

# Detection of Methicilin Resistant (MECA) Gene from Staph Aureus by PCR

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## ABSTRACT

**Aims:** To detect *mecA* gene by polymerase chain reaction (PCR) and to demonstrate an efficient method for the detection of *mecA* gene of MRSA.

**Methodology:** In the present study, 60 MRSA strains on disc diffusion culture plates were collected from three laboratories from October, 2012 to January, 2013. The DNA of MRSA was extracted by phenol / chloroform extraction method and specimen were processed by the standard PCR technique.

**Results:** The *mecA* gene was detected in 31 strains from 60 MRSA isolates on disc diffusion. The prevalence of (Mutated?) MRSA strains among MRSA strains on disc diffusion culture plates isolates was 51.7%. These samples give 310 bp products on 1% agarose gel and others 29 samples gave no product.

**Conclusion:** This study shows that the PCR for *mecA* gene is an efficient method to detect the methicillin resistance *S. aureus* and *mecA* gene is necessary for the resistance in methicillin resistance *S. aureus*.

**Keywords:** MRSA, *mecA* gene, PCR

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## INTRODUCTION

Resistance is primarily mediated by the production of an altered penicillin-binding protein (PBP2a) which catalyses the transpeptidation reaction that cross-links the peptidoglycan of the bacterial cell wall. There are four penicillin-binding proteins: PBP1, PBP2, PBP3 and PBP4. PBPs have a high affinity for  $\beta$ -lactam antibiotics, which covalently bind to the PBPs and inhibit transpeptidation, preventing the terminal step in the peptidoglycan synthesis. MRSA differs genetically from Methicillin-Sensitive *S. aureus* (MSSA) by the presence of a chromosomal *mecA* gene (2007 bp) which encodes the 76 KDa PBP2a. PBP2a is unlike other constitutive PBPs, having a low affinity for  $\beta$ -lactam antibiotics. Infections caused by MRSA are increasing in prevalence in adults and children, and the detection of MRSA has important implications for the therapy and management of patients infected with this organism<sup>1</sup>.

The last decade has seen an alarming increase in MRSA infections in Pakistani hospitals. Pakistan's Armed Forces Institute of Pathology provides laboratory services to a 1,500-bed tertiary-care hospital in Rawalpindi and is the main reference

laboratory in northern Pakistan. According to that database, the frequency of MRSA among all nosocomial isolates of *S. aureus* increased from 39% (212/543) in 1996 to 51% (516/1,018) in 2003<sup>2</sup>.

## METHODOLOGY

The study was conducted in Lahore, the capital of Punjab province and the second largest city of Pakistan. Following laboratories are included: Chughtai Lahore Laboratory, SIMS laboratory, PGMI Laboratory. All culture plates positive for MRSA on disc diffusion were included and culture plates of MRSA having contamination were excluded from the study.

## RESULTS

The detail of results is given in tables 1 and 2

Table 1: Frequency of MRSA Positive & Negative Samples

	Frequency	%age
Positive	31	51.7
Negative	29	48.3
Total	60	100

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Table 2: Frequency of positive & negative samples from different labs

	Chughtais Lahore Lab		SIMS		PGMI	
	Frequency	%age	Frequency	%age	Frequency	%age
Positive	14	56	11	55	06	40
Negative	11	44	09	45	09	60
Total	25	100	20	100	15	100

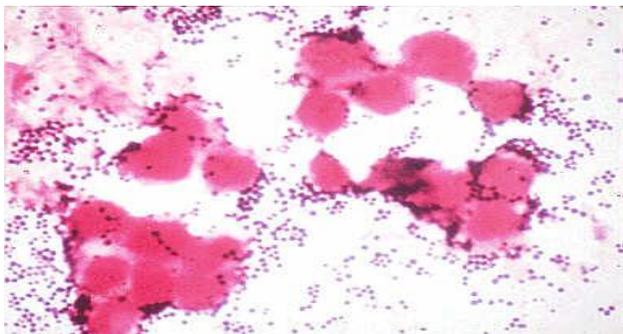


Fig 1: Gram stain of Staph. aureus

## DISCUSSION

Antimicrobial resistance results in increased morbidity, mortality, and costs of health care. The early and immediate detection of resistant microorganisms will reduce these adverse effects and their attendant costs. The *mecA* gene, which is responsible for this resistance, is often associated in-vitro with resistance to all Beta-lactams antibiotics. In the present study, *mecA* gene, by using the PCR based amplification technique was detected in 51.7% of total initially characterized methicillin resistant by oxacillin disc diffusion test.

Ozumba in 2005 studied the resistance of bacteria to antibiotics; particularly those used for first-line therapy and determined the prevalence of antibiotic resistance among common pathogens. Most of the *S. aureus* and coagulase negative staphylococci were resistant to common anti-staphylococcal drugs<sup>3</sup>.

In Pakistan, many studies have been conducted on the emergence of antimicrobial resistance in *S. aureus*. In this part of the review, Hafiz et al. (2002) determined the frequency of MRSA in major cities of Pakistan, Karachi, Peshawar.

Lahore, Sukhar, Islamabad, Quetta and Mirpur Azad Kashmir. Of total 792, *S. aureus* isolates from eight laboratories methicillin resistant *S. aureus* were 42% and no vancomycin resistant *S. aureus* was encountered<sup>4</sup>.

Anwar with his colleagues studied 714 *S. aureus* isolates from different clinical samples at Lahore from both community and hospital samples. It was observed that community acquired *S. aureus* were more among hospital samples. Methicillin resistance was high [31.7%] among the hospital strains<sup>5</sup>.

## CONCLUSION

PCR for *mecA* gene is an efficient method to detect the methicillin resistance *S. aureus* and *mecA* gene is necessary for the resistance in methicillin resistance *S. aureus*.

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