Skirrows Medium
(See Campylobacter Agars)

Snyder Test Agar

Intended Use
Snyder Test Agar, also known as BCG Dextrose Agar, is used for estimating the relative number of lactobacilli in saliva based on acid production.

Summary and Explanation
Tooth decay (dental caries) is a localized, progressive demineralization of the hard tissues of the crown and root surfaces of teeth. Streptococcus mutans and possibly lactobacilli ferment dietary carbohydrates that produce acids that cause the demineralization. The organisms reside in dental plaque, which is a gelatinous material that adheres to the surfaces of teeth. Demineralization of the tooth alternates with periods of remineralization. If demineralization exceeds remineralization, a subsurface carious lesion becomes a clinical cavity with extension of the decay into the dentine.

Snyder described a test procedure for determining, by colorimetric analysis, the rate and amount of acid produced by microorganisms in saliva. The procedure uses an agar medium that is known as Snyder Test Agar. Alban simplified the procedure, used it extensively and reported it to be more accurate than Snyder’s original procedure.

Principles of the Procedure
Snyder Test Agar contains peptones as sources of carbon, nitrogen, vitamins and minerals. Dextrose is the carbohydrate. Bromcresol green is the pH indicator. Agar is the solidifying agent.

Microorganisms that use the dextrose in the medium acidify the medium and the pH indicator, bromcresol green, changes color from blue-green to yellow.

Formula
Difco™ Snyder Test Agar

Approximate Formula* Per Liter
Proteose Peptone No. 3 ........................................... 10.0 g
Pancreatic Digest of Casein ......................................10.0 g
Dextrose ...................................................................20.0 g
Sodium Chloride ........................................................ 5.0 g
Agar .........................................................................20.0 g
Bromcresol Green ....................................................... 0.02 g

Directions for Preparation from Dehydrated Product
1. Suspend 65 g of the powder in 1 L of purified water. Mix thoroughly.

User Quality Control

Identity Specifications
Difco™ Snyder Test Agar
Dehydrated Appearance: Cream with green tint to light green, free-flowing, homogeneous.
Solution: 6.5% solution, soluble in purified water upon boiling. Solution is dark emerald green, very slightly to slightly opalescent.
Prepared Appearance: Dark emerald green, slightly opalescent.
Reaction of 6.5% Solution at 25°C: pH 4.8 ± 0.2

Cultural Response
Difco™ Snyder Test Agar
Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-72 hours.

<table>
<thead>
<tr>
<th>ORGANISM</th>
<th>INOCULUM</th>
<th>RECOVERY</th>
<th>ACID PRODUCTION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactobacillus rhamnosus</td>
<td>9595</td>
<td>Good</td>
<td>+</td>
</tr>
<tr>
<td>Lactobacillus fermentum</td>
<td>9338</td>
<td>Good</td>
<td>+</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>25923</td>
<td>None to poor</td>
<td>–</td>
</tr>
</tbody>
</table>
2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
3. Autoclave at 121°C for 15 minutes.
4. Test samples of the finished product for performance using stable, typical control cultures.

**Procedure**
Specimens should be collected preferably before breakfast, lunch, or dinner, and before the teeth are brushed. This procedure can be done just before lunch or dinner.

**Snyder Procedure**
1. Collect specimens of saliva in a sterile container while patient is chewing paraffin for 3 minutes.
2. Shake specimens thoroughly and transfer 0.2 mL to a tube of sterile Snyder Test Agar melted and cooled to 45°C. (Prepared medium in tubes is heated in a boiling water bath for 10 minutes and cooled to 45°C.)
3. Rotate the inoculated tubes to mix the inoculum uniformly with the medium and allow to solidify in an upright position.
4. Incubate at 35°C. Observe color at 24, 48 and 72 hours.

**Alban Modification**
1. Collect enough unstimulated saliva to just cover the medium in the tube. When specimen collection is difficult, dip a sterile cotton swab into the saliva under the tongue or rub on tooth surfaces and place the swab just below the surface of the medium.
2. Incubate the inoculated tubes and an uninoculated control at 35°C.
3. Examine tubes daily for 4 days.
4. Observe daily color change compared to control tube.

**Expected Results**
**Snyder Procedure**
Observe tubes for a change in color of the medium from bluish-green (control) to yellow. A positive reaction is a change in color so that green is no longer dominant; record as ++ to ++++. A negative reaction is no change in color or only a slight change (green is still dominant); record as 0 to +.

**Alban Modification**
a. No color change
b. Color beginning to change to yellow from top of medium down (+)
c. One half of medium yellow (+++)
d. Three fourths of medium yellow (++++)
e. The entire medium is yellow (++++)

The final report is a composite of the daily readings, for example; –, +, ++, ++++. The readings indicate the rapidity and amount of acid production.

**Limitations of the Procedure**
1. The data indicate only what is happening at the time the specimen was collected.
2. At least two specimens collected within 2-4 days must be obtained to establish a base-line or reference point.
3. Only when two or more specimens have been cultured can any reliability or prediction be obtained.
4. The clinician must study enough cases by use of periodic laboratory data to establish the value or significance for the purpose intended.

**References**

**Availability**
**Difco™ Snyder Test Agar**
Cat. No. 224710 Dehydrated – 500 g

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**Sodium Hippurate Broth**

**Intended Use**
Sodium Hippurate Broth is used in the determination of the ability of an organism to hydrolyze sodium hippurate by enzymatic action.

**Summary and Explanation**
Ayers and Rupp discovered that hemolytic streptococci from human and bovine sources could be differentiated by their ability to hydrolyze sodium hippurate. 1 Facklam et al. modified the test procedure in their study of the presumptive identification of groups A, B and D streptococci. 2

The ability of an organism to hydrolyze sodium hippurate is one of a number of tests that aid in the differentiation of bovine beta-hemolytic group B Streptococcus (S. agalactiae) from human beta-hemolytic group B Streptococcus species. 3 Differentiation of beta-hemolytic group B streptococci from beta-hemolytic group A streptococci and nonenterococcal group D streptococci is also aided by the determination of hippurate hydrolysis. 3