Abstract

In present study thirty one of bacterial isolates were isolated from twenty six samples collected from different patients suffered with skin infections. The bacteria isolated on culture media (Mannitol salt agar , MacConkey agar and blood agar), identified and determined the antibiotic resistance by vitek 2 system. The results showed that most frequent infection were impetigo, boil and chronic folliculitis 5(19%) at age group (2-17), (18-55) and (18-38) years respectively. According to the gender the infections were more in females 15(57.6%) than males 11(42.3%), of all those infections, impetigo and boil were more common in females 4(15%), while skin cancer and chronic folliculitis were more common in males 3(12%). The results of bacterial isolation showed that 26 (83.87%) of isolates were Gram positive bacteria while 5(16.13%) were Gram negative, The high frequent genus in identified by Vitek 2 system was *Staphylococcus* sp. followed by *Acinatobacter* sp., *Aeromonas* sp., and *Sphingomonas* sp., the most dominant species was *staphylococcus aureus* 11 (35.5%) followed by *Staphylococcus. epidermidis* 8(25.8%). The results of antibiotic resistance to (48) antibiotics from different groups revealed that out of (31) isolates (23) were multi-drug resistance bacteria, the more resistance isolates were *Staphylococcus aureus* (R3) was resist to (12) classes of antibiotics followed by *Staphylococcus hominis* (R16) and *Staphylococcus epidermidis* (R18) (9) classes. There has been a concerning rise in the antibiotic-resistant pattern of skin infection-causing *Staphylococcus aureus*. These days, it is imperative to utilize antibiotics wisely and to put antimicrobial stewardship into practice.

Key words: Skin infection, bacteria, drug resistance

Introduction

Skin is the largest organ in human body and basic barrier, serving as the first line of defense against bacterial infections. It's not only blocking the pathogens from entry to body from the environment, but also providing a large-scale biological niche for a wide range of bacteria (Ibrahim *et al*., 2015). Skin and soft tissue infections (SSTIs) are a major cause of morbidity and mortality, they are characterized by a microbial invasion of the skin layers and underlying soft tissues which range in severity from mild to life threatening, the infections can occurs from the uncomplicated cellulitis, to abscesses, deep tissue necrosis and necrotizing fasciitis (Esposito *et al*., 2016 ; Allaw *et al*., 2023). Bacterial SSTIs are mainly caused by Gram-positive bacteria like *Staphylococcus aureus* and β-hemolytic streptococci and many coagulase-negative *Staphylococcus*, Gram-negative Enterobacteriaceae, non-fermentative bacteria such as *Pseudomonas* spp. and *Acinetobacter bumaniin* can also cause (Ramirez *et al*., 2020 ; Allaw *et al*., 2023). Necrotizing fasciitis, scarlet fever, erysipelas, erythrasma, abscesses, folliculitis, furunculosis, and impetigo have all been related to a numerous of bacterial skin infections (Al-Kahfaji, 2022).

Cellulitis is an acute bacterial infection that is inflaming the subcutaneous tissue around it as well as the deep dermis (Brown and KL, 2019). The bacteria that most commonly involved of cellulitis are *S. aureus* and group A streptococci, also Gram negative bacteria *Pseudomonas aeruginosa* and Enterobacteriaceae group (Sari *et al*., 2022 ; Joseph, *et al*., 2022). Erysipelas is a severe Streptococcal infection of the skin primarily spreading through the lymphatic vessels (Jendoubi *et al*., 2019). It's a soft tissue infection involving the upper dermis, it has more distinct margins when compared with other soft tissue skin infections. Impetigo is a superficial soft tissue skin infection infect the epidermis. Is a most common bacterial skin infection in children two to five years of age. Is caused by *S. aureus* or *S. pyogenes* (Linz *et al*., 2023). Currently, folliculitis is a common skin condition, which an inflammation of the tiny pockets in the skin of hair grow (hair follicles) and the per follicular tissue that can affect healthy people of any age group or any sex. It occurs when bacteria infect hair follicles. Some of resolve spontaneously, but others progress to furuncles (boils),

involve adjacent tissue. That is distinguished with red swelling ,hard and painful lumps filled with pus. *S. aureus* are most common bacteria cause of these infection (Nasr, 2018 ; Jappa, and Kutre, 2018 ; Lin *et al*., 2021).

Resistance to antibiotics is one of the crucial issues related to public health. and one of the most vital threats to the healthcare sector is the rise of antibiotic-resistant microbes. Multidrug-resistant bacteria (MDR) that are deadly pathogenic are rising day by day and pose a very serious threat to human health. Whereas earlier, such antibiotic resistance was only found in nosocomial infections, but it is now become a common phenomenon (Jubeh *et al*., 2020 ; Bharadwaj *et al*., 2022). The spread of "superbugs" that are now resistant to several antibiotics is a serious issue (Davies and Davies, 2010). These include the ESKAPE pathogens (*Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa, and Enterobacter* spp.) (Khan and Khan, 2016). The total deaths from methicillin-resistant *S. aureus* (MRSA) are now comparable to those caused by HIV, and it is predicted that by 2050, antimicrobial resistance would be responsible for at least 10 million annual deaths (De Kraker *et al*., 2016). So the present study aimed to evaluate the clinical and bacteriological profiles of bacterial isolates in skin infections, including the number and type of potential bacteriological pathogens as well as antibiotic susceptibility and pattern of isolates because of limited data are available regarding skin and soft tissue infection, mortality rate, and antibiotic susceptibility of bacteria in our hospital settings.

Material and Methods

Samples collection

A total of 26 samples were collected from patients undergoing skin infections includes cellulitis, erysipelas, impetigo, folliculitis, furuncles and acne in Al-Sadr Teaching Hospital, Al-Fyhaa Teaching Hospital, AL-Mauana Teaching Hospital and Tumor center in Basrah province south of Iraq at the period from the beginning of October 2022 to the end of

December 2022. The age of patients ranged between 3 - 70 years old. It should be mentioned that all patients in this study were selected depending on their history and clinical examination by specialist doctors. Samples that taken were collected from deep parts of the infection (pus) by a cotton sterile media swab. Then all samples were collected in sterilized containers and transported to a laboratory for isolation.

Culturing of bacteria

All swabs were streaking on the media of blood agar , MacConkey agar and Mannitol salt agar the plates incubated at 37 ^{o}C for 24 hrs. Then observed for the presence of isolated colonies. re-cultured on nutrient agar plates for purification and subsequent experiments.

Identification of bacterial isolates

The vitek 2 system was performed at Al-Bayan laboratory in Basra city by using Vitek 2 kit (NG REF 21 341) (pincus, 2006).

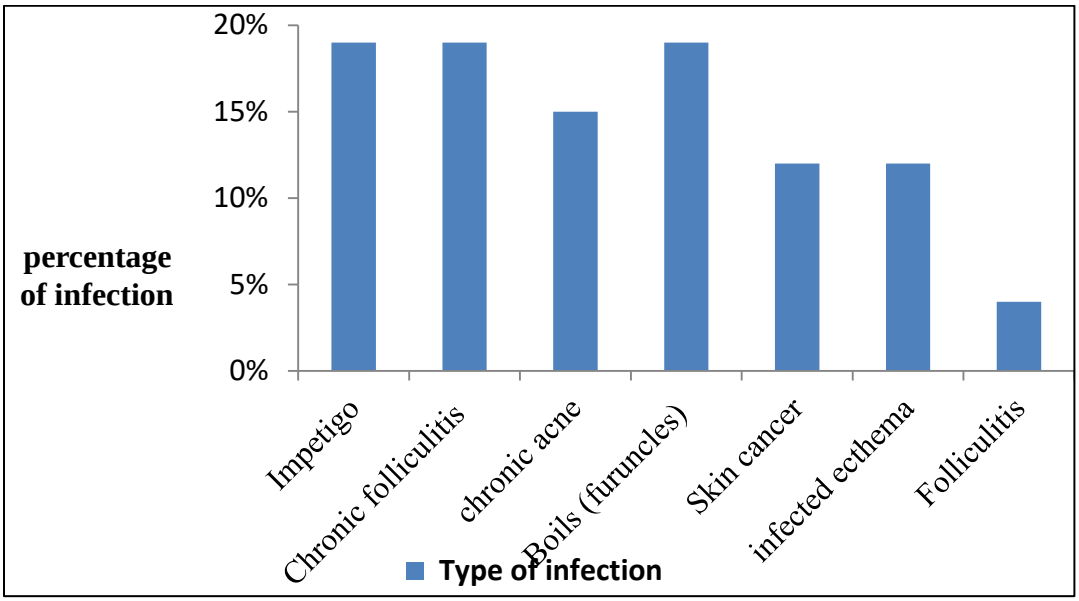
Antibiotic sensitivity test

The sensitivity and resistance of isolates were tested by using VITEK 2 system (Piddok, 1991).

Results

Skin infection frequency

The result of skin infection frequency showed that impetigo was the most frequent infection with percentage 5(19%) followed by chronic folliculitis and boils (furuncles) 5(19%), while folliculitis infection recorded the lowest frequent 1(4%) figure (1).



Figure(1): Percentage of skin infection.

Skin infection percent according to the age

Table (1) revealed the results of skin infection according to the age. Where the percentage of impetigo, boils (furuncles) and chronic folliculitis was 5(19%) at (2-17), (18-55) and (18-38) years respectively. Folliculitis infection showed the lowest percentage 1(4%) at the age of 73 years old .

Table (1): Percentage of skin infections according to the age

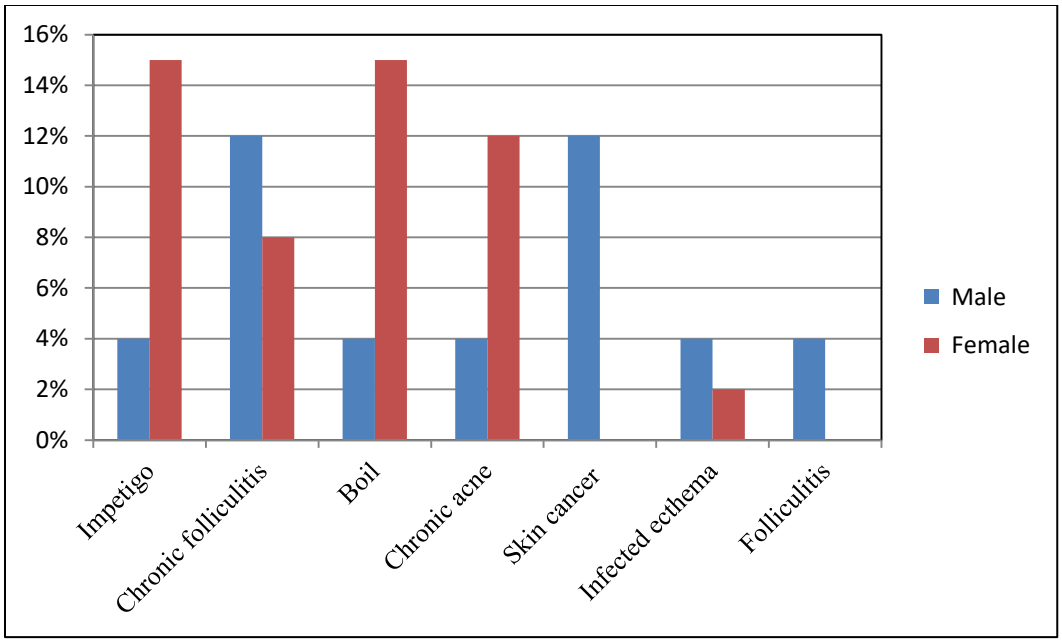
Type of infection	**No.of infections per(%)**	**Age of patients**
Impetigo	5 (19%)	2-17
Chronic folliculitis	5 (19%)	18-38
Skin cancer	3 (12%)	25-35
Boils (furuncles)	5 (15%)	18-55
Folliculitis	1 (4%)	73
infected ecthema	3 (12%)	9-50
chronic acne	4 (15%)	27-25
Total	**26**	

Skin infection percent according to the gender

The results showed, that the frequent of skin infection in female (15) was more than male (11), the impetigo and boils (furuncles) showed the highest percentage 4(15%) in female compared to the male where the skin cancer and chronic folliculitis recorded 3(12%) table (2) and figure (2).

Table (2): percentage of skin infection according to the gender

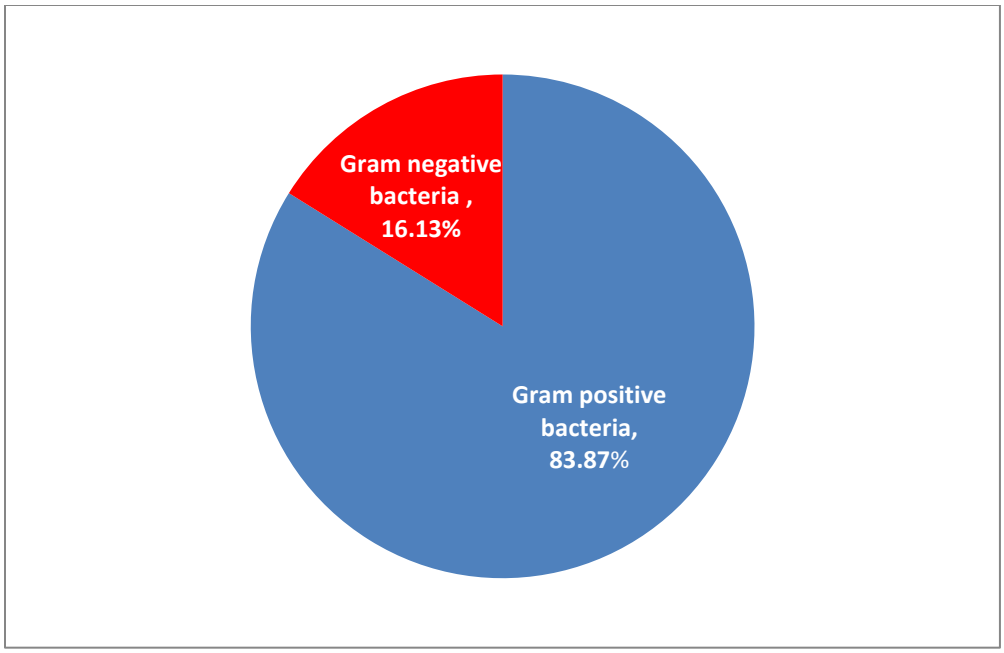
Type of infection	**Male**	**Female**
Impetigo	1 (4%)	4(15%)
Boils (furuncles)	1(4%)	4(15%)
Skin cancer	3(12%)	0
Chronic folliculitis	3(12%)	2(8%)
Folliculitis	1(4%)	0
infected ecthema	1(4%)	2 (8%)
chronic acne	1(4%)	3(12%)
Total	**11 (42.3%)**	**15(57.6%)**



Figure(2): percentage of infections according to the gender.

Culturing of samples and isolated of bacteria

All samples were cultured on blood base agar, Mannitol salt agar, and MacConcy agar media with duplicate. Out of 26 samples, 25 samples showed bacterial growth while one sample showed no. The total of isolates that obtained were 31 and depending on Gram staining, the percentage of Gram positive was high 26 (83.87%) compared to Gram negative 5 (16.13%) figure (3).



Figure(3): percentage of Gram positive and Gram negative bacteria isolated from skin infections.

Identification of bacterial isolates

Thirty-one isolates were identified by Vitek 2 system analysis. The results showed identified 4 genera and 10 species with identity percent about (99-86%) table (3).

Table(3):Identification of bacterial isolates by vitek 2 system

Sample No	Type of bacteria	Identity
R1	*S. hominis ssp hominis*	94%
R2	*A. baumannii complex*	99%
R3	*S. aureus*	86%
R4	*S. aureus*	95%
R5	*S. aureus*	91%
R6	*S. aureus*	92%
R7	*S. aureus*	87%

R8	*S. epidermidis*	96%
R9	*S. aureus*	92%
R10	*S. aureus*	99%
R11	*S. aureus*	95%
R12	*S. aureus*	92%
R13	*S. aureus*	99%
R14	*S. aureus*	99%
R15	*S. epidermidis*	92%
R16	*S. hominis ssp hominis*	95%
R17	*S. epidermidis*	92%
R18	*S. epidermidis*	99%
R19	*A. baumannii complex*	99%
R20	*A. baumannii complex*	99%
R21	*S. warneri*	87%
R22	*S. lentus*	94%
R23	*S. epidermidis*	95%
R24	*S. epidermidis*	95%
R25	*S. epidermidis*	99%
R26	*S. epidermidis*	99%
R27	*S. hominis ssp hominis*	92%
R28	*S. xylosus*	99%
R29	*Sphingomonas paucimobilis*	97%
R30	*S. lugdunensis*	
R31	*A. hydrophila/ Punctate (caviae)*	98%

The species of *S. aureus* showed significantly superior percentage 11(35.5%) followed by *S. epidermidis* 8(25.8%), *S. hominis* and *A. baumannii* were 3(9.7%) table (4) and figure (4).

Table (4): frequency and percentage of bacterial species identified by vitek 2 system.

Vitek 2 system	N	Per(%)
Staphylococcus aureus	11	35.5 %
S. epidermidis	8	25.8 %
S. hominis ssp hominis	3	9.7 %
S. lugdunensis	1	3.2 %
S. lentus	1	3.2 %
S. warneri	1	3.2 %
S. xylosus	1	3.2 %
Acinetobacter baumannii complex	3	9.7 %
Aeromonas hydrophila/punctata (caviae)	1	3.2 %
Sphingomonas paucimobilis	1	3.2 %

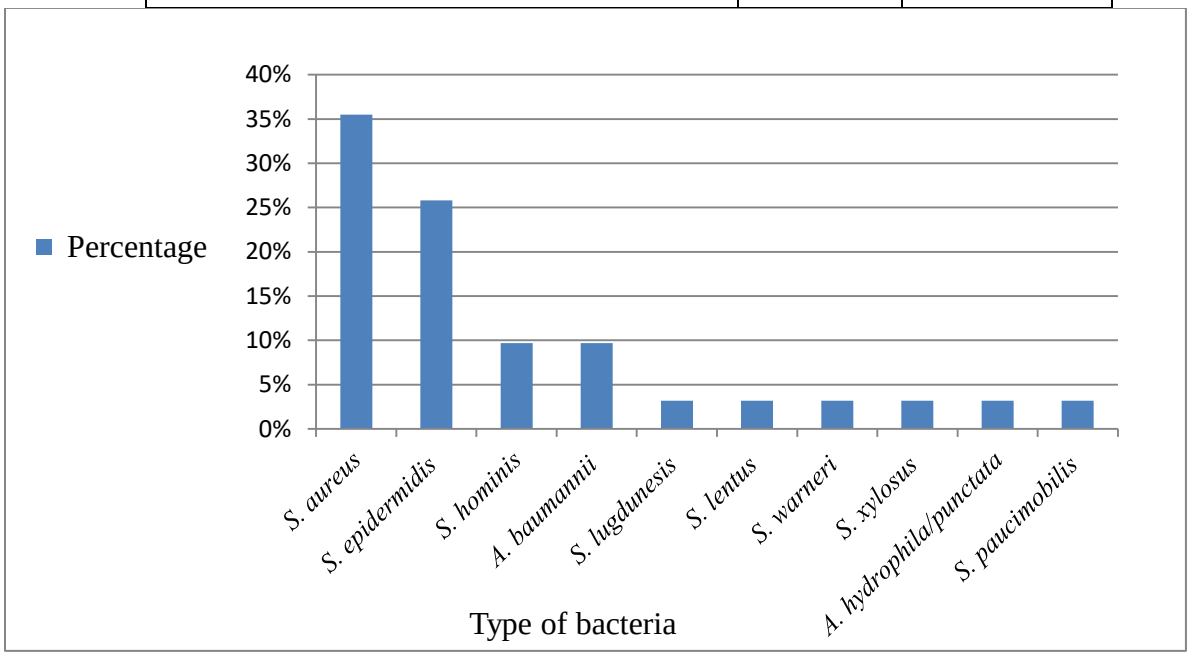


Figure (4): Percentage of bacterial species identified by vitek 2 system.

Antibiotic Susceptibility test by Vitek 2 System analysis

Antibiotic Susceptibility of all bacterial isolates was determined by using Vitek 2 Compact System analysis. Bacterial isolates were tested for susceptibility to different antibiotics from different groups. Out of 31 bacterial isolates 23 were shown to be multiple drug resistant (MDR), were resistant to more than three classes of antibiotics and were classified as MDR as shown in table (5). The isolate of R3 (*S. aureus*) was resistance for 12 classes of antibiotics followed by R16 (*S. hominis*) and R18 (*S. epidermidis*) resist for 9 classes of antibiotic, table (5).

Table (5):Number of antibiotics classes that resistance by bacteria isolated from skin infections.

Bacterial species	**Number of antibiotics class resistance by bacteria**	**Classes of antibiotics**
***S. hominis ssp hominis* R1**	7	Cephalosporins, Penicillins, Carbapenems, Quinolones, Macrolides, Lincosamides, and Tetracyclines
***A. baumannii complex* R2**	**6**	Penicillins, Cephalosporins, Carbapenems, Tetracyclines, polymyxine and Antifolate
***S. aureus* R3**	**12**	Cephalosporins, Penicillins, Carbapenems, Aminoglycosides, Quinolones, Macrolides, Lincosamides, Tetracyclines,

		Antifolate, Rifamycins , Fusidane and Sulphonamides
S. aureus **R4**	**5**	Cephalosporins, Penicillins, Carbapenems, Macrolides and Lincosamides
S. aureus **R5**	**7**	Cephalosporins, Penicillins, Carbapenems, Macrolides, Tetracyclines Fusidane and Lincosamides
S. epidermidis **R8**	**7**	Cephalosporins, Penicillins, Carbapenems, Aminoglycosides, Macrolides, Tetracyclines and Fusidane
S. aureus **R9**	**3**	Cephalosporins, Penicillins, and Carbapenems
S. aureus **R10**	**3**	Cephalosporins, Penicillins, and Carbapenems,
S. aureus **R11**	**6**	Cephalosporins, Penicillins, Carbapenems, , Aminoglycosides, Quinolones, and Macrolides
S. aureus **R12**	**3**	Cephalosporins, Penicillins, and Carbapenems
S. aureus **R13**	**3**	Cephalosporins, Penicillins, and Carbapenems
S. aureus **R14**	**3**	Cephalosporins, Penicillins, and Carbapenems
S. epidermidis **R15**	**7**	Cephalosporins, Penicillins, Carbapenems, , Aminoglycosides, Lincosamides, Glycopeptides and Fusidane

***S. hominis ssp hominis* R16**	**9**	Cephalosporins, Penicillins, Carbapenems, Glycopeptides , Quinolones, Lincosamides, Fusidane, Rifamycins, and Macrolides
***S. epidermidis* R17**	**7**	Cephalosporins, Penicillins, Carbapenems, Macrolides, Lincosamides, Tetracyclines and Fusidane
***S. epidermidis* R18**	**9**	Cephalosporins, Penicillins, Carbapenems, Quinolones, Macrolides, , Tetracyclines, Antifolate, Sulphonamides and Fusidane
***S. warneri* R21**	**5**	Cephalosporins, Penicillins, Carbapenems, Macrolides, and Fusidane
***S. lentus* R22**	**6**	Cephalosporins, Penicillins, Quinolones, Macrolides, Sulphonamides and Fusidane
***S. epidermidis* R23**	**7**	Cephalosporins, Penicillins, Carbapenems, , Aminoglycosides, Lincosamides, Glycopeptides and Fusidane
***S. epidermidis* R24**	**6**	Cephalosporins, Penicillins, Macrolides, Lincosamides Glycopeptides and Fusidane
***S. epidermidis* R25**	**4**	Cephalosporins, Penicillins, Lincosamides, and Glycopeptides
***S. epidermidis* R26**	**3**	Cephalosporins, Penicillins, and Carbapenems
S. hominis ssp hominis	**6**	Cephalosporins, Penicillins,

R27		Lincosamides, Macrolides, Glycopeptides and Fusidane

Discussion

In the current study, Out of twenty six samples collected from various skin infections, Impetigo and chronic folliculitis were the most frequent infection 5(19%) figure (1), according to the age the results showed that most of impetigo infections were between 2-17 years, table (1) this was agreed with previous studies that showed impetigo common in children under 18 years (Loadsman *et al*., 2019 ; Bowen *et al*., 2015). Children between the ages of two and five are typically affected by impetigo, a highly contagious superficial skin illness that comes in two varieties: nonbullous impetigo and bullous impetigo (Brown *et al*., 2003 ; Manje *et al*., 2023).

Skin infection is a prevalent illness that affects children. Geographical location, climate and season, social status, and personal hygiene all have an impact (Kelbore *et al*., 2019; García *et al*., 2020). Alkubaisi *et al*. (2020) suggests that because this age group of "schoolchildren" has access to the same healthcare services, and practices almost identical personal hygiene, they are more likely to be exposed to infectious skin diseases. In a developing country like Iraq, it is a complicated big health issue. Temperature, humidity, poor personal hygiene, a lack of good water sources, and low levels of education allow the disease to spread among children in the community. One of the contributing reasons to skin infections and infectious diseases is personal cleanliness (Gauchan *et al*., 2015; Aggarwal *et al*., 2021).
For chronic folliculitis the infection was between (18-38) years table (1), this was compatible with Srinivas and KA, (2020) that observed most commonly in the age group of (21-40) years.

The infections were significantly higher in female 15(57.6%) compared to male 11(42.3%), table (2), this finding is consistent with previous studies conducted in Brazil and Egypt (Ferreira *et al*., 2011; Abdel-Hafez, *et al*., 2003) and disagreement with other study conducted in Iraq by Alkubaisi *et al*. (2020). According to the gender

the results showed that prevalence of impetigo infections were significantly higher among female (Little girls) 4(15%) than male (Little boy) table (2) and figure (2), while in chronic folliculitis the results showed that the infections were high in male 3(12%) compared to the female 2(8%) this was in agreement with Srinivas and KA, (2020). Immune modulation is influenced by progesterone, testosterone, and estradiol. They impact a wide range of cells, such as lymphocytes, macrophages, and dendritics (Muenchhoff and Goulder , 2014). It is well known that testosterone suppresses the immune system, which may lower interferon-γ levels (IFN-γ), Conversely, estradiol has the ability to increase T-helper 1 (Th1) immunity (Jacobsen and Klein, 2021). So, the difference in the level of estradiol and testosterone between males and females could effected.

Depending on Gram staining, 26 (83.87%) were Gram positive, whereas 5 (16.13%) Gram negative. The result come in agreement with Ahmed *et al*. (2020) where Gram positive cocci recorded (65.4%) while 68 (34.5%) was Gram negative bacilli. The bacteria was identified biochemically by an automated microbiology vitek 2 system, it's used for microbial identification provide highly accurate and reproducible results that approved by multiple independent studies. The result revealed that isolated bacteria with probability range between (99-86%) the frequency showed high percentage of *S. aureus* 11(35.5). This outcome is consistent with research conducted by previous studies (Matiny *et al.*, 2012 ; Marko *et al*., 2012; Sanaa, 2017), which used the Vitek 2 method to identify a variety of bacterial species.

The result showed that *Staphylococcus* genus was the most frequent with high percent (35.5%) of *S. aureus* this compatible with previous studies (Mohanty *et al*., 2018; Zhao *et al*., 2021 ; Manje *et al*., 2023) . Major Gram-positive bacterial pathogen *S. aureus* is responsible for a broad range of clinical illnesses, from endocarditis and localized soft-tissue infections to potentially fatal bacteremia (Mohanty *et al*., 2018). Human skin is frequently home to the commensal bacteria *S. epidermidis*. In the present study this species showed the percent of (25.8 %) .This bacteria viewed as a key member of the healthy skin microbiota, involved in the fight against pathogens, influencing the immune system, and implicated in wound repair. Concurrently, it is the

second source of nosocomial infections, and skin conditions including atopic dermatitis have been linked to an overgrowth of this bacteria (Landemaine *et al*., 2023).

Antibiotic susceptibility testing was performed using the Vitek- 2 Compact system analysis , (48) antibiotics were used. the results revealed that bacterial isolates had different patterns of resistance to antibiotics table (5). (23) isolates of bacteria were MDR out of (31) isolates . Widest spectrum of resistance was observed of *S. aureus* (R3) among the Gram positive bacterial isolates, this isolate showed more resistance to antibiotics than any other where resist (12) classes of antibiotics such as aminoglycosides, penicillins, macrolides, quinolones, lincosamides , tetracyclines, antifolate and sulphonamides the result was compatible with previous studies in Botswana (Truong *et al*., 2011; Alter *et al*., 2019). The bacteria of *S. epidermidis* (R18) was resist for (9) classes of antibiotics include penicillins , cephalosporins , tetracyclines, macrolides, antifolate and sulphonamides this result agreed with previous studies (Chabi and Momtaz, 2019; Siciliano *et al*., 2023; Leili *et al*., 2024).

Conclusion

According to the current investigation, *S. aureus* and *S. epidermidis* are the two Gram-positive bacteria isolates most frequently associated with skin infections. The concerning increase in Gram-positive bacterial infections that are resistant to many drugs highlights the critical need to determine the sensitivity pattern and epidemiology of the bacteria causing infections of the skin and soft tissues. Adhering to antibiotic stewardship can assist in preventing the development of drug resistance and minimizing the use of ineffective antibiotics.

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