Antimicrobial Susceptibility Patterns Of Blood Culture Isolates From Cancer Patients After Anti-Cancer Therapy.

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Abstract: Blood stream infections (BSI) have a major impact on cancer patients. Antimicrobial patterns in pathogens should be analyzed by routine surveillance since they vary among different healthcare facilities and geographical area. This study focuses on epidemiology, clinical features, and antimicrobial profiles of cancer patients with BSI after anticancer therapy. A Descriptive cross-sectional study was conducted within a period of 4 months. The causative agents and antibiotic profiles were studied according to Clinical Laboratory Standards institutes (CLSI) and VITEK automated system. Antibiotic profiles of organisms and antibiotic resistance patterns were analyzed in terms of frequency. The prevalence of BSI was 11.75%. Gram negative bacilli (GNB) were higher than gram positive cocci (GPC). Among GNB the most prevalent was *Klebsiella pneumoniae* and among GPC it was, *Staphylococcus aureus*. Prevalence of multi-drug resistance (MDR) was 78.8%. High resistance to Erythromycin was seen in patients with hematological cancers while high resistance to Benzyl penicillin was seen in patients with solid cancers. Chemotherapy had no significant impact on presence of multi-drug resistance while the association between CRP with ANC and presence of fever was not significant. Among different antibiotic resistant groups, ESBL and Carbapenem resistances were noted. According to our study high prevalence of antimicrobial resistance in Gram-negative isolates and emergence of MDR pathogens is alarming. Investigation of novel antibiotics, usage of combination therapy and usage according to the standard antimicrobial susceptibility testing may help to decrease or prevent the emergence of antibiotic resistance.

Keywords: - Cancer, Blood-stream infections, Antimicrobial resistance.

Introduction:

Blood stream infections (BSI) have a significant impact on cancer patients. It is the most common complication seen in patients with hematological malignancies such as lymphomas, leukemia, multiple myeloma and febrile neutropenia (Trecarichi *et al*., 2009) and in patients with oncological malignancies such as solid tumors. The prevalence of BSI lies between 11 to 38% while mortality reaches 40% (Montassier *et al*., 2013).

Cancer patients have a high risk of infection due to prolong neutropenic conditions, damages caused due to cytotoxic agents and altered gut flora due to antibiotic usage (Saghir *et al*.,2009). Apart from surgical operations, radiation therapy, myelo-suppressive cytotoxic chemotherapy, frequent use of invasive procedures such as central venous catheters and immunosuppression status caused by the

malignancy itself will make the patients more vulnerable to BSI.(Marin *et al*.,2014, Rolston,2017). Previous studies have mostly focused on BSI due to hematological malignancies, but patients with solid cancers that undergo surgery too possess a high potential in acquiring BSI. It could be acquired endogenously such as normal flora near operative sites or exogenously via hospital environment such as air, medical staff and equipment (Homsi *et al*.,2000, Nurain *et al*.,2015).

The development of BSI hinders the ongoing anticancer therapies such as chemotherapy and radiation therapy by delaying its administration and by reducing the dosage that can be applied. This will also lead to lengthening of hospital stay (Marin *et al*.,2014, Montassier *et al*.,2013) and increase costs related to patient care while significantly increasing morbidity and mortality (Rani *et al*.,2017).

A previous study (Chandrasiri *et al*.,2013) carried out in Sri Lanka stated that adult leukemia was the most frequent (15%) clinical condition that supported BSI while contribution of solid tumor was 9.7%. Centers for disease control and prevention (CDC) in USA has estimated that out of the patients who undergo chemotherapy about 10% are subjected to infections (Telliant *et al*.,2015).

Fever is considered as the principle indicator and sometimes the only clinical presentation of BSI. But it may also be shown as part of the flu like syndrome in cancer patients receiving chemical and biological therapy. Therefore, attention should be given for cancer patients with fever since it serves as an indicator of early diagnosis (Nejad *et al*.,2010). CRP is a statistically significant predictor for BSI in adults (Al-Mulla *et al*.,2014). Neutropenia refers to Absolute Neutrophil Count (ANC) less than 500 cells/mm3 and it also plays a significant role in BSI. It is considered as a risk factor in up to 25% of cancer patients with death rates extending to 24% in high income countries and 33% in low middle-income countries (Lubwama *et al*., 2019). All these clinical data play a significant role as indicators of BSI.

Recent studies show that gram negative bacilli (GNB) are common in BSI in cancer patients during aggressive therapy (Saghir *et al*.,2009). According to (Marin *et al*.,2014) the shift from gram positive to gram negative is dependent on the geographical area. In US Latin America gram negatives have been found to be frequent while in Europe it was gram positive. When considering gram positive bacteria Genus staphylococcus were more frequently isolated while *Klebsiella pneumoniae* , *E.coli* and *Pseudomonas aeruginosa* were isolated as gram negative bacilli (Fentie *et al*.,2018).

Antimicrobial patterns in pathogens should be analyzed by routine surveillance since they vary among different healthcare facilities and geographical area and also due to the increase of Antimicrobial resistance (AMR) in bacterial agents. BSI requires immediate antibiotic treatment. Empiric antibiotic therapy is used as the standard practice until culture results are available. This Empiric therapy is developed based on knowledge of institution specific patterns of microbial prevalence and resistances. Thus, such studies are required to assist antimicrobial therapy and control of infections at different institutions. (Lubwama *et al*.,2019). According to previous studies information on BSI in solid cancer patients is scarce and comparative studies are also limited (Marin *et al*.,2014). Therefore, this study focuses on epidemiology, associations between clinical features, causative organisms of BSI, antimicrobial resistance between two populations namely oncological and hematological malignancies of cancer patients with BSI and the association

between chemotherapy and antimicrobial resistance.

Methodology:

A descriptive Cross-sectional prospective study was conducted at National cancer institute Maharagama from August 2019 to December 2019. Ethical clearance was obtained from the Ethical review committee, Faculty of Medicine, General Sir John Kotelawala Defense University, Rathmalana and Informed consent was taken from National Cancer Institute, Maharagama. The total number of 309 positive blood cultures that met the inclusion criteria was examined.

Information on age, sex, clinical data, (White Blood Count, Absolute Neutrophil Count, C-Reactive Protein, Fever, Anticancer therapy, Antibiotics given prior to culture and blood collection site for each patient was recorded.

Samples sent to the Microbiology laboratory for routine diagnosis were used for the project. All positive blood culture samples were detected by BD BACTEC™ FX Automated blood culture analyzer. Microbial identification was performed using the biochemical tests following Laboratory Manual in Microbiology, 2011 and VITEK 2 compact automated system. Hematological reports were assorted under hospital permission. Full blood count was done using Sysmex XN - 1000 hematology analyzer and Absolute neutrophil count was obtained from full blood count report. Nine antibiotic classes named Penicillin, Aminoglycosides, Cephalosporins, Carbapenems, Fluoroquinolones, Macrolides, Lincosamides, Glycopeptides, Beta-lactam inhibitors were considered in this study.

Antibiotic susceptibility was tested using disc diffusion method following Clinical Laboratory Standard Institute (CLSI) guidelines and VITEK 2 – compact automated system. According to the CLSI recommendations, the screening of Methicillin- Resistant Staphylococcus aureus (MRSA), ESBL production, Vancomycin Resistant Enterococci (VRE) and Carbapenem Resistant Enterobacteriaceae (CRE) was conducted. In this study we considered Carbapenem resistant Enterobacteriaceae (CRE), extended spectrum beta lactamase (ESBL) regarding gram negative isolates and Methicillin Resistant Staphylococcus aureus (MRSA) and Vancomycin Resistant Enterococci/Staphylococci regarding gram positive isolates.

All data was analyzed using descriptive and inferential statistics by IBM SPSS Statistics 20 software. Antibiotic profiles of organisms and antibiotic resistance patterns were analyzed in terms of Frequency. Correlation between CRP-Fever, ANC–CRP, & MDR organisms and No. Of chemotherapy drugs were analyzed respectively by using Regression, Pearson correlation & Cochran's Mantel – Haenzel test respectively.

Results:

The prevalence of Blood stream infections (BSI) among cancer patients was 11.75%. The number of positive blood cultures examined in this study was 309. The study population consisted of patients aging from 2 months to 82 years. The majority of BSI occurred in hematological malignancies (57.9%) while BSI in patients with solid tumors were lower (42.1%).

An extended range of 41 species of causative agents were isolated. Majority of the organisms isolated were gram negative bacilli (60.1%) out of which *Klebsiella pneumoniae* (n=41, 12.5%) was dominant followed by, *Escherichia coli* (n=38, 11.6%), *Acinetobacter baumannii* (n=19, 5.8%) 37.8% gram positive cocci and 2.1% gram positive bacilli were also isolated. Predominant gram positive cocci were *Staphylococcus aureus* (n=28, 8.5%) followed by *Staphylococcus hominis* (n=18, 5.5%), Coagulase negative *Staphylococcus* (n=17,

5.2%) and *Staphylococcus hemolyticus* (n=15, 4.6%). The prevalence of poly-microbial bacteremia was 6.1% (n= 19). Some of the rare species named *Achromobater xylosoxidans*, *Bordetella hinzii*, *Burkholderia cepacia*, *Elizabethkingia meningoseptica*, *Alcologens faecalis*, *Ochrobactrum anthropic* were also isolated.

Gram negative organisms were predominant over gram positive organisms among both solid and hematological malignancies. In our study, antimicrobial resistance was high in hematological malignancies than solid tumors. Among hematological malignancy patients, GNB's such as *Enterobacter cloacae*, *Acinetobacter baumannii*, group named as LF other(Lactose fermenting),

Escherichia coli and *Klebsiella pneumoniae* and GPC's such as *Staphylococcus hemolyticus*, *Staphylococcus hominis*, *Streptococcus spp*. and Diptheroids showed high rate of resistance to most of the antibiotics. Among solid tumor patients, GNB's such as *Escherichia coli*, *Klebsiella pneumoniae* and group named as LF other and GPC's such as Coagulase Negative Staphylococcus, *Staphylococcus spp*. and *Staphylococcus hemolyticus* showed high rate of resistance to most of the antibiotics.

In this study, for gram negative organisms, Extended spectrum beta-lactamase (ESBL), Carbapenem resistant Enterobacteriaceae (CRE) and for gram positive organisms, Methicillin resistant *Staphylococcus aureus* (MRSA), Vancomycin resistant Enterococci (VRE) were considered as antimicrobial resistance patterns. Overall, 28.57% gram negative organisms were positive for ESBL. Rate of occurrence of ESBL was tested in *Klebsiella pneumoniae* (24.4%) and *Escherichia coli* (34.5%). But both were less than 50%. CRE was considered with regard to Imipenem and Meropenem. The highest resistance to Imipenem was showed by *Enterobacter spp*. (53.3%) while *Klebsiella pneumoniae* showed 73.2% against Meropenem. When considering VRE, *Enterococci spp*. showed no resistance to Vancomycin (100% sensitive) but 12.8% Vancomycin resistant *Staphylococcus spp*. were isolated. Only 4 cases (14.3%) of MRSA was observed. In our study, the total prevalence of MDR organisms was 218 (78.7%).

The total prevalence of MDR organisms was 78.7%. Majority was MDR gram negative lactose fermenting organisms (90.90%) followed by MDR gram positive organisms (73.73%) and nonlactose fermenting organisms (61.40%).

No significant association was found (p=0.562) between number of chemotherapy drugs given to the patient and the presence of MDR bacteria. Furthermore, no association was noted between absolute neutrophil count and C-reactive protein values (p=0.154). A regression model was developed to find the association between C-reactive protein value and presence of fever. The model was insignificant (p=0.376) indicating that no significant association between the two variables.

Polymyxin B is the most effective drug for gram negative organisms while most effective drugs for GPC are Fusidic acid and Teicoplanin.

Discussion:

The overall prevalence of BSI among cancer patients in this study was 11.75%. This finding is in line with many other studies which indicated a prevalence between 11-38% (Montassier *et al*., 2013, Fentie *et al*.,2018, Rani *et al*.,2017).

In this study patients ranging from 2 months to 82 years were considered. Associations between certain clinical features such as CRP, fever and ANC were assessed in this study. No significant association was observed in

the present study though literature review indicated otherwise.

When considering the bacterial profiles majority of the organisms isolated in this study were gram negative bacilli (GNB) (60.1%) while gram positive cocci and gram positive bacilli were 37.8% and 2.1% respectively. Our results were in accordance with many other studies. (Lubwama *et al*.,2019, Gudiol *et al*.,2014 and Montassier *et al*.,2013).

In a previous study, lower MDR prevalence was recorded (20) (46.5%) while 5 (11.6%) organisms were sensitive to all tested antibiotic classes and no organism was resistant to all tested antibiotic classes (Fentie, A. *et al*.,2018). Very high antibiotic resistance in our setting may be due to several reasons. The calculation of MDR prevalence changes according to the MDR definition as different research groups have followed different definitions. Patients taking antibiotics over the counter without a proper prescription and also the irrational use of antibiotics in our setting may also have played a role in high rates of MDR. A high degree of resistance to Cephalosporins among Enterobacteriaceae in the present study could be because Cephalosporins are one of the most used antibiotics for inpatients as well as for outpatients in developing countries. Papanicolas *et al*.,2017 has proposed a model stating that chemotherapy is a contributing factor for emergence of antibiotic resistant bacteria in gut. It is also mentioned that chemotherapy together with antibiotics has the potential to promote pathogen overgrowth and cause translocation into the blood stream. In our study this fact was taken into consideration and we attempted to find out any association between number of chemotherapy drugs given to a patient and the presence of multi drug resistant bacteria, however the association obtained was insignificant (p=0.562).

Conclusion:

In conclusion, this study provides information on antibiotic resistance of blood isolates which may be a useful guide for physicians initiating empirical therapy. According to our study high prevalence of antimicrobial resistance in Gram-negative isolates and emergence of MDR pathogens is alarming. This highlights the importance of investigating novel antibiotics to overcome the issue. Furthermore, evaluation of Minimum inhibitory concentration (MIC) of effective antibiotics could be performed since it depicts the extent of resistance and possibility of using the drug at a higher dosage for treatment.

Routine surveillance of baseline resistance, formulation of hospital antibiotic policy, usage of combination therapy and usage according to the standard antimicrobial susceptibility testing may help to decrease or prevent the emergence of antibiotic resistance. We also suggest that the association between chemotherapy and multi-drug resistant pathogens to be further investigated with a large sample size and a control group with similar characteristics such as age, gender etc. for better understanding.

References:

Chandrasiri, P, Elwitigala, J, Nanayakkara, G, Chandrasiri, S, Patabendige, G, Karunanayaka, L,Perera, J, Somaratne, P, and Jayathilleke, K (2013):A multi centre laboratory study of Gram negative bacterial blood stream infections in Sri Lanka, Ceylon Medical Journal, 58(2), p.56.

D, R. R, and Chaitanya, S (2017):Retrospective Analysis of Blood Stream Infections andAntibiotic Susceptibility Pattern of Gram Negative Bacteria in a Tertiary Care Cancer Hospital, International Journal of Medical Research & Health Sciences, 6(12): pp. 19–26.

Fentie, A, Wondimeneh, Y, Balcha, A, Amsalu, A and Adankie, B (2018) :Bacterial profile,antibiotic resistance pattern and associated factors among cancer patients at

University of Gondar Hospital, Northwest Ethiopia, Infection and Drug Resistance, Volume 11, pp.2169-2178

Gudiol, C, and Carratalà, J (2014):Antibiotic resistance in cancer patients, Expert Review ofAnti-infective Therapy, 12(8), pp.1003-1016.

Homsi, J, Walsh, D, Panta, R, Lagman, R, Nelson, KA, Longworth, DL (2000): Infectiouscomplications of advanced cancer, Support Care Cancer,8(6),pp.487–492.

Lubwama, M, Phipps, W, Najjuka, C, Kajumbula, H, Ddungu, H, Kambugu, J and Bwanga, F(2019):Bacteremia in febrile cancer patients in Uganda, BMC Research Notes, 12(1).

Marin, M, Gudiol, C, Ardanuy, C, Garcia-Vidal, C, Calvo, M, Arnan, M and Carratalà, J(2014):Bloodstream infections in neutropenic patients with cancer: Differences between patients with hematological malignancies and solid tumors, Journal of Infection, 69(5),pp.417-423.

Montassier, E, Batard, E, Gastinne, T, Potel, G, and Cochetière, M (2013):Recent changes inbacteremia in patients with cancer: a systematic review of epidemiology and antibiotic resistance, European Journal of Clinical Microbiology & Infectious Diseases, 32(7), pp.841-850.

Nejad, ZE, Ghafouri, E, Farahmandi-Nia, Z, Kalantari, B and Saffari, F (2010): Isolation,Identification, and Profile of Antibiotic Resistance of Bacteria in Patients with Cancer, Iran Journal of Medical Sciences,35(2),109–115.

Nurain, AM, Bilal, NE and Ibrahim, ME (2015):The frequency and antimicrobial resistancepatterns of nosocomial pathogens recovered from cancer patients and hospital environments, Asian Pacific Journal Tropical Biomedicine, (12),1055–1059.

Papanicolas, L, Gordon, D, Wesselingh, S and Rogers, G (2018):Not Just Antibiotics: Is CancerChemotherapy Driving Antimicrobial Resistance,Trends in Microbiology, 26(5), pp.393-400.

Rolston, KV, Yadegarynia, D and Kontoyiannis, DP (2006):The spectrum of Grampositivebloodstream infections in patients with hematologic malignancies, and the in vitro activity of various quinolones against Gram-positive bacteria isolated from cancer patients, International Journal of Infectious Disease, 10,pp. 223-30

Saghir, S, Faiz, M, Saleem, M, Younus, A and Aziz, H (2009):Characterization and anti microbial susceptibility of gram - negative bacteria isolated from bloodstream infections of cancer patients on chemotherapy in pakistan, Indian Journal of Medical Microbiology, 27(4), p.341.

Teillant, A, Gandra, S, Barter, D, Morgan, D and Laxminarayan, R (2015):Potential burden ofantibiotic resistance on surgery and cancer chemotherapy antibiotic prophylaxis in the USA: a literature review and modelling study,The Lancet Infectious Diseases, 15(12), pp.1429-1437.

Trecarichi, E, Tumbarello, M, Spanu, T, Caira, M, Fianchi, L, Chiusolo, P, Fadda, G, Leone, G,Cauda, R and Pagano, L (2009):Incidence and clinical impact of extendedspectrum-β-lactamase (ESBL) production and fluoroquinolone resistance in bloodstream infections caused by Escherichia coli in patients with hematological malignancies,Journal of Infection, 58(4), pp.299-307.