

High Carrying Rate of Extended-Spectrum Beta-Lactamase (ESBL) Producing Enterobacteriaceae by Slaughterhouse Workers in Lomé, Togo in 2019

ABSTRACT

Introduction: Extended-spectrum beta-lactamase-producing Enterobacteriaceae (ESBL) represent a real public health concern because of their spread. The role of agri-food chains in transmitting of digestive ESBL-producing bacterial strains in the community, has been demonstrated but little work has been done in our settings (Togo, west Africa). The aim of this study was to estimate the rate of digestive carrying ESBL producing enterobacteria in slaughterhouse workers in Lomé, Togo.

Materials and Methods: This is a cross-sectional study carried out in three slaughterhouses in Lomé. Fresh stools of 60 slaughterhouse workers and socio-demographic data were collected during the period of September to October 2019 after obtaining the consent of each participant.

The bacterial strains of interest were isolated on the selective medium Purple Bromocresol + Ceftazidime at 6µg/l. Uriselect[®] and API 20E media were used for identification. Antibiotic susceptibility test was performed in Mueller-Hinton agar plate diffusion method (Kirby Bauer technic) and according to CASFM-EUCAST recommendations.

Results: The digestive carriage rate of ESBL producing bacteria among professionals of three slaughterhouses of Lomé was 80% (n=48/60). *Escherichia coli* was the main bacteria 78.2% (n = 43/55) followed by *Klebsiella pneumoniae* 16.4% (n = 9/55) and *Enterobacter cloacae* 5.4% (n = 3/55). The antibiotic profile of ESBL producing enterobacteriaceae showed resistance to Amoxicillin + Clavulanic Acid (26%), Ticarcillin + Clavulanic Acid (86%), Piperacillin + Tazobactam (14%), Cefoxitin (7%) Ciprofloxacin (63%), Levofloxacin (49%), Nalidixic Acid (42%), Chloramphenicol (33%), Gentamicin (21%), Sulfamethoxazole-Trimetoprim (93%). These bacteria are 100% sensitive to Imipenem, Ertapenem, Amikacin and Fosfomycin.

Conclusion: This study reveals a very high carriage rate of ESBL producing *Escherichia coli* amongst Slaughterhouse Workers in Lomé. It confirms the major potential role of the agri-food chains in the spread of ESBL producing bacteria in the Community.

Keywords: Digestive Carrying ESBL producing bacteria, Workers, Slaughterhouse, Lomé

1. INTRODUCTION

Over the past decade, antibiotic resistance in Enterobacteriaceae has increased dramatically worldwide. This situation is mainly due to an increased prevalence of enterobacteria producing extended spectrum β -lactamases (ESBLs) (Ilse Overdevest et al., 2011).

The emergence of multi-drug resistant bacteria (MDRBs), in particular the enterobacteria producing β -extended-spectrum lactamase (ESLB), is a concerned phenomenon in developing countries (Rogers BA et al., 2011).

Extended-spectrum β -lactamases (ESBLs) have been detected in meat samples and transmission of ESBLs from livestock to humans through the food chain has been suggested (Jouini A et al.; Overdeest I et al.; Leverstein-van Hall MA et al. 2011; Kola A et al. 2012). ESBL-producing Enterobacteriaceae can be transferred from animals to humans through food or direct contact (Dohmen W, et al., 2015; Kola A, et al., 2012). Direct contact with livestock mainly occurs in an occupational setting. In farmers, carriage of ESBL-producing Enterobacteriaceae is associated with the presence of ESBL-producing Enterobacteriaceae in animals (Dohmen W, et al., 2015; Dierikx C, et al. 2013; Hammerum AM, et al., 2014).

Slaughterhouse workers might also be occupationally exposed to ESBL-producing Enterobacteriaceae. Depending on the job task, slaughterhouse workers have frequent contact with live animals, animal carcasses or animal products (Geser N, et al., 2011).

Studies have shown that one of the risk factors for the acquisition of extended spectrum beta lactamase (ESBL)—producing organisms is the abuse of antibiotics in poultry production

(Agyare C et al. 2019). Globally, there have been an increase in the challenges related to treatment of human and animal bacterial infections attributed to the development of antimicrobial resistance (Jasovsky D et al. 2016). Resistance to commonly used antibiotics has major socioeconomic and public health implications.

The socioeconomic implications of AMR include increased cost and duration of treatment while the public health implications include decreased ability to treat common infections resulting in increased human suffering and ultimately death (Li B, Webster TJ, 2018; Prestinaci F et al. 2015; Michael CA, 2014).

The possibility of antimicrobial resistance genes circulating among humans, animals and the environment constitutes a direct threat to public health. This is why it is critical to develop new approaches and institute strict control measures in accessing and using antimicrobials in humans and animals (Miles TD et al. 2006).

A few rare studies in Africa have investigated the fecal carriage of ESBL-producing enterobacteria in the community and in hospitalized patients (Abdoul-Salam Ouedraogo et al., 2016, Oumar Ouchar Mahamat et al. 2019). In Senegal, Michaud et al. in 2014 studied the ESBL carriage among workers and pigs. Nevertheless, carrying among slaughterhouse workers is poorly described.

In Togo, authors have described the phenomenon of bacterial resistance due to ESBLs in the hospital environment without addressing the carrying of ESBLs in the agro-food chain (Toudji Akouetevi et al. 2017, Salou et al., 2011).

The purpose of this study is to estimate the prevalence of ESBL-producing Enterobacteriaceae carriage among workers in three slaughterhouses in Lomé.

2. MATERIAL AND METHODS

Study area

Our study was undertaken in three slaughterhouses in Lomé, the capital city of Togo (west Africa), which is located in the south-west of the country along the coast of the Gulf of Guinea with an area of 90 Km square. It has 1,477,660 inhabitants according to the 2010 census.

The slaughterhouse in the port area ONAF (National Office of refrigerated slaughterhouse of Lomé) is located in district II of Lomé. The slaughter areas of Agoè-Zongo and Gbossimè are located in the Agoè district and district V of Lomé respectively.

The samples were analyzed at the microbiology laboratory of Sylvanus Olympio Teaching Hospital of Lomé.

Study design

This is a cross-sectional study that took place from 1st September to 31th October 2019. The study was conducted among volunteer slaughterhouse workers who are selected upon their consent. Any slaughterhouse worker who gave his consent to participate to the study was included. After an explanation about the aim of the study, each worker gave his consent by signing a consent form. Socio-demographic data and risk factors were collected by using an individually questionnaire.

Stools sample were collected from each participant in a clean jar. Each participant gave consent to the study after explanation and signed the consent form for participation in the study.

Ethical approval for this study was obtained from the Scientific and Ethical committee of the

Health Research Ethics Committee (Approval Number: 048/2019/CBRS). Permission was sought from the management of each study site prior start the study. Confidentiality of information obtained was assured.

Sampling method

A refrigerated slaughterhouse and two slaughter areas have been selected: refrigerated slaughterhouse of the harbor area (ONAF), the slaughter areas of Agoè-Zongo and Gbossimè.

A letter of request was sent to the Livestock Direction of Lomé, Togo, which sent correspondence to ONAF to allow the study.

At the study site, an explanation is given to employees who are urged to participate in the study. Only those who have given their informed consent by signing the consent form are included in the study. A clean jar is given to each participant for this purpose, who will bring fresh stool the next day. The stool is transported in a cooler to the bacteriology laboratory at Sylvanus Olympio Teaching Hospital within 2 hours of collection. The samples are analyzed for the presence of Enterobacteriaceae producing Extended Spectrum Beta-Lactamase (ESBL).

Questionnaire Survey

The identification of risk factors for ESBLs includes the administration of a questionnaire to slaughterhouse professionals. The questionnaire includes socio-demographic data such as age, gender, marital status, neighbourhood of residence, education level and risk factors for carrying ESBLs, occupation, water used on site, working hours in the area, I, self-medication, hospitalization in the previous year, antibiotic use in the previous three months

Microbiological analysis

Sample pre-treatment

Briefly about one Gram of human fresh stool sample was inoculated in enrichment broth (thioglycolate broth) and incubated for 5 hours at 37°C. A loop-full culture from enrichment broth was streaked onto BCP selective agar + 6 µg/l Cefotaxime previously dried and incubated at 37°C for 18-24 hours.

Isolation and identification of ESBL-producing strains

Enterobacteriaceae grown on the selective medium BCP + 6 µg/l of Cefotaxime are selected according to their morphological appearance and re-isolated on the Uriselect® medium for identification. Non-ESBL strains do not grow on this selective medium.

Escherichia coli strains are pink in colour on the Uriselect® medium. Other strains are identified by biochemical commercialized gallery API 20E (Biomérieux, France).

Antimicrobial susceptibility profiling

The antibiotic susceptibility patterns of Enterobacteriaceae isolates strains were tested using the disk diffusion method (Kirby Bauer method) on Mueller Hinton Agar following standard zone size interpretative criteria recommended by CASFM-EUCAST (European Committee of Antibiogram susceptibility Testing) 2019 (Richard BONNET et al., 2019)

One colony from overnight culture on Mueller Hinton (MH) medium was picked using sterile Pasteur loop and emulsified in 5 ml of sterile normal saline to adjust the inoculum density equal to that of 0.5 Mc Farland turbidity standards (1.5×10^8 CFU/ml). Using a sterile swab, the bacteria was spread on Mueller Hinton agar to obtain a lawn culture within 24 hours. After air drying, commercially available antibiotics discs (Oxoid, UK) were placed on the medium 30 mm apart and 15 mm away from the edge of the plate and incubated at 37°C for 24 hours. The agar was then dried. Inhibition zone diameter was measured with a double decimeter, recorded and values interpreted according to the recommendations of EUCAST 2019.

The isolates were tested using a panel of 21 antibiotics from different families commonly used to treat human bacterial infections namely Ampicillin 10 µg, Ticarcillin 75 µg, Amoxicillin + Clavulanic Acid 30 µg, Ticarcillin + Clavulanic Acid 85 µg, Cefoxitin 30 µg, Piperacillin 30 µg, Piperacillin + Tazobactam 36 µg, Ceftazidime 10 µg, Ceftriaxone 30 µg, Aztreonam 30 µg, Cefepime 30 µg, Imipenem 10 µg, Ertapenem 10 µg, Amikacin 15 µg, Gentamicin 10 µg, Chloramphenicol 30 µg, Sulfamethoxazole-Trimetoprim 25 µg, Nalidixic Acid 30 µg, Ciprofloxacin 5 µg, Levofloxacin 5 µg, Fosfomycin 200 µg. A

standard reference strain of *Escherichia coli* ATCC25922, sensitive to all antimicrobial drugs being tested, was used as a control.

Detection of the ESBL phenotype by the double disk method

All isolated strains of Enterobacteriaceae were tested for ESBL production by the double disk method during the antibiotic susceptibility testing (Carter MW et al., 2000). Combination disk Amoxicillin + clavulanic acid 20+10 µg or Ticarcillin + clavulanic acid 75+10 µg was applied at the center of Mueller Hinton Agar which was inoculated with the test strain. Ceftazidime 10 µg and Cefepim 30 µg disks frame Ticarcillin + Clavulanic Acid 75 + 10 µg disc 30 mm apart from each other and 15 mm from the edge of the plate. After 18-24 hours of incubation at 37°C, isolate that showed increase of ≥ 5mm the zone of inhibition of the combination disks in comparison to that of the Ceftazidime and Cefepim disk was considered an ESBL. *Escherichia coli* ATCC 25922 and a *Klebsiella pneumoniae* ATCC 700603 were used as negative and positive controls, respectively.

Statistical Analysis

The collected data on a survey form are entered into the Excel software. The statistical analysis was done by the GraphPad Prism 8 and Excel software. The data are analyzed by calculating proportions, prevalence and P value. Statistical significance tests of risk factor for carrying enterobacteriaceae producing ESBL were performed using the Chi-square /Fisher's exact test at 5% level of significance (GraphPad Prism software, version 8).

3. RESULTS

During the study period, we included and screened 60 slaughterhouse workers. They were male (100.0%). The age of the participants ranged from 23 to 76 years with a median age of 39 years. Almost (93.3%) (n= 56) of participants are married and 60.0% (n= 36) of them have a secondary school education (Table 1)

Prevalence of carrying ESBL

The prevalence of ESBL-producing Enterobacteriaceae carriage is 80% (n = 48) among 60 screened professionals. Upon 48 workers screened positive, 55 strains of ESBL-producing Enterobacteriaceae were isolated. *Escherichia coli* was the majority strain (78.2%) (n = 43) followed by *Klebsiella pneumoniae* (16.4%) (n = 9) and *Enterobacter cloacae* (5.4%) (n = 3) (Figure 1). In 7(11.7%) professionals, an association two ESBL bacteria has been found (Table 2). Of the seven cases of association of bacteria, 5 (71.4%) were from Agoè Zongo slaughter

Table 1: Socio-demographic characteristics of slaughterhouse workers in Lomé-Togo, 2019

| Characteristics | N (%) | % |
|-----------------|-------|-------|
| Marital status | | |
| Married | 56 | 93.3% |

| | | |
|---------------------------------------|----|-------|
| Single | 4 | 6.7% |
| Education level | | |
| Uneducated | 4 | 6.7% |
| Primary | 14 | 23.3% |
| Secondary | 36 | 60.0% |
| Superior | 6 | 10.0% |
| Profession | | |
| Butcher/ pork butcher | 31 | 51.7% |
| Skilled and specialized worker | 16 | 26.7% |
| Veterinarian | 4 | 6.7% |
| Inspection Officers | 5 | 8.3% |
| Tax collector | 3 | 5.0% |
| Security guard | 1 | 1.7% |
| Duration of work (years) | | |
| 0-9 | 22 | 36.7% |
| ≥ 10 | 38 | 63.3% |
| Locality | | |
| Slaughterhouse ONAF | 29 | 48.3% |
| Slaughter area of Agoè Zongo | 20 | 33.3% |
| Slaughter area of Gbossime | 11 | 18.3% |

Table 2: Distribution of ESBL producing bacteria association by slaughterhouse area

| Area | <i>Escherichia coli</i> and <i>Klebsiella pneumoniae</i> | <i>Escherichia coli</i> and <i>Enterobacter cloacae</i> | <i>Escherichia coli</i> and <i>Escherichia coli</i> * | Total |
|-------------------------------------|--|---|---|-------|
| Slaughterhouse ONAF | 1 | 1 | 0 | 2 |
| Slaughter area of Agoè Zongo | 3 | 0 | 2 | 5 |
| Slaughter area of Gbossime | 0 | 0 | 0 | 0 |

| | | | | |
|--------------|---|---|---|---|
| Total | 4 | 1 | 2 | 7 |
|--------------|---|---|---|---|

*phenotype was different

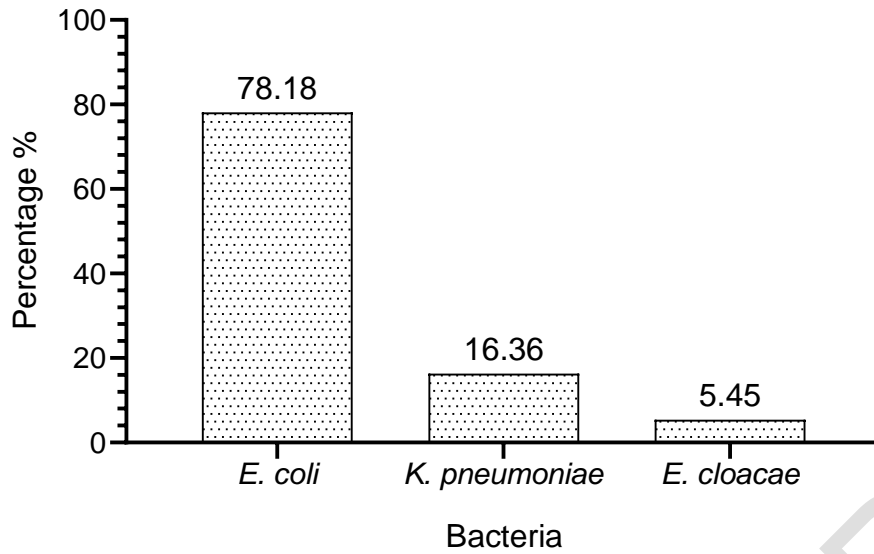


Figure 1: Proportion of isolated enterobacteria from slaughterhouse workers in Lomé, 2019

Carrying risk factors analysis

Table 3: Distribution of slaughterhouse workers and ESBL carriage over different contact exposure in Lomé, 2019

| | Total | ESBL Carriers | Non ESBL Carriers |
|--|-------|---------------|-------------------|
| | | | |

| | | | | |
|----------------|---|----|----|---|
| High exposure | Contact with carcass | 17 | 15 | 2 |
| | Contact with stomach content | 5 | 5 | 0 |
| | Contact with live animals | 6 | 6 | 0 |
| | Contact with live animals, carcass, stomach content, animal faeces, blood | 27 | 18 | 9 |
| | Contact with carcass, stomach content | 1 | 0 | 1 |
| Lower exposure | Tax collector | 3 | 3 | 0 |
| | security guard | 1 | 1 | 0 |
| | | | | |

Table 4: Overview of participant characteristics and P value from the Fisher exact test for the probability of ESBL carriage in Lomé, 2019

| ESBL Carrying risk factors | ESBL carriers | Non ESBL carriers | <i>P</i> value |
|----------------------------|---------------|-------------------|----------------|
| Age | | | |
| 23-29 | 8 (100%) | 0(0%) | <0,0001 |
| ≥ 30 | 40 (77%) | 12 (23%) | |
| Matrimonial statut | | | |
| Married | 44 (79%) | 12 (21%) | <0,0001 |

| | | | |
|---------------------------------------|-----------|----------|---------|
| Single | 4 (100%) | 0 (0%) | |
| Education Level | | | |
| Less than Secondary | 15 (83%) | 3 (17%) | 0,5891 |
| Secondary and above | 33 (79%) | 9 (21%) | |
| Duration of work | | | |
| 0-9 years | 21 (91%) | 2 (9%) | 0,0015 |
| ≥ 10 years | 27 (73%) | 10 (27%) | |
| Source of water used on the work site | | | |
| Borehole | 30 (97%) | 1 (3%) | <0,0001 |
| National water supply(TDE*) | 18 (62%) | 11 (38%) | |
| Work Exposure | | | |
| High exposure | 44 (79%) | 12 (21%) | <0,0001 |
| Lower exposure | 04 (100%) | 0 (0%) | |
| Antibiotics Self-medication | | | |
| Yes | 31 (91%) | 3 (9%) | <0,0001 |
| No | 17 65% | 9 (35%) | |
| Hospitalization previous year | | | |
| Yes | 2 (100%) | 0 (0%) | <0,0001 |
| No | 46 (79%) | 12 (21%) | |
| Antibiotics in the previous 3 months | | | |
| Yes | 11 (73%) | 04 (27%) | 0,1751 |
| No | 37 (82%) | 08 18%) | |

*Togolaise des eaux

Resistance profile of isolated strains

The antibiotic profile of *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter cloacae* producing ESBL isolated showed 100% resistance to this bêta lactamin: Ampicillin, Ticarcillin, Piperacillin, Ceftazidim, Ceftriaxon, Aztreonam and Cefepim. *Escherichia coli* showed high resistance over 50% to Ticarcillin + Clavulanic Acid (79,07%), Ciprofloxacin (58,14%), and Sulfamethoxazole-Trimetoprim (86,05%).

The resistance rate of this antibiotics is less than 50%: Amoxycillin + Clavulanic Acid (13,95%), Piperacillin + Tazobactam (9,30%), Piperacillin + Tazobactam (9,30%), Cefoxitin (2,33%), Levofloxacin (39,53), Nalidixic Acid (44,19%), Chloramphenicol (30,23%), Gentamicin (9,30%).

Klebsiella pneumoniae showed resistance to Amoxicillin + Clavulanic Acid (55,56%), Ticarcillin + Clavulanic Acid (100%), Piperacillin + Tazobactam (55,56%), Cefoxitin (7%), Ciprofloxacin (66,67%), Levofloxacin (33,33), Nalidixic Acid (22,22%), Chloramphenicol (22,22%), Gentamicin (44,44%), Sulfamethoxazole-Trimetoprim (88,89%). Bacteria are 100% sensitive to Imipenem, Ertapenem, Amikacin.

Susceptibility of strains

Table 5: Resistance profile of isolated strain from slaughterhouse workers in Lomé, 2019

| Family of antibiotics | Name of antibiotic disk | Percentage of resistance (%) | | |
|-----------------------------|--|--------------------------------|------------------------------------|-----------------------------------|
| | | <i>Escherichia coli</i> N = 43 | <i>Klebsiella pneumoniae</i> N = 9 | <i>Enterobacter cloacae</i> N = 3 |
| Bêta-Lactamin | Ampicillin (10µg) | 100 | 100 | 100 |
| | Amoxicillin-clavulanate (20-10µg) | 13.9 | 55.6 | 66.7 |
| | Ticarcillin (75µg) | 100 | 100 | 100 |
| | Ticarcillin + Clavulanate (75-10µg) | 79.1 | 100 | 66.7 |
| | Pipêracillin (30µg) | 100 | 100 | 100 |
| | Pipêracillin + Tazobactam (36µg) | 9.3 | 55.6 | 0 |
| | Céfoxitin (30µg) | 2.3 | 0 | 66.7 |
| | Ceftazidim (10µg) | 97.7 | 100 | 100 |
| | Ceftriaxon (30µg) | 100 | 100 | 100 |
| | Céfépim(30µg) | 100 | 100 | 100 |
| | Aztréonam (30µg) | 100 | 100 | 100 |
| | Imipénèm (10µg) | 0 | 0 | 0 |
| | Ertapénèm (10µg) | 0 | 0 | 0 |
| | Aminosids | Amikacin (30µg) | 0 | 0 |
| Gentamicin (10µg) | | 9.3 | 44.4 | 66.7 |
| Sulfonamides and associates | Sulfaméthoxazole-Triméthoprim (27,75-1,25µg) | 86.0 | 88.9 | 100 |
| Phénicolés | Chloramphénicol (30µg) | 30.2 | 22.2 | 66.7 |

| | | | | |
|----------------|-------------------------|------|------|------|
| Quinolon and | Nalidixique Acid (30µg) | 44.2 | 22.2 | 33.3 |
| Fluoroquinolon | Ciprofloxacin (5µg) | 58.1 | 66.7 | 66.7 |
| | Lévofoxacin (5µg) | 39.5 | 33.3 | 0 |
| Other family | Fosfomycin (200µg) | 0 | 33.3 | 0 |

Discussion

The present study aimed to examine the prevalence and risk factors for carrying ESBL-producing enterobacteria among apparently healthy slaughterhouses professional in Lomé and their resistance profile to antibiotics.

Studies shown that there is an increase of prevalence of ESBL faecal carriage over the world. Studies from three different areas in Spain (Madrid, Barcelona and Zaragoza) showed the prevalence of faecal carriage between 5.5% and 8.1% during 2002 and 2004 (Valverde A *et al.* 2004; Miro´ E *et al.*, 2005; Castillo Garcia FJ *et al.*, 2007). The prevalence of faecal carriage was 1.4% in York (UK) in 2003, 7 2.4% in Lebanon (Moubareck C *et al.*, 2005) and 7% in India (Rodrigues C *et al.*, 2005). A higher rate was found in Saudi Arabia (15.4%) (Kader AA *et al.*, 2007).

This study showed a very high carrying rate (80%) of these ESBL-producing enterobacteria. This is similar to the findings from Luvsansharav U *et al.* in Japon (69.3%) in 2012 on Prevalence of and risk factors associated with faecal carriage of CTX-M b-lactamase-producing Enterobacteriaceae in rural Thai communities (Luvsansharav U *et al.*, 2012). A high prevalence above 50% has also been observed among healthy individuals (57%) for *Escherichia coli* in a study conducted by Saleem *et al.* in 2017 in Pakistan. On the other hand, studies conducted by Michaud *et al.* in Senegal in 2014 among farm workers showed a low digestive carrying rate (9.4%). This high frequency of ESBL-producing enterobacteria in our study could be explained by permanent contact with the animals and their unhealthy working environment. It may be correlated to the poor hygienic measures observed in our study. This situation reveal a veritable problem of public health.

Escherichia coli is the most isolated ESBL producing enterobacteria with 78.18% followed by *Klebsiella pneumoniae* 16.36%). This is similar to the findings from a study conducted in Thailand on faecal specimens from the healthy individuals reported that majority of the isolates were ESBL-producing *Escherichia coli* (85.1%) (Sasaki T. *et al.*, 2010). This is confirmed by the work of Toudji *et al.* in 2017 which also reported *Escherichia coli* 51.13% as the majority strain of isolated ESBL followed by *Klebsiella pneumoniae* 30.10%. This result is far from the findings from poultry workers in the Federal Capital Territory, Abuja, Nigeria which reported low prevalence of ESBL producing *E. coli* (16,7%) (Aworh M. K. *et al.*, 2019). It is the same for findings from a survey of households and chicken farms in the Mekong Delta in Vietnam with Nguyen V. T *et al.* who reported a very low prevalence of ESBL producing *E. coli* (3.2%).

Our study showed an association of two bacteria producing of ESBL in two slaughterhouses areas particularly the slaughter area of Agoè Zongo which gathered 71,43% (5/7) of the associations.

Factors associated with ESBL carrying among slaughterhouse workers in our study were the age under 30 years, singles, the duration of work under 10 years, the use of only borehole water on the site, the lower work exposure, hospitalization in the past year and self-medication with antibiotics.

High consumption of antibiotics in self-medication may be a risk factor for the high prevalence of ESBL-producing Enterobacteriaceae. In this regard, previous studies have reported correlations between antimicrobial resistance and antibiotic usage (Goossens *et al.*, 2005; Tian *et al.*, 2008).

Our risk factor analyses show that even in the community setting, duration of work and self-medication with antibiotics have a major contribution to the faecal carriage of resistant bacteria. A history of hospitalization in the past year, the age, the marital status, the source of water use on the site and work exposure indicated higher risks of carrying ESBL-producing Enterobacteriaceae. Our study showed that the lower work exposition with animals and carcass like tax collection and safety guardian is associated with ESBL carriage. This is different from findings from Dohmen *et al.* which study revealed that slaughterhouse workers were more likely to carry ESBL when working in the early slaughtering steps (before chilling of the pig carcasses) than slaughterhouse workers working from this slaughter step forward, i.e. working in the cooling, cutting and deboning area (Dohmen *et al.*, 2017).

Besides the describe risk factors, none of the analyzed determinants such as education level, antibiotics use in the previous three months were found to be risk factors for ESBL carriage in slaughterhouse workers.

Some studies showed that prior hospitalization or previous use of antimicrobial drugs was irrelevant for the faecal carriage of ESBL-producing Enterobacteriaceae in healthy people (). Our results regarding hospitalization aspect for faecal carriage differ from previous findings.

Hospitalization and the use of antimicrobials, which have been described as possible risk

factors in previous studies (Otter JA et al. 2019; Sogaard M et al., 2017), is confirmed in our study and show an association with ESBL-carriage.

Our study also observed that ESBL producing *Escherichia coli* isolates from slaughterhouse workers showed high resistance rates for sulfamethoxazole-trimethoprim, chloramphenicol, nalidixic acid and fluoroquinolones, however, we observed low resistance rates for gentamicin 9,30% and susceptibility à 100% for Amikacin and Fosfomycin. *Klebsiella pneumoniae* strains isolated shown high resistance for gentamicin sulfamethoxazole-trimethoprim, chloramphenicol, fosfomycin, nalidixic acid and fluoroquinolones. Studies of Saleem R. et al. in Pakistan have shown that extended-spectrum β -lactamases (ESBL)—producing *Escherichia coli* are resistant to several antibiotics especially penicillins and cephalosporins however, they are susceptible to cephamycins and carbapenems (Saleem R. et al., 2017). This can be confirmed by our study where all strains studied were 100% sensitive to imipenem, ertapenem. *Escherichia coli* strains were 97.67% sensitive to cefoxitin, and *Klebsiella pneumoniae* were 100% sensitive. A research done on Parisian checkup centre showed the sensitivity pattern in which none of the ESBL-producing *E. coli* isolate was resistant to piperacillin-tazobactam, imipenem or amikacin (Chanoine M. et al., 2013). As these bacteria are multi-resistant to several families of antibiotics, an infection with these germs would be difficult to treat and would force the use of the latest generation of antibiotics such as carbapenems and also cephamycin.

4. CONCLUSION

This study carried out in three main slaughterhouses in the city of Lomé revealed a high rate of carrying of multi-resistant enterobacteria, mainly ESBL producing *E. coli*, among the staff of these workplaces. This confirms the existence of a health risk in terms of biosecurity and biosafety. It is urgent to take steps to promote good animal husbandry practices for animal welfare and good hygiene practices in Togo's slaughterhouses.

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