TM 1195 - SOB MEDIUM (HANAHAN'S BROTH)

INTENDED USE

For cultivation of recombinant strains of *Escherichia coli*.

PRODUCT SUMMARY AND EXPLANATION

Transformation is a process involving the uptake of foreign genetic material, which on subsequent recombination event results into genetically altered cell. The ability of a bacterium to take up exogenous DNA from the extracellular environment is termed as competence of the bacterium. Factors affecting the cell surface are important to competence, particularly changes in the membrane permeability, so as to allow the foreign DNA to enter the recipient cell. Bacteria undergoing transformation need to be cultured on a rich, isotonic medium to overcome or recover from the process of transformation by mending the perforations caused by transformation and undergo replication. Hanahans Broth developed by Hanahan is used for the cultivation of these recombinant Escherichia coli strains that have undergone transformation.

Hanahans Broth is a nutritionally rich growth medium for use in the preparation and transformation of competent cells. For generation of competent cells, the bacteria are grown in Hanahans Broth to the desired turbidity and subjected to standard procedures such as electroporation or treatment with CaCl2 in chilled conditions to achieve competence. For the survival of such perforated, competent cells, a rich, isotonic environment is needed. Hanahans Broth with 0.4% dextrose is used in the final stage of transformation, which provides carbon and energy source for mending the perforations and subsequent replication.

Tryptone and yeast extract in the medium supply nitrogenous compounds and growth factors for the recombinant *E. coli*. Potassium and sodium chloride maintains isotonic conditions. Magnesium sulphate is added to the medium as the necessary component for DNA replication.

COMPOSITION

Ingredients	Gms / Ltr
Tryptone	20.000
Yeast extract	5.000
Sodium chloride	0.500
Magnesium sulphate	2.400
Potassium chloride	0.186

PRINCIPLE

Tryptone and yeast extract in the medium supply nitrogenous compounds and growth factors for the recombinant *E. coli*. Potassium and sodium chloride maintains isotonic conditions. Magnesium sulphate is added to the medium as the necessary component for DNA replication.

INSTRUCTION FOR USE

- Dissolve 28.08 grams in 1000 ml distilled water.
- Heat if necessary to dissolve the medium completely.
- Dispense in tubes and sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Cream to yellow homogeneous free flowing powder.
Appearance of prepared medium : Medium amber coloured, clear solution without any precipitate.
pH (at 25°C) : 7.0±0.2

INTERPRETATION

Cultural characteristics observed after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Incubation Temperature	Incubation Period
Escherichia coli	53868	50-100	Good-luxuriant	35-37°C	18-24 Hours

PACKAGING:

In pack size of 500 gm bottles.

STORAGE

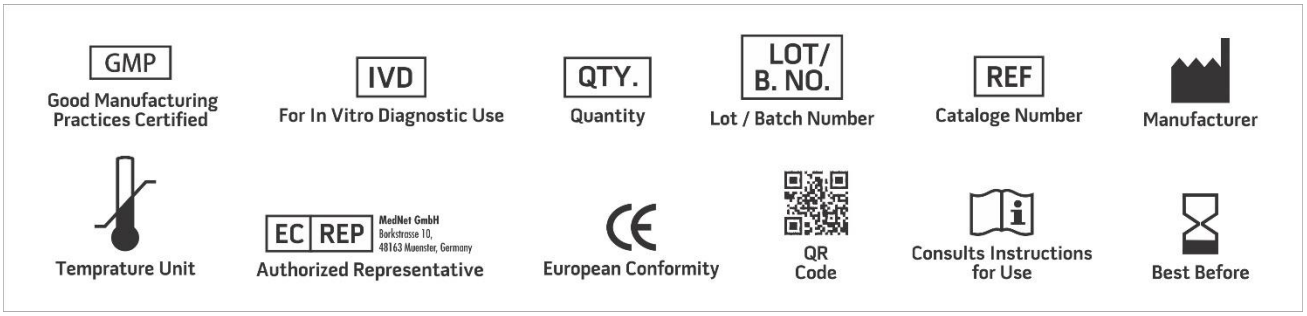
Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. Alcamo I. E., 2001, Fundamentals of Microbiology, 6th Edition, Jones and Bartlett Publishers.
2. Hanahan D., 1983, J. Mol. Biol., 166:557.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
5. Sambrook J., Fritsch E. E. and Maniatis T., 1989, Molecular Cloning: A Laboratory Manual, 2nd Ed., Cold Spring Harbor Lab. Press; Cold Spring Harbor, N.Y.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.
***For Lab Use Only**

Revision: 08 Nov., 2019