Staphylococcal infections among leukemic patients

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Abstract:

Staphylococcus are cause hospital community acquired infection and they are an important cause of health –care associated infection.The Coagulase positive Staphylococcus are *Staphylococcus aureus* which can implicated in toxic shock syndrome. Methicillin and Vancomycin *Staphylococcus aureus* resistant (MRSA, VRSA) become major cause of hospital- acquired infection and community acquired infection.Coagulase negative staphylococcus emerged as major cause of infection in immunocompromised patients.

The main objective of this study was to evaluate the distribution of Staphylococci among leukemic patients since it is well known that leukemic patients are prone to be infected easily due to their immunosuppressed status.

This study was undertaken between oct. 2009 and Jun 2010 at Iraqi center of hematology and medical genetics. 140 clinical specimen(aspirated wound,superficial wound,urine, blood) have deen collected carefully from leukemic patients and subjected to well known established microbiological methods for diagnosis and identification of the isolates .All isolates were tested for their susceptibility to antimicrobials according to Kirby –Bauer technique.

Out of 140 clinical specimen collected from leukemic patients, it was possible to obtain(63) bacterial isolates form which(43) of Coagulase negative staphylococci (CONS) and (20) of Coagulase positive staphylococci. Out of 43(CONS) isolates has been found that *S.epidermidis* constitutes (28)the highest of all isolates. Antimicrobial susceptibility reveald that *S.aureus* is highly sensitive to Gentamycin (85%), Erythromycin (80%), while it is resistant to the drugs Cefotaxim (45%), Choramphenicol(40%),and Tetracycline(20%). *S.epidermidis* show highly sensitive to Erythromycin(100%),Vancomycin (100%), and Cefotaxim(70%) and highly resistant to the drugs Chloromphenicol(45%), Augmentin(45%),Gentamycin (10%), and Tetracycline(10%).

It is concluded that *S.epidemidis* rankes the first(28)among the isolates and S.aureus ranke the 2nd .All isolates were highly resistant to Chloramphenicol and highly sensitive to Erythromycine.

Key words: Staphylococcal infection, (CoNS), leukemic patients.

Introduction:

Staphylococcus are cause of hospital and community acquired infection and they are important cause of health – care associated infections [1,2].The main Coagulase positive Staphylococci is *Staphylococcus auraus* which can survive on dry surfaces increasing the chance of transmission and it is implicated in toxic shock syndrome [3,4].Methicillin and Vancomycin *Staphylococcus aureus* resistant (MRSA, VRSA) become major of hospital –acquired infection and being recognized with increasing frequency in community –acquired infection [5].

S.epidermidis, Coagulase negative staphylococcus species (CoNS) are commensal of the skin but cause

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severe infection in [6,7,8] immunosuppressed patients and those with central venous catheter. *S. saprophyticus,* another Coagulase negative species that is part of the normal vaginal flora and is implicated in genitourinary tract infection in sexually –active young women [9, 10, 11]. Several other Staphylococcus species have been implicated in human infections notably *S.caprae, S.schleiferi* and *S.xylusus* [12, 13, 14]. Coagulase negative Staphylococcui (CoNS) is the leading cause of noscomial bacteremia in most pediatric hospitals [15]. Patients at high risk of infections by (CoNS) are those requiring prolonged intravenous access for chemotherapy or paranteral nutrition because of serious underlying disease [16]. In immunocompromised patients, CoNS have emerged as major cause of infection, [11].The culture of *S.epidermidis* should always be considered potentially hazardous in immunocompromised patients[17].

The aim of our study is to find the incidence of Staphylococci in clinical materials from immunocompromised patients and study their antibiotic sensitivity pattern by using commersal type to put forth the importance of Coagulase negative Staphylococci which are otherwise throught as non_pathogenic;and to help the clinicians choose an effective antibiotic against Staphylococcal infections.

Materials and Methods:

Patients: - one hundred leukemic patients were the source of 140 clinical specimens which were collected from different sites from bodies of patients according to the type of the clinical case.

Methods:

Specimens collection:-Each specimen (aspirated wounds, superficial wounds, urine, 5ml of blood) have been collected carefully and aseptically from leukemic patients suffering from infections following chemotherapy for the purpose of isolation and identification of isolates.

Specimens processing:- Isolates were thoroughly diagnosed according to well established microbiogical methods in which specimen were cultured on blood agar and MacConky. Blood culture of 5ml blood collected aseptically and carefully and cultured on brain heart infusion broth .Specimen on blood agar and brain heart infusion broth cultured in aerobic and anaerobic conditions. Each isolate has been tested by biochemical reactions. Analytic profile index system (API-staph) was followed for differentiation between Coagulase negative staphylococcus species.

Antimicrobial susceptibility test: Kirby-Bauer [18] was applied for detection of susceptibility of the isolates for the commonly used antimicrobial agents. The results of an isolate whether sensitive or resistant were compared to a standard zone of growth inhibition table(1).

Table 1: Interpretation of zone inhibition using Kirby and Bauer method (disc diffusion method.)

Antimicrobial agent	Code	Disc potency Mcg/Disc	Diameter of zone inhibition(mm)		
			Resistant	Intermediate	Sensitive
Ampicillin	AM	10	<=11	12-13	>=20
Cefotaxim	CTX	30	<=14	15-22	>=23
Cephalexin	KF	30	<=14	15-17	>=18
Chloramphenicol	C	30	<=12	13-17	>=18
Ciprofloxcin	C IP	10	<=15	16-20	>=21
Clindamycin	CN	2	<=12	13-17	>=18
Tobremycin	TM	10	<=13	13-14	>=15
Erythromycin	E	15	<=13	14-17	>=18
Ampiclox	AMP	30	<=14	15-16	>=17
Gentamycin	GN	10	<=12	13-14	>=15
Nalidixic acid	NAL	30	<=13	14-18	>=19
Pencillin-G	PG	6	<=20	21-28	>=29
Rifampicin	RA	5	<=16	17-19	>=20
Co-Trimoxazole	SXT	25	<=18	19-2	24-32
Amoxicillin	AMX	10	<=19	-	>=29
Amikacin	AN	30	<=14	15-16	>=17

Results:

This work resulted in obtaining 140 clinical specimens collected from different body sites of 100 leukemic patients. Out of the 140 specimen it was possible to obtain (63)well diagnosed microorganism from which (43)isolates were (CoNS) while *S.aureus* which was Coagulase positive were (20)isolates.

Concerning the site of sample collection as in table(2) it was seen that superficial wounds yielded the highest collection i.e (18) while aspirated wounds and urine were almost equal in yielding isolates in which (16) form aspirated wounds and (17) isolates form urine. blood revealed the lowest (12) isolates as compared to the other three sites of specimens collection.

Table 2: Distribution of isolates according to site of collection

Site of specimens	No. of patients	No. isolates	*S.aureus*	*S.epidermidis*	*S.chromogenes*	*S.xylusus*	*S.caprae*
Aspirated wound	25	16(25.4%)	9	7	-	-	-
Urine	25	17(27%)	5	8	1	1	2
Superficial wound	30	18(28%)	3	8	3	1	3
Blood	20	12(19%)	3	5	-	2	2
Total	100	63	20	28	4	4	7

Our results in **table(2)** showed that the 20 isolates of *S.aureus* have been distributed according to their collection in which(9)isolates form aspirated wounds, while (5)isolates form urine ,and(3)isolates form each of superficial wound and blood respectively. Out of 43 isolates of (CoNS). It has been found that *S.epidermidis* ranks the highest (28)from which(7)isolates from aspirated wounds and (8)isolates from each of urine and superficial wounds respectively, while blood yielded (5)isolates.

The other (CoNS) which were the least among the isolates in which no isolates of *S.chromagenes* , *S.ylusus*, and *S.caprae* from aspirated wound while urine yielded isolates of each *S. chromogenes*, and *S.ylusus* respectively, and (2) isolates of *S.caprae*. Superficial wounds yielded (7) isolates of (CoNS) other than

S.epidermidis from which (3)isolates of each of *S.chromogenes* and *S.caprae* while one isolate of *S.xylusus*.

Blood samples (12)were free from *S.chromogenes* while *S.xylusus* and *S.caprae* were (2)isolates of each, respectively, and (5) isolates of *S.epidermidis*.

Antimicrobial susceptibility test:-

Detection of whether an isolate sensitive or resistant was through a comparison between the resultant zone of growth inhibition with those of standard of zone of growth inhibition as in table (1). Well known technique [18] was applied to detect the susceptibility of the isolates to the commonly used antimicrobials. Table (3) shows that *S.aureus* is highly sensitive to Gentamycin(85%) , and Erythromycin(80%), while it is resistant to each of the drugs Cefotaxim(45%), Chloramphenicol (40%) and Tetracycline (20%)and intermediate to Augmentin (60%) and Vancomycin (50%). The results showed that the isolates *S.epidermidis*, *S.chromogenes* and S.*xylusus* are highly sensitive toVancomycin and Erythromycin while *S.caprae* is less sensitive to Vancomycin(60%) and *S.chromogenes* is resistant to Erythromycin (50%). Also the results showed that *S.epidermidis* *S.chromogenes* ,and *S.xylusus* are highly resistant to each of the drugs Gentamycin and Tetracycline except *S.caprae* which showed moderate sensitively (75%)to Tetracycline .All isolates of (CoNS) and *S.aureus* are highly resistant to the drug chloramphenicol ranging from(40% to50%) and all isolates showed variability in their susceptibility to Cefotaxime and Augmentin.

Table 3: The susceptibility of the Staphylococcal isolates to antimicrobial agents in percentage

Bacterial isolates	E	VA	CE	GM	T	C	AMC
S.aureus	80	50	45	85	20	40	60
S.epidermidis	100	100	70	10	10	45	45
S.chromogenes	50	100	100	Zero	Zero	50	50
S.xylusus	100	100	50	Zero	50	50	60
S.caprae	85	60	60	zero	75	50	40

E=Erythromycine, VA=Vancomycine, CE=Cefotaxime, GM=Gentamycine,T=Tetracycline, C=Chloramphenicol, **AMC=Augmentin**.

Discussion:

The results of this study clearly show that out of (43) (CoNS) isolates, *S.epidermidis* was (28)which is the highest among the bacterial isolates. These results are fully agreed with other (10) who proved that most isolates from clinical samples *S.epidermidis* specially from urine and blood. It is seen from table(2) that *S.epidermidis* represent (8)isolates from each of urine and superficial wounds respectively since *S.epidermidis* is mostly carried as skin commensals in addition to that it is pathogenic role [10, 11,12].

Isolation of *S.aureus* or *S.epidermidis* from the blood is a hazardous condition since a positive blood culture yielding *S.epidermidis* should always be considered partially hazardous in immunocompromised patients[13].To obtain pure culture and to role out the growth of the suspected contaminant of (CoNS) in blood cultures, the specimen were incubated up to 48h before reading the results since it has been mentioned by several workers [13, 14, 15] that pathogenic bacteria grow in a shorter time than contaminants in blood culture. The least of the isolated (CoNS) from which no isolates have been obtained include *S.chromogenes* ,*S.xylusus* and *S.caprae* from aspirated wound .Even if the rate of isolates of (CoNS) other than *S.epidermidis* from the other sites in this work is low, its pathogenic role

is not excluded since it has been supported by others [16,17,19]who claimed that there are number of recent reports which states that (CoNS) are the most common pathogen in blood stream and urinary tract infection in immunocopmromised patients. Another important isolates in this study is *S.aurues* which rank the 2nd 20 among the isolates a result which agree with other [20, 21] who found that *S.aureus* ranks the 2nd in their isolates.The incidence of CoNS and CoPS was related to multiple factors such as decreased granulocyte(phagocyte) number or function ;decreased lymphocyte number or function; defects in mechanical barrier to colonization and infection; and contact or exposure to pathpgenic organisms. Inaddition to leukemia itself affect the immune system and residue of normal cells due to exposed to aggressive chemotherapy [21, 22].

The pattern of antimicrobial susceptibilities in our work revealed that. *S.aureus* is sensitive to Gentamycin and Erythromycin ;and highly resistant to the drugs Cefotaxim, Chloramphenicol ,and to Tetracycline while it is intermediate to Vancomycin a result which disagree with other [17] who found that *S.aureus* is highly sensitive to Vancomycin, this clearly demonstrate the acquisition of drug resistance through plasmids or conjunction.

The results revealed that (CoNS) were highly sensitive to the drugs Vancomycin and Erythromycin respectively except *S.caprae* showed intermediate to Vancomycin , and *S.chromogenes* showed resistante to Erythromycin. All (CoNS) resistante to Gentamycin,Tetracycline,and Chloramphenicol except S.caprae showed resistante to Tetracycline and to Chloramphenicol but all isolates variable in their susceptibilities against Cefotaxim and Augmentin.

It cannt be advised that Vancomycin is the drug of choice in infections caused by CoNS and CoPS due to it's a poteintial toxic drug and has long term side effect since it s very effective against staph infection by inhibiting synthesis of bacterial cell wall. These effects include to hearing loss, abnormal liver function, nephrotoxic, and affect white blood cell production to include leukopenia and eosinophillia which can make patients more susceptible to another dangerous side effect of Vancomycin use called superinfections. Erythtomycin reveald good effect, it kills bacteria by blooking their protein synthesis. In conclusion, our study revealed that (CoNS) cannt be neglected as commensals since they have been isolated in significant number, also variability in the antibiotic susceptibility pattern of (CoNS) reflects the different protocols and panels of antibiotics being used in different hospitals, and difference in gegraphical location from were these isolates have been obtained ,as a aresult of that, it may be essential to determine its species and antibiotics sensitivity as no practicular pattern can be predict in any problematic situation.

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اصابات البكترية العنقودية في مرضى سرطان الدم

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الخلاصة:

تمهيد:المكورات العنقودية هي السبب في الالتهابات التي تحدث في المستشفيات و المجتمع.العنقوديات الذهبية المقاومة للمشليين والفانكومايسيين اصبحت لها الدور الرئيسي في الالتهابات المكتسبة في المستشفيات والمجتمع. اما العنقوديات السالبة لفحص الكواكيولييز فهي تكافايية على الجلد ولكن تسبب التهاب شديد لمرض نقص المناعة.

الهدف:تقييم انتشار العنقوديات عند مرضى سرطان الدم لكون المصابيين بهذا المرض يكون الجهاز المناعي لديهم مثبط ولذا فهم عرضة للاصابة بمختلف الجراثيم.

طريقة البحث:هذه الدراسة اجريت للفترة من الاول من تشرين الثاني 2009 الى نهاية حزيران2010 في المركز العراقي لامراض الدم والوراثة الطبية .كل عينة تم جمعها من مصادرمختلفة من جسم مرضى سرطان الدم ،وتم تشخيص مكوناتها الجرثومية بالطرق العملية المعروفة لتشخيص الجراثيم .كل العزلات خضعت لاجراء فحص الحساسية للمضادات الحيوية.

النتائج:من مجموع 140 عينة تم جمعها من مرضى سرطان الدم، تم الحصول على 63عزلة بكتيرية مشخصة منها 43عزلة تمثل العنقودية السالبة لفحص Coagulase و20 عزلة تمثل العنقودية الموجبة لفحص Coagulase. من مجموع 43 عزلة سالبة لفحص الCoagulase تم الحصول على 28 عزلة وهي البشروية والتي تشكل اعلى نسبة من العنقوديات سالبة لفحص Coagulase.فحص حساسية الجراثيم للمضادات الحيوية اظهرت ان العنقودية الذهبية عالية الحساسية للجنتامايسين (85%) والارثرومايسيين(80%) ،بينما اظهرت مقاومة شديدة للفانكومايسيين(50%)والسيفوتاكسام(45%)والكلورامفينيكول(40%) والتتراسايكليين(20%) في حين ان البشروية اظهرت حساسية عالية جدا لكل من الارثرومايسيين(100%) والفانكومايسيين(100%) والسيفوتاكسام(70%) بينما اظهرت مقاومة شديدة جدا لكل من الكلورامفينيكول(45%)والاوكمنتين(54%)و الجنتامايسيين(10%)والتتراسايكليين(10%).

الاستنتاج:يستخلص من البحث ان العنقوديات البشروية تمثل اعلى نسبة 28من بين كل العزلات وان العنقودية الذهبية تمثل النسبةالثانية(20)،وكل العزلات اظهرت مقاومة شديدة للكلورامفينيكول وحساسية عالية للارثرومايسين.