto one XLD Agar plate and one *Brilliance* Salmonella Agar plate to give a total of 4 plates.
- 5. Incubate the plates at 37°C for 24h ± 3h
- 6. Confirm presumptive red colonies with black centres from XLD and purple colonies from *Brilliance* Salmonella Agar as *Salmonella* spp by appropriate biochemical and serological methods. Refer to ISO 6579:2002 +A1:2007³.

The following Oxoid products may be useful to confirm a presumptive *Salmonella* identification: Nutrient Agar (CM0003), TSI Agar (CM0277), Urea Agar (CM0053 & SR0020K), Lysine Decarboxylase Medium (CM0308), Methyl-Red Voges-Proskauer (MRVP) Medium (CM0043), Tryptone Soya Broth (CM0129), Agglutinating Anti-sera (see Oxoid product list for codes); Oxoid Salmonella Latex Test Kit (DR1108), Microbact (see Oxoid product list for codes); O.B.I.S. Salmonella (ID0570). Contact your local Oxoid supplier for prepared media codes.

Testing clinical samples

Inoculate the plate directly with pea sized bead or loopful of specimen. Incubate plates at 37°C or for 24h±3h. Alternatively, pre-enrich in suitable selective broth prior to inoculation onto a *Brilliance* Salmonella plate. Use an incubation protocol appropriate to the broth chosen. Purple colonies should be confirmed as *Salmonella* spp by appropriate biochemical and serological methods.

For further instructions on the use and interpretation of *Brilliance* Salmonella Agar with clinical samples, <u>simply download the data sheet (885KB)</u> in PDF format. Please confirm prepared media codes with your local supplier.

Storage Conditions and Shelf Life

Brilliance Salmonella Agar should be stored in the tightly capped original container at 10-30°C. Salmonella Selective Supplement should be stored in the dark at 2-8°C. When stored as directed, the un-opened products will remain stable until the expiry date printed on the packaging.

Locally prepared *Brilliance* Salmonella Agar plates can be stored for up to 2 weeks when made from CM1092 and SR0194 according to the manufacturer's instructions and stored at 2-8°C, out of direct sunlight. A longer shelf life may be attainable, but should be validated under the relevant, local manufacturing and storage conditions.

Appearance

Dehydrated medium: White/cream coloured, free-flowing powder

Selective supplement: White, freeze-dried pellet Prepared medium: White/cream coloured opaque gel

Quality control

Positive controls:	Expected results
Salmonella Typhimurium ATCC°14028*	Good growth; purple colonies
Klebsiella pneumoniae ATCC®13883*	Good growth; blue colonies
Negative controls:	
Enterococcus faecalis ATCC® 29212*	Inhibited
Escherichia coli ATCC [®] 25922*	Inhibited

^{*}This organism is available as a Culti-Loop®

Precautions

Brilliance Salmonella Agar is for *in vitro* diagnostic use only, by experienced microbiologists. It must not be used beyond the stated expiry date, or if the product shows any sign of deterioration. Sterilise specimens, equipment and media properly after use.

Limitations

It should be noted that, as with all *Salmonella* media, organisms with atypical enzyme patterns may give anomalous reactions on *Brilliance* Salmonella Agar. A small number of atypical strains that may give a weak reaction or fail to grow, especially when low numbers are present in the sample. *Brilliance* Salmonella Agar is not recommended for the isolation of *Salmonella typhi* and *Salmonella paratyphi*. Please use Oxoid Salmonella Culture Medium (OSCM) CM1007 as your chromogenic medium for this purpose.