



RapidResa Polymyxin Acinetobacter NP® Test

Device for Rapid Detection of Colistin Resistance in *Acinetobacter baumannii*.

ENGLISH

DESCRIPTION

Acinetobacter baumannii primarily causes colonization, but may be an opportunistic pathogen associated with hospital-acquired infections. *A. baumannii* is intrinsically resistant to many antibiotic agents and it can also rapidly develop or acquire resistance to virtually all antibiotics. High percentage of carbapenem-resistant *A. baumannii* have been reported. Treatment options to multidrug-resistant bacteria are limited, with colistin (polymyxin E) being one of the most important last-resort antibiotics.

RapidResa Polymyxin Acinetobacter NP® Test is a panel containing desiccated biochemical substrates for rapid detection of colistin-resistant/-susceptible *Acinetobacter baumannii* isolates.

A panel allows testing of eight (8) different isolates.

Test results are evaluated by visual assessment of color development in the wells.

CONTENT OF THE KIT

- 2 panels of RapidResa Polymyxin Acinetobacter NP® Test (panels individually packed in foil with silica gel desiccant)
- 16 Tubes of Mueller Hinton II Broth (3.6 ml)
- 1 Vial of Resazurin Solution (1.2 ml)
- Resealable Bag
- Sealing Film
- Instructions Sheet

PRINCIPLE OF THE METHOD

This assay is based on the utilization of resazurin (7-hydroxy-3H-phenoxazin-3-one-10-oxide) as cell viability indicator and a comparison of bacterial growth in medium with or without a defined concentration of colistin. There is a direct correlation between the reduction of resazurin in the growth medium and proliferation of cells. Resazurin is blue in color when it is in an oxidized state and turns pink/violet when reduced by viable cells.

CONFIGURATION

Tests are placed in columns: each test consists of three (3) wells, each indicated with a letter:

○ a	○ a	○ a	○ a	○ a	○ a	○ a	○ a	Well a is the negative control
○ b	○ b	○ b	○ b	○ b	○ b	○ b	○ b	Well b does not contain any antimicrobial agent
○ c	○ c	○ c	○ c	○ c	○ c	○ c	○ c	Well c contains colistin

COLLECTION AND STORAGE OF THE SAMPLE

Colonies to be examined must be properly isolated and taken from a solid culture medium used for isolation of the test organism.

TEST PROCEDURE

Preparation of the specimen

1. Suspend well-isolated colonies from an overnight agar plate into a vial of Muller Hinton II Broth provided in the kit.
2. Adjust the suspension to achieve a turbidity equivalent to a 0.5 McFarland standard. This results in a suspension containing approximately $1-2 \times 10^8$ CFU/mL. Optimally, use inoculum within 15 minutes.

Inoculation of the panel

3. Take a panel from its envelope and leave it at room temperature for 10 min.
NOTE Use scissors to cut off the envelope at one end in order to maintain its integrity and functioning. Do not discard the envelop until all the 8 tests have been carried out.
4. Add 200 µl of Mueller Hinton II Broth to well **a** (sterility control).
5. Add 200 µl of isolate suspension to wells **b** and **c**.
6. Add 20 µl of the resazurin solution to each well in a single column, i.e. **a**, **b** and **c**.
7. Mix the reagent with the medium by pipetting up and down
8. Cover the panel with the lid provided and incubate at $36 \pm 1^\circ\text{C}$ for 4 hours in ambient air.













EVALUATING THE RESULTS

At the end of incubation period, observe the color change in the wells and interpret results as follow:

Well	Susceptible	Resistant
a	blue	blue
b	pink/violet	pink/violet
c	blue	pink/violet

If all the wells in a column appear either pink/violet or blue the result cannot be reported for that particular test. In that case, check the test procedure and the viability of the colonies. Repeat the test using a new columns in the same panel or a new panel and a microbial culture of recent growth.

Examples of Color Reactions for 4 *Acinetobacter baumannii* isolates:

	Isolate 1	Isolate 2	Isolate 3	Isolate 4
a				
b				
c				
Interpretation	Colistin-resistant	Colistin-susceptible	Invalid test	Invalid test

NOTE If not all the 8 tests have been performed, use the film provided in the kit to seal the inoculated columns so to prevent any leakage of contaminated fluids. Then, return the panel into its own desiccant envelop and subsequently into the resealable bag provided with the kit. Store into the refrigerator until time of testing (see STORAGE).

USER QUALITY CONTROL

Quality control of RapidResa Polymyxin Acinetobacter NP® Test is performed using colistin-resistant and colistin-susceptible *A. baumannii* strains.

FACTORS THAT MAY INVALIDATE THE RESULTS

Contaminated culture; poor standardization of the inoculum; clinical material unsuitable; use of expired panels or expired supplementary reagents; non compliance with temperatures and times of incubation.

PRECAUTIONS

RapidResa Polymyxin Acinetobacter NP® Test does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. It is nevertheless recommended to consult the safety data sheet for its correct use. The product is a disposable device to be used for professional use only. It must be used in the laboratory by properly trained personnel, using approved aseptic and safety methods for handling pathogenic agents.

STORAGE

Store RapidResa Polymyxin Acinetobacter NP® Test at 2-8°C in the original packaging. Once an envelope is opened the panel should be used within 15 days. Keep away from direct sunlight and direct heat. Do not use the panels beyond the expiry date indicated on the label. Eliminate without using if there are signs of deterioration.

DISPOSAL OF USED MATERIAL

After use, RapidResa Polymyxin Acinetobacter NP® Test and material that has come into contact with the sample must be decontaminated and disposed of in accordance with guidelines used in the laboratory for decontamination and disposal of potentially infected material.

REFERENCES

1. Lescat M, Poirel L, Tinguely C, Nordmann P. A Resazurin Reduction-Based Assay for Rapid Detection of Polymyxin Resistance in *Acinetobacter baumannii* and *Pseudomonas aeruginosa*. J Clin Microbiol. 2019;57(3).

TABLE OF SYMBOLS

LOT Batch code	 Do not reuse	 Manufacturer	 Contains sufficient for <n> tests	 Temperature limits
REF Catalogue number	 Fragile, handle with care	 Use by	 Caution, consult accompanying documents	

Product	Packaging	Ref.
RapidResa Polymyxin Acinetobacter NP® Test	2 panels (2 x 8 tests)	76046



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