



MOLECULAR CHARACTERIZATION OF STAPHYLOCOCCUS AUREUS OBTAINED FROM BLOOD CULTURES

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Abstract:

Infections of the skin and soft tissues (SSTIs), pneumonia, bloodstream infections (BSI), and sepsis are only some of the illnesses that are caused by methicillin-resistant Staphylococcus aureus (MRSA), or MRSA. These bloodstream infections are linked to a large amount of patient morbidity and mortality all around the world. Isolates of MSSA included a greater number of virulence factors. Those MRSA isolates that had SCCmec-IV exhibited a wider diversity of virulence factors, and they were resistant to a greater number of antibiotics that were not beta-lactam types. The majority of S. aureus isolates belong to the CC5 strain.

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Introduction:

Infections that are obtained in hospitals and those that are acquired in the community are most commonly caused by *Staphylococcus aureus*, which belongs to the Gram-positive bacteria family. Significant infections, including bacteremia, sepsis, endocarditis, and osteomyelitis, can be brought on by *S. aureus*. These infections can range from moderate skin and soft tissue infections to severe infections. Resistance to antibiotics is one of the most significant obstacles that must be overcome in order to treat these diseases. The prevalence of MRSA is reported to range between 6 and 80 percent in Latin America, 52 to 57 percent in Mexico, 50 percent in China, and 0.9 to 26.8 percent in Europe [1,2]. Despite the fact that the global average prevalence of MRSA is forty percent, there are significant variations among various geographical places on a global scale. Molecular characterisation of *Staphylococcus aureus* has evolved into a method that may be utilized for the purpose of investigating and identifying clones that are circulating and epidemic in both the hospital and the community. Using SCCmec, MLST, CC, and PFGE, as well as the presence of virulence factors, including Pantón-Valentine leucocidin (PVL), these clones may be classified into four different types. It has been found that MRSA clones that have SCCmec elements I, II, and IV of ST5 and CC5 are related with HIV infections.

It is also important to note that the expression of virulence factors is connected to the pathogenicity of this particular bacteria (3). In point of fact, *Staphylococcus aureus* is capable of producing a wide range of heat-resistant extracellular protein toxins that operate as virulence factors. These toxins include superantigens, hemolyzerins, and leukocidins. The Pantón-Valentine leukocidin (PVL), which is a cytotoxin that produces holes in the membrane and has been linked to boils, skin abscesses, and severe necrotic skin infections [4], is the most significant of these factors. PVL is the most important of these factors. Some of the strains of *Staphylococcus aureus* that are considered to be pathogenic also include the gene that codes for staphylococcal toxic shock toxin (TSST-1). The antigenic specificity of these poisons is not a factor in their ability to activate polyclonal T-cells; rather, they possess superantigenic capabilities. There is a clear connection between this activation and the clinical manifestations of staphylococcal toxic shock. It has been demonstrated that the release of these toxins by strains that are known to be associated with illnesses poses a potential threat to extended durations of hospitalization [5].

Enzymes known as secreted glycerol ester hydrolase (lipase) may be produced in large quantities by *S. aureus*. These enzymes are responsible for the release of free fatty acids from triglycerides. In point of fact, many triglycerides include extra harmful fatty acids that appear to interfere with the proliferation of cells by modifying the permeability of the cell, preventing oxidative phosphorylation, or obstructing electron transport [6]. Due to the great genomic plasticity of *S. aureus*, the bacteria have an outstanding capacity to integrate exogenous genetic material from other strains, which enables them to gain new features such as antibiotic resistance and virulence [7]. This vast variety of harmful profiles is connected to the high genomic plasticity of *S. aureus*. *S. aureus* is a prevalent nosocomial infection among immune-compromised patients in hospital environments and health care institutions [7]. This is due to the fact that all of these conditions contribute to its prevalence.

Review:

The use of the molecular approach demonstrated a significant amount of relevance, since seven of the strains were not *S. aureus*. It is possible to say that the 16S-23S ribosomal RNA gene is a target of choice and that it is helpful for improved strain identification [8]. The existence of multiple copies of this gene in *S. aureus* strains is the cause of the genetic polymorphism (band size and number of bands) that was identified in our research. This polymorphism was connected to the 16S-23S rRNA gene. Research conducted by a number of writers has demonstrated that *S. aureus* is the species that possesses the greatest number of polymorphisms [8].

On the subject of the pathogenicity of *Staphylococcus aureus*, one of the tactics that host cells employ in order to react to an infection is the creation of fatty acids and lipids, which result in the formation of microscopic holes in the membrane of the bacterial cell. *S. aureus*, on the other hand, is capable of producing enzymes known as lipases, which are responsible for the destruction of these fatty acids prior to causing damage to the bacterial membrane [9]. It seems that 14.28% of the strains of *S. aureus* that we have are capable of producing lipase. One possible explanation for this outcome is that the detergents that are used for surface cleaning include a relatively low amount of lipids in their composition. Additionally, the stimulating impact of Ca²⁺ that was present in the medium in *S. aureus* is responsible for the generation of lipase in the test that was conducted with Tween 80 solution. According to the findings of one study [9], bivalent cations like calcium frequently boost the enzymatic

activity of lipase. This finding lends credence to the aforementioned assertion. Recent research carried out by other researchers has documented the synthesis of lipase by strains of *Staphylococcus aureus*, *Staphylococcus hycus* USA 300, and *Staphylococcus* sp. Lp12 [10]. Due to the fact that microbial lipases are extremely resistant to temperature, detergents, and certain proteolytic enzymes [10], this virulence factor should receive a greater amount of attention.

The phenotypic findings on the absence of PVL, ETA, and ETB toxins produced by our *S. aureus* strains provide some degree of reassurance. The same observations were found in *S. aureus* strains that were obtained from catheters at the CNHU of Cotonou [11]. These strains were isolated from the hospital environment in central Benin. On the other hand, a significant number of clinical strains isolate from bodily fluids are capable of producing PVL at a rate of up to fifty percent of the bacterium that is separated [11]. There is a possibility that this difference is related to the fact that strains that have been isolated from bodily fluids are frequently exposed to the immune system, which has resulted in the development of this virulence factor. Molecular PCR experiments, on the other hand, revealed that just one strain of *S. aureus* has the PVL gene, whereas three strains demonstrated the presence of the TSST-1 gene. In addition, the fact that these strains originated from the pediatric and maternity wards is cause for concern due to the close proximity of the two wards inside the hospital as well as the possibility of gene transfer [11].

According to the genetic features of *S. aureus*, it has been demonstrated that its clonal distribution might vary from one geographical location to another. It is not possible to obtain this level of differentiation only by the use of the PFGE approach; other mechanisms, such as MLST and CC typing, are required. According to the findings of a number of research, the most prevalent form of CC found in Asia and the Americas is CC5 [12]. The similar pattern was seen by us, especially considering that out of the 14 clones, fifty percent were classified as CC5 (69%), followed by CC8 (8.4%). It has been found that CC5 is more prevalent in MRSA that has SCCmec-II, while CC8 has been linked to MRSA-SCCmec-IV isolates that have PVL. This results in more severe clinical problems, such as fulminant pneumonia or deep vein thrombosis. In one study, 14.7% of the MRSA isolates and 18% of the MSSA isolates were unable to be classified into any CC. This was due to the fact that the method that was utilized could only identify the six most prevalent CCs that were found all over the world [13]. The MRSA epidemiology may be determined by a variety of approaches, including PFGE, MLST,

CC, spa typing, SCCmec, CC, and virulence factor detection. The information obtained from these methods can have an effect on the therapies that are administered to patients. In our collection, ST5 was one of the most common, which is consistent with what has been documented in other Latin American countries, such as Brazil (89%) and Guatemala (95%). However, it is distinct from what has been observed in Colombia (79%) and Ecuador (72%), where ST8 is more prevalent [13].

It is known that MRSA has evolved resistance to the majority of antibiotics that are used to successfully treat and manage its infections. However, it has been shown that CA-MRSA is resistant to a smaller number of drugs than HA-MRSA. Because of the presence of a wide range of virulence factors, CA-MRSA is typically considered to be more virulent than HA-MRSA [14]. It has also been found that increasing virulence in CA-MRSA is related with higher expression of a variety of genetic components. Panton-Valentine leukocidin (PVL), for example, is a significant cytotoxin that is released by *S. aureus*. PVL is encoded by *lukS-PV* and *lukF-PVG*, which are both essential mechanisms. In addition, PVL is present in the majority of CA-MRSA strains, while it is only sometimes discovered in HA-MRSA strains in Europe and the United States. However, the situation was different in certain parts of China [15]. The positivity rates for the staphylococcal enterotoxin (SE) genes, the arginine catabolic mobile gene (*arcA*), the leukocidin gene (*lukE*), the hemolysin genes (*hla*, *hly*, *hld*, *hlg*, and *hly2*), and the adhesion genes (*clfA*, *icaA*, *sdrC*, *sdrD*, and *sdrE*) were discovered to have a close relationship with the various types of MRSA isolates. In addition, it was shown that the genetic distribution of certain virulence genes, in particular those that produce enterotoxins, was connected to several MRSA lineages. On the other hand, it was discovered that CC1 and CC5 isolates included a greater number of enterotoxin genes than other CC isolates, which suggests that various CCs have distinct virulence profiles. On the other hand, the importance of these virulence factors in MRSA bacteremia has not been established with sufficient clarity [16].

Conclusion:

The identification of MRSA strains among food isolates need to be regarded as a possible threat to the healthcare of the general public. As a result, it is imperative that antibiotic prescriptions be regulated in order to utilize them in a sensible manner in the treatment of human and animal illnesses. CC 80-MRSA- IV was found to be a prominent pathogen among clinical isolates, as

demonstrated by the findings of the DNA microarray analysis that was performed in this investigation. A number of virulence factors, including lukF/lukS-PV, edinB, and sea, as well as antibiotic resistance genes, including sat, aphA3, far1, tetK, and blaZ, were discovered to be related with this particular clone. In addition, it was discovered that the majority of food isolates that were classified to CC1 and CC97 included genes that were resistant to antibiotics and genes that produced enterotoxins. It was also noticed that practically all clones were discovered in their normal host; nevertheless, it was fascinating to see that certain nasal carriers had isolates attributed to CC705 that were supposed to be missing in humans.

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