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Pathogen Profile and Antimicrobial Susceptibility Patterns in Patients with Peritonitis: A Prospective Hospital-Based Study in Northern Tanzania

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Abstract

Background

Peritonitis is a life-threatening surgical emergency and a major cause of morbidity and mortality worldwide. Appropriate antibiotic therapy is critical, yet misuse of antimicrobial agents contributes to the rise of resistant strains, especially in developing countries. Resistance leads to treatment failure and imposes financial burdens on both patients and healthcare systems. This study aimed to assess intra-abdominal pathogens and their antimicrobial susceptibility patterns among patients with peritonitis.

Methods

This was a hospital-based descriptive cross-sectional study conducted at Kilimanjaro Christian Medical Centre (KCMC), northern Tanzania, between October 2019 and April 2020. All patients with peritonitis admitted to the surgical department were enrolled. Intraoperative peritoneal swabs were collected and cultured. Antimicrobial susceptibility testing was performed using the Kirby-Bauer method on Müller-Hinton agar and interpreted according to Clinical and Laboratory Standards Institute (CLSI) guidelines. Data were summarized using descriptive statistics and supplemented with information from patients' clinical notes.

Results

A total of 39 patients were identified, of whom Thirty-five patients met eligibility criteria. The median age was 32 years (IQR: 20–48), and 51.4% were male. The most common isolates were *Escherichia coli* (42.4%) and *Klebsiella pneumoniae* (21.2%). Both were highly susceptible to amikacin and meropenem (100%), but showed low susceptibility to ceftriaxone (22% and 20%, respectively).

Conclusion and recommendations

High levels of resistance to commonly used empirical antibiotics were observed, highlighting the need to revise empirical therapy for peritonitis in this setting. Rational use of antibiotics, alongside close monitoring of treatment responses, is essential to reduce morbidity, mortality, and the spread of antimicrobial resistance.

Keywords: *Antimicrobial Susceptibility, Intra-Abdominal Pathogens, Peritonitis, Tanzania.*

Introduction

Peritonitis is a surgical emergency and a leading cause of surgical emergency admissions. Globally, peritonitis is a major cause of non-trauma death in emergency settings and the second leading cause of sepsis in critically ill patients (1). The causes can vary, but bacterial peritonitis can be classified into primary and secondary. Primary bacterial peritonitis is rare, but there is bacterial infection without loss of integrity of gastrointestinal tract (GIT). Secondary peritonitis is more common, where the integrity of the GIT is lost. Aerobes and anaerobes are often involved. Another cause of peritonitis is chemical leak, i.e., perforated gastric ulcer. These often progress to bacterial peritonitis due to the transfer of microorganisms (2). Peritonitis is often polymicrobial in origin. In the past, the rate of *E. coli* accounted for about 25%, while gram-positive cocci in over 30%. Currently, in secondary peritonitis, gram-negative bacteria can be found in 60% of the cases; more than 40% being *E. coli*, followed by *Klebsiella pneumoniae* (3).

Despite the advancements in surgical techniques, antimicrobial therapy, and intensive care support, management of peritonitis continues to be highly demanding, challenging, and complex. The mortality rate is greater than 40% if the source of infection is not controlled, and the failure rate is as low as 6% when controlled (4). The contamination of the peritoneal cavity can lead to a cascade of infection, sepsis, multisystem organ failure, and death if not treated in a timely manner, preferably with a multidisciplinary approach (5).

In Tanzania, the prevalence of peritonitis is reported to be 11.6% and mortality in developing countries like Tanzania ranges from 13-43%; 15.5% reported by Mabewa et al. Peritonitis is the commonest cause of admission in the surgical ward, associated with significant workload, complications and deaths (6,7).

Antibiotic resistance is rising in developing countries due to wrong medical practices like self-prescription, poor adherence, and availability over-the-counter. These also increase mortality and financial consequences (8). The prevalence of antimicrobial-resistant pathogens differs geographically due to the differences in the consumption of antibiotics, and as a result the guidelines for prophylaxis and treatment should vary between countries based on their respective antibiotic-resistant rates (9).

Peritonitis remains a major public health concern regardless of its etiology due to its high morbidity and mortality globally. The use of appropriate antibiotic use is equally vital in the management in addition to surgery. Understanding the bacterial pathogens and the susceptible pattern will ensure rational antimicrobial therapy according to the bacteriology and drug susceptibility profiles, guide clinicians to establish a protocol and reduce the risk of the evolution of drug-resistant bacteria. Currently third generation cephalosporins are mostly in

use and the overall proportion of gram-negative bacteria resistant to them was 35.9% (10). This study aimed at identifying the common pathogens and their susceptibility patterns among patients with peritonitis at KCMC.

Methods

Design, setting and participants

This was a descriptive cross-sectional study conducted from October 2019 to April 2020 at Kilimanjaro Christian Medical Centre (KCMC). KCMC is a referral hospital in the Northern part of Tanzania, in Kilimanjaro Region with a catchment population of about 20 million from the northern Tanzania and nearby countries. Being a tertiary facility, KCMC provides emergency surgical services ranging from neurotrauma, orthopedics and trauma, gynecological emergencies and emergency general surgery including pediatric cases. Cases of peritonitis are managed acutely from the emergency department throughout post operative period. Once clinical and/or radiological diagnosis is made, patients are stabilized and prepared for emergency surgery. A number of cases are managed post operatively in the surgical intensive care unit depending on their clinical parameters while most are nursed in the surgical ward until discharge.

All patients who were diagnosed with peritonitis and underwent surgery in the general surgical ward during the study period were enrolled in the study. Patients who had a laparotomy at a peripheral health facility and were re-operated at KCMC were excluded from the study.

Data collection procedures

The socio-demographic and clinical data of all participants was collected using a data collection sheet. Additional clinical data were extracted from patients' medical records. Baseline investigations including complete blood count, blood grouping & crossmatch, serum electrolytes, serum urea and creatinine, abdominal X-ray, and ultrasonography were performed upon the patient's arrival at the emergency department or department of general surgery, and managed accordingly if any abnormalities were found. Follow-up control investigations including complete blood count, serum electrolytes, serum protein levels, urine dipstick were carried out on the third day post-operatively to all study participants.

A thorough examination to identify clinical features of chest infection and surgical site infection was conducted daily, documented and managed where necessary. Patients having clinical features of chest infection, surgical site infection (SSI), or sepsis, additional tests such as chest X-ray, pus swab and blood specimen for culture and antimicrobial susceptibility testing were performed respectively.

Patient follow-up was conducted in the surgical wards and/or surgical intensive care units until death or discharge. The seven-day outcome was monitored at surgical outpatient units or via telephone if the patient was discharged before seven days post-operation.

Sample collection, Transport, and Processing

Under strict aseptic conditions, peritoneal swabs were collected intra-operatively using sterile cotton swab sticks and transported to the clinical laboratory using Stuart's transport media. Data were analyzed using Microsoft Excel and SPSS® (22.0; SPSS Inc.,) and reported in descriptive charts. The susceptibility was calculated by adding up the results of the columns adjusted by the absolute number in the column.

Culture

The following culture media were used: 5% blood agar, chocolate agar, and Mac Conkey agar. All inoculated plates were incubated in aerobic conditions and were examined for growth at 24 hours, 48 hours, and 5 days then discarded. A significant bacterium was considered if culture yield ≤ 10^5 colony-forming unit (CFU/ml).

Antimicrobial susceptibility testing

The antimicrobial agents tested included Gentamycin (10µg), Amikacin, Piperacillin, Amoxicillin & Clavulanic acid (10µg), Meropenem, Ciprofloxacin (5µg), Ampicillin, Ceftriaxone (30µg), Trimethoprime & Sulphamethoxazole and Ceftazidime. The Kirby-Bauer method was used for antimicrobial susceptibility testing on Müller-Hinton Agar, and the results were interpreted according to Clinical and Laboratory Standards Institute (CLSI) (11). Media and discs were tested for quality control using standard stains. ISL regulations were maintained and adhered to. Reference strains of *E. coli* ATCC 25922 was used for quality control for antimicrobial susceptibility tests.

Results

Baseline characteristics

The median age of participants at enrolment was 32 years (Interquartile Range (IQR): 20-48 years). Slightly more than half (51.4%) of the patients were males. The mean (standard deviation (SD)) pre-operative hemoglobin level of the participants was 11.3g/dl (±2.1). The majority (42.9%) of the participants were ASA-1, and 31.4% presented two days after their first symptoms. No patient was admitted to the intensive care unit (ICU) before the operation, but 45.7% needed ICU admission postoperatively. The duration of surgery ranged from 100

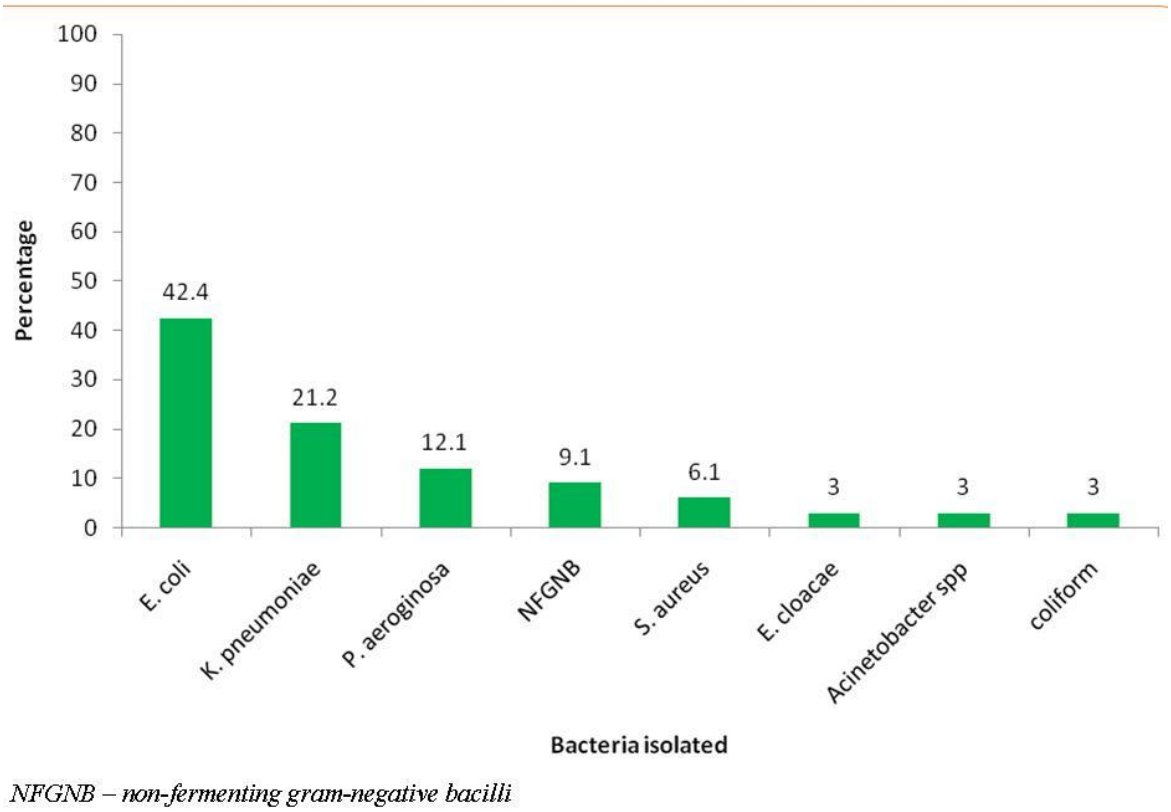
to 150 minutes. More than half of the patients (60%) developed complications post-surgery. The average (±SD) hospital stay was 5.6 (±2.7) days. Table 1 shows the baseline characteristics of the study participants.

Table 1: Baseline characteristics of study participants (N=35)

Characteristics	n	%
Age category (years)		
0 – 18	6	17.1
19 – 49	21	60.0
50 and above	8	22.9
Median age (IQR)	*32 (20-48)*	
Sex		
Male	18	51.4
Female	17	48.6
ASA classification		
1	15	42.9
2	11	31.4
3	5	14.3
4 or above	4	11.4
Pre-operative haemoglobin level (g/dl)		
10.9 or less	13	37.1
More than 10.9	22	62.9
Mean haemoglobin (±SD)	*11.3 (±2.1)*	
Duration of illness (days)		
1	8	22.9
2	11	31.4
3	9	25.7
4	7	20.0
Mean duration (±SD)	*2.4 (±1.1)*	
Duration of surgery (minutes)		
Less than 120	11	31.4
120 or more	24	68.6
Median duration (IQR)	*120 (100-150)*	
Post-operative complications		
Yes	21	60.0
No	14	40.0

Isolation of pathogens

Figure 1 shows the species of bacteria isolated. A total of 33 pathogenic bacteria were isolated from 27 positive cultures. Six samples did not grow, and two simultaneous bacterial growth in six participants occurred. *Escherichia coli* (42.4%) *Klebsiella peumoniae* (21.2%) were the most common intra-abdominal pathogens isolated from abdominal peritoneal swabs.



NFGNB – non-fermenting gram-negative bacilli

Figure 1. Species of bacteria isolated from peritoneal swabs in patients with peritonitis (N=33)

Antimicrobial susceptibility pattern

Coliforms and *Enterobacter cloacae* demonstrated 100% susceptibility to Amikacin and Gentamycin, while *E. coli* showed 100% susceptibility to Amikacin and Meropenem. *Acinetobacter spp.* also showed good susceptibility to Amikacin. Despite Ceftriaxone being prescribed to most patients, susceptibility testing revealed that it had the poorest effectiveness against the isolated organisms. Table 2 below details these findings.

Table 2: Antimicrobial susceptibility pattern for peritoneal isolates

Antibiotics	Bacterial isolates (%)							
	Escherichia coli	***Enterobacter cloacae***	***Kliebsiella pneumoniae***	**NFGNB***	***Pseudomonas aeroginosa***	***Staphylococcus aureus***	***Acinetobacter spp.***	**Coliform**
Gentamycin	72.7	100	57.1	50.0	66.0	100	0	100
Amikacin	100	100	100	100	66.0	-	100	100
Piperacillin	0	0	100	-	100	-	0	100
Amoxicillin & clavulanic acid	62.5	0	33.0	33.0	0	-	0	-
Meropenem	100	-	100	-	50.0	-	0	-
Ciprofloxacin	0	-	60.0	-	66.0	0	0	100
Ampicillin	20	0	0	0	-	-	0	0
Ceftriaxone	22.2	0	20.0	0	0	-	0	0
Trimethoprim & sulphamethoxazole	33.0	-	100	0	-	-	0	0
Ceftazidime	0	-	0	100	50.0	-	-	-

**NFGNB – non-fermenting gram-negative bacilli*

Discussion

This study aimed at identifying the microorganisms present in cases of peritonitis and their antimicrobial susceptibility pattern to antibiotics commonly used in empiric treatment in order to evaluate and recommend the most appropriate treatment options. *Escherichia coli* and *Klebsiella pneumoniae* were the most common intra-abdominal pathogens isolated from peritoneal swabs of patients with peritonitis and were found to be less susceptible to Ceftriaxone. This indicates a need to revise empirical antimicrobial guidelines to improve treatment efficacy.

Studies by Sartelli et al. and Sahani et al. also identified *E. coli* and *K. pneumoniae* as the predominant gram-negative microorganisms in cases of peritonitis, with *S. aureus* and *Enterobacter spp*. being the least isolated microorganism (12,13). A study by Nithya et al., and Seni et al., found that, *E. coli* and *K. pneumoniae* were the most common organisms (14,15). This aligns with a study by Kuftinec et al., whereby *E. coli*, Klebsiella species, and Enterobacter species were identified as the common bacteria causing both spontaneous and secondary peritonitis. The authors emphasized that if secondary bacterial peritonitis is diagnosed, prompt surgical intervention is crucial for improving survival (16). However, in our study, all participants underwent emergency laparotomy, underscoring the importance of timely surgical intervention in managing secondary peritonitis.

Antibiotic resistance is a rising health threat in developing countries due to various reasons, hence imposing a socio-economic burden. In recent years, there has been a rapid emergence of resistant pathogens, leading to treatment failure due to multi-resistant bacteria (8). Few new antimicrobial agents are coming up while other drugs are being withdrawn from the market due to expanding antimicrobial resistance. In secondary peritonitis the rate of resistant organisms at initial operation is 30% (17).

In the study by Sahani et al., the authors highlighted the superior efficacy of Meropenem compared to Cephalosporins combined with Salbactam (13). Given the economic constraints and the need to preserve the efficacy of certain antibiotics, the preference for using Cephalosporins, Gentamycin, and Ciprofloxacin over Meropenem can be justified. This approach helps to reserve Meropenem and Amikacin for more severe cases or when resistance to other antibiotics is encountered.

Similarly, the gram-negative organisms isolated in both the current study and Nithya's study showed high susceptibility of *E. coli* and *K. pneumoniae* to Imipenem. However, authors reported poor susceptibility of *E. coli* to Ampicillin and Amoxicillin-clavulanic acid contrary to our study findings showing a 62.5% susceptible to Amoxicillin-clavulanic acid (14). Higher

susceptibility to Amoxicillin-clavulanic acid observed in our study should not create a false sense of security, as the high levels of interaction between the two geographical areas suggest that it is only a matter of time before resistance increases here as well. Both studies demonstrated high susceptibility to meropenem and gentamicin, supporting their continued use.

Five out of six WHO regions had more than 50% resistance to third generation cephalosporins (E.g. Ceftriaxone) of *E. coli* and *K. pneumoniae* along with others in hospital settings. 45% of deaths across Africa and South-East Asia were attributed to multi-drug resistance. Furthermore, 77% of deaths in Africa were associated with *K. pneumoniae* resistant to third-generation cephalosporins. In addition to that, a recent WHO report states that it is estimated that 10 million deaths will be accountable to antimicrobial resistance by 2050 (8).

Antimicrobial resistance is growing in developing countries at an alarming rate due to inappropriate use of drugs, availability over-the-counter and unregulated supply, self-prescription and poor compliance which has also led to drug resistance. There is a need of regular community-based antimicrobial surveillance data because resistance rates can vary in one region of a country over time. This is useful to health care professionals in particular communities or regions to treat infections with specific susceptible antimicrobials and such surveillance needs to be conducted regularly and continuously (18).

Empirical antimicrobial therapy should be supported based on local epidemiology and antimicrobial stewardship, clinical severity of infection and source of infection. The treatment should also cover for less common organisms along with the habitual organisms (3). In our study, *E. coli* and *K. pneumoniae* were the commonest isolates hence treatment should include Gentamycin and Meropenem and Amikacin and Piperacillin base can be used for cases with severe forms of sepsis.

Conclusion and recommendations

The most common bacteria isolated in patients with secondary peritonitis were gram-negative *Escherichia coli* and *Klebsiella pneumoniae,* less susceptible to Ceftriaxone. This study suggests that the current local regime needs to be reassessed due to the high number of gram-negative bacteria resistant to third generation cephalosporins which is the local first-line treatment. In addition to early source control, appropriate empiric treatment is crucial for the effective management of peritonitis.

A proper and adequate understanding is necessary for policy making locally, hence continuous surveillance and antimicrobial stewardship are urgently required to curb rising

resistance and improve peritonitis outcomes in Tanzania. Without effective antimicrobials, various medical procedures such as surgery, invasive diagnostic and treatment procedures, and transplantation medicine will be severely hindered with the corresponding increase in morbidity and mortality from secondary microbial infections.

Further research is needed to further analyze specific microorganisms such as extended spectrum beta-lactamases (ESBL), anaerobes and fungi with a broader range of antibiotics for testing. Multicentred national level research is also recommended to perform molecular characterization so that evidence-based data can be utilized to improve infection prevention control (IPC) in the country. Furthermore, there is a need to use newer generation antimicrobial agents and newer combination agents as well as with beta-lactamase inhibitors.

Study Limitations

Anaerobic organisms and fungi were not tested due to the lack of proper facilities, which may have led to an underestimation of the bacteria involved in secondary peritonitis. Additionally, the limited time frame and the COVID-19 pandemic limited recruitment, resulting in a smaller sample size and reduced statistical power, limiting the scope for advanced statistical analysis. This constraint also restricted the research to a few variables, and important co-morbidities such as diabetes mellitus, Human Immunodeficiency Virus (HIV) infection, and hypertension were not systematically recorded.

Ethical Approval and Consent to Participate

Ethical approval was obtained from the Kilimanjaro Christian Medical University Research Ethical Committee (PG 024/2019), and permission was obtained from the Executive Director of Kilimanjaro Christian Medical Centre. Written informed consent was obtained from all participants enrolled in this study and for publication. Informed consent was obtained from the parents/ legally authorized representatives of subjects that are under the age of 18. All methods were carried out in accordance with relevant guidelines and regulations.

Abbreviations

ASA	American Society of Anesthesiologists
ATCC	American Type Culture Collection
CFU	Colony Forming Unit
CLSI	Clinical and Laboratory Standards Institute
COVID-19	Coronavirus Disease 2019

ESBL	Extended Spectrum Beta-Lactamase
GIT	Gastrointestinal Tract
HIV	Human Immunodeficiency Virus
ICU	Intensive Care Unit
IPC	Infection Prevention and Control
IQR	Interquartile Range
ISL	International Standards Laboratory
KCMC	Kilimanjaro Christian Medical Centre
SD	Standard Deviation
SPSS	Statistical Package for the Social Sciences
SSI	Surgical Site Infection
WHO	World Health Organization

Declarations

Data Availability

Due to ongoing analyses, the supporting data are available from the corresponding author on a reasonable request.

Conflicts of Interest

The authors declare that they have no competing interests.

Authors' Contributions

JL and DM designed the study and collected the data. JL and DJD analyzed the data and drafted the manuscript. JL, DJD, DM, RP and KC contributed to the final draft of the manuscript. All authors gave their final approval to the manuscript.

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