Changing Trends in Prevalence and Antibiotics Resistance of Uropathogens in Patients Attending the Gondar University Hospital, Northwest Ethiopia

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1. Background

Urinary tract infection is one of the commonest bacterial infections encountered in daily clinical practice [1]. It has been estimated that worldwide about 150 million people suffer from asymptomatic and symptomatic UTIs each year [2]. In most parts of the sub-Saharan Africa, as well as in other developing parts of the world, UTI is among the most common health problems occurring both in the community and hospitalized patients [3]. Since the last two to three decades, just as many community and hospital acquired bacterial infections, UTIs due to multidrug resistant uropathogens have caused a growing concern worldwide [1, 4–6]. Investigators [1, 7, 8] explained that the drug resistance problem in Africa stems from factors like indiscriminate use of antibiotics, inappropriate advertisement, and erratic prescription by unqualified drug sellers.

Since the previous two decades, the problem of UTIs due to uropathogens resistant to the commonly used antibiotics was reported by many authors in Ethiopia in general and in Gondar region in particular [1, 9–12]. Consequently, the prevalence of urinary tract pathogens and their resistance to the different antibiotics may have changed over the years in the study area. Hence, studies are needed as a guide in the community and hospital health care settings. The objectives of this study therefore were to assess the changing prevalence and resistance patterns of the uropathogens to commonly used antibiotics in a two-year study period.

2. Materials and Methods

2.1. Study Design and Area. This hospital based cross-sectional study was conducted at the Gondar University hospital from September 1, 2011, to June 30, 2012. Gondar University
hospital is found in Gondar town, located 727 km from Addis Ababa to the Northwest Ethiopia. Gondar town has a population of about 207,000 [13].

2.2. Sample Collection and Processing. Urine samples were collected by a clean-catch midstream, catheterization, or use of urine bags in a sterile container from each study participant whom the clinicians suspected for UTI and who has not received antibiotic therapy during the previous 14 days. Isolation of uropathogens was done by a surface streak procedure on Cysteine Lactose Electrolyte Deficient (CLED) medium using a 0.001 mL calibrated procedure on Cysteine Lactose Electrolyte Deficient (CLED) days. Isolation of uropathogens was done by a surface streak has not received antibiotic therapy during the previous 14 days. Isolation of uropathogens was done by a surface streak with the suspension using sterile cotton swap and the antibiotic discs were placed over the agar and left for 30 minutes for diffusion of the antibiotics in the disc. The plates were inverted upside down and incubated at 37°C for 18 to 24 hours. The zones of inhibition were then read as resistant and sensitive using calibrated ruler and compared with the standard chart [15]. Intermediate results were few in number and therefore were considered as resistant for convenience. Antibiotics agents employed for susceptibility testing were ampicillin (10 μg), amoxicillin (30 μg), co-trimoxazole (25 μg), gentamicin (10 μg), ciprofloxacin (5 μg), penicillin (10 IU), and erythromycin (15 μg) (Oxoid, Ltd, UK).

This study was done using the same method conducted by Moges et al., 2002 [1], 10 years back in the same study area, and the present results were compared with the previous report [1].

2.4. Quality Control. Each batch of the culture media used was tested for sterility. Standard control strains of E. coli ATCC 25922 and S. aureus ATCC 25923 were used during cultivating and antibiotics susceptibility testing as a control throughout the study.

2.5. Data Analysis. Data were checked for completeness, cleaned manually, entered, and analyzed using SPSS version 16 statistical software. The chi-square test (χ²) was used to measure the association and a P value less than 0.05 was considered statistically significant.

2.6. Ethical Clearance. Ethical clearance was obtained from the research and publication office of the University of Gondar, College of Medicine and Health Sciences. The data were collected after a written informed consent was sought from each study participant.

3. Results

Of the total 538 consecutive urine samples cultured, 284 (52.8%) gave significant bacteriuria. The majority (58.8%) of the positive cases were females while the remaining (41.2%) were males. The age of the study participants ranged from 1 year to 50+ years with the median age of 24. The frequency of positive urine cultures (28.2%) was high in the age group of 20–29 years followed by the age group of 50+ years (Table 1).

As it is seen from Table 2, the most common isolate of the uropathogens was E. coli (42.3%), followed by Klebsiella spp. (14.4%), CoNS, (12.3%), S. aureus (9.2%), Proteus spp., and Enterobacter spp. (5.6%). Klebsiella spp. showed the highest rate of resistance to amoxicillin (97.7%), ampicillin (95.1%), tetracycline and cotrimoxazole (82.9%), and chloramphenicol (78%) compared to E. coli to the same antibiotics. Citrobacter, Enterobacter, and Proteus species revealed higher rate of resistance patterns to tetracycline, ampicillin, amoxicillin, cotrimoxazole, and chloramphenicol. S. aureus and CoNS also demonstrate higher resistance rates to tetracycline, cotrimoxazole, amoxicillin, ampicillin, and penicillin. All gram negative uropathogens were resistant to 25–60% ciprofloxacin, while 53.8% S. aureus and 54.3% CoNS were resistant to this antibiotic.

### Table 1: Frequency of positive urine cultures for bacteria by age and sex in UTI suspected patients attending Gondar University Hospital, Northwest Ethiopia, 2012.

| Variable           | Frequency of positive isolates N (%) | χ² | P value |
|--------------------|-------------------------------------|----|---------|
| Sex                |                                     |    |         |
| Male               | 117 (41.2)                          | 45.8 | 0.007 |
| Female             | 167 (58.8)                          |    |         |
| Total              | 284 (100.0)                         |    |         |
| Age group in years |                                     |    |         |
| <5                 | 29 (10.2)                           | 2.1 | 0.32   |
| 5–9                | 13 (4.6)                            |    |         |
| 10–14              | 19 (6.7)                            |    |         |
| 15–19              | 11 (3.9)                            |    |         |
| 20–29              | 80 (28.2)                           |    |         |
| 30–39              | 40 (14.0)                           |    |         |
| 40–49              | 29 (10.2)                           |    |         |
| 50+                | 63 (22.2)                           |    |         |
| Total              | 284 (100.0)                         |    |         |

χ²: chi-square test.
Referring to Table 3, of the 282 total urinary bacterial isolates tested for antibiotics resistance patterns, 86.5% have shown multidrug resistance (resistant to ≥2 antibiotics). The rest, Enterobacter, Proteus, Streptococcus, Providencia, and Pseudomonas spp., were resistant to five antibiotics tested, whereas Citrobacter spp. were resistant to four antibiotics.

Most of the uropathogens’ isolates compared in the two study periods (2002 and 2012) showed no statistically significant differences in the isolation rates, except S. aureus which revealed a statistically significant decrease (18% to 9.2%) (P = 0.002) and Enterobacter spp. which revealed a significant increase (1.7% to 5.6%) (P < 0.001) in the current study (Table 4).

Table 5 shows that the resistance rate (34.2%) of E. coli for gentamicin in the present study was significantly higher than the rate (14.1%) of ten years back (P = 0.04). Similarly, the resistance rate (38.4%) of S. aureus isolates for ciprofloxacin in our study is significantly higher than the rate (6.5%) of the previous study (P = 0.03). The uropathogens (E. coli, Klebsiella spp., CoNS, Citrobacter spp., and Proteus spp.) in the previous study were found to be 100% sensitive to ciprofloxacin except the few isolates of S. aureus 2 (6.5%).

Table 6 shows that E. coli was more resistant to the tested antibiotics in our study than the previous ones. Klebsiella spp. was resistant to seven antibiotics in this study than the previous ones which were resistant to five antibiotics. Previous isolates of Proteus spp. were less resistant to three and four antibiotics than in the present study. Citrobacter spp. of the present study were resistant to six antibiotics, whereas they were resistant to five in the previous study. CoNS and S. aureus each showed resistance to seven antibiotics in both study periods.

### 4. Discussion

In this study, the overall rate of isolation of the uropathogens was significantly higher than the previous rate (41%) reported from the same study area [1] but lower than the rate reported in Nepal (71.7%) [17]. This disparity of rates may be attributed...
| Isolate      | N (%) | R0 (%) | R1 (%) | R2 (%) | R3 (%) | R4 (%) | R5 (%) | R6 (%) | R7 (%) |
|-------------|-------|--------|--------|--------|--------|--------|--------|--------|--------|
| E. coli     | 120 (42.3) | 15 (12.5) | 3 (2.5) | 15 (12.5) | 18 (15) | 20 (16.7) | 18 (15) | 21 (17.5) | 9 (7.5) |
| Klebsiella spp. | 41 (14.4) | 0 (0) | 4 (9.8) | 4 (9.8) | 5 (12.2) | 12 (29.3) | 10 (24.4) | 4 (9.8) | 2 (4.9) |
| CoNS        | 35 (12.3) | 1 (2.9) | 6 (17.4) | 8 (22.9) | 5 (14.3) | 3 (8.6) | 5 (14.3) | 5 (14.3) | 2 (5.7) |
| S. aureus   | 26 (9.2) | 1 (3.8) | 1 (3.8) | 3 (11.5) | 2 (7.7) | 11 (42.3) | 3 (11.5) | 3 (11.5) | 1 (3.8) |
| Enterobacter spp. | 16 (5.6) | 1 (6.3) | 3 (18.8) | 2 (12.5) | 2 (12.5) | 2 (12.5) | 4 (25) | 2 (12.5) | 0 (0) |
| Proteus spp. | 16 (5.6) | 1 (6.3) | 1 (6.3) | 2 (12.5) | 2 (12.5) | 5 (31.3) | 3 (18.8) | 2 (12.5) | 0 (0) |
| Streplococcus spp. | 5 (1.5) | 0 (0) | 3 (30) | 0 (0) | 3 (30) | 2 (20) | 1 (10) | 1 (10) | 0 (0) |
| Citrobacter spp. | 10 (3.5) | 1 (10) | 1 (10) | 1 (10) | 2 (20) | 2 (20) | 1 (10) | 0 (0) | 2 (20) |
| Providencia spp. | 5 (1.8) | 1 (20) | 0 (0) | 1 (20) | 1 (20) | 2 (40) | 2 (40) | 0 (0) | 0 (0) |
| Pseudomonas spp. | 5 (1.8) | 0 (0) | 0 (0) | 0 (0) | 1 (20) | 1 (100) | 0 (0) | 0 (0) | 0 (0) |
| Serratia spp. | 2 (0.7) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 1 (100) | 0 (0) | 0 (0) | 0 (0) |
| Salmonella spp. | 1 (0.4) | 0 (0) | 0 (0) | 0 (0) | 2 (50) | 1 (100) | 0 (0) | 0 (0) | 0 (0) |

R: resistant to 0, 1, 2, 3, 4, 5, 6, and 7 antibiotics, respectively.

Table 4: Comparison of urinary bacterial isolates from UTI suspected patients at Gondar University Hospital in 2002 and 2012.

| Isolate      | 2002* N (%) | 2012 N (%) | \(\chi^2\) | P value |
|-------------|-------------|-------------|-------------|---------|
| E. coli     | 78 (45.3)   | 120 (42.3)  | 1.15        | 0.29    |
| Klebsiella spp. | 18 (10.5) | 41 (14.4)   | 1.08        | 0.3     |
| CoNS*       | Not isolated | 35 (12.3)   | 0           | 0       |
| S. aureus   | 31 (18)     | 26 (9.2)    | 9.64        | 0.002   |
| Enterobacter spp. | 3 (1.7) | 16 (5.6)    | 3.94        | <0.001  |
| Proteus spp. | 7 (4.1)     | 16 (5.6)    | 0.4         | 0.53    |
| Streptococcus spp. | 4 (2.3) | 5 (1.8)     | 0.4         | 0.53    |
| Citrobacter spp. | 10 (5.5)  | 10 (3.5)    | 1.6         | 0.2     |
| Pseudomonas spp. | 1 (0.6)   | 5 (1.8)     | 0           | 0.42    |
| Salmonella spp. | 3 (1.7)    | 1 (0.4)     | 0           | 1.00    |
| Serratia spp. | 2 (1.2)     | 2 (0.7)     | 0           | 0.63    |
| Hafnia alvei | 1 (0.6)     | Not isolated | 0           | 0       |
| Others       | Not isolated | 7 (2.5)     | 0           | 0       |
| Total        | 172 (41)    | 284 (52.8)  | 19.7        | <0.001  |

* Moges et al., 2002 (East African Journal) [1]; ** coagulase negative Staphylococci.

to the differences in the study samples and improper collection and processing of specimens. In our study, women have higher rate of UTI than men (Table 1), because in females the urethra has been known to be shorter and closer to the anus [18]. Other investigators have also reported similar findings to ours [5, 19]. The high prevalence of UTI in age groups of 20–29 and more than 50 years may be related to sexually active age group and older study participants whose immune system may be impaired, respectively.

The distribution of the uropathogens isolated in our study was almost similar to the results reported previously [1] except with S. aureus. The most commonly isolated uropathogen in the present study was E. coli and it is consistent with the previous study done in the same study area [1]. However, the frequency of E. coli in urine samples varies in different studies [20, 21]: these may be due to the large variation of different species of bacteria in the study and variations in specimen collection and processing.

Antibiotic resistance is a major clinical problem in treating infections caused by different bacterial pathogens. Resistance to antibiotics has increased over the years. In our present study, a higher proportion of isolates of members of Enterobacteriaceae was in average resistant to ampicillin (84%), tetracycline (83%), amoxicillin (80%), cotrimoxazole (77%), chloramphenicol (69%), gentamicin (50%), and ciprofloxacin (45%), whereas the resistance patterns of these
Table 5: Percentage of isolates with antimicrobial resistance in 2002 and 2012.

| Antibiotic       | Escherichia coli (%) | Klebsiella spp. (%) | CoNS # (%) | Staphylococcus aureus (%) | Citrobacter spp. (%) | Proteus spp. (%) |
|------------------|----------------------|---------------------|------------|---------------------------|----------------------|------------------|
|                  | 2002 | 2012 | 2002 | 2012 | 2002 | 2012 | 2002 | 2012 | 2002 | 2012 | 2002 | 2012 | 2002 | 2012 | 2002 | 2012 |
| Erythromycin     | NA   | NA   | NA   | NA   | 71.5**| 37.2 | 19.4 | 50.0* | NA   | NA   | NA   | NA   | NA   | NA   | NA   | NA   | NA   |
| Gentamicin       | 14.1 | 34.2*| 33.3 | 39   | 21.4**| 0    | 22.6**| 0    | 50   | 50   | 28.6 | 25   | 25   | 25   | 25   | 25   |
| Penicillin       | NA   | NA   | NA   | NA   | 57.1 | 48.6 | 38.7 | 50.0 | NA   | NA   | NA   | NA   | NA   | NA   | NA   | NA   | NA   |
| Chloramphenicol  | 42.3 | 48.3 | 77.8 | 70.7 | 50   | 0    | 51.6**| 0    | 80   | 50   | 71.4 | 56.2 | 56.2 | 56.2 | 56.2 | 56.2 |
| Tetracycline     | 68   | 62.5 | 77.8*| 61   | 71.4 | 80.8 | 73.1 | 80   | 80   | 100  | 62.4 | 62.4 | 62.4 | 62.4 | 62.4 | 62.4 |
| Co-trimoxazole   | 56.4 | 70   | 72.2 | 78   | 57.1 | 57.1 | 48.4 | 73.1 | 90   | 80   | 71.4 | 75   | NA   | NA   | NA   | NA   |
| Ampicillin       | 69.2 | 56.7 | 94.4 | 70.7 | 21.4 | 48.6 | 25.8 | 50   | 100**| 50   | 71.4 | 62.4 | 62.4 | 62.4 | 62.4 | 62.4 |
| Ciprofloxacin    | 0    | 30*  | 0    | 24.4*| 0    | 25.7*| 6.5  | 38.4*| 0    | 30*  | 0    | 18.8*| 18.8*| 18.8*| 18.8*| 18.8*|
| Amoxicillin      | NA   | 37   | ND   | 43.9 | ND   | 28.6 | NA   | 38.5 | ND   | 20   | NA   | 50   | NA   | NA   | NA   | NA   | NA   |
| Total            | 78 (45.3) | 120 (42.3) | 18 (10.5) | 41 (14.4) | 14 (8.1) | 35 (12.3) | 31 (18) | 26 (9.2) | 10 (5.8) | 10 (3.5) | 7 (4.1) | 16 (5.6) | *P < 0.05 compared with results for 2002; **P < 0.05 compared with results for 2012; #coagulasenegative Staphylococci; ND: not done; NA: not applicable.

Table 6: Comparison of multidrug resistance patterns of urinary isolates from UTI suspected patients at Gondar University Hospital in 2002 and 2012.

| Isolate/study periods | N (%) | R2 | R3 | R4 | R5 | R6 | R7 |
|----------------------|-------|----|----|----|----|----|----|
| **E. coli**          |       |    |    |    |    |    |    |
| 2002                 | 78 (45.3) | 5 (6.4) | 19 (24.4) | 14 (17.9) | 11 (14.1) | 1 (1.3) | 1 (1.3) |
| 2012                 | 120 (42.3) | 15 (12.5) | 18 (15) | 20 (16.7) | 18 (15) | 21 (15.5) | 9 (7.5) |
| **Klebsiella spp.**  |       |    |    |    |    |    |    |
| 2002                 | 18 (10.5) | 1 (5.6) | 1 (5.6) | 7 (38.9) | 5 (27.8) | 0 | 0 |
| 2012                 | 41 (14.4) | 4 (9.8) | 5 (12.2) | 12 (29.3) | 10 (24.4) | 4 (4.9) | 2 (4.9) |
| **Proteus spp.**     |       |    |    |    |    |    |    |
| 2002                 | 7 (4.0) | 0 | 0 | 0 | 4 (57.1) | 1 (14.3) | 0 |
| 2012                 | 16 (5.6) | 2 (12.5) | 2 (12.5) | 5 (31.3) | 3 (18.8) | 2 (12.5) | 0 |
| **Citrobacter spp.** |       |    |    |    |    |    |    |
| 2002                 | 10 (5.8) | 1 (10.0) | 1 (10.0) | 2 (20.0) | 5 (50.0) | 0 | 0 |
| 2012                 | 10 (3.5) | 1 (10) | 2 (30) | 2 (20) | 1 (10) | 1 (10) | 0 |
| **CoNS**             |       |    |    |    |    |    |    |
| 2002                 | 14 (8.1) | 2 (14.3) | 3 (21.4) | 1 (7.1) | 0 | 1 (7.1) | 1 (7.1) |
| 2012                 | 35 (12.3) | 8 (22.9) | 5 (14.3) | 3 (8.6) | 5 (14.3) | 5 (14.3) | 5 (5.7) |
| **S. aureus**        |       |    |    |    |    |    |    |
| 2002                 | 31 (18.0) | 1 (3.2) | 3 (9.7) | 7 (22.6) | 2 (6.5) | 2 (6.5) | 3 (9.7) |
| 2012                 | 26 (9.2) | 3 (11.5) | 2 (7.7) | 11 (42.3) | 3 (11.5) | 1 (3.8) | 1 (3.8) |

*Resistant to 2, 3, 4, 5, 6, and 7 antibiotics, respectively.

isolates, a decade ago [1], were ampicillin (81%), tetracycline (78%), cotrimoxazole (67%), gentamicin (36%), and ciprofloxacin (0%). As has been clarified here, the most significant change among uropathogens in 10 years period has been the significant increase of resistance to gentamicin, and the emergence of resistance to ciprofloxacin from zero in the study conducted 10 years ago, to 45% in our case.

In comparing the two periods of study (2002 versus 2012) (Table 5), the resistance rate of *E. coli* (44.2%) for gentamicin in the present study was significantly higher than the rate (14.1%) of the previous study (*P* = 0.04) [1] which is supported by a report from teaching hospital in Saudi Arabia where there was high ciprofloxacin resistance in gram positive cocci, particularly *S. aureus* [24]. Hence, the overall trend of the present study indicates the emergence of increasing number of different multidrug resistant isolates of uropathogens than those in the previous study. Increased resistant patterns of the isolates for ciprofloxacin may be explained by the fact that the widespread use of the drug in the area might have favored the resistant isolates due to selective pressure.

Multiple drug resistance clinical isolates are increasing every time and some of the drugs are approaching to be no more essential for the treatment of UTI patients [17, 24]. Similarly, the resistance rate of *S. aureus* isolates for ciprofloxacin in our study is significantly higher than the rate (6.5%) of the previous study (*P* = 0.03) [1] which is supported by a report from teaching hospital in Saudi Arabia where there was high ciprofloxacin resistance in gram positive cocci, particularly *S. aureus* [24]. Hence, the overall trend of the present study indicates the emergence of increasing number of different multidrug resistant isolates of uropathogens than those in the previous study. Increased resistant patterns of the isolates for ciprofloxacin may be explained by the fact that the widespread use of the drug in the area might have favored the resistant isolates due to selective pressure.
The overall trends of the present study indicated that there are increasing multidrug resistant spp. in the present study (Table 3) compared to the previous isolates [1]. These studies urge a need for a large scale monitoring of drug resistance problems in different parts of the country and evaluate susceptibility patterns of the isolates. This would have a paramount importance in using empiric treatment which would be safe, effective, and economical for the patient.

5. Conclusion

This study has showed more resistant E. coli and Klebsiella spp. and S. aureus and CoNS to seven antibiotics tested over a ten-year period. The resistance rates of E. coli for gentamicin and S. aureus for ciprofloxacin in the present study were also higher than the rates of a decade ago. The uropathogens (E. coli, Klebsiella spp., CoNS, Citrobacter spp., and Proteus spp.) ten years back were found to be all sensitive to ciprofloxacin except the few isolates of S. aureus. Gentamicin and ciprofloxacin have been recommended for empirical treatment of urinary tract infections in areas where diagnostic bacteriologic services are not available.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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