

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE**

A. 510(k) Number:

k062461

B. Purpose for Submission:

New product *Candida albicans* PNA FISH™

C. Measurand:

C. albicans specific 26S ribosomal RNA

D. Type of Test:

Fluorescent In Situ Hybridization (FISH) using protein nucleic acid (PNA) probes

E. Applicant:

AdvanDx, Inc.

F. Proprietary and Established Names:

C. albicans PNA FISH™, *Candida albicans* Culture Identification Kit

G. Regulatory Information:

1. Regulation section:

21 CFR Part 866.2660 – Microorganism differentiation and identification device

2. Classification:

Class I

3. Product code:

JXB – Kit, Identification, Yeast

NZS – FISH (Fluorescent In Situ Hybridization) kit, protein nucleic acid, rna, yeast

4. Panel:

83 Microbiology

H. Intended Use:

1. Intended use(s):

C. albicans PNA FISH™ is a qualitative nucleic acid hybridization assay intended for identification of *Candida albicans* from blood cultures.

2. Indication(s) for use:

C. albicans PNA FISH™ is a qualitative nucleic acid hybridization assay, intended for the identification of *Candida albicans* from blood cultures.

3. Special conditions for use statement(s):

Prescription use only.

4. Special instrument requirements:

AdvanDx Teflon-coated Microscope Slides. A fluorescent microscope equipped with an AdvanDx Dual Band Filter.

I. Device Description

The *C. albicans* PNA FISH™ Culture Identification Kit contains a 3 mL bottle of fixation solution, a 1.5 mL bottle of fluorescein-labeled PNA probe in hybridization solution, a 50 mL bottle of concentrated wash solution, which must be diluted prior to use, and a 3 mL bottle of mounting medium. The one-well, Teflon-coated microscope slides, glass coverslips and the external quality control organism slides are sold separately. User-prepared quality control organism slides are acceptable. After processing, the slides must be examined within two hours by using a fluorescent microscope equipped with a dual band filter.

J. Substantial Equivalence Information:

1. Predicate device name(s):

- a. Conventional Methods-similar Indication for Use
- b. BD Affirm™ Microbial Identification Test – similar Indication For Use
- c. *S. aureus* PNA FISH – similar technology

2. Predicate K number(s):

- a. K931374
- b. K060099

Note: A pre-amendment device was also referenced.

3. Comparison with predicate(s):

| Item | Test Device | Pre-Amendment | Primary Predicate | Secondary Predicate |
|-----------------------------|---|---|--|--|
| Product Name | <i>C. albicans</i> PNA FISH | Conventional Routine Methods | Affirm VPIII | <i>S. aureus</i> PNA FISH |
| Intended Use | Identification of <i>Candida albicans</i> | Identification of yeast from colonies isolated on solid media | Detection and identification of <i>Candida</i> species | Identification of <i>Staphylococcus aureus</i> |
| Technology Method | Fluorescence in situ hybridization | Phenotypic, Biochemical | Colorimetric DNA probe hybridization | Fluorescence in situ hybridization |
| Time to result | 2.5 hours from time of smear preparation | 1 – 7 days | 1 hour | 2.5 hours from time of smear preparation |
| Sample | Smear of blood cultures | Yeast colonies isolated on solid media | Vaginal fluid | Smear of blood cultures |
| Control organisms | <i>Candida albicans</i> and <i>Candida glabrata</i> | Test dependent | Positive Control and Negative Control beads | <i>S. aureus</i> and <i>S. epidermidis</i> |
| Mechanism of identification | <i>C. albicans</i> specific 26S rRNA | Test dependent | DNA capture probe and Color development probe | <i>S. aureus</i> specific 16 S rRNA |
| Interpretation of results | Qualitative fluorescent microscopy | Qualitative-Test dependent | Visual Qualitative | Qualitative fluorescent microscopy |

K. Standard/Guidance Document Referenced (if applicable):

Not applicable

L. Test Principle:

One drop of fluorescein-labeled, *C. albicans*-specific PNA probe is added to a methanol, heat, or flame fixed smear, prepared from liquid blood culture media with GPCC. Hybridization is performed during a 90 +/- 5 minute incubation at 55 +/- 1° C, in an incubator or on a slide warmer. The slide is examined by fluorescent microscopy within two hours of staining. *C. albicans* is identified as multiple bright green

fluorescent yeast cells in multiple fields on a reddish background, whereas non-*C. albicans* cells will not fluoresce.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

The performance data were generated by multiple, geographically distinct, clinical laboratories. Each laboratory compared the device results to corresponding results obtained by standard culture identification procedures. These laboratories reported the sensitivity and specificity results in peer reviewed literature articles, and during presentations performed at professional meetings (posters).

a. ***Precision/Reproducibility:***

Inter-laboratory and Intra-laboratory testing demonstrated >95% reproducibility. The ten isolate study described in the guidance document was used. (10 organisms tested 3 times on 3 days at 3 sites.)

b. ***Linearity/assay reportable range:***

Not applicable

c. ***Traceability, Stability, Expected values (controls, calibrators, or methods):***

The recommended QC isolates, *Candida albicans* and *Candida glabrata* as user-prepared slides from stock or liquid blood cultures, are recommended for testing in parallel for each batch of tests performed. At one testing site (Ohio) the *C. albicans* PNA FISH user-prepared organism control slides were tested a sufficient number of times to demonstrate that the device can produce acceptable quality control results >95% of the time (244/244).

The stability of the *C. albicans* PNA FISH reagents were evaluated using real-time data. The sponsor periodically removed samples from five lots of *C. albicans* PNA FISH reagent from storage for testing. The analytical fluorescence performance and a functional performance of each lot were tested 5 times over 30 months after the manufacturing date. The fluorescent dye on the probe is light sensitive and is considered the least stable component of the kit. There was no change in performance for at least 24 months when stored at 2 - 8° C and protected from light, as indicated in the product labeling. This study is on-going and data are being collected to potentially extend the shelf life.

d. ***Detection limit:***

The claimed detection limit for *C. albicans* in blood cultures was determined to be approximately 1×10^5 colony forming units (CFU) per mL by serial dilutions of a *C. albicans* positive culture. This is consistent with the analytical sensitivity of slide-based staining techniques and is not limited by the test itself, but rather by the general requirement for 1×10^5 CFU/mL for interpretation by standard light microscopy.

e. **Analytical specificity:**

Specificity of *C. albicans* PNA FISH probe was evaluated using cultures of 23 reference strains (13 *C. albicans* strains and 10 strains of other species), 25 spiked blood culture bottles and 76 clinical isolates. The analytical specificity for *C. albicans* strains positive by PNA FISH was 100% (13/13), and 100% (111/111) of the other strains were negative.

An Advanced BLAST search of the GeneBank nr-database (www.nlm.nih.gov/blast) showed that the target sequence is unique for *C. albicans*, and is not found in other species. The *C. albicans* PNA probe targets a ribosomal sequence, which is well-suited for the design of species-specific probes. Some *C. albicans* sequences have a single mismatch to the probe, but no *C. albicans* sequences have more than one mismatch.

f. **Assay cut-off:**

Not applicable

2. **Comparison studies:**

a. **Method comparison of device to conventional methods, as the reference method:**

The performance of *C. albicans* PNA FISH was evaluated using two different automated blood culture media systems at multiple sites. The data demonstrates that the *C. albicans* PNA FISH™ is compatible with two of the three major blood culture systems, and results from testing are comparable to results obtained by conventional methods. The results are displayed in the table below.

Automated blood culture system media evaluation comparing device results to results obtained by subculture and subsequent identification by standard methods, by study site

| Blood Culture System Evaluated | Sensitivity | Specificity | Study |
|--------------------------------|--------------------|---------------------|--------------------------|
| BacT/Alert | 100.0% (9/9) | 100% (24/24) | A |
| BACTEC BacT/Alert | 99.0% (100/101) | 100.0% (143/143) | B |
| BacT/Alert | 100.0% (29/29) | 100.0% (28/28) | C |
| Total | 99.3% (138/139) | 100.0% (195/195) | 334 Total isolates |

BacT/Alert = BacT/Alert Blood culture system (bioMérieux, Durham, NC)

BACTEC = BACTEC Blood culture system (Becton Dickenson, Sparks, MD)

