Paradigm Diagnostics Salmonella Indicator Broth (PDX-SIB)
User Guide
Salmonella Test Kit

Intended Use

Paradigm Diagnostics Salmonella Indicator Broth is intended for use in screening environmental samples for the presence of viable Salmonella species. Positive samples will exhibit a color change from blue to yellow if Salmonella bacteria are growing in the liquid medium. The color change indicates a presumptive positive. Positive samples should be followed up with characterization by cultivation on a selective agar, immuno-diagnostic or genetic analysis such as PCR for confirmation.

Scientific Principal of the Test

PDX-SIB contains selective agents to prevent the growth of competitive microflora which providing nutrients for growth of the resistant Salmonella species. Salmonella spp. are further differentiated from any active background microflora by metabolism of a specific energy source metabolized primarily by Salmonella species. As the population of Salmonella metabolizes the growth substrate, the compound is fermented to an acidic by-product, which turns the pH indicator in the broth from blue to yellow. A yellow color observable after incubation at 37°C for 24-48 hours is deemed presumptively positive.

Diagnostic Performance Parameters

PDX-SIB was subjected to a panel of more than 100 Salmonella and non-salmonella organisms. The test exhibited 98.5% sensitivity with 95% specificity. The tests were conducted with inocula at both low (<10 CFU per sample) as well as high (>100 CFU per sample).
Additional Notes

- Environmental samples are best collected with pre-wetted swabs or sponges such as those available from 3M Company, or Whirl-Pak Corporation.
- Water samples containing low Salmonella counts should be diluted 10:1 with either 10X strength lactose broth or Tryptic Soy Broth and held at 37°C for three hours to ensure viable Salmonella population is present before exposure to harsh selective agents.

Materials and Equipment Required

- An incubator capable of maintaining 37°C such as an environmental cabinet, a water bath or a heating block.
- Sample collection devices such as sterile swabs, sponges or vials for liquid samples.
- Swabs and sponges should be pre-wetted with commonly employed Salmonella recovery media such lactose broth or buffered peptone water.

Confirmation Step

Confirmation of presumptive positive samples can be carried out utilizing selective agars such as those described in the Bacterial Assay Manual\(^1\) or by utilization of commercially available immuno-diagnostic assays or PCR analysis.

Disposal

Decontaminate the PDX-SIB by autoclave, bleach or other disinfectant in accordance with local, state and federal regulations.

Product Shelf Life

PDX-SIB is a developmental product and as such no documented shelf life is reportable. The product should be stored at refrigerator temperatures (4°C) to obtain maximum useful life.

Precautions

1. Most Salmonella species are human pathogens and enriched samples should be handled employing Good Microbiological Practices.
2. False positives have been observed with high-level inoculations of some Klebsiella species underscoring the need for confirmation on a selective agar.

\(^{1}\) http://www.cfsan.fda.gov/~ebam/bam-10.html.
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Instruction for PDX-SIB Use

1. **Sampling:** Water samples should be first diluted into 10X strength Lactose broth in 20 mL screw-cap tubes. Add 4.5 mL to the concentrated Lactose broth and place the sample in an incubator adjusted to 37°C for three to four hours. This step permits recovery of any sub-lethally injured cells and permits some pre-enriched growth of the bacterial population.

2. **Transfer the contents of the screw-cap tube to the 50 mL conical tube containing 45 mL of SIB (blue solution).** Place the sample back into the incubator and hold for ~ 24 hours at 37°C.

3. **Interpretation:** Presumptively positive samples will turn **yellow** due to the accumulation of acidic metabolic products.

4. **Positive samples can be confirmed by streaking onto a selective agar, such as the enclosed Salmonella indicator agar (available from ChromAgar).** Plates should be incubated at 37°C a minimum of 18 hours. Salmonella spp. colonies produce
an insoluble magenta color. Alternative confirmation methods are described in the most recent publication of the Bacteriological Assay Manual\textsuperscript{2}

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\item \textsuperscript{1} \url{http://www.cfsan.fda.gov/~ebam/bam-10.html}
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