



Research Article

## Assessment of the Coagulase Test in the Identification of *Staphylococcus Aureus* Strains

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### Abstract

**Introduction:** *Staphylococcus aureus* are among the most important and several pathogens in human infections. The objectives of this study are to evaluate the efficacy of the coagulase test in the detection of

*Staphylococcus aureus* in the laboratory and to identify the factors associated with Staphylococcal infections.

**Methods:** It is a prospective study of 69 strains of Staphylococci in a period of six months from January

2019 to June 2019 in the University Hospital of Befelatanana.

**Results:** Among of the 69 strains of Staphylococci, 47 (68%) were represented by *Staphylococcus aureus*. Concerning the prediction performance of the catalase test, It has a sensitivity of 93.6%, a specificity of 100%, a positive predictive value (PPV) of 100% and a negative predictive value (NPV) of 88%. Concerning the associated factors, the men (71.1%) ( $p=0.46$ ; NS), the patients aged 60 and over (76.5%) ( $p=0.63$ ; NS), in resuscitation department (93.3%) ( $p=0.01$ ) and with suppuration (100%) ( $p=10^{-3}$ ) were the most affected by *Staphylococcus aureus*.

**Conclusion:** Coagulase test can replace the standard gold test for the detection of *Staphylococcus aureus*. In case of doubt, the bacteriological characteristics and the factors associated with Staphylococcal infections may be helpful in the diagnosis of *Staphylococcus aureus* infection.

**Keywords:** Coagulase; Baird-parker; *Staphylococcus aureus*; Suppuration

## 1. Introduction

Staphylococci are among the most important and severe pathogens in human infections. They are observed in multiple clinical situations, in community acquired infections and nosocomial infections [1]. Among staphylococci, *Staphylococcus aureus* is one of the major human pathogens in which it is responsible for toxic shocks, food borne diseases and especially a wide spectrum of suppurative infections [2]. The laboratory of medical biology plays a very important role in the diagnosis of infections, especially bacterial infections. Indeed, the laboratory highlights the infectious agent and can determine its sensitivity to antibiotics [3]. Of these most isolated bacteria, staphylococci including

*Staphylococcus aureus* are the most common [4]. There are several methods for identifying *Staphylococcus aureus* strains in the laboratory. In the laboratory of the University Hospital of Befelatanana, we used simultaneously the baird-parker culture medium and the coagulase test for the identification of *Staphylococcus aureus* strains. The identification on baird-parker culture medium represents the gold standard test [5-8]. But this method is more difficult compared to the coagulase test because it requires more consumables and reagents. On the other hand, the coagulase test is simple, easy to do and does not require a lot of reagents and consumables. In this study, we tried to compare the results of these 2 tests to see if the coagulase test is as reliable as the baird-parker test which is the gold standard test. If the coagulase test is reliable, we can stop doing the baird-parker test, which is difficult to perform. Thus, the aims of this study are to evaluate the efficacy of the coagulase test in the detection of *Staphylococcus aureus* in the laboratory and to identify the factors associated with Staphylococcal infections in order to improve the care of the patient.

## 2. Materials and Methods

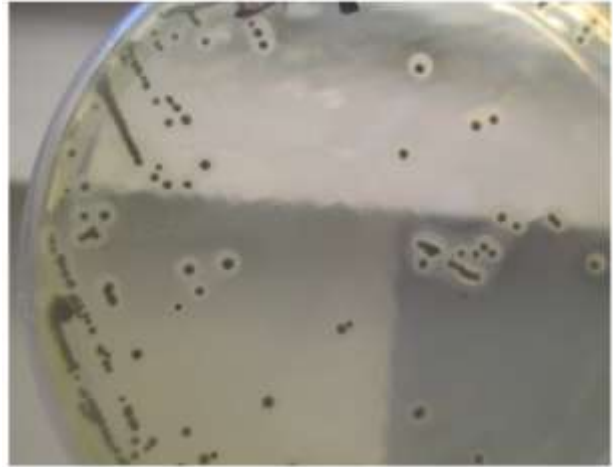
It is a prospective study of 69 strains of Staphylococci in a period of six months from January 2019 to June 2019 in the University Hospital of Befelatanana. In the beginning, the isolates were identified according to appearance, odor and color of bacterial colonies. Then, the identification of Gram-positive cocci on microscopic examination and the positivity of catalase confirms the diagnosis of Staphylococcal infection. For the diagnosis of *Staphylococcus aureus* infection, we performed 2 tests simultaneously. The first test is the inoculation of the strain on the baird-parker culture medium. The second is the coagulase test. Both tests require incubation at 37°C in an oven for 24 hours. The identification on baird-parker culture medium represents the gold standard test [5-8]. But this test requires several

conditions that make it more difficult to perform compared to the coagulase test. Indeed, a petri dish by strain is necessary for the test. In addition, we need to buy a box of baird-parker culture medium that is quite expensive. Finally, the egg yolk tellurite emulsion must be used, which will be mixed with the baird-parker agar during the preparation of the culture media. The egg yolk tellurite emulsion is necessary for the good growth of *Staphylococcus aureus* strains. Indeed, strains of Staphylococci will reduce tellurite that will become a black tellurium causing a black staining of staphylococci colonies. Similarly, there is proteolysis of egg yolk proteins, resulting in a clear (transparent) halo around the colony. The presence of this halo is specific to strains of *Staphylococcus aureus* (Figure 1 and 2). Concerning the coagulase test, we can simply put in a glass tube a few milliliters of citrated plasma and put some bacterial colonies therein. After 24 hours incubation in the oven, the positivity of the test shows the appearance of coagulum in the glass tube. This test shows that the bacterial strain present in the tube possessed an enzyme, coagulase, which is characteristic of strains of *Staphylococcus aureus*. Study parameters were age, gender, clinical information, hospitalization departments, baird-parker and coagulase test results. For ethical reasons, the authorization of the director of the establishment was obtained before the data were collected in the registers. The seizure was done anonymously to maintain confidentiality. The statistical significance threshold used was  $p=0.05$ .

### 3. Results

Among of the 69 strains of Staphylococci, 47 (68%) were represented by *Staphylococcus aureus* (Figure 3). The identification of these strains of *Staphylococcus aureus* was performed by the baird-parker test. Concerning the prediction performance of the catalase test, it has a sensitivity of 93.6%, a specificity of 100%,

a positive predictive value (PPV) of 100% and a negative predictive value (NPV) of 88% (Table 1). Concerning the associated factors, the men (71.1%) ( $p=0.46$ ), the patients aged 60 and over (76.5%) ( $p=0.63$ ), in resuscitation department (93.3%) ( $p=0.01$ ) and with suppuration (100%) ( $p=10^{-3}$ ) were the most affected by *Staphylococcus aureus* (Table 2).



**Figure 1:** Strains of *Staphylococcus aureus* in positive baird-parker test.



**Figure 2:** Strains of non-aureus *Staphylococcus* in negative baird-parker test.

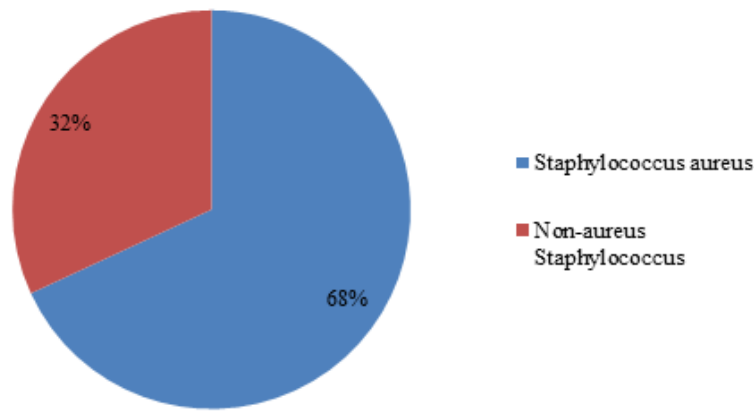


Figure 3: Distribution of staphylococcal strains.

	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Coagulase test	93.6	100	100	88

Table 1: Prediction performance of the catalase test.

Factors	Staphylococcus aureus		Non-aureus Staphylococcus		Total	p-value
	n	%	n	%		
<b>Age</b>						
<20 years	14	73.7	5	26.3	19	0.63
[20-39] years	10	62.5	6	37.5	16	
[40-59] years	10	58.8	7	41.2	17	
≥ 60 years	13	76.5	4	23.5	17	
<b>Gender</b>						
Female	15	62.5	9	37.5	24	0.46
Male	32	71.1	13	28.9	45	
<b>Department</b>						
Surgery	1	33.3	2	66.7	3	0.01
Out-patients	3	33.3	6	66.7	9	
Medecine	29	69.0	13	31.0	42	
Resuscitation	14	93.3	1	6.7	15	
<b>Clinical information</b>						
Others	5	71.4	2	28.6	7	10 <sup>-3</sup>
Infection	20	57.1	15	42.9	35	
Suppuration	20	100.0	-	-	20	
urinary disorders	2	28.6	5	71.4	7	

Table 2: factors associated with Staphylococcal infections.

#### 4. Discussion

In this study, strains of *Staphylococcus aureus* are more frequent than strains of *non-aureus Staphylococcus*. Indeed, the identification of *non-aureus Staphylococcus* is due in the majority of cases to skin contaminations during sampling [9]. Good sampling conditions eliminate the contaminating bacteria and identify the true bacteria that are responsible for the infection. The low prevalence of *Staphylococcus non-aureus* strains in our study shows that the samples were taken under good conditions.

Regarding the performance of the catalase test, this study showed a specificity and a positive predictive value of 100%. Thus, there is never a false positive in the identification of strains of *Staphylococcus aureus*. Similarly, the coagulase test will never make a diagnostic error. Thus, this result makes it possible to properly adjust the treatment corresponding to the germ and the patient is taken care of properly. On the other hand, the coagulase test is not 100% sensitive and the negative predictive value is lowered. This situation is probably due to the citrated plasma used for the coagulase test. In fact, we use citrated plasmas from patients hospitalized in the hospital or patients doing a health check. Thus, some citrated plasmas may contain few coagulation factors due to diseases presented by patients. And the result of the coagulase test can be falsely negative. Thus, we propose the use of citrated plasma of patients whose coagulation tests are normal (time of Quick and Activated Cephalin Time). The dosage of other coagulation factors should be made if possible. In short, in case of negativity of the coagulase test, we can compare the bacteriological result with the factors associated with the bacterial infection. And if in doubt, we can repeat the catalase test using another citrated plasma.

Concerning the associated factors, men and elderly subjects are most affected by *Staphylococcus aureus* infection without significant difference. Thus, *Staphylococcus aureus* infections can affect both genders without distinction. However, the high frequency of older subjects in this study may be due to their higher vulnerability and their greater immune deficiency [10]. This immunodeficiency will allow the germ to become more virulent resulting in the appearance of the infection. Concerning the department and the clinical information, patients hospitalized in the departments of resuscitation and presenting with suppurations are the most affected by *Staphylococcus aureus* with a very significant difference. These cases are mainly represented by patients with postoperative suppurations in the intensive care unit. Other studies have also found a high prevalence of *Staphylococcus aureus* infections in the suppurations [11-12]. A study carried out at the Niamey hospital found a prevalence of 31% of *Staphylococcus aureus* strains in surgical site infections [13]. Contamination of the surgical site most often occurs during the operating period, either caused by the patient's flora present before the incision, either by the flora of the staff or by the antiseptic solutions or the contaminated instruments [14].

The results of this study showed that the coagulase test can replace the baïrd-parker test which is more difficult and more expensive to perform. Even if the baïrd-parker remains the reference method for the identification of *Staphylococcus aureus* strains [5-8], all laboratory can use the coagulase test to identify these strains. And in case of doubt, technicians can verify the reliability of the results by comparing the bacteriological result with the factors associated with the patient's infection. Similarly, the coagulase test can be redone while respecting pre-analytical conditions.

## 5. Conclusion

This study highlighted the possibility of using only the coagulase test in the identification of *Staphylococcus aureus* strains. Indeed, this test showed a good result from the point of view of sensitivity and specificity compared to the bairst-parker test which is the reference method. Similarly, laboratory technicians can compare bacteriological findings with factors associated with the bacterial infection presented by the patient. The coagulase test is an easy-to-use test and does not require a lot of reagents and consumables. The use of the coagulase test will reduce expenditure in laboratories in developing countries and reduce the cost of bacteriological examination. Thus, the patient will be treated properly and his life expectancy will be improved.

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## Conflicts of Interest

The authors do not declare any conflict of interest.

## References

1. Zahlane K, Haouach K, Zouhdi M. Staphylococci: actual state of epidemiology and antibioresistance in University Hospital Center of Rabat. *Maroc Médical* 29 (2007): 279-285.
2. Donnio P-Y, Gaschet A, Cady A. Treatment news antibiotic of *Staphylococcus aureus* infection. *Médecine Tropicale* 16 (2010): 244-251.
3. Nounagnonhou M, Amadou PE. Problem of Enterobacteriaceae colonies with metallic

reflection on EMB agar. End of training memory for obtaining a professional degree. Polytechnic school of Abomey-Calavi (EPAC), University of Abomey-Calavi (UAC). Bénin (2008): 24.

4. Agon SIA, Olowo PP. Determination of species and antibiotic resistance profiles of strains of Staphylococci isolated from pathological samples at CNHU-HKM. End of training memory for obtaining a professional degree. Polytechnic school of Abomey-Calavi (EPAC), University of Abomey-Calavi (UAC). Bénin (2012): 53.
5. European Pharmacopoeia. Control of microbial contamination in non sterile products - Solution and recommended culture media. Council of Europe (2007).
6. The United States Pharmacopeia (USP 31)-NF 26. Microbial Limit Tests. United States Pharmacopeia Convention Inc. Rockville, MD. USA (2008).
7. ISO 6888-1 (V 08-014). October 1999. Microbiology of Foods. Horizontal method for the enumeration of coagulase positive staphylococci. (*Staphylococcus aureus* and other species). Part 1: Technique using the Baird-Parker agar medium (1999).
8. ISO 22718. September 2009. Cosmetics - Microbiology - Detection of *Staphylococcus aureus* (2009).
9. Zaine El Houssaini, Nadia Harrar, Khalid Zerouali, Houria Belabbes, Naima Elmdaghri. Prevalence of coagulase-negative staphylococci in blood cultures at the Ibn-Rochd University Hospital in Casablanca. *The Pan African Medical Journal* 33 (2019): 193.
10. Gilles Berrut Laure de Decker. Immunosenescence: a review. *Geriatr Psychol Neuropsychiatr Vieil* 13 (2015): 7-14.

11. Kanassoua KK, Kassegne I, Sakiye E, et al. Surgical site infections in general surgery in a regional hospital in Togo. *Revue de Cames Santé* 3 (2015): 50-54.
12. Abalo A, Walla A, Ayoubu G, et al. Surgical Site Infection in Orthopedic Surgery in a Developing Country. *Revue de Chirurgie Orthopédique Traumatologique* 96 (2010): 112-117.
13. Ousmane Abdoulaye, Mahaman Laouali Harouna Amadou, Oumarou Amadou, et al. Epidemiological and bacteriological features of surgical site infections (ISO) in the Division of Surgery at the Niamey National Hospital (HNN). *Pan African Medical Journal* 31 (2018): 33.
14. Faye-Kette H, Kouassi MY, Akoua-Koffi G, et al. Microbial Epidemiology of Surgical Site Infections (ISO) in Abidjan Trauma Department and Sensitivity of Germs to Antibiotics. *Revue Bio-Africa* 6 (2008): 25-31.



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