**Strep-A-Chek™ Kit**

**Reagent Strips and Color Developer**

Cat. No.: 13-050-00

**Intended Use**

Strep-A-Chek™ Kit is intended for use in the detection of pyrrolidonyl arylamidase (PYR) from beta-hemolytic colonies grown on blood agar plates, as an aid in the presumptive identification of Group A Streptococcus.

**Description**

Strep-A-Chek™ consists of Strep-A-Chek™ Reagent Strips impregnated with a chromogenic substrate for the detection of pyrrolidonyl arylamidase (PYR), an enzyme reported to be present in Group A beta-hemolytic Streptococcus, and EY-20™ Reagent Tubes which contain a diazo dye color developer, Fast Garnet. The PYR enzyme has been shown to be accurate in differentiating Group A streptococci and enterococci from other Streptococcus species. Strep-A-Chek™ Kit when used in conjunction with other tests such as CAMP, hiruprate, and bile-esculin, may be used for the presumptive identification of beta-hemolytic streptococci or enterococci from any source.

**Chemical Principle**

Hydrolysis of the chromogenic substrate impregnated on the Strep-A-Chek™ Reagent Strips by pyrrolidonyl arylamidase (PYR) releases a free beta-naphthylamide derivative. This complex with a diazo dye, Fast Garnet, the color developer present in EY-20™ Reagent Tubes, to produce a PINK/RED color, which is indicative of a positive result.

**Materials Supplied**

100 Strep-A-Chek™ Reagent Strips impregnated with 0.1% L-Pyrrol glutamyl-β-naphthylamide.
20 EY-20™ Reagent Tubes containing 0.35% Fast Garnet.
1 Material Safety Data Sheet (MSDS)

**Recommended Quality Control Organisms and Expected Results**

Good laboratory practices include the use of control specimens to ensure proper kit performance. Positive and negative organisms should be tested according to the laboratory’s established Quality Control program.

<table>
<thead>
<tr>
<th>ORGANISM (not supplied)</th>
<th>ATCC#</th>
<th>EXPECTED RESULTS</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Streptococcus pyogenes</em></td>
<td>19615</td>
<td>PINK/RED color change</td>
</tr>
<tr>
<td>Group C streptococci</td>
<td>12449</td>
<td>No color change</td>
</tr>
</tbody>
</table>

**Precautions**

Strep-A-Chek™ is intended for IN VITRO DIAGNOSTIC USE only and should be used by properly trained, qualified laboratory personnel. Normal precautions should be taken against dangers of microbial hazards.

**Storage and Stability**

Store Strep-A-Chek™ Reagent Strips and EY-20™ Reagent Tubes desiccated and in the original box at 2-8°C. This product should not be used passed the expiration date. Allow Strep-A-Chek™ Kit components to come to room temperature (20°C-28°C) before using. Protect EY-20™ Reagent Tubes from light and moisture. DO NOT use EY-20™ Reagent Tubes if visibly wet.

**Specimen Collection**

1. A GRAM STAIN and CATALYST™ TEST MUST be performed on the specimen before using Strep-A-Chek™ Group A Streptococcus are gram positive and catalase negative.

2. Only beta-hemolytic colonies should be selected from blood agar plates.

**Procedure**

1. Allow the Strep-A-Chek™ Kit components to come to room temperature (20°C-28°C) before using.

2. Reconstitute the contents of an EY-20™ Reagent Tube by adding 1.0 ml of distilled or deionized water to the tube and agitate. 1 ml of EY-20™ solution is sufficient for more than 5 tests.

**Notes**

- Store reconstituted EY-20™ Reagent at room temperature (20°C-28°C) protected from light. Use within 8 hours of reconstitution.

3. Remove Reagent Strip from its container. Remove at least 5 well isolated beta-hemolytic streptococci colonies from the blood agar plate using a wooden applicator stick or inoculation loop.

4. Inoculate reagent strip by rubbing colonies onto filter paper area of strip.

5. Add 1 drop of EY-20™ solution to the inoculated area. Incubate at room temperature (20°C-28°C) for up to 10 minutes.

6. View for color formation. Formation of a PINK/RED color in the test area indicates the detection of pyrrolidonyl arylamidase (PYR), a POSITIVE result for the presumptive identification of Group A Streptococcus. A NEGATIVE result should be recorded if there is no color change after 10 minutes.

**Interpretation of Results**

<table>
<thead>
<tr>
<th>OBSERVATION</th>
<th>INTERPRETATION</th>
<th>RESULT</th>
</tr>
</thead>
<tbody>
<tr>
<td>PINK/RED color change</td>
<td>Pyrrolidonyl arylamidase (PYR) detected</td>
<td>Presumptive identification of Group A Streptococcus</td>
</tr>
<tr>
<td>No color change</td>
<td>Pyrrolidonyl arylamidase (PYR) NOT detected</td>
<td>NEGATIVE</td>
</tr>
</tbody>
</table>

**Limitations of Test**

It must be emphasized that only pure cultures with characteristics listed in SPECIMEN COLLECTION should be tested with the Strep-A-Chek™ system. Some Leuconostoc and Streptococcus strains may appear coccobacillary, even rod shaped, and are often confused with members of the genus Lactobacillus. These strains may also be gram positive and catalase negative. The source of the specimen and clinical symptoms are important. Further biochemical and serological testing is necessary for definitive identification.

**Performance Characteristics**

In a clinical trial by Yajko, *et al.* comparing Strep-A-Chek™ with bacitracin disk susceptibility test for accuracy in the presumptive identification of *Streptococcus pyogenes* (Group A *Streptococcus* from a primary blood agar plate the sensitivity and specificity was 100%. Strep-A-Chek™ was evaluated using a total of 320 clinical isolates of beta-hemolytic streptococci (see table). These included 169 group A, 42 group B, 38 Group C, 21 group F, 39 group G and 11 beta-hemolytic streptococci which did not agglutinate with antisera to groups A, B, C, D, F, or G with the Strepex Latex agglutination test.

**Comparison of Bacitracin with Strep-A-Chek™**

<table>
<thead>
<tr>
<th>NO.</th>
<th>BACITRACIN SENSITIVE (%)</th>
<th>NO. PYR</th>
</tr>
</thead>
<tbody>
<tr>
<td>167</td>
<td>167</td>
<td>167</td>
</tr>
<tr>
<td>GROUP A</td>
<td>42</td>
<td>0</td>
</tr>
<tr>
<td>GROUP B</td>
<td>38</td>
<td>0</td>
</tr>
<tr>
<td>GROUP C</td>
<td>21</td>
<td>0</td>
</tr>
<tr>
<td>GROUP F</td>
<td>39</td>
<td>0</td>
</tr>
<tr>
<td>GROUP G</td>
<td>11</td>
<td>0</td>
</tr>
<tr>
<td>S. MILLER (GROUP A)</td>
<td>2</td>
<td>0</td>
</tr>
</tbody>
</table>

False positive rate for Bacitracin = 15%

In another clinical trial by Daly, *et al.* comparing Strep-A-Chek™ with Streptex and Luminex milk reduction for identification of *Streptococcus* the sensitivity and specificity was also 100%. A total of 311 isolates were evaluated and included 176 group A, 43 group B, 8 group C, 9 group F and 9 group G. 100% of 52 group D enterococci and 100% of 14 group D non-enterococci were identified by Strep-A-Chek™.

**Bibliography**

MATERIAL SAFETY DATA SHEET
Effective Date: March 31, 2006
Revision 3
Page 1 of 2

PRODUCT IDENTIFICATION
Name: Strep-A-Chek™ Kit
Catalog Number: 13-050-00

EMERGENCY INFORMATION
EY Laboratories, Inc.
107 North Amphlett Blvd.
San Mateo, CA 94401
EMERGENCY PHONE: 650-342-3296

HAZARDOUS COMPONENTS
MATERIAL CONCENTRATION
Pyroglutamic Acid-4-methoxynaphthylamide 0.01% solution dried on paper strip.
(PYR)
Fast Garnet GBC salt 0.35% (w:w) Fast Garnet:Glucose
CAS #: 101-89-3

HEALTH HAZARD INFORMATION
EXPOSURE LIMITS
None established. The toxicological properties of these chemicals have not been thoroughly investigated.

EFFECTS OF OVEREXPOSURE
The chemical may cause local irritation if allowed to contact skin. Irritation may result if affected skin is allowed to contact the eyes or mucous membranes of the nose or mouth.

ROUTES OF EXPOSURE
Fast Garnet may be harmful by inhalation, ingestion, or absorption through the skin. The chemical is supplied as a powder in the tube. The primary route of exposure would be by inhalation of the powder or by contact with the solution after reconstitution.

PHYSICAL CHARACTERISTICS
APPEARANCE
Fast Garnet, Light orange / brown powder.

FORM
Fast Garnet powder mixed with glucose.

SOLUBILITY in H₂O
100%.

FIRE AND EXPLOSION HAZARDS
Not considered to be a fire hazard.

EXTINGUISHING MEDIA
Water spray, CO₂, or dry chemical powder.

SPECIAL FIRE FIGHTING NOTE
Wear protective equipment to prevent contact with skin, eyes, and respiratory tract.

REACTIVITY DATA
STABILITY
Stable. Decomposition products are not known to be hazardous.

HAZARDOUS POLYMERIZATION
Will NOT occur.

INCOMPATIBILITY
Strong oxidizing agents, moisture, light, and alkaline conditions.

SPILL / LEAK PROCEDURES
MATERIAL RELEASE / SPILL
Avoid contact with material. Clean up spill and place all waste in a bag for disposal. Ventilate area.

WASTE DISPOSAL
Dissolve powder in water or buffer. Autoclave for 1 hour. Dispose of in accordance with all Local, State, and Federal regulation.

EMERGENCY FIRST AID PROCEDURES
May be harmful if swallowed, inhaled, or allowed to absorb through the skin. Wash contacted area with water for 15 minutes. If inhaled remove to fresh air. Report exposure to the appropriate safety official. Consult physician as necessary.

SPECIAL HANDLING PRECAUTIONS
VENTILATION
Mechanical exhaust recommended.

EYE PROTECTION
Safety glasses recommended.

RESPIRATORY PROTECTION
OSHA approved respirator.

PROTECTIVE GLOVES
Required.

ADDITIONAL INFORMATION
Avoid skin contact.

SPECIAL PRECAUTIONS
This material is for in vitro diagnostic use only. It is not intended for food, drug, household, agricultural, or cosmetic use. All material should be handled only by technically qualified individuals experienced with working with potentially hazardous chemicals. The above information is correct to the best of our knowledge. The user should make independent decisions regarding completeness of the information, based on all sources available. EY Laboratories, Inc. shall not be held liable for any damage resulting from handling or contact with the above product.