### GERM TUBE TEST

### Principle

Germ tubes are produced by Candida albicans and Candida dubliniensis. The formation of germ tubes is associated with increased synthesis of protein and ribonucleic acid. The essential requirements for this synthesis are contained in tryptic soy broth and fetal bovine serum.

#### **Clinical Significance**

The germ tube test is used as a screening procedure to identify Candida albicans/dubliniensis.

#### Specimen

**Biosafety Level 2** 

• Any isolated yeast colony may be used.

#### Reagents

Remel's Germ Tube Solution

Prepared by rehydration of the vial with 25 ml sterile distilled water. Swirl to dissolve. Aliquots of 0.5 ml are placed in sterile 12 x 75 plastic capped tubes and frozen at -20°C. These are good for 4 months.

#### Instrumentation/Equipment

Pasteur pipet White light microscope (10X & 40X objectives) 12 x 75 mm slide & 22 x 22 mm Coverslip 35°C heating block

#### **Quality Control**

- Quality control is performed on each new lot # of Remel Tube Solution when reconstituted, aliquoted, and frozen.
- <u>Positive Control</u>: <u>Candida albicans ATCC 10231</u> Germ tubes formed.
- <u>Negative Control</u>: <u>Candida glabrata ATCC15126</u> No germ tubes formed.

## Procedure

- 1. Remove a frozen 0.5 ml aliquot of germ tube solution for each germ tube test. Allow to thaw and reach room temperature.
- 2. With a Pasteur pipet, lightly touch the surface of a yeast colony. A heavy inoculum will result in a decreased percentage of cells exhibiting germ tubes.
- 3. Place the inoculated pipet into one of the 0.5 ml aliquots of germ tube solution.
- 4. Incubate in a 35°C heating block for 4 hours.
- 5. After the incubation time, place one drop of the yeast-germ tube solution on a microscope slide and cover slip.
- 6. Examined under low and high powers for germ tube production.
  - **POSITIVE**: Germ tubes appear as thin filaments coming from the yeast cells which are not constricted at their point of origin on the yeast cells. (See Appendix 1.)
  - **NEGATIVE**: A negative germ tube test will show yeast cells only or pseudohyphae which have constrictions at the point of origin and along the filament. (See Appendix 1.)

# **Reporting Results**

- 1. Germ tube positive isolates report as: <u>Candida albicans, differentiation from Candida</u> <u>dubliniensis cannot be ruled out.</u>
- 2. Germ tube negative isolates report as: Yeast, probable not Candida albicans. If indicated or necessary, the yeast may be completely identified to genus and species using the API 20C.
- 3. Record results in Result Entry in LIS..

## Procedural Notes/Problem-Solving Tips

Some isolates of C. albicans will not produce germ tubes, but will identify as Candida albicans on the API 20C. If germ tube is negative and an API C is going to be performed, do not report "yeast, not Candida albicans. Please report "yeast to be identified".

#### References

- 1. *Remel Germ Tube Solution Technical Information*, TI No. 21066.
- 2. Balows, A.; Hausler, W. J.; Herrmann, K. L.: Isenberg, H. D.; Shadomy, H. J., *Manual of Clinical Microbiology*, 5th edition, 1991, pp. 620-621.

POLICY CREATION :					
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REVISION HISTORY					
Rev	Description of Change	Author	Effective Date		
1	Added revision history chart, reviewed and signed	T Smith	5-23-11		
2	Revised Negative Control material used.	B. Pestien	4/1/14		
3	Added dubliniensis	T. Nuese	4/12/16		
4	Changed read time to 4 hours. Changed reporting of negative germ tube when API is going to be performed	T Nuese	12/4/17		

#### Reviewed by

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