

**CAUTION!**

1. Do not attempt to eat this product, touch the growth area with bare hands or allow the medium to get into eyes.
2. Please make sure to read the precautions and instructions in this Instruction Manual before attempting to use the device and exercise extreme caution when using it.

**Product Description and Intended Use**

Easy Plate EB is a microbiological culture device made up of a waterproof sheet, a readymade dry medium on the sheet and a transparent cover over the medium. The Easy Plate EB method is intended to indicate the level of Enterobacteriaceae bacteria in food and beverage products. It is compact and easy to use and reduces the total amount of waste produced during testing. Easy Plate EB is manufactured at an ISO (International Organization for Standardization) 9001 certified site.

**Contents of Product**

One box of this product contains 100 sheets.

- 25 sheets per bag
- 4 bags in a box

**Materials Required but Not Provided\***

- Incubator ( $35 \pm 1^\circ\text{C}$  or  $37 \pm 1^\circ\text{C}$ )
- Stomacher or Blender
- Sampling bag
- Pipette or Pipettor and pipette tips
- Maximum recovery Diluent (MRD) or appropriate diluents according ISO 6887

**Instructions for Use****1. Sample preparation**

User should choose an appropriate method for the sample preparation. Refer to ISO 6887, FDA BAM or other regulatory guidelines.

The following method is an example.

- 1) Weigh each 10 g test portion into a stomacher bag or a filter bag.
- 2) Add 90 g MRD or appropriate diluent according to ISO 6887
- 3) Prepare all decimal dilutions with 9 mL MRD plus 1 mL previous dilution.
- 4) Shake 25 times.
- 5) Adjust the pH of the diluted sample to 6.5-7.5. Use 1N HCl or NaOH to adjust the pH.

**2. Inoculation**

- 1) Allow the bag of Easy Plate EB to room temperature ( $15-25^\circ\text{C}$ ). Then, remove required number of the sheets from the bag under aseptic conditions.
- 2) Place the sheet on a flat surface.
- 3) Lift the cover and place 1 mL sample suspension onto the center of the plate.
- 4) Lower the cover onto the sheet and allow the sample to

spread evenly. (If the cover sheet on the plate becomes bent, discard and replace the entire plate. A bent cover sheet will prevent uniform spreading of the sample)

- 5) Allow the plate to settle for 3 minutes or more on a horizontal surface. Do not tilt the sheet until solidification of the suspension is complete. Once solidification is complete, the plate can be handled normally.
- 6) Hold both ends of the sheet and place it into an incubator.

**3. Incubation**

Incubate the plate(s) at  $35 \pm 1^\circ\text{C}$  or  $37 \pm 1^\circ\text{C}$  for  $24 \pm 1$  hours. Up to 25 plates can be stacked for space saving and convenience.

**4. Interpretation**

Count all red colonies regardless of size or intensity. The suitable colony counting range is 1-250. In the case of growth area entirely colors orange and red spots cannot be seen, it is judged as negative.

**Troubleshooting for counting colonies:**

- a) If the growth area entirely colors red purple but the red spots cannot be seen, record as too numerous to count (TNTC).
- b) When a bubble disrupts a colony so that the colony outlines the bubble, count it as one colony.
- c) When a colony is spreading, count it as one colony.
- d) When the sample is not clear (i.e. cloudy or dark), prepare a higher dilution.
- e) When colonies are extremely small or light, incubate continuously for a few hours to make the colony size larger or the colony color intensity darker.
- f) When the entire growth area become red purple due to food components or pH solution, prepare a higher dilution.
- g) This product cannot measure the activity of oxidase. Therefore, gram-negative bacteria of oxidase positive may grow. Please conduct the oxidase test if necessary.
- h) If an estimated count is required, count colonies within 1-3 squares (1 cm x 1 cm) printed on the cover and calculate an average. Multiplying the average number by 20 provides the estimated count since the circular growth area is approximately 20 cm<sup>2</sup>.

**5. Colony isolation**

Lift the cover and pick a single colony from the gel.

**Precautions**

- 1) This product is not to be used for clinical testing.
- 2) This product has not been tested with all possible food

- products, food processes, test protocols or strains.
- 3) Do not open the cover until just before inoculation.
  - 4) Do not use the product after its expiry date.
  - 5) Do not use any plates that show damage or are deformed, discolored, or show presence of foreign materials.
  - 6) Do not expose the product to direct sunlight.
  - 7) Use caution when lowering the cover onto the media and spreading the sample suspension. Avoid direct pressure on the plate cover and avoid causing the suspension to spill out from the growth area.
  - 8) If the sample suspension does spill out from the growth area, discard the plate and repeat the inoculation with a new plate.
  - 9) Always wear safety eyewear when performing inoculations and analysis. If medium or reagents get into the eyes or mouth, flush thoroughly with water and seek medical attention.
  - 10) Analysis needs to be performed under the control of a skilled microbiologist. Refer to Good Laboratory Practices\* or ISO 7218.

\*Ex) U.S. Food and Drug Administration. Code of Federal Regulations, Title 21, Part 58. Good Laboratory Practice for Nonclinical Laboratory Studies.

### **Storage**

Store in the refrigerator (2-8°C).

The product can be stored in the product packaging up to 25°C for up to 14 days or up to 30°C for up to 5 days before opening the packaging.

### **Storage after Opening the Packaging**

Put any unused sheets back into the packaging, fold the end of the packaging over twice, and seal with tape. The shelf life under refrigerated condition is 3 months after opening.

### **Shelf Life**

Expiry date is specified on the side of the box and the back side of the bag after the word "EXP". The shelf life of the product is if it is stored properly in unopened packaging.

### **Disposal**

Any and all media, supplements, and reagents must be sterilized by autoclaving after use, and then disposed as industrial waste according to local laws and regulations.

### **Warranty**

Kikkoman Biochemifa Company warrants the products to have a certain level of quality. This warranty guarantees that Kikkoman Biochemifa Company shall replace defective products should any be found. This warranty does not provide any other guarantees. Kikkoman Biochemifa

Company shall not be liable for any damages, including special or consequential damages, or expenses arising directly or indirectly from the use of this product.

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