

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

A. 510(k) Number:

K093024

B. Purpose for Submission:

To obtain a SE determination for a modification of the assay procedure for Yeast Traffic Light 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:

Candida tropicalis, *Candida albicans* and *Candida parapsilosis*, *Candida glabrata* and *Candida krusei* specific ribosomal RNA sequences

D. Type of Test:

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

E. Applicant:

AdvanDx, Inc

F. Proprietary and Established Names:

AdvanDx Yeast Traffic Light PNA FISH Culture Identification Kit

G. Regulatory Information:

1. Regulation section:

866.2660

2. Classification:

Class I

3. Product code:

NZS

4. Panel:

83 Microbiology

H. Intended Use:

1. Intended use(s):

Yeast Traffic Light PNA FISH is a multicolor, qualitative nucleic acid hybridization assay intended for the identification of *Candida albicans* and/or *Candida parapsilosis*, identification of *Candida tropicalis*, and identification of *Candida glabrata* and/or *Candida krusei* on smears made from positive blood cultures containing yeasts observed on Gram stain or other microbiological stains. The test does not distinguish between *C. albicans* and *C. parapsilosis*. The test does not distinguish between *C. glabrata* and *C. krusei*.

Subculturing of positive blood cultures is necessary to recover organisms for susceptibility testing, differentiation between *C. albicans* and *C. parapsilosis*, differentiation between *C. glabrata* and *C. krusei*, and/or differentiation of mixed growth.

Yeast Traffic Light PNA FISH is indicated as an aid in the diagnosis of *Candida tropicalis*, *Candida albicans* and *Candida parapsilosis*, and *Candida glabrata* and *Candida krusei* fungemia.

2. Indication(s) for use:

Yeast Traffic Light PNA FISH is a multicolor, qualitative nucleic acid hybridization assay intended for the identification of *Candida albicans* and/or *Candida parapsilosis*, identification of *Candida tropicalis*, and identification of *Candida glabrata* and/or *Candida krusei* on smears made from positive blood cultures containing yeasts observed on Gram stain or other microbiological stains. The test does not distinguish between *C. albicans* and *C. parapsilosis*. The test does not distinguish between *C. glabrata* and *C. krusei*.

Subculturing of positive blood cultures is necessary to recover organisms for susceptibility testing, differentiation between *C. albicans* and *C. parapsilosis*, differentiation between *C. glabrata* and *C. krusei*, and/or differentiation of mixed growth.

Yeast Traffic Light PNA FISH is indicated as an aid in the diagnosis of *Candida tropicalis*, *Candida albicans* and *Candida parapsilosis*, and *Candida glabrata* and *Candida krusei* fungemia.

3. Special conditions for use statement(s):

Prescription use only

4. Special instrument requirements:

Dual Band Filter (Cat. No. AC003)
 Microscope Slides (Cat. No. AC001)

I. Device Description:

Yeast Traffic Light PNA FISH is a fluorescence *in situ* hybridization (FISH) method using PNA probes hybridizing to specific ribosomal RNA sequences of *Candida albicans*, *C. parapsilosis*, *C. glabrata*, *C. krusei*, and *C. tropicalis*. The test provides rapid identification of *C. albicans* + *C. parapsilosis*, *C. tropicalis*, and *C. glabrata* + *C. krusei* on smears made from yeast positive blood cultures within 90 minutes.

The PNA FISH technology in the Yeast Traffic Light PNA FISH (new) is similar to the technology used for Yeast Traffic Light PNA FISH (k080719) differing only in the elimination of the ethanol step in the sample preparation and the reduction of hybridization time from 90 to 30 minutes.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Yeast Traffic Light PNA FISH

2. Predicate 510(k) number(s):

K080719

3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Technology	Fluorescence In Situ Hybridization (FISH) using protein nucleic acid (PNA) probe	Same
Sample	Positive blood culture	Same
PNA Probes	<i>C. albicans</i> PNA/Flu <i>C. parapsilosis</i> PNA/Flu <i>C. tropicalis</i> PNA/Flu <i>C. tropicalis</i> PNA/Tam <i>C. glabrata</i> PNA/Tam <i>C. krusei</i> PNA/Tam	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):

Non applicable

L. Test Principle:

A mixture of fluorophore-labeled PNA probes is added to a smear prepared from a culture. Hybridization is performed at 55°C for 30 minutes. The hybridization is followed by a post-hybridization wash at 55°C for 30 min. with a stringent Wash Solution to remove unbound PNA probe, then the smear is mounted with Mounting Medium and examined by fluorescence microscopy.

The *C. albicans*-specific PNA probe and the *C. parapsilosis*-specific PNA probe are labeled with fluorescein (green). Both the *C. albicans* and the *C. parapsilosis* are identified as green fluorescent cells. The test does not distinguish between *C. albicans* and the *C. parapsilosis*.

Two *C. tropicalis*-specific PNA probes of the same sequence are labeled with fluorescein (green) and rhodamine (red), respectively. *C. tropicalis* is identified by combined read and green fluorescent probes binding in cells which then appear yellow by using the dual band filter.

The *C. glabrata*-specific PNA probe and the *C. krusei*-specific PNA probe are labeled with rhodamine (red). *C. glabrata* and *C. krusei* are identified as red fluorescent cells. The test does not distinguish between *C. glabrata* and the *C. krusei*.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

The assay was performed on 13 slides (19 isolates) in triplicate on three separate days at three separate sites. Each batch was run independently by at least two different operators at each site. The reproducibility was >95%.

b. *Linearity/assay reportable range:*

Not applicable

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

Positive Control:

<i>C. albicans</i> ATCC 18804	Green
<i>C. glabrata</i> ATCC 2001	Red
<i>C. tropicalis</i> ATCC 750	Yellow

Negative Control:

S. cerevisiae ATCC 18824 Negative

All results were as expected.

d. *Detection limit:*

The detection limit was determined to be approximately 10^5 CFU/mL by serial dilutions of *C. tropicalis*, *C. albicans*, *C. parapsilosis*, *C. glabrata* and *C. krusei* cultures. The average number of colonies per mL (CFU/mL) was calculated from three plates. The data sets showed a minimum of 10^5 CFU/mL to produce a positive result for the Yeast Traffic Light PNA FISH™ assay.

e. *Analytical specificity:*

Yeast Traffic Light PNA FISH has been evaluated on 75 laboratory and reference strains comprising *Candida* species and other closely related yeasts species and a variety of other frequently isolated organisms. The results demonstrated:

All (10/10) *C. albicans*, including two *C. stellatoidea* (a variant of *C. albicans*), and (4/4) *C. parapsilosis* strains were green-positive.

Both (2/2) *C. tropicalis* were yellow-positive.

All (6/6) *C. glabrata*, (1/1) *Issatchenkia orientalis* (teleomorph of *C. krusei*) and (3/3) *C. krusei* strains were red-positive.

Candida nivariensis, *Candida bracarensis*, and *Kluyveromyces delphensis* cross-reacted and produce a red signal. *Candida orthopsilosis* (3/3) and *Candida metapsilosis* cross-reacted to create a green signal. One of two strains of *Candida sojae* cross-reacted and produce a yellow signal. All other (27/27) fungal and (13/13) bacteria strains were negative.

f. *Assay cut-off:*

Not applicable

2. Comparison studies:

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

The new modified assay procedure was compared to the original assay procedure and the conventional culture methods.

b. *Matrix comparison:*

Not applicable

3. Clinical studies:

The new procedure was compared to the conventional culture method, and the original procedure. A total of 158 positive blood bottles (114 prospective + 41 seeded with clinical isolates) were tested at three sites. The seeded isolates included 20 *C. tropicalis*, five *C. glabrata*, six *C. krusei*, eight *C. lusitaniae*, one *C. utilis* and one *C. guilliermondii* at a concentration of 2.5×10^4 CFU/mL. The studies included two commercially available, continuously monitoring blood culture systems (i.e. BacT/ALERT and BACTEC).

Performance Data for Yeast Traffic Light PNA FISH (New) vs. Routine Identification Methods on Yeast-positive Blood Culture Bottles

Study	Sensitivity <i>C. albicans</i> / <i>C. parapsilosis</i>	Sensitivity <i>C. tropicalis</i>	Sensitivity <i>C. glabrata</i> / <i>C. krusei</i>	Specificity	Blood Culture System
A	100% (20/20) ¹	100% (1/1)	100% (4/4) ¹	100% (4/4)	BacT/Alert
B	100% (11/11)	100% (5/5)	100% (17/17)	100% (2/2)	BacT/Alert
C	100% (28/28) ²	100% (25/25)	93.3% (25/26) ^{2,3}	100% (15/15)	BACTEC
Total	100% (59/59) 95% CI (95.1-100)	100% (31/31) 95% CI (90.8-100)	97.9% (46/47) 95% CI (88.7-99.6)	100% (21/21) 95% CI (86.7-100)	N= 158

¹Includes 1 mixed culture: *C. albicans*/*C. glabrata*

²Includes 2 mixed cultures: *C. krusei*/*C. parapsilosis*

³One *C. glabrata* was missed by both Yeast Traffic Light PNA FISH (New) and Yeast Traffic Light (K080719) with the BACTEC PEDS Plus/F bottle

Performance Data for Yeast Traffic Light PNA FISH (New) vs. Yeast Traffic Light PNA FISH (K080719) on Yeast-positive Blood Culture Bottles (Clinical and Seeded with Clinical Isolates)

Study	Positive Agreement <i>C. albicans</i> / <i>C. parapsilosis</i>	Positive Agreement <i>C. tropicalis</i>	Positive Agreement <i>C. glabrata</i> / <i>C. krusei</i>	Negative Agreement	Blood Culture System
A	100% (20/20) ¹	100% (1/1)	100% (4/4) ¹	100% (4/4)	BacT/Alert
B	100% (11/11)	100% (5/5)	100% (17/17)	100% (2/2)	BacT/Alert
C	100% (28/28) ²	100% (25/25)	100% (25/25) ²	100% (16/16)	BACTEC
Total	100% (59/59) 95% CI (95.1-100)	100% CI (31/31) 95% CI (90.8-100)	100% (46/46) 95% CI (93.7-100)	100% (22/22) 95% (87.3-100)	N= 158

¹Includes 1 mixed culture: *C. albicans*/*C. glabrata*

²Includes 2 mixed cultures: *C. krusei*/*C. parapsilosis*

a. *Clinical Sensitivity:*

See sensitivity chart above

b. *Clinical specificity:*

See specificity chart above

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

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

C. albicans and *C. parapsilosis* cells: multiple bright green fluorescent cells in multiple fields

C. glabrata and *C. krusei* cells: multiple bright red fluorescent cells in multiple fields

C. tropicalis cells: multiple bright yellow fluorescent cells in multiple fields

The expected *C. albicans* + *C. parapsilosis*, *C. tropicalis*, and *C. glabrata* + *C. krusei* positive result rate from yeast positive blood culture bottles is approximately 50%, 9%, and 31%, respectively, based on AdvanDx clinical studies, but may vary depending on institution and patient population.

N. Proposed Labeling:

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

O. Conclusion:

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