

K9140 NGP WEE-TABS

PRINCIPLE/DISCUSSION:

It is known that approximately 50% of all clinical isolates and 80% of gram-negative bacilli are from the family *Enterobacteriaceae*(1); the most common of which is *Escherichia coli*. Most *Enterobacteriaceae* can be recognized by their reactions to a few chemical compounds. Though each may be used alone for specific applications, NGP Wee-Tabs provide three tests which, combined with urease and OMPI Wee-Tabs, will identify approximately 95% of such organisms. The tablets detect the presence of enzymes which hydrolyze various chromogenic substrates and employ the following principles:

1. The primary test is p-nitrophenol N-acetyl- α d-glucosaminide (NAG). Organisms producing the necessary enzymes hydrolyze this substrate, releasing the yellow nitro-phenol.
2. Enzymes acting on 4-methylumbelliferyl- β -D-glucoside (β -GLU) release the fluorescent methyumbelliferone which can be observed under a Wood's lamp.
3. The third test is proline aminopeptidase (PRO). Organisms containing the enzymes necessary for hydrolysis of the Proline- β -naphthylamide release free β -naphthylamide which is detected by the addition of aminopeptidase reagent.

MATERIAL SAFETY DATA:

K9140: NGP Wee-Tabs contain: NAG (p-nitrophenol-N-acetyl- α -D glucosaminide), β -GLU (4-methylumbelliferyl- β -D-glucoside), PRO (proline- β -naphthylamide), and other inert ingredients necessary for tableting. None of the substrates are harmful in this form.

MATERIALS REQUIRED:

NGP tablets are sold ready to use, 28 tubes per bottle. Usage requires 24 hour growth on media appropriate for the specimen. Consult a current reference manual for the correct media to use. The following items are also required but not provided: Microbiological loop or needle, Purified water, pH 6.5 - 7.5, Long-wave fluorescent light KS1699 or equal, and K2375 PEP reagent.

PROCEDURE:

- 1) Add 0.3-0.5 ml (about 5-8 drops) of water to the tube.
- 2) Inoculate the tube with a single colony from a primary agar plate (eg. Blood agar or MacConkey, etc). Mix well. OPTIONAL: To read at 2 hours or for brighter results, inoculate with 10-15 colonies.
- 3) Incubate @ 35-37C for 4 hours. Though finished at 4 hours, tests may be held for up to 28 hours. Do not read after more than 28 hours as false positives may occur.

INTERPRETATION:

- 1) **NAG:** Observe for a yellow color indicating a positive test. Colorless is negative.
- 2) **β -GLU:** Observe the tube for fluorescence, using a long-wave ultra-violet light. A positive 4-MU shows a **bright blue** fluorescence. **IMPORTANT-pale** fluorescence should be considered negative if PYR is positive. Pale fluorescence is only considered as a positive test when the PYR is negative.
- 3) **PRO:** Add 2 drops of PEP to the tube and reincubate for 15 minutes. A bright pink or red color indicates a positive test. After all tests are completed, refer to the chart.

NAME	TESTS USING COMBINATION TEST TABLETS OR DISCS								CONFIRMATION	
	ONPG	MUG	INDOLE	IPA	PYR	NAG	BGLU	PRO	UREA	LACTOSE
<i>Cedecea sp.</i>	+	-	-	-	-	+	+	-	-	V-
<i>Citrobacter sp.</i>	+	-	+(m)	-	+	-	+(m)	-	-	V
<i>Edwardsiella sp.</i>	-	-	+	-	-	+	-	+	-	-
<i>Enterobacter sp.</i>	+	-	-	V-	V+	V+	+	-	V-	V+
<i>Escherichia coli</i>	+	+(n)	+	-	-	-	-	-	-	+
<i>Escherichia sp.</i>	+	-	+	-	+	-	V-	-	-	+
<i>Hafnia alvei</i>	-	-	-	-	-	V+	-	+	-	-
<i>Klebsiella sp.</i>	+(a)	-	-(b)	-	+	-(c)	+	-	+(d)	V
<i>Kluyvera sp.</i>	+	-	+	-	-	-	+	-	-	+
<i>Leminorella sp.</i>	-	-	-	-	-	-(e)	-	-	-	-
<i>Morganella morgani</i>	-	-	+	+	-	-	-	-	+	-
<i>Proteus sp.</i>	-	-	-(f)	+	-	-	-	-	+	-
<i>Providencia sp.</i>	-	-	+	+	-	-(g)	-	-	V+	-
<i>Rahnella sp.</i>	+	-	-	+	+	-	+	+	-	+
<i>Salmonella sp.</i>	-(h)	V	-	-	-	-	-	-	-	-
<i>Serratia sp.</i>	+	-	-	-	+	+	+	+	+	-(i)
<i>Shigella sonnei</i>	+	+	-	-	-	-	-	-	-	-
<i>Shigella sp.</i>	-	V	V	-	-	-	-	-	-	-
<i>Yersinia sp.</i>	V	-	V	V-	-	V-	+(j)	-(k)	-	-

REFERENCES/FOOTNOTES:

- 1) Manual of Clinical Microbiology, 5th Edition, Chapter 36
- a) *K. rhinoscleromatis* is negative.
- b) *K. oxytoca* and *K. ornithinolytica* are positive.
- c) *K. ornithinolytica* is positive.
- d) *K. ozaenae* and *K. rhinoscleromatis* are negative.
- e) *Lem. grimonti* is positive.
- f) *P. vulgaris* may be weakly positive.
- g) Only *Prov. stuarti* is positive.
- h) *Salmonella arizonia* is positive.
- i) *Ser. rubidea* is usually positive.
- j) *Yer. enterocolitica* is negative.
- k) *Yer enterocolitica* is positive.
- m) *C. freundii* is negative.
- n) *E. coli* 0157 is MUG negative.



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