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Evaluation of secondary bacterial infection of skin diseases in Egyptian in- & outpatients & their Sensitivity to antimicrobials

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Abstract

Background: Organisms causing bacterial infections complicating dermatoses differ among in- and outpatients. They constantly change their antibiotic sensitivity, thereby posing additional concern about disease outcome.

Objective: The aim of the present study was to detect the types of bacteria commonly complicating skin diseases of Egyptian in- and outpatients of the dermatology department and to test their sensitivity to a panel of the most commonly used antibiotics.

Patients & Methods: Direct film was prepared and samples were cultured aerobically and anaerobically from the deeper parts of the suppurative exudate of secondarily infected skin of 37 outpatients and 23 inpatients suffering from various dermatoses. This was followed by antibiotic sensitivity testing in addition to testing of gram positive and gram negative organisms for beta lactamase and extended beta lacamase (ESβL) production, respectively.

Results: Staphylococcus aureus (83.3%) and Gram-negative enteric bacteria (21.7%) were the most common isolated organisms from all cases. Streptococcus pyogenes, Pseudomonas aeruginosa and Enterococci were detected in 15%, 6.7% and 5% of cases respectively. There was significant difference between in- and/outpatients as regards the antibiotic sensitivity pattern of both S.aureus and the Enterobacteriaceae group. S.aureus strains isolated from inpatients showed more resistance to amoxicillin/clavulonic acid, cefaclor, fusidic acid, methicillin, ofloxacin and tobramycin ($p<0.05$). The rate of methicillin resisitant staph aureus (MRSA) in hospitalized patients was (26.3%) versus (9.7%) in outpatients ($p<0.05$). Enterobacteriaceae strains from hospitalized patients were more resistant to cefaclor, levofloxacin and ofloxacin ($p<0.05$).

Collectively, the incidence of β-lactamase production by Gram-positive cocci was (96%) in both groups, while the incidence of ESβL production by Gram-negative bacilli was (29.4%), being higher in inpatients than outpatients.

In conclusion, this study shows that S.aureus is the most common cause of secondary infection in all skin lesions. The incidence of Enterobacteriaceae infection was more in inpatients with higher levels of ESβL production. Resistance of different bacterial isolates to antibiotics was also higher in inpatients.

Introduction

An intact stratum corneum prevents invasion of skin by normal skin flora or pathogenic microorganisms. Skin diseases that are usually complicated by secondary bacterial invasion can be broadly classified into itchy skin conditions in which scratching provides a portal of entry to microorganisms such as scabies and pediculosis, and those characterized by absence of skin barrier, such as eczema, pemphigus and ulcers [1].

The most common causes of secondary bacterial infections of the skin are staphylococci and streptococci. Secondary infections to skin lesions can be potentially life threatening and may progress rapidly; therefore, their early recognition and proper medical and surgical management are important [7].

The current work aims at isolation and identification of bacteria causing secondary infection of skin diseases in Egyptian patients visiting the outpatient clinic, or admitted as inpatients in the dermatology department, Ain Shams University Hospital. Determination of antibiotic sensitivity for these bacteria, testing the β-lactamase production by the Gram-positive cocci, and the extended spectrum β-lactamase (ESβL) production by the Gram-negative bacilli will be done in an attempt to detect whether there are differences among outpatients, which represent patients in the normal community and inpatients, who reflex nosocomial skin diseases.

Subjects And Methods

Patients: Included in the present study were patients attending the outpatient clinic or those admitted as inpatients in the dermarology department, Ain Shams University Hospital suffering from different skin diseases which are complicated by secondary bacterial infections. The study was conducted during the period from April 2005 to December 2005. All patients were subjected to full history taking, clinical and bacteriological examination.

Methods: A sample from the suppurative exudates of infected skin lesions was taken by means of sterile disposable swab and inoculated into peptone water as transport medium for aerobic bacteria between the clinic and the bacteriological laboratory. Another sample was taken by another swab and inoculated into thioglycolate broth as transport medium for anaerobic bacteria.

Direct films were prepared from the samples and were stained with Gram stain and examined by oil immersion of light microscope for the presence and morphology of microorganisms.

The samples were cultured aerobically at 37°C for 18-24 hours on Blood agar medium and Mac Conkey's agar medium. The second sample was cultured anaerobically on Colombia blood agar medium in anaerobic jar using an anaerobic gas pack for 48 hours at 37°C. Bacteriological

identification of the colonies was done according to Colee et al.[14].

All isolated organisms were tested for antibiotic sensitivity by disc diffusion method using commercially prepared discs 6 mm in diameter (Oxoid-England). Interpretation of results was done according to National Committee for Clinical Laboratory Standards [30,31].

Gram-positive cocci were tested for production of β-lactamase by nitrocefin discs (Mast Diagnostic-England) for rapid detection of β-lactamase. Positive results: Development of a red colour in the area of the disc where the culture was applied. Negative results: No colour change.

All Gram-negative bacilli were tested for the production of extended spectrum β-lactamase (ESβL) enzyme by ESβL detection discs (Mast Diagnostics-England), which contain three paired sets of cartridges, each cartridge containing 50 discs. The diameter of any observed zones of inhibition was measured and recorded:

$$\frac{\textbf{*CAZ/CLAV}\ [\text{zone diameter (mm)}]}{\textbf{CAZ}\quad [\text{zone diameter (mm)}]}$$

*CAZ: Ceftazidime, CLAV: Clavulonic acid

An index number > 1.5 means positive ESβL and < 1.5 indicates negative ESβL. The calculations were repeated with the results from the remaining sets of discs. A positive result from any or all of the sets of MAST ID ESβL detection discs indicated ESβL production [16].

Stastical analysis

Analysis of the data was performed by SSPS version 12, where the data were expressed as mean ± standard deviation. Unpaired student t-test was used for between groups comparison of numerical variables, while Chi square test or Fischer exact test were used for comparison between categorical variables. P-value < 0.05 was considered significant.

Results

This study was conducted on 60 patients suffering from secondary bacterial infection of skin diseases; 37 outpatients (61.7%) and 23 inpatients (38.3%). Thirty five patients were males (58.3%) and 25 were females (41.7%), their ages ranged from 2 months to 75 years (mean 27 19).

Outpatients suffered from eczema (n=10), pediculosis (n=6), scabies (n=5), papular urticaria (n=4), sweat rash (n=4), kerion (n=3) and viral infections of the skin (herpes simplex and chicken pox, n=3). The inpatient group was suffering from pemphigus (14 patients), psoriasis (two with generalized pustular psoriasis and one with extensive chronic plaque psoriasis) in addition to 6 patients with skin ulcers due to pyoderma gangrenosum, vasculitis, lymphoedema and dermatitis artefacta **(Table1)**. All inpatients, with the exception of those with lymphoedema and dermatitis artefacta, were receiving

immunosuppressive therapy.

Organism		Pemphigus	Eczema	Skin ulcer	Pediculosis	Scabies	Papular uiticaria	Sweat rash	Viral infection	Kerion	Psoriasis	Total (Org.)	P-value
Staph aureus	**No. of cases**	**13**	**9**	**4**	**5**	**4**	**4**	**4**	**1**	**3**	**3**	**50**	**> 0.05**
	Percentage	92.9%	90%	50%	83.3%	80%	100%	100%	33.3%	100%	100%	83.3%	
Strept. pyogenes	**Count**	**1**	**4**	**0**	**0**	**2**	**0**	**0**	**1**	**0**	**1**	**9**	**> 0.05**
	Percentage	7.1%	40%	0%	0%	40%	0%	0%	33.3%	0%	33.3%	15%	
CONS	**Count**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**1**	**0**	**0**	**1**	**> 0.05**
	% within	0%	0%	0%	0%	0%	0%	0%	33.3%	0%	0%	1.7%	
Enterococci	**Count**	**1**	**0**	**0**	**2**	**0**	**0**	**0**	**0**	**0**	**0**	**3**	**> 0.05**
	Percentage	7.1%	0%	0%	33.3%	0%	0%	0%	0%	0%	0%	5%	
Entero-bacteriacae	**Count**	**5**	**2**	**3**	**0**	**1**	**0**	**0**	**2**	**0**	**0**	**13**	**> 0.05**
	Percentage	35.7%	20%	37.5%	0%	20%	0%	0%	66.7%	0%	0%	21.7%	
Pseudomonas	**Count**	**2**	**0**	**2**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**4**	**> 0.05**
	Percentage	14.3%	0%	25%	0%	0%	0%	0%	0%	0%	0%	6.7%	
Anaerobic	**Count**	**1**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**1**	**> 0.05**
	Percentage	7.1%	0%	0%	0%	0%	0%	0%	0%	0%	0%	1.7%	
No Organism	**Count**	**1**	**0**	**0**	**1**	**1**	**0**	**0**	**0**	**0**	**0**	**3**	**> 0.05**
	Percentage	7.1%	0%	0%	16.7%	20%	0%	0%	0%	0%	0%	5%	
Total (Primary Lesion)		**14**	**10**	**6**	**6**	**5**	**4**	**4**	**3**	**3**	**3**		

Table (1): Incidence of infection by different bacterial isolates in the different primary skin lesions

Bacterial isolates from secondarily-infected skin diseases:

The commonest encountered organism was Staphylococcus aureus (61.8%) followed by Gram-negative bacilli of the family Enterobactericeae (16%), then Strept.pyogenes (11.1%), Pseudomonas aeruginosa (5%), Enterococci (3.7%), and the least isolated were anaerobic Gram-positive cocci and coagulase negative staphylococci (CONS) (each representing 1.2%).

As regards Enterobactericeae, E.coli and Proteus species were the most commonly isolated organisms, (30.8%) for each. Other less frequently encountered organisms included Serratia marcescens (15.3%), while Klebsiella pneumoniae, Citrobacter freundii and Enterobacter aerogens were the least isolated (each represented 7.7%).

On comparing the type of primary skin lesion with the organism causing secondary bacterial infection, a non-statistically significant difference was detected ($p>0.05$). S.aureus was the commonest

organism isolated from all cases (83.3%) followed by the Enterobacteriaceaea group (21.7%), Strept.pyogenes (15%) and P.aerugnosa was isolated from (6.7%) of the cases. Enterococci, CONS and anaerobic Gram-positive cocci were the least isolated (5%, 1.7%, and 1.7% respectively). Mixed infections are responsible for overlap in percentage **(Table 1)**.

In pemphigus, S.aureus was the most commonly isolated organism (92.9%), followed by Enterobactericeae (35.7%) and Pseudomonas (14.3%), while Enterococci, Strept.pyogenes and anaerobic Gram-positive cocci were the least isolated, only one isolate of each was detected. In eczema patients, S.aureus was the commonest isolated organism (90%) followed by Strept.pyogenes (40%) and Enterobacteriaceae (20%), while no other organisms were isolated. In skin ulcers the commonest isolated organism was also S.aureus (50%), followed by Enterobacteriaceae (37.5%) and P.aeruginosa (25%) no other organisms were found. In scabies, S.aureus was the commonest isolated organism (80%), followed by Strept.pyogenes (40%), and Enterobacteriaceae (20%).However in viral infections of the skin Enterobacteriaceae were the most common isolated organisms (66.7%). In kerion, papular urticaria and sweat rash S.aureus was the only isolated organism **(Table 1)**.

As regards the distribution of bacterial isolates and the anatomical site from where they were detected, Enterobacreiaceae group and Strept.pyogenes were found more frequently in the lower limbs than in other sites **(Table 2)**

Site of secondary infection	Bacterial isolates						
	S.aureus	CONS	Strept. pyogenes	Enterococci	Enterobac-teriaceae	Pseudomonas	Gram +ve anaerobic cocci
Upper limbs	14	0	2	0	2	2	1
Lower limbs	18	0	6	0	7	2	0
Head and neck	13	1	0	2	2	0	0
Trunk	5	1	1	1	2	0	0

Table (2): Distribution of bacterial isolates according to the anatomical site of secondary infection.

As we compared the type and incidence of secondary bacterial infection between in- and outpatients, only Enterobactericeae group were more common among inpatients ($P<0.05$). There was no statistically significant difference ($P>0.05$) between inpatients and outpatients as regard percentage of mixed infection although it was higher in inpatients **(Table3)**.

Parameter	No (%)		P	S
	Inpatients -23	Outpatients (37)		
S.aureus	19 (82.6%)	31 (83.8)	>0.05	NS
CONS	0 (0%)	1 (2.7%)	---	---
Strept.pyogenes	2 (8.7%)	7 (18.9%)	>0.05	NS
Enterococci	1 (4.3%)	2 (5.4%)	---	---
Enterobactericeae	8 (34.8%)	5 (13.5%)	<0.05	S
Pseudomonas	2 (8.7%)	2 (5.4%)	---	---
Anaerobic gram +ve cocci	1 (4.3%)	0 (0%)	---	---
No organism	1 (4.3%)	2 (5.4%)	---	---
Mixed infection	9 (39.1%)	10 (27%)	> 0.05	NS

P: P value S: Significant NS: Non-significant

Table (3): Comparison between inpatients and outpatients as regards the incidence of secondary infection by the different bacterial isolates

Testing for antibiotic sensitivity of isolated organisms:

On testing for the antibiotic sensitivity of staphylococci, all S.aureus isolates had good sensitivity to clindamycin, chloramphenicol, and vancomycin, while they were resistant to penicillin, ampicillin, tetracycline and cefotaxime; without a significant difference between in- and outpatients. A significant difference between in- and outpatients was found as regards their sensitivity to amoxicillin/clavulonic acid, cefaclor, fusidic acid, methicillin, ofloxacin and tobramycin ($p<0.05$). Both groups were resistant to erythromycin, incidence in inpatients being higher. A larger percentage of inpatients were resistant to amoxicillin/clavulonic acid and fusidic acid compared to outpatients. Both groups were sensitive to cefaclor, methicillin, ofloxacin and tobramycin, but a larger percentage of the outpatient group was sensitive to these antibiotics **(Table 4)**.

Antibiotic	Inpatient			Outpatient			P value	Sig
	R	S	I	R	S	I		
Amoxycillin + clavulonic a.	85%	15%	---	58.10%	41.90%	---	< 0.05	S
Ampicillin	95%	5%	---	93.50%	0%	6.50%	> 0.05	NS
Cefaclor	25%	55%	20%	6.50%	93.50%	0%	< 0.05	S
Cephazolin	40%	45%	15%	12.90%	74.20%	12.90%	> 0.05	NS
Cefotaxime	75%	10%	15%	71%	9.70%	19.40%	> 0.05	NS
Chlor-amphenicol	10%	85%	5%	0%	96.80%	3.90%	> 0.05	NS
Clindamycin	15.80%	84.20%	---	12.90%	87.10%	---	> 0.05	NS
Erythromycin	57.90%	5.30%	36.80%	22.60%	0%	77.40%	< 0.05	S
Fusidic acid	63.20%	31.60%	5.30%	12.90%	80.60%	6.50%	< 0.05	S
Methicillin	26.30%	63.20%	10.50%	9.70%	90.30%	---	< 0.05	S
Ofloxacin	55%	45%	---	3.20%	96.80%	---	< 0.05	S
Penicillin	94.70%	5.30%	---	100%	---	---	> 0.05	NS
Tetracycline	79%	10.50%	10.50%	71%	25.80%	3.20%	> 0.05	NS
Tobramycin	50%	50%	---	6.50%	93.50%	---	< 0.05	S
Vancomycin	0%	100%	---	6.50%	93%	---	>0.05	NS

R: Resistant S: sensitive I: Intermediate Sig: Significance
S: Significant NS: Non significant

Table (4): Comparison between inpatients and outpatients as regards antibiotic sensitivity of

Staphylococci

As regards the antibiotic sensitivity of Gram-negative enteric bacilli strains, a statistically significant difference (P<0.05) to cefaclor, levofloxacin and ofloxacin between inpatients and outpatients was found. Outpatients were more resistant to cefaclor and more sensitive to levo- and ofloxacin. There was no statistically significant difference (P>0.05) in sensitivity to the other antibiotics, but resistance to amoxicillin/clavulonic acid, chloramphenicol, sulphamethoxazole/trimethoprim and tobramycin was more in the inpatient group, while outpatients showed same or higher resistance to cephazolin, cefoperazone and cefotaxime. All strains were resistant to ampicillin and piperacillin and showed good sensitivity to amikacin, aztreonam and imipenem **(Table 5)**.

Antibiotic	Inpatient			Outpatient			P	Sig
	R	S	I	R	S	I		
Amoxicillin + clavulonic acid	83.30%	16.70%	---	60%	40%	---	> 0.05	NS
Ampicillin	100%	---	---	100%	---	---		
Amikacin	28.60%	71.40%	---	---	100%	---	> 0.05	NS
Aztreonam	14.30%	71.40%	14.30%	16.70%	83.30%	---	> 0.05	NS
Cefaclor	42.90%	57.10%	---	50%	---	50%	< 0.05	S
Cephazolin	85.70%	14.30%	---	100%	---	---	> 0.05	NS
Cefoperazone	28.60%	42.90%	28.60%	16.70%	50%	33.30%	> 0.05	NS
Cefotaxime	42.90%	14.30%	42.90%	66.70%	33.30%	---	> 0.05	NS
Chloramphenicol	28.60%	71.40%	---	---	100%	---	> 0.05	NS
Imipenem	14.30%	85.70%	---	---	100%	---	> 0.05	NS
Levofloxacin	42.90%	57.10%	---	---	100%	---	< 0.05	S
Ofloxacin	57.10%	42.90%	---	---	100%	---	< 0.05	S
Piperacillin	100%	---	---	100%	---	---		
Sulphamethoxazole trimethoprim	57.10%	42.90%	---	---	83.30%	16.70%	> 0.05	NS
Tobramycin	57.10%	42.90%	---	16.70%	83.30%	---	> 0.05	NS

R: Resistant S: Sensitive I: Intermediate Sig: Significance S: Significant NS: Non-significant

Table (5): Comparison between inpatients and outpatients as regards antibiotic sensitivity of the Enterobacteriaceae

All Pseudomonas aeruginosa isolates (n=4) were sensitive to amikacin, cefoperazone and levofloxacin while they were resistant to cefepime. Resistance to cefotaxime, imipenem and piperacillin was more in the inpatient group while resistance to cephazolin was more in the outpatient group. Sensitivity to aztreonam was more in the outpatient group.

β-lactamase enzyme production was present in 96% of the total isolated Gram-positive cocci, which was as follows: 97.8% of the isolated staphylococci, while 66.7% of the isolated Enterococci.

E.coli (2/4) and Serratia (1/2) were the most common ESβL-producing organisms among members of the family Enterobacteriaceae followed by Proteus spp. (1/4) and Pseudomonas aeruginosa (1/4) while no ESβL production was detected among Klebsiella, Enterobacter and Citrobacter. ESβL production by Gram-negative bacilli was higher in inpatients, but due to small numbers of isolates, no statistical analysis could be performed.

Discussion

The present study aimed at isolating and identifying bacteria causing secondary infection in skin lesions, at determining the antibiotic sensitivity for these bacteria and at testing for β-lactamase and ESβL production by Gram-positive cocci and Gram-negative bacilli respectively in the inpatients and outpatients of the dermatology department Ain Shams University Hospital.

The results revealed that Staphylococcus aureus was the most common organism causing secondary infection of different skin lesions (eczema, pemphigus, skin ulcers, pediculosis, scabies, kerion, papular urticaria, psoriasis, sweat rash and viral infections). It was found in (83.3%) of all cases and represents (61.8%) of total isolated organisms.

Similarly, Ochsendorf et al. [33] in Germany, Brook [7] in USA and Lee and Tay [26] in Singapore mentioned that S.aureus was the commonest organism causing secondary infection of skin lesions and represented 67%, 43.5% and 45% of all positive cultures respectively. This might be related to the inhibitory effect of serum exuding from denuded skin on liolenic acid. Linolenic acid is an essential free fatty acid normally present on intact skin, which is responsible for inhibition of Staph colonization [25].

In the present study, enteric Gram-negative bacilli were the second most common pathogens causing secondary infection of skin lesions and were found in (21.7%) of all cases and represented (16%) of the total isolated organisms. They were followed by Strept.pyogenes which was isolated from (15%) of all cases and represented (11.1%) of total isolated organisms.

These results agree with those of Brook [7] who found that enteric Gram-negative bacilli together with Strept.pyogenes were the second most common causes of secondary infection where each of them represented (23%) of the total isolated organisms. On the other hand, they differ from those reported by Lee and Tay [26] who found that Strept.pyogenes was the second most common cause of

secondary infection and was identified in (21%) of cases. This difference may be because 38.3% of our patients were inpatients, and the incidence of Enterobacteriaceae among the inpatients was significantly higher than in outpatients.

On comparing the difference in bacterial isolates in inpatients to outpatients, the inpatients showed more common mixed infection and a statistically higher secondary infection by Enterobacreiaceae. Similarly, Gentry et al. [21] reported that Gram-negative bacilli were the second most common pathogen following S.aureus in inpatients. Other studies have shown that the second most common organisms causing secondary bacterial infections are Streptococcus pyogenes and Pseudomonas [26,33]. Differences in findings might be attributed to the fact that most of our inpatients were pemphigus patients who stayed for a long time in hospital.

Mixed infection was 31.7% with a rate of 1.35 pathogen/ patient with no significant difference between inpatients and outpatients, although it was higher in inpatients (39.1%) than in outpatients (27%). Mixed secondary bacterial infections of skin diseases have been reported [7,21,26]. The polymicrobial nature could be due to a potential for bacterial synergy [9]. In addition, being in a hospital for a long time facilitates acquisition of a number of organisms and immunosuppression may contribute to the occurrence of mixed infection.

Concerning the distribution of different bacterial isolates among the different anatomical sites, we found that there was no difference in the distribution of S.aureus among the different sites, whereas; the Enterobacteriaceae group was found more commonly in the lower limbs. Similarly, Brook [6] and Brook et al.[10,11] stated that organisms that reside in the mucous membranes close to the lesions predominated in infections next to these membranes. Enteric Gram-negative bacilli were found most often in buttock and leg lesions, the probable source of these organisms are the rectum and the vagina where they normally reside.

On the other hand our study disagrees with that of Brook[6,7], since he reported that streptococci were most commonly found in lesions of the head, face, neck and fingers. Strept reached these sites from the oral cavity, where they usually reside. We found that Streptococcus pyogenes was more commonly found in the lower limbs than in the other sites. These variations may be because we isolated Strept. pyogenes mainly from scabies and eczema. There is no clear explanation for the predominance of Strept in eczema lesions. As for scabies, Hay [22] noticed that Strept shows a special affinity to infect scabies lesions.

Regarding secondary infection of eczema lesions, the present study shows that S.aureus was the commonest (90%), followed by Strept.pyogenes (40%), while the Enterobacteriaceae were isolated from 20% of the cases. Similarly Brook et al. [10,11] reported that Staph aureus followed by Strept pyogenes were the most common aerobic or facultative organisms isolated both from infected atopic dermatitis as well as poison ivy dermatitis lesions respectively. Besides, most of our eczema cases were due to atopic dermatitis, it is therefore expected that Staph aureus is the predominant pathogen; since it colonizes atopic dermatitis patients even in the absence of skin lesions and may play a role in its pathogenesis. It is also the most common organism causing secondary bacterial infection of atopic dermatitis and other eczemas [27,28,29].

Denuded pemphigus lesions, were most commonly infected by S.aureus (92.9% of cases), followed by the Enterobacteriaceae group (35.7% of cases), then Pseudomonas aeruginosa (14.3%). Enterococci, Strept.pyogenes, anaerobic gram-positive cocci were found less commonly and were detected in (7.1% of cases each). As we can observe, the Enterobacteriaceae group represents the second most common organism and there is a high rate of mixed bacterial infections, owing to the fact

that all our pemphigus patients were inpatients and were under immunosuppressive therapy. Ahmed Moy [1] stated that S.aureus was the commonest organism isolated from secondarily infected pemphigus lesions and that septicemia resulting from it is the most important cause of death. Therefore, cautious observation of and antiseptic care for pemphigus lesions, in addition to a judicious use of steroids is mandatory.

Pseudomonas aeruginosa was the third most commonly detected organism in skin ulcers 25% following S.aureus (50%) and Enterobacteriaceae (37.5%). No other bacterial isolates were detected. Apart from Dissemond et al [18] who isolated mainly S.aureus from skin ulcers, various other studies reported prevalence of Pseudomonas aeroginosa. This is mostly related to the preference of Pseudomonas aeroginosa in colonizing and causing secondary bacterial infections of burns and skin ulcers [8,15,38]

As we examined the sensitivity of isolated bacteria to antibiotics, there was a significant difference between inpatients and outpatients as regards sensitivity of Staphylococci to antibiotics. S.aureus strains isolated from inpatients showed more resistance to amoxicillin/clavulonic acid, cefaclor, erythromycin, fusidic acid, ofloxacin and tobramycin. A significant difference in MRSA was detected between in- and outpatients, being much higher in the former (26.3%) than in the latter (9.7%) these isolates were vancomycin sensitive. On the other hand, both groups were almost equally sensitive to clindamycin, chloramphenicol and vancomycin and both groups were resistant to ampicillin, penicillin, cefotaxime and tetracycline. Variations in sensitivity are related to the frequency of usage of the individual antibiotics in hospitals compared to outpatients.

High resistance of S. aureus to ampicillin and penicillin (>90%) coincides with the high incidence of β-lactamase production by staphylococci in the current study which was also >90%. These organisms not only survive penicillin therapy but can also protect penicillin-susceptible bacteria from penicillin by releasing the free enzyme into the infected tissue or pus [7].

Our results contrast with those performed by El Kholy et al [19] who studied the antibiotic sensitivity of bloodstream isolated organisms in a number of hospitals in Cairo, Egypt including Ain Shams Specialized hospital. In their retrospective study, they found that only 29% of S.aureus isolates oxacillin/methicillin susceptible. Staphylococci were less sensitive to clindamycin (64%), cephazolin (29%) and chloramphenicol (53%) and almost equally resistant to erythromycin (51%) compared to our study. Differences in results may be due to differences in study design, where they did a retrospective in which they could not differentiate between nosocomial and community acquired cases.

Other studies have similarly detected that staph skin infections in outpatients responded well to amoxicillin/clavulonic acid, oxacillin and clindamycin, whereas erythromycin and ampicillin were less active. MRSA was lower in outpatients compared to inpatients , were it ranged from 1.5-21.5% in the former compred to 31-75% in the latter [13,20,23,32,38]. The prevalence of MRSA has increased worldwide, as it is evident from many surveillance studies [17,24,36]. However there are considerable differences between countries. The highest rates have been noted in developed countries and especially in Western Pacific regions [17], both in community acquired and nosocomial infections compared to developing countries such as Madagascar [35].

It is worth mentioning that in recent years, community-associated pathogens have increased dramatically and are likely to cause life-threatening systemic infections, especially in children and elderly individuals, and may also cause serious skin and soft-tissue infections in healthy individuals. Compared with nosocomial strains, community-associated MRSA isolates are associated with

increased virulence and currently are more likely to be susceptible to a variety of antibiotics [2].

Fusidic acid resistance was high in our inpatient group compared to the outpatient group. This may be due to the extensive use of topical fusidic acid in our inpatients suffering from secondary skin infections. Likewise, Shah and Mohanraj [37] in the UK detected high level of fusidic acid resistance to S.aureus isolated from dermatology outpatients 50%, which rose to 78% in inpatients with atopic dermatitis. This incidence was much higher than that detected in the in- and outpatients of other departments, being only 10%. Fusidic acid topical preparations, alone or in combination with topical corticosteroids, have been used in atopic dermatitis patients for prolonged durations, to suppress staphylococci, which colonize eczematous and non-eczematous atopic skin and their toxins play a pivotal role in pathogenesis of the disease [28]. Therefore, different strategies in the treatment of atopic and other dermatologic conditions should be undertaken regarding the prolonged or prophylactic use of antibiotics. They may be substituted by topical antiseptics to reduce the emergence of resistant strains [12].

Outpatients generally showed higher antibiotic sensitivity to Enterobacteriaceae group compared to inpatients, although a statistical significant difference was only observed with levofloxacin and ofloxacin. A statistical difference was also detected with cefaclor, but 57.1% of inpatients were resistant and outpatients were either resistant (50%) or showed intermediate sensitivity (50%). Enterobacteriaceae from both inpatients and outpatients showed complete resistance to ampicillin and piperacillin and most of them were resistant to cephazolin. These findings, apart from ampicillin which is not effective against Gram-negative bacilli, could be attributed to the more use of these antibiotics in our hospital.

El Kholy et al., [19] reported higher susceptibility of Enterobacteriaceae to imipenem (98.2%), ciprofloxacin (>79.2%), ampicillin-sulbactam (38%) and cefazolin (40%) compared to our inpatients. Preboji et al., [34] in Cameroon reported a high incidence of resistance to amoxicillin (85%), piperacillin (75%) and trimethpoprim/sulphamethoxazole (71%) in their inpatients.

In the present study, only four isolates of Pseudomonas aeruginosa were found, although other studies have shown them to be commonly isolated from wounds and chronic venous ulcers [8]. Therefore, statistics could not be done to compare in-/and outpatients as regards the antibiotic sensitivity pattern.

Concerning the ESβL production by Gram-negative bacilli, our study showed that the incidence of ESβL production by Gram-negative bacilli was (29.4%) with no significant difference between the inpatients and the outpatients although it was higher in inpatients (40%) than in outpatients (14.2%). Failure to detect a significant difference despite this apparent difference is because of the small number of isolates from patients. Bouchillon et al. [5], in USA conducted a study including 38 centers from 17 countries to detect ESβL production by nosocomial Enterobacteriaceae and found that the production rate for the combined Enterobacteriaceae was (10.5%), being highest in Egypt (38.5%), coinciding with our study, and in Greece (27.4%). It was lowest in the Netherlands (2%) and Germany (2.6%). However, another study showed a higher incidence of ESβL production by Gram-negative bacilli in Germany (15%) [4].

In conclusion, our current study detected that S.aureus is the most common cause of secondary infection in all skin lesions and was isolated from all body sites with nearly equal prevalence in inpatients and outpatients. The incidence of Enterobacteriaceae infection was more in inpatients with higher levels of ESβL production. Resistance of different bacterial isolates to antibiotics was also higher in inpatients. We, therefore, recommend further studies on large scale to estimate the exact

incidence of the different bacterial organisms implicated in secondary infection of different skin diseases, bacterial culture of specimens from secondary infected skin diseases should be performed to confirm the bacterial etiology and administer the proper treatment, limit the misuse of antimicrobials to prevent the emergence of resistant bacterial strains, antimicrobial susceptibility testing should be considered when prescribing antimicrobial therapy and the estimation of ESβL production by Gram-negative bacilli should be applied to every case of hospital acquired infection to prevent spread of infection by these resistant organisms.

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