Pseudalert®

QUANTIFICATION - PROCESS STEPS

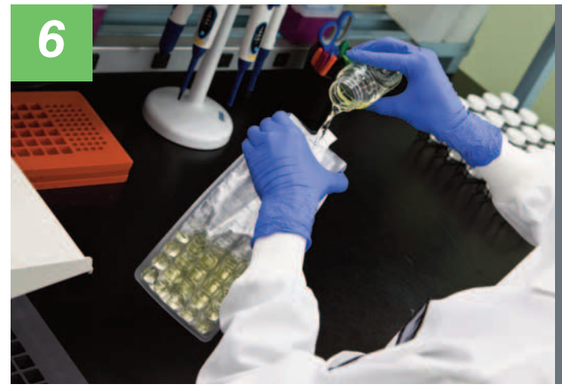
1 Tear Pseudalert® reagent snap pack from the strip.

2 Snap open the snap pack.

3 Add Pseudalert reagent to 100 ml water sample.

4 Cap the vessel and shake to dissolve.

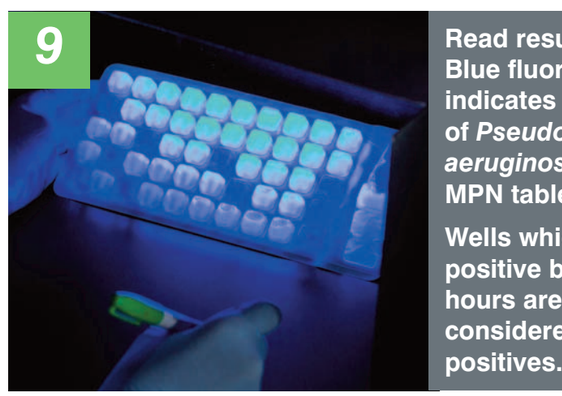
5 Add 2 drops of IDEXX antifoam to the vessel **OR** use antifoam vessels.



6 Pour sample into Quanti-Tray®.

7 Seal the Quanti-Tray® using the IDEXX Sealer Plus.

8 Incubate the Quanti-Tray® for 24-28 hours at 38⁰C±0.5⁰C.



9 Read results:
Blue fluorescence indicates the presence of *Pseudomonas aeruginosa*. Refer to MPN table.

Wells which appear positive before 24 hours are also considered confirmed positives.

The process steps above are intended as a guide only. Please refer to the current product insert for detailed instructions.

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IDEXX

ISO 9001:2015 CERTIFIED
ISO 14001:2015 CERTIFIED

ISO 11133:2014 COMPLIANT
ISO 17025:2005 ACCREDITED

Pseudalert* Test Kit

Introduction

The Pseudalert* test detects the presence of *Pseudomonas aeruginosa* in water samples. The test is based on a bacterial enzyme detection technology that signals the presence of *Pseudomonas aeruginosa* through the hydrolysis of a substrate present in the Pseudalert reagent. *Pseudomonas aeruginosa* cells rapidly grow and reproduce using the rich supply of amino acids, vitamins and other nutrients present in the Pseudalert reagent. Actively growing strains of *Pseudomonas aeruginosa* have an enzyme that cleaves the substrate to produce a blue fluorescence under ultraviolet (UV) light. Pseudalert detects *Pseudomonas aeruginosa* at 1 cfu in either 100 mL or 250 mL samples within 24 hours.

Storage

Store at 2–30°C away from light

Presence/Absence (P/A) Procedure

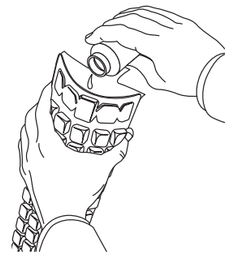
1. Add contents of one appropriately sized snap pack to either a 100 mL or 250 mL sample in a sterile, transparent, nonfluorescing vessel.
2. Cap vessel and shake.
3. Incubate at 38±0.5°C for 24 to 28 hours.
4. Read results according to Result Interpretation table below.

Quanti-Tray* Enumeration Procedure (100 mL Samples Only)

1. Cap vessel and shake until dissolved.
2. Add 2 drops of IDEXX Antifoam Solution[1] to the sample/reagent mixture.
   NOTE: IDEXX 120 mL sample vessels[2] containing antifoam are also available.
3. Pour sample/reagent mixture into a Quanti-Tray* or Quanti-Tray*/2000 and seal in a Quanti-Tray* Sealer.
4. Place the sealed tray in a 38±0.5°C incubator for 24 to 28 hours.
5. Read results according to the Result Interpretation table below. Count the number of positive wells and refer to the MPN table provided with the trays to obtain a Most Probable Number.

NOTE: Quanti-Tray* Enumeration Procedure for 250 mL available from IDEXX.

Result Interpretation



Appearance	Result
No blue fluorescence	Negative for *Pseudomonas aeruginosa*
Blue fluorescence†	Positive for *Pseudomonas aeruginosa*

†Presence of blue fluorescence greater than the amount present in a negative control sample

- Look for blue fluorescence with a 6-watt, 365-nm UV light held within 5 inches of the sample in a dark environment. Face light away from your eyes and toward the sample.
- Refer to –/+ fluorescence read guide on kit box. Chart colors are as seen under a UV light.
- Pseudalert results are definitive at 24–28 hours. In the presence/absence procedure, positives for *Pseudomonas aeruginosa* observed before 24 hours, and negatives observed after 28 hours are also valid.

Procedural Notes

- Use only sterile, nonbuffered, oxidant-free water for dilutions.
- For comparison, an incubated sterile water blank containing Pseudalert reagent (negative control) can be used when interpreting results.
- This insert may not reflect your local regulations. For compliance testing, be sure to follow appropriate regulatory procedures.
- Pseudalert is a primary water test. Pseudalert performance characteristics do not apply to samples altered by preenrichment or concentration.
- Pseudalert has not been validated for use with flavored bottled water, marine water or carbonated water samples.
- Aseptic technique should always be followed when using Pseudalert. Dispose of materials in accordance with Good Laboratory Practices.
- The presence of a high mineral content (especially magnesium and/or calcium) can cause the Pseudalert reagent mixture to become cloudy, but this does not affect the outcome.
- Interpret any blue fluorescence as positive, even if the fluorescent signal is weak
- If you are unsure about a well or vessel with weak fluorescence at 24 hours, incubate for another 1-4 hours.

Quality Control Procedures

1. One of the following quality control procedures is recommended for each lot of Pseudalert:
   A. IDEXX-QC *Pseudomonas*[3]: *Pseudomonas aeruginosa*, *Escherichia coli*, *Pseudomonas fluorescens*.
   B. Additional Quality Control Methods.
      i. For each of the American Type Culture Collection (ATCC)[4] bacterial strains, (*Pseudomonas aeruginosa* ATCC 27853 (WDCM 00025) or 10145 (WDCM 00024), *Escherichia coli* ATCC 25922 (WDCM 00013) and *Pseudomonas fluorescens* ATCC 13525 (WDCM 00115)) streak the culture onto labeled TSA or blood agar plates and incubate at 35±0.5°C for 18–24 hours.
      ii. For each bacterial strain, touch a sterile 1 µL inoculating loop to a colony and use it to inoculate a labeled test tube containing 5 mL of sterile deionized water. Close cap and shake thoroughly.
      iii. For each bacterial strain, take a 1 µL loop from the test tube and use it to inoculate a labeled vessel containing either 100 mL or 250 mL of sterile deionized water. These are your controls.
2. Follow the P/A Procedure or Quanti-Tray Enumeration Procedure above.
3. Results should match the Result Interpretation table above.

NOTE: IDEXX internal quality control testing is performed in accordance with ISO 11133:2014. Quality Control Certificates are available at idexx.com/water.

1. IDEXX Antifoam Solution catalog number: WAFDB
2. IDEXX 120 mL Shrink-banded Vessels with Antifoam catalog number: WV120SBAF-200
3. IDEXX-QC *Pseudomonas*, IDEXX Catalog #UN3373-WQC-PSE
4. American Type Culture Collection, 1-800-638-6597 atcc.org