

COMPARISON OF MILK AGAR WITH CHOCOLATE AGAR FOR SAMPLING OF HOSPITAL AIR AND DOOR SWAB SAMPLES

Sayan Bhattacharyya^{*1}, Dhioendra Kumar², Shweta Singh³, Abhishek Sengupta⁴, Asim Sarfraz⁵, Nahid Anjum⁶, Ankur Kumar⁷ & Priyadarshini⁸

*¹Assistant Professor, Microbiology, AIIMS Patna

^{2,4,5&6}Senior Resident, Microbiology, AIIMS Patna

^{3,7&8}Tutor, Microbiology, AIIMS Patna

Abstract

Air sampling and hospital environmental sampling are routine procedures for hospital infection control measures and nosocomial infection surveillance. Samples were taken from general wards, outpatient departments and laboratories, thrice from each site. Chocolate agar made from human blood and milk agar were used. Milk agar was superior to chocolate agar in retrieving *S. aureus* and *Bacillus cereus*, which produced distinct golden yellow pigment and colonies with surrounding halo on it, respectively. Hence milk agar can be a good replacement for chocolate agar and other media for air and other hospital environmental sampling.

Keywords: Milk Agar, Chocolate Agar, Door Swab etc.

Introduction

Air sampling and hospital environmental sampling are routine procedures for hospital infection control practice and nosocomial infection surveillance protocols(1). Different methods are there in the literature for air sampling, like settle plate method, slit sampling and others(2). Settle plate method is a means of passive air sampling by which particles containing aerial bacteria deposit on surface of nutrient media at the rate of about 0.46 cm/second(3). Usually the 'complete particle' floating in air (microorganism in association with the 'carrier' particle) is 12 µm in diameter or more(4). The Settle plate method is very suitable for operation theatres air sampling because they do not create any turbulence. Chocolate agar handling may be hazardous and it can be difficult to make, and nutrient agar may be not as sensitive in recovery of colonies from air as compared to chocolate agar. Milk agar is a good enriched medium that enhances pigment production in Staphylococci and other bacteria. So our study was aimed at comparing milk agar with chocolate agar for settle plate air sampling and recovery of colonies from swabbing door of hospital wards and departments

Materials and methods

This was a laboratory based observational study carried out in the Department of Microbiology of the institute from August 2016 to december 2016. Samples were taken from general wards, 2 different outpatient departments and 3 laboratories, thrice from each site. Air was sampled by settle plate method while door swab sample was taken by sterile cotton swab moistened with saline. In settle plate method, chocolate agar and milk agar plates were kept open in the respective sites, 1 meter above floor and 1 metre way from walls. Air sampling and door swab inoculation was done on buffalo milk agar (composition: Nutrient agar 67 ml + buffalo milk 34 ml), sterilised by autoclaving at 110 deg C and 10 lbs/in² pressure for 10 minutes. Chocolate agar was prepared by adding 10 ml outdated human blood from blood bank to autoclaved nutrient agar (HiMedia, India; 90 ml) at 75 deg C temperature and subsequently pouring the plates. Plates were incubated at 37 deg C for 24 hours. Door swab samples were streaked on both these media, and sample was collected using sterile cotton swab stick moistened with sterile peptone water. Samples were taken thrice from each site. *Staphylococcus aureus* was identified by positive slide and tube coagulase tests from colonies showing Gram positive cocci in clusters by Gram stain(5). *Bacillus cereus* was identified by Gram positive bacilli with subterminal non-bulging spore on Gram stain from colonies, and positive motility on semi solid agar and positive lecithinase activity on Egg yolk agar, and positive mannitol fermentation reaction with no hemolysis on blood agar(6).

Results and discussion

Milk agar was superior to chocolate agar in retrieving *S. aureus* and *Bacillus cereus*, which produced distinct golden yellow pigment and dry, flat colonies with surrounding halo on it, respectively. Also it was better for growing filamentous fungi like *Aspergillus* spp., which formed cottony colonies at a faster rate on this medium as compared to chocolate agar. Gram negative bacilli produced translucent colonies on both media.

Number of colonies were same in both media on all occasions. Details are given in figures 1 and 2 (see bottom).

Conclusion

Milk agar can be a good replacement for chocolate agar and other media for air and other hospital environmental sampling. Gillespie in his book said that lactose milk agar can be a good medium for observing lactinase activity of bacteria as there was a zone of opalescence around the colonies on this medium in otherwise lecithinase positive bacteria(7). Milk agar has been used in routine clinical microbiology lab for a long time, mainly as litmus milk agar for characterisation of anaerobic bacteria(8). This is all the more important because handling chocolate agar made from human blood may be dangerous on account of fear of transmission of bloodborne infections, and can also lead to suboptimal colony recovery due to indiscriminate human consumption of antibiotics(9).

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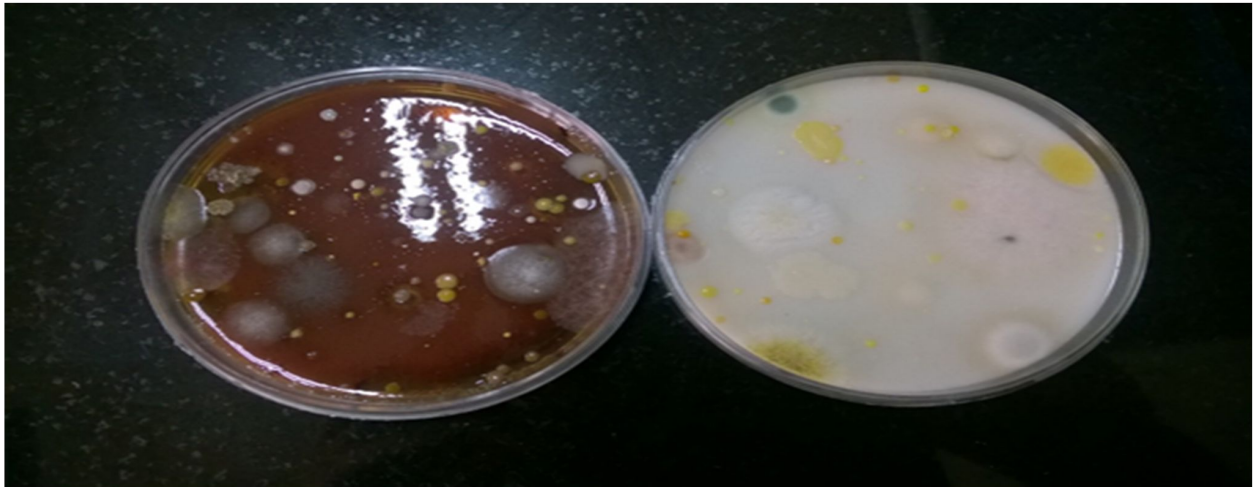


Fig. 1: Bacterial and Fungal colonies recovered from air by settle plate method using chocolate agar (left) and milk agar (right)



Fig. 2: Colonies from door swab using both media.