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Evaluation of Routin Microbiological Media and a Selective Fungal Medium for Recovery of Yeast from Mixed Clinical Specimens

[1]Abbas Ali Jafari1, [2]Abdul Hossein Kazemi and [3]Hossein Zarrinfar

[1]Parasitology and Mycology Group, College of Medicine,
Yazd Medical University, Safaieh Bo Ali Street, Yazd, Iran
[2]Tabriz Medical University, Immunology Group, Tabriz, Iran
[3]Parasitology and Mycology Group, College of Medicine,
Yazd Medical University, Safaieh Bo Ali Street, Yazd, Iran

Abstract: The recovery of yeast from clinical specimens cultured on routine bacteriological media was compared with recovery on a selective fungal medium. Overgrowing of bacteria in mixed bacterial and yeast specimens was suppressing the yeast growth on bacteriological media. Totally 229 pus specimens used for evaluating of bacteriological and fungal selective media. The specimens were cultured on bacteriologic media (blood and chocolate agar) as well as on Sabouraud agar containing Chloramphenicol (50 mg lG[1]) to inhibit bacterial growth. Finally the yeast growth was reported semi-quantitatively as light, moderate, or heavy and the results analyzed using SPSS software. The number of specimens yielding yeast growth in bacterial and sabouraud agar was compared using Chi-square test. Using Sabouraud agar particularly in cases of mixed infections was very useful for recovering of yeast, because yeast was only recovered from 29.3% of 41 yeast-positive pus specimens (Chi-square = 7.74, P_{val} = 0.005) and also from 24% of 25yeast-positive throat specimens (Chi-square = 11.09, P_{val} = 0.00008) using bacteriologic cultures. Using selective fungal medium for culturing of specimens containing a mixture of bacteria and yeasts is very helpful and necessary for accurate detection of yeasts. Clinicians must request yeast isolation when clinical manifestatation of patient showed probable yeast infection and in this case the microbiological laboratory must add a selective fungal medium to isolate yeast etiological agents. In clinical studies of yeast infections, it is also important to use selective fungal cultures.

Key words: Pus specimen % yeast % bacteria % media

INTRODUCTION

In routine bacteriological media such as blood agar and chocolate agar plates, yeast can be grown, but in specimens, which yeast was mixed with bacterial populations, there is a possibility that bacteria will suppress the yeast growth. Therefore usually it is necessary to use a selective fungal medium such as Sabouraud agar. Microbiological laboratories usually use such media if the clinician requested isolation of fungal or yeast culture, but if such a request has not been made, fungal selective media are often omitted. It seems that Sabouraud dextrose agar is a common and suitable media for isolation of yeast and other fungi [1, 2].

Sabouraud agar is also a choice media for recovering of dermatophytes from skin swabs, which sent for bacterial culture. Gillian *et al*. showed that using sabouraud agar containing chloramphenicol (50 mg lG[1]) and cyclohxamid (500 mg lG[1]) is very useful for recovering of dermatophytes from skin swabs sent for bacterial culture [3]. Physicians usually requested recovering of fungi from hospitalized patients and usually they requested for fungal and bacterial isolation [4].

In order to show the important role of adding a selective fungal medium in such cases, we compared the efficiency of yeast recovery from both routine bacteriological and Sabouraud dextrose agar. Since in our study we only decided to evaluate the impact of selective fungal medium for isolating the yeast from clinical samples rather than the clinically importance of isolated yeast, we designed a simple study to evaluate and compare the isolation rate of fungal yeast from mixed samples in both microbiological and fungal selective medias.

Corresponding Author: Dr. Abbas Ali Jafari, Parasitology and Mycology Group, College of Medicine, Yazd Medical University, Safaieh Bo Ali Street, Yazd, Iran

Table 1: Comparison of bacteriological media and Sabouraud dextrose agar for the recovery of yeast from 41 yeast-positive pus specimens

Medium	Type of recovered bacteria	No. of specimens from which yeast were isolated	No. of specimens yielding yeast growth rate		
			Heavy	Moderate	Light
Bacteriological media	No bacterial growth	5	1	2	2
	Aerobic gram-negative rods	4	1	2	1
	Aerobic gram-positive cocci	3	1	0	2
Sabouraud agar only	No bacterial growth	14	8	3	3
	Aerobic gram-negative rods	9	3	5	1
	Aerobic gram-positive cocci	6	2	2	2
Total		41	16	14	11

Table 2: Comparison of bacteriological media and Sabouraud dextrose agar for the recovery of yeast from 25 yeast-positive throat specimens

Medium	Type of recovered bacteria	No. of specimens from which yeast were isolated	Growth rate		
			Heavy	Moderate	Light
Bacteriological media		6	2	2	2
Sabouraud agar only		19	8	6	5
Total		25	10	8	7

MATERIAL AND METHODS

In the current study totally 229 pus specimens received from patients' abscesses, wound and drain secretions, etc., used for cultivation onto bacteriological and fungal selective media. The specimens were cultured on blood agar, chocolate agar as well as on Sabouraud dextrose agar containing chloramphenicol (50 mg l^{-1}) to inhibit bacterial growth. All specimens were inoculated by one laboratory technician and incubated at 37°C for 48 h. Isolated yeast and bacteria were identified to genus and species level using macroscopic, microscopic and microbiologic analysis. Finally the yeast growth was reported semi-quantitatively as light, moderate, or heavy and results compared using SPSS software.

RESULTS

From 229 cultured specimens, yeast colonies were recovered in 41 specimens (18%) (Table 1) including 19 pure cultures (46.3%) isolated on all media, mostly on Sabouraud dextrose agar (73.7%). The other 22 yeast-positive specimens had mixed yeast and bacteria and for most of these specimens, yeasts were recovered from Sabouraud agar that means this medium to be most useful for isolation of yeast (Table 3). In specimens that had yeast with aerobic, gram negative bacteria, such as *Klebsiella* sp., *Enterobacter* sp., *Escherichia coli* or *Pseudomonas* sp., irrespective colonies of yeast was isolated. On the Sabouraud dextrose agar, only few rods like gram-negative bacteria grew that could be as a result

Table 3: Recovery of yeast from 41 yeast-positive pus specimens on bacteriological media and Sabouraud dextrose agar

Medium types	Culture results			
	Positive		Negative	
	Number	Percent	Number	Percent
Bacteriological medium	12	29.3	217	52
Sabouraud dextrose agar	29	70.7	200	48
Total	41	100	417	100

Table 4: Recovery of yeast from 25 yeast-positive throat specimens on bacteriological media and Sabouraud dextrose agar

Medium	Culture results			
	Positive		Negative	
	Number	Percent	Number	Percent
Bacteriological medium	6	24	26	66.7
Sabouraud dextrose agar	19	76	13	33.3
Total	25	100	39	100

of possible suppression of chloramphenicol and yeast on bacteria on some of these plates. There was seen a statistically significant differences between the numbers of specimens yielding yeast growth on sabouraud dextrose agar in compare with bacteriological media (Chi-square = 7.74, P_{val} = 0.005).

Also in current study 32 throat samples, which had mixed bacterial and yeast, were cultivated on blood,

chocolate agar plates and on Sabouraud agar. Yeast were isolated from 25 (78%) of the throat specimens (Table 2), of which most of them (76%) were recovered from Sabouraud agar only (Table 4). There was seen a statistically significant differences between the number of specimens from which yeast were recovered on Sabouraud dextrose agar in compare with bacteriologic medium (Chi-square = 11.09, P_{val} = 0.00008). The comparison of semi-quantitative yeast growth rates on bacteriologic and Sabouraud agar didn't show any statistical differences (P_{val} = 0.324).

From 37 yeast isolate recovered in this study, 25 isolates were identified to species level using assimilation and fermentation of sugar tests as follows: Candida albicans (19 isolates), Candida tropicalis (3 isolates), Candia krusei (2 isolates) and Saccharomyces cerevisiae (1 isolate).

DISCUSSION

However yeasts can be recovered from pure cultures on bacteriological media, but yeast growth may be suppressed in pus specimens that yeasts are frequently present with different bacteria. Also yeast can be inhibited the growth of the slow-growing bacteria in bacteriologic media. In our study we isolated yeast from 49% of 41 yeast-positive pus specimens and from 24% of 25 yeast-positive throat specimens containing mixed cultures.

Our results indicated that often yeast weren't isolated on bacteriological media in case of pus specimens, because yeast growth was suppressed by bacteria on these media. Overgrowing of yeast by bacteria and also bacterial toxins may kill yeast in those media [5, 6]. Therefore it is very important that the clinicians requested yeast recovery in clinical cases, which assumed for fungal infections as etiological agent with or without bacterial infections. There are many studies, which indicated that, the abdominal pus specimens taken from patients with intra-abdominal perforation [7, 8] and specimens collected from burned lesions have important yeast pathogens [9]. In such cases microbiological laboratory personnel must be employed a selective fungal medium like Sabouraud dextrose agar to isolated yeast species rather than bacterial agents. Sabouraud dextrose agar is a basic and useful media for recovering of opportunistic moulds and yeast. Jaimez *et al*. in one study for detection of yeast and aflatoxinogenic strains of aspergillus reported that there wasn't seen any statistical significant between Sabouraud agar and new culture modified mediums [10].

CONCLUSIONS

Results of current study indicate that using of selective media is very useful and necessary for recovering of yeasts from pus specimens and also for culturing of many mixed clinical specimens in microbiology laboratories.

REFERENCES

1. Merz, W.G. and G.D. Roberts, 1995. Detection and recovery of fungi from clinical specimens, Pfaller F.C. Tenover and R.H. Yolken (ed.). Manual of clinical microbiology, American Society for Microbiology, Washington, D.C., pp: 709-722.
2. Silva, J.O., S.A. Franceschini, M.A. Lavrador and R.C. Candido, 2004. Performance of selective and differential media in the primary isolation of yeasts from different biological samples. Mycopathologia, 157: 29-36.
3. Gillian, M., S. Jocelyn and M. Arthur, 2004. Clinical Benefit of Recovering Dermatophytes from Skin Swabs Sent for Bacterial Culture. J. Clin. Microbiol., 42: 4838-4839.
4. Arthur, J., L. Morris, C. Terry, L. Byrne and F.J. Madden, 1996. Duration of Incubation of Fungal Cultures. J. Clin. Microbiol., 34: 1583-1585.
5. Hockey, L.J., N.K. Fujita, T.R. Gibson, D. Rotrosen, J.Z. Montgomerie and J.R. Edwards, 1982. Detection of fungemia obscured by concomitant bacteremia: *in vitro* and *in vivo* studies. J. Clin. Microbiol., 16: 1080-1085.
6. Kerr, J.R., 1994. Suppression of fungal growth exhibited by Pseudomonas aeruginosa. J. Clin. Microbiol., 32: 525-527.
7. Montravers, P., R. Gauzit, C. Muller, J.P. Marmuse, A. Fichelle and J.M. Desmonts, 1996. Emergence of antibiotic-resistant bacteria in cases of peritonitis after intraabdominal surgery affects the efficacy of empirical antimicrobial therapy. Clin. Infect. Dis., 23: 486-494.
8. Pittet, D., M. Monod, P.M. Suter, E. Frenk and R. Auckenthaler, 1994. Candida colonisation and subsequent infections in critically ill surgical patients. Ann. Surg., 220: 751-758.

9. Slotman, G.J., E. Shapiro and S.M. Moffa, 1994. Fungal sepsis: multisided colonization versus fungemia. Am. Surg., 60: 107-113.

10. Jaimez, J., C.A. Fente, C.M. Franco, A. Cepeda and B.I. Vazquez, 2003. Application of a modified culture medium for the simultaneous counting of molds and yeasts and detection of aflatoxinogenic strains of Aspergillus flavus and Aspergillus parasiticus. J. Food Prot., 66: 311-318.