MDPI

*Article*

# High Fecal Carriage of Multidrug Resistant Bacteria in the Community among Children in Northwestern Tanzania

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**Abstract:** Colonization of multidrug resistant (MDR) bacteria is associated with subsequent invasive infections in children with comorbidities. This study aimed to determine the resistance profile and factors associated with MDR pathogen colonization among HIV− and HIV+ children below five years of age in Mwanza, Tanzania. A total of 399 (HIV− 255 and HIV+ 144) children were enrolled and investigated for the presence of MDR bacteria. The median [IQR] age of children was 19 (10–36) months. Out of 27 *Staphylococcus aureus* colonizing the nasal cavity, 16 (59.5%) were methicillin resistant while 132/278 (47.2%) of Enterobacteriaceae from rectal swabs were resistant to third generation cephalosporins, with 69.7% (92/132) exhibiting extended spectrum beta lactamase (ESBL) phenotypes. The proportion of resistance to gentamicin, amoxicillin/clavulanic acid and meropenem were significantly higher among HIV+ than HIV− children. A history of antibiotic use in the last month OR 2.62 [1.1, 6.9] ($p = 0.04$) and history of a relative admitted from the same household in the past three months OR 3.73 [1.1, 13.2] ($p = 0.03$) independently predicted ESBL rectal colonization. HIV+ children had significantly more fecal carriage of isolates resistant to uncommonly used antibiotics. There is a need to strengthen antimicrobial stewardship and Infection Prevention and Control (IPC) programs to prevent the emergence and spread of MDR pathogens in children.

**Keywords:** children; MDR/ESBL colonization; human immunodeficiency virus

## 1. Introduction

Antimicrobial resistance (AMR) is a global public health threat; it is estimated that it will kill about 10 million people annually by year 2050 [1]. AMR affects all countries, but the burden is disproportionately higher in low- and middle-income countries (LMIC) than in high-income countries (HIC) [2]. In many LMIC, a high burden of infectious diseases, poverty, weak governance and health systems, and low awareness remain major challenges in the fight against AMR [3,4].

Infection with AMR leads to serious illnesses, treatment failure, prolonged hospital stays and increased healthcare costs [5,6]. LMICs bear the consequences of AMR owing to the high cost and poor access to effective antibiotics to treat MDR infections [7]. MDR colonization is a risk factor for subsequent MDR infections [8–10] due to the fact that these MDR pathogens may serve as endogenous reservoirs for overt clinical infections. Overt subsequent clinical infections following colonization are more common in critically ill patients with comorbidities such as HIV, Diabetes Mellitus and Sickle Cell Disease [9,11].

HIV infection has been reported to be a risk factor for colonization with MDR pathogens [12]; this has partly been attributed to trimethoprim/sulfamethoxazole prophylaxis that is routinely given to prevent opportunistic infections [13,14]. The high prevalence of HIV and the emergence of MDR complicate efforts to control bacterial infections in many LMIC settings.

Evidence with regards to AMR is limited; however, it is known that HIV infected children below five years of age are particularly at risk of colonization with methicillin resistant *Staphylococcus aureus* (MRSA) and extended spectrum beta-lactamase producing Enterobacteriaceae (ESBL-PE) [15–17]. Therefore, this study aimed to determine the resistance profile and factors associated with MDR pathogen colonization among HIV− and HIV+ children below five years of age in Mwanza, Tanzania.

## 2. Results

### 2.1. Socio-Demographic and Clinical Characteristics of Enrolled Children

A total of 399 children below five years of age were enrolled with a median [interquartile range [IQR] age of 19 [10–35] months and 212 (53%) were males. The gender distribution was almost equal between two groups ($p$ = 0.463). Antibiotic uses in the past one month (excluding SXT) was 89% in HIV− group and 90.3 in HIV+ group, $p$ = 0.694 (Table 1). Among the enrolled HIV+ children, 143 (99.3%) were on ART.

**Table 1.** Socio-demographic and clinical characteristics of enrolled participants.

| Characteristics | | HIV− (N = 255) | | HIV+ (N = 144) | | $p$-Value |
|---|---|---|---|---|---|---|
| | | Frequency (n)/ Median [IQR] | Percentages (%) | Frequency (n)/ Median [IQR] | Percentages (%) | |
| Median [IQR] age in months | | 14 (9–22) | | 34 (22–44) | | <0.001 ** |
| Gender | Male | 139 | 44.5 | 73 | 49.3 | |
| | Female | 116 | 45.5 | 71 | 50.7 | 0.463 |
| Residence | Urban | 250 | 98.0 | 130 | 90.3 | |
| | Rural | 5 | 2 | 14 | 9.7 | <0.001 ** |
| Immunization schedule | Completed | 203 | 79.6 | 139 | 96.5 | |
| | Not Completed | 52 | 20.4 | 5 | 3.5 | <0.001 ** |
| Source of water | Tap water | 241 | 94.5 | 122 | 84.7 | |
| | Pond/lake water | 14 | 5.5 | 22 | 15.3 | 0.001 ** |
| Type of toilet | Modern toilet | 244 | 95.7 | 120 | 83.3 | |
| | Pit latrine | 11 | 4.3 | 24 | 16.7 | <0.001 ** |
| Family size | Median | 4 (3–5) | | 5 (3.5–6) | | 0.068 |
| Care taker leve of edication | Primary | 168 | 65.9 | 111 | 77.1 | |
| | Secondary | 87 | 34.1 | 33 | 22.9 | 0.019 |
| Type of house | Brick | 226 | 88.6 | 108 | 75 | |
| | Mud | 29 | 11.4 | 36 | 25 | <0.001 ** |
| Animal keeping | Yes | 55 | 21.6 | 30 | 20.8 | |
| | No | 200 | 78.4 | 114 | 79.2 | 0.863 |

**Table 1.** *Cont.*

| Characteristics | | HIV− (N = 255) | | HIV+ (N = 144) | | *p*-Value |
|---|---|---|---|---|---|---|
| | | Frequency (n)/ Median [IQR] | Percentages (%) | Frequency (n)/ Median [IQR] | Percentages (%) | |
| Duration of breastfeeding | Median [IQR] months | 6 (5–6) | | 6 (6–6) | | 0.002 ** |
| Use of antimalaria in the last month | Yes | 26 | 10.2 | 8 | 5.6 | |
| | No | 229 | 89.2 | 136 | 94.4 | 0.111 |
| Relative hospital admission in same household past 3 months | Yes | 7 | 2.7 | 5 | 3.5 | |
| | No | 248 | 97.3 | 139 | 96.5 | 0.683 |
| Antibiotics use over the past one month | Yes | 227 | 89.0 | 130 | 90.3 | |
| | No | 28 | 11.0 | 14 | 9.7 | 0.694 |
| ART us | Yes | NA | NA | 143 | 99.7 | |
| | No | NA | NA | 1 | 0.3 | NA |
| MUAC | SAM | 34 | 13.3 | 3 | 2.1 | |
| | MAM | 221 | 86.7 | 141 | 97.9 | <0.001 ** |

Key: MUAC = Mid Upper Arm Circumference, SAM = Severe Acute Malnutrition, MAM = Moderate Acute Malnutrition, NA = not applicable, ** Significant *p* < 0.01.

### 2.2. Isolated Bacteria Pathogens from Nasal, Oral and Rectal Swabs

Of 399 enrolled children, 27 (6.8%) had nasopharyngeal colonization with *Staphylococcus aureus*. Of them, 16 (59.5%) were found to be MRSA. No significant difference of MRSA colonization was observed among HIV− children and HIV+ (13, 61.9% vs. 3, 50.0%; *p* = 0.662). The predominant pathogen colonizing the oral cavity was *K. pneumoniae* 11 (40.7%), followed by *E. coli* 5 (18.5%) and *S. pneumoniae* 4 (14.8%). Of 278 (69.7%) children screened for colonization of ESBL-PE, 92 (33.1%) were found to be colonized. The predominant isolates were *E. coli* 177 (63.7%) and *K. pneumoniae* complex 29 (10.4%) (Table 2).

**Table 2.** Isolated pathogens from different sources among 399 participants by HIV status.

| Isolate | Oral (N = 27) | | Nasal (27) | | Rectal (N = 278) | |
|---|---|---|---|---|---|---|
| | HIV− | HIV+ | HIV− | HIV+ | HIV− | HIV+ |
| *K. pneumoniae complex* | 3 | 8 | | NA | 12 | 17 |
| *E. coli* | 4 | 1 | | NA | 136 | 41 |
| *S. aureus* | | NA | 21 | 6 | NA | |
| MRSA | | NA | 13 | 3 | NA | |
| *S. pneumoniae* | 2 | 2 | | NA | NA | |
| Others | 3 * | 4 * | | NA | 45 ** | 27 ** |
| Total | 12 | 15 | 21 | 6 | 193 | 85 |

NA = Not Applicable; MRSA = methicillin resistant *Staphylococcus aureus*. * Others Oral; *A. hydrophila* (n = 1), *Acinetobacter* spp. (n = 1), *C. freundii* (n = 1), *S. pyogenes* (n = 1), *Pantoea agglomerans* (n = 1), Unidentified Gram-negative rods (n = 2). ** Others rectal; *A. hydrophila* (n = 17), *Citrobacter* spp. (n = 15), *E. cloacae* complex (n = 15), *Enterobacter* spp. (n = 1), *Pantoea agglomerans* (n = 5), Unidentified Gram-negative rods (n = 19).

### 2.3. Patterns of Resistance by Source of Bacteria

Gram negative bacteria from rectal isolates were significantly more resistant to ciprofloxacin, amoxicillin/clavulanic acid and ceftriaxone than Gram negative bacteria from oral cavity (48% vs. 16.7%, *p* = 0.007; 48.5% vs. 22.2%. *p* = 0.025; and 47.5% vs. 16.7% *p* = 0.009), respectively. Gram negative enteric pathogens from rectal isolates exhibited higher ESBL phenotype than those from the oral cavity (33.1% vs. 11.1% *p* = 0.039), (Table 3).

**Table 3.** Percentage resistance and ESBL phenotypes of Gram-negative bacteria isolated from oral and rectal swabs.

| Antibiotic | ORAL SWAB N = 18 (%) | RECTAL SWAB N = 278 (%) | *p* Value |
|---|---|---|---|
| Amoxicillin/clavulanic acid | 4 (22.2%) | 135 (48.7%) | 0.025 * |
| Ceftriaxone | 3 (16.7%) | 132 (47.5%) | 0.009 * |
| Sulfamethoxazole/Trimethoprim | 15 (83.3%) | 233 (83.8%) | 0.583 ** |
| Tetracycline | 10 (55.6%) | 186 (66.9%) | 0.230 ** |
| Gentamicin | 2 (11.1%) | 40 (14.4%) | 0.516 ** |
| Ciprofloxacin | 3 (16.7%) | 135 (48.6%) | 0.007 * |
| Ceftazidime | 2 (11.1%) | 103 (25.8%) | 0.019 * |
| Meropenem | 2 (11.1%) | 20 (7.2%) | 0.394 ** |
| ESBL | 2 (11.1%) | 92 (33.1%) | 0.039 * |

* Significant, ** Not significant.

*2.4. Comparison of Percentage Resistance of Gram-Negative Bacteria Colonizing Gastrointestinal Tract (GIT) of HIV Infected and Non-HIV Infected Children*

Out of 278 isolates from GIT, isolates from HIV+ children had high resistance to gentamicin (28.2% vs. 8.3%, $p < 0.001$), amoxicillin/clavulanic acid (62.4% vs. 42.5%, $p = 0.002$) and meropenem (16.5% vs. 3.1%, $p < 0.001$) compared to those isolates from HIV− children. Although not statistically significant, HIV+ children were more colonized with ESBL-PE than HIV− children (42.4% vs. 30.6%, $p = 0.056$), (Table 4).

**Table 4.** The resistance patterns of GIT isolates in HIV and non-HIV infected children (N = 278).

| Antibiotic Agents | HIV− (n = 193) | HIV+ (n = 85) | *p* Value | Overall Resistance |
|---|---|---|---|---|
| Sulfamethoxazole/Trimethoprim | 158 (81.9%) | 75 (88.2%) | 0.184 | 233 (83.8%) |
| Tetracycline | 125 (64.8%) | 61 (71.8%) | 0.253 | 186 (66.9%) |
| Ciprofloxacin | 92 (47.7%) | 43 (50.6%) | 0.654 | 135 (48.6%) |
| Gentamicin | 16 (8.3%) | 24 (28.2%) | 0.000 ** | 40 (14.4%) |
| Ceftriaxone | 87 (45.1%) | 45 (52.9%) | 0.226 | 132 (47.5%) |
| Amoxicillin/clavulanic acid | 82 (42.5%) | 53 (62.4%) | 0.002 ** | 135 (48.6%) |
| Ceftazidime | 66 (34.2%) | 37 (43.5%) | 0.138 | 103 (37.1%) |
| Meropenem | 6 (3.1%) | 14 (16.5%) | 0.000 ** | 20 (7.2%) |
| ESBL | 59 (30.6%) | 36 (42.4%) | 0.056 | 95 (34.2%) |

** Significant $p < 0.01$.

*2.5. Multidrug Resistance Patterns among HIV and No HIV Infected Children below Five Years of Age*

Among 278 GIT isolates, HIV+ children had significantly higher multidrug resistance to SXT/TET/CN (22.4% vs. 6.2% $p = 0.000$), CRO/CIP/CN (16.5% vs. 6.7% $p = 0.012$), TET/CN/CRO (17.7% vs. 5.2% $p = 0.001$), CN/TET/CIP (18.8% vs. 5.2% $p = 0.0001$), and SXT/TET/CN (22.4% vs. 6.2%, $p < 0.001$) (Supplementary File S1).

*2.6. Factors Associated with ESBL Colonization among HIV− and HIV+ Children below Five Years of Age*

On univariate logistic regression analysis, a history of antibiotic use in the last month (3.45, 95% CI: 1.39–8.58, $p = 0.007$) and a relative admitted from same household in the past three months (OR 4.8, 95% CI: 1.47–15.36, $p = 0.009$) were significantly associated with ESBL producing enteric bacteria GIT colonization in children below five years of age.

No significant difference was observed regarding ESBL colonization among HIV− and HIV+ children (25.0% vs. 23.14%); however, we found the proportion of ESBL-producing isolates to be higher in HIV+ children than in HIV− children, although this did not reach

statistical significance (42.4% vs. 30.6%, $p$ = 0.056). On multivariable logistic regression analysis, antibiotic use in the last month (OR 2.62, 95% CI: (1.1–6.9), $p$ = 0.04) and history of a relative admitted from the same household in the past three months (OR 3.73, 95% CI (1.1, 13.2), $p$ = 0.03) remained significant factors associated with ESBL producing enteric Gram-negative GIT colonization in children below five years of age (Table 5). The effect of trimethoprim/sulfamethoxazole (TSX) prophylaxis on colonization with EBSL could not be assessed as all HIV+ children were on TSX prophylaxis.

**Table 5.** Factors associated with ESBL rectal colonization (N = 399).

| Variable (N) | ESBL Colonization (n, %) | Univariate OR (95%CI) | $p$ Value | Multivariable OR (95%CI) | $p$ Value |
|---|---|---|---|---|---|
| **A: Child factors** | | | | | |
| Age (months) | * 19, IQR (10–29) | 0.98 (0.97, 1.00) | 0.154 | 0.98 (0.96, 1.00) | 0.091 |
| Sex | | | | | |
| Male (212) | 56 (26.4) | 1 | | | |
| Female (187) | 39 (20.9) | 1.36 (0.85, 2.17) | 0.194 | | |
| HIV status | | | | | |
| Negative (255) | 59(23.1) | 1 | | | |
| Positive (144) | 36(25.0) | 1.11 (0.69, 1.78) | 0.675 | 1.43 (0.81–2.52) | 0.212 |
| Residence | | | | | |
| Urban (380) | 89(23.4) | 1 | | | |
| Rural (19) | 6(31.6) | 1.51 (0.56, 4.09) | 0.418 | | |
| Antibiotics usage last month | | | | | |
| No (357) | 79 (22.1) | 1 | | | |
| Yes (42) | 16 (38.1) | 3.45 (1.39, 8.58) | 0.007 | 2.62 (1.1, 6.9) | 0.04 |
| **B: Family factors** | | | | | |
| Caretaker Education level | | | | | |
| Secondary (120) | 30 (25.0) | 1 | | | |
| Primary (279) | 65 (23.3) | 0.91 (0.55, 1.50) | 0.714 | | |
| Relative Admitted | | | | | |
| No (387) | 88 (22.7) | 1 | | | |
| Yes (12) | 7 (58.3) | 4.8 (1.47, 15.36) | 0.009 | 3.7 (1.1, 13.2) | 0.03 |

* Median age of children colonized with ESBL-PE.

## 3. Discussion

This study reports the burden and associated factors of nasopharyngeal, oral, and rectal colonization of MDR pathogens in HIV− and HIV+ children below five years of age. About 4% of the enrolled children had nasopharyngeal colonization with MRSA, and 33.1% (92/278) were orally and GIT colonized by ESBL-PE, with no significant differences between HIV− and HIV+ children.

This study has shown a MRSA prevalence of 4%, similar with the values reported in China (3.9%) [18] and Taiwan (3.5–3.8%) [19], but lower than those reported in India (16.5–23.5) [19]. The low prevalence in the current study could be due to improvements in Infection Prevention and Control (IPC) procedures. It should be noted that the current study was conducted during the midst of the coronavirus disease 2019 (COVID-19) pandemic, where IPC, especially hand hygiene, was largely strengthened. In most developed countries such as Switzerland, the very low prevalence of MRSA colonization is largely related to routine screening, stringent IPC procedures and antimicrobial stewardship measures. The

literature has documented that MRSA colonization occurs in individuals who have frequent exposure to healthcare settings, and in those with frequent antibiotic usage as well as those with immunodeficiencies [20,21].

In contrast to the low prevalence of MRSA colonization, the current study observed that almost two-thirds of children were colonized with potential ESBL-PE with a predominance of *E. coli* and *K. pneumoniae* complex. Similar findings regarding the colonization of ESBL producing *E. coli* and *K. pneumoniae* complex were reported in the same region among admitted neonates and street children [8,22], as well as other settings in LMIC [23–27] among non-HIV infected children. However, the prevalence was twice as high as that reported in HIC among non-HIV infected children [28], which could be attributed to poor hygiene, infrastructure and water sanitation, as documented previously in a meta-analysis [29]. Moreover, the misuse of antimicrobials is common in LMICs; it has been previously found to be associated with ESBL-PE carriage [2], as was confirmed in this study.

In this study we found that history of recent antibiotic use in the last month and a relative admitted living from same household in the last one month independently predicted ESBL producing enteric bacteria colonization. These findings are in line with studies done on admitted children in HIC, which documented that colonization was found to increase with a hospitalized family member in the three previous months [30,31]. Previous studies observed that children who had recent antibiotic usage had three times the odds of rectal colonization of ESBL-PE as opposed to those who had not been on antibiotics [24,32]. A family member with a history of hospital admission could carry and transmit a resistant pathogen to other family members due to poor hygiene and sanitation. One review documented that antibiotics commonly used in primary health care influence the composition of the gastrointestinal microbiota [33], increasing selection pressure on MDR pathogens. This process may be further complicated by poor IPC practices and hygiene which favor the spread of resistant bacteria in both the community and healthcare settings. Therefore, exposure to antibiotics and hospital admission increase the risk of becoming colonized with ESBL-GNB.

### Limitations

A correlation between colonization of ESBL and MRSA strains and subsequent occurrence of invasive infections was established in the current study. In addition, due to the fact that participants were children up to 5 years of age and most of information was retrieved from the parents, there is a possibility of recall bias, especially on the data regarding breastfeeding. We only collected data on overall duration of breastfeeding rather than duration of exclusive breastfeeding in order to mitigate this bias.

## 4. Materials and Methods

### 4.1. Study Design, Duration and Study Area

This cross-sectional study was conducted from April to December 2020 at Baylor Children's Hospital and Nyamagana District hospital in Mwanza Tanzania. The study was designed to compare the burden and characteristics between HIV− and HIV+ children. Baylor Children's Hospital is a patient-centered, pediatric HIV prevention and treatment clinic which serves the districts of Mwanza regions and other nearby regions. The clinic attends an average of 100 children per week. Nyamagana District Hospital runs a Reproductive and Child Health (RCH) clinic daily, attending approximately 30 children per day.

### 4.2. Study Population and Inclusion Criteria

All children aged six months to five years of age attending HIV outpatient clinic at Baylor Children's Hospital and the Reproductive and Child Health Center at Nyamagana District Hospital were enrolled. The study included all children with no clinical signs and symptoms of systemic infection such as fever, chest wall indrawing, difficulty in breathing, crying during micturition, and diarrhea among other symptoms [34]. Children who had

no documentation of HIV status on their RCH card and who failed to provide sufficient samples were excluded.

### 4.3. Sample Size Estimation, Sampling Technique

The study aimed at enrolling 125 children per group using a two independent proportions formula [35]. We used 16.9% as proportion 1 from the Tellevik study [24] and aimed to observe a 25% minimum difference; therefore, proportion 2 in the formula was assumed to be 42% with a beta level of 90% and an alpha level of 5%, giving a minimum sample size of 68 participants per group.

### 4.4. Data Collection and Sampling Procedures

A convenience sampling method was used to select the clinics based on proximity to a reference laboratory at Bugando Medical Centre. Children attending these clinics were enrolled serially until the sample size was reached. A data collection tool was customized in epicollect5 and used to collect socio-demographic characteristics (age, sex, socio-economic, household size etc.) and clinical data (duration on HIV treatment, history of antibiotics usage, vaccination history, vitals and anthropometric measurements and other comorbidities).

Anthropometric parameters which included height and weight were measured using a measuring tape and mechanical weight scale (SECA, Benson Avenue, New York, NY, USA) while the ages of the children were obtained from mothers/caregivers. In every enrolled child the Mid Upper Arm Circumference (MUAC) was measured and interpreted accordingly. Acute malnutrition is divided into severe acute malnutrition (SAM), being defined as a weight-for-height Z-score (WHZ) $<-3$ or a mid-upper arm circumference of <11.5 cm, or moderate acute malnutrition (MAM), with a WHZ between $-2$ and $-3$ Z-scores or a MUAC between 11.5 and 12.5 cm [36].

### 4.5. Sample Collection and Laboratory Procedures

4.5.1. Samples Collection

Specimens (oral swab, nasal pharyngeal swab and stool/rectal swabs) were collected from all enrolled children by trained medical personnel. Sterile swabs (Mast Diagnostica GmbH, Reinfeld, Germany) in Amies transport media were used to collect a single time swab from every participant. Samples were then transported to the Microbiology Laboratory of the Catholic University of Health and Allied Sciences (CUHAS) within four hours of collection for laboratory analysis.

4.5.2. Screening of MDR Bacteria

Stool/rectal swab samples were directly inoculated on MacConkey agar (MCA; HiMedia, Mumbai, India) supplemented with cefotaxime 2 µg/mL (MCA-C) for screening of extended spectrum β-lactamase producing Gram-negative bacteria (ESBL-GNB). Nasopharyngeal swabs were directly inoculated on 5% sheep blood agar (BA; HiMedia, Mumbai, India) for isolation of *S. aureus* while oral swabs were directly inoculated on BA and MCA plates for the isolation of bacteria pathogens. Plates were incubated aerobically at 37 °C for 18–24 h. Gram negative bacteria (GNB) isolated from oral swabs were further sub-cultured on MCA-C for screening of potential ESBL-PE. In-house prepared biochemical identification tests (Triple sugar Iron test [TSI] for sugar fermentation, $CO_2$ and $H_2S$ production; Sulphur, Indole and Motility [SIM] for $H_2S$ and indole production and motility; Simmons citrate agar for utilization of citrate as the sole source of carbon; oxidase test for production of oxidase enzyme; and Christensen's urea agar for urease enzyme production) were used for identification of GNB. For identification and confirmation of *S. aureus* (golden yellowish with β-hemolysis on BA plates), catalase, coagulase tests and DNAse agar were used. All procedures were performed as per standard operating procedures (SOPs) as previously documented [37].

### 4.5.3. Antibiotics Susceptibility Testing

The disc diffusion technique, as documented by Kirby-Bauer, was used for antibiotics susceptibility testing (AST). Bacteria were suspended in sterile saline and turbidity were adjusted to 0.5 McFarland standard, then swabbed on Mueller Hinton agar (HI Media, New Delhi, India) plates. Antibiotics discs of ciprofloxacin (CIP 5 μg), gentamicin (CN 10 μg), tetracycline (TE 30 μg) and trimethoprim/sulphamethoxazole (STX 1.25/23.75 μg) (HiMedia, New Delhi, India) were tested for Gram negative bacteria from oral swab and stool/rectal swab samples. For Gram positive bacteria erythromycin (E 15 μg), tetracycline (TE 30 μg), gentamicin (CN 10 μg), ciprofloxacin (CIP 5 μg), clindamycin (CD 2 μg) and cefoxitin (FOX 30 μg) (HiMedia, New Delhi, India) were tested. Inoculated plates of MHA were incubated aerobically at 37 °C for 18–24 h. Interpretation of zones of inhibition measured in millimeters were performed as recommended by Clinical and Laboratory Standards Institute (CLSI, 2020) guidelines [38].

### 4.5.4. ESBL Confirmation

Isolates were confirmed to be ESBL producers using disc approximation methods as previously described [39]. Two cephalosporins discs (ceftriaxone and ceftazidime) were seeded edge-by-edge with amoxicillin/clavulanic discs on MHA plates at a distance of 15 mm, inhibition of cephalosporins discs enhanced towards the disc of amoxicillin/clavulanic acid were interpreted as positive for ESBL production [40].

### *4.6. Data Management and Analysis*

Data were collected using Epicollect5 (https://five.epicollect.net/), accessed on 10 August 2021 and analyzed using STATA version 15 software. The categorical variables were summarized as proportions while continuous variables were summarized as median [IQR; interquartile range] or mean (±SD; standard deviation) depending on the distribution. Chi square test was used to compare socio-demographic characteristics, ESBL-PE and MRSA colonization among HIV− and HIV+ children below five years of age. A stepwise variable selection algorithm was used was used to determine factors associated with ESBL- PE colonization. A *p* value of <0.05 was considered statistically significant. All analyses were conducted using STATA version 15.

### 5. Conclusions

Overall, the MDR carriage in children below five years of age was high and was increased by recent antibiotic use and a relative living in same household being admitted to hospital in the last three months. HIV+ children were more colonized by isolates resistant to uncommonly used antibiotics in the community. Antibiotic stewardship and guidance on appropriate use is extremely important in this setting to prevent resistant bacteria from further spreading in the community and healthcare facilities. There is a need to expand antibiotic stewardship programs, strengthening IPC practices to prevent the emergence and further spread of MDR pathogens in LMIC. Further studies are needed to establish the genotypes and mobile genetic elements responsible for persistence and transmission of MDR pathogens among children in the city of Mwanza, Tanzania.

**Author Contributions:** D.R.M. and S.E.M. conceived and designed the study; D.R.M., V.S. and T.M. supervised execution of the study; S.E.M., B.R.K. and D.R.M. analyzed data. M.M.M., B.T.M., B.S., E.B., E.L., J.B. and S.E.M. critically reviewed study findings. D.R.M. wrote the initial draft of the manuscript which was critically reviewed and approved all authors. All authors have read and agreed to the published version of the manuscript.

**Institutional Review Board Statement:** The protocol for conducting this study was sought from the joint CUHAS/BMC research ethics and review committee for ethical clearance number CREC/418/2020. Permission to conduct the study was requested from all hospital authorities. Written informed consent was sought from guardians/parents of these children after full disclosure of the protocol. Confidentiality was ensured and maintained throughout the study.

**Informed Consent Statement:** Written informed consent has been obtained from the patient(s) to publish this paper.

**Data Availability Statement:** Summary statistics have been included in this manuscript; raw data are available upon request from the Director of Research and Innovation of the Catholic University of Health and allied Sciences.

**Conflicts of Interest:** The authors declare that they have no competing interests.

## Abbreviations

| | |
|---|---|
| AIDS | Acquired Immunodeficiency Syndrome |
| CD4 | Cluster of Differentiation 4 |
| CSF | Cerebrospinal Fluid |
| ESBL-PE | Extended –spectrum beta lactamase-producing Enterobacteriaceae |
| FBP | Full Blood Picture |
| HIC | High Income countries |
| HIV | Human immunodeficiency virus |
| IPC | Infection Prevention and Control |
| IQR | Interquartile Range |
| LMIC | Low- and Middle-Income Countries |
| MDR | Multidrug resistant |
| MRSA | Methicillin Resistant Staphylococcus aureus |
| MUAC | Mid-Upper Arm Circumference |
| NICU | Neonatal Intensive Care Unit |
| OI | Opportunistic infection |
| PICU | Pediatric Intensive Care Unit |
| PIC | Prevention Infection Control |
| SD | Standard Deviation |
| SIM | Sulphur, Indole Motility |
| TSI | Triple Sugar Iron Test |
| WBC | White Blood Cells |
| WHO | World Health Organization |

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