

PRO DISC

CK1532

Background

Enzymatic hydrolysis of L-proline- β -naphthylamide releases free β -naphthylamine which is detected and shown by the colour change after adding PEP reagent. This test is especially of use in screening for *Clostridium difficile* from CCEY. A negative test indicates the organism cannot be *C. difficile*. The test is also of value when used with other tests in the speciation of anaerobes, both Gram-positive and Gram-negative, and certain other organisms such as enterics.

Instructions

- (1) Place 1 disc onto a slide or into a small test tube. Moisten slightly with distilled water. (Do not oversaturate.)
- (2) Inoculate heavily with a loopful of organism from a fresh pure 24-hour culture plate.
- (3) Wait 2 minutes (at room temperature) then add 1 drop of PEP/Indole reagent (CK9053). Wait 1-2 minutes for colour to develop. Do not read after more than 2 minutes.

Interpretation

The appearance of a dark pink to red colour is positive.

Limitations

Indole reactions of organisms grown on any media containing tryptophane (e.g. blood agar) may interfere but will still produce predictable reactions. From such media,

positive PRO reactions will range from dark pink or red (PRO+/indole-) to very dark blue or purple (PRO+/indole+). Green or turquoise (indole+) and yellow (indole -) are both PRO negative.

Quality Control

Each lot should be tested with known positive and negative organisms. Some suggested strains are *C. difficile* ATCC#9689 (positive) and *C. perfringens* ATCC#13124 (negative). Positive PRO and Indole may be shown by testing with *C. sordelli*.

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

Storage

Store discs and reagent tightly covered with desiccant in a dry place at 2-8°C. Discs do not have to be warmed before use.

Health & Safety Information

The discs contain L-proline- β -naphthylamide: Naphthylamides have been identified as possible carcinogens however when used as directed the discs present no hazard. PEP/indole reagent contains hydrochloric acid, is poisonous, mildly corrosive and stains clothing and hands. Handle with care. Consult poison control centre if ingested.

References

- 1, Manual of Clinical Microbiology, Fifth Edition, Chapter 36, Enterobacteriaceae
- 2, Kilian M and Bulow P. 1976. Rapid Diagnosis of Enterobacteriaceae, Acta Path microbiol, Scand, Sect B, 84;245-251
- 3, Wadsworth Anaerobic Bacteriology Manual, 5th Edition, 1993, Glucosidase tests, page 152