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Gujarat Cancer Society Research Journal

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Journey So Far : "GCRI Bulletin …….Gujarat Cancer Society Research Journal"

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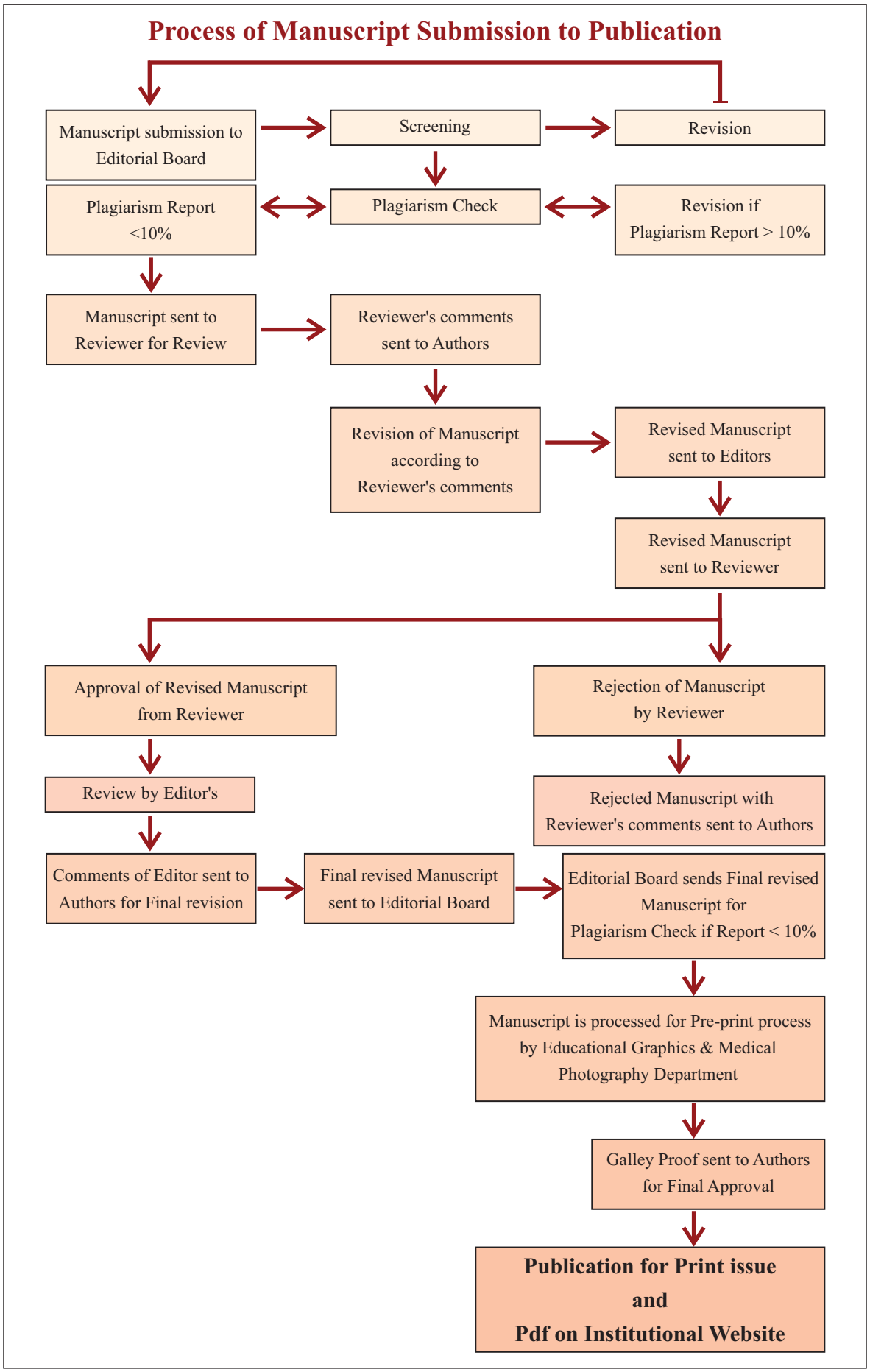
Research and publication complement teaching and training, clinical care, and public health works. Equally, 'Success in Academic' unequivocally depends on "research and publications". It is documented as lifelong learning process to communicate one's ideas to scientific peers, and invite them to express an informed view. Thus **"Publication of your work is de rigueur." A journal is an academic phenomenon of clinical research, hard work, talent, and creativity, skilfully showcasing the excellence of the science that it embodies.** Hence, in the year 1991, the GCRI Teacher forum put forth a proposition to have a journal published from the institute emphasizing the need to create platform to enable learners seeking knowledge to thrive through such type of communications. This challenge was undertaken by Dr. Dilip D Giri, Former Professor of Pathology and by Dr. Siddharth Adhvaryu, Former Chief Research Officer, and finally, they set the ball rolling on to transform scientific and clinical research knowledge in form of **GCRI BULLETIN.** Supportively, the Gujarat Cancer Society most generously accepted the responsibility to finance for the publication of the Journal and translating suggestion into reality. On December 1991, the journey began and the first issue of GCRI Bulletin was launched with contributions from academic and Research GCRI staff members. **Dr. M. R. Das,** Director Grade Scientist from CCMB, Hyderabad was considerate for allowing to publish his Dr. T. B. Patel Oration lecture as a lead article in the inaugural journal. Though, the focus of the journal was solely on publishing research articles since inception, all the same, to project academic activities carried out by various Departments of the Institute summaries of papers and articles published in various journals and other scientific publications, as well as account of the work presented in the clinical meetings and the journal clubs were included amongst the range of articles as one of the features in the journal. **Upto May 2003, Volume 8, it was published as "GCRI Bulletin", thereafter, from April 2004 (Volume 9-10) to April 2011 (Volume 13), it was published as** 'GCS Research Bulletin'.

In the year 2012, one small step forward, was, it's metamorphose into the 'Gujarat Cancer Society Research Journal. This, one can say was recreation of the former 13 editions which already had established the style and standard, both scientific and aesthetic. It was, and so shall be the present form, the official mouth-piece and the scientific ambassador of the Gujarat Cancer Society (GCS) and the Gujarat Cancer and Research Institute (GCRI) portraying their research and academic pursuits and efforts. One may presume, that this one small step can be a giant leap into future. The Journal of "Gujarat Cancer Society Research Journal" reached its 10th year of publishing in 2021.

What is new? (1) The frequency of publication from annually is increased to bi-annually, (2) On December 12, 2012, the "National Institute of Science Communication and Information Resources" New-Delhi, registered the **"Gujarat Cancer Society Research Journal" and assigned ISSN (International Standard Serial Number) number.** ISSN is assigned to serials published in India by the NSL being National Centre for ISSN. (2) **Introduction of Peer review Process**: "Gujarat Cancer Society Research Journal" content is peer-reviewed. A scientific journal requires constant and stringent quality control, therefore, a strict **peer review process** is necessary for maintaining quality. "Gujarat Cancer Society Research Journal" follows a double blind review process, therein the reviewer/s and authors are unaware of each other's identity. This scientific review process although strict yet friendly and benevolent, as well as pure and un-envious eyed, balanced review of need, content and presentation of the research material given for publication (3) Its **indexed** with Index Copernicus since 2013. (4) It is also indexed with **NLM** (National library of Medicine) catalog ie an alternative search interface to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the bibliographic records in Locator Plus in the NCBI database. (5) **Plagiarism** is considered **as serious professional/scientific/publication misconduct.** Therefore, one and all manuscript submitted to the Gujarat Cancer Society Research Journal are subjected to thorough plagiarism check with professional plagiarism detection 'iThenticate software as well as scrutiny by the editorial team before processing the manuscript.

What more? Inclusion of "About the Journal and Instruction to Authors", Guidelines for Reviewer, Contributors Form, Organisation Information, interesting anecdotal reports under the heading "Brain Waves" and Reviewers Board.

Connecting Digital Identifier with publication: GCS Research Journal, April 2021, Volume 22 has integrated author`s ORCID ID with their publication. "ORCID iDs" are permanent identifiers for researchers. It (ORCID iD) will significantly **improve discoverability** and will help increase the chances of a researcher being recognized for their work. Can be used in various scenarios like an application for grant submission or receiving funds, manuscript submission to a journal, or filling professional details on a researcher profile page.

Most important is its **Accessibility**: "Gujarat Cancer Society Research Journal" archives (2012; Volume 14 onwards), and current volumes are freely available on GCRI website in Pdf format on the given link(https://www.gcriindia.org/gcs_research_journal.html). Volume 21, 2019 onwards, one will be able to access, download or print singly full details (Pdf format) of all original articles, cases reports, anecdotal reports, etc.

What is forthcoming?

1) The present day scope is to publish the work done at GCS and GCRI during a specified period of time but later on it may be expanded to include former GCRIans' work done at GCRI. It will continue to serve as an academic-research bridge between the basic sciences and the applied sciences, viz. various disciplines of medicine within and outside GCSGCRI.

2) Obtain e_ISSN: The Journal is available on institution website, therefore, next step would be to get registered e-issn number. Also, all the articles would be available online in both html and Pdf format.

3) To go in for other major indexers such as DOAJ, PMC, Medline , etc.

We thank all of our authors, past editorial board members, reviewers, and staff who have made the past decade successful for this journal. We dream of quality papers that address public health needs and are written in English to an international standard. Papers that are of a high scientific and ethical standard with appropriate statistical analysis.

Please work with us to achieve our above dreams and the dream of good health of the people of our region.

Looking forward for your support and inputs in making this journal an enriching academic fountain ahead…

Prevalence of New Delhi Metallo - Beta - Lactamase (NDM) and Other Carbapenemases in Commonly \Isolated Gram-negative Bacilli by Multiplex PCR Isolated from Cancer Patients Suffering with Infections

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Summary

Life threatening infections are caused by antibiotic resistant bacteria leading to a significant mishap in antimicrobial treatment. The boundless utilization of carbapenems has brought about the development of carbapenemase producing bacteria. Among carbapenemase, metallo-beta lactamases are of prime importance produced by gene clusters, carried on the mobile plasmids. This study was aimed to know the prevalence of metallo-beta-lactamase(MBL) by PCR in commonly isolated pathogenic Enterobacteriaceae and non-lactose fermenting bacilli like Acinetobacter spp. and pseudomonas spp. This study was for seven months (May to November 2018). The pathogenic Enterobacteriaceae, Acinetobacter and Pseudomonas were collected and stored. After preliminary identification by Vitek 2 (AES analysis) the isolates were subjected for DNA testing (multiplex PCR) using targeted primers for different MBL genes. Out of the 5062 clinical samples, we selected 160 random isolates of interest for further study. 66.25% of GNB were resistant to imipenem, 56.87% to meropenem and 81.25% to doripenem. The results of selected 104 different GNB carbapenemase positive showed that 24 had blaNDM genes, 1 each of blaVIM & blaIMP genes & VIM family, 2 had SIM family, 19 had IMP family & 22 had detectable GIM-1 genes. There were co-existing genes either two or three together in single isolate. Out of the six isolates of Klebsiella spp. five isolates had blaNDM-1 and blaGIM-1, three isolates had blaNDM and VIM family. There was also presence of blaNDM, blaIMP, IMP family and GIM in one isolate. Our study found that Klebsiella pneumonia is the principle supply of different plasmid borne beta lactamase qualities coding for carbapenemase hydrolyzing all beta lactam antibiotics. This is of concern because the mobile resistant plasmids are prevalent in the CRE in hospitals and also colonize the normal flora. This has a potential for outbreak infections due to these resistant bacteria.

In conclusion, these infecting bacteria having MBL, isolated from our patients causing morbidity and mortality are enthralling attention of the clinicians leading to vast awareness on the existence of multi drug resistant super bugs in day-to-day practice. It is a requirement of the day to have antibiotic policies for the hospitals and having strict infection prevention policies to prevent the spared of these super bugs in the hospitals from HCWs & patients to patients.

Keywords: Metallo-beta-lactamase (MBL), blaNDM, CRE (Carbapenem resistant Enterobactereceae), Multidrug resistance, Antibiotic policy

Introduction

NDM represents New Delhi metallo-beta-lactamases (MBL), which is an enzyme that makes gram negative bacteria (GNB) not sensitive to a wide range of beta lactam antibiotics. These incorporates antibiotics having a place with carbapenem family which are viewed as pillar for the treatment of diseases cause by antibiotic resistant bacteria. These includes antibiotics belonging to carbapenem family which are considered main stay for the treatment of antibiotic resistant bacterial infections. This enzyme has been reported most commonly from India and Pakistan. The concern to health industry in that it is spreading throughout the world as people travel from country to country.[1]

Carbapenem antibiotics (imipenem, ertapenem, meropenem, doripenem) are members of the beta lactam class of antibiotics like penicillin's & cephalosporins have broad spectrum of activity against the gram positive, gram negative and anaerobic bacteria. Carbapenem are unique because they relatively are resistant to hydrolysis by most of beta-lactamases or are inhibitors of beta lactamases.[1]

The beta-lactamases are classified based on their functional and molecular properties. Focusing on the molecular properties, they are classified into molecular classes A, C and D which includes serene beta lactamases, though atomic class B beta-lactamases are all metallo enzymes (NDM) with dynamic site zinc molecular class B beta lactamases are all metallo enzymes (NDM) with and active site zinc, and these enzymes are inhibited by ethylene diamine tetra acetic acid (EDTA) thereby called as metallo beta lactamases.[2]

It has been observed that over more than a decade the commonly isolated Enterobacteriaceae have become resistant to almost all generation of cephalosporin and carbapenems due to the wise actions taken by the superbugs fighting against the antibiotics used in the patient treatment be it in the ICU or treating the immunocompromised patients. They are called carbapenem resistant Enterobacteriaceae (CRE). This as a major challenge

being faced by the treating physicians and has become a major cause of morbidity and increased stay in the hospital also impacting the financial expenditure for the hospital stay and expensive antibiotics.

Therefore, this study was designed to know the prevalence of metallo-beta-lactamase (NDM) by molecular method called multiplex polymerase chain reaction (m-PCR) in commonly isolated pathogenic Enterobacteriaceae and non-lactose fermenting bacilli like Acinetobacter spp and Pseudomonas spp.

Materials and Method

This prospective study was conducted over a period of seven months from May to November 2018 in the laboratory of microbiology of The Gujarat Cancer & Research Institute, Ahmedabad, Gujarat, India. Pathogenic bacteria were isolated from different types of specimens received, like blood for blood cultures in patients suffering from bacteremia, pus material received on swab in complicating post-operative wound infections and urine culture from patients suffering from urinary tract infections.

The pathogenic isolates selected for further study belonged to commonly isolated Enterobacteriaceae family and non-lactose fermenters like Acinetobacter spp, Burkholderia Spingomonas spp and Pseudomonas spp, which were initially identified by automated identification and susceptibility testing instrument called Vitek-2 compact and advance expert system (AES) of the software of Vitek. Formal ethical clearance and approval was taken from the institutional ethics committee before starting the study.

Method:

Carbapenemase production was noted from the results produced by the advance expert system of Vitek-2 Software which flagged the carbapenemase producers.

Genotypic detection of metallo-beta lactamase genes was done by multiplex polymerase chain reaction as follows:

Focus was on the detection of blaNDM-1, blaVIM, blaIMP and IMP family, VIM family, GIM-1, SPM-1 genes, and documentation of amplicons was done by gel documentation system.

Extraction: DNA extraction was finished utilizing the spin column method (QIAGEN: GmbH, Hilden, Germany) according to the company guidelines.

Multiplex Polymerase chain reaction: Hi-Chrome PCR master mix, a ready to use mix of ChromTaq DNA Polymerase, Buffer, MgCl2 and dNTPs (HIMEDIA, MolBioTM) was used. All lysates were exposed to multiplex PCR utilizing preliminaries focusing on blaNDM-1. The conjunction of other MBL encoding genes specifically blaVIM and blaIMP were searched for by utilizing consensus primer. Other multiplex PCR primers introductions for the identification of other MBL qualities were for SIM, IMP, SPM, VIM, and GIM. The primers used in the multiplexing (MP-3 and MP-5) are listed in Table-1. MP-3 is where the forward and backward primers used were three, whereas in MP-5 they were five

Amplification for the PCR assay was carried out in a thermal cycler (Applied Biosystems Gene amp 9700 PCR system) with following protocol: Starting with DNA release and denaturation for 5 minutes at 94℃ temperature, trailed by 40 patterns of 94°C temperature for 30 second, 52°C temperature for 40 second and 72°C temperature for 50 second, trailed by a solitary, last extension step at 72°C temperature for 5 minutes.

Gel Electrophoresis: PCR product containing amplicons was analyzed in a 2.5% agarose gel in 1x TAE buffer at 130V and 180mA current for 1.5 hr and the band were visualized with ethidium bromide using a Gel Documentation UV illuminator system (Gel. Luminax, BioZen). A DNA size marker of 100bp was used for comparative analysis.

Table 1: Primers used in Multiplexing PCR (Shanthi et al. 2014)

Multiplex	Sr. No.	Oligonucleotide	Nucleotidesequence, 5'–3'	Ampliconsize (Basepair)
MP-3	1	BlaNDM-1–F BlaNDM-1–R	GGGCAGTCGCTTCCAACGGT GTAGTGCTCAGTGTCGGCAT	475
	2	BlaVIM–F BlaVIM–R	TTTGGTCGCATATCGCAACG CCATTCAGCCAGATCGGCAT	500
	3	BlaIMP–F BlaIMP–R	GTTTATGTTCATACWTCG GGTTTAAYAAAACAACCAC	432
MP-5	4	IMPfamily-F IMPfamily-R	GGAATAGAGTGGCTTAAYTCTC CCAAACYACTASGTTATCT	188
	5	VIMfamily-F VIMfamily-R	GATGGTGTTTGGTCGCATA CGAATGCGCAGCACCAG	390
	6	GIM-1-F GIM-1-R	TCGACACACCTTGGTCTGAA AACTTCCAACTTTGCCATGC	271
	7	SPM-1-F SPM-1-R	AAAATCTGGGTACGCAAACG ACATTATCCGCTGGAACAGG	477
	8	SIM-1-F SIM-1-R	TACAAGGGATTCGGCATCG TAATGGCCTGTTCCCATGTG	570

Table 2: Infecting Gram-Negative Bacilli and Carbapenem Resistance (n=160)

Total GNB	Imipenem		Meropenem		Doripenem	
n=160	R	%	R	%	R	%
	106	66.25%	91	56.87%	130	81.25%

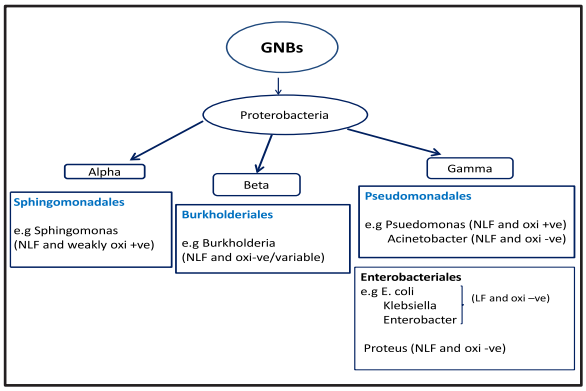

Figure 1: Classification of commonly isolated bacterial pathogens from cancer patients

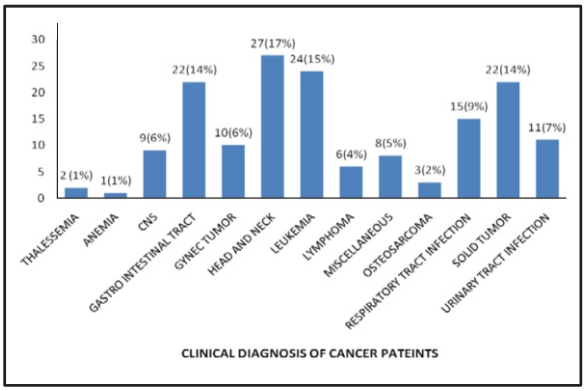

Figure 3: Gram negative bacilli infections in cancer patients enrolled in the study

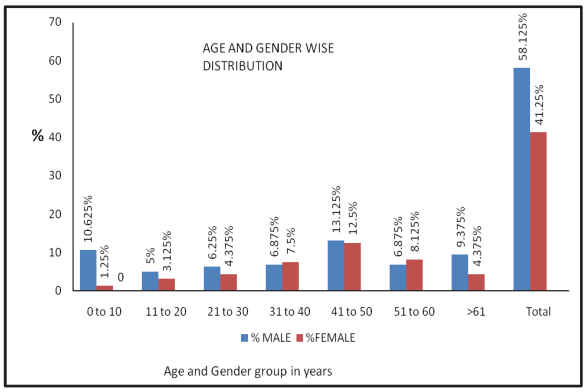

Figure 2: Age group and gender of patients enrolled in the study

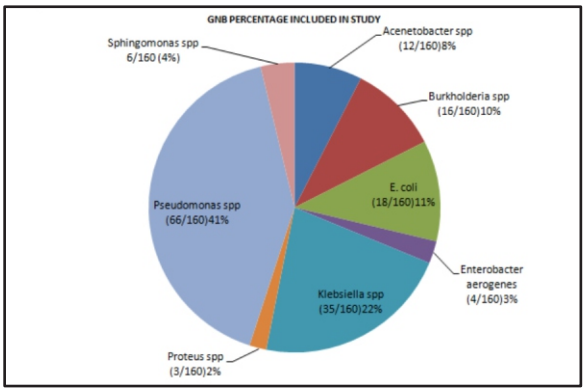

Figure 4: Types of organisms isolated in study group

Results

During the 6 months of study, a total number of 5062 clinical samples from various units were received for culture and sensitivity. From this randomly 160 isolates of different Gram-Negative bacilli (GNB) were included in the study. For the ease of identification in the laboratory commonly isolated bacterial pathogen is as classified below in the Figure 1.

Age and gender distribution of the patients who had infections is as per Figure 2. Highest percentage of infection was seen in 13.125% of male in the age group 41-50 yrs, followed by 10.625% in the age group of 0-10 years. 12.5% of the females having infections were seen in the age group of 41-50 years followed by 8.125% in age group of 51-60 years. Least percentage of infections (5%) in males was seen in the age group 11-20 years, while 1.25 % females having infections belonged to age group 0-10 years which is shown in the Figure 2.

Another finding of the study was patients with different cancers having infections showed that highest infections were seen in head and neck cancer 17% (27/160), followed by leukemia 15%(24/160). GI and solid tumor showed 14% (22/160), followed by respiratory cancer 9% (15/160) and urinary tract infections 7% (11/160), rest were less than 10% which is shown in Figure 3.

Out of 865 GNB were isolated, 160 isolates were randomly picked for future study. Out of the 160 isolates 41% (66/160) were Pseudomonas spp, 22% (35/160) were Pneumonia, 11% (18/160) were E. coli, 10% (16/160) were Burkholderia cepacia, 8% were Acinetobacter spp., Spingomonas paucimovilis 4%, Proteus spp 2% and Enterobacter aerogenes and Enterococcus spp showed 1 % each. (Figure 4)

According to Table 2, the carbapenem resistance of GNB shows 66.25% to imipenem, 56.87% to meropenem and 81.25% to doripenem, respectively. More than 57% of them were resistant to carbapenems.

It was an observation that 83% (10/160) of Acinetobacter spp. were carbapenemase producers. Similarly, Burkholderia spp. (6%), E. coli (94%), Enterobacter aerogenes (100%), Klebsiella spp. (97%), Proteus spp. (67%), Pseudomonas (52%) and Spingomonas spp. (33%) were carbapenemase

Table 3: Flagging of Carbapenamase activity shown by Advance Expert System (AES) of Vitek-2 (n=160)

Sr.No.	Name	Positive*	%	Negative	%
1	Acinetobacter spp (n=12)	10	83%	2	17%
2	Burkholderia spp (n=16)	1	6%	15	94%
3	E.coli (n=18)	17	94%	1	6%
4	Enterobacteraerogenes (n=4)	4	100%	0	0%
5	Klebsiella spp (n=35)	34	97%	1	3%
6	Proteus spp (n=3)	2	67%	1	33%
7	Pseudomonas spp (n=66)	34	52%	32	48%
8	Sphingomonas spp (n=6)	2	33%	4	67%
	Total	104	65%	56	35%

**Flagged by AES of Vitek-2 compact.*

Table 4: Carbapenamase by AES of Vitek system and Genotypic Comparison

	Carbapenamase detection by AES*1	Carbapenamase(MBL) detection by Multiplex PCR (Genotypic)							Total isolates	
		MP-3*2			MP-5*3					
NAME	Carbapenamase +ve	BlaNDM-1	BlaVIM	BlaIMP	VIM Family	SIM 1	IMP Family	SPM 1	GIM 1	
Acinetobacter spp (n=12)	10	-	-	-	-	-	-	-	-	0
Burkholderia spp (n=16)	1	-	-	-	-	-	-	-	-	0
E.coli (n=18)	17	7	-	-	-	-	8	1	8	14
Enterobacter aerogenes (n=4)	2	1	-	-	-	-	-	-	1	1
Klebsiella spp (n=35)	34	15	-	1	5	1	6	-	12	24
Proteus spp (n=3)	2	-	-	-	-	-	-	-	-	0
Pseudomonas spp (n=66)	34	1	1	-	3	1	5		1	5
Spingomonas spp (n=6)	2	-	-	-	-	-	-	-	-	0
Total=160	104	24	1	1	8	2	19	1	22	

*1AES-Advance expert system-Vitek-2 compact, *2 MP-3= multiplexing for 3 primers, *3MP-5= multiplexing for 5 primers

producers. On the whole there was 65% (104/160) of GNBs which were carbapenemase producers, flagged by AES of Vitek-2 compact.

As per Table 4, twenty-four of the GNB showed blaNDM-1 genes, twenty-two had GIM-1, nineteen had IMP family, eight had VIM family, two had SIM-1 and one each had blaVIM, blaIMP and SPM-1.

At the same time, it was also observed during the study that out of 35 Klebsiella spp., in 15 isolates blaNDM-1 was present and in 12 isolates GIM-1 was found. The carbapenamase genes were also detected in E.Coli, Enterobacter aeroginosa, Klebsiella spp. and Pseudomonas spp. These genes were not detected in Acinetobacter spp., Burkholderia cepecia, Proteus spp. and Sphingomonas spp.

The presence of single type of carbapenemase genes was found in E.coli, Klebsiella and Pseudomonas. Commonly found genes in E.coli were blaNDM-1 and IMP family, in Klebsiella there was blaNDM-1, IMP family and GIM-1 genes, and in Pseudomonas there was presence of only IMP family. Whereas there was also an observation that more than one gene was co-existing in single isolate. For example, in one species of E.coli there was presence of three genes i.e., blaNDM-1, IMP family and GIM-1 and in three isolates there was presence of two genes like IMP family and GIM-1, and blaNDM-1 and GIM-1. Likewise, there was also presence of four genes blaNDM-1, blaIMP, IMP family and GIM1 family in one strain of Klebsiella spp. In all there was presence of two to four genes in single isolate of gram-negative bacilli. (Table-5)

Metallo-beta-lactamase (MBL) resistant genes which were detected in this study using multiplex PCR are illustrated on agarose gel

Table 5: Shows presence of single Metallo-beta-lactamase (MBL) genes and Co-Existing genes found in GNB.

Strain	Single gene type								Co-existing										
									No.of genes										
									3	2	2	3	2	2	4	3	2	3	2
	BlaNDM-1	BlaVIM	BlaIMP	VIMFamily	SIM1	IMPFamily	SPM1	GIM1	BlaNDM-1,IMPFamily,GIM1	IMPFamily,GIM1	BlaNDM-1,GIM1	BlaNDM-1,GIM1,SPM1	BlaNDM-1,VIMFamily	VIMFamily,GIM1	BlaNDM-1,BlaIMP,IMPFamily,GIM1	IMPFamily,VIMFamily,GIM1	IMPFamily,SIM1	BlaNDM-1,VIMFamily,SIM1	BlaVIM,VIMFamily
E.coli	2					4			1	3	3	1							
Klebsiella spp	6					2		3		1	5		3	1	1	1	1		
Pseudomonas spp						2				1								1	1
Enterobacter aerogenes											1								

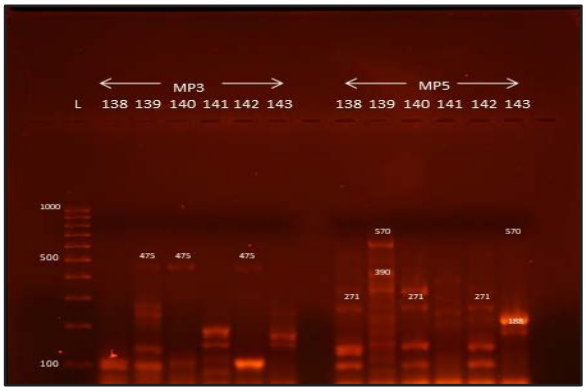

Figure 5: Gel documentation for the amplified products: 138,140,141 & 143 – klebsiella, 139-Pseudomonas, 142-E.Coli

electrophoresis.(Figure 5) Multiplex-3 had primers for blaNDM-1, blaVIM and blaIMP and MP-5 had primers for IMP, VIM, GIM, SPM and SIM genes. Three isolates of Klebsiella spp showed presence of either single MBL gene or double genes. Whereas Pseudomonas showed the presence of three genes namely blaNDM, SIM & VIM. E.coli showed blaNDM and GIM genes. There was presence of blaNDM genes in pseudomonas, Klebsiella and E.coli.

Discussion

Antimicrobial resistance is a major threat to patients admitted in the hospital. Carbapenems have been the last resort against the multidrug resistant gram-negative pathogens, cephalosporinase or extended spectrum beta-lactamase producing Enterobacteriaceae. In the line with the increased global burden of Carbapenemase Producing Enterobacteriaceae (CPE) over the period of recent years, and the production of various types of carbapenemase are increasingly reported in Klebsiella pneumonia and Escherichia coli.

The adaptable carbapenemase is the New-Delhi Metallo-beta-lactamase (NDM-1) is an adaptable molecular class B lactamase. The other normal metallo-beta-lactamase which has been a sensational expansion in the spread of obtained or adaptable families incorporates SIM, IMP, GIM and VIM proteins. The history of VIM (Verona Integron encoded MBL) is that it was isolated in France in 1997 and is most widespread MBL in Pseudomonas aeruginosa. SPM-1 (for Suo Paulo metallo beta lactamase) isolated from Sao Paolo, Brazil. This has caused multiple hospital outbreaks with high mortality in Brazil. The most recent group of procured metallo beta-lactamase from Korea is enzyme SIM-1 which means "Seoul imipenamases" has the nearest character to the IMP family was found when imipenem safe Pseudomonas spp. also, Acinetobacter spp were screened. Adaptable imipenem obstruction was first identified in Japan in 1990 in Pseudomonas aeruginosa and in B.fragilis. IMP-1(for dynamic on imipenem) found adaptable plasmid in P.aeroginosa clinical separates was found on S.marcescence and other Enterobacteriaceae. Its hydrolysis imipenem, penicillin's, extended spectrum cephalosporins but not aztreonam. Now IPM family has been found throughout the world. Identification of GIM-1 (for German imipenamases) has 30% homology to VIM, 43% to IMP & 20% to SPM. When this was isolated in 2007, it was not reported in elsewhere in the world. It is found in clones of Pseudomonas aeruginosa within class1 integrons in plasmid. Since their underlying

discovery (2007), SPM, SIM, GIM metallo-beta-lactamase have not spread past their nations of beginning. In any case, IMP and VIM keep on being distinguished around the world, the pattern moving past the metallo-beta-lactamases from P.aeruginosa and into the Enterobactereceae from P.aeruginosa.[2]

Our study showed out of 160 isolates of gram-negative bacilli , 41% were pseudomonas, 22% were Klebsiella pneumonia, 11% were E-Coli, 10% were Burkholderia cepeceae , 6% were Acinetobacter Bahmani, 4% were spingomonas paucimovilis etc. All these isolates were resistance to Imipenem, Meropenem and Doripenem and the percentage resistance was 66.25 %, 56.87 % & 81.25 % respectively. The study conducted by V Manchanda et al showed 90 % and 71 % to Imipenem and meropenem resistances respectively, in contrast to our study.[3]

The amplification product of polymerase chain reaction assay showed 475 bp amplicon product for NDM-1 gene. Out of the 104 GNB which were resistant to carbapenems, 24 isolates (23 %) were having the blaNDM-1 genes in our study. The isolates were from samples received from different infectious sites of patients.

Study conducted by Shyam sundar grover et al showed 50 % presence of blaNDM-1 gene in three of six isolates of E.coli.[4] Several other studies from India reported high incidence of blaNDH like enzyme production among carbapenem resistant E-coli from hospitals. Deshpande et al reported blaNDM-1 in nine E.coli isolates among 24 carbapenem resistant Enterobacteriaceae (37.5%).

In the study done by Mariappan shanthi et al in 2014 showed that among the Pseudomonas isolates blaNDM-1 coexisted with blaVIM in one isolate. They got blaVIM alone in 33 isolates and 2 isolates conveyed the blaIMP quality alone. Further blaGIM, blaSPM, blaSIM were not identified in any of their reviews isolates.[5] This study was only on the isolates of Pseudomonas spp, whereas our study included Gram negative bacilli belonging to Enterobacteriaceae and others, where in we found single genes of blaNDM-1 and blaIMP and blaGIM in E.coli, Klebsiella and Pseudomonas spp. coexistence of MBL genes was in E coli, Klebsiella, Pseudomonas and Enterobacter aerogenes. Among carbapenemase, metallo-β-lactamases (MBLs) are of prime significance for the locale under study in view of the development of new variations of MBL, for example, New Delhi metallo-β-lactamase (NDM) and different IMP variations from the subcontinent. MBLs have a place with class B carbapenemase as per the Ambler grouping framework. blaVIM, blaNDM, and blaIMP are significant MBL quality groups that are conveyed by versatile plasmids viable with a huge range of clinically significant microbes.[6]

The previously mentioned system of conjunction of genes has been accounted for in Klebsiella pneumoniae, Escherichia coli, Salmonella spp. and Enterobacter spp. Plasmids containing the blaNDM-1 gene have been seen to coexhibit gene for CTX-M, OXA-1, and TEM-1enzymes. Major ESBL and MBL qualities, including blaSHV, blaCTX-M, blaTEM, and blaOXA-51, and genes for the IMP-family and VIM family, have been accounted for to exist together in clinically safe Acinetobacter baumannii in Iran.[7] Notwithstanding, one review inferred that there was no critical connection among ESBL and MBL creation genes.[7] Ertapenem-resistant, ESBL-creating Klebsiella pneumoniae isolates have been accounted for in Italy and displayed to convey novel porin variations that added to the decreased vulnerability of disengages to meropenem and imipenem.Our study didn't include detection of ESBL thus there is no mention of these genes. Surprisingly, Acinetobacter baumanii and Burkholderia cepeceae did not demonstrate the presence of MBL genes.

In our study K. pneumonia is the fundamental supply of assorted plasmid-borne bla genes coding for carbapenemase, i.e., B-lactamases that hydrolyze practically all accessible b-lactams including carbapenems. The most significant being the KPCs and the metallo-b-lactamases (MBLs) NDM, VIM, IMP, GIM, and SIM happening separately or in existing together structures.

Treatment Agents: Polymyxins and tigecycline are the antimicrobials regularly utilized for the treatment of contaminations brought about via carbapenem-resistance Enterobacteriaceae. Polymyxin B has been utilized in clinical practice. This was found in excess of 50 yrs. back, has been re-found as an important helpful specialist with viability against multidrug-resistance GNB because of lack of new antimicrobials with exercises against these living beings. Exactly the same thing works out in a good way for the utilization of colistin provoking clinicians to rethink its utilization and is utilized in carbapenem-safe microscopic organisms. Colistin and tigecycline don't synergistically affect carbapenem-resistance Klebsiella in vitro, they are not hostile and may have an added substance impact when utilized together and forestall the rise of protection from these antibiotics. their mix or monotherapy can be utilized in multi-drug resistance P aeruginosa because of efflux. The constraint of tigecycline as a treatment choice in urinary tract contaminations and circulatory system diseases is notable.[8]

Clinical impact and Infection control: The issue of carbapenemase-intervened resistance escalated once genes for these compounds became related with obtained hereditary versatile components like plasmids and integrons. Natural organic entities might give hereditary material as a wellspring of these

enzymes and clinical strains might discard this data with the assistance of a portable piece of hereditary material (transposon) inside the medical clinic setting and into the general climate. Contamination with carbapenemase-creating Enterobacteriaceae (CRE) is arising as a significant test in the medical care setting. The rise and spread of carbapenem-resistance GNB forms are troubling general wellbeing advancement and highlight the prompt requirement for forceful location and control systems. Patients with unnoticed CRE colonization in the normal flora have filled in as repositories for transmission during medical care related episodes. Outbreaks with carbapenemase producing organisms have been due to lack of adherence of infection control measures. The flare-up of diseases with CRE recommends that early identification using designated observation and the presentation of severe contamination control measures including support of hand cleanliness and contact safeguards can assist with controlling the spread of these microbes.[8]

Conclusion

Taking everything into account, MBL-delivering bacterial isolates are arising quickly around the world. An incredible number of carbapenem-safe clinical bacterial species are impervious to a large portion of the usually utilized antibacterial agents, showing the ascent of super-microbes and their skillet protection from antimicrobial treatment. Deciding the resistance systems and the main driver for their end are vital. Carry out the normal screening of MBLs and ESBLs in lab methodology before anti-microbial treatment starts. Further investigations are needed to determine different sorts of quality variations pervasive among clinical isolates in our area for the ramifications of medicine in clinical settings. The present study indicated that the Pseudomonas and Enterobacteriaceae (E. Coli and Klebsiella) were carbapenemase producers in our group of cancer patient. The data presented showed that not only blaNDM-1 were responsible but also the other genes co-existed which contributed to the resistance.

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Conflicts of interest: There are no irreconcilable situations.

References

1. Papp-Wallace KM, Endimiani A, Taracila MA, Bonomo RA: Carbapenems: past, present, and future. Antimicrobial Agents and Chemotherapy 2011; 55:4943-4960.
2. Queenan AM, Bush K: Carbapenemases: the versatile β-lactamases. Clinical Microbiology Reviews 2007; 20:440-458
3. Manchanda V, Rai S, Gupta S, et al: Development of TaqMan real-time polymerase chain reaction for the detection of the newly emerging form of carbapenem resistance gene in clinical isolates of Escherichia coli, Klebsiella pneumoniae, and Acinetobacter baumannii. Indian Journal of Medical Microbiology 2011; 29:249-253
4. Grover SS, Doda A, Gupta N, et al: New Delhi metallo-β-lactamase-type carbapenemases producing Escherichia coli isolates from hospitalized patients: A pilot study. The Indian Journal of Medical Research 2017; 146:105-110
5. Shanthi M, Sekar U, Kamalanathan A, Sekar B: Detection of New Delhi metallo beta lactamase-1 (NDM-1) carbapenemase in Pseudomonas aeruginosa in a single centre in southern India. The Indian Journal of Medical Research 2014; 140:546-550
6. Ain NU, Iftikhar A, Bukhari SS, et al: High frequency and molecular epidemiology of metallo-β-lactamase-producing gram-negative bacilli in a tertiary care hospital in Lahore, Pakistan. Antimicrobial Resistance & Infection Control 2018; 7:1-9
7. Nordmann P, Dortet L, Poirel L: Carbapenem resistance in Enterobacteriaceae: here is the storm! Trends in Molecular Medicine 2012; 18:263-272
8. Codjoe FS, Donkor ES: Carbapenem resistance: A review. Medical Sciences 2018; 6:1

Isolation of Different Non-Lactose Fermenting Gram Negative Bacilli (NLFGNB) and their Antimicrobial Resistant Pattern: A Retrospective Analysis

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Summary

The emergence of gram-negative bacilli causing infections has created untreatable problems due to increasing antibiotic resistance thus complicating cancer treatment, prolonging stay in the hospital and escalating the burden of cost on the patients. Our study of retrospective analysis was focused on the common and uncommon pathogenic isolates of non-lactose fermenter gram negative bacilli (NLFGNB) to know the prevalence of different isolates which were less (42%) in contrast to lactose fermenter gram negative bacilli (58%). All the standard microbiological methods were followed including the identification (ID) and antibiotic susceptibility testing (AST) by Vitek-2 and the analysis was for a period of two years (2019-2020). The isolates identified were Pseudomonas 59%, Acinetobacter 23%, Burkholderia 10%, and least isolation was of Sphingomonas 5% and proteus 3%. The most uncommon NLFGNB isolated were only two, namely Achromobacter xylosoxidans and Elizabethkingia. Antimicrobial resistance showed that more than 50% of them were MDR (Multiple Drug Resistance) (MDR is shown by at least one antimicrobial drug in three or more antimicrobial category). Even though there were two unusual bacilli isolated (Achromobacter, Elizabethkingia) they were sensitive to most of the antimicrobitics but 100% of Achromobacter spp. (single isolate) was resistant to gentamycin and aztreonam. Forty to fifty percent of pseudomonas spp were resistant to carbapenems, aminoglycosides, and quinolones. Thirty to forty percent of them were resistant to betalactam+betalactamase inhibitors (BL+BLI) and to also third and fourth generation cephalosporins. Acinetobacter species had 6.6% to 28% resistance to tigecycline, minocycline and colistin. In other words, around 93.4% to 72% were sensitive and can be drug of choice to treat infections caused by them. Ninety percent of Burkholderia spp were resistant to betalactam+betalactamase inhibitors (BL+BLI) and 25% - 28% of them were resistant to carbapenems. Ninety to hundred percent of Proteus spp. were resistant to minocycline and tigecycline. To carbapenems there was low and high resistance like 27.6% to meropenem and 77.4% to imipenem. Sphingomonas Paucimobilis showed 39.7% to 77.4% resistant to most of the panel of antibiotics used. In conclusion, there was isolation of NLFGNBs which are multidrug resistant and complicating the treatment of cancer patients. There in a need for development of clinico-microbiological meetings and discussions to prevent the spread of antibiotic resistant NLFGNB from patient to patient and form antibiotic policies through antibiotic stewardship program (ASP).

Keywords: NLF, GNB, Antibiotic Resistance, Cancer, MDR.

Introduction

Non-Lactose fermenting gram-negative bacilli (NLFGNB) emerged as important health care associated infections leading to morbidity in the patients. Risk factors associated with the surge of these infections are prolonged hospital stay, lack of antibiotic policies, lapses in asepsis and unhygienic conditions prevailing in most of the hospitals.

Most common NLFGNB isolated from the patients are Pseudomonas aeruginosa, Acinetobacter baumanii, Burkholderia cepacia, Morganella morganii, Proteus mirabilis and Salmonella typhi.[1] Pseudomonas aeruginosa is an important and common pathogen in hospitalized patients, causing treatment failure due to its multiple resistant mechanisms in critically ill patients specifically in intensive care units and in wards because of its ubiquitous nature and ability to survive in moist hospital environment. It has multifactorial resistance mechanism like mutations in genes encoding porins, efflux pumps, penicillin-binding proteins, and chromosomal β-lactamase production, ESBL (Extended Spectrum Beta- Lactamases), carbapenemase etc.[2] Strains of P. aeruginosa are the cause of several diseases predominantly pneumonia, bacteraemia, meningitis, urinary tract infections, as well as skin and soft-tissue infections Sphingomonas paucimobilis, also an opportunistic pathogen that take advantage of underlying conditions and causes infectious disease. Burkholderia cepacia is an aerobic gram-negative bacillus found in various aquatic environment and has low virulence and is a frequent colonizer of fluids used in the hospital (e.g., irrigation solutions, intravenous fluids).

Thus, the purpose of this study was to know the prevalence of non-lactose fermenting gram negative bacilli causing super or opportunistic infections and its antibiotic susceptibility pattern in

cancer patients, thereby helping the clinicians for treating the patients

Methods and Materials

This retrospective study was conducted at the microbiology laboratory of The Gujarat Cancer & Research Institute for a period of 2 years (2019 to 2020). Patient's infectious samples were received in the laboratory who were suffering with different types of infections in clinically diagnosed cancers. All standard Microbiological methods were followed for isolation, preliminary identification, and further identification (ID), and antibiotic susceptibility testing (AST) performed by automated system called Vitek-2 compact. The results were entered in software of WHO Net, and data was analysed for antibiotic resistance pattern of NLFGNB.

Results

A total of 3121 gram-negative bacilli were isolated during the retrospective analysis. Out of these 58 % (1820/3121) were lactose fermenters and 42 % (1301/1321) were non lactose fermenter. (Figure 1) The isolation of NLF gram negative bacilli were from different patients' diagnosis having post operate infection (34%), leukaemia patient (20%), head & neck cancers (18%), gynec & gastrointestinal cancer (8%).(Figure 2)

As per Figure 3, out of 1301 NLFGNB, Pseudomonas isolated were 59%, Acinetobacter spp. were 23%, Burkholderia were 10%, Sphingomonas was 5%, and Proteus were 3%. There were two rare bacilli which were isolated. One each of Achromobacter zylosoxidans and Elizabethkingia.

Analysis of antimicrobial resistance was done for all 1299 NLFGNB (in general) and individual isolates. Combined antimicrobial resistance of all NLF GNB showed that 77.4% were resistant to aztreonam, around 53-55% resistance was observed for co-trimoxazole and ciprofloxacin. Around 40-49% resistance was seen for antibiotics like cefaperazone/sulbactam, ticarcillin /clavulanic acid, piperacillin/tazobactam, ceftazidime, cefepime carbapenems. There was 14-27% resistance to tigecycline, minocycline and colistin respectively.(Figure 4) The two isolates of NLF GNB, showed sensitivity to all the antibiotics, except single isolate of Achromobacter species showed 100% resistant to gentamycin & aztreonam.

Now looking into the individual species, antimicrobials resistance of pseudomonas spp (n=763) showed that 54% resistance was there for co-trimoxazole, around 40-50% resistance to carbapenem, aminoglycosides, quinolone, and 30-40% resistant to BL (Beta Lactamases) +BLI (Beta

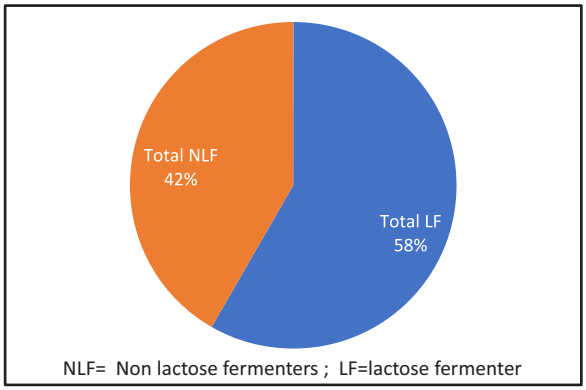

Figure 1: Differential growth of gram-negative bacilli on MacConkey agar (n=3121)

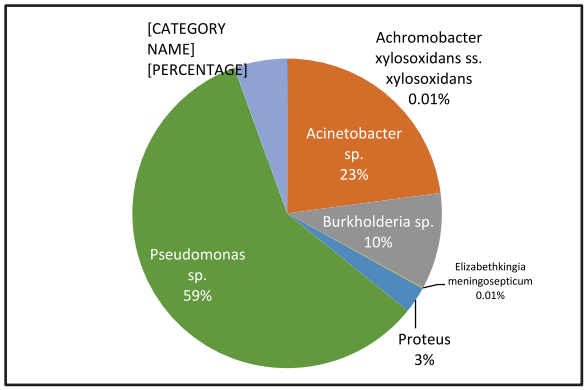

Figure 3: Different Isolates of Non-lactose fermenter Gram negative bacilli (n=1301)

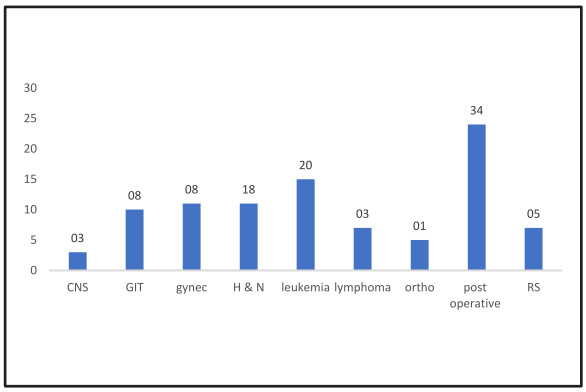

Figure 2: Percentage of Non-lactose fermenter GNBs in different cancers (n=1301)

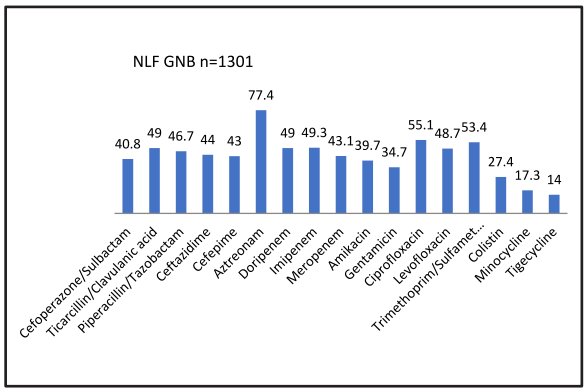

Figure 4: Antibiotic Resistant pattern of all NLF GNBs.

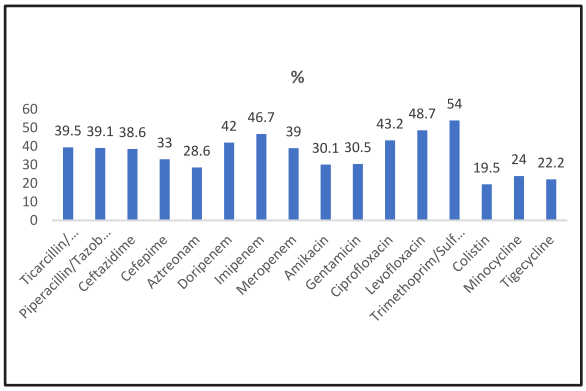

Figure 5: Antibiotic Resistant pattern of Pseudomonas Spp (n=763)

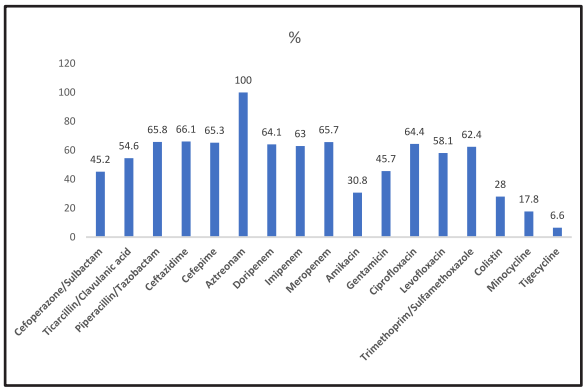

Figure 6: Antibiotic Resistant pattern of Acinetobacter spp (n =297)

Lactamases Inhibitor) like ticarcillin clavulanic acid, piperacillin / tazobactam, 3rd and 4th generation cephalosporins.(Figure 5)

In the case of Acinetobacter baumanii (n=297) isolates, they were 100% resistant to aztreonam, (50-66.1%) ticarcillin/clavulanic acid, piperacillin/tazobactam (65.8%), ceftazidime (66.1%), cefepime (65.3%), carbapenems (65.7%), ciprofloxacin (64.4%), levofloxacin (58.1%), co-trimoxazole (62.4%). Nonetheless, there was least resistance between 6.6% to 28% to tigecycline (6.6%) minocycline (17.8%) and colistin (28%).(Figure 6)

The NLF Burkholderia spp showed more than 90% resistance to BL+BLI. It was good that there was less resistance to carbapenems (25-28%), levofloxacin (32.2%) and tigecycline (21.3%) and minocycline (9.5%).(Figure7)

Non-lactose fermenting Proteus species have always been an issue and complicates admitted patients causing hospital acquired infections. The scenario is contrary to other NLFs showing that 90-100% resistance is seen to antibiotics like colistin, minocycline & tigecycline, to carbapenems there was low & high resistance to meropenem (27.6%) and imipenem (77.4%). To rest of the antibiotics like BL-BLI, Proteus was less resistant varying from 20 to 24.2%. (Figure 8)

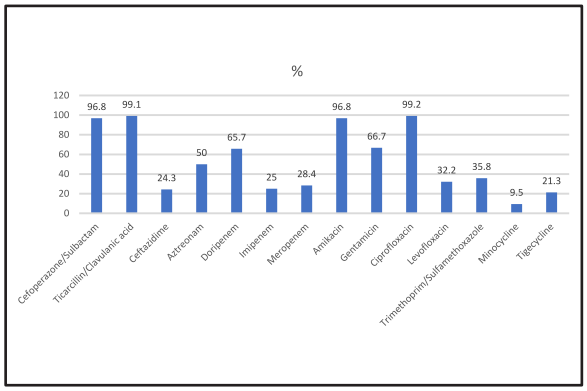

Figure 7: Antibiotic Resistant pattern in Burkholderia Sp.(n=131)

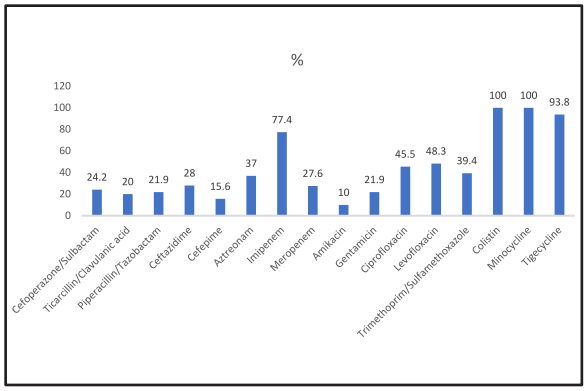

Figure 8: Antibiotic Resistant pattern of Proteus sp. (n=36)

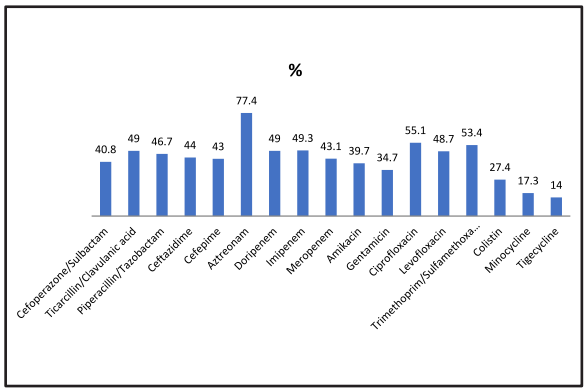

Figure 9: Antibiotic resistant pattern of Sphingmonas sp. (n=72)

There were (72) isolates of Sphingomonas paucimobilis and there was Sphingomonas showing 39.7% to 77.4 % resistance to most of the antibiotics.(Figure 9) But to tigecycline, minocycline and colistin the percentage resistance was 14%, 17.3% and 27.4% respectively.

As per Table 1 comparison was done between NLF GNB in total and singly isolated bacilli. Grouping of percentage resistance was done having above 50% and below 50% and were again grouped as per the Table 1. Fifty percent antibiotic resistance of other single NLFGNBs which ranged from 10-20%, for Pseudomonas was colistin and minocycline, for Acinetobacter and Sphingomonas was tigecycline, for

Table 1: Comparative antibiotic Resistance of all NLF GNBs in General and Individual Isolates

Resistant Range	AllNLF	Pseudomonas	Acinetobacter baumanii	Burkholderia	Proteus	Sphingomonas
Above 50%	Aztreonam-774% Ciprofloxacin-55.1 % Co-trimoxazole-53.4%	Co-trimoxazole–54%	Ticarcillin / Clavulanicacid Piperacillin/ Tazobactam Ceftazidime Cefepime	Cefoperazone/ Sulbactam Ticarcillin/ Clavulanicacid Aztreonam Doripenem Amikacin Ciprofloxacin Gentamicin Piperacillin/ Tazobactam Ceftazidime Cefepime Levofloxacin Trimethoprim/ Sulfamethoxazole	Imipenem Colistin Minocycline Tigecycline	Aztreonam Ciprofloxacin Co-trimoxazole
40–50%	Cefoperazone/ Sulbactam Ticarcillin/ Clavulanicacid Piperacillin/ Tazobactam Ceftazidime Cefepime Doripenem Imipenem Meropenem Levofloxacin	Doripenem Imipenem Ciprofloxacin Levofloxacin	Cefoperazone/ Sulbactam Gentamicin		Ciprofloxacin Levofloxacin	Cefoperazone/ Sulbactam Ticarcillin / Clavulanicacid Piperacillin / Tazobactam Ceftazidime Doripenem Imipenem Meropenem Levofloxacin Cefepime
30–40%	Amikacin Gentamicin	Ticarcillin / Clavulanicacid Piperacillin / Tazobactam Ceftazidime Cefepime Meropenem	Amikacin	Levofloxacin Co-trimoxazole	Aztreonam Co-trimoxazole	Amikacin Gentamicin
20–30%	Colistin	Cefoperazone/ Sulbactum Aztreonam Minocycline Tigecycline	Colistin	Cefepime Imipenem Meropenem Tigecycline Colistin	Cefoperazone / Sulbactam Ticarcillin / Clavulanicacid Piperacillin / Tazobactam Ceftazidime Meropenem Gentamicin	Colistin
10–20%	Minocycline Tigecycline	Colistin	Minocycline Tigecycline	Minocycline	Amikacin Cefepime	Minocycline Tigecycline

Burkholderia was minocycline and for proteus was amikacin and cefepime.

Discussion

Non-Lactose fermenting gram negative bacilli (NLFGNB) have now become multidrug resistant to most of the panels of antibiotics been used for invitro antibiotic susceptibility testing given as per CLSI guidelines. Once considered as contaminants are now gaining importance and emerging as health care associated infections.

In our study, the NLFGNB isolation was 42% in contrast to the study conducted by Grewal et al, where they found 11.6% (216) yield of NLF GNB out of 1854 culture-positive samples.

Two hundred and sixteen (11.6%) yielded NFGNB.[4] Since our institute is dedicated cancer centre and we have tumour surgeries done the isolation of NLFGNB were maximum (34%) from post-operative wounds.

Isolation of Pseudomonas sp. by Grewal et al was 87.96% whereas in our analysis we found much less (59%) when compared to them.[4] The other NLFGNB like Acinetobacter sp. isolation was more with us (23%) than when compared to study of Grewal et al, where the isolation of Acinetobacter was 7.87%.

The current study analysed antimicrobial sensitivity testing by focusing on the resistance pattern of all the NLF GNB in general and individual species and analysis showed that 40-50 % of them were resistant to BL + BLI, 3rd generation, 4th generation cephalosporin, carbapenems and

fluroquinolone, levofloxacin. Overall, the best drugs that can be used for treatment are minocycline, tigecycline (tetracycline group) and colistin (CLSI 2019-2020). But the latest guideline of CLSI 2021 does not interpret sensitivity to colistin. Only interpretation as intermediate and resistant in given. In such cases the recommendation for pseudomonas is to give loading dose and maximum renal adjusted dose. Clinical and PK/PD data demonstrate colistin and polymyxin B have limited clinical efficacy even if intermediate result is obtained. Alternative agents were strongly preferred. Consultation with an infection diseases specialist is recommended in such cases.

Antimicrobial resistance of pseudomonas species was much less (<40%) to most of the antibiotics like ticarcillin/clavulanic acid, piperacillin/tazobactam, ceftazidime, minocycline, tigecycline, aztreonam, cefoperazone/sulbactam etc, unlike the study conducted by Grewal et at, where they had MDR pseudomonas aeruginosa. Our analysis showed that Burkholderia spp were multidrug resistant where in the resistance was more than 50% to most of the antibiotics and only 25% resistant to imipenem (i.e.,75% sensitive) and the results of Grewal et at showed 10% sensitivity to imipenem.[4]

A.baumanii showed maximum susceptibility to the imipenem (88.2%) followed by cefoperazone/sulbactam in Grewal et at study.[4] Whereas our study showed maximum susceptibility 37% (as 63% were resistant) to imipenem and to cefoperazone/sulbactam 54.8% (as 45.2% were resistant) were sensitive. Since our study focused on resistant pattern, we could analyse that Acinetobacter showed maximum sensitivity to minocycline, tigecycline (10-20% resistance) and amikacin (30-40 % resistant). Therefore, these drugs can be considered to patients who are critically ill.

Antimicrobial stewardship: In the early era of antibiotics there were only fourteen new classes of antibiotics between 1935 and 2003. This led to overuse and misuse and the impact is the development of antimicrobial resistance. After the exhaustion of the development of newer drugs there is now a method of conserving the antibiotics. Therefore, antimicrobial stewardship (ASP) has come into being wherein there is optimal selection of doses and duration of antimicrobial treatment that results in the best clinical outcome for the treatment and prevention of infection with minimal toxicity to the patient or minimal impact on subsequent resistance. Thus, the goals of ASP are three:

1) To work with the health care practitioner to help each patient receive the most appropriate antibiotic with correct dose and treatment.
2) To prevent antimicrobial overuse, misuse, and abuse.
3) To have prescription audits to know the prevalent use of different antibiotics.
4) To minimise the development of resistance both at the individual patient level and at the community level.

Conclusion

Infections caused by gram negative bacilli (GNB) is gradually increasing the morbidity and mortality. Unresolved postoperative infections caused by opportunistic GNB are increasing hospital stay and expenditure on costly antibiotics. Immunocompromised leukemic patients are prone to opportunistic infections due to the multidrug resistant bacteria leading to mortality.

Our study focused on the infections caused in cancer patients by NLF GNB complicating the cancer treatment, which accounted for around 42% isolation amongst all the Gram-negative bacilli.

Though Sphingomonas, Proteus and Burkholderia isolation is less as compared to Pseudomonas and Acinetobacter spp they were multidrug resistant, and this raises concern of rapidly spreading of these bacteria in the hospital leading to emergence of outbreak of uncontrolled infection.

Thus, it is necessary to have frequent and ongoing screening of these bacteria, regular assessment of antibiotic susceptibility profiles and judicious use of antibiotics are recommended for effective management of infection caused by NLF GNB or any other bacteria and limiting emergence of multi drug resistance.

References

1. Lambert P A: Mechanisms of Antibiotic Resistance in Pseudomonas aeruginosa. Journal of the Royal Society of Medicine 2002; 95:22-26
2. Ozer B, Tatman-Otkun M, Memis D, Otkun M: Characteristics of Pseudomonas aeruginosa Isolates from In-tensive Care Unit. Central European Journal of Medicine 2009; 4:156-163
3. Wroblewska M: Novel Therapies of Multidrug-Resistant Pseudomonas aeruginosa and Acinetobacter spp. Infections: The State of the Art. Archive Immunological et Therapiae Experimentalise 2006; 54:113-120
4. Grewal US, Bakshi R, Walia G, Shah PR: Antibiotic susceptibility profiles of non-fermenting gram-negative bacilli at a tertiary care hospital in Patiala, India. Nigerian Postgraduate Medical Journal 2017; 24:121-125

Its our Choice…
Be a Duck who Quack or an Eagle to Fly High…

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सर्वं परवशं दुःखं सर्वमात्मवशं सुखम्।
एतद् विद्यात् समासेन लक्षणं सुखदुःखयोः॥

Everything that is in other's control is painful. All that is in self-control is happiness. This is the definition of happiness and pain in short.

जो सब अन्यों के वश में होता है, वह दुःख है।
जो सब अपने वश में होता है, वह सुख है।
यही संक्षेप में सुख एवं दुःख का लक्षण है।

Covid-19 a pandemic that shock the world in 2019.. 2020.. and still continuing its impact on all of us. All of us were affected socially ,economically and mentally and many of dear, near ones were lost by the deadly wave of this disease. Life had taken a U turn and reset button was placed on the speedy thinking of human brain.

We can see this pandemic in two ways: a disaster that destroyed our life and economy and many more things or a lesson to rebound and rise from ashes due a calamity that can struck us any time without a notice.

Covid patient care was a totally new encounter for the physicians across the globe. As a clinician we came across a variety of incidents during the management of these patients. The treatment started with use of various medications including steroids, HCQ and many.. many drugs and we succeeded . we failed and stood back on our legs to learn a new lesson on managing a new disease. There were incidents of drug shortages, shortage of ventilators, limited PPE kits and limited oxygen at various medical centres but this didn't limit the medical professionals to fight back.

The medical industry rose to the occasion and a mass production of PPE kits and medical devices were manufactured at fastest possible speed. A team approach by nursing professionals working day and night, hospital staff support and doctors as key stone fought hard to defeat this disease. Vaccine research, production and distribution at super fast speed and mass vaccination drive to millions was made possible due to efforts of many many known and unknown hands.

Any untoward incident or problem in life is handled by us with anger and complaint against these things. While driving a car on a road, a rash car driver overtakes us without proper traffic rules and we say that traffic in our area is very poor. Secondly at a traffic signal we see the signal turning red just in front of us. These things annoy us and start getting disturbed before start of our work. A second example of a car tyre deflated or technical snag of a gadget used in a car driver and this makes the driver irritated. But there is way around to see the traffic problem; we can enjoy our driving in tough traffic situation by enjoy music on radio and think of making driving better in tough situations. The lessons learnt by this events can lead us to calm and cool life.

The nature got reset due to lock down, limited traffic along the road and the pollution level in various cities went to normal level. On the other hand, travel restriction along with minimal human mobility has impacted the natural environment positively. The carbon emission level got reduced and the air quality had got improved. The COVID-19 has resulted in schools shut down and the children were out of the classroom. Online school, various new teaching modules got invented and improved education system dramatically. There was a rise of e-learning and various teachers learned new skills, knowledge dissemination got globally transmitted. This new approach of online teaching can lead to better and faster spread of information and newer skills.

The choice is ours to behave like a duck to quack and complain regularly about various problems in life leading to tough and distressed life. We can react to a problem with holistic approach and fight with it like an eagle to fly high in life.

आ नो भद्राः क्रतवो यन्तु विश्वतः ।

Let noble thoughts come to me from all directions.

सभी दिशाओं से नेक विचार मेरी ओर आएँ।

Aleukemic Leukemia Cutis: A Rare Case Report with Review of Literature

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Summary

Acute myeloid leukemia can present as an extramedullary disease without bone marrow involvement. There are two extramedullary manifestations of acute myeloid leukemia: (1) Myeloid sarcoma and (2) Leukemia cutis. It is a very unusual presentation and is mostly associated with acute myeloid leukemia. Very rarely it presents with primary skin lesions without bone marrow involvement in such a case it is called aleukemic leukemia cutis. Here we report a 57-year-old male patient who presented with skin lesions and was diagnosed with the help of excisional skin biopsy and immunohistochemistry. Bone marrow examination was normal. He was treated with standard induction chemotherapy "7+3" followed by remission documentation followed by consolidation with intermediate-dose cytarabine.

Keywords: aleukemic leukemia cutis, myeloid sarcoma, acute myeloid leukemia, chemotherapy, primary skin lesions

Introduction

Extramedullary (EM) presentation of acute myeloid leukemia (AML) is unusual. Myeloid sarcoma (MS) and leukemia cutis (LC) represent two well-known EM manifestations.[1] LC presenting before bone marrow involvement of leukemia is very rare. We hereby present a middle-aged male presented with biopsy-proven isolated skin involvement.

Case Report

A 57-year-old male without any comorbidities, ECOG performance status 1 presented with a chief complaint of skin lesions all over the body for 2 months. Skin lesions were nonpruritic nontender, erythematous to violaceous maculopapular, and nodular of varying size scattered all over the trunk, abdomen, upper back, proximal arm, and thigh. The largest lesion over the trunk was approximately 3x3cm. (Figure 1) Lesions were gradually increasing in size and pigmentation. There was no history of fever, weight loss, easy fatigabilitys and cough. On examination, there were multiple erythematous to violaceous macular patches, papules, and nodules over the trunk, abdomen, upper back, proximal arm, and thigh. The remaining general and systemic examinations were within normal limits. On laboratory evaluation, his complete blood count and bone marrow examination were within normal limits.

This patient underwent excision biopsy from skin lesion over trunk which on morphological examination showed basket wave hyperkeratosis and slight irregular acanthosis in the epidermis. There was a dense pan dermal infiltrate composed of medium to large atypical cells displaying a high N/C ratio, round to irregularly contoured nuclei, finely dispersed chromatin, conspicuous nucleoli, and scant cytoplasm. Atypical cell infiltration into the subcutis was consistent with leukemia cutis. (Figure 2)

Immunohistochemical (IHC) staining was suggestive of atypical cells that were diffusely positive for CD34 and MPO (patchy). (Figure 3 and 4) Ki67 was 70-75%. LCA showed weak patchy positivity among atypical cells. Minor populations of reactive lymphoid cell population were seen in the background showing positivity for CD3 CD20 CD4 CD8 CD5 and negative for TDT CK CD30 CD56 CD68 CD117 consistent with LC.

Bone marrow culture metaphase karyotype showed balanced reciprocal translocation between the long arm of the chromosome 16 and 22 between the regions q23 and q13 respectively {karyotype 46 XY, t16;22) (q23; q13)}. (Figure 5) Fluorescent in situ hybridization (FISH) inv16, t(4;11), t(9;11), t(8;21), t(15;17), t(9;22) were not detected. So the diagnosis of aleukemic LC was made. The patient was treated with 7+3 (cytarabine 7 days plus daunorubicin 3 days) AML-specific chemotherapy. Post-chemotherapy on day 21 there was a significant reduction in size, induration, and pigmentation of the skin lesions. (Figure 6)

To document remission whole-body ^{18}FDG PET CT was done, which was suggestive of low-grade FDG avid ill-defined soft tissue thickening along the subcutaneous plane of the right anterior chest wall in the infraclavicular region (SUVmax 2.4), and left lower anterior chest wall (SUVmax 2.5) possibility of neoplastic etiology. Low-grade FDG avid ill-defined soft tissue thickening involving the subcutaneous plane of the sole of right foot at the level of 1st toe inflammatory etiology likely. No evidence of

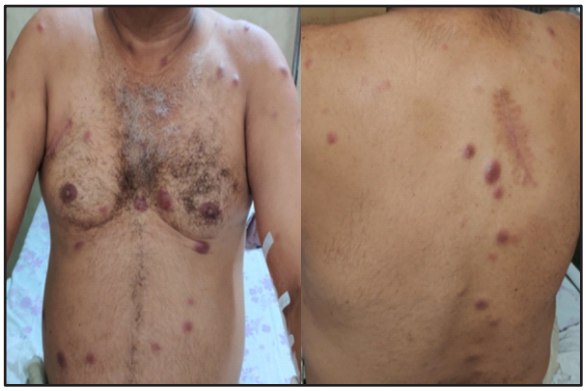

Figure 1: Erythematous to violaceous maculopapular and nodular lesions of varying size over trunk, abdomen, upper and lower back seen at the time of presentation

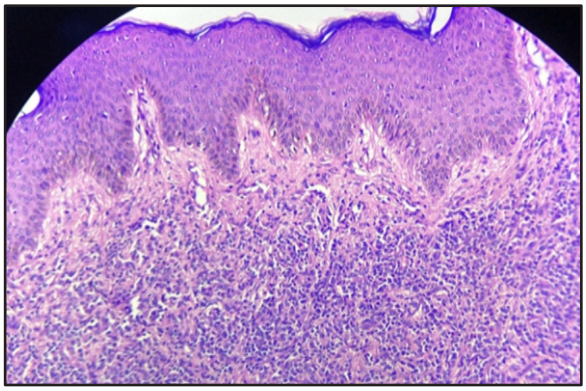

Figure 2: Skin excisional biopsy Hematoxylin and Eosin stain-Dermis showing diffuse infiltration of malignant tumor cells of varying sizes. The tumor cells are medium to large with an increased N:C ratio and irregular nuclear contour. (magnification 200x)

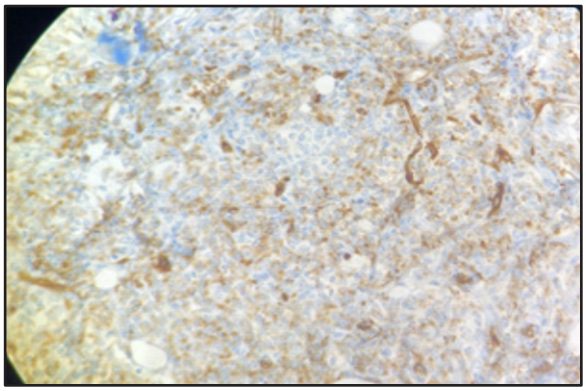

Figure 3: Immunohistochemistry staining-CD 34 positive staining (magnification 100x)

metabolically active disease elsewhere in the body. Repeat biopsy was done from the left anterior chest wall, suggestive of no residual tumor evidence. His Lumbar puncture was negative for malignant cells. After remission documentation cytarabine consolidation was started. The patient has received 2 dosages of intermediate-dose cytarabine as a consolidation therapy till date currently he is clinically well and shows no evidence of disease. The patient has been counselled for the potential role of

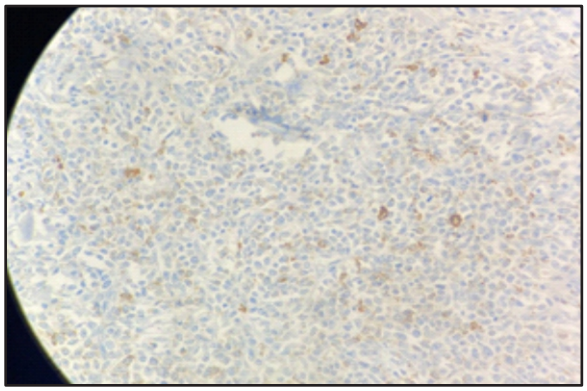

Figure 4: Scattered cells with cytoplasmic MPO positive (magnification 100x)

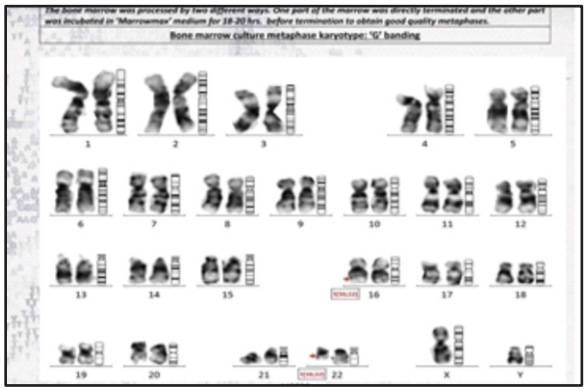

Figure 5: Cytogenetics -46XY, t(16;22)(q23;q13)

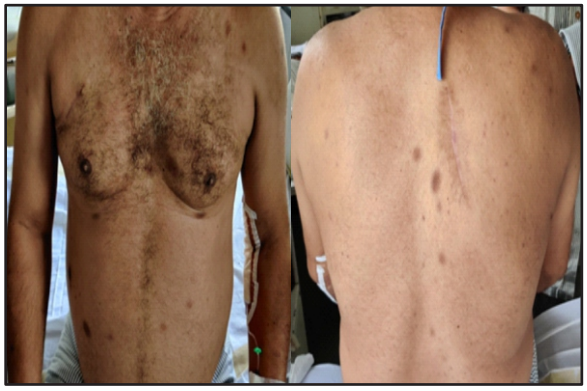

Figure 6: Significant reduction in size, induration, and pigmentation of lesions seen on post-induction chemotherapy day 21

allogeneic hematopoietic stem cell transplantation (HSCT).

Discussion

AML can present as an EM disease without bone marrow disease.[1] EM presentation is a very rare scenario and presents diagnostic challenges. There are two EM manifestations of acute leukemia (1) MS and (2) LC.

MS is a rare EM tumor of immature myeloid cells.[1] The term was first coined in 1811[2] and later called "chloroma" by King[3] in 1853 due to its green color caused by the presence of myeloperoxidase.[3]

LC occurs due to infiltration of the skin by the leukemic myeloid cells resulting in a nodular rash which is also known as cutaneous granulocytic sarcoma.[1] It is seen with higher frequency in children than adults. Approximately 25-30% of infants with congenital leukemia presents with skin involvement,[4] two-third of which are AML.[5] LC is seen in approximately 3% of patients with AML and uncommonly in chronic myeloproliferative diseases.[6] Approximately 50% of cases are of acute myelomonocytic leukemia or acute monocytic leukemia, also known as FAB subtypes AML M4 and M5 respectively.[6]

The usual mode of presentation of LC is as a diffuse papulonodular rash that is more common on the lower limbs followed by upper limbs and then the trunk.[1] AML can also presents with wide range of non-specific skin lesions such as are macules, papules, vesicles, pyoderma gangrenosum, vasculitis, neutrophilic dermatitis (Sweet syndrome), cutis verticis gyrata, and erythema multiforme or nodosum.[7] LC can present with, following, or very seldomly preceding systemic leukemia.[8] When it precedes systemic leukemia it is called "aleukemic leukemia cutis"(ALC).[8] Due to the non-specific presentation of the disease, the skin biopsy can be extremely helpful in the diagnostic work-up,[9] which on histopathology appears as diffuse infiltration of large cells with large nuclei and plentiful cytoplasm (myeloblast).[1] For confirmation of the diagnosis IHC stains, flow cytometry, FISH, and molecular analysis plays a crucial role.[1] Isolated EM AML is considered as a forerunner of medullary AML. Median time to progress to medullary AML ranges from 5-12 months.[1]

In our case, the patient has consulted a dermatologist at a private hospital and took treatment for around 1 month. After not seeing any improvement dermatologist advised to do a biopsy of the skin lesion, which was suggestive of atypical myeloid cell infiltration. He was referred to a private oncology hospital for further treatment, where IHC was done, suggestive of leukemia cutis. After that, patient has consulted our hospital for further treatment. ALC must be differentiated from other lymphoproliferative disorders by doing the IHC test.[7] In our case, the IHC report demonstrated positivity for CD34 and MPO, which was confirmative of the diagnosis of LC.

The management of isolated EM AML is similar to medullary AML with remission induction therapy. The goal is to eradicate EM leukemia and any clinically obscure disease in the marrow.[10] There is no predefined consolidation approach but chemotherapy-based consolidation can be considered in fit patients.[10] The role of allogeneic HSCT has not been evaluated very well in this setting but it is an option for fit patients who can tolerate the procedure. The aim should be eradicating the systemic disease with intensive chemotherapy and/or HSCT.[10] At present, the recommendation is to consider treatment similar to that of patients with the medullary disease.[10] When HSCT is not planned, a patient needs to be treated like conventional AML-type chemotherapy according to standard age, cytogenetic, and molecular-based risk stratifications.[11] LC in patients with AML is associated with decreased overall survival and leukemia-specific survival suggestive of poor prognosis.[12]

Conclusion

Aleukemic leukemia cutis is a very rare presentation of AML. High suspicion, early diagnosis, and treatment can have good outcomes. At present, there are very few case reports of this rare entity so large randomized studies comparing various treatment modalities are the need of the hour.

Declaration of Patient Consent

The authors certify that they have obtained all appropriate patient consent forms. In the form the patient has given his consent for his images and other clinical information to be reported in the journal. The patient understands that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

References

1. Solh M, Solomon S, Morris L, Holland K, Bashey A: Extramedullary acute myelogenous leukemia. Blood Rev 2016; 30:333-339
2. Burns A: Observations of Surgical Anatomy in Head and Neck. Edinburgh, United Kingdom Thomas Royce & co 1811;364-366
3. King A: A case of chloroma. Monthly J Med 1853; 8:97-104
4. Resnik KS, Brod BB: Leukemia-cutis in congenital leukemia analysis and review of the world literature. Arch Dermatol 1993; 129:1301-1306
5. Cho-Vega JH, Medeiros LJ, Prieto VG, Vega F: Leukemia cutis. Am J Clin Pathol 2008; 129:130-142
6. Agis H, Weltermann A, Fonatsch C et al: A comparative study on demographic, hematological, and cytogenetic findings and prognosis in acute myeloid leukemia with and without leukemia cutis. Ann Hematol 2002; 81:90-95
7. Yonal I, Hindilerdenc F, Coskun R, Doganb O I, Nalcaci M: Aleukemic Leukemia Cutis manifesting with disseminated nodular eruptions and a plaque preceding acute monocytic

leukemia: A Case Report. Case Rep Oncol 2011; 4:547–554
8. Su WP: Clinical, histopathologic, and immunohistochemical correlations in leukemia cutis. Sem in Dermatol 1994; 13:223-230
9. Du X A, Hung T, Surmanowicz P, Gniadecki R: Diagnostic challenge of aleukemic leukemia cutis preceding acute myelogenous leukemia: A case report. JCMS Case Reports 2020; 8: 1–4
10. Dohner H, Estey EH, Amadori S et al: Diagnosis and management of acute myeloid leukemia in adults: recommendations from an international expert panel, on behalf of the European Leukemia Net. Blood 2010; 115:453–74
11. Burnett A, Wetzler M, Lowenberg B: Therapeutic advances in acute myeloid leukemia. J Clin Oncol 2011; 29:487-494
12. Wang CX, Pusic I, Anadkat MJ: Association of leukemia cutis with survival in acute myeloid leukemia. JAMA Dermatol 2019; 155:826-832

Presentations at the Clinical Meetings (January 2021 to June 2021)

Sr No.	Date	Speaker/Department	Title
1	13.01.2021	Jamsheer V. T. Surgical Oncology	Newer Evidence and Literature on Oropharyngeal Carcinoma for Minimally Invasive Surgery
		Shivaraj M. L. Radiodiagnosis	Role of MRI in Diagnosis Staging and Follow up of Patients with Carcinoma Cervix
2	27.01.2021	Shah Kajal Medical & Pediatric Oncology	Impact of Covid-19 Pandemic on Cancer Care
		Patel Shruti Cancer Biology	A Comprehensive DNA Panel Next Generation Sequencing Approach Supporting Diagnostic and Therapy Prediction in Neurooncology
3	10.02.2021	Dave Drishti Anaesthesiology	New Dimensions in Airway Management: Risks for Healthcare Staff
		Vemanamandhi Priyanka Gynaecological Oncology	Randomized Trial of Primary Debulking Surgery Versus Neoadjuvant Chemotherapy for Advanced Epithelial Ovarian Cancer (Scorpion-Nct01461850)
4	24.02.2021	Salunke Abhijeet Surgical Oncology	A to Z RAM (Radiological Assessment Method) for Bone Tumors: A Useful Guide for Orthopedic Sugeons and Radiologists
		Rajvik Kruti Immunohaematology	PDL-1 expression in Cancer and its Association with Clinical Outcomes
5	10.03.2021	Modi Nikhil Neuro Oncology	Endoscopic Pituitary Surgery- Learning Curve and Anatomical Variant
		Joshi Jigna Stem Cell Biology	Clinical Impacts of EGFR Mutation Status: Analysis of 5780 Surgically Resecyed Lung Cancer Cases
6	24.03.2021	Khoja Jasmin Physiotherapy	Development of an Exercise Intervention for the Prevention of Musculoskeletal Shoulder Problems after Breast Cancer Treatment: The Prevention of Shoulder Problems Trial (UKPROSPER)
		Patel Dharmesh Cytogenetics	Cytogenetic Exploration of Ewing Sarcoma with Emphasis on Variant Translocations: A 12- Year Study at a Regional Cancer Centre in South Asia
7	09.06.2021	Haresh Surgical Oncology	Primary Chest Wall Sarcoma: A Single Institution Experience of 3 Years
		Patel Foram Microbiology	A Seven Year Reterospective Study of Prevelance of Viral Infections in Diverse Cancer Patients Admitted for Treatmentina Regional Cancer Centre

About the Journal and Instructions to Authors

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Gujarat Cancer Society Research Journal is a biannually (April and October) peer-reviewed journal published by the Gujarat Cancer Society (formerly published as GCS Research Bulletin). The journal's full text is available online at http://www.cancerindia.org

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The Journal intents to cover basic, clinical, clinico-basic research and medical education carried out by the staff of the Gujarat Cancer Society and Gujarat Cancer and Research Institute related to human well being including ethical and social issues in the field of Oncology. The Journal gives preferences to original scientific papers, case reports, anecdotal reports and minireviews. It may comprise invited review articles, publish oration speeches and work presented in the clinical meetings and the journal clubs. Hence it will continue to serve as an academic-research bridge between the basic sciences and the applied sciences, viz. various disciplines of medicine within and outside GCS-GCRI.

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This section is not mandatory, but can be added to the discussion.

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Standard Journal
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Online journal article
Miyamoto O, Auer RN: Hypoxia, hyperoxia, ischemia and brain necrosis. Neurology [serial online] 2000; 54:362-71. Available at: www.neurology.org. Accessed February 23, 2000.

Chapter in a book
Weinstein L, Swartz MN: Pathogenic properties of invading microorganisms. In: Sodeman WA Jr, Sodeman WA, eds. Pathologic Physiology: Mechanisms of Disease.Philadelphia: Saunders, 1974: 457-472

Online book or website
Garrow A, Weinhouse GL: Anoxic brain injury: assessment and prognosis. In: Up To Date Cardiovascular Medicine [online] Available at: www.UpToDateInc.com/card. Accessed February 22, 2000.

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Lillywhite HB, Donald JA: Pulmonary blood flow regulation in an aquatic snake. Science. In press.

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Role of Hospital Pharmacy Service in Healthcare

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Introduction

Hospital pharmacy is the health care service, which compromises the art, practice and profession of procuring, storing, maintaining and dispensing medicines and medical devices, advising patients, doctors, nurses and other healthcare professionals on their safe, effective and efficient use. It is a specialized field of pharmacy which forms an integrated part of patient health care in a health facility. It is the profession that strives to continuously maintain and improve the medication management and pharmaceutical care of patients to the highest standards in a hospital setting. The pharmacist is the custodian of medicines and pharmaceutical products throughout the hospital. Procurement and distribution play a critical role in the provision of cost-effective quality pharmaceutical care.

The missions of the hospital pharmacists:

- To be part of the medication management in hospitals, which encompasses the entire way in which medicines are selected, procured, delivered, prescribed, administered and reviewed to optimize the contribution that medicines make for producing informed and desired outcomes;

- To enhance the safety and quality of all medicine related processes affecting patients of the hospital to ensure the **7 "rights"** are respected: right patient, right dose, right route, right time, right drug with the right information and right documentation.

Pharmacy Department of the Gujarat Cancer & Research Institute was established initially with few Anticancer Drugs and some general medicines with only three dispensing windows on ground floor. Medicines were dispensed maintaining manual registers for each drug with each and every patient register numbers. Currently, we have an enriched Drug Formulary of 534 Drugs & Medicines, Surgical Dressing Materials, Disposable items and Surgical Items. Pharmacy Department is working in 5 wings and three satellite Pharmacy at Bhavnagar, Rajkot and Siddhpur to distribute these drugs using digital platform.

Purchase of Medicines

A list of medications appropriate for the patients and as per the scope of the organization's clinical services is developed collaboratively by the multi-disciplinary committee. As per the formulary, medicines and surgical items are purchased from n'procure portal taking care of quality, assuring required documents. Even from GEM Portal/GMSCL basis/ Government or Semi Government institutional basis many items can be procured.

The Pharmacy Department of the Gujarat Cancer & Research Institute is functioning in five sections. (i) Main Pharmacy, (ii) Indoor Ward Pharmacy, (iii) Indoor Patient Pharmacy, (iv) Outdoor Patient Pharmacy and (v) Chemotherapy Pharmacy.

I. Main Pharmacy: It purchases Drugs and Medicines through e-tender for patients of Gujarat Cancer and Research Institute, using the e-tendering platform of (n) Code Solutions in accordance with Tender committee. It follows well defined procedure of acquisition of medications. The process included various issues of vendor evaluation, indenting process, generation of purchase orders and receipt of goods. Goods received are supplied to the sub pharmacies as per their requirement. About 534 medicines are dispensed from this department.

II. Indoor Ward Pharmacy: It follows the drug distribution system of Floor Stock System in which General/Common Drugs/Surgical Disposable items are supplied to wards using the digital platform of Ward Management System. Requisition in form of authorized indents from various departments and wards are supplied from Indoor Ward Pharmacy.

III. Indoor Patient Pharmacy: Patients admitted at GCRI wards are dispensed their medication requirement from this pharmacy. Average 650 patients are supplied medicines daily.

IV. Outdoor Patient Pharmacy: Pharmacy services are arranged in such a way that the outdoor patients and discharged patients can collect their medicines

(tablets/capsules) from ground floor Room No. 25, which is just near to cash collection counter. Average 650 patients are supplied medicines daily.

V. Chemotherapy Pharmacy: Patients receiving day care chemotherapy are supplied their medicines from this location.

Pharmacy Department follows the HIMS, i.e. Hospital Information Management System of organization for purchase and distribution of medications. In addition to all these, distribution of all items are also arranged to dispense at our allied Cancer Care Centers of our Siddhpur, Rajkot and Bhavnagar branches too.

Narcotics & Psychotropic Substances

A substance used for the pain management of patients with moderate to severe pain. Following Narcotic and Psychotropic Substances Act 1985 (NDPS Act), Narcotics are purchased and stored in double-locked cupboards and keys are managed by two different designated persons. Documented procedures guide the use of narcotic drugs and psychotropic substances which are in consonance with local and national regulations.

Activities

The pharmacy computers are linked with all bill collection counters for paid patients. With the provision of silent billing system, Pharmacy Department provides medicines, costly drugs, chemotherapy medicines, as well as surgical items, ortho surgical items free of cost to various categorized patients like PMJAY (including MA-YOJANA), SC, ST, etc. Patients under GCS's Poor Patient Fund are provided free pharmacy services as per rules of the institute. Number of patients under School Health Programme is markedly increased and all medicines are purchased and supplied to them also for free of cost. Distinguished services are provided to special room patients, patients taking treatment under BMT and Government employees.

Stock Maintenance

Through Pharmacy Management System, the pharmacy department supports the distribution and management of drugs, medical device inventory, and maintains stock management in precise and accurate manner.

Responsibility

Medication safety management and the evaluation of a patient's medication-related needs by determining the indication, safety and effectiveness of therapy as part of the dispensing process is a key responsibility. Furnishing of information and advice to both patients and other healthcare professionals also form part of our role.

During COVID times, all our pharmacy staff members showcased supererogation by ensuring all indoor and outdoor ward patients have access to stock of formulary and non-formulary medication and surgical disposable items and used digital technology like WhatsApp platform to handle prescription to prevent spread of Covid infection among healthcare staff.

Training Programmes

Internal training programmes are held in yearly frequency in alignment with required skills and updates of knowledge, ethical practice and improving medication management.

Rational Use of Antimicrobial

In an era of a concerning increase in antimicrobial resistance, infections are becoming harder to treat. We, Pharmacists play a critical role in ensuring that antimicrobials such as antibiotics are being used responsibly in our hospitals and only when necessary. We ensure that the combination and duration of antimicrobial therapy is appropriate for each individual patient to optimise therapy, and that it is discontinued when it is no longer needed in order to prevent a further increase in antimicrobial resistance.

Future Planning

The ultimate goal for the pharmacy department is to reduce the patient's trouble of loaded information from various sources and absence of streamlined process which makes the patient's health care management difficult. Our ultimate goal is to serve the patient with full stock of medication supplies and ensure delivery of exceptional quality of pharmaceutical care. This goal can be achieved through various initiatives starting with dispensing of medications in central distribution system from where patients and care takers have access to all the requirements without going to various windows for different needs.

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Outdoor Patient Pharmacy - A - 4, Gr. Floor, New Building GCRI

Chemotherapy Pharmacy - A-406, 4th Floor, New Building GCRI

Indoor Patient Pharmacy - R. No. 210, 2nd Floor, Nr. New BMT, Old Building

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