

Raka Ray No.3 Agar



Medium used for the isolation and cultivation of lactic acid bacteria from beer and brewing processes.

• CONTENTS (Liter)

| | |
|------------------------------|--------|
| Tryptone | 20.0 g |
| Yeast Extract | 5.0 g |
| Liver Extract | 1.0 g |
| Maltose | 10.0 g |
| Fructose | 5.0 g |
| Glucose | 5.0 g |
| Betaine Hydrochloride | 2.0 g |
| Diammonium Citrate | 2.0 g |
| L-Aspartic Acid | 2.5 g |
| Magnesium Sulfate | 0.98 g |
| Manganese Sulfate | 0.42 g |
| Dipotassium Phosphate | 2.0 g |
| N-Acetyl Glucosamine | 0.5 g |
| Potassium Glutamate | 2.5 g |
| Agar | 19.0 g |
| Final pH = 5.4 ± 0.2 at 25°C | |

• PROCEDURE

Suspend 77.9 G of powder in 1 L of distilled or deionized water. Add 2 vials of Raka Ray No.3 Agar supplement (MB-R0746). Heat to boiling until completely dissolved. Sterilize by autoclave at 121°C for 15 minutes. Cool to 45 - 50°C in water bath. Mix well. Pour into petri dishes.

Raka Ray No.3 Agar supplement

1vial contents (each vial is sufficient for 500mL of medium)

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|---------------------|----------|
| Cycloheximide | 0.0035 g |
| Phenylethyl Alcohol | 1.5 g |
| Tween 80 | 5.0 mL |

• INTERPRETATION

Raka Ray No.3 Agar is a medium used for the isolation and cultivation of lactic acid bacteria from beer and brewing processes. Tryptone, yeast extract and liver extract provide carbon, nitrogen, vitamins, amino acids and essential nutrients. Maltose, fructose and glucose are carbon energy sources. Betaine hydrochloride, N-acetyl glucosamine and tween 80 stimulate growth of lactobacilli. Diammonium citrate and dipotassium phosphate are the buffering agents. L-aspartic acid and potassium glutamate are sources of amino acids. Magnesium sulfate and manganese sulfate are added as trace elements. Agar is the solidifying agent. Cycloheximide and phenylethyl alcohol made the medium more selective by inhibiting yeast and Gram negative organisms.

• TECHNIC

Inoculate the specimen using a sterile loop to the medium or using the overlay technique. Incubate at 30 ± 2°C for 48 hours up to 7 days under anaerobic condition. Refer appropriate references for recommended test procedure.

• QUALITY CONTROL FOR USE

Dehydrated medium

Appearance: free-flowing, homogeneous

Color: light beige

Prepared medium

Appearance: slightly opalescent with slight precipitates

Color: light amber

Incubation conditions: 30 ± 2°C / 48 hours up to 7 days under anaerobic condition

| Microorganism | ATCC | Inoculum CFU | Growth |
|---|-------|------------------|-----------|
| <i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i> | 11842 | 50-100 | good |
| <i>Lactobacillus fermentum</i> | 9338 | 50-100 | good |
| <i>Lactobacillus plantarum</i> | 8014 | 50-100 | good |
| <i>Escherichia coli</i> | 25922 | ≥10 ³ | inhibited |
| <i>Saccharomyces cerevisiae</i> | 76625 | ≥10 ³ | inhibited |

• STORE

The powder is very hygroscopic. Store the powder at room temperature, in a dry environment, in its original container tightly closed and use it before the expiry date on the label. Store prepared medium at 2 - 8°C.

• REFERENCES

1. Saha R. B., Sondag R. J. and Middlekauff J. E., 1974, An improved medium for the selective culturing of lactic acid bacteria, Proceedings of the American Society of Brewing Chemists, 9th Congress, p. 9-10.
2. Methods of Analysis of ASBC, 1976, 7th Edi., The Society, St. Paul Mn USA
3. European Brewing Congress, EBC Analytica Microbiologica, 1981, J. Inst. Brewing 87:303-321. Murray, P. R., E. J. Baron, M. A. Pfaller, F
4. Hsu W. P., and Taporowsky J. A., 1977, Breweries Digest, 52 : 48.
5. Hug H. , Schlienger E. and Pfenniger H., 1978, Braveri- Rundschau, 89.145
6. Lawrence D. R. and Leedham P. A., 1979, J. Inst. Brewing, 85. 119.

• PACKAGE

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| Cat. No : MB-R1334 Raka Ray No.3 Agar | 500 G |
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