

K320/K1320 INDOLE-IPA

PRINCIPLE/DISCUSSION:

Key Indole-IPA Tablets contain the amino acid tryptophan, which is degraded by many organisms. The end product of the deamination is indole, which will react with an aldehyde reagent to form a colored end product. Members of the *Providencia* and *Proteus* species will also oxidize tryptophan to indole-pyruvic acid, which will produce color after the addition of ferric chloride.

MATERIALS REQUIRED:

Each tablet contains tryptophan with inert fillers. This product does not contain any material known at this time to be hazardous. K1320 are provided ready to use in tubes, 28 per pack. Tubes are required for K320 which is sold 50 tablets bulk packed. The tests require fresh 24 hour growth on appropriate media. Consult a microbiology manual for suggestions. The following items are required but not provided:

- Inoculating loop
- Distilled water, pH 7.0-7.2
- Key Kovacs Reagent Solution (Catalog # K170 or K980170, sold separately)
- Key Ferric Chloride Solution (Catalog #K190 or K980190, sold separately)

STORAGE:

Store tightly covered, with desiccant, in a dry place at room temperature.

PROCEDURE:

- (1) Add 5 drops of water to the WEE-TAB tube containing the tablet or place large tablet in tube, adding 1 ml of water.
- (2) Inoculate heavily. (5-6 colonies)
- (3) Incubate @35-37C for 4 hours for WEE-TABS or 24 hours for large tablets. If either the indole or IPA test are being done singly, follow the classic test directions below. If both tests are needed, do an indole swab test followed by the IPA. Positive indole indicates the test is finished and the IPA may be done. Negative indole results require more incubation time before calling the negative. Do not proceed with the IPA test, but return the tube to the incubator for up to 24 hours. Then repeat the swab test and proceed with the IPA test. This test cannot be used with spot indole reagent.
 - ♦ **Indole Swab test:** Dip a swab into the tube and drop it into another tube containing 2-3 drops of Kovacs' reagent or drop Kovac's directly onto the swab. If the test is positive the swab will turn red.
 - ♦ **Classic Indole Test:** After incubation, add 1-2 drops of Kovacs' reagent to the WEE-TAB or 5 drops to K320 without shaking. Observe for color.
 - ♦ **IPA Test:** After overnight incubation, add 1 drop of Key Ferric Chloride Solution to the WEE-TAB or 3-4 drops to K320. Observe for color.

INTERPRETATION:

Indole Test: After the addition of Kovacs Reagent, a positive test should develop a red color in the top layer within 3-4 minutes.

IPA Test: After addition of the Ferric chloride, a positive test will develop to dark red within 5-10 minutes.

QUALITY CONTROL:

Each bottle should be tested with known positive and negative organisms.

Dispose of all used material in a manner appropriate for biohazardous material.

Some common reactions are shown on the chart.

ORGANISM	INDOL	IPA
<i>Citrobacter freundii</i>	-	-
<i>Citrobacter diversus</i>	+	-
<i>E. coli</i>	+	-
<i>Enterobacter cloacae</i>	-	-
<i>Morganella morgani</i>	+	+
<i>Proteus mirabilis</i>	-	+
<i>Proteus vulgaris</i>	+	+
<i>Providencia spp.</i>	+	+
<i>Salmonella/Shigella spp.</i>	-	-

REFERENCES:

- (1) Manual of Clinical Microbiology, Fifth Edition, Chapter 36, "Enterobacteriaceae".
- (2) Bailey and Scott's Diagnostic Microbiology, Seventh Edition, Chapter 27 "Enterobacteriaceae".

