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DETECTION OF BACTERIAL URINARY TRACT INFECTIONS IN PREGNANT WOMEN WITH GESTATIONAL DIABETES AND STUDYING THE ROLE OF SOME VIRULENCE FACTORS OF S. AUREUS IN THOSE INFECTIONS

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Abstract:

Background: UTIs are prevalent among a significant proportion of the population. At some stage in their life, over half of all women will have a urinary tract infection (UTI); one-third of these instances require antibiotic therapy.

Aim: The present study aimed to evaluate the prevalence of UTIs among pregnant women with and without gestational diabetes and to study different virulence factors of Staphylococcus aureus involved in their pathogenicity in UTIs.

Materials and methods: This cross-sectional study enrolled 292 pregnant women aged 17 to 45 at a gynecological and pediatric hospital in Kirkuk City. Urine samples were collected midstream, clean-catch. Urine samples underwent 24-hour culture on blood agar and MacConkey agar, followed by microscopic examination and various biochemical tests to identify bacteria. Sensitivity testing using antibiotic discs and subsequent subcultures facilitated the isolation of pure microbial cultures. Bacterial identification relies on colour, shape, size, and edge characteristics. Additional cultures were conducted to assess virulence factors, including biofilm formation. Gestational age determination for each participant was based on the first day of their last menstruation, further verified by prior medical records and routine ultrasonographic measurements. Comprehensive medical information was extracted from patient files for a thorough and meticulous analysis. A modified approach for screening biofilm formation by Staphylococci involved using a specialised medium consisting of Congo red.

Results: Among the total of 292 pregnant women included in the study, 91 individuals (31.16%) were diagnosed with gestational diabetes, while the majority, comprising 201 individuals (68.84%), did not exhibit this condition. Out of the 108 pregnant women who were diagnosed with positive urine culture, 78 (72.22%) had gestational diabetes, and 30 (27.78%) had gestational diabetes. Of the 184 pregnant women who have negative urine cultures, on the other hand, 13 (7.07%) had gestational diabetes, and 171 (92.93%) did not have gestational

diabetes. The study demonstrated that Staphylococcus aureus was the most prevalent, accounting for 25.64% of the isolates from GDM pregnant women, followed closely by Escherichia coli at 24.36%. Enterococcus faecalis constituted 17.95%, Klebsiella pneumonia and Staphylococcus saprophyticus represented 16.67% and 5.13%, respectively. Pseudomonas aeruginosa and Proteus mirabilis comprised 3.85%, and Enterobacter cloaca accounted for 2.56%. The study also showed that all isolates exhibit 100% positivity for coagulase, urine, Sinophobe, and gelatinase production, underscoring their potential for pathogenicity. In conclusion, these findings contribute to understanding the interplay between gestational diabetes, urinary health, and bacterial infections. Further research is warranted to address the discrepancies and inform targeted interventions for this vulnerable population.

Keywords: UTI, pregnancy, gestational diabetes, virulence factors, S. aureus detection.

Introduction

Urinary tract infections are widespread in the general population, with an estimated 50% of women encountering at least one occurrence in their lifetime, prompting the need for antibiotic intervention in roughly 30% of cases (1). Simultaneously, gestational diabetes mellitus (GDM), identified during pregnancy, is characterized by impaired carbohydrate metabolism, posing significant health risks for both the mother and the developing fetus (1). The ramifications extend to pregnancy-related complications, including dorsal dystocia, delivery traumas, newborn hyperbilirubinemia, hypoglycemia, and respiratory distress syndrome, potentially necessitating a caesarean section and elevating the risk of pre-eclampsia, marked by elevated blood pressure, fluid retention, and albuminuria (2). A pivotal risk factor for the future onset of type 2 diabetes mellitus (T2DM) is hyperglycemia during pregnancy, commonly known as gestational diabetes mellitus (GDM) (3). GDM affects around 5% of pregnancies, particularly prevalent in high-risk groups (4). The heightened susceptibility in women can be attributed to anatomical factors, such as shorter and wider urethras, facilitating the movement of microorganisms and making them more prone to urinary tract infections, one of the most prevalent ailments across all age groups (3). Infections of the urinary system can manifest in either the upper or lower urinary tract, with cystitis representing a syndrome characterized by painful urination, frequency, and occasional pain above the pubic area (4). Over 95% of UTIs result from a single bacterial species, with Escherichia coli being the predominant causative agent during acute infections (5). Various bacteria contribute to UTIs, including coagulase-negative staphylococci, Enterococcus faecalis, and specific Gram-positive cocci (CoNS). Furthermore, Gram-negative microbes like Klebsiella species, Proteus species, Pseudomonas aeruginosa, and Enterobacter species are implicated in UTIs (6). There must be at least 10^5 Colony Forming Units (CFU) per milliliter of pathogenic organism in the urine for it to be considered a urinary tract infection. (7,8). An infection caused by S. aureus is caused by a mix of toxins and adhesion factors. Urinary tract infections (UTIs) are one of the most common infections found in both outpatients and hospitalized patients. One of the things that can cause

UTIs is S. aureus (9). Not only does S. aureus create coagulase, but it also produces additional hemolysins and other virulence factors. The present study aimed to evaluate the prevalence of UTI among pregnant women with and without gestational diabetes and to study different virulence factors of Staphylococcus aureus involved in its pathogenicity in UTI.

Materials and Methods

In this cross-sectional study from April to September 2023, 292 pregnant women aged 17 to 45 were enrolled in a gynaecological and pediatric hospital in Kirkuk City. Mid-stream, clean-catch urine samples were meticulously collected in sterile containers. To ensure the reliability of culture results, participants refrained from antibiotics 72 hours before sample collection. The collected specimens were swiftly delivered to the laboratory within an hour; any delayed samples were refrigerated at 4°C and analysed within 6 hours. Urine samples underwent a 24-hour culture on both blood agar and MacConkey agar. Subsequently, microscopic examination and various biochemical tests were employed to identify the bacteria isolated from the cultures. Sensitivity testing using antibiotic discs and subsequent subcultures facilitated the isolation of pure microbial cultures. Bacterial identification relies on colour, shape, size, and edge characteristics. Additional cultures were conducted to assess virulence factors, including biofilm formation. All procedures strictly adhered to the manufacturer's instructions and the standard operating procedures of microbiology laboratories.

The first day of her last period was used to find out how far along a woman was in her pregnancy. Medicine records and regular ultrasounds showed this to be true. Many medical data from patient files was taken out to be looked at carefully and completely. This told them how old the mother was, how many kids she had, how many abortions she had, and her medical and reproductive history.

A different way to test for biofilm formation by Staphylococci bacteria used a special medium of 0.8g of Congo red, 20g of agar, 50g of sucrose, and 1000ml of brain heart infusion broth. Congo red was made as a concentrated water solution and autoclaved at 131°C for 15 minutes without the other parts of the medium. Once it got down to 55°C, it was added. After adding the cells, the plates were incubated aerobically at 37°C for 24 to 48 hours. Hi Media in Mumbai was used to get the chemicals for the medium.

Positive results were shown by black colonies that were dry and crystalline, while pink colonies meant that weak slime production was happening. The darkening of colonies that didn't have a dry, crystalline shape pointed to a result in the middle. For reliability's sake, the experiment was done three times, each time in duplicate. As controls, S. epidermidis ATCC 35984 (which makes a lot of slime) and S. epidermidis ATCC 12228 were used (non-slime producer).

Four or five colonies were taken from overnight growth on blood agar and put into 5ml of nutrient broth for the inoculation step. The turbidity was measured using the McFarland 0.5 standard (1.5 x 108 cfu/ml of bacteria). This was done after incubating at 37°C for 2 to 6 hours. After that, 0.01 ml of the standardized suspension was added to each tube, making a final inoculum of about 106 cfu/ml. A growth control was used for each test, and a small amount of the inoculum was plated to check its purity and density. As a control, S. aureus ATCC 25923

was used, and all the tubes were kept at 37°C for 18 to 24 hours. This method gave a strong and controlled way to check for biofilm formation by Staphylococci.

Statistical Analysis

The IBM SPSS ver 23.1 statistical program was used to do the computerized statistical analysis. Chi-square (X2) and T-Test were used to compare things. As long as the P value was less than 0.05, it was statistically significant. If it was greater than 0.05, it was not statistically significant.
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Results

Table 1 summarizes key clinical characteristics of 168 pregnant women. Maternal age averaged 33.4 years, gestational age 32.4 weeks, and parity 2.2.

Table 1: Clinical features of studied women

Parameters	**Pregnant Women (n:292)**
Maternal Age (years)	**33.4 ± 4.14**
Gestational Age (weeks)	**32.4 ± 4.33**
Parity	**2.2 ± 1.33**

Table 2 shows how the pregnant women are spread out based on whether they have gestational diabetes or not. Out of the 292 pregnant women who were part of the study, 91 (31.16%) were diagnosed with gestational diabetes. The other 201 (68.84%) did not have this condition.

Table 2: Distribution of pregnant women according to gestational diabetes

Gestational diabetes	No.	%
Present	91	31.16
Absent	201	68.84
Total	292	100

Out of the 108 pregnant women who were diagnosed with positive urine culture, 78 (72.22%) had gestational diabetes and 30 (27.78%) haven't gestational diabetes. For the 184 pregnant women who have negative urine cultures, on the other hand, 13 (7.07%) had gestational diabetes and 171 (92.93%) did not have gestational diabetes. It's important to note that the reported p-value (P<0.001) shows a very strong link between gestational diabetes and the results of the urine culture.

Table 3: Distribution of pregnant women according to Urine culture result of gestational diabetes

Pregnant women	Positive urine culture		Negative urine culture	
	No.	%	No.	%
With Gestational diabetes (n:91)	78	72.22	13	7.07
Without Gestational diabetes (n:201)	30	27.78	171	92.93
Total	108	100.	184	100

P<0.001

Table 4 shows the breakdown of pregnant women who were diagnosed with gestational diabetes mellitus (GDM) based on urine culture results and the number of trimesters they were in. The data shows a strong link between these factors, since the number of positive urine cultures in pregnant women with GDM changes from trimester to trimester. In the first trimester, 12.82 percent of cultures were positive. This number rose to 32.05 percent in the second trimester and reached a high point of 55.13 percent in the third trimester. On the other hand, the percentage of urine cultures that are negative changes, which shows that GDM, urinary health, and pregnancy progression are all connected in a dynamic way.

Table 4: Distribution of GDM pregnant women according to Urine culture result and trimester of pregnancy

Trimester of pregnancy	Pregnant women with gestational diabetes			
	Positive urine culture		Negative urine culture	
	No.	%	No.	%
1st Trimester	10	12.82	3	23.08
2nd Trimester	25	32.05	6	46.15
3rd Trimester	43	55.13	4	30.77
Total	78	100	13	100

P<0.001

The study showed that Staphylococcus aureus was the most common type of bacteria found in GDM pregnant women, making up 25.64 percent of the isolates. Escherichia coli came in second, making up 24.36 percent. A lot of the bacteria that were found were enterococcus faecalis (17.95%), Klebsiella pneumonia (16.67%), and Staphylococcus saprophyticus (5.13%). 3.85 percent was made up of Pseudomonas aeruginosa and Proteus mirabilis, and 2.56 percent was made up of Enterobacter cloacae. .

Table 5: Distribution of bacterial isolates causing UTI among GDM pregnant women

Bacteria	No.	%
S. aureus	20	25.64
E. coli	19	24.36
E. faecalis	14	17.95
K. pneumonia	13	16.67
S. saprophyticus	4	5.13
P. aeruginosa	3	3.85
Proteus mirabilis	3	3.85
Enterobacter cloaca	2	2.56
Total	78	100

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Figure 1: Growth of *Klebsiella pneumoniae* isolates on Chromogenic agar

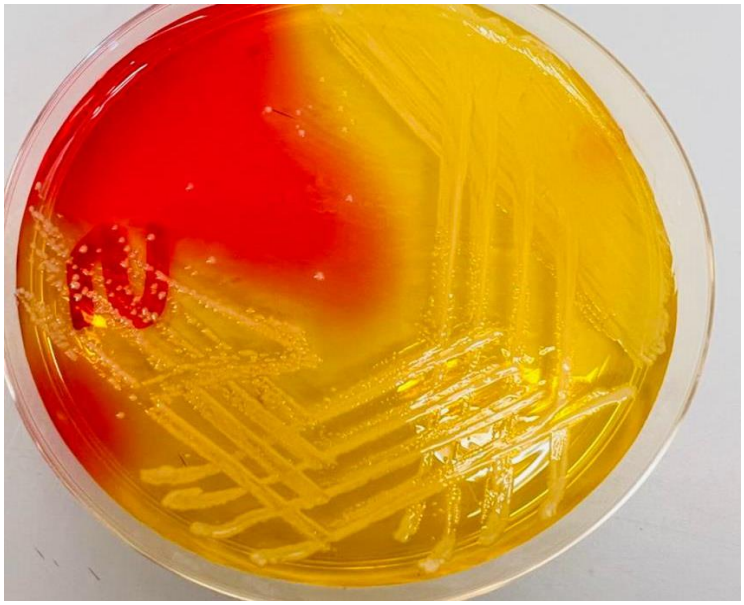

Figure 2: Growth of S. aureus on mannitol salt agar.

In the study, urinary tract infections (UTIs) in GDM pregnant women were described in terms of their symptoms, which can be seen in Table 6. When asked if they felt pain or discomfort while urinating, 76.92 percent said yes, while 23.08 percent said they didn't. Half of the people who were questioned said they didn't have back pain, while the other half said they did. 7.69 percent of people said they had recurrent UTIs, but 92.31% said they did not have recurrent episodes.

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Table 6: The study revealed the clinical features of UTIs among the GDM pregnant women

Clinical features of UTI		No.	%
Discomfort or pain urine	Yes	60	76.92
	No	18	23.08
	Total	78	100
Back pain	yes	39	50
	no	39	50
	Total	78	100
Recurrent UTI	yes	6	7.69
	no	72	92.31
	Total	78	100

Due to its persistent and strong resistance (up to 100 percent) against Gentamicin, Trimethoprim, Levofloxacin, Ampicillin, Ceftriaxone, and Cefixime, the only antibiotic that can penetrate S. saprophyticus is Imipenem. On the other hand, S. aureus reacts differently; 85% of it is resistant to ampicillin and 75% to gentamicin. Gentamicin and ampicillin are two of the many medications to which Proteus mirabilis has developed a remarkable resistance. All strains of Pseudomonas aeruginosa are resistant to antibiotics. It utterly evades several antibiotics, including ciprofloxacin, ceftriaxone, cefotaxime, ampicillin, and trimethoprim. However, this infection is well treated with imipenem and levofloxacin. K. pneumoniae does not respond to other antibiotics. The resistance profile of certain antibiotics poses a challenge. Ciprofloxacin, amoxiclav, levofloxacin, ampicillin, amikacin, ceftriaxone, cefotaxime, cefixime, and trimethoprim prove ineffective against the target. However, imipenem stands as a notable exception to this resistance trend. E. cloaca commonly exhibits resistance to ciprofloxacin, amoxiclav, trimethoprim, levofloxacin, ampicillin, amikacin, ceftriaxone, cefotaxime, cefixime, and ciprofloxacin. Conversely, ampicillin and imipenem demonstrate efficacy against this strain. In contrast, E. faecalis displays high resistance to Ampicillin, Trimethoprim, and Cefotaxime. Caution in antibiotic selection is crucial. Moreover, strategic consideration is essential as E. coli increasingly resists Ampicillin, Trimethoprim, and Cefotaxime

Table 7: Antibiotics profile of bacteria isolated from GDM pregnant women with UTI

Antibiotics		***S. saprophyticus***	***S. aureus***	***Proteus mirabilis***	***P. aeruginosa***	***K. pneumonia***	***E. cloaca***	***E. faecalis***	***E. coli***
Gentamicin	Resistant	4(100%)	15(75%)	3(100%)	3(100%)	9(69.23%)	2(100%)	11(78.57%)	17(89.47%)
	Sensitive	0(0%)	5(25%)	0(0%)	0(0%)	4(30.77%)	0(0%)	3(21.43%)	2(10.53%)
Trimethoprim	Resistant	4(100%)	19(95%)	0(0%)	3(100%)	11(84.62%)	1(50%)	14(100%)	11(57.89%)
	Sensitive	0(0%)	1(5%)	3(3%)	0(0%)	2(15.38%)	1(50%)	0(0%)	8(42.11%)
Amoxiclav	Resistant	3(75%)	13(65%)	3(100%)	2(66.67%)	9(69.23%)	2(100%)	10(71.43%)	8(42.11%)
	Sensitive	2(25%)	16(35%)	0(0%)	2(33.33%)	7(30.77%)	0(0.0%)	8(28.57%)	9(57.89%)
Levofloxacin	Resistant	4(100%)	11(55%)	3(100%)	0(0%)	2(15.38%)	0(0%)	11(78.57%)	10(52.63%)
	Sensitive	0(0%)	9(45%)	0(0%)	3(1%)	11(80.77%)	2(1%)	3(21.43%)	9(40.53%)

Ampicillin	Resistant	0(0%)	17(85%)	3(100%)	3(100%)	11(84.62%)	0(0%)	14(100%)	12(63.16%)
	Sensitive	4(100%)	3(15%)	0(0%)	0(0%)	2(15.38%)	2(100%)	0(0%)	7(36.84%)
Amikacin	Resistant	3(75%)	14(70%)	1(33.33%)	2(66.67%)	5(38.46%)	0(0%)	10(71.43%)	8(42.11%)
	Sensitive	1(25%)	6(30%)	2(66.67%)	1(33.33%)	8(61.54%)	2(100%)	4(28.57%)	11(57.89%)
imipenem	Resistant	0(0%)	7(35%)	2(66.67%)	1(33.33%)	1(7.69%)	0(0%)	6(42.86%)	7(36.84%)
	Sensitive	4(100%)	13(65%)	1(33.33%)	2(66.67%)	12(92.31%)	2(100%)	8(57.14%)	12(63.16%)
Ceftriaxone	Resistant	1(100%)	17(85%)	3(100%)	0(0%)	9(69.23%)	2(100%)	11(78.57%)	9(47.37%)
	Sensitive	0(0%)	3(15%)	0(0%)	3(100%)	4(30.77%)	0(0%)	3(21.43%)	10(52.63%)
Cefotaxime	Resistant	3(75%)	18(90%)	1(33.33%)	3(100%)	9(69.23%)	2(100%)	13(92.86%)	11(57.89%)
	Sensitive	1(25%)	2(10%)	2(66.67%)	0(0%)	4(30.77%)	0(0%)	1(7.14%)	8(42.11%)
Cefixime	Resistant	4(100%)	15(75%)	0(0%)	3(100%)	7(53.85%)	2(100%)	12(85.71%)	7(36.84%)
	Sensitive	0(0%)	5(25%)	3(100%)	0(0%)	6(46.15%)	0(0%)	2(14.29%)	12(63.16%)
Ciprofloxacin	Resistant	2(50%)	16(80%)	0(0%)	2(66.67%)	7(53.85%)	0(0%)	8(57.14%)	9(47.37%)
	Sensitive	2(50%)	4(20%)	3(100%)	1(33.33%)	6(46.15%)	2(100%)	6(42.86%)	10(52.63%)

Based on the investigation, it was shown that all of the isolates produce the same amounts of potentially hazardous enzymes: coagulase, uriease, Sinophobe, and gelatinase. In addition to beta-hemolysis, capsule synthesis, DNase production, and gelatinase production are all present in significant concentrations in the majority of the isolates (100 percent). that is 100%. But the percentages of Beta-lactamase and lipase production vary throughout isolates, suggesting that these bacteria may be resistant to antibiotics and capable of tissue injury. The percentages of other virulence factors, like lecithin and protease synthesis, also vary (76.47 percent and 47.06 percent , respectively) .

Table 7: Pathogen virulence factor distribution in Staphylococcus aureus samples taken from pregnant women experiencing urinary tract infections.

Virulence	Positive		Negative	
	No.	%	No.	%
Coagulase	17	100	0	0
Beta-hemolysis	15	88.24	2	11.76
Capsule production	15	88.24	2	11.76
Urease	17	100	0	0
Sinophobe	15	88.24	2	11.76
Beta-lactamase production	14	82.35	3	0
DNase production	17	100	0	0
Invasiveness (Growth on Congo red Agar)	14	82.35	3	0
Lecithin’s production	13	76.47	4	0
Lipase production	14	82.35	3	0
Gelatinase production	17	100.00	0	0
Protease production	8	47.06	9	52.94

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Figure 3: Biofilm formation on Congo red agar for S. aureus

Discussion

Complications from urinary tract infections (UTIs) can be harmful to the mother and the baby, thus pregnant women with gestational diabetes should take special precautions to avoid them (1). The risk of urinary tract infections in diabetics has long been believed to be higher (2). Out of the 292 pregnant women who were part of the study, 91 (31.16%) were diagnosed with gestational diabetes. The other 201 (68.84%) did not have this condition. This study clarified the importance of the link between GDM and the frequency of UTIs in pregnant women, which is supported by another study that found married women with GDM are more likely to have UTIs (5). Different parts of the world were found to have the highest rates of infection (7-11). One reason for the difference in the prevalence could be the change in nutritional status and socioeconomic status. The antibiotic used depends on the types of uropathogens that it can kill, how well it works for this purpose, and any side effects it might cause (2,3). Taking into account the stage of pregnancy, this study found that most of the pregnant women who had a positive UTI were in their third trimester. This was in line with another study's findings that the percentage of infections rose as the pregnancy went on (12). Another study from Oluyoro Catholic Hospital in Ibadan, South-Western Nigeria, found that UTI is still a big health issue for pregnant women, especially in the third trimester (13). The most common type of bacteria found in this study was Staphylococcus aureus, which made up 25.64 percent of the isolates from GDM pregnant women. Escherichia coli came in second, with 24.36 percent. A lot of the bacteria that were found were enterococcus faecalis (17.95%), Klebsiella pneumonia (16.67%), and Staphylococcus saprophyticus (5.13%). 3.85 percent was made up of Pseudomonas

aeruginosa and Proteus mirabilis, and 2.56 percent was made up of Enterobacter cloacae. In a similar study, Shakir (14) found that among pregnant women, Escherichia coli made up 26.36 percent of the bacteria that were isolated. This was followed by Staphylococcus aureus, which made up 25.18 percent. A previous study from Nigeria found that E. coli was the most common type of bacteria found in pregnant women with UTI (42.1%) (15). In their study, Ekuma et al. (16) found that there was no significant difference between the two patient groups when it came to the prevalence of E. coli, which was found in 14.1% of pregnant women and 20.9% of non-pregnant women. E. coli usually get into the bladder from the perineum and beat the body's natural defenses (5). Research from Iraq on the spread of disease showed that S. saprophyticus urinary tract infections (UTIs). This type of Staphylococcus makes an extracellular enzyme complex that can help both gram positive and gram negative bacteria grow. Escherichia coli and S. aureus are the most common bacteria that have been isolated. The high rate of infection by these bacteria may be because they can fight off bad conditions in the urinary tract by using virulence factors like adhesion and resistance (1,5,14). Some changes in the body's structure and function that happen during pregnancy make it easier for women who have bacteriuria to get symptomatic UTIs(161). Because of muscle relaxation caused by higher progesterone levels during pregnancy and the ureters getting obstructed by the growing uterus, the renal collecting system, especially the ureters, had less peristalsis. Progesterone may make you have more space in your bladder (12).

Table 6 shows the study's findings on the clinical features of urinary tract infections (UTIs) in pregnant women with gestational diabetes mellitus (GDM). These findings add to what is already known about the topic. Previous research has often pointed out that pregnant women, especially those with GDM, are more likely to get UTIs because their bodies and immune systems are changing. The fact that 76.92 percent of GDM pregnant women said they felt pain or discomfort while urinating supports the idea that these kinds of symptoms are common in this group of people who have UTIs. Also, the percentage of people who have back pain and recurrent UTIs is similar to what other studies have found (5,14). These common results from different studies show how important it is to keep an eye on and help pregnant women with GDM who are having certain types of UTIs so that both the mother and the baby are healthy (18,19). The current study shows that E. coli is more resistant to Ampicillin, Trimethoprim, and Cefotaxime. This shows how important it is to use smart treatment methods when dealing with this pathogen. S. aureus, on the other hand, has different reactions to different drugs. For example, 85% of times it is resistant to Ampicillin and 75% of times it is resistant to Gentamicin. The complexities go beyond these organisms. For example, S. saprophyticus is highly resistant to a wide range of antibiotics, and P. aeruginosa consistently resists many agents but responds to Imipenem and Levofloxacin. It was shown by Shakir (14) that is more resistant to ampicillin, trimethoprim, and cefotaxime, but very sensitive to ceftazidim (93.02 percent). S. aureus has different reactions to different drugs. It is very sensitive to ciprofloxacin and oxacillin (88.23%), but it is completely resistant to ampicillin and lincomycin. These differences in susceptibility show how hard it is to fight infections caused by these bacteria and how important it is to use antibiotics that are specifically designed for each person. The findings

showed that the bacteria samples were very resistant to beta lactams. This is because they make the enzyme β-Lactamase. Antibiotics lose their effectiveness when this enzyme cuts the β-Lactam ring in them. Resistance was also linked to changes in how easily bacteria's cell membranes let things pass through them. The study showed many different ways that antibiotics are not working, such as genetic changes that affect parts of the plasma membrane that help antibiotics enter cells and move between them. Additionally, the part plasmids play in moving resistance genes around through transformation, conjugation, and transduction was emphasized (20). The study's results show a full analysis of the virulence factors in the bacterial isolates, highlighting how dangerous they could be. First, all isolates tested positive for Coagulase, Urease, Sinophobe, and Gelatinase production, which means they are very likely to cause disease. The majority of isolates also showed high levels of other virulence factors, with 88.24 percent showing Beta-hemolysis and Capsule production. Also, all of the isolates were positive for both DNase and Gelatinase production, which means they can break down nucleic acids and proteins, respectively. A lot of the isolates showed that they could make Beta-lactamase (82.35%) and Lipase (82.35%), which means they might not be sensitive to antibiotics and could damage tissue. Some virulence factors, like Lecithin production (76.47%) and Protease production, showed different percentages, though (47.06 percent). In a study conducted by Nordin (21), it was determined that 42.2% of coagulase-positive staphylococci exhibited positive results for the DNase test, 53.3% for the presence of a capsule, and 74.4% for hemolysis. Staphylococcus aureus has emerged as a significant human pathogen, contributing to both hospital- and community-acquired infections over an extended period (5). This pathogen is implicated in various infections, including septicemia, pneumonia, wound sepsis, septic arthritis, osteomyelitis, and post-surgical toxic shock syndrome, presenting high rates of morbidity and mortality (14).

The success of this human pathogen is attributed to its adaptability, appearing at different times and locations and displaying distinct clonal types and antibiotic resistance patterns regionally and nationally. Numerous virulence and pathogenicity factors in S. aureus have been identified, enhancing the bacteria's ability to colonize patient tissues, reducing susceptibility to antibiotics, and exacerbating infections (22).

Al-Khafaji's study (23) also found that 42.2% of coagulase-positive staphylococci were positive for the DNase test, 53.33% for the presence of a capsule, and 74.4% for hemolysis. Abo-Shama et al.'s research (24) revealed that 85.1% of S. aureus isolates exhibited β-hemolysis and DNAse synthesis, aligning with our own findings. The composition of red blood cells in the culture media is pivotal in determining the bacteria's capability for β-hemolysis, among other factors. The presence of serum and cholesterol in the blood used for the test, along with the testing method itself, influences the ability to detect the bacteria's β-hemolysis capacity, subsequently impeding the hemolysis process (25).

Staphylococcus aureus can produce hemolysin for various reasons, including its osmotic properties, its ability to create cell perforations, and its cytotoxic effects, enabling the destruction of specific human blood cells (26). Consistent with the majority of findings, S. aureus isolates exhibit the production of extracellular enzymes such as DNase, lecithinase, and

protease (27). These findings provide a more detailed understanding of the individual pathogenicity profiles of the isolates, emphasizing the need for specific therapeutic strategies to address these infections.

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