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A Comprehensive Overview of *Staphylococcus aureus* Isolates from Cancer Patients

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Abstract
Staphylococcus aureus (S.aureus) a gram positive commonly isolated pathogen is known for multi drug resistance and nosocomial infection. A 3 year review study of S.aureus isolated from clinical samples of cancer patients (lymphohaematological malignancies and solid tumors) was done in the department of microbiology in a government referral cancer hospital in bangalore, South India from January 2014 to December 2016.The study was to know the isolation percentage of S.aureus in cancer patients and to identify the most common gram positive pathogen causing infection in cancer patients. A review on S.aureus infections in cancer patients done for the first time in South India.

Materials and Methods: *A meticulous survey of clinical samples comprising of blood, urine, e.n.t (ear, nose, throat), sputum, pus from cancer patients including all clinical oncology services was reviewed in microbiology laboratory. Bacterial culture was processed by conventional method and antibiotic susceptibility testing performed by the Stokes's disc diffusion technique as per Clinical and Laboratory Standards Institute (CLSI) recommendations.*

Results: *The total number of growth from all clinical samples was blood 7441(15.4%), pus 929(32.8%), sputum 2480(17.8%),urine2357(13.4%)and e.n.t 1552(24.8%) of which gram positive pathogens from pus 773(32.8%), sputum 266(17.8%), e.n.t 250(24.8%), blood 230(15.4%)and urine 829(13.4%). Amongst which S.aureus was isolated from pus187 (24.2%), urine 75(91.5%), sputum 74(27.8%), blood 64(27.8%), and e.n.t 52(20.8%). MRSA from pus, e.n.t, sputuzm, blood, urine was 32.8%,24.8%17.8% 15.4%, and 13.8% respectively. Other gram positive isolates were enterococci, β-steptococci, co-agulase negative staphylococci, streptococcus pneumonia and aerococcus.*

Conclusion: *S.aureus was the most common gram positive bacteria isolated and gram negative infections was more common than gram positive infections as analysed from infections in cancer patients at our regional referral hospital. Demographic and epidemiological changing pattern of bacterial strains in cancer patients need to be updated frequently.*

Keywords: *Gram positive pathogen, Staphylococcus aureus (S.aureus), Cancer, MRSA.*

Introduction

Staphylococcus genus is a heterogeneous group of bacteria consisting of 30 species of which *S.aureus* has been found to be the most clinically important species and since its discovery during the 1880s *S.aureus* has emerged as a potential

pathogenic gram-positive bacterial species that cause various infections such as bacteraemia, infective endocarditis, sepsis, toxic shock syndrome, and skin and soft tissue infections.[1] and also known for nosocomial infections with a high propensity for methicillin resistance called Methicillin Resistant S*taphylococcus Aureus (*MRSA). Many organisms causing catheter-related infections are predominantly those that colonize human skin and approximately 70–80% are gram-positive with *Staphylococcus* species as the most isolated[2]. *S.aureus* generally are opportunistic pathogens or commensals on host skin and may act as pathogens on entry into the host tissue through a trauma to the cutaneous barrier, inoculation by needles, the implantation of medical devices or in cases in which the microbial community is disturbed or in immunocomp-romised individuals[3,4,5] *S.aureus* infections in cancer and non-cancerous patients vary primarily due to the underlying immune status and to multiple risk factors which is exaggerated in cancer patients and *S.aureus* infections in cancer patients can be more complicated considering the rigorous chemotherapy, radiotherapy and to surgical interventions. The ability of *S.aureus* to produce metastatic or secondary infections such as endocarditis, osteomyelitis, and septic arthritis has been documented in few studies[6,7,8,9]. while it is observed that infectious complications are a serious cause of morbidity and mortality in cancer patients especially those with underlying hematological malignancies where autopsy studies demonstrate that approximately 60 % of deaths are infection related[10,11,12,13,14,15,16]. Fewer data exist on infectious mortality in patients and in solid organ tumors approximately 50 % of these patients are estimated to have an infection as either the primary or an associated cause of death [12,14,15,16,17,18] Bacterial infections predominate during the early phases of a neutropenic episode, whereas fungal infections occur more often in patients with prolonged neutropenia[19]

A study of infections in solid tumors done by Kenneth V[20] et al mention that several factors increase the risk of infection in patients with solid tumors and the presence of multiple risk factors in the same patient is not uncommon. These include obstruction (most often caused by progression of the tumor), disruption of natural anatomic barriers such as the skin and mucosal surfaces, and treatment-related factors such as chemotherapy, radiation, diagnostic and/or therapeutic surgical procedures and the increasing use of medical devices such as various catheters, stents, and prostheses.[2] While Susan. N et al in their study on hematologic malignancies state that one of the most common complications involved in treating patients with hematologic cancer is infection.[21] requiring prompt treatment. Risk factors such as break in mucosal barriers, pressure sores [22], in-situ catheters, prolonged hospital stay, over use of multi antibiotics, underlying disorders like diabetes, cancer increase the chances of infection with *S.aureus*. Identifying risk factors in haematological and solid tumors can prevent infections, health care costs and decreasing morbidity and mortality rates. The most important risk factor for infection with resistant pathogens is prior colonization or infection by resistant organisms due to unnecessary use of multiple higher broad spectrum antibiotics.

Surgery, medical procedures, radiation therapy and the widespread and increasing use of catheters and other devices is often associated with the development of infection[23]and may contribute as risk factors during pre or post interventions in immunocompromised patients.

S.aureus infection arises due to a breech in skin or mucosal barrier and in cancer patients additional external factors such as chemotherapy and radiotherapy cause denudation of mucosal barriers implicating more reasons for early infections. Infections in cancer patients can be minimized considering the risk factors and advocating the right antibiotic protocol at the right time and since most bacteria carry multiple resistance genes against commonly used antibiotics, they show multiple antibiotic resistance patterns thereby giving rise to treatment problems.[24]

S.aureus is predominantly the commonest isolate which most often give rise to secondary bacterial infection depending on the site of infection. Prompt treatment of monobacterial *S.aureus* infections in cancer need to be evaluated to prevent polybacterial *S.aureus* infections requiring alternate antibiotics which inherently lead to MRSA and with underlying malignancy MRSA can become the prime risk factor for secondary and polymicrbial infections. As infections in cancer patients may present at any point of time relevant criteria becomes crucial to avoid recurences such as depressed immune system, multiple eitiological factors due to underlying disease, counts status, presence or absence of chemotherapy, co-existing secondary infections and dual malignancies and rigourous external anti-cancer treatment. Antibiotic policy and addressal of relevant cancer associated risk factors may reduce the burden of infection.

The role of *S.aureus* toxins in cancerous infections may be considered as a hidden potential to exaggerate the status of infection.

Materials and Methods

Culture positive clinical samples from oncology patients of both haematological malignancies and solid tumors were subjected to bacterial culture. Blood culture was processed by BACTEC (Biomeriux), while pus, e.n.t, sputum, urine were processed by conventional culture methods.

Inclusion criteria were

1. Clinical samples from in-patient and outpatient.
2. Neutropenic and non-neutropenic status of patients
3. Patients on antibiotics
4. Haematolymphoid malignancies and solid tumors
5. Patients receiving chemotherapy, radioth-erapy chemotherapy and radiotherapy.

Exclusion criteria

1. Non cancer patients
2. Age below 5 years and above 70 years.
3. Surveillance cultures.

We analyzed all samples to know the true infection of *S.aureus* irrespective of risk and co factors of malignancy in our patients.

Cultures were subjected to morphological identification and bio typing to identify different bacterial isolates according to standard microbial-ogical methods. *S.aureus* was phenotypically identified on the basis of culture characteristic, morphology and biochemical tests[25]

Detection of MRSA was done phenotypically employing cefotoxin antibiotic disc.

Clinical samples were processed by conventional method and antibiotic sensitivity test by Stoke's method as per CLSI guidelines.[26] with reference strain *S. aureus* ATCC 29213 was used. Fungal growth was not included in the study. This is a review study of a large sample size therefore molecular work could not done. The study is a retrospective review data that involved no diagnostic, therapeutic or intervention and as there was no direct patient contact hence informed patient consent was not required as per our institutional review board.

Statistical Analysis

Chi-square test was used to analyze for the frequency data with 5% level of significance the respective p-value is given in the tables.

Results:

All clinical samples from cancer patients were analysed to get the true estimate of *S.aureus* infections. Pus had the highest number of culture positivity. All urine samples yielded *S.aureus* isolation but there was no MRSA. While least yield of *S.aureus* was in e.n.t sample

The results were: Total blood cultures 7441; pus 2929; sputum 2480; urine 2357 ent 1552. Total number of all clinical samples and growth positivity shown in table 1. Total gram positive isolates from all samples depicted in Table 2. and of which *S.aureus* was 64(27.8%), 187(24.2%), 74(27.8%), 75(91.5%) 52(20.8%) respectively.

Maximum resistance of *S.aureus* was to ciprofloxacin in all samples except urine where

the maximum resistance was to rifampicin and vancomycin. Vancomycin resistance was to all urine samples7 (100%) followed by pus15(4.4%) 15(4.4%) sputum 6(5%),e.n.t6(4.3%)blood sample 1(1.3%); Less sensitivity to erythromycin and sulhpamethaxazole was observed in blood and sputum while *S.aureus* was more sensitivity to gentamicin and rifampicin except urine as in Table III. Rifampicin is used when a multi drug resistant *S.aureus* infection is present after ruling out necessary risk factors. Gram positive pathogens isolated were *enterococci, β-steptococci, co-agulase negative staphylococci, streptococcus pneumonia and aerococcus.* Other gram positive isolates were 46.5%, 67.2%, 58.6%, 66.55%, 8.5% from all samples respectively.

MRSA from pus, e.n.t, sputum, blood, urine was 32.8%, 24.8%17.8% 15.4%, and 13.8% respectively. MRSA isolation was highest in blood and nil in urine sample. Chi square could not be analyzed for urine sample due to less sample size.

A comparative isolation of *S.aureus* and MRSA is depicted in Table 111.

Solid tumors were 98.7%while lymphoroliferative and hematological malignancies were 1.3%.

The low incidence of haematological malignancies were mostly from the paediatric population.

Fig 1: Percentage of Gram Positive Isolates

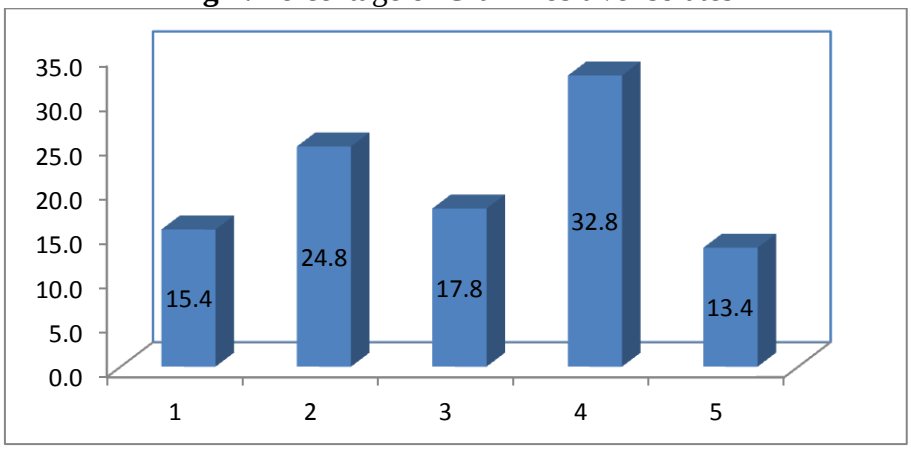

Table No I: Number and Percentage of Culture

Culture	Total	Growth positivity	Percentage
Blood	7441	1489	20.0%
ENT	1552	1008	64.9%
Sputum	2480	1493	60.2%
Pus	2929	2357	80.5%
Urine	2357	612	26.0%

Table II: No and Percentages of Staphylococcus aureus

Culture	Gram Positive	Staph aureus		MRSA		Other GPC		P-value
		#	%	#	%	#	%	
Blood	230	64	27.8	59	25.7	107	46.5	0.598
ENT	250	52	20.8	30	12.0	168	67.2	0.008
Sputum	266	74	27.8	36	13.5	156	58.6	0.002
Pus	773	187	24.2	172	22.3	514	66.5	0.366
Urine	82	75	91.5	0	0.0	7	8.5	0.000

GPC=Gram Positive cocci

Table III: Sensitive and Resistance Pattern of *Staphylococcus aureus*

	SENSITIVE		RESISTANT		Total	P-Value
Blood	#	%	#	%		
Ciprofloxacin	34	43.6	44	56.4	78	0.110
Erythromycin	40	51.3	38	48.7	78	0.749
Gentamicin	67	85.9	11	14.1	78	0.000
Levofloxacin	70	89.7	8	10.3	78	0.000
Rifampicin	71	91.0	7	9.0	78	0.000
Sulphamethaxazole	54	69.2	24	30.8	78	0.000
Vancomycin	77	98.7	1	1.3	78	0.000
E.N.T						
Ciprofloxacin	28	20.3	110	79.7	138	0.000
Erythromycin	107	77.5	31	22.5	138	0.000
Gentamicin	130	94.2	8	5.8	138	0.000
Levofloxacin	134	97.1	4	2.9	138	0.000
Rifampicin	136	98.6	2	1.4	138	0.000
Sulphamethaxazole	120	87.0	18	13.0	138	0.000
Vancomycin	132	95.7	6	4.3	138	0.000
Sputum						
Ciprofloxacin	27	22.5	93	77.5	120	0.000
Erythromycin	76	63.3	44	36.7	120	0.000
Gentamicin	118	98.3	2	1.7	120	0.000
Levofloxacin	116	96.7	4	3.3	120	0.000
Rifampicin	117	97.5	3	2.5	120	0.000
Sulphamethaxazole	99	82.5	21	17.5	120	0.000
Vancomycin	114	95.0	6	5.0	120	0.000
PUS						
Ciprofloxacin	75	21.9	267	78.1	342	0.000
Erythromycin	211	61.7	131	38.3	342	0.000
Gentamicin	311	90.9	31	9.1	342	0.000
Levofloxacin	330	96.5	12	3.5	342	0.000
Rifampicin	325	95.0	17	5.0	342	0.000
Sulphamethaxazole	270	78.9	72	21.1	342	0.000
Vancomycin	327	95.6	15	4.4	342	0.000
Urine						
Ciprofloxacin	2	28.6	5	71.4	7	
Erythromycin	3	42.9	4	57.1	7	
Gentamicin	5	71.4	2	28.6	7	
Levofloxacin	4	57.1	3	42.9	7	
Rifampicin	0	0.0	7	100.0	7	
Sulphamethaxazole	5	71.4	2	28.6	7	
Vancomycin	0	0.0	7	100.0	7	

Discussion

Gram negative infections predominated over gram positive infections from our patients. The source of *S.aureus* infections as reviewed are from patient's own endogenous microflora particularly from those residing on the mucosal surfaces of the mouth and gastrointestinal tract unlike *S.aureus* infections from non-cancerous patients who get infected from hospital environment and community acquired infections as reported from numerous studies. The prevalence of *S.aureus* as a cause of infection in cancer patients varies widely depending on the specific population,the type of infection studied and geographic location and observed that *S.aureus* has a major clinical impact on patients with malignancy.[27] Changing demographic strains may have an implication in the pattern of infection from the

immuncompromised. Studies from Montassier E et al report isolation rate of *S.aureus* was 28% in their cancer patients. [28] The data analysis of more isolation of gram-negative bacilli than gram-positive cocci from our region does not comply with studies in the west as reported by several authors. A shift of pathogenic microorganism from gram-negative bacilli to gram-positive cocci which now account for 3 of every 4 bacteremic isolates in patients with hematological malignancies.[29,30,31,32,33,34,]

Treatment for *S.aureus* infections with routine first line of antibiotics is implemented for *S.aureus* and vancomycin and is part of the antibiotic protocol towards our cancer patients.

Inherent risk factors like obesity, diabetes type II predispose to infections in patients with decreased immune status and underlying malignancy. Interestingly cancer patients have multiple predisposing factors that increase the risk of infection such as chemotherapy, radiation therapy, surgery, stem cell transplantation, bone marrow transplantation or steroids in addition to suppressed immune system. Cuervo SI [35] et al in their review found that the risk factors in cancer patients do not differ considerably from non-cancerous patients. Mucosal barriers due to intervention of anti malignant chemotherapy, radiotherapy and progressive neoplastic disease predispose to risk factor. A detailed cohort study on essential risk factors is required to prevent common infections with major pathogens like *S.aureus and Escherichia coli (E.coli)* which can effect a radical change by decreasing morbidity and mortality to a great extent in cancer patients.

Our results correlate and are similar to studies done by other cancer centers in our country and one study from middleeast. [36,37,38,39,40] Unlike most western countries report the rate of isolation of gram positive infections more common than gram negative infections in cancer patients[41]

The leading cause of invasive bacterial disease in cancer patients are broad range of gram-positive bacteria causing serious infections with the greatest burden of disease being due to staphylococci, streptococci, and enterococci. [42]

The variant epidemeological strains need to be considered in a cancer patients while *S.aureus* still remains as the potential gram positive pathogen causing infection isolated amomgst bacteria in our cancer patients. Kang CI[43] et al study mention that S. aureus infections in cancer patients are serious clinical conditions with high mortality rates, even in non-neutropenic patients. Co-factors and underlying physiological imbalance in an immunosuppresion milieu compound to infection in cancer patients. Most common pathogen like *S.aureus* infections in cancer patients can affect treating modality despite chemotherapy protocols.

Conclusion:

Gram negative pathogen infections is more common than gram positive pathogen infections from our cancer patients at our cancer referral hospital in south India. Infections in cancer patients is challenging to specific population and demographic locations and with changing epidemeological strains, predictable risk factors, anti malignant chemotherapy and antibiotic protocols a successful treatment outcome in cancer patient can be a reality.

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