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Editorial

Workplace Security and Violence Against Physicians in Bangladesh

Workplace security, especially for healthcare professionals, is a critical issue worldwide. In Bangladesh, the prevalence of violence against physicians has become a significant concern, affecting the quality of healthcare and the well-being of medical professionals. This article explores the current state of workplace security for physicians in Bangladesh, the factors contributing to violence, and potential solutions to mitigate these issues.

Physicians in Bangladesh face numerous challenges, including inadequate infrastructure, heavy workloads, and insufficient remuneration. However, the threat of violence, both verbal and physical, has emerged as one of the most pressing issues. According to a study published in the Bangladesh Medical Journal, approximately 54% of physicians in Bangladesh have experienced some form of workplace violence, ranging from verbal abuse to physical assault.[1]

A 2019 survey conducted by the Bangladesh Medical Association (BMA) revealed that more than 70% of doctors in government hospitals reported experiencing violence in their workplace at least once in their careers.[2] This widespread issue has far-reaching implications for both healthcare providers and patients.

Contributing Factors

Several factors contribute to the high incidence of violence against physicians in Bangladesh:

1. **Patient Dissatisfaction:** Overcrowded hospitals and long waiting times often lead to patient frustration. When expectations for quick and effective treatment are not met, this frustration can sometimes manifest as violence against healthcare providers.[3]
2. **Poor Healthcare Infrastructure:** The healthcare system in Bangladesh struggles with inadequate facilities and resources. This inadequacy can lead to treatment delays and suboptimal patient care, further fueling discontent among patients and their families.[4]
3. **Lack of Security Measures:** Many hospitals and clinics lack adequate security personnel and systems to protect staff from violent incidents. The absence of a robust security infrastructure leaves physicians vulnerable to attacks.[5]
4. **Cultural and Social Norms:** There is a general lack of awareness and respect for the professional boundaries and rights of healthcare workers. Cultural norms that do not discourage aggressive behavior towards physicians exacerbate the problem.[6]
5. **Legal and Administrative Gaps:** The legal framework in Bangladesh often fails to provide sufficient protection or recourse for physicians who are victims of violence. Administrative inefficiencies also mean that incidents of violence are not always properly recorded or prosecuted.[7]
6. **Media Influence:** Sensationalist media reporting can sometimes contribute to negative public perceptions of healthcare professionals, leading to increased tensions and potential violence.[8]

Impact on Healthcare Delivery

The violence against physicians has profound implications for the healthcare system in Bangladesh:

- **Mental Health:** Continuous exposure to violence can lead to significant psychological stress and burnout among physicians, affecting their mental health and overall job performance. A study found that 45% of doctors who experienced violence reported symptoms of anxiety and depression.[9]
- **Professional Attrition:** Fear of violence can deter medical professionals from practicing in high-risk areas, leading to a shortage of experienced physicians in regions where they are most needed. This attrition further strains the healthcare system.[4]
- **Quality of Care:** The threat of violence can result in physicians practicing defensive medicine, where they avoid high-risk procedures or patients, potentially compromising the quality of care provided. Defensive medicine increases healthcare costs and reduces patient satisfaction.[3]

Measures to Address the Issue

Addressing workplace violence against physicians in Bangladesh requires a multifaceted approach :

1. **Enhancing Security Measures:** Hospitals and clinics should invest in better security infrastructure, including surveillance systems and trained security personnel. Establishing a visible security presence can deter potential aggressors.[5]
2. **Legal Reforms:** Strengthening laws to protect healthcare workers and ensuring strict enforcement can provide a deterrent against violence. Quick and decisive legal action against perpetrators can also reassure physicians.[6]
3. **Improving Healthcare Infrastructure:** Upgrading healthcare facilities and ensuring adequate resources can reduce patient frustration and improve the overall patient experience, indirectly reducing the risk of violence.[4]
4. **Training and Awareness Programs:** Conducting regular training sessions for healthcare workers on de-escalation techniques and for the public on the importance of respecting medical professionals can help in preventing violent incidents.[7]
5. **Support Systems for Victims:** Establishing support systems, including counseling and legal aid for physicians who experience violence, can help them cope with the aftermath and continue their practice with confidence.[9]
6. **Community Engagement:** Involving community leaders and local organizations in awareness campaigns can foster a culture of respect and cooperation between healthcare providers and the community, reducing the likelihood of violent incidents.[3]

Violence against physicians in Bangladesh is a significant barrier to providing quality healthcare. Addressing this issue requires concerted efforts from the government, healthcare institutions, and society at large. By improving security measures, enacting stronger legal protections, enhancing healthcare infrastructure, and fostering a culture of respect towards healthcare workers, Bangladesh can create a safer and more conducive environment for its physicians, ultimately benefiting the entire healthcare system.

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Original article

Clinico-aetiological Profile of Urinary Tract Infection in Pediatrics Department in a Tertiary Care Hospital

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Abstract:

Background: Urinary tract infection (UTI) is one of the important cause of infection in pediatric age group. ***Objectives:*** The study was designed to assess the clinical profile, common bacterial microorganisms causing UTI and their antimicrobial susceptibility patterns at Bashundhara Ad-din medical college and hospital. ***Materials and Methods:*** A cross-sectional study involving children aged 1 month to 15 years who had UTI symptoms was carried out in the pediatric department from January to June 2023. Among suspected patients, standard urine testing and microscopic examination were carried out. Urine cultures and sensitivity tests were subsequently done for the pyuria patients (n = 110). This study included 44 patients with UTIs who tested positive for culture. Patients that were both indoors and outpatients provided clinical data. ***Result:*** Among 317 suspected cases only 110 had pyuria and among them only 44 (13.8%) having significant culture positive result. Fever, poor feeding, vomiting, irritability were most common symptoms below One year while children presented with fever, abdominal pain and increased frequency of micturition mostly. E.coli was reported as the most common (59%)etiological agent followed by Proteus(12%),Staphylococcus aureus (9%), Pseudomonas(7%), klebsiella (5%). E.coli was sensitive mostly to Ampicillin (70%),Nitrofurantoin (68.5%),Meropenem and Amikacin (61.5%).Klebsiella was 100% sensitivetoAmikacin and Nitrofurantione. Proteus was mostly sensitive to Meropenem (84%),Imipenem (73%). Pseudomonas was 100% sensitive to Piperacillin-Tazobactum,Meropenem (66.7%). Enterococcus was 100% sensitive to Linezolid and vancomycin (90%). Staphylococcusaureus wassensitive to Nitrofurantoin (98%) and Gentamycin (75%),Vancomycin (50%). Coagulase negative staphylococcus was 100% sensitive to Vancomycin, 91% sensitive to Gentamycin. Acinetobacter was 90% sensitive to Piperacilli-Tazobactum combination and 85% to Nitrofurantoin. Staphylococcus saprophyticus was 90% sensitive to Linezolid,Vancomycin. ***Conclusion:*** Though various microorganisms are responsible for UTI in children, E. coli is the most common causative agent. Rational use of antibiotics must be encouraged and restriction of antibiotic abuse should be done to retard development of further drug resistance.

Keywords: UTI, bacterial Isolates, antibiotic susceptibility.

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Introduction:

Urinary tract infection (UTI) is one of the most common bacterial infection seen in pediatric population, which is a significant cause of morbidity in children.[1,2] The overall prevalence of UTIs among infants and young children is estimated to be approximately 2%–20%.[3,4] Since UTI can present with non-specific clinical features, UTI is underdiagnosed. [5,6] UTI is associated with renal parenchymal scarring in approximately 10-30% of pediatric patients presenting with febrile UTI.[7,8] Hence it is necessary to clinically suspect UTI and start the children on appropriate empirical antibiotics at an early stage.

Children's UTIs are frequently caused by bacteria, with E. coli being the most common isolate pathogen in the pediatric age range.[9] Proteus mirabilis, Staphylococcus aureus, Enterococci, and Klebsiella spp. are additional bacteria that can cause UTIs. [10,11] Many antibiotics are not effective against the majority of these infections. [11-13].Treatment requires an understanding of the microorganisms involved and the antibiotic susceptibility of uropathogens in each geographic environment.[14]

The gold standard for diagnosing UTIs is urine culture and sensitivity pattern.[16] In girls' midstream urine, a colony count of more than 105CFU/ml of a single species and more than104 CFU/ml in boys' urine is regarded as confirmatory for UTI.17Significant bacteriuria is defined as any growth from suprapubic aspiration or a pure growth of 102/ml from a catheterized urine sample.[15]

Although E. coli has been reported to account for most of the cases of symptomatic UTI in children.[16,17] Studies from some other parts of the world however, have shown a changing trend in the bacteriology of UTI.[18-20] The spectrum of etiologic agents causing UTI and their antimicrobial resistance pattern have been continuously changing over the years, both in community and in hospitals.[20] It is especially true for developing countries where antibiotics are prescribed often irrationally. [21] In Bangladesh, most of the centers use antibiotics empirically due to unavailibity of standard therapeutic guidelines and local susceptibility data. In this perspective this study was designed to investigated the incidence, the various clinical presentation, the microbiological profile and antibiotic sensitivity pattern of UTI in children at a tertiary teaching institute.

Materials and Methods:

A cross sectional study was conducted in the department of Pediatrics,Bashundhara Ad din Medical College Hospital. The study was done for a period of 6 months(January 2023 to June 2023). The study protocol was approved by the ethical committee of the institution. The objective of the study was to analyze the clinical presentation of UTI in children between 1month to 15 years of age, to analyze the causative microorganism and their drug susceptibility in urinary tract infection in children.

Inclusion criteria: Children between the age group of 1month to 15 years of age who presented with symptoms like fever, abdominal pain, dysuria, urgency, frequency, poor feeding, vomiting, irritability during the study period, who visited both the outpatient and inpatient department was included in the study. Exclusion criteria: Recurrent UTI, other causes of pyuria like glomerulo-nephritis, vasculitis (SLE and others), known urinary malformations,children on antibiotics within last seven days of sample collection and samples having mixed collections were excluded. Alsorepeated samples from same patient who has already been included and those samples with evidence of perineal contamination was excluded from the study. Children were categorized according to age group like neonate (0 to 28 days), infant(birth to 1 year), toddler(1 to 3 years), preschool age (3 to 5 years), children (5 to 15 years).

Patient details including age, sex, clinical presentation, previous history of UTI and any congenital anomaly was collected from suspected indoor and outdoor patients after taking consent and entered in the predesigned proforma. Urine culture and sensitivity report was collected from the patients and analyzed. Urine sample collected by clean catch midstream technique or catheter sample was included. Urine sample showing significant growth that is more than or equal 105 CFU/ml of single micro-organism in presence of symptoms was considered significant and processed for further identification and susceptibility testing.

Antibiotic susceptibility test was done by conventional method and interpreted according to Clinical and Laboratory Standards Institute Guidelines (CLSI) 2019 and 2020. Antibiotics tested were Ampicillin, Cephalosporins, Amikacin, Gentamicin, Co-trimoxazole, Nitrofurantoin, Piperacillin- tazobactam, Fluoroquinolones and Carbapenems for gram-negative organisms and Ampicillin, Gentamicin, Nitrofurantoin, Norfloxacin,Llinezolid and Vancomycin for gram-positive organisms.

Statistical analysis was done using Statistical package for social sciences (SPSS) Software version [16].

Result:

Among 317 suspected cases only 110 had pyuria and among them only 44 having significant growth in culture media result accounting 13.8% of total sample studied. Occurance of UTI was highest (n=22,50%) in children (5-15 yrs) and lowest in below 1 yr age group (n=3,6.81%).

Fever (66.6%), poor feeding (66%), vomiting (33%), irritability(33%) were most common symptoms in neonate and infants while children presented with fever(86.3%), abdominal pain (65.9%) and increased frequency of micturition (72.7%) mostly (table l).

E.coli was reported as the most common (59%) etiological agent followed by Proteus (12%), Staphylococcus aureus (9%), Pseudomonas (7%), Klebsiella (5%), Coagulase

negative staphylococcus (2%), Staphylococcus saprophyticus(2%), Enterococcus (2%) (tablelV). According to age group E.coli, klebsiella and Proteus was found mostly in neonates and infant and E.coli, Proteus, Pseudomonas ,Enterococcus were found mostly in children (table ll). E.coli was sensitive mostly to Ampicillin (70%),Nitrofurantoin (68.5%),Meropenem (61.5%) and Amikacin (61.5%). Klebsiella was 100% sensitivetoAmikacin and Nitrofurantione and 90% sensitive to Emipenem, Meropenem. Proteus was mostly sensitive to Meropenem (84%),Imipenem (73%), Piperacilin-Tazobactum (81%),ceftriaxone (75.5%). Pseudomonas was 100% sensitive to Piperacillin-Tazobactum and Meropenem (66.7%), Amikacin (84%) Ceftriaxone, Ciprofloxacin. Enterococcus was 100% sensitive to Linezolid and vancomycin (90%). Staphylococcus aureus sensitive to Nitrofurantoin (98%) and Gentamycin (75%),Vancomycin (50%). Coagulase negative staphylococcus was 100% sensitive to Vancomycin, 91% sensitive to Gentamycin. Acinetobacter was 90% sensitive to Piperacilli-Tazobactum combination and 85% toNitrofurantoin. Staphylococcus saprophyticus was 90% sensitive to Linezolid, Vancomycin.(table lll).

Table 1: Clinical presentation according to age category (n=44)

Symptoms	Neonate & infant		Toddler		Preschool		Children		Total	
	N=3	%	N=7	%	N=12	%	N=22	%	N=44	%
Fever	2	66.66%	5	71.29%	10	83.33%	21	95.45%	38	86.36%
Dysuria	-	-	-	-	2	16.66%	13	59%	15	34%
Frequency	-	-	4	57.14%	8	66.66%	20	90.9%	32	72.72%
Urgency	-	-	-	-	3	25%	13	59%	16	36.36%
Abdominal Pain	-	-	2	28.57%	10	83.33%	17	77.27%	29	65.9%
Vomiting	1	33.33%	2	28.57%	7	58.33%	4	18.18%	14	31.82%
Poor feeding	2	66.66%	3	42.86%	3	25%	8	36.36%	16	36.36%
Irritability	1	33.33%	3	42.86%	2	16.66%	5	22.72%	11	25%

Table 2: Distribution and frequency of uropathogens according to age category

Neonatesandinfants			Toddler		Pre-school		Children	
	Count	%	Count	%	Count	%	Count	%
Enterococci	0	0.0%	1	2.3%	0	0.0%	1	2.3%
Staphylococcusaureus	0	0.0%	0	0.0%	0	0.0%	4	9.1%
Coagulase–ve staph	0	0.0%	0	0.0%	0	0.0%	1	2.3%
Escherichiacoli	1	2.3%	6	13.6%	9	20.5%	10	22.7%
Klebsiella	1	2.3%	0	0.0%	0	0.0%	0	0.0%
Proteus	1	2.3%	0	0.0%	2	4.5%	2	4.5%
Pseudomonas	0	0.0%	0	0.0%	0	0.0%	3	6.8%
Acinetobacter	0	0.0%	0	0.0%	0	0.0%	1	2.3%
S.saprophyticus	0	0.0%	0	0.0%	1	2.3%	0	0.0%

Table 3: Antimicrobial sensitivity pattern for gram positive organism%

	Enterococci	Staphylococcus aureus	Coagulase –ve staphylococcus	Staphylococcussaprophyticus
Ampicillin	50%	35%	25%	25%
Gentamycin	71%	75%	91%	91%
Nitrofurantoin	76%	98%	10%	0%
Linezolid	100%	10%	20%	99%
Vancomycin	90%	50%	98%	90%

Table 4: Antimicrobial sensitivity pattern for gram negative organism%

	E.coli	Proteus	Pseudomonas	Klebsiella	Acinetobacter
Ampicillin	70%	76%	0%	0%	0%
Ceftazidime	35%	32%	33.3%	0%	7.7%
Cefuroxime	11%	50%	33.3%	30%	0%
Cefotaxime	19%	20.8%	19%	36%	0%
Ceftriaxone	30.8%	75.5%	33.3%	80%	0%
Cefixime	26.9%	0%	33.3%	0%	0%
Amikacin	61.5%	72%	84%	100%	0%
Gentamycin	50%	60%	21%	0%	10%
Cotrimoxazole	60%	0%	33.3%	0%	15%
Nitrofurantoin	68.5%	60%	0%	100%	85%
Piperacillin+ Tazobactum	46.2%	81%	100%	0%	90%
Ciprofloxacin	42.3%	20%	33.3%	20%	410%
Imipenem	36%	73%	33.3%	90%	72%
Meropenem	61.5%	84%	66.7%	90%	72%

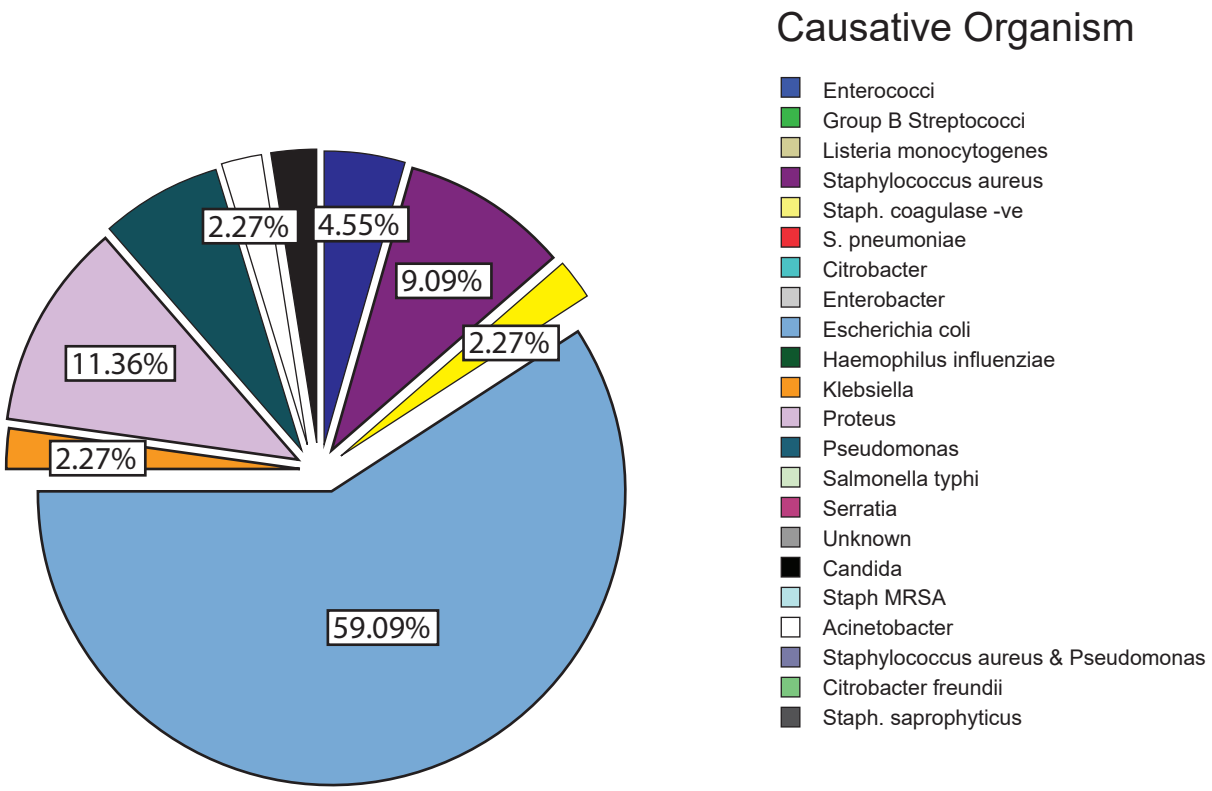

Figure: Causative organism (%)

Discussion

UTI is a common health problem in children and most common cause of morbidity and mortality especially in 2 years of life.[22] Urinary culture positive rate was 13.8% in this study which was similar to rates of 19.3% and 22.2% in previous studies. [23,24] Fever and abdominal pain was the most common symptoms in children which was similar to other studies.[25-27] Children with UTI usually present with non-classical clinical features and these are difficult to diagnose.[28] In our study, fever, poor feeding and irritability were the common clinical features in neonates while the older children presented with fever and urinary symptoms which agree with other reports where fever, abdominal pain, vomiting, dysuria, poor feeding, and irritability are reported as frequent signs and symptoms of UTI. [29,30] Diagnosis of UTI is really challenging due to its vague presenting symptoms, especially in young children. Thus, a high index of suspicion is appropriate when a young child presents with fever.[28]

E. coli was the most common organism isolated (59.1%) in our study. This was in accordance with other studies in which E. coli was isolated from 61.0% to 72.8%.[26,31-34] However, Yüksel et al and Chakupurakal et al reported a very high percentage (87.0%) and (92.0%) of E. coli in their study.[35,36] Proteus was second isolate of our study which was occupying 11.4% of the total isolates. Different studies have shown the growth of Proteus in urine from 5.8% to 12.4%.[37,38] In this study Pseudomonas was isolated in only 6.8% cases , Enterococci in 4.5% and Klebsiella 2.3%. E. coli was followed by Enterobacter spp. (16.7%), and Pseudomonas (11.1%) in a study of Philippine39 and followed by Proteus (20%), Klebsiella (5.4%) and Pseudomonas (1.8%) in a study of Nepal.[23] E.coli was sensitive mostly to Ampicillin (70%),Nitrofurantoin (68.5%), Meropenem and Amikacin (61.5%) in our study while Shrestha et al. reported E. coli as most sensitive to Nitrofurantoin (84.6%), Amikacin (80.7%), Gentamicin (73%) and Ofloxacin(53.8%).[23] Klebsiella was 100% sensitive to Amikacin, Nitrofurantione and 90% sensitive toEmipenem, Meropenem in this study while In a study done in S.S.G hospital India, Klebsiella was the second most common organism and was found to be most sensitive to Ofloxacin, Amikacin and Piperacillin+Tazobactum.[21] Proteus was mostly sensitive to Meropenem (84%),Imipenem (73%), Piperacilin-Tazobactum (81%),ceftriaxone (75.5%), Nitrofurantoin (60%) where Proteus was sensitive to nitrofurantoin and norfloxacin in 33.1% and 25.0% respectively in another study.[40] Pseudomonas was sensitive to Piperacillin-Tazobactum (100%) and Meropenem (66.7%), Amikacin (84%),Ceftriaxone, Ciprofloxacin and it is similar to a study done in Gujarat, India.[41] As like our studyGram-positive organisms like Coagulase negative Staphylococcus, Staphylococcus aureus, Streptococci have also been reported by other authors. Lok et al and Muoneke et al reported Staphylococcus aureus as the second most common uropathogen isolated in their study.[14]

Limitation

Study group was very small.

Conclusion

As the UTI in children usually presents with non-specific features, it demands the urine test for the diagnosis. Though, our data is small, it suggests providing treatment only after the proper microbiological investigations. E. coli being the commonest bacteria and exhibiting the changing drug resistance pattern, it is advisable to perform the antibiotic susceptibility testing as well. Finally, this type of study should be repeated periodically to assess the pattern of microorganisms causing UTI and then antimicrobial susceptibility which will guide in choosing antibiotics for the empiric treatment.

Conflict of interest

The authors thereby declare no conflict of interest.

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Original article

Interplay of Viral RNAemia, Interleukin-6 and Interleukin-6 Gene Polymorphism with COVID-19 Patients

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Abstract

Background: Coronavirus Disease 2019 (COVID-19) represents a formidable global pandemic attributed to the severe acute respiratory syndrome coronavirus type-2 (SARS-CoV-2). Elevated IL-6 levels, detectable serum SARS-CoV-2 viral load (RNAaemia) in critically ill patients, IL-6 gene polymorphismcontribute to excessive yet ineffective host immune responses, culminating in impaired lung function and expedited mortality among COVID-19 patients. ***Objectives:*** The primary objective of this study is to scrutinize the associations between IL-6, SARS-CoV-2 viral load (RNAaemia), IL-6 gene polymorphismconcerning disease severity in COVID-19 patients. ***Methodology:*** Conducted within the Department of Microbiology and Immunology at Bangabandhu Sheikh Mujib Medical University (BSMMU), this cross-sectional study spanned from March 2021 to January 2022. A cohort comprising 84 confirmed COVID-19 patients via positive RT-PCR and 28 healthy subjects was enrolled. Peripheral venous blood samples were procured for the detection of SARS-CoV-2 viral RNA (RNAemia) through Real-time reverse transcription polymerase chain reaction assay (RT-PCR), serum IL-6 levels via the chemiluminescence method, single nucleotide polymorphisms (SNPs) of IL-6 through sequence-specific primer polymerase chain reaction (SSP-PCR). ***Result:*** Serum IL-6 levels (pg/ml) were markedly elevated in critical patients (102.02±149.7) compared to severe (67.20±129.5) and moderate (47.04±106.5) patients. The prevalence of serum SARS-CoV-2 nucleic acid-positive cases was predominantly observed in critical patients (39.28%), and a strong correlation between extremely high IL-6 levels, RNAaemia, and high mortality was established (R=.912, P<0.001). Genotype distribution for IL-6 174G/C (rs 1800795) gene indicated.CC and GC genotypes exhibited a robust association with the severity of COVID-19 when compared to the GG genotype. A significant statistical difference in genotypes was discerned between critical and moderate groups (p<0.001, OR-10.316, CI-3.22-23.86), with the CC genotype being linked to COVID-19 severity and mortality. ***Conclusion:*** Serum IL-6, IL-6 174 G/C gene, and SARS-CoV-2 RNAaemia may prove valuable in clinical practice for risk assessment, disease progression and mitigate morbidity and mortality.

Keywords: RNAemia, Interleukin-6, gene polymorphism, COVID-19 patients

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Introduction

The COVID-19 pandemic, initially detected in December 2019 in Wuhan, Hubei Province, China, has become a worldwide public health issue.On December 31, 2019, Chinese authorities notified the World Health Organization (WHO) about multiple instances of pneumonia of unknown origin.[1] By January 7, 2020, it was established that the cause of these reported cases was the novel coronavirus known as 2019-nCoV, later renamed severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), leading to the designation of the outbreak as COVID-19.[1-2] Cytokine release syndrome (CRS) emerged as the primary cause of morbidity in patients with SARS-CoV and it is also responsible for inducing acute respiratory distress syndrome (ARDS) in those infected with SARS-CoV-2.[3] The presence of ARDS is associated with elevated serum IL-6 levels and unfavorable clinical outcomes. IL-6, a versatile cytokine, plays a crucial role in regulating a wide array of cellular activities and serves as a major immune-modulatory agent. It actively participates in the control of acute phase reactions, the activation of T helper cells, the inhibition of T regulatory (Tregs) cells, and the differentiation of B cells, thereby coordinating both innate and adaptive immune responses. [3-4]During the early stages of infectious inflammation, monocytes and macrophages, stimulated by TLRs, produce IL-6. In the acute stage of SARS-CoV-1 infection, high levels of both interleukin 6 (IL-6) and interleukin 8 (IL-8) were detected, particularly in association with lung lesions.4Notably, IL-6 can induce a hyper-innate inflammatory response as a result of SARS-CoV-1 invading the respiratory tract.[4-5] Immune dysregulation, particularly, is induced by Interleukin-6 (IL-6) rather than Interleukin-1β (IL-1β). Elevated IL-6 levels have been observed in hospitalized patients, particularly those in critical condition, and are linked to ICU admission, respiratory failure, and a bleak prognosis. The real-time reverse-transcription polymerase chain reaction (RT-PCR) assay for detecting SARS-CoV-2 in throat swab samples has been widely employed for diagnosing COVID-19 patients. Nucleic acids have been identified in serum or plasma samples for all novel coronaviruses, although the duration of viremia remains unclear. SARS-CoV-2 RNA exhibits relative stability in plasma, even though its presence may not necessarily indicate an active. Viral RNA in blood has been detected in COVID-19 patients within the first 2 to 3 days after the onset of symptoms; however, there is no evidence regarding the viral load in plasma and serum during the incubation period. [5-6] The presence of a substantial IL-6 level, coupled with a meaningful Ct (cycle threshold) value of viral RNA in serum samples, can be regarded as a reliable and precise biomarker revealing adverse outcomes. Detectable serum SARS-CoV-2 viral load (RNAemia) was identified exclusively in the critically ill group, and IL-6 levels in critically ill patients showed a significant increase, nearly tenfold compared to other patients. The notably elevated IL-6 level was closely associated with the detection of RNAemia.[7-8] The combination of IL-6 levels and serum viral RNA Ct-value may be considered an effective indicator for standard clinical assessments, offering a high level of accuracy in predicting impending adverse outcomes.[9] Several studies have reported the association of IL-6 with SARS-COV-2 RNAemia in critically ill COVID-19 patients, linked IL6 gene polymorphism to susceptibility and the severity of pneumonia, and established a connection between lymphocytes, inflammatory monocytes, and elevated IL-6 expression, leading to an excessively ineffective host immune response, resulting in lung functional impairment and swift mortality.[9-10] The objective of this study is to investigate the relationships between Interleukin-6, SARS-COV-2 viral RNA (RNAemia), Interleukin-6 gene polymorphism with the severity of the disease in COVID-19 patients.[11]SARS CoV-2 exist in blood or rectal swabs, may not to be detected in the throat swab and is important to examine samples from different sources to validate the infection.[12]

Materials and methods:

This was a cross-sectional study which was conducted in the Dept. of Microbiology & Immunology, Dept. of Anaesthesia, Analgesia and Intensive Care Medicine and COVID Unit, Bangabandhu Sheikh Mujib Medical University (BSMMU), Shahbag, Dhaka andsubjects were selected from the persons who attended Fever clinic, BSMMU. This study was conducted from March 2021 to January 2022. Informed written consent was obtained from patients and control and ethical clearance was sanctioned by the Institutional Review Board (IRB) at BSMMU.Purposive sampling procedure was followed.According to MORGAN'S table for sample size was calculated and sample size was = 28 x 4 = 112 where 28 was severe COVID-19 category admitted into BSMMU ICU. Data were collected by the pre-designed data collection sheet. All statistical analysis was performed using the statistical package for social science (SPSS) program, 26 versions.Theconfidentiality of data and the privacy of the respondentswere maintained strictly.A total of 84 COVID-19 patients confirmed by positive RT-PCR for SARS-CoV2 from nasopharyngeal or oropharyngeal swab were enrolled in this study. The moderate, severe and critically ill COVID-19 patients were recruited from Intensive Care Unit (ICU) and COVID unit of BSMMU, Shahbag, Dhaka and categorized according to National Guideline on Clinical Management of COVID-19, version 9.0, published on 6th May, 2021, Bangladesh. SARS-COV-2 viral RNA (RNAemia) detection by Real-time reverse transcription polymerase chain reaction assay (RT-PCR) and detection of IL-6 gene polymorphism by single plex SSP- PCR, Detection of serum level of IL-6 by chemilumi-

nescence Assay. Inclusion criteria were following accordingto National Guideline on Clinical Management of COVID 19, version 10.0 published on 9th February, 2022, Bangladesh.[13]

Result:

The overall median age was 55 years (interquartile range 40–69) and Individuals with detectable RNAemia were significantly older than those without RNAemia (63 vs 50 years; P = .04).

Table 1: Demographic, Laboratory Characteristics of 84 Patients with SARS-CoV-2 RNAemia

Variables		Total patient n=84	Detectable SARS-CoV-2 RNAemia (n = 11)	Nondetectable SARS-CoV-2 RNAemia (n = 73)	P value
Age, median (IQR)		55 (40–69)	63 (47.5–71)	50 (37–67)	**.04**
Sexno. (%)	Male	28 (33.33)	3 (10.71)	25 (89.28)	.703
	Female	56 (66.67)	8 (14.28)	48 (85.71)	
Lymphopenia at presentation no. (%)	No	35 (41.67)	3 (27.27)	32 (43.83)	.721
	Yes	49 (58.33)	8 (72.71)	41 (56.16)	.961
Median Ct (IQR)		36.4 (34.0-38.5)			

Table 1 shows, Fisher's exact test, p value<.05 is significant.

Table 2: Association of serum level of IL-6 with severity of COVID-19 infection and healthy controls

Category of patient	IL-6(pg/ml) Mean ± SD	Mean rank	P value
Control groupn=28	3.5±1.8	17.9	<0.001
Moderate groupn=28	47.04±106.5	32.5	
Severe groupn=28	67.2±129.5	63.7	
Critical groupn=28	102.02±149.7	76.4	

Table 2 shows, Kruskal Wallis test, P<.05 indicated statistical significance.

In control, the mean IL-6 was 3.5 ±1.81, in moderate group of patients it was 47.04±106.5, in severe group of patients, it was 67.2±129.5and in critical group of patients it was 102.02±149.7. Kruskal Wallis test showed that there was significant statistical difference among the groups regarding IL 6 as p<0.001.

Table 3: The correlation analysis of RNAaemia and vital signs withserum IL-6 level

Variables		All patients n=84	IL-6 (＜100 pg/ml)	IL-6 (100＞ pg/ml)	R	P-value
RN Aaemia	**Negative**	73	68	5	0.912	＜0.001
	Positive	11	1	10		
Vital Signs	**Death**	5	0	5	0.983	0.102
	Alive	79	69	10		

Pearson correlation, P<.05 indicated statistical significance.

Table 3 shown that, the incidence of RNAemia was closely correlated with IL-6 high in critically ill patients (R=0.912).

Table 4: Allele and genotypic frequencies of IL-6 174G/C genes in COVID-19 patients and healthy controls

Gene and SNP	Genotype /Allele	Patient (n=84) (%)	Healthy controls (n=28) (%)	P value	OR (95% CI)
IL-6 174G/C (rs 1800795)	**GG**	59 (70.24)	23 (82.14)	.927	1.027 (0.45-4.65)
	GC	23 (27.38)	5(17. 85)	.896	2.043 (0.3-3.45)
	CC	2 (2.38)	0	-	-
	G allele	141 (83.92)	52(91.07)	.706	0.847 (0.32-3.24)
	C allele	27 (16.07)	5(8.92)		

Note: OR = Odds ratio, 95 % CI= 95% Confidence interval. P value and Odds ratio were calculated using Chi-square test for (3 x 2 and 2 * 2) contingency table. P value< 0.05 was considered as statistically significant. IL-6 174 G/C polymorphism in different categories of patients and controls. Table 4.5 shows frequency of GG, GC and CC genotype in patients were 70.24%, 27.38% and 2.38% respectively which were not significantly different from healthy control .Frequency of G and C allele was 83.92% and 16.07% respectively in patients; 91.07% and 8.92% in healthy control Any of these associations were not statistically significant.

Table 5: Allele and genotypic frequencies of IL-6 174G/C gene with different category of study population

Study population	IL-6 174G/C gene				
	GGn (%)	GC&CCn (%)	OR	CI	p value
Moderate Control	25 (89.20) 23 (82.14)	3 (10.71) 5 (17.85)	3.47	.851-5.290	.821
Severe Control	19 (67.85) 23 (82.14)	9 (32.14) 5 (17.85)	3.106	0.89-8.24	.092
Critical Control	15 (53.57) 23 (82.14)	13 (46.42) 5 (17.85)	.015	0.09-3.23	.023
Moderate Severe	25 (89.20) 19 (67.85)	3 (10.71) 9 (32.14)	4.983	1.25-4.36	.018
Moderate Critical	25 (89.20) 15 (53.57)	3 (10.71) 13 (46.42)	8.316	3.22-9.86	.019
Severe Critical	19 (67.85) 15 (53.57)	9 (32.14) 13 (46.42)	3.327	0.823-12.01	.854

Note: OR = Odds ratio, 95 % CI= 95% Confidence interval. P value and Odds ratio were calculated using Chi-square test for (3 x 2 and 2 * 2) contingency table. P value< 0.05 was considered as statistically significant.In the control, 23 (82.14%) had GG genotype while in moderate group, 25 (89.20%)patients had GG genotype (p=0.821). severe group, 19 (67.85%)patients had GG genotype. critical group, 15 (53.57%)had GG genotype. In the moderate group, 25 (89.20%) patients had GG genotype while in severe group, 19 (67.85%) patients had GG genotype (p=.018). Odds ratio expressed that in the severe group, the odds of having GC and CC was 4.983times compared to moderate group. In the moderate group, 25 (89.20%) patients had GG genotype while in critical group, 15 (53.57%) GGgenotype (p=.019). Odds ratio expressed that in the critical group, the odds of having GC and CC was 8.316times compared to moderate group. In the severe group, 19 (67.85%) patients had GG genotype while in critical group, 15 (53.57%)GGgenotype. There was no significant statistical difference between the groups regarding IL-6 174G/C SNP gene as p=0.854.

Table 6: Correlation between IL-6 and IL-6 174G/C genotype according to disease severity

Category of patients		**IL-6 174G/C genotype**	
IL-6	**Moderate group**	Pearson Correlation	.067
		p-value	1.01
	Severe group	Pearson Correlation	**.481***
		p-value	**.05**
	Critical group	Pearson Correlation	**.651****
		p-value	**<.001**

Here, Correlation is significant at the 0.05 level (2-tailed).Correlation is significant at the 0.01 level (2-tailed). Correlation is significant at the 0.001 level (2-tailed).Serum IL-6 level significantly correlated with IL-6 174G/Cgenotype in critical group (p<.001).

Figure 1: Mean level of IL-6 according to genotype distributions

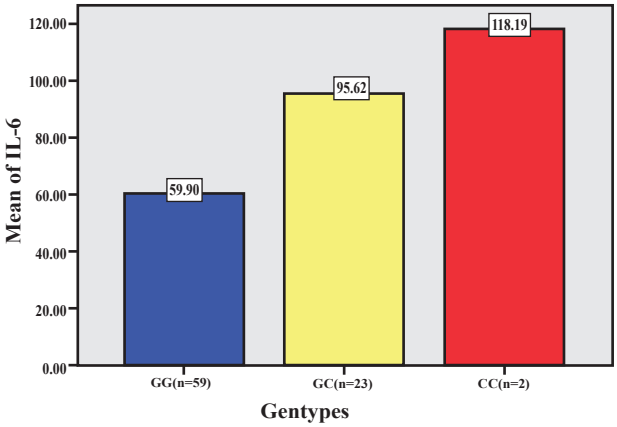

Figure 1 shows bar diagram showing the mean level of IL-6 according to genotype distributions of IL-6 174G/C. Statastical association found between the mean level of IL-6 and GC and CC genotypes (P-0.03).

Discussion:

In this study, the distribution of serum SARS-CoV-2 nucleic acid positive cases in various patients groups, and the result showed that those cases were confirmed mostly in critically ill patients group (39.28%), the extremely high IL-6 level was closely correlated with the incidence of RNAaemia and the mortality. (R=.912, P<0.001) Chen et al., (2020) showed that serum SARS-CoV-2 viral load and IL-6 level could serve as an indicator of poor prognosis.[14] The association between RNAemia, ICU admission (p=.04) and invasive mechanical ventilation (p=.02) suggests the potential utility of plasma SARS-CoV-2 RNA testing as a prognostic indicator .[15] Preliminary data from a limited number of patients have shown an association between RNAemia and severe COVID-19 and support the presence of RNA in extrapulmonary sites .[14] A similar association between detection of SARS-CoV-1 RNA in serum and clinical

complications including oxygen desaturation, mechanical ventilation, and mortality was noted in various types of studies.[16] Thus, in the absence of a robust COVID-19 clinical scoring system, plasma may be considered as a complementary modality for the early identification of individuals likely to develop severe COVID-19.[17] In this study serum level of IL-6 was found statistically significant ($p<0.001$) among different COVID-19 patient groups. But another studies show found that IL-6 level increased in critically ill group than moderate and has statistically significant association ($p<0.001$) with disease severity.14 Different types of studies showed that serum level of IL-6 in severe and critical patients compared with moderate and mild patients has significant statistical difference ($p=0.004$).[18] In this study, patients with critical Covid-19 had mean IL-6 level76.4 pg/mL compared to 32.5 pg/mL in moderate and 63.7 pg/mL in severe Covid-19 group. A metanalysis of 9 studies concluded that increased IL-6 is highly associated with severe disease and patients with severe Covid-19 had mean IL-6 higher (58pg/mL) compared to mild disease (17 pg/mL).[19] A study by Herold et al. found that IL-6 > 80 pg/mL predicts respiratory failure and need for mechanical ventilation in Covid-19. Among hospitalized patients with COVID-19, patients with high IL-6 ($R = -0.535$, $P < 0.001$)levels at admission are at increased risk of developing a severe form of the disease, requiring mechanical ventilation and ICU, and progressing to respiratory distress syndrome and multiorgan failure.[20] In this study, IL-6 174 genotype and allele frequency in total patients were not significantly different from healthy control.Comparison of genotype and allele frequency among moderate and severe and critical groups showed similar pattern: significantly reduced GG genotype and G allele, along with increased GC genotype and C allele in severe and critical group. This denotes that genotype GG and G allele were protective while genotype GC and C allele increased the odds of severe disease and mortality. It is likely that IL-6 174 C allele is a risk factor for critical illness and mortality and IL-6 GG genotype and G allele seems to be protective. [21] Findings of this study support the pilot analysis by Rajkumar (2020) and Ulhaq and Soraya (2020) across several nations where IL-6 174 G allele was negatively correlated with COVID-19 prevalence and mortality and IL-6 174 C allele was significantly associated with the severity of COVID-19 associated pneumonia.[21-22] Similar results were found in several studies on community acquired pneumonia (CAP) and ARDS. [23]

Conclusion:

IL-6 increased in critically ill COVID-19 patients compared to moderate group and healthy groups and statistically significant among different category of COVID-19 patients.Extremely high IL-6 level correlated with the incidence of RNAemia and the mortality.IL-6 174 GC genotype may be one of the most important risk factors for severe COVID-19 disease and mortality.

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Conflict of interest:

The authors declare that no conflict of interest exists.

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Original article

Isolation ESBL-Producing Enterobacteriaceae Species in Dhaka City's Red Meat: Phenotypic Analysis and Antibiotic Resistance Profiles

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Abstract

Background: It is generally acknowledged that one of the most significant issues facing modern human medicine is the fast and irreversible rise of antibiotic-resistant pathogenic bacteria, which has been documented over the past two decades. This circumstance raises concerns in many areas. When treating different types of infections, there are resistance mechanisms that prevent the use of last-resort medications. Many resistance mechanisms that develop in bacterial populations and propagate are those with broad activity spectra that render all or most medicines in a particular treatment category ineffective. ***Objective:*** The goal of this study is to evaluate the microbiological quality of raw meat,the frequency of production of Extended- spectrum-beta-lactamases (ESBL) in Enterobacteriaceae and the antibiotic susceptibility of these isolates in vitro. ***Methodology:*** A comparative cross-sectional approach was employed in this study to examine 40 food samples of beef and mutton from different areas of Dhaka. For pure culture, Mac Conkey agar plates were utilized after sample collection. Muller-Hinton agar media (MHA) was used for antibiotic susceptibility testing after completion of organism detection test on Eosin-methylene blue (EMB) agar media. ***Results:*** Among 40 samples, 95% of the bacteria discovered belonged to the Enterobacteriaceae family. Five (or 25%) of the beef samples showed resistance against the combination of Ceftazidime (CAZ) and Amoxicillin-Clavulanic acid (AMC). About the mutton, 20 (100%) cases were found resistant to CAZ+AMC (100%), and 12 instances were resistant to CTX (Cefotaxime) + AMC (60%). Several strains (17.5%) in our investigations demonstrated phenotypic positive for producing the ESBL enzyme. Of 40 dietary samples, 35% of the strains that create ESBL are identified in beef and mutton, respectively, with 30% and 5% 0f theses strains producing ESBL. ***Conclusion:*** Antibiotic resistance is an important health issue due to the potential pathogenicity of antibiotic-resistant bacteria associated with animals, their case of transmission to mankind through food chains, and their extensive environmental dispersion through animal wastes. These might cause patient to develop complicated, long-lasting, incurable illnesses as well as animal drug resistance gene might be transferred from livestock to human being, which could decrease the life expectancy and increase the cost of treatment even death.

Keywords: ESBL, phenotypic characteristics, AMC

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Introduction:

Antimicrobial resistance is a burgeoning global concern. Annually, 33,000 individuals succumb fatal infections caused by bacterial antibiotic resistance within the European region. According to published article, it has been projected that the global population is going to face a significant threat to public health and economic stability in the coming years due to anti-microbial resistance.[1] By the year 2050, it is estimated that approximately 10 million lives per annum and a cumulative financial output of 100 trillion USD could be jeopardized due to the emergence of drug-resistant infections. We must urgently seek proactive measures to mitigate the progression of drug resistance to safeguard public health and economic prosperity.[2] It is worth noting that the production of ESBLs is often associated with the emergence of multi-drug resistance.[3] Meat is a good substrate for many organisms to thrive because it is high in moisture, rich in nitrogenous compounds (such as amino acids, peptides, and proteins),and abundantly provided with mineral & auxiliary growth agents.[4] Food continue to endanger both human and animal health, despite the fact that industrialized nations have improved the hygienic of all meat production procedures. Food borne illnesses spread to humans when tainted food is consumed and toxins enter the body. This becomes one of the major global public health issues & both social and economic stability will be endangered.[5]

Generally speaking, unclean slaughter and sale facilities can harbor microorganisms resistant to antibiotics that infect raw meat. Eating meat tainted with microorganisms that are resistant to antibiotics can have serious health effect[6] and because of cultural and religious concerns, many here consume red meat.

This study proposes to assess the microbiological quality of raw meat, the incidence of Extended spectrum-beta-lactamases (ESBL) Production in *Enterobacteriaceae* isolates from raw meat, and the susceptibility of these isolates to antibiotics.

Materials and methods

Samples of beef and mutton were utilized in this cross-sectional investigation.To properly isolate the bacteria, beef and mutton samples were obtained straight from the slaughterhouse, placed in a sterile Ziploc bag, and then delivered to the lab immediately without freezing. There were 40 tissue samples collected from various portions of cows and goats. After that, samples were immediately swabbed directly with a sterile cotton swab over MacConkey agar using the direct cotton swab technique. After that, overnight incubation was performed at 37 degrees Celsius aerobically & maintaining the same procedures subculture was done. For stock preparation, Luria-Bertani broth, a nutrient-rich medium frequently used for the growth of bacteria and 500 microliters of 50% glycerol were used. At last, -20 and -80 degrees Celsius were utilized to store two copies of the stock for each of these copies. Organism detection tests were done on Eosin-methylene blue media and Kirby-Bauer, a double-disk diffusion test was done over Mueller-Hinton agar media using two different antibiotics along with beta-lactamase inhibitor.[7] Bauer used the disc diffusion technique to assess the susceptibility of ESBL-producing *E. Coli* by Clinical Laboratory Standards Institute recommendations (CLSI 2018). Microsoft Excel was used for the statistical analysis.

Result:

Among 40 samples, 95% of the pieces exhibited strain positivity & 5% no growth over MacConkey agar plates (Figure 1). Eosin methylene blue (EMB) medium organism detection test findings show that all 20 samples of beef are Klebsiella spp, whereas 10 and 8 samples of mutton are *E.coli* and *Klebsiella* spp (Table 1).

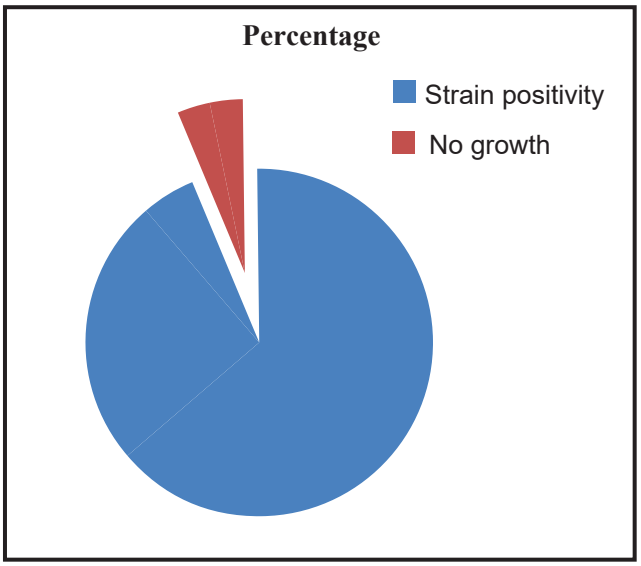

Figure 1: A graphic illustrates the appearance of *Entero-bacteriaceae* in total samples onMacConkeyagar plates by direct swab approach.

Table 1: Distribution of micro-organisms in beef & mutton samples.

Name of organism	Beef (n=20)	Mutton(n=20)
E.coli	0	8
klebsiella	20	10
No growth	0	2

During the screening process, it was observed that a total of 7 strains exhibited positive for ESBL producing bacteria among the 40 overall meat samples obtained (Table 2) & Five (or 25%) resisted the CAZ and AMC combos in case of beef (All are *klebsiella* spp).Regarding the mutton, there were 20 that were resistant to CAZ+AMC (100%), and 12 were resistant to CTX+AMC (60%) (Figure 2).

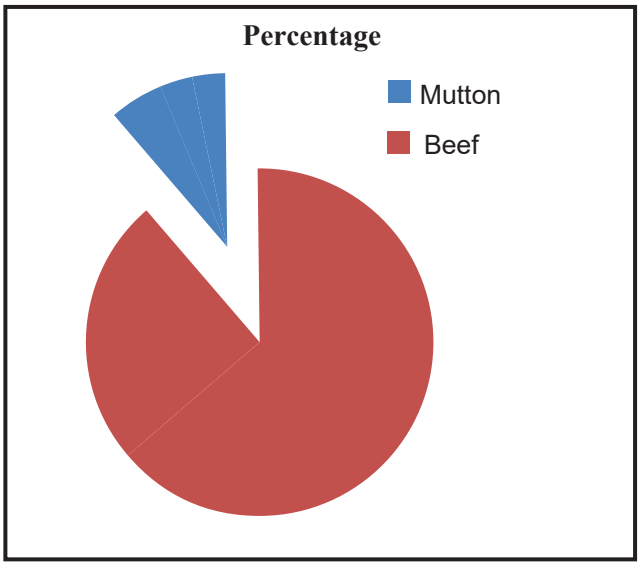

Figure 3: This pie chart represents percentage distribution of ESBL producers in animal based foods after Double-disc synergy tests.

Table 2: Double-disk diffusion test results only for ESBL producing bacteria (Zone diameter-mm).

Sample ID	Gram-negative bacteria	CAZ (30µg/ml)	CAZ(30µg/ml) + AMC(30µg/ml)	CTX (30µg/ml)	CTX(30µg/ml) + AMC (30µg/ml)	Results
P3B	*Klebsiella* spp.	0	25	28	25	ESBL
P7B	*Klebsiella* spp.	23	25	15	27	ESBL
P9A	*Klebsiella* spp.	23	23	20	26	ESBL
P9B	*Klebsiella* spp.	25	27	15	29	ESBL
P10A	*Klebsiella* spp.	30	30	7	30	ESBL
P10B	*Klebsiella* spp.	30	30	11	30	ESBL
BaPA	*E.coli*	22	24	4	20	ESBL

* CAZ = Ceftazidime, CTX= Cefotaxime, AMC= Amoxicillin- Clavulenic acid

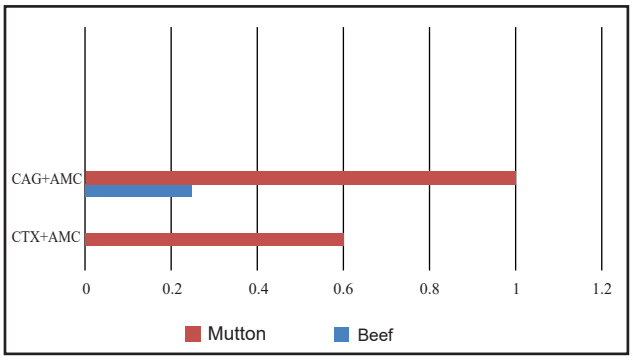

Figure 2: This column chart represents AMC resistance results in overall samples.

The distribution of ESBL-producing bacteria among various food types .The strains that produce ESBL account for 35%, of which 30% and 5% are found in beef and mutton, respectively among 40 food samples (Figure 3).

Table 3: Shows the antimicrobial drug resistance profile of microorganisms (*klebsiella* Species) isolated from beef samples in several Dhaka market locations

Sl No.	Strain name	CAZ (30µg/ml)	CAZ (30µg/ml) + AMC(30µg/ml)	CTX (30µg/ml)	CTX (30µg/ml) + AMC (30µg/ml)	Results
1.	P1A	30	25	35	35	None
2.	P1B	0	0	33	33	None
3.	P2A	26	26	30	28	None
4.	P2B	24	23	27	30	None
5.	P3A	0,	0	27	28	None
6.	P3B	1	25	28	25	None
7.	P4A	0	0	30	30	None
8.	P4B	0	0	30	30	None
9.	P5A	0	0	30	29	None
10.	P5B	0	0	25	20	None
11.	P6A	25	25	27	27	None
12.	P6B	28	28	15	20	None
13.	P7A	25	22	31	32	None
14.	P7B	23	25	15	27	ESBL
15.	P8A	32	30	30	32	None
16.	P8B	25	25	34	34	None
17.	P9A	23	23	20	26	ESBL
18.	P9B	25	27	15	29	ESBL
19.	P10A	30	30	7	30	ESBL
20.	P10B	30	30	11	30	ESBL

- P=Piece of sample, A= Anterior side of the sample, B= Posterior side of the sample
- Antibiotics susceptibility zone diameter in mm.
- On EMB media Klebsiella species purple in color

Tables 3, 4a, and 4b list 40 distinct samples, their sensitivity to cefotaxime (CTX), ceftazidime (CAZ), and clavulanic acid, along with the strain name and abbreviations for each.

Table 4 (a): Strain abbreviations & their meanings.

Strain name	Full form	Strain name	Full form
T	Thigh	B	Belly
Ta	Tail	Lu	Lungs
H	Head	C	Chest
L	Liver	Ba	Back
LA	Leg anterior	LB	Leg back

Table 4 (b): Shows the antimicrobial drug resistance profile of microorganisms isolated from mutton samples in several Dhaka market locations

Sl No.	Strain name	Organism	CAZ(30 µg/ml)	CAZ(30µg/ml) + AMC(30µg/ml)	CTX(30µg/ml	CTX(30µg/ml) + AMC (30µg/ml)	Results
1	LBPA	*E.coli*	0	0	25	27	None
2	BaPA	*E.coli*	0	0	4	20	ESBL
3	BPA	*E.coli*	0	0	0	0	None
4	BPB	*E.coli*	0	0	0	0	None
5	LuPA	*Klebsiella* spp.	0	0	20	15	None
6	LuPB	*E.coli*	0	0	25	25	None
7	LAPA	*Klebsiella spp*.	0	0	0	0	None
8	LAPB	*Klebsiella* spp.	0	0	0	0	None
9	LPA	*Klebsiella* spp.	0	0	0	0	None
10	LPB	*E.coli*	0	0	0	0	None
11	CPA	*Klebsiella* spp.	0	0	13	13	None
12	CPB	*Klebsiella* spp.	0	0	22	22	None
13.	TaPA	*E.coli*	0	0	0	0	None
14	TaPB	*E.coli*	0	0	0	0	None
15	HPA	*Klebsiella* spp.	0	0	22	22	None
16	HPB	*Klebsiella* spp.	0	0	20	20	None
17	TPA	*Klebsiella* spp.	0	0	0	0	None
18	TPB	*Klebsiella* spp.	0	0	0	0	None
19	LBPB	Nil	0	0	0	0	None
20	BaPB	Nil	0	0	0	0	None

- P=Piece of the sample, A=Anterior side of the sample, B=Posterior side of the sample.
- On EMB media *Klebsiella* species purple in color & *E.coli* metallic sheen green in colour.

Discussion:

In the Dhaka metropolitan area, specifically in the neighborhood markets of Dhaka, a total of 40 animal-based foods were assessed to assess ESBL production. According to the Dhaka Tribune, red meat accounts for more than 50% of all meat consumption in Bangladesh. Additionally, it is believed that food obtained from the local markets in Dhaka may harbor pathogens capable of causing food borne illnesses. The published article, [8] brought attention to the potential health risks associated with retail beef due to inadequate sanitation and hygiene practices during processing and handling. *Escherichia coli (E. coli)* is thought to spread through meat because of the people and tools used to process it, such as knives and cutting boards, as well as poor hygiene and sanitation practic-

es.[9] The transmission of contamination during the slaughtering process may occur through various means, including the hands of the slaughter men, water, and equipment utilized. Among the 40 samples derived from the red meat, 95% belong to the family Enterobacteriaceae, which supports the study in Ghana in 2012,[10] where 65% Enterobacteriaceae was found in food samples. Among all the strains, 100% and 50% of *Klebsiella* spp were found in beef and mutton samples, respectively, however, this study strongly aligns with the investigations in Athens.[11] In this study, a number of strains (17.5%) showed phenotypic positivity for making the ESBL enzyme. This is different from other studies, which found conflicting results (45.9%) for this phenomenon, which was seen in China.[12] Five (or 25%) resisted the CAZ and AMC combos in case of beef (All are *klebsiella* spp). Regarding the mutton, there were 20 that were resistant to CAZ+AMC (100%), and 12 were resistant to CTX+AMC (60%). The mutton findings closely match the Nigerian study, [13] where 91 samples (82.7%) were resistant to AMC.

A variety of enteric illnesses, including diarrhea and endocarditis, as well as infections of the skin, soft tissues, joints, bones, eyes, respiratory tract, and urinary tract, are caused by the *Enterobacteriaceae* family.Consequently, it is now a growing problem. We only got 40 pieces of meats, so the sample size was really small. The sample was not collected from throughout the nation but rather from different parts of Dhaka city. The weight, size, and nutritional value of the meats are not taken into account in this investigation. We are unable to utilize human samples due to a lack of resources and time. The World Health organization has designated several Enterobacteriaceae superbugs because of their increased production and spread of Carbapenamaeses & ESBL.This study aims to assess the incidence of Enterobacteriaceae microorganisms that cause ESBL and their phenotypic characterization & antibiotic resistance pattern.

Conclusion:

This study shows that the ESBL-producing Enterobacteriaceae found in raw meat in Dhaka, Bangladesh, have high antibiotic resistance. The rising rate of antibiotic resistance is highlighted by the need to enact policies to rationalize antibiotic usage and build a national resistance surveillance system. These policies will help local communities produce antibiotic therapy guidelines. The most significant degree of public health and laboratory techniques must be significantly advanced to stop the spread of bacteria resistant to medications.

Conflict of interest:

The authors hereby declare that no conflict of interest exists.

Acknowledgment:

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July 2024, 2(2):23-26

Original article

The Role of Blood Group and Demographic Factors in Dengue Infection: Implication of Public Health in Bangladesh

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Abstract

Background: Dengue fever, caused by the Dengue virus, has become a global health concern, with its prevalence expanding significantly over the decades. ***Objective:*** This study aimed to investigate the relationship between ABO blood groups and susceptibility to dengue infection. ***Methodology:*** A cross-sectional study was conducted in Dhaka city, Bangladesh, from March to September 2023, involving 1200 individuals, including Dengue positive and negative cases. ***Result:*** The results showed that the blood group had a significant association ($p<0.05$*) with the frequency of Dengue virus infection. While blood type A had the lowest susceptibility to dengue infection, those with blood groups O and AB had a much higher risk. Age group and living in an urban area were found to be significant factors related to dengue infection ($p<0.05$*). ***Conclusion:*** These results underscore the importance of considering demographic characteristics and blood groups in understanding Dengue transmission dynamics and devising effective public health strategies for its control.

Keywords: Dengue virus, Dhaka, ABO blood types

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Introduction:

Before 1970, only nine countries experienced significant dengue epidemics. Now, dengue is endemic in over 100 countries across Africa, the Americas, the Eastern Mediterranean, South-East Asia, and the Western Pacific. The Americas, Southeast Asia, and the Western Pacific bear approximately 70% of the burden, with Asia being the most affected region.[1] Dengue virus is a RNA virus (Flaviviridae family) with four distinct serotypes (DENV 1-4) that are common in tropical climates.[2] Between January 1st and August 7th, 2023, Bangladesh reported 69,483 confirmed dengue cases and 327 deaths, with a higher than usual case fatality rate (0.47%). Notably, 63% of cases and 62% of deaths occurred in July 2023. This surge, starting earlier than usual, deviates from past patterns and indicates an unusual seasonality.[3] The transmission of these viruses occurs through the bite of an infected mosquito. Female mosquitoes, in particular, feed on the blood of various hosts, such as birds or mammals. During this feeding process, mosquitoes release saliva before and during the bite. Once the virus is transmitted, the newly infected host may experience symptoms.[4] Genetic factors are also

involved, along with environmental factors, incases of illness susceptibility. Despite exposure, not everyone gets infected, as individuals may have varying levels of susceptibility or resistance. Therefore, factors like genetics, i.e. blood group, and environmental conditions can affect disease development.[5] Various research studies speculate that the human leukocyte antigen (HLA) haplotype determines the propensity to dengue infection, but they have not identified any clear or precise polymorphisms. The ABO blood type is a component of innate immunity, and people with various ABO blood groups have varied susceptibility to or resistance to viral and bacterial infections and illnesses. Kaipainen and Vuorinen proposed a correlation between blood groupings and disease in 1960. The ABO blood group plays a considerable role in making a person susceptible or resistant to different bacterial or vector-borne diseases.[6] There are four major blood groups (blood types): A, B, AB, and O. Each blood group have RhD positive or RhD negative, for a total of eight blood groups and few specific blood groups are the most prevalent for vector-borne diseases i.e, dengue and chikungunya.[7] A person's susceptibility or resistance to dengue infections might be determined by testing their blood type. The primary objective of this study was to identify blood groups that may be correlated with an individual's resistance to or susceptibility to dengue infection.

Materials and Methods:

This was a retrospective observational study conducted in Dhaka city during the period from March to September 2023. Data regarding 600 dengue NS1 antigen/dengue immunoglobulin M (IgM) antibodies or both (IgM & IgG) positive serum samples as well as 600 dengue negative serum samples as a control group were collected by Simple Random Sampling method irrespective of age and sex from the clinical laboratory of Bashundhara Ad-din Medical College and Rushmono Specialized Hospital, Dhaka.

All patients with serological confirmation of dengue (NS1, IgM/IgG positivity) were done by immunochromatographic tests (qDetect™ dengue NS1 antigen and qDetect™ dengue IgM/IgG test kit (OMCH, Dhaka, Bangladesh), respectively. ABO and Rh blood grouping reports were collected from dengue patients as well as from dengue-negative persons. General information about dengue fever awareness was recorded through an interview with a semistructured, pre-tested questionnaire from the participants. Proper written consent was obtained from all the participants before data collection. A predesigned questionnaire was used in data collection. All collected data was processed and analyzed using Microsoft Excel version 10. Ethical clearance was obtained from the Ethical Committee of Bashundhara Ad-din Medical College and Hospital, Dhaka.

Results:

The present study investigated the relationship between demographic factors, including age group, sex, residency, ABO blood groups, and dengue infection status among a sample of 1200 individuals. Notably, individuals aged 40-59 years exhibited the highest proportion(42.67%) of dengue-positive cases and age group was significantly ($p<0.05$) associated with dengue infection. Additionally, urban residency was also significantly ($p<0.05$) correlated with a higher incidence of dengue infection. Dengue cases were higher in males (60.83%) compared to females (39.17%). However, there was no significant difference in dengue infection rates according to sex.

Table 1: Socio-demographic status of study participants

	Dengue Negative (n=600)	**Dengue Positive (n=600)**	**P Value**
Age group			
<20 years	173(28.83%)	98(16.33%)	**<0.05**
20-39years	214 (35.67%)	167(27.83%)	
40-59 years	129 (21.5%)	256(42.67%)	
60 years	84 (14%)	79(13.17%)	
Sex			
Male	387(64.5%)	365(60.83%)	0.189
Female	213(35.5%)	235(39.17%)	
Residency			
Urban	487(81.17%)	324(54%)	**<0.05**
Rural	113(18.83%)	276(46%)	

* Chi-square test done
* P value <0.05 is considered as significant

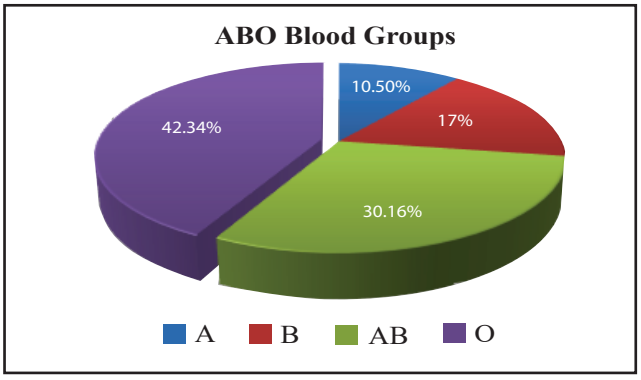

Figure 1: Pie chart showing the frequency distribution of ABO blood groups among dengue-positive cases

Table 2: ABO blood group distribution in dengue positive and negative cases

Blood Group	Dengue Positive (n=600)	Dengue Negative (n=600)	P Value
A	63	125	**<0.05**
B	102	180	
AB	181	138	
O	254	157	

* Chi-square test was used to compare the dengue positive cases and dengue negative controls
* P value <0.05 is considered as significant

About 63 of the 600 dengue-positive cases belonged to blood group A, 181 to blood group AB, and 254 to blood group O (Table 2). Table 2 also shows the distribution of blood types among the Dengue Negative or the control group. A significant association was found between the blood group with dengue positivity (p-value < 0.05).Among the dengue-positive cases, blood type O and AB had the highest rates of dengue infection 42.34% and 30.16% respectively,whereas blood group A had the lowest susceptibility to dengue infection (10.5%).

Discussion:

The emergence and re-emergence of viral diseases transmitted by vectors raise worldwide concerns about the hazard to health and the feasibility of prevention and control. Dengue (DENV) is the most frequent vector-borne viral (*Flaviviridae* family) illness in Asia and the sub-Asian region. It has drawn attention in recent years due to its rising prevalence, extending the geographical range and potential consequences from circulation and unexpected health issues and social burdens.[8] International tourists who visit endemic and epidemic areas have the possibility of acquiring an arbovirus infection. Dengue virus is the most often diagnosed arboviral illness in visitors and locals, whereas the specific arbovirus risk varies with geography.[5] In Bangladesh, there have been several dengue epidemic outbreaks throughout the previous few years. Changing anthropological behaviour, climate change and high mutation frequency are important determinants of arthropod-borne virus emergence. Arthropod-borne viruses adapt readily to new susceptible hosts by alteration of receptor specificity, transmission efficiency, antigenicity, ecological and environmental conditions.[9] Besides all these, there are one of the most important hereditary qualities of an individual is blood. The blood type-disease relationship may be highlighted by the ABO and rhesus (Rh) blood grouping systems. The basis for genetic and evolutionary research and disease may be the blood grouping system.[10] Even though few studies have documented correlations between dengue infections and specific blood groups, relatively very few have related the issue. In Bangladesh the phenotypic frequency of blood group A+ (21.3%), A- (1.6%), B+ (30.1%), B- (1.6%), AB+ (4.4%), AB- (0.7%), O+ (37.3%) and O- (3.0%) whereas Rh positive and Rh negative were 93.1% and 6.9%, respectively.[10] The present study included 1200 subjects among them 600 dengue positive patients (positive for dengue NS1 antigen/dengue immunoglobulin M (IgM) antibodies or both (IgM & IgG) and about 600 were dengue negative people irrespective of age and sex as a control group. Among them aged people (40 to 59 years) were more infected in dengue (Table 1) most probably due to increased prevalence of chronic diseases and other comorbidities in the elderly persons. In modern-day years, epidemiological studies from regions that traditionally demonstrated a classical pattern.[11] A seroprevalence study of adults aged 18–79 years in Singapore comparing the prevalence of anti-dengue IgG antibodies in the population in 2004 and 2010 showed that the age-standardized rates of seroprevalence were significantly lower in 2010 (54.4%) compared to 2004 (63.1%).[12] In this study, males (60.83%) are more infected than female and indicating the heightened risk of transmission in urban environments compared to rural areas (Table 1). It may be influenced by various factors such as sociocultural differences and differences in behavior and activities. Further research is needed to explore these factors and develop targeted prevention strategies. Dengue infection is significantly higher in urban areas ($p > 0.05$) than in rural areas might be due to rural environments based on population size, population density, housing density, infrastructure or surface cover type (impervious surfaces, vegetation), access to urban areas or distance to a road/urban center, environmental changes (including changes to landscapes, rural production systems, climate, land use, and transportation infrastructure) or agricultural practices.[13]

The result of the study demonstrated that individuals with blood group O (42.34%) and AB (30.16%) are more susceptible to dengue infection, while blood group A appeared to be protective against the virus. A research finding of Ravichandran et al.(2019),[6] individuals with blood group O were much less susceptible to dengue fever. In contrast, according to the research by Khode et al.(2013)[14] blood type O is a risk factor for dengue infections and AB blood group is statistically significantly associated with dengue fever as compared to the control group.[15]

Usually host genetic factors and the immune system play a pivotal role in the prevention of viral infections. Two

genetic variables, HLA and ABO blood categories, have been associated with vulnerability or resistance to infectious illnesses. Blood-type antigens are carbohydrate -based, with N-acetyl-d-galactosamine being the immunodominant sugar for A determinants and d-galactose for B determinants.[16] Galactosyltransferases synthesize these sugars. The primary antibody that detects these sugars is natural IgM. DENV patients develop IgM antibodies against glycosylated dengue viral proteins, which may cross-react with host cells. Further research is needed to determine whether an individual's ABO blood type and natural IgM antibody levels impact dengue illness or not.[6] Although previous studies have established a correlation between HLA and dengue disease, no specific polymorphisms have been identified that exhibit an unequivocal association with the severity of the disease. Therefore, it is important to determine whether a correlation exists between the severity of dengue disease and a polymorphism in the galactosyltransferase gene.

The research was constrained by its retrospective case record–based design, which prevented the investigation of several parameters.

Conclusion:

This study concludes that the O and AB blood group is associated with a higher risk of developing dengue fever than those with other blood groups. These results may provide valuable insights into the complex interplay between demographic characteristics and ABO blood groups in dengue infection. Understanding these associations can inform public health interventions aimed at controlling the spread of dengue virus infection.

Conflict of interest:

The authors hereby declare that no conflict of interest exists.

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Original article

A Retrospective Analysis of Antimicrobial Resistance Patterns of Predominant Pathogens Causing Neonatal Bloodstream Infection in Tertiary Care Hospitals of Dhaka City

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Abstract

Background: Neonatal bloodstream infections (BSIs) kill most poor country babies. Effective therapy requires time. A retrospective research in two tertiary hospitals in Dhaka, Bangladesh, examined the incidence of bacterial pathogens causing newborn BSIs and their antibiotic resistance patterns. ***Methodology:*** A total of 1825 blood samples were obtained from patients who were admitted at the Neonatal Intensive care unit of Ad-din Women's Medical College & Hospital, Dhaka & Rushmono specialized Hospital, Dhaka, Bangladesh from July 2020 to December 2021. All the blood samples were processed for culture using a BACT/Alert blood culture machine. Further identification & antimicrobial susceptibility tests were performed using standard microbiological procedure. ***Result:*** The incidence of bloodstream infection (BSI) in 1825 newborn intensive care facility blood samples was 17.2%. Acinetobacter spp. (23.9%) and coagulase-negative Staphylococci (CoNS) (52.7%) were the most common isolates. Staphylococcus species were resistant to ampicillin, cephradine, and erythromycin but sensitive to imipenem, vancomycin, and linezolid. 31.5% of Staphylococcus aureus and 47% of CoNS are methicillin-resistant. Staphylococcus epidermidis resists methicillin better than MRSA. One-tenth of isolated Staphylococcus species are vancomycin-resistant. Acinetobacter spp. was responsive to colistin, meropenem, piperacillin-tazobactum, and amikacin but resistant to ampicillin, cephradine, cefuroxime, and cefixime. 48% of identified Acinetobacter species are cephalosporin, fluoroquinolone, and aminoglycoside resistant, 14% are meropenem resistant, and 2.7% are Colistin resistant. ***Conclusion:*** The study emphasizes the importance of antibiotic stewardship for emerging resistance patterns and improves neonatal outcomes. Judicial use of antimicrobial agents in NICU is mandatory. These may provide valuable insights for healthcare professionals and researchers to develop effective antibiotic policies to combat neonatal sepsis and reduce associated morbidity and mortality.

Keywords: BSI: blood stream infection, ARO: antimicrobial-resistant organisms, NICU: neonatal intensive care unit, Coagulase-negative staphylococci (CONS), Acinetobacter

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Introduction:

One of the main causes of newborn death in underdeveloped nations is bloodstream infection. Nearly 50% of patients in neonatal critical care units in certain towns get bloodstream infections.[1,2] About ten million newborns are thought to pass away in the first five days of life, according to estimates from the World Health Organization. One in five newborns in certain poor nations is said to be affected by septicemia.[3] Neonatal infections can be obtained through vertical transmission from microbes in the birth canal or through environmental exposure due to inadequate health facilities. Neonatal septicemia is a clinical syndrome characterized by bacteremia, symptoms, and clinical indications that manifest during the first months of life. The delay in its diagnosis and treatment results in mortality.[4]

The compromised immune systems of neonates render them highly susceptible to infection. Neonatal sepsis is challenging to diagnose at the time of presentation and is associated with substantial morbidity and mortality. For this reason, individuals who are suspected of having sepsis are initiated on empiric antibiotic therapy until sepsis is definitively ruled out. Antimicrobial-resistant organisms (ARO) are the consequence of antibiotic overuse. ARO infection leads to a delay in the commencement of effective antibiotic therapy, a reduction in the number of treatment options, and an increase in morbidity and mortality. This is accompanied by a protracted hospital stay and increased hospitalization costs.[5]

Gram-positive organisms are responsible for up to 70% of nosocomial infections in neonates in numerous hospitals, with coagulase-negative staphylococci (CoNS) causing up more than half of this total.[6,7] Conversely, in certain developing nations, neonatal pathogens may be significantly more prevalent in gram-negative organisms, which are associated with a higher prevalence of antimicrobial resistance.[3] For instance, A. baumannii described the first outbreak of multiple-drug-resistant in particularly low birth weight neonates in the United States in 2004. Pathogens also exhibit variability over time.[8] But now a days, Acinetobacter is one of the most important microorganism responsible for neonatal blood stream infection in our country.[9]

Coagulase-negative staphylococci (CoNS) are common colonizers of human skin; but now adays it has become true pathogens, rather than simply culture contaminants, causing cardiovascular, joint, and bloodstream infections. Several authors have demonstrated CoNS as the most frequent cause of blood stream infection.[10–12]

The incidence, risk factors, pattern, antimicrobial sensitivities of pathogens, and mortality of neonatal sepsis vary across various regions and countries due to epidemiological differences.[13] Empiric antibiotic therapies are predicated upon the monitoring of antimicrobial sensitivity patterns in culture isolates. To expedite the prevention of neonatal morbidity and mortality, it is necessary to implement particular strategies customized to each country's specific circumstances. This may include the prevention and treatment of neonatal sepsis. To prevent the development of resistant microorganisms and limit inappropriate antibiotic use, it is recommended that antibiotic stewardship be implemented, which includes the appropriate selection and administration of antibiotics, de-escalation of therapy, and a multidisciplinary team approach to neonatal sepsis management. In addition, neonatal survival can be enhanced by the identification of risk factors, early diagnosis, and the implementation of therapy following local epidemiology and antimicrobial resistance patterns.

The objective of this investigation was to determine the most frequently encountered bacterial pathogens that contribute to neonatal BSI in two tertiary health care hospitals in Dhaka city that have NICU facilities. Additionally, we identified the antibiotic resistance patterns of the most prevalent pathogens and analyzed the pattern to ascertain the emergence of multidrug resistance in this region.

Materials and Methods

In this retrospective study, blood samples were obtained from patients who were admitted at the Neonatal Intensive care unit of Ad-din Women's Medical College & Hospital, Dhaka & Rushmono specialized Hospital, Dhaka, Bangladesh. A total of 1825 blood samples were processed from July 2020 to December 2021. All the blood samples were processed for culture the use of a BACT/Alert blood culture device to find out the presence of bacterial pathogens. Manual method has been utilized as well. Antimicrobial susceptibility tests were performed on the isolated pathogens using Kirby-Bauer disk diffusion method.

Bacterial isolation: Collected blood samples were directly inoculated into pediatric FAN blood culture bottle. Bottles were incubated in the BACT/Alert machine for 5 days. One drop of blood from growth positive culture bottles were directly inoculated onto MacConkey (MC) agar and blood agar (5% sheep blood) plates. Blood agar plates and MacConkey plates were then incubated at 37 °C in aerobic condition. The bacterial isolates were identified and confirmed by using standard microbiological and biochemical tests like Gram staining, growth on selective media, colony morphology on culture media, lactose fermentation, indole, and citrate utilization, H2S production, catalase, coagulase, oxidase, and urease test.All Staphylococci isolates were identified using colony morphologic analyses on Blood agar, Gram staining, and catalase and coagulase testing. Coagulase positive strains are considered as Staphylococcus aureus. Coagulase Negative Staphylococcus are further tested for Novobiocin susceptibility. Novobiocin sensitive strains ae considered as Staphylococcus epider-

midis. And remaining other species are classified as other CoNS. All tests are performed according to guidelines of World Health Organization.[14]

Antimicrobial Susceptibility Testing: According to Clinical and Laboratory Standards Institute (CLSI) guidelines of 2019 antimicrobial susceptibility testing was performed by using disc diffusion (Kirby-Bauer's) technique on Mueller Hinton agar (Merck, Germany).[15] The antibiotic discs of ampicillin (Amp), cephradine (Ceph), cotrimoxazole (Cot), ciprofloxacin (Cip), levofloxacin (Lev), nalidixic acid (NA), ceftriaxone (CTR), chloramphenicol (Clo), amoxyclav (AMC), cefixime (CXM), cefotaxime (CTX), gentamicin (Gen), amikacin (AK), azithromycin (Az), ceftazidime (CAZ), meropenem (Mero), piperacillin-tazobactam (PIT), colistin (Col) were used for Gram negative bacteria and ampicillin (Amp), cephradine (Ceph), cotrimoxazole (Cot), ciprofloxacin (Cip), levofloxacin (Lev), cefotaxime (CTX), ceftriaxone (CTR), amoxyclav (AMC), gentamicin (Gen), amikacin (AK), imepenem (Ime), cefuroxime, cefixime (CXM), oxacillin (Ox), cloxacillin (Clox), erythromycin (Ery), Novobiocin, doxycycline (Do), vancomycin (Van), linezolid (Lz) were used for Gram positive bacteria. All antibiotic discs are obtained from Oxoid Ltd, Bashingstore, Hampire, UK.

Microsoft Excel program were used for statistical analysis and figure generation.

Results

The frequency of newborn blood stream infection from 1825 blood culture samples from neonatal intensive care unit patients is shown in Table-1. 17.2% (313/1825) had bloodstream infection. About 52.7% are coagulase negative Staphylococcus (CoNS), 23.9% are Acinetobacter, 6.1% are Staphylococcus aureus and rests are others. No Salmonella Typhi or paratyphi was found. These data indicate that Coagulase-negative Staphylococci Spp. and AcinetobacterSpp. are the main causes of newborn blood stream infection, but Salmonella speceis not (Table 1, Figure 1).

Most Neonatal blood stream infections were caused by Staphylococci, and CoNS were most often isolated. Among the CoNS 124 was identified as S.epidermidis. They were sensitive to imipenem (86.06%), vancomycin (90.30%), and linezolid (100%) but resistant to ampicillin (92.73%), cephradine (83.03%), and erythromycin (52.12%) (Figure 2).

Resistance pattern of various Staphylococcus spp is categorized in detail in Table 2(a), 2(b), and 2(c).Staphylococcus species which showed resistance to penicillin, oxacillin andcloxacillin are considered as methicillin resistance. Among the S.aureus 31% are detected MRSA and 15.8% are VRSA [Table 2(a)]. Almost 55% of S.epidermidis were found Methicillin resistant, whereas vancomycin resistance was detected in 13% S.epidermidis [Table 2(b)]. CoNS than S.epidermidis show Methicillin resistance among 22% cases; but no vancomycin resistance is found [Table 2(c)].

Gram negative pathogens most frequent in newborn blood stream infections include Acinetobacter spp. They responded better to colistin (100%), meropenem (90%), piperacillin-tazobactam (92%), and amikacin (82%). Ampicillin (90%), cephradin (80%), cefuroxime (90%) and cefixime (80%) are more resistant (Figure 3). The identified Acinetobacter species are 48% cephalosporin, fluoroquinolone, and aminoglycoside resistant, 14% meropenem resistant, and 2.7% Colistin (Polymixin E)

Table 1: The water quality parameters of different stations of the four studied rivers

Pathogen	Number =N
CoNS	165
Acinetobacter	75
Staphylococcus aureus	19
Klebsiella	17
Enterobacter	18
E.coli	5
Proteus	5
Pseudomonas	4
S.pneumoniae	3
Enterococci	2
Salmonella typhi	0
Salmonella paratyphi	0
Total	313

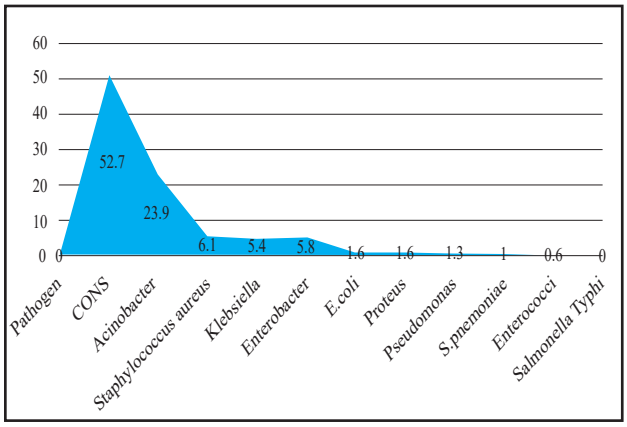

Figure 1: Distribution of bacterial pathogens causing neonatal bloodstream infection (Percentage)

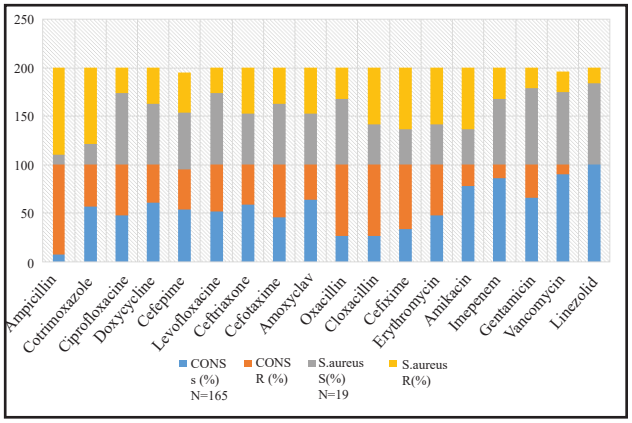

Figure 2: Susceptibility pattern of CONS spp. to different antimicrobial agents (n=165)

Table 2 (a): Categorization of Staphylococcus aureus spp. (N=19) according to their antibiotic resistance pattern:

Definition	Number	Percentage
MRSA(resistance to ampicillin, oxacillin, cloxacillin)	6	31.6%
MRSA +Resistance to Aminoglycosides, Tetracycline, Chloramphenicol, Macrolides)	4	21.1%
VRSA (Vancomycin resistant)	3	15.8%
Non-specific sensitivity pattern	6	31.6%

Table 2 (b): Categorization of *Staphylococcus epidermidis* spp. (N=124) according to their antibiotic resistance pattern

Definition	Number	Percentage
MRSE(resistance to ampicillin, oxacillin, cloxacillin)	68	54.9%
MRSE +Resistance to Aminoglycosides, Tetracycline, Chloramphenicol, Macrolides)	15	12.1%
VRSE(Vancomycin resistant)	16	12.9%
Non-specific sensitivity pattern	25	20.2%

Table 2 (c): Categorization of CONS spp. Other than *Staphylococcus epidermidis* (N=41) according to their antibiotic resistance pattern:

Definition	Number	Percentage
MRCONS(resistance to ampicillin,oxacillin, cloxacillin)	9	22%
MRCONS +Resistance to Aminoglycosides, Tetracycline,Chloramphenicol, Macrolides)	6	14.6%
VR CONS(Vancomycin resistant)	0	0
Non-specific sensitivity pattern	26	63.4%

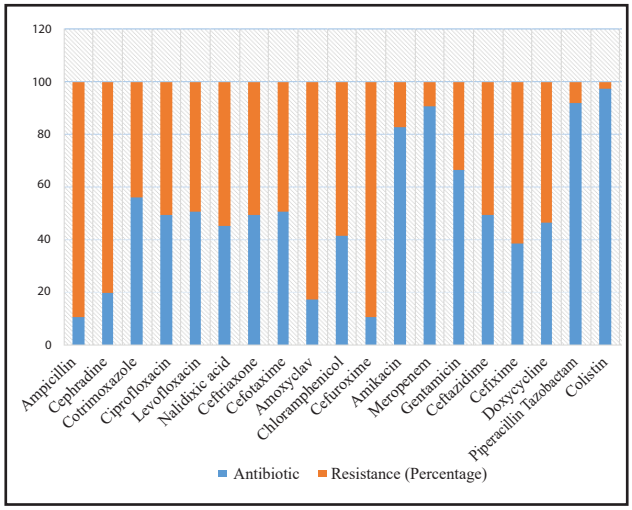

Figure 3: Susceptibility pattern of *Acinetobacterspp.* to different antimicrobial agents (n=75)

Table 3: Categorization of *Acinetobacter spp.* (N=75) according to their antibiotic resistance pattern:

Category	Definition	Number	Percentage
MDR	Resistant to- • All cephalosporin and inhibitor combination • Fluoroquinolones • Aminoglycoside	36	48%
XDR	MDR+ resistant to carbapenems	14	18.7%
PDR	XDR+ resistant to Polymixin E (Colistin)	02	2.7%
Unclassified	Miscellaneous pattern of sensitivity	23	30.6%

Discussion:

Neonatal bloodstream infections and antibiotic resistance are global concerns. This investigation detected 17.2% bloodstream infection without anaerobic culture. ICU patients are more likely to develop nosocomial BSIs. Due to vertical transmission from dealing, neonates are more susceptible to infection. The most common pathogens isolated from neonates were Coagulase-negative Staphylococci Spp(52.7%, 165/313) and AcinetobacterSpp (24%, 75/313) (Table 1, Figure 1).

CoNS are the main pathogen in Late-Onset Neonatal Sepsis (LONS), especially in premature babies. Several publications found that Gram-positive microorganisms cause more neonatal hospital-acquired bloodstream infections than gram-negative and yeast.[16–18] Within the first week of life, neonates become rapidly colonized by environmental pathogen.[19,20] With CoNS &Acinetobacter infection, central venous catheters (CVC), mechanical ventilation, parenteral nourishment, and other invasive skin or mucosa-breaching treatments, BSI risk increases significantly.[21,22]Thus, hospitalized newborns get most of the germs from parents, caregivers, and their surroundings.[23] Hospital workers may spread endemic strains of organisms for long durations.[24] Antibiotic resistance in skin-residing bacteria is minimal at birth but rises significantly in the first week of hospitalization. Perinatal antibiotic exposure selectively affects neonatal microorganism spectrum and antibiotic resistance.[25] CoNS &Acinetobacter spp. blood infection can occur in the babies without being under intensive care or antibiotics, mechanical ventilation or having indwelling catheters.[26]

However, we have observed an increase of susceptibility against Cotrimoxazole (50%) than the studies of previous decades.[27] If this trend continues, cheaper first-line medications for newborn blood stream infections may be attainable.

This research found high ampicillin, cephradine, and erythromycin resistance in Staphylococci spp., the main pathogen of newborn BSI. (Table 3). CONS were sensitive to amikacin (78%), imipenem (86%), vancomycin (90%) and linezolid (100%). S. epidermidis (55%), Staphylococcus aureus (31.5%) are methicillin-resistant.
MR Staphylococcus strains introduced into healthcare settings are transmitted and persistent-based on the availability of vulnerable patients, selective pressure from antimicrobial use, colonization pressure from larger numbers of colonized or infected patients, and hospital device implementation.[28]

Methicillin-resistant S. epidermidis far exceeds MRSA. S. epidermidis is linked to methicillin resistance more than S. aureus.[29] Since many S.aureus, S. epidermidis, and other CONS isolates are methicillin-resistant (MR), then glycopeptides are indicated for therapy.[30] However, about 10% of isolated Staphylococcus species are vancomycin-resistant. Other developing country research found similar results.[18,31]Currently, vancomycin and linezolid are effective treatments for CONS and S. aureus. The rise of Vancomycin-resistant staphylococcus (10%) worries medical professionals. hence CoNS infections must be properly diagnosed and treated.

We have found CoNS spp. are 46-67% more Cephalosporin-resistant. Previous investigations have demonstrated that antimicrobial resistance patterns resemble antibiotic usage in a hospital unit, and our NICU's substantial use of Beta-lactams and aminoglycosides may have selectively pressured commensal CoNS.[32]

Acinetobacter spp. is the second-most prevalent newborn blood stream pathogen (23.9%). In Bangladesh, Acinetobacterwas the most prevalent neonatal BSI isolate for decades. We found increased sensitivity to colistin (98%), meropenem (97%), piperacillin-tazobactum (80%), and amikacin (77.8%) (Figure 3). Many Bangladeshi and Indian investigations found similar susceptibility patterns.[9,33]Acinetobacter species in isolation are 48% MDR (cephalosporin, fluoroquinolone, and aminoglycoside), 14% meropenem (XDR), and 2.7% PDR (Colistin or Polymixin E) (Table 3)

Cefepime, ceftazidime, aztreonam, ciprofloxacin, gentamicin, and tobramycin were considered MDR, whereas amikacin, ampicillin sulbactam, imipenem meropenem, and pipeacillinTazobactum were sensitive.[34] Several reports showed that the normal flora are affected byusage of broad spectrum antibiotics and induced MDR A. baumannii.[35]

Carbapenem resistance in Acinetobacter spp. isolated from clinical samples ranged from 14-35% in Bangladesh and India.[9,35,36]The imipenem resistance of Acinetobacter spp. varied from 0 to 40% in the last decade, according to regional antibiogam data.[37] First-line carbapenem treatment for sepsis or suspected sepsis is currently given to many Bangladeshi newborns. Additionally, community-wide antibiotic usage complicates newborn sepsis treatment. As new antimicrobials become ineffective, Acinetobacter sp. are spreading fast. They gain resistance faster than Gram-negative organisms.[38] At present, the therapeutic options for infections caused by antibiotic resistant strains are limited. This suggests using antibiotics sparingly to treat Acinetobacter infections.

Limitation of the Study

Resource restrictions prevented us from distinguishing indoor and outdoor patient samples. So we couldn't identify nosocomial from community-acquired BSI. We couldn't get patient data on clinical symptoms or any infant BSI risk variables.

Conclusion

Our investigation identifies leading bacterial pathogens causing newborn bloodstream infections (BSI) in Dhaka city across various age groups and their antibiotic susceptibility patterns. Our data shows that CoNS are the main neonatal pathogen. CoNS strains were resistant to ampicillin, cephradine, erythromycin, or sensitive to imipenem, vancomycin, and linezolid. More Staphylococcus spp. are now resistant to methicillin and vancomycin, while Acinetobacter is resistant to third-generation cephalosporins and carbapenems, compared to a decade ago. Clinicians and policymakers should focus on this. Our results could assist healthcare personnel give better treatment to their patients and researchers and policymakers establish suitable antibiotic policies to tackle future infectious disease concerns.

Conflict of interest

The authors have no relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript.

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Review article

Unraveling the Complexity of ESBL and MBL: Insights into Pathogenesis, Mechanisms, and Health Ramifications

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Abstract

The emergence and spread of extended-spectrum β-lactamases (ESBLs) and metallo-β-lactamases (MBLs) among bacterial pathogens pose a formidable threat to global health. This review article provides a comprehensive examination of ESBL and MBL-related health hazards, focusing on their pathogenesis, mechanisms of action, and clinical implications. Through an extensive analysis of current literature, this review elucidates the intricate interplay between bacterial enzymes and antimicrobial agents, delineating the molecular mechanisms underlying ESBL and MBL-mediated resistance. Furthermore, it explores the epidemiology of ESBL and MBL-producing bacteria, encompassing transmission dynamics, risk factors, and global prevalence patterns. The clinical impact of ESBL and MBL-related infections is also addressed, including challenges in diagnosis, treatment options, and patient outcomes. By synthesizing insights from microbiology, epidemiology, and clinical medicine, this review aims to provide a comprehensive understanding of ESBL and MBL-related health hazards. Additionally, it discusses strategies for infection control, antimicrobial stewardship, and public health policy to mitigate the escalating threat posed by ESBL and MBL-producing bacteria..

Keywords: ESBL, MBL, AMC

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Introduction

The widespread dissemination of antibiotic resistance among bacterial pathogens presents a formidable challenge to global public health.[1] Among the diverse mechanisms of antimicrobial resistance, the emergence and spread of extended-spectrum β-lactamases (ESBLs) and metallo-β-lactamases (MBLs) pose significant threats to the efficacy of β-lactam antibiotics, a cornerstone of modern medicine.[2] ESBLs and MBLs are enzymes capable of hydrolyzing a broad spectrum of β-lactam antibiotics, including penicillins, cephalosporins, and carbapenems, thereby conferring resistance and limiting treatment options for bacterial infections.[3] The complex interplay between bacterial enzymes and antimicrobial agents underlies the pathogenesis, mechanisms, and health implications of ESBL and MBL-mediated antibiotic resistance. Understanding the molecular underpinnings of ESBL and MBL production is essential for unraveling the transmission dynamics, epidemiology, and clinical impact of multidrug-resistant pathogens.[4] The genetic basis of ESBL and MBL production underscores the remarkable adaptability of bacterial pathogens in response to selective pressures imposed by antibiotic use. Through horizontal gene transfer and genetic recombination, bacteria acquire genetic elements encoding ESBLs and MBLs, facilitating the dissemination of resistance determinants among diverse bacterial species and geographic locations.[5] Characterizing the evolutionary origins and genetic determinants driving ESBL and MBL production is critical for elucidating the epidemiology and evolution of antibiotic resistance.[6] At the molecular level, ESBLs and MBLs employ diverse mechanisms to confer resistance to β-lactam antibiotics.

ESBLs typically reside in the periplasmic space of Gram-negative bacteria, where they hydrolyze β-lactam antibiotics and render them ineffective against bacterial targets.[7] MBLs, on the other hand, utilize zinc ions as cofactors to catalyze the hydrolysis of β-lactam antibiotics, thereby circumventing the inhibitory effects of β-lactamase inhibitors.[8] The clinical implications of ESBL and MBL-related infections extend beyond individual patient outcomes to encompass broader public health concerns.[9] ESBL and MBL-producing bacteria are associated with increased morbidity, mortality, and healthcare costs, necessitating comprehensive infection control measures and antimicrobial stewardship programs to mitigate their impact.[10] Furthermore, the global spread of ESBL and MBL-mediated antibiotic resistance underscores the urgent need for collaborative efforts to address this growing threat on a global scale. In light of the escalating challenge posed by ESBL and MBL-producing bacteria, this review aims to provide a comprehensive exploration of their pathogenesis, mechanisms, and health implications. By synthesizing insights from microbiology, epidemiology, and clinical medicine, this review seeks to inform strategies for infection control, antimicrobial stewardship, and public health policy in the fight against antibiotic resistance.

Mechanisms of ESBL and MBL Production

The mechanisms underlying the production of extended-spectrum β-lactamases (ESBLs) and metallo-β-lactamases (MBLs) are pivotal to understanding the development of antibiotic resistance in bacterial pathogens.ESBLs and MBLs are enzymes that confer resistance to β-lactam antibiotics through various molecular mechanisms. The genetic basis of ESBL production involves the acquisition of resistance genes encoding these enzymes, often facilitated by mobile genetic elements such as plasmids and transposons. Similarly, MBLs utilize zinc ions as cofactors to catalyze the hydrolysis of β-lactam antibiotics, rendering them ineffective against bacterial targets.[11] Understanding the molecular structure and function of β-lactamases, along with the genetic determinants driving ESBL and MBL production, is essential for elucidating the mechanisms of antibiotic hydrolysis and resistance.[12]

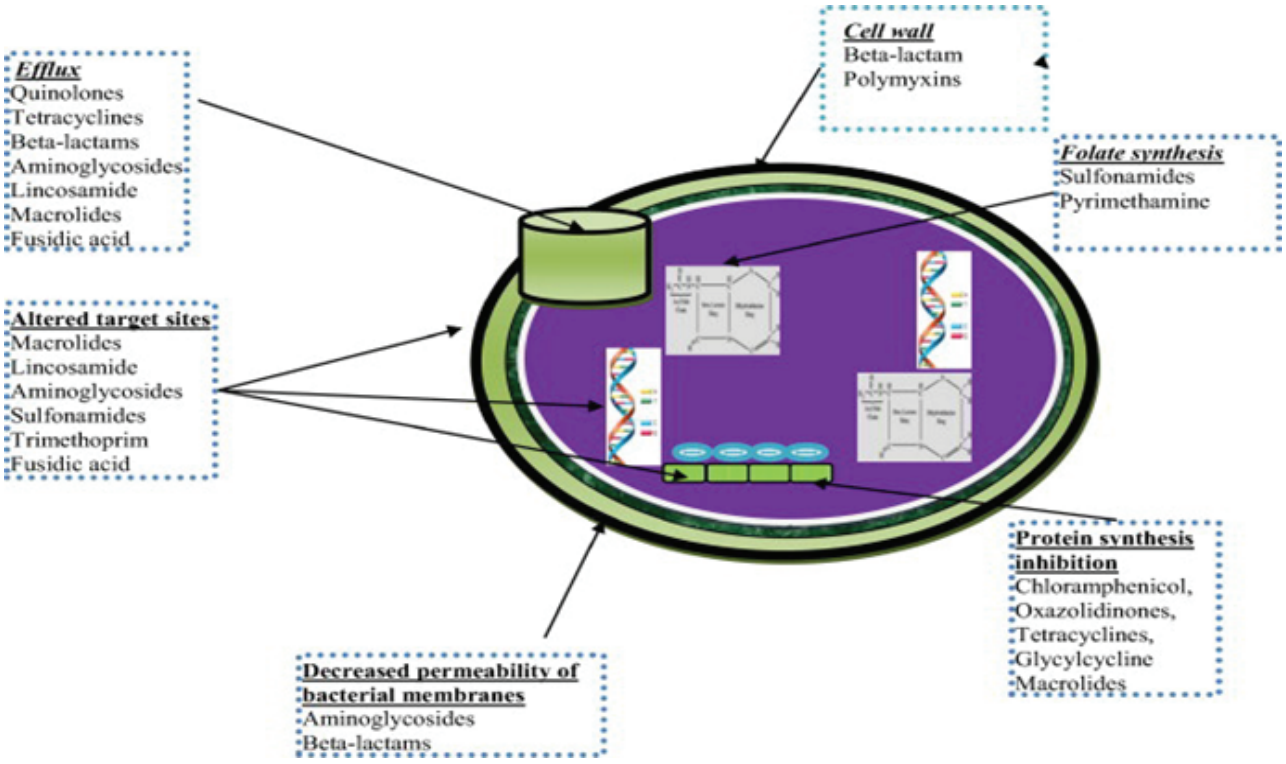

Figure 1: Antibiotic resistance vs. antimicrobial activity mechanism.[7]

This knowledge provides insights into the evolutionary dynamics of antibiotic resistance and informs the development of novel therapeutic strategies aimed at combating multidrug-resistant pathogens.

Pathogenesis of ESBL and MBL-Producing Bacteria:

Understanding the pathogenesis of extended-spectrum β-lactamase (ESBL) and metallo-β-lactamase (MBL)-producing bacteria is essential for elucidating their role in colonization, infection dynamics, and virulence.[13] Colonization by ESBL and MBL-producing bacteria often precedes the development of clinical infections and serves as a reservoir for transmission within healthcare settings and the community.[14] Once established, these bacteria interact with the host through complex mechanisms, including adhesion, invasion, and evasion of host immune defenses. Virulence factors such as adhesins, toxins, and biofilm formation contribute to the ability of ESBL and MBL-producing bacteria to cause invasive infections and evade host immune responses.[15]

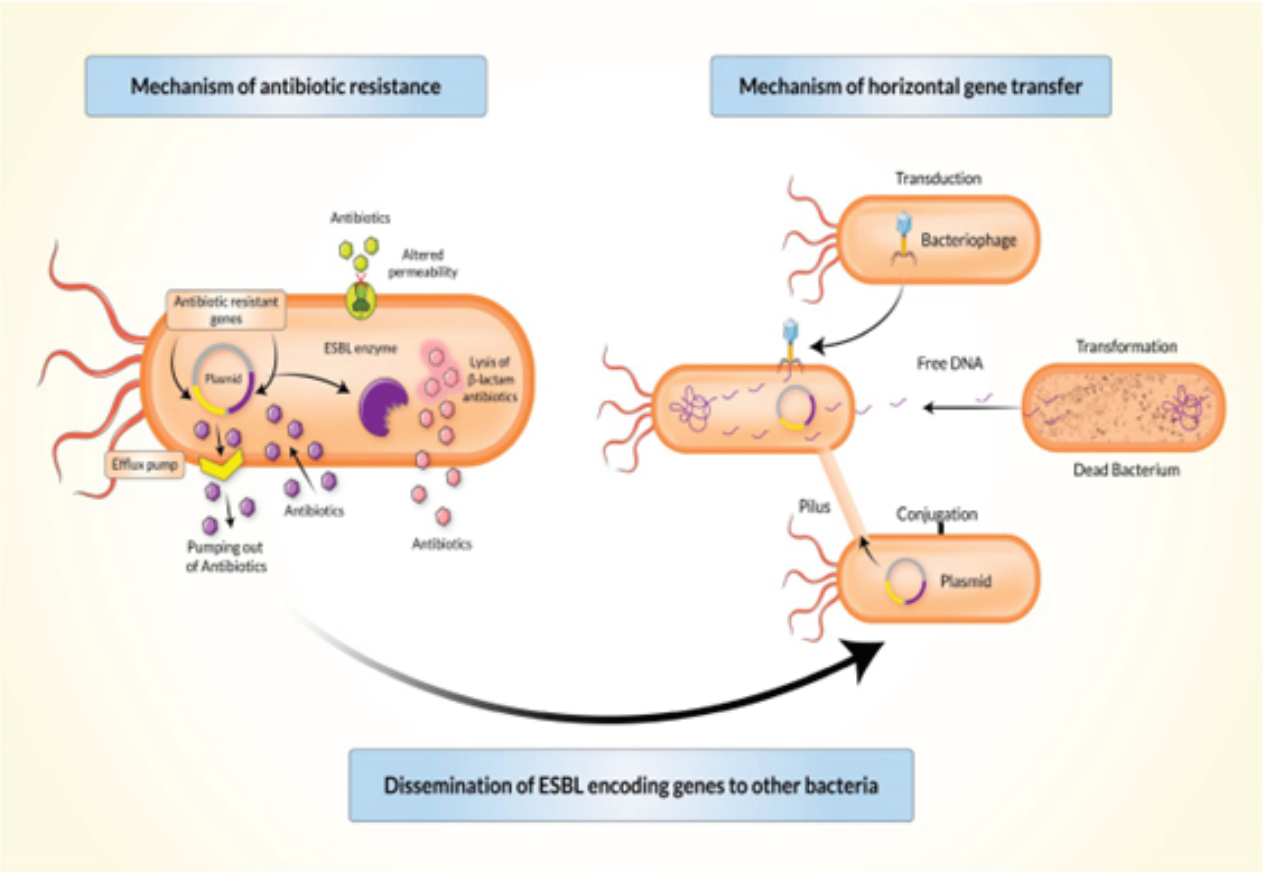

Figure 2 : Mechanism of antibiotic resistance & horizontal gene transfer[2]

Furthermore, the acquisition of antibiotic resistance genes may confer a fitness advantage, allowing these bacteria to thrive in the presence of selective pressure exerted by antimicrobial agents.[16] Understanding the interplay between colonization dynamics, host-pathogen interactions, and virulence factors is critical for devising strategies to prevent and control ESBL and MBL-related infections.

Epidemiology and Transmission Dynamics

The epidemiology and transmission dynamics of extended-spectrum β-lactamase (ESBL) and metallo-β-lactamase (MBL)-producing bacteria play a crucial role in shaping the global burden of antibiotic resistance.[17] Distinctions between healthcare-associated and community-acquired infections highlight the diverse settings in which ESBL and MBL-producing bacteria can emerge and spread.[18] Healthcare-associated infections often occur in hospital settings, where factors such as prolonged hospital stays, invasive procedures, and exposure to antimicrobial agents contribute to the selection and dissemination of multidrug-resistant pathogens.[19]

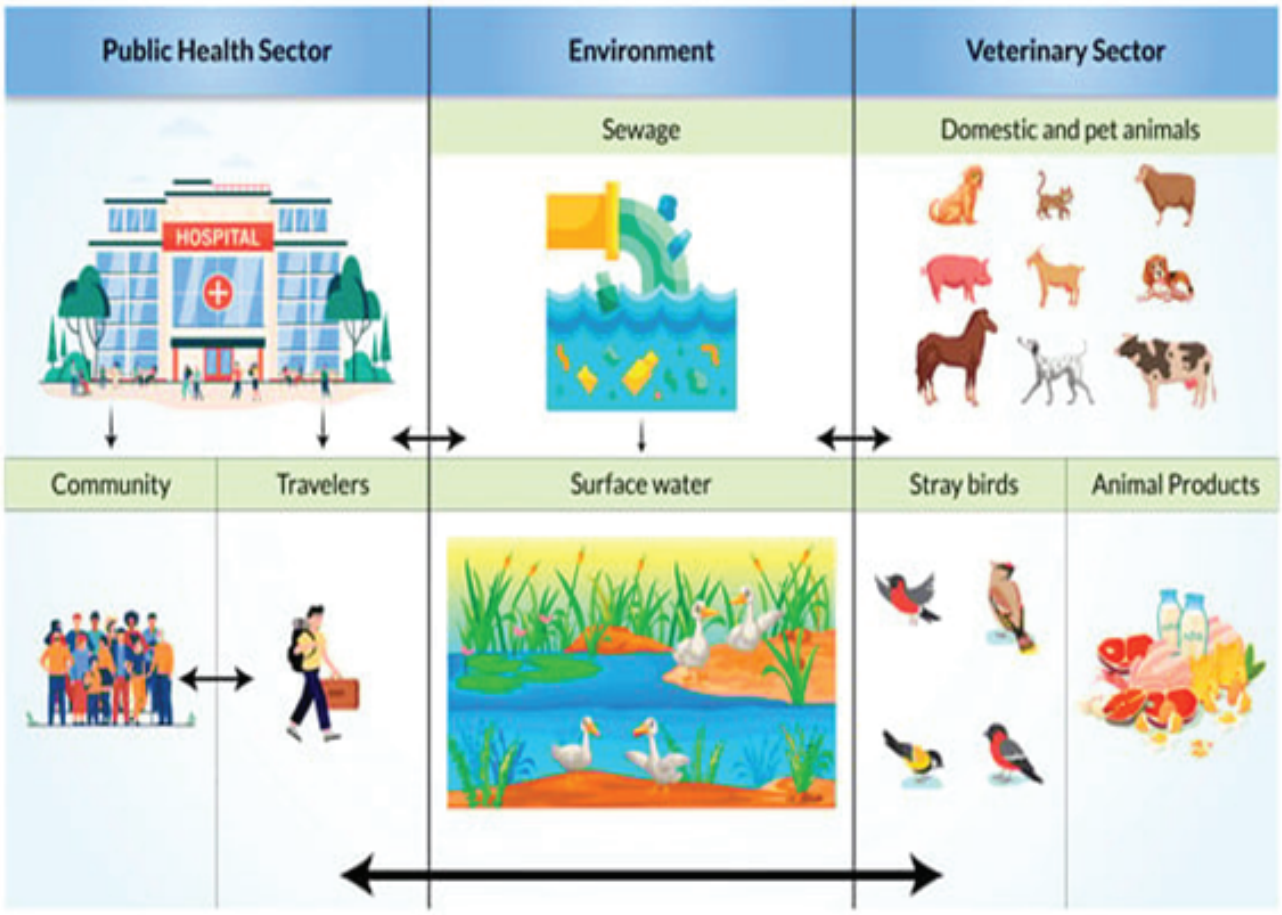

Figure 3 : Possible transmission pathways of Extended-Spectrum β-Lactamase (ESBL)-producing bacteria[2]

Conversely, community-acquired infections may arise in individuals with no recent healthcare exposure, underscoring the importance of community-based surveillance and infection control measures. Global surveillance data provide valuable insights into the prevalence and distribution of ESBL and MBL-producing bacteria, facilitating the identification of emerging hotspots and trends in antimicrobial resistance.[20] Factors contributing to the transmission and spread of ESBL and MBL-producing bacteria include inadequate infection control practices, international travel, and the dissemination of resistance genes through mobile genetic elements.[7] Addressing these challenges requires a multifaceted approach that encompasses enhanced surveillance, infection prevention strategies, and antimicrobial stewardship efforts.

The clinical implications of extended-spectrum β-lactamase (ESBL) and metallo-β-lactamases

The clinical implications of extended-spectrum β-lactamase (ESBL) and metallo-β-lactamase (MBL)-producing bacteria extend beyond individual patient outcomes to encompass broader challenges in healthcare delivery and antimicrobial stewardship.9 Infections caused by ESBL and MBL-producing bacteria are associated with increased morbidity, mortality, and healthcare costs compared to infections caused by susceptible strains.[21] The limited treatment options for multidrug-resistant pathogens further compound these challenges, often necessitating the use of last-line antibiotics with associated risks of toxicity and treatment failure. Antimicrobial stewardship efforts face significant hurdles in the management of ESBL and MBL-related infections, including difficulties in empiric therapy selection, optimal dosing regimens, and the potential for treatment failure due to resistance mechanisms.[22] Furthermore, the global dissemination of ESBL and MBL-producing bacteria complicates the landscape of infectious diseases, requiring collaborative efforts among healthcare providers, policymakers, and researchers to develop and implement effective strategies for infection control and antimicrobial stewardship.[2]

Health Hazards Associated with ESBL and MBL

The health hazards associated with extended-spectrum β-lactamase (ESBL) and metallo-β-lactamase (MBL)-producing bacteria are profound, encompassing increased morbidity and mortality rates, complications in vulnerable populations, and broader implications for public health and

infection control.19 Infections caused by ESBL and MBL-producing bacteria are often associated with adverse clinical outcomes, including treatment failure, prolonged hospital stays, and increased healthcare costs.[23] Vulnerable populations, such as elderly individuals, immuno-compromised patients, and those with underlying medical conditions, are at particular risk of experiencing severe complications from ESBL and MBL-related infections. Moreover, the global dissemination of multidrug-resistant pathogens poses significant challenges for infection control efforts, requiring stringent measures to prevent transmission within healthcare settings and the community.[24] Addressing the health hazards associated with ESBL and MBL-producing bacteria necessitates a multifaceted approach that encompasses enhanced surveillance, antimicrobial stewardship, and infection prevention strategies to mitigate the impact of antibiotic resistance on public health.[25]

Strategies for prevention and control for ESBL & MBL

Extended-spectrum beta-lactamase (ESBL) and metallo-beta-lactamase (MBL) producing bacteria pose significant challenges in healthcare settings due to their resistance to multiple antibiotics. Effective prevention and control strategies are essential to mitigate their spread and impact.[2] Key measures include stringent infection control practices such as hand hygiene, environmental cleaning, and appropriate use of personal protective equipment. Antibiotic stewardship programs play a crucial role in optimizing antibiotic use, thereby reducing selective pressure for resistance development.[26] Surveillance for ESBL and MBL-producing organisms, along with timely detection through laboratory testing, facilitates early intervention and containment efforts.[27] Additionally, implementing measures to prevent horizontal gene transfer, such as limiting unnecessary antimicrobial exposure and employing molecular typing techniques, can aid in understanding transmission dynamics.[28] Comprehensive strategies integrating these components are pivotal for combating the dissemination of ESBL and MBL-producing bacteria in healthcare settings, ultimately safeguarding patient outcomes and public health.[25]

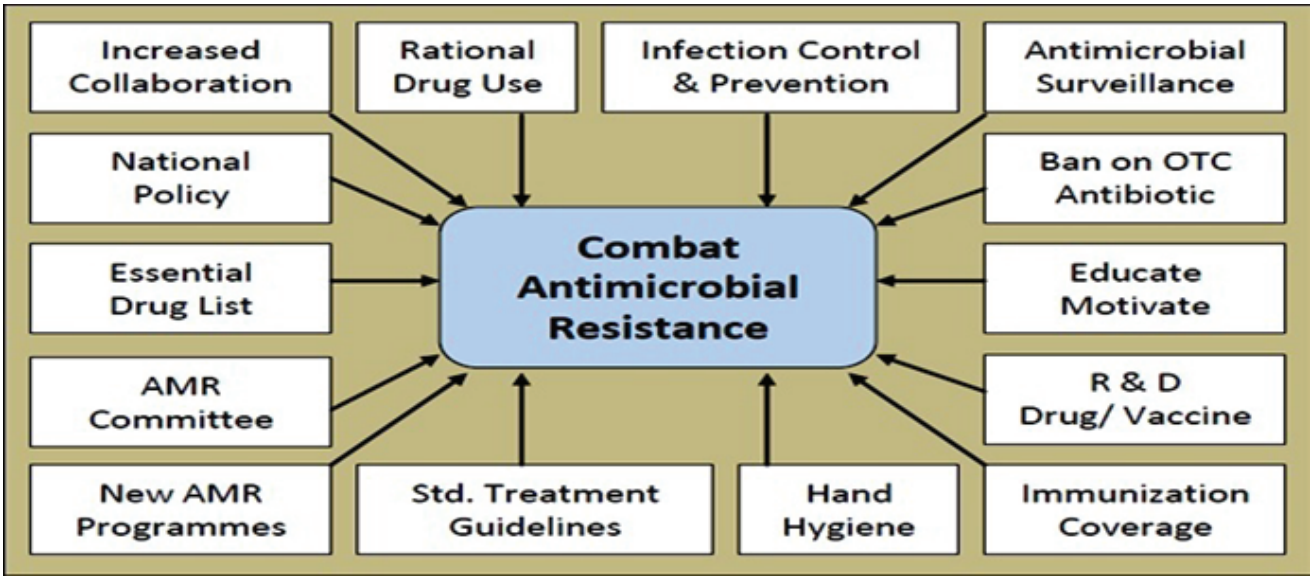

Figure4: The key points to AMR control strategies[29]

Unveiling the Pathogenesis, Mechanisms, and Imperatives of ESBL and MBL Resistance

Our journey through the pathogenesis and mechanistic intricacies of Extended-Spectrum Beta-Lactamase (ESBL) and Metallo-Beta-Lactamase (MBL) resistance has illuminated the formidable challenges posed by these enzymes, stemming from genetic mutations within bacterial populations.[13] ESBLs neutralize penicillins and cephalosporins, while MBLs extend resistance to carbapenems and beyond, showcasing the adaptability of bacterial pathogens.4The ramifications are profound, elevating morbidity, mortality rates, and straining healthcare resources.[30] Urgent action is warranted through enhanced surveillance, infection control protocols, and novel therapeutic strategies. Looking ahead, a unified, multidisciplinary approach involving healthcare providers, researchers, policymakers, and the public is crucial in combating this global threat, preserving antimicrobial efficacy, and safeguarding public health.[31] In closing, our exploration underscores the imperative for concerted action, determination, and resilience in confronting antimicrobial resistance.

Conclusion

In conclusion, our journey through the intricacies of ESBL and MBL resistance has provided invaluable insights into their pathogenesis, mechanisms, and health ramifications. As we unravel the complexity of these enzymes, it becomes increasingly clear that they pose significant challenges to public health, necessitating urgent action. By enhancing surveillance, implementing stringent infection control measures, and fostering innovation in therapeutic strategies, we can hope to mitigate the impact of multidrug-resistant bacterial strains. However, confronting antimicrobial resistance requires a unified, multidisciplinary effort involving healthcare professionals, researchers, policymakers, and the public. Together, we must remain vigilant, resilient, and committed to preserving the efficacy of antimicrobial agents for the well-being of current and future generations.

Conflict of interest

The authors hereby declare that no conflict of interest exists.

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Case report

Pyrexia of Unknown Origin in a Bangladeshi Citizen

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Abstract

The term pyrexia of unknown origin refers to a condition in which the patient has an elevated temperature (fever) but, despite investigations by one or more qualified physicians, no explanation is found. It was introduced by Petersdorf and Beeson in 1961. A 36-year-old male patient, attended in Bashundhara Ad-din Medical College Hospital (BAMCH), presented with a two-months' history of fever. Notably, his fever was unaccompanied by other symptoms such as cough, runny nose, changes in appetite, chills, abdominal pain, urinary issues, diarrhea, weight loss, chest pain, dyspnea, or palpitations. Over this period, he underwent treatment with multiple antibiotics including Azithromycin, Ceftriaxone, Meropenem, Vancomycin, Gentamycin, and Doxycycline. While empirical antibiotic therapy can be life-saving in certain situations, it may also impede accurate diagnosis. Typically, drug-induced fever manifests within 7–10 days, though this timeframe can vary considerably from hours to several months. This case describes that while medicine is often viewed as a remedy for illnesses, it can inadvertently contribute to their onset. In spite of the fact that medicine is sometimes thought of as a cure for ailments, this case illustrates how it might unintentionally hasten their development.

Key word: Pyrexia of unknown origin, Drug fever, Quantiferon TB Gold Test.

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Introduction

Pyrexia of unknown origin (PUO) refers to fever that is not resolved spontaneously, while its etiology cannot be determined despite extensive diagnostic workup.[1] The term PUO was first introduced by Petersdorf and Beeson in 1961. It is characterized by a fever exceeding 38.3 °C (> 101 °F) on multiple occasions, lasting for over three weeks, and the inability to diagnose specifically after one week of hospital investigations.[2] There are many causes of PUO like Malignant: lymphoma, renal cell carcinoma, Infectious diseases: milliary TB, brucellosis, Q fever, Inflammatory disorders: Adult still's disease, juvenile rheumatoid arthritis, and Giant cell arteritis. Miscellaneous disorders: drug fever, cirrhosis, and Idiopathic.

Case Summary

Mr. Nazrul (pseudonym), a 36-year-old male patient, presented in the outpatient department of BAMCH with a two-month history of fever, which was not associated with cough, runny nose, changes in appetite, chills, rigor, abdominal pain, burning sensation during micturition, diarrhea, or weight loss, chest pain, dyspnea, palpitation, orthopnea, and any urinary symptoms. He had been admitted to various hospitals in different locations, including Chittagong, Khulna, Barishal, and Cumilla, for approximately 32 days. During this period, he received several antibiotics, including Azithromycin, Ceftriaxone, Meropenem, Vancomycin, Gentamycin, and Doxycycline.

Mr. Nazrul revealed no known drug allergies or any other comorbidities. He lived in the Middle East for around 12 years and had a family history of pulmonary tuberculosis. He also reported a previous episode of enteric fever two months back. Apart from the raised temperature, his blood pressure was 110/70 mmHg, and his pulse rate was 76 beats/min. All other physical and systemic examinations revealed no abnormalities.

Past investigations reveal persistently raised Erythrocyte sedimentation rate, C reactive protein. Other investigations, including Serum electrolyte, Serum creatinine, Routine microscopic examination of Urine, Dengue Antibody, Urine culture & and sensitivity, Blood culture & and sensitivity, ICT for malaria-kala azar, Triple antigen, Chest X-ray P/A view, reveal no significant abnormalities (Appendix 1). We have advised for HIV screening, Bone marrow study, and QuantiFERON TB Gold test, where QuantiFERON TB Gold test was positive.

As there were no compelling indications, and for observation purpose, we had decided to stop all drugs. The fever was subsided within the next three days. As no sign of active TB, instead of starting Anti TB drugs, we put the patient on observation. Then we made a follow-up visit after 15 days with CBC, CRP (6 mg/l), Serum creatinine, Peripheral blood film, Urine R/M/E, CXR P/A view, and acute phase reactants like ESR become normal.
The patient was apyretic for 14 days and all other investigations with ESR became almost normal

Discussion

Evaluation of a patient with PUO is difficult. Initial assessment involves a thorough history covering occupation, travel, animal exposure, family diseases, and past illnesses. Misdiagnosis often stems from inadequate evaluation, by delayed testing and inappropriate investigations. Repeated interviews and file reviews can reveal crucial diagnostic clues.[1] A fever of unknown origin without any identifiable source of infection was observed in our patient, Mr. Nazrul.

While initiating empirical antibiotics might be life-saving in some circumstances, it might also pose a barrier to confirming a diagnosis. Typically, the onset of drug-induced fever occurs within 7–10 days, but this timeframe can vary from hours to several months.[3] Upon discontinuation of the responsible drugs, fever resolution usually takes around 2–3 days. However, if other hypersensitivity symptoms like a widespread rash accompany the fever or if a particular agent is slow to be eliminated, the fever may persist for additional days or even weeks. Our patient used a variety of antibiotics, including beta-lactamase, over an extended period and the cessation of all antibiotics resulted in the resolution of fever within 72 hours of hospitalization. The possibility of drug fever could be considered in this case due to several factors, such as Mr. Nazrul has been treated with various antibiotics over a significant period without resolution of his fever, which could suggest an underlying cause other than infection. The absence of symptoms commonly associated with infections, such as cough, runny nose, diarrhea, or urinary symptoms, despite the prolonged fever, may raise suspicion for an alternative diagnosis like drug fever. Despite extensive investigations, including blood and urine cultures, dengue antibody test, and chest X-ray, HIV screening, Bone marrow study no significant abnormalities were found, which could indicate that the fever is not due to an infectious process. Positive QuantiFERON TB Gold test is suggestive of latent tuberculosis infection, it is not conclusive evidence of active tuberculosis. Therefore, other potential causes for the fever, such as drug reactions, should still be considered.

The following reasons suggest that this may not be a case of drug fever, such as despite discontinuation of various antibiotics, the fever persisted, which is not typical for drug fever. Drug fever usually resolves once the causative medication is stopped. Normal findings on HIV screening and bone marrow studies suggest that infectious and hematologic causes for the fever are less likely, which could potentially rule out drug-induced reactions as well. Positive QuantiFERON TB Gold test indicates latent tuberculosis infection. However, it does provide another potential explanation for the fever. Our patient demonstrated a positive QuantiFERON TB Gold test result. It is a simple blood test that aids in the detection of Mycobacterium Tuberculosis. QFT is an interferon¬ Gamma (IFN¬Y) release assay, commonly known as an IGRA, and is a modern alternative to the Tuberculin skin test (TST, PPD, or Mantoux). A positive QFT can't distinguish between active TB disease and latent TB infection and is intended for use with risk assessment, radiography, and other medical and diagnostic evaluations. Like any diagnostic aid, QFT can't replace clinical judgment.[4] QFT is more sensitive and specific than TST.[5,6] QFT Gold test is 94.1% sensitive and 97.3% specific, on the other hand, TST is 68.9% sensitive and 59% specific. QFT TB Gold test may result in a false positive in Previous TB vaccination with BCG and Infection with non-tuberculosis mycobacteria.

The diagnosis of TB relies on a combination of clinical evaluation, radiological findings, and other laboratory investigations, including the QFT Gold test. A positive result indicates exposure to Mycobacterium tuberculosis but does not necessarily mean active disease. In our case Mr. Nazrul, while the QFT is positive, it's crucial to consider the absence of clinical symptoms suggestive of active TB, along with normal findings on other investigations such as chest X-ray and bone marrow study.

Given the absence of symptoms consistent with active TB and normal findings with other tests, the decision is not to initiate anti-TB treatment. However, regular monitoring and follow-up may be necessary to ensure early detection and management or development of active TB in the future, especially considering his history of living in a high TB burden area and family history of pulmonary TB.

Conclusion

Our case highlights the paradox that while medicine is often seen as the solution to diseases, it can also inadvertently invite and contribute to their occurrence. This emphasizes the importance of careful consideration in medical decisions to ensure optimal patient outcomes.

Conflict of interest

The authors hereby declare that no conflict of interest exists.

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Appendix 1: Investigations and their reports

Date	Test Name	Findings
30/12/23	QuantiFERON-TB Gold test	Positive
30/12/23	Bone marrow Study	Normal
30/12/23	HIV Screening	Negative
26/12/23	Complete blood count with Erythrocyte sedimentation rate	Hemoglobin 8.4 gm/dl, White cell count 15100/µL , Neutrophil 82.8%, Erythrocyte sedimentation rate 100mm/hour, Platelet count 768000/µL
26/12/23	Serum creatinine	0.7mg/dl
26/12/23	Routine microscopic examination of Urine	Normal
25/12/23	Chest Xray P/A view	Normal
17/12/23	Serum electrolytes	Normal
10/12/23	Urine culture & sensitivity	Normal
10/12/23	Blood culture & sensitivity	Normal
09/12/23	Reticulocyte	1.5%
07/12/23	Serum lactate dehydrogenase	437.6U/L
07/12/23	Complete blood count with Erythrocyte sedimentation rate	Hemoglobin 10.2 gm/dl, White cell count 21500/µL , Neutrophil 79%, Erythrocyte sedimentation rate 100mm/hour, Platelet count 470000/µL
06/12/23	Complete blood count with Erythrocyte sedimentation rate	Hemoglobin 11.4 gm/dl, White cell count 17689/µL , Neutrophil 77%, Erythrocyte sedimentation rate 120mm/hour, Platelet count 420000/µL
05/12/23	MRI of Lumbo sacral spine	Disc Bulge at L4-L5 level abutting the L5 transversing nerve root on both side straightening of lumber lordotic curve. Hypo intense marrow singnal intensity in both T1-T2
04/12/23	ICT for malaria-kala azar	Negative
02/12/23	Tripple antigen	Normal
20/11/23	Dengue Antibody	Negative
19/11/23	Duplex scan of left lower limb	Suggestive of incompetence of sapheno popliteal valve of left lower limb. No DVT or Varicosity of superficial vein present
14/11/23	Ultrasonography of whole abdomen	Mild hepatomegaly
(follow up inv.) 15/01/24	CBC, Serum creatinine, Peripheral blood film, Urine r/m/e, CXR P/A view,CRP	Normal
	ESR	22 mm in first Hour

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