

Isolation and Identification of Staphylococcus Aureus from Al-Kut Hospital in Kut City

<https://doi.org/10.31185/wjps.VolX.IssX.XX>

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Abstract— Samples were collected from the hospital in 2019-2020, where the collection and diagnosis of samples about period from 2019/11 to the last diagnosis in the laboratory 2020/2/1, samples were harvested from patient rooms, waiting rooms and door handles. The samples were isolated with 100 samples and placed in special tubes to bacteriology laboratory. Isolation and diagnosis of bacteria include grow in different media like nutrient and Mannitol Salt agar and in selective Media like Baird Parker Agar with staining with gram stain to show bacteria shape and characteristic. The antibiotic's sensitivity to these bacteria was detected by observing and measuring the area of inhibition of bacterial growth with the presence of different types of antibiotics. The purpose of this research to know the amount of pathological bacteria present in the hospital and classify them for the purpose of avoiding the danger to human life, to control them and diagnose them in easy and quick ways, and to know their sensitivity against different types of antibiotics.

Keywords—Staphylococcus aureus, antibiotic is sensitivity, pathological bacteria

1 Introduction:

Staphylococcus aureus is one of the most clinically important species, It's a Gram positive bacteria, spherical cells are arranged irregularly similar to clusters, non-motile, aerobic electives [1], [2].

Although it represents 30% of the normal flora in the human body, it is present in different areas such as skin and respiratory tracts, but it is an opportunistic pathogen [3],[4],[5] .

Staphylococcus aureus accounted for nearly 40% of all pyogenic diseases recorded in medical centers worldwide, especially in hospitalized patients with weakened body defenses [6].

Purulent infections caused by S. aureus bacteria range in severity, including superficial skin diseases such as Impetigo, boils, eye injuries to systemic and deep life-threatening infections such as Septicemia as well as acute intestinal infections [7].

Pathogenesis is due to its ability to multiply rapidly, and has many virulence factors such as the production of toxins and enzymes that play an important role in the invasion of host tissues and the spread of bacteria [8].

Bacteria are characterized by the ability to make holes in the cell walls of cells, which facilitates the spread of bacteria in the tissues of the host and increases their pathogenicity by resist the host's immune system defenses, thus proving itself in host tissues, in addition to its ability to produce toxins and enzymes that play a role in infection and the appearance of disease symptoms and blood plasma coagulation [9].

Staphylococcus aureus is a natural germination of humans and some lactobacilli, it is also an opportunistic bacterium [10].

These bacteria enter the bloodstream and cause infections in the circulatory system [11].

It is also one of the most important bacterial species involved in hospital infection nosocomial infection, as reported by the national nosocomial Infection Surveillance to increase the incidence of these staphylococci to 70% [12].

Exposure of bacteria to more than one antibiotic contributes to reduce the rate of resistance and raise the level of sensitivity to antibiotics, especially when exposed to two antibiotics, one of which affects the construction of the cell wall and the other affects the protein structure [13].

The infection is described as nosocomial when the patient is hospitalized, There are two main types of infection: infection caused directly by a therapeutic procedure and those caused by a hospital environment [14].

These infections occur as a result of treatment procedures when microbes enter one of the sites of the body, as occurs during surgery where the germs are allowed to enter the tissues or organs in the patient's body, where it stabilizes and reproduces the infection [15].

Epidemiological studies have shown that hospital infections are more frequent in university hospitals than in other public and private hospitals [16].

To understand how hospital infections occur and identify effective ways to prevent them, we need to know the pathophysiology of these infections. An infection may be caused by a microorganism naturally carried by the gastrointestinal flora and oropharyngeal bacterial flora and is considered to be endogenous infection; It consists of a very large number of microbes that live with the human being without having a beneficial or harmful effect in their host. They may be found in the external environment such as water and soil [14].

2 Materials and Methods

-Several samples of *staphylococcus* bacteria were collected from various places, including hospitals, bedrooms and some nationals. After examination of these samples, these bacteria were detected using scientific methods used in laboratory tests. Most of the bacteria found were *staphylococcus aureus*, which is a pathogen of humans. Nutrient agar and blood agar base were used in primary isolation, and Mannitol salt agar in final isolation. The dishes were incubated at a temperature of 37 ° C for 24 hours

Another 24 hours before the result was considered negative, isolates were diagnosed based on phenotypic traits and biochemical tests [17].

- Cultural diagnosis

After taking samples from the patients at Al-kut Hospital and placing them in clean, dry and airtight tubes, they were isolated and many media were prepared, the agricultural characteristics of developing colonies were determined on the cultivated media including the MaCconkey agar, blood agar, nutrient agar, Baird Parker Agar and mannitol salt agar [17].

-Microscopy:

The samples were stain with gram stain and showed the microscopic examination to show the shape and gram stain [17].

-Biochemical tests

These tests were conducted according to Quinn et al., (2004), to detect the presence and diagnosis of bacteria *Staphylococcus aureus*, the most important tests like (catalase test, Oxidase test, Coagulase test) [17].

-Antibiotic susceptibility test

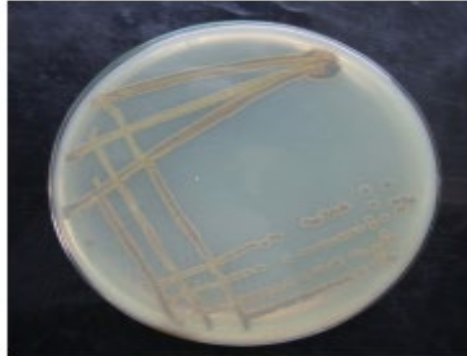
The isolation sensitivity was tested against ten antibiotics selected based on NCCLS (2000), including: Ciprofloxacin (CIP5), pyelonephritis(N10), (ATH15)azithromycin ,Oxytetracycline T30), Ceftriaxone (CRO30), Augmentin AUG30), Chloramphenicol (CM10), Amikacin(AK30). Antibiotics are prepared depending on Sambrook and his group (1989) in the center of Müller-Huntten. After MIC was determined for each antagonist, allergies were determined depending on NCCLS (2000)[18].

3 Results and Discussion

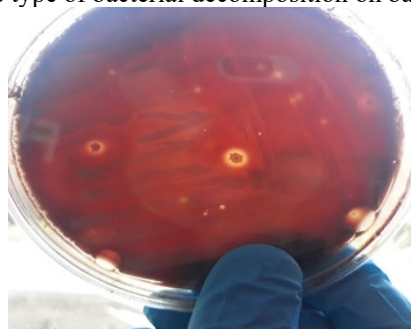
-Cultural characteristics

After collecting the samples, the bacteria were transplanted in several mediums, including nutrient agar, mannitol salt agar MaCconkey agar and Baird Parker Agar then incubated at 37 ° C for 24 hours. This bacterium is positive for gram stain and did not grow on the center of MaCconkey agar and in Baird parker agar Characteristic appearance of jet black colonies surrounded by a white halo were considered to be presumptive *S. aureus* [19].

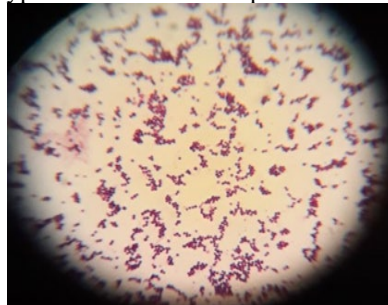
Morphological characteristics: The smear was prepared from the isolated culture on clean grease free microscopic glass slide and stained with Gram's method of staining, was observed under microscope, spherical cells arranged in irregular clusters resembling to bunch of grapes.



A picture showing the type of bacterial decomposition on baird parker agar



A picture showing the type of bacterial decomposition on the center of blood.



Microscopic exam show bacterial morphology.

-Biochemical identification

The results were confirmed by the Catalase test and the appearance of a positive result where the bacteria interacted with the added reagent H_2O_2 .

The virulence of the bacteria was examined by the secretion of the enzyme. Coagulase with blood plasma and positive result. The results are consistent with Quinn et al., 2004.

- Antibiotic susceptibility test

Bacterial isolates from clinical specimens for different places of patients' rooms and waiting rooms in Kut women's hospital. The results of phenotypic tests and biochemical tests confirmed the isolation of these bacteria

S. aureus. In the antibiotic sensitivity test, most of the isolates showed high resistance to Oxytetracycline and Ceftriaxone as shown in Table 1-3. This is consistent with [20].

The results of this study showed less sensitivity to the antibiotic pyelonephritis as shown in Table (1), and the reason is that the bacteria resistant to beta-lactam secretes beta-lactamase enzymes. The first is NCCLS (2000).

Table 1 shows the sensitivity of *S. aureus* to different antibiotics

Table 1. the sensitivity of S

Type of antibiotic	Bacterial hypersensitivity staphylococcus aureus
Ciprofloxacin (CIP5)	4 cm
(N10) pyelonephritis	2.5 cm
azithromycin (ATH15)	3.5 cm
T30 (Oxytetracycline)	4.5 cm
Ceftriaxone (CRO30)	4.5 cm
AUG30(Augmentin)	3.5 cm
CM10) (Chloramphenicol	4 cm
(AK30) Amikacin	3.5 cm



(3): Picture showing antibiotic sensitivity test on Muller Hinton agar medium.

4 Conclusion:

Samples were collected from the hospital in (2019-2020) where the process of collection of samples and diagnosis from 1/11/2019 to the last diagnosis in the laboratory 2/1/2020, after isolation and identification show 70 samples were negative and 30 sample were positive that mean many contamination in hospital room

The antibiotic sensitivity of these bacteria was detected by observing and measuring the inhibition of bacterial growth in the presence of different types of antibiotics.

The purpose of writing the research: To know the amount of pathogenic bacteria in the hospital and classification for the purpose of avoiding its danger to human life. Methods of control and diagnosis in easy and quick ways.

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Article submitted 16 December 2021. Published as resubmitted by the authors 12 January 2022.