Comparison of Germ Tube Production By Candida Albicans In Various Media

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Abstracts: Introduction: Candidiasis is one of the important opportunistic fungal infections in human. Candida albicans (C.albicans) is the predominant Candida species isolated from clinical samples. In routine laboratory, C.albicans is diagnosed on the basis of culture, staining morphology, germ tube production, & chlamydospore formation on cornmeal agar. The objective of this study was to assess the reliability of different media for germ tube production. Material & Methods: The study was carried out on various clinical samples received in Microbiology department of M.P.Shah medical college, Jamnagar from January to July 2011. Among all clinical samples, 100 isolates of C.albicans were compared for germ tube production in 4 different media (sterile horse serum, pooled human serum, trypticase soy broth and Brain heart infusion (BHI) broth. Candida species were also identified by using culture, staining morphology, & chlamydospore formation on cornmeal agar. Results: In our study among 100 C.albicans isolates, sterile horse serum gave 100% germ tube production, pooled human serum gave 93%, BHI gave 63% and trypticase soy broth gave 60% germ tube formation at the end of 2 hours of incubation. Conclusion: This study shows that sterile horse serum is best medium for germ tube production of C.albicans and it can replace human serum which has its disadvantage in being bio hazardous with false negative reporting [Makawana G et al NJIRM 2012; 3(2) : 6-8] Key words: Candida Albicans, Germ tube production, Sterile horse serum, Pooled human serum

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Introduction: Candidiasis is the commonest fungal disease found in humans affecting mucosa, skin, nails, and internal organs of the body. It is also a common opportunistic infection in immunocompromised patients. The genus Candida contains more than 160 species but only 10 are regarded as important human pathogen. Among the candida species causing the human infection, the most pathogenic is C.albicans. It is the most pathogenic yeast isolated in clinical laboratories. Germ tube is a characteristic morphology observed in C.albicans and confirmation of germ tube is available as a rapid method for identification of C.albicans. Germ tube formation was first reported by Reynold and Braude in 1956. Germ tube formation is also produced by Candida dubliensis. In our study we have isolated C.albicans from various clinical samples and 100 such isolates were compared for germ tube production in four different media (Pooled human sera, sterile horse serum, BHI broth and Trypticase soy broth).

Material and Methods: In the present study we compared germ tube formation in four different media; Pooled human serum, sterile horse serum, BHI broth and Trypticase soy broth.

All the four media were taken in 4 different sterile test tubes and inoculated with light inoculums of yeast and incubated at 37°C. These results were read after 2 hours. Germ tube production is indicated as long tube like projection extending from the yeast cells (figure 1) & budding yeast cell with pseudohyphae (figure 2).
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Result: In the present study, out of 100 C.albicans isolates, germ tube production was seen in all 100 isolate in horse serum (100%), in 93 isolate in human serum (93%), in 63 isolate in BHI broth (63%), and in 60 isolate in Trypticase soy broth (60%). So in our study horse serum was found to be best giving 100% positive result for germ tube production followed by human serum which gave positive result in 93 % cases. BHI broth gave positive result in 63 % cases while it was positive in 60% cases in trypticase soy broth.

Discussion: Germ tube production is one of the important characteristic of C.albicans which is helpful for its rapid presumptive identification. A variety of media have been used for germ tube test. The medias used for germ tube test are pooled human serum, sterile horse serum, BHI broth, Trypticase soya broth, raw egg white, tissue culture media, peptone media, sheep serum, N-acetyl D-glucosamine, proline mixture of amino acids such as lee’s medium and modified Sabourad’s dextrose agar. As compared to other media serum is more sensitive for germ tube production.

Pooled human sera which is routinely used in diagnostic laboratory has effect of biological inhibitors present in it, so there may be chances of false negative result. It also has disadvantage of biohazard.

Sterile Horse serum which is commercially available has more sensitivity (100%) as compared to Pooled human sera used for germ tube production. There is no risk for biohazard in using sterile horse serum. Germ tube formation in animal serum has been reported to be very sensitive to cell concentrations greater than 10⁶ cells per ml² although Mackenzie³ reported that there is only a minor effect until the cell concentration is greater than 10⁷cells per ml. When human serum was stored at 4°C for 15days there was a 50% decrease in germ tube formation. This is in agreement with Taschdjian et al.⁴ who suggested that only freshly prepared or frozen human serum be used in germ tube test. Because of the time required to prepare human serum and inherent safety problems concerned with its use, many clinical laboratories have started to use non-human serum, germ tube media, with mixed results⁵. We report a new stable germ tube induction medium which was as effective as pooled human serum for the identification of C. albicans, without the extensive time required for the preparation and testing of pooled human serum and without the medical dangers associated with handling human serum⁶. In addition to human serum, a number of other mixtures induce germ tube formation, including plasma⁷, egg white⁸, saliva⁹, tissue culture medium 199 (Difco Laboratories, Detroit, Mich.)⁹, sheep serum¹⁰, Trypticase soy broth (BBL MicrobiologySystems, Cockeysville, Md.)⁹, and various peptone media can be used.

Conclusion: Thus our study showed horse serum as best medium for germ tube production of C.albicans and it can replace human serum which has its disadvantage in being bio hazardous & false negative reporting.

References:
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