MedCrave
Step into the World of Research

Journal of Bacteriology & Mycology: Open access

Research Article

Open Access

CrossMark

Comparative study of C-reactive protein and complete blood count in cancer and non-cancer patients followed by antibiogram analysis of isolated bacterial pathogens

Abstract

Cancer is a group of diseases involving abnormal cell growth with the potential to invade or spread to other parts of the body. This study focuses on the analysis of blood cell and their proteins followed by antibiotic susceptibility and ESBL detection in Cancer and non-Cancer patients. In this study, a total 200 blood and urine samples were screened out for bacteria especially ESBL producing bacteria. Out of the tested samples, different bacterial pathogens were identified and among the isolates *E coli* were (13.33%), *S. aureus* (11.66%), *P. aeruginosa* (11.66%), *salmonella* (10%), *bacillus spps* (9.16%), *Enterobacter spps* (8.33%), *Mycobacterium Spps* (7.5%), *S. Pyogene* (7.5%), *H. pylori* (6.66%), *Klebsiella spp.* (5.83%), *S. epidermidis* (4.16 %) and *Shigella* was (4.16%). The ESBL producing bacteria among the isolated 120 bacterial species were only 14 bacterial isolates are ESBL producers which are *E. coli* (06), *P. aeruginosa* (04). Enterobacter spp (04), *Klebsiella spp* (03) and *Shigella* were only (01). The 100 blood samples are also analysis through CRP test in which 50 blood samples taken from cancer patients and 50 from non-cancer patients. In total 50 cancer sample CRP analysed that 21 patents have the Lowest (0.92 mg/L), 08 was (0.92-1.93 mg/L) 11 was (1.94-3.69 mg/L) and10 patients were on the Highest (>3.69 mg/L) risk. In 50 non-cancer blood sample the CRP level are 09 individual were (0.92 mg/L), 11 individual (0.92-1.93 mg/L), 25 individual (1.94-3.69 mg/L) and 03 patients were Highest (>3.69 mg/L) risk. Maximum resistivity (91%) was showed by penillion (P) and maximum sensitivity (78%) was showed by rifampicin (RD) against isolated bacterial pathogens. It is recommended that Continuous ESBL screening and supervision are necessary at hospital settings to observe and develop approaches for observing and controlling the spread of ESBL generating bacteria.

Keywords: blood, cancer, bacteria, pathogens, CRP, ESBL

Volume 11 Issue 1 - 2023

Asif Iqbal,[1] Dr. Riaz Muhammad,[2] Bakht B Khan,[3] Aleesha Jamshed,[4] Muhammad IK Rehman,[2] Sawaira Iqbal[5]

[1]Department of Microbiology, Hazara University Mansehra, Pakistan
[2]Department of Microbiology, Abasyn University Peshawar, Pakistan
[3]Professor Peshawar Medical College, Pakistan
[4]Departments of Environmental Sciences, COMSATS University, Abbottabad Campus, Pakistan
[5]Department of botany, GPGC Nowshera kpk Pakistan

Correspondence: Asif Iqbal, Department of Microbiology, Hazara University Mansehra, Kp Pakistan-21300, Tel +923305988671, Email microbiologist11@gmail.com

Received: December 30, 2022 | **Published:** January 11, 2023

Introduction

The most important part of cancer research over the past numerous years have been complete on the comprehensive study of the mechanisms of malignancy etiology.[1] Cancer is an extremely difficult progression. Carcinogens (cancer causing agents) such as viruses and some of the 200 chemicals found in cigarette smoke are processed in the body and then cause modifications to DNA.[2] This stage of carcinogenesis[3] is known as initiation. In the subsequent stage-promotion cells that carry damaged or mutated DNA may undertake alterations that allow them to progress into a cancer. Many factors modify these processes, such as hormones, genetics, body weight, and the many constituents present in the diet. CRP was discovered nearly 70 years ago by scientists exploring the human inflammatory response.[4] White Blood Cells (WBC) are the main and important type of blood cells which circulate throughout the blood vessel and the lymphatic system. These are very important and defensive cell of immune system, because WBCs perform very important role in defence against microbial antigen.[5] CRP is a significant acute phase complementary system associated protein which is present in normal serum of humans and produced by liver due to the macrophage which release cytokines (IL-1, IL-6, IL-8, TNF etc.) when bacteria is engulf by macrophage.[6] CRP is the first protein of complementary system which binds to antigen.[7] The bacteria present in various types of cancer patients were *Salmonella, Shigella, Campylobacter*, *H. pylori*, *Mycobacterium tuberculosis*, *Escherichia coli*, Listeria, *Actinobacter baumannii*, *Streptococcus pyogene*, *Streptococcus mutant*, *Salmonella*, *Staphylococcus aureus*, *Vibrio cholera* and *Yersinia* etc.[8] These bacterial species cause different opportunistic infections in cancer patients due to the compromised immune system and many other reasons. Among β-lactamases, Extended-Spectrum Beta–Lactamases (ESBLs), Ampciline C-type β-lactamases, in addition to carbapenemase are of abundant apprehension.[9] Antibiotic resistance is rapidly increase in that regions where the people have no proper hygiene and immoral use of antibiotics.[10] One main reason is the rapid traveling of people from one country to another for different purposes especially from that part of the world where resistant bacteria exist especially ESBL generating Bacteria are present in some parts of Asia, Africa and South America.[11] The current paper focuses on the analysis of blood cell and antibiotic susceptibility of bacterial pathogens in Cancer and non-Cancer patients.

Methodology

The current research work will be carried out in MRL in the Department of Microbiology and Biotechnology, Abasyn University Peshawar and in Microbiology and Hematology Laboratory of Pathology Department HMC from March 2017 to December 2017.

Total 200 blood and urine samples were collected from cancer patients of various types (Liver, Lungs, Blood, Skin and Stomach cancer) and non-cancer patients within one to two month. The samples were collected from different hospital located at Peshawar City i.e. mostly from HMC, IHP and LRH Peshawar.

Submit Manuscript | http://medcraveonline.com

©2023 Iqbal et al. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and build upon your work non-commercially.

CBC and CRP

Blood samples each with 4.5ml blood were collected in green EDTA tubes then the samples were first mixed. Then the blood sample was run in Sysmex and cell dyne ruby (CDR) blood counting apparatus.[12] The C-reactive protein (CRP) test was performing accordingly.[13,14]

Urine analysis

Total 100 Urine sample were collected and out of these 50 samples was taken from cancerous patient and 50 from non-cancer patient. Then the physical, chemical and Microscopic analysis was conducted for the evaluation of patient Blood cells and for the presence of bacteria in patients' Urine.[15]

Identification of isolated bacteria

The different culture Media was used i.e., Nutrient agar, blood agar, CLED agar, MacConkey agar, Eosin Methylene Blue (EMB), *Salmonella, Shigella* Agar (SSA). Pathogenic bacteria were isolate though serial dilution method on nutrient agar. The pathogenic bacteria form a colony; from colony different bacterial species were isolated according to their morphology, Gram's reaction and biochemical test.[16]

Antibiotic susceptibility testing

Disc diffusion assay

Antibiotic sensitivity was done through Kirby-Bauer disc diffusion technique. Standard protocols mentioned in Clinical and Laboratory Standards Institute (CLSI) 2015 was followed. Different antibiotics (Amoxicillin, cyclosporine, Vancomycin, Gentamycin, Tazobactum, Cefotaxime, Ciprofloxacin, Rifampicin, Doxycycline, Chloramphenicol, Polymaxin-B, Carbapenam, tetracycline, etc.) of various concentrations was evaluated. According to the type of isolated bacteria various types of antibiotics was tested against isolated pathogenic bacteria according to the Clinical and Laboratory Standards Institute (CLSI) 2015 protocol.

In the disk diffusion test, the bacterial isolate is inoculated uniformly onto the surface of an agar plate and a filter disk impregnated with a standard amount of an antibiotic is applied to the surface of the plate, resulting in a gradient of the antibiotic surrounding the disk. Following incubation, a bacterial lawn appears on the plate and zones of inhibition of bacterial growth would be present around the antibiotic disk. The test is performed under standardized conditions hence the size of the inhibition zone is dependent on the degree of sensitivity of the microorganism to the antibiotic.[17]

Detection of ESBL producing bacterial isolates

A 0.5 MacFarland's suspension of every single insulates was spread on a Muller – Hinton agar (MHA) plate Ceftazidime (30 µg) Ceftazidime and clavulanic acid (30 µg/ 10 µg) discs was positioned aseptically on the agar having plate. The space of approximately 15mm was retained in the middle of the two discs (edge to edge) and the cultures were incubated at 37 C overnight. The observation was equal or greater than 5mm rise in the zone diameter for the antimicrobial agent which was tested in combination with clavulanic acid, against its zone thickness when tested only, confirmed the occurrence of Extended Spectrum Bata Lactamase (ESBL) manufacture by the bacteria.[18]

Results and discussion

Present study was carried out to determine the current scenario of CRP, CBC, and bacteria resistance present in cancer and non-cancer patients. In this study, a total 200 blood and urine samples were screened out. Out of the tested samples, different bacterial pathogens were identified and among the isolates *E coli* were (13.33%), *S. aureus* (11.66%), *P. aeruginosa* (11.66%), *salmonella* (10%), *bacillus spps* (9.16%), *Enterobacter spps* (8.33%), *Mycobacterium Spps* (7.5%), *S. Pyogene* (7.5%), *H. pylori* (6.66%), *Klebsiella spp*. (5.83%), *S. epidermidis* (4.16 %) and *Shigella* was (4.16%). Similarly a study also by Helde-Frankling et al.,[19] isolated bacteria in various varieties of cancer are *Salmonella, Shigella, Campylobacter*, *H. pylori*, *Mycobacterium tuberculosis*, *Escherichia coli*, *Listeria*, *Actinobacter baumannii*, *Streptococcus pyogene*, *Streptococcus* mutant, *Salmonella*, *Staphylococcus aureus*, *Vibrio cholera* and *Yersinia*. Our study shows the some indication of viral and fungal spps as well. However we don't search out viral and fungal infections. The outcomes of Sengar et al.[20] are somehow in confirmatory with our results where the frequency of Gram-Negative Bacteria remained (51.59%), in which *E. coli* (12.79%) *Pseudomonas* species were (9.71%) *Salmonella* (8%), and gram positive (40.72 %) in which *staph aureus* were (9.23%) *Bacillus* spps (9.16%) respectively. However the outcomes of Parveen et al.[21] also reported the Spiral or curved bacilli isolates and genus Campylobacter (Table 1 & 2).

Table 1 Positive sample distribution of bacterial isolates in various samples

Total samples	Negative samples	Total positive bacterial isolates	Total bacterial isolates in cancer samples	Total bacterial isolates in non-cancer samples	Total bacteria isolates from blood samples	Total bacteria isolates from urine samples
200	80	110	51	59	30	80

Table 2 Percentage distributions of various bacterial pathogens identified in both cancer and non-cancer samples and ESBL bacterial Isolates

S no.	Name of bacteria isolated	Count	Percentage (%)	ESBL positive isolates
01	*E. coli*	16	13.33	06
02	*S. aureus*	14	11.66	Nil
03	*P. aeruginosa*	14	11.66	04
04	*Salmonella*	12	10	Nil
05	*bacillus spp.*	11	9.16	Nil
06	*Enterobacter spp.*	10	8.33	04
07	*Mycobacterium Spp.*	09	7.5	Nil
08	*S. pyogene*	09	7.5	Nil
09	*H. pylori*	08	6.66	Nil
10	*Klebsiella spp.*	07	5.83	03
11	*S. epidermidis*	05	4.16	Nil
12	*Shigella*	05	4.16	01
Total		120	100	18

In the present research work we also detect ESBL producing bacteria among the isolated 120 bacterial species. Out of these only 14 bacterial isolates are ESBL producers which are *E. coli* (06), *P. aeruginosa* (04), *Enterobacter spp* (04). *Klebsiella spp* (03) and *Shigella* were only (01). The ESBL producing isolates were detect though double disk diffusion assay in which Ceftazidime (30 µg) Ceftazidime and clavulanic acid (30 µg/ 10 µg) discs were used. The Findings of Bos et al.[22] also isolated ESBL producing bacteria from cancer patients with bacteremia (Table 3).

Table 3 Susceptibility profile of various isolated bacteria against various antibiotics

Microbes	Count	VA		CN		CAZ		TZP		TE		AMC		CTX		CIP		RD		DO		C		PB		Pen	
-	-	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S
E. coli	16	40	60	62	38	30	70	18	82	53	47	53	47	47	53	23	77	51	49	32	68	54	46	53	47	81	19
Staph aureus	14	46	54	53	47	38	62	26	74	44	56	43	57	41	69	36	64	32	72	41	59	46	54	41	59	79	21
P. aeruginosa	14	30	70	40	60	29	71	24	76	32	68	31	69	43	57	42	58	19	81	40	60	42	58	42	58	70	30
Salmonella spps	12	51	49	31	69	52	48	45	55	43	52	42	58	24	74	25	75	41	59	30	70	40	60	21	79	65	35
bacillus spp.	11	32	68	32	68	43	57	37	73	29	71	35	65	53	47	35	65	22	78	39	61	22	78	39	61	91	09
Enterobacter spps	10	21	79	12	88	34	56	28	72	46	56	44	56	62	38	53	47	41	59	37	63	32	68	43	57	75	25
Mycobacterium Spps	09	48	52	18	82	51	49	51	49	48	52	39	61	44	56	38	62	33	77	32	68	40	60	59	41	79	21
S. pyogene	09	30	70	36	64	41	59	39	61	45	55	27	73	53	47	43	57	19	81	38	62	20	80	61	39	59	41
H.pylori	08	46	54	27	73	52	48	44	56	28	72	45	55	32	67	50	50	22	78	36	64	42	58	46	56	76	24
Klebsiella spp.	07	40	60	45	55	35	65	12	88	23	77	26	74	61	39	29	71	40	60	40	60	61	39	49	51	62	38
S. epidermidis	05	25	75	33	77	24	76	29	71	38	62	48	52	41	59	24	76	29	71	57	43	45	55	48	52	45	55
Shigella	05	31	69	39	61	19	81	50	50	21	79	30	70	60	40	46	54	38	62	50	50	31	69	40	60	61	39

In the current study 100 blood samples were also screened for CRP. Out the of the total sample 50 blood samples taken from cancer patients and 50 from non-cancer patients. In total 50 cancer sample CRP analysed that 21 patents have the Lowest (0.92 mg/L), 08 was (0.92-1.93 mg/L) 11 was (1.94-3.69 mg/L) and 10 patients were on the Highest (>3.69 mg/L) risk. In 50 non-cancer blood sample the CRP level are 09 individual were (0.92 mg/L), 11 individual (0.92-1.93 mg/L), 25 individual (1.94-3.69 mg/L) and 03 patients were Highest (>3.69 mg/L) risk. Juan et al.[23] also studied High levels (>3 mg/L) of CRP were associated with an increased risk of incident cancer (hazard ratio, 1.4; 95% CI, 1.1 to 1.7) compared with persons with low levels (<1 mg/L. Although CRP seems to affect several cancer sites, the association was strongest for lung cancer (hazard ratio, 2.8; 95% CI, 1.6 to 4.9). Decreased CRP levels was associated with an increased cancer risk of 2.6 (95% CI, 1.6 to 4.4) in homozygous carriers. The cancer and non-cancer samples were also screened for CBC analysis through Sysmex and cell dyne ruby. Durnaś et al.[24] also reported the Calculation of blood sample permanence used for Complete Blood Count (CBC) by means of Sysmex XN-9000 also through Mindray BC-6800 analyzers. Similar screening method also showed by Daniluk et al.[25] that the partiality for haematocrit, mean corpuscular haemoglobin attentiveness, mean corpuscular volume, and Red Blood Cell (RBC) sharing width coefficient of variant was greater than the critical dissimilarity after 8 hours. In the consequence the same analyzers were used (Figure 1 & 2).

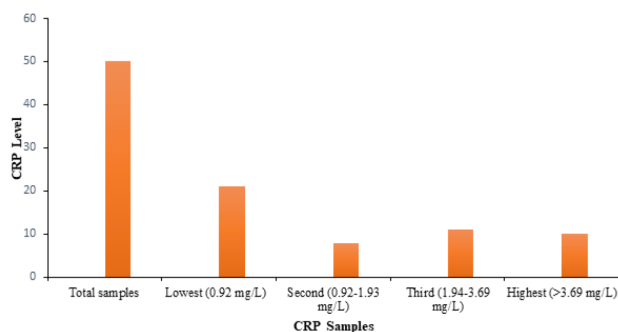

Figure 1 CRP analysis for cancer samples.

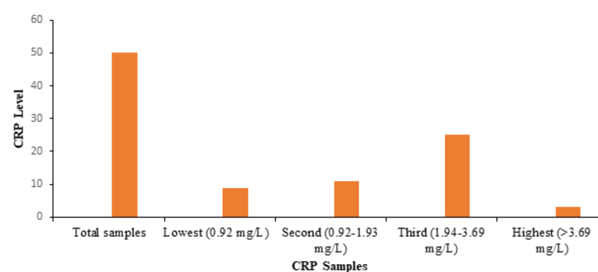

Figure 2 CRP analysis for non-cancer samples.

Conclusions and recommendations

It is concluded that the maximum isolated bacteria were *E. coli*, *S. aureus*, *P. aeruginosa* and *Salmonella*. The ESBL producing bacteria were *Enterobacter spp. E. coli, Klebsiella spp*, *P. aeruginosa*, and *Shigella*. The blood analysis were performed for cancer patients in which 60 % patient were high level of WBC in cancer and 56% were High level of WBC in non-cancer patients. In urine samples the presence of bacteria in cancer which is 86% and 78% in non-cancer samples.

It is recommended that Continuous ESBL screening and supervision are necessary at hospital settings to observe and develop approaches for observing and controlling the spread of ESBL generating bacteria. Community awareness is required for use and mismanagement of antibiotics. Alternate approaches must be hopeful to find out novel sources of antibacterial for control of bacterial resistance. More research studies are required to study cancer and non-cancerous patients on microbiological, molecular and immunological level. Selective pressure for resistance through antimicrobial use is important, infection control practices are critical to limiting the spread of resistant organisms. The life-threatening nature of bacteremia and sepsis underscores the importance of using timely surveillance data to develop rational antimicrobial therapy recommendations and to design strategies to help control antimicrobial resistance.

Acknowledgments

None.

Conflicts of interest

Authors declare that there are no conflicts of interest.

References

1. Batista Jr ML, Henriques FS, Neves RX, et al. Cachexia associated adipose tissue morphological rearrangement in gastrointestinal cancer patients. *J Cachexia Sarcopenia Muscle*. 2016;7(1):37–47.
2. Castagnola E, Mikulska M, Viscoli C. Prophylaxis and empirical therapy of infection in cancer patients. *Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases*. 2015;3395–3413.
3. Iqbal A, Raza MH, Jadoon H, et al. Applications of nanomaterials for health and environment protection. *MOJ Eco Environ Sci*. 2022;7(3):84–87.
4. Christiansen MN, Chik J, Lee L, et al. Cell surface protein glycosylation in cancer. *Proteomics*. 2014;14(4–5):525–546.
5. Bang CS, Yoon JH, Kim YJ, et al. Clinical impact of body mass index on bactibilia and bacteremia. *BMC Gastroenterol*. 2014;14:104.
6. Zembower TR. Epidemiology of infections in cancer patients. *Cancer Treat Res*. 2014;161:43–89.
7. Študentová H, Vitásková D, Šrámek V, et al. Correlations of neutrophil-to-lymphocyte, lymphocyte-to-monocyte and platelet-to-lymphocyte ratios with biomarkers of atherosclerosis risk and inflammatory response in patients with a history of breast cancer. *Pteridines*. 2015;26(4):161–172.
8. Thomas AM, Jesus EC, Lopes A, et al. Tissue-associated bacterial alterations in rectal carcinoma patients revealed by 16S rRNA community profiling. *Front Cell Infect Microbial*. 2016;6:179.
9. Pomar V, Benito N, López-Contreras J, et al. Characteristics and outcome of spontaneous bacterial meningitis in patients with cancer compared to patients without cancer. *Medicine (Baltimore)*. 2017;96(19):e6899.
10. Noskin GA. *Management of Infectious Complication in Cancer Patients*. Springer. 2007;96.
11. Papo N, Shai Y. Host defense peptides as new weapons in cancer treatment. *Cell Mol Life Sci*. 2015;62(7–8):784–790.
12. Cammarota G, Ianiro G. Gut microbiota and cancer patients: a broad-ranging relationship. *Mayo Clin Proc*. 2017;92(11):1605–1607.
13. Agrawal A. CRP after 2004. *Mol Immunol*. 2005;42(8):927–930.
14. Iqbal A, Ali S, Asad M, et al. Perspectives and Applications of Plant Microbe Interactions in Post Genomic Era. *Annals of the Romanian Society for Cell Biology*. 2022;26(01):1216–1226.
15. Lo WCJ, Luther DG. Detection of cancer cell death mediated by a synthetic granzyme B-like peptide fluorescent conjugate and the same peptide binding in bacteria. *J Fluoresc*. 2014;24(2):465–471.
16. Gómez MG, Guío L, Hernández JL, et al. Bacteraemia due to extended-spectrum beta-lactamases (ESBL) and other beta-lactamases (ampC and carbapenemase) producing Enterobacteriaceae: association with health-care and cancer. *Rev Esp Quimioter*. 2015;28(5):256–262.
17. Juárez PC, Compte DV, Horton AG, et al. Hospital-acquired infections at an oncological intensive care cancer unit: differences between solid and hematological cancer patients. *BMC Infect Dis*. 2016;16(1):274.
18. Zhang Q, Li D, Bai C, et al. Clinical prognostic factors for time to positivity in cancer patients with bloodstream infections. *Infection*. 2016;44(5):583–588.
19. Frankling MH, Bergqvist J, Bergman P, et al. Antibiotic treatment in end-of-life cancer patients-a retrospective observational study at a palliative care center in Sweden. *Cancers (Basel)*. 2016;8(9):84.
20. Sengar M, Kelkar R, Jain H, et al. Frequency of bacterial isolates and pattern of antimicrobial resistance in patients with hematological malignancies: A snapshot from tertiary cancer center. *Indian J Cancer*. 2014;51(4):456–458.
21. Parveen A, Sultan F, Raza A, et al. Bacteraemia caused by Escherichia coli in cancer patients at a specialist center in Pakistan. *J Pak Med Assoc*. 2015;65(12):1271–1276.
22. Bos MMEM, Smeets LS, Dumay I, et al. Bloodstream infections in patients with or without cancer in a large community hospital. *Infection*. 2013;41(5):949–958.
23. Juan H, Qijun W, Junlan L. Serum CRP protein as a differential marker in cancer. *Cell Biochemistry and Biophysics*. 2012;64(2):89–93.
24. Durnaś B, Wątek M, Wollny T, et al. Utility of blood procalcitonin concentration in the management of cancer patients with infections. *Onco Targets Ther*. 2016;9:469–475.
25. Daniluk T, Fiedoruk K, Sciepuk M, et al. Aerobic bacteria in the oral cavity of patients with removable dentures. *Adv Med Sci*. 2006;51(Suppl 1):86–90.

Citation: Iqbal A, Muhammad R, Khan BB, et al. Comparative study of C-reactive protein and complete blood count in cancer and non-cancer patients followed by antibiogram analysis of isolated bacterial pathogens. *J Bacteriol Mycol Open Access*. 2023;11(1):1–4. DOI: 10.15406/jbmoa.2023.11.00334