

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY**

A. 510(k) Number:

K092655

B. Purpose for Submission:

SE determination for a modification of the assay procedure for *C. albicans* PNA FISH. The specific modifications are: elimination of the 5-10 minutes ethanol step in smear preparation; a reduction of the hybridization time from 90 minutes to 30 minutes.

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:

Product Code	Classification	Regulation Section	Panel
NZA	Class 1	21 CFR 866.2660	83 Microbiology

H. Intended Use:

1. Intended use:

C. albicans PNA FISH™ is a qualitative nucleic acid hybridization assay intended for identification of *Candida albicans* on smears made from yeast positive blood cultures containing yeast observed on Gram stain or other microbiological stains. Subculturing

of positive blood cultures is necessary for susceptibility testing and/or differentiation of mixed growth.

2. Indication for use:

C. albicans PNA FISH™ is a qualitative nucleic acid hybridization assay, intended for the identification of *Candida albicans* on smears made from yeast positive blood cultures containing yeast observed on Gram stain or other microbiological stains. Subculturing of positive blood cultures is necessary for susceptibility testing and/or differentiation of mixed growth.

3. Special conditions for use statement:

Prescription use only.

4. Special instrument requirements:

AdvanDx Teflon-coated Microscope Slides. (Cat. AC001) A fluorescent microscope equipped with an AdvanDx Dual Band Filter (Cat. No. AC003)

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 cover slips 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:

C. albicans PNA FISH™

2. Predicate K number

K062461

3. Comparison with predicate:

Similarities		
Technology	Fluorescence In Situ Hybridization (FISH) using protein nucleic acid (PNA) probe	Same
Sample	Positive blood culture	Same
PNA Probes	Fluorescein-labeled <i>C.albican</i> specific PNA probe	Same

Differences		
Item	Device	Predicate
Fixed smear treatment	None	Ethanol for 10 minutes and air dried
Hybridization at 55°C	30 minutes	90 minutes

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 a positive blood culture bottle. Hybridization is performed during a 30 +/- 5 minute incubation at 55 +/- 1⁰ 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:

a. *Precision/Reproducibility:*

A reproducibility study for *C. albicans* PNA FISH™ assay was performed by using ten reference yeast isolates in different concentrations, once per day with positive and negative controls, over a period of three days at three different sites, by at least two different operators at each site. Results showed 100% precision and reproducibility between and within sites.

Summary of Reproducibility Results by Sites Across 3 Days

	Site 1	Site 2	Sites 3	Total Agreement
Positive Agreement	54/54	54/54	54/54	100 % (162/162)
Negative Agreement	63/63	63/63	63/63	100% (189/189)
Total Agreement	100% (117/117)	100% (117/117)	100% (117/117)	100% (351/351)
Positive/Negative Control	9/9	9/9	9/9	27/27

b. Linearity/assay reportable range:

Not applicable

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

Positive and negative control slides were performed at each testing site. All results were as expected.

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 53 laboratory and reference strains (47 fungal strains and 6 strains of other species), the analytical specificity for *C. albicans* strains positive by PNA FISH™ was 100% (18/18), and 100% (35/35) 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.

The *C. albicans* PNA probe sequence is highly specific for identification of *C. albicans*.

f. Assay cut-off:

Not applicable

2. Comparison studies:

a. *Method comparison with predicate device:*

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 comparable 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.

Performance Data for C. albicans PNA FISH (New) vs. C. albicans PNA FISH (K062461) on Yeast-positive Blood Culture Bottles (Clinical and Seeded Samples)

Study	Positive Agreement <i>C. albicans</i>	Negative Agreement	Blood Culture System
A	20/20	34/34	BactT/Alert
B	9/9	51/51	BactT/Alert
C	12/12	39/39	BACTEC
Total	100% (41/41) 95% CI (93.0-100)	100% (124/124) 95% CI (97.6-100)	N = 165

Performance Data for C. albicans PNA FISH (New) vs. C.albicans PNA FISH (New) vs. Routine Identification Methods on Yeast-positive Blood Culture Bottles (Clinical Samples)

Study	Sensitivity <i>C. albicans</i>	Specificity	Blood Culture System
A	<i>15/15</i>	<i>14/14</i>	BactT/Alert
B	<i>4/4</i>	<i>31/31</i>	BactT/Alert
C	<i>12/12</i>	<i>39/39</i>	BACTEC
Total	<i>100% (31/31)</i> <i>95% CI (90.8-100)</i>	<i>100% (84/84)</i> <i>95% CI (96.5-100)</i>	N = 115

b. *Matrix comparison:*

Not applicable

3. Clinical studies:

a. *Clinical Sensitivity:* Not applicable

b. *Clinical specificity:* Not applicable

c. Other clinical supportive data (when a. and b. are not applicable):

Not applicable

4. Clinical cut-off:

Not applicable. Qualitative visual color change.

5. Expected values/Reference range:

The expected *C. albicans* positive result rate from yeast positive blood culture bottles is 25% - 50%, depending on institutional and patient population

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.