

# Antimicrobials Resistance of Bacteria Isolated from Three Bathing Waters in Southern Côte d'Ivoire

Thérèse Kouassi-Agbessi<sup>1,2,\*</sup>, Timothée Ouassa<sup>2,3</sup>, Anderson Richmond Djatchi<sup>2,3</sup>,  
Yessé Nanga Zinzendorf<sup>2,4</sup>, Aubin Tchapelé Gbagbo<sup>1</sup>, Christophe N'cho Amin<sup>1,2</sup>

<sup>1</sup>Laboratory of Water and Food Analysis, National Institute of Public Health, Abidjan, Côte d'Ivoire

<sup>2</sup>Department of Pharmaceutical and Biological Sciences, Félix Houphouët-Boigny University, Abidjan, Côte d'Ivoire

<sup>3</sup>Center for Diagnosis and Research on AIDS, University Hospital Center of Treichville, Abidjan, Côte d'Ivoire

<sup>4</sup>Laboratory of Microbiology, National Laboratory of Public Health, Abidjan, Côte d'Ivoire

## Email address:

aktherese@hotmail.com (T. Kouassi-Agbessi), timoth2@yahoo.com (T. Ouassa), djatchirichmond@yahoo.fr (A. R. Djatchi),  
dorfange@gmail.com (Y. N. Zinzendorf), aubintg2007@yahoo.fr (A. T. Gbagbo), christophe.amin@gmail.com (C. N'cho Amin)

\*Corresponding author

## To cite this article:

Thérèse Kouassi-Agbessi, Timothée Ouassa, Anderson Richmond Djatchi, Yessé Nanga Zinzendorf, Aubin Tchapelé Gbagbo, Christophe N'cho Amin. Antimicrobials Resistance of Bacteria Isolated from Three Bathing Waters in Southern Côte d'Ivoire. *Frontiers in Environmental Microbiology*. Vol. 5, No. 4, 2019, pp. 92-99. doi: 10.11648/j.fem.20190504.12

Received: October 2, 2019; Accepted: October 21, 2019; Published: October 25, 2019

**Abstract:** Waters used for recreational activities in order to ensure the health of populations who practice their recreational activity. Disease or infection risk associated with recreational water areas is mainly related either to faecal contamination or to domestic or hospital effluents. The present study aimed assessment of microbiological contamination of surface waters (rivers and lake) of three cities in southern Côte d'Ivoire used for recreational activities, by bacteria generally found in humans and animal's digestive tract, and then assesses their resistance to commonly antimicrobial drugs used in human therapy. Water samples have been taken from different identified sites over a 13-months period. A microbiological analysis based on numbering of germs was performed then isolated strains were assayed for antibiotic sensitivity tests. Data analysis was performed using SPSS 22.0 software. Results have shown the presence of *Escherichia coli*, *Pseudomonas aeruginosa* and *Enterococcus faecalis* in waters analysed. *E. coli*, a faecal contamination marker, was present in all the analysed waters with levels higher than standards defined by the 2006/7 /EC European Directive for bathing water, making them unsuitable for swimming. *E. coli* strains showed high levels of resistance to amoxicillin, ticarcillin, nalidixic acid and cotrimoxazole. Moreover, a beta-lactamase producing strain was isolated, suggesting probability of contamination by hospital or domestic effluents. The results of this study show the importance of microbiological monitoring of surface.

**Keywords:** Antimicrobial Resistance, Bathing Waters, Faecal Contamination, Côte d'Ivoire

## 1. Introduction

Stakes related to water quality have always been a challenge for human beings who may be exposed to water microbial pollution through drinking water but also through aquatic recreational activities. Faecal contamination of recreational waters may expose to various pathogens, including bacteria, parasites and viruses, for human populations [1]. During the nineteenth century, waterborne diseases were responsible for large epidemics of dysentery, typhoid fever, cholera, among others [2].

In recent years, several studies in Europe and North America have demonstrated antibiotic-resistant bacteria or genes encoding resistance in different aquatic environments [3-9]. In Gabon, a phenotypic study of bacteria isolated from the waters of lakes and rivers has revealed in these bacteria the phenomenon of multi-resistance to antibiotics [10]. Other studies in hospital effluents in both developed and developing countries have also highlighted this multi-resistance phenomenon and the presence of extended spectrum

beta-lactamase-producing Enterobacteriaceae (ESBLs) [11-13]. A public health problem arises because man, in the different environments where he lives, can be exposed to these multi-resistant bacteria, especially through water. The presence of these multi-resistant bacteria in the environment could lead to the transfer of resistance genes to bacteria in the environment, thus causing an even greater spread of these antibiotic resistances. Moreover, the presence of antibiotic-resistant pathogens in surface waters increases the risks incurred during a waterborne infection since the therapeutic possibilities are diminished. In Côte d'Ivoire, studies carried out on the quality of surface water have shown that these waters are polluted by human activities (agriculture, industry) [14-16]. Specifically, however, few studies have focused on the microbiological quality of recreational waters [17].

The aim of this study was to research for multidrug-resistant bacteria in surface waters used for recreational purposes in southern Côte d'Ivoire and to determine their phenotypes of resistance to some antibiotics.

## 2. Materials and Methods

### 2.1. Material

#### 2.1.1. Study Site

This experimental study was conducted on recreational surface waters in southern of Côte d'Ivoire (Abidjan, Jacqueville and Agboville) from December 2017 to December 2018. The sampling sites were located according to GPS apparatus. In Abidjan, were concerned: the waters of the Banco (longitude: 05°21'697", latitude: 004°21'532"), in Jacqueville and Agboville, these are the waters of the lake (longitude: 004°24'730", latitude: 05°12'222") and the river located in the Moutcho neighbourhood (longitude: 05°57'610", latitude: 004°11'610") which were concerned.

#### 2.1.2. Analytical Materials

The main equipment consists of:

1. a membrane filtration device (Sartorius, France),
2. a benchtop autoclave (Certoclav connect, France),
3. a water bath (Raypa, Spain),
4. Three incubators (Selecta, Spain) set at 30°C, 37°C and 44°C.

Various consumables were used including:

1. 0.45 µm filtrating membranes (Merck, Germany),
2. 45 mm and 90 mm petri dishes (Corning, France),
3. sterile graduated pipettes of 1ml, 2ml, 5ml, 10ml (Deltalab, Espagne),
4. sterile Pasteur pipettes (Marque, France),
5. sterile swabs (Citoswab, Chine),
6. hemolysis tubes,
7. Antibiotic discs (Bio-Rad, France).

The culture media used were as follows:

1. Rapid'E. coli 2 Medium (Bio-Rad, France),
2. BEA (Bile Esculin Azide) agar (Bio-Rad, France),
3. Pseudosel (Liofilchemr, Italy) agar for the enumeration of bacteria,
4. Mueller-Hinton agar (MH) (Bio-Rad®, France) to perform the sensitivity tests.

### 2.2. Methods

#### 2.2.1. Sampling

Sampling campaigns were carried out on the three water sources from December 2017 to December 2018 with a monthly withdrawal per source of water. Samples were taken according to WHO / UNEP recommendations. The water samples were collected at 0.5 meter from the surface of the water body in 500 ml sterile glass flasks of and transported to the laboratory in a cooler containing icebox to maintain the cold chain and a thermometer to guarantee the compliance with the temperature range from + 4°C to + 8°C. Analyses were carried out within the following four hours after sampling. Bacterial strains were stored at -20°C until the day of sensitivity testing.

#### 2.2.2. Enumeration of Bacteria

Bacteria was isolated from the samples according to the membrane filtration method [18]. A hundred of milliliters (100 ml) of water sample were filtered on a membrane. The membrane was then placed on a petri dish containing the corresponding culture medium and was incubated for 18 to 24 hours at 30°C for total coliforms, 37°C for *Pseudomonas aeruginosa* and 44°C for thermo-tolerant coliforms and enterococci. The identification of coliform colonies was based on biochemical characters on the reduced Le Minor gallery.

#### 2.2.3. Study of the Antibiotics Sensitivity

Sensitivity tests of bacteria to antimicrobials were performed using diffusion method of in gelled media of antibiotic-laden disc at known concentration from Kirby-Bauer, applied on Mueller-Hinton agar according to the standards of Antibiogram Committee of French Society of Microbiology (CASFM) in 2019. For the detection of extended-spectrum beta-lactamase (ESBLs) phenotype, third (3rd) generation of cephalosporin (cefotaxime (CTX), ceftazidime (CAZ) and cefepime (FEP) and aztreonam (ATM) were placed at 1.5 cm from clavulanic acid-associated amoxicillin (AMC) disc. A synergistic image shows the production of beta-lactamase. The quality control of the antibiograms was carried out using reference strains: *Escherichia coli* ATCC 25922, *P. aeruginosa* ATCC 27853 and *Enterococcus faecalis* ATCC 29212. The list and corresponding load of antibiotics tested are recorded in the tables 1, 2 and 3.

**Table 1.** List of antibiotic discs tested on *Enterobacteriaceae*.

Antibiotics (load in µg)	Abbreviations	S ≥	I	R <
Amoxicillin (20)	AMX	19	-	19
Amoxicillin + clavulanic acid (20/10)	AMC	19	-	19
Ticarcillin (75)	TIC	23	-	20
Cefoxitin (30)	FOX	19	-	15
Cefuroxime (30)	CXM	19	-	19
Cefotaxime (30)	CTX	20	-	17
Ceftazidime (10)	CAZ	22	-	19
Cefepime (30)	FEP	27	-	21
Aztreonam (30)	ATM	26	-	11
Imipenem (10)	IMP	22	-	16
Amikacin (30)	AKN	16	-	13
Netilmicin (30)	NET	15	-	12
Gentamicin (15)	GMI	17	-	14
Co-trimoxazole (Trimethoprim+Sulfamethoxazole (1,25/23,75))	SXT	14	-	11
Nalidixic acid (30)	NAL	14	-	14
Ciprofloxacin (5)	CIP	26	-	24
Levofloxacin (5)	LVX	23	-	19

**Table 2.** List of antibiotics tested on *P. aeruginosa*.

Antibiotics (load in µg)	Abbreviations	S ≥	I	R <
Ticarcillin (75)	TIC	18	-	18
Ticarcillin + clavulanic acid (75/10)	TCC	18	-	18
Ceftazidime (10)	CAZ	16	-	16
Cefepime (30)	FEP	19	-	19
Aztreonam (30)	ATM	25	-	22
Imipenem (10)	IMP	20	-	17
Amikacin (30)	AKN	18	-	15
Netilmicin (30)	NET	12	-	12
Gentamicin (15)	GMI	15	-	15
Ciprofloxacin (5)	CIP	26	-	26
Levofloxacin (5)	LVX	22	-	22
Co-trimoxazole (Trimethoprim+Sulfamethoxazole (1,25/23,75))	SXT	14	-	11

**Table 3.** List of antibiotic discs tested on *E. faecalis*.

Antibiotics (load in µg)	Abbreviations	S ≥	I	R <
Ampicillin (10)	AMP	10	-	8
Imipenem (10)	IMP	21	-	18
Gentamicin (15)	GMI	15	-	15
Doxycycline (30)	DOX	18	-	15
Erythromycin (15)	ERY	23	-	14
Streptomycin (500)	STR	14	-	14
Vancomycin (30)	VAN	12	-	12
Co-trimoxazole (Trimethoprim+Sulfamethoxazole (1,25/23,75))	SXT	14	-	11

#### 2.2.4. Statistical Analysis

Data entries were done on Excel 2013 and statistical analysis of data was realized using SPSS 22.0 software.

## 3. Results and Discussion

### 3.1. Results

#### 3.1.1. Distribution of Bacterial Strains

Samples were taken from 39 freshwater samples. After enumeration, 39 were positive for thermo-tolerant and *Enterococcus faecalis*, and 10 for *Pseudomonas aeruginosa*. After transplanting, 26 strains of *E. coli*, 13 non-*E. coli* Enterobacteria, 10 *P. aeruginosa* and 10 *E. faecalis* were positive. Table 4 shows the distribution of isolated bacteria in samples.

**Table 4.** Distribution of bacterial strains studied.

Strains	Frequency	Percentage (en %)
<i>E. coli</i>	26	44.0
Enterobacteriaceae none <i>E. coli</i>	13	22.0
<i>P. aeruginosa</i>	10	17.0
<i>E. faecalis</i>	10	17.0
Total	59	100.0

#### 3.1.2. Enumeration of Bacteria

The means of microbial densities obtained in Banco waters were 6,364 CFU/100 mL for thermo-tolerant coliforms, 5,518 CFU/100 mL for *E. coli*, 9,115 CFU/100 mL for *E. faecalis* and 158 CFU/100 mL for *P. aeruginosa*. These mean densities were respectively 4,885 CFU/100 mL for thermo-tolerant coliforms, 4,315 CFU/100 mL for *E. coli*, 4,548 CFU/100 mL for *E. faecalis* and almost none for *P. aeruginosa* in the

Moutcho River while they were 1,616 CFU/100 mL for thermo-tolerant coliforms, 1,543 CFU/100 mL for *E. coli* 1,048 CFU/100 mL for *E. faecalis* and almost none for *P. aeruginosa* in lake waters.

### 3.1.3. Antimicrobial Sensitivity Test

The study of antibiotic sensitivity was performed on bacterial strains isolated from the different sampling sites and the results of the sensitivity tests were recorded in tables 5, 6 and 7.

#### i. Antimicrobial Sensitivity of *Enterobacteria*

Table 5 revealed a high sensitivity of enterobacteria to most of the first and second generation betalactamins with levels

generally higher than 95.0%, unlike amoxicillin and ticarcillin which have considerable resistance rates especially for *Enterobacteriaceae* different from those of *E. coli*.

On the regard of sensitivity to other antibiotics, a very good sensitivity of all enterobacteria to aminoglycosides had been observed with sensitivity percentages ranking from 96.2 to 100.0%. Quinolones also showed very good activity on *Enterobacteriaceae* other than *E. coli*. However, slight resistance of *E. coli* was observed with resistance levels of 23.1% and 11.5% respectively for nalidixic acid and ciprofloxacin.

Table 5. Distribution of enterobacteria according to their sensitivity to antibiotics.

Disc of antibiotics	<i>Escherichia coli</i> (n=26)		<i>Enterobacteriales non Escherichia. Coli</i> (n=13)	
	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)
Amoxicillin	13 (50.0)	13 (50.0)	0 (0)	13 (100)
Ticarcillin	16 (61.5)	10 (38.5)	1 (7.7)	12 (92.3)
Cefuroxime	25 (96.2)	1 (3.8)	13 (100)	0 (0)
Cefoxitin	22 (84.6)	4 (15.4)	10 (76.9)	3 (23.1)
Cefotaxime	25 (96.2)	1 (3.8)	13 (100)	0 (0)
Ceftazidime	25 (96.2)	1 (3.8)	13 (100)	0 (0)
Cefepime	25 (96.2)	1 (3.8)	13 (100)	0 (0)
Aztreonam	25 (96.2)	1 (3.8)	13 (100)	0 (0)
Imipenem	26 (100)	0 (0)	13 (100)	0 (0)
Amoxicillin+Clavulanic acid	22 (84.6)	4 (15.4)	10 (76.9)	3 (23.2)
Amikacin	26 (100)	0 (0)	13 (100)	0 (0)
Gentamicin	25 (96.2)	1 (3.8)	13 (100)	0 (0)
Netilmicin	26 (100)	0 (0)	13 (100)	0 (0)
Nalidixic acid	20 (76.9)	6 (23.1)	12 (92.3)	1 (7.7)
Levofloxacin	23 (88.5)	3 (11.5)	13 (100)	0 (0)
Ciprofloxacin	23 (88.5)	3 (11.5)	13 (100)	0 (0)
Co-trimoxazole	20 (76.9)	6 (23.1)	10 (76.9)	3 (23.1)

#### ii. Antimicrobial Sensitivity of *P. aeruginosa*

*P. aeruginosa* sensitivity test showed variability in betalactamin resistance with resistance ratio of 30.0% and 100% respectively for aztreonam and ticarcillin. No resistance was observed with imipenem. Similar to aminoglycosides such as netilmicin, amikacin and gentamicin that was tested no resistance was observed with quinolones. In contrast, all strains of *P. aeruginosa* were resistant to the sulfamethoxazole-trimethoprim combination (table 6).

Table 6. Sensitivity of *P. aeruginosa* strains to antibiotics.

Disc of antibiotics	<i>P. aeruginosa</i> (n=10)	
	Sensitive (%)	Resistant (%)
Ticarcillin	0 (0.0)	10 (100.0)
Ticarcillin + clavulanic acid	10 (100.0)	0 (0.0)
Ceftazidime	10 (100.0)	0 (0.0)
Cefepime	10 (100.0)	0 (0.0)
Aztreonam	7 (70.0)	3 (30.0)
Imipenem	10 (100.0)	0 (0.0)
Amikacin	10 (100.0)	0 (0.0)
Gentamicin	10 (100.0)	0 (0.0)
Netilmicin	10 (100.0)	0 (0.0)
Ciprofloxacin	10 (100.0)	0 (0.0)
Levofloxacin	10 (100.0)	0 (0.0)
Co-trimoxazole	0 (0.0)	10 (100.0)

#### iii. Antimicrobial Sensitivity of *E. faecalis*

Table 7 showed that all *E. faecalis* strains tested had good susceptibility to penicillins, aminoglycosides and fluoroquinolones, but these strains were all resistant to co-trimoxazole.

#### iv. Phenotypes of resistance to beta-lactams

*E. coli* strains producing a low-level penicillinase showing acquired resistance phenotype were the most found with a ratio of 30.8%. However, wild-type strains were the most dominant at 42.3% (table 8).

Concerning *P. aeruginosa*, only high level penicillinase phenotypes and impermeability were found with respectively 69.2% and 30.8% (table 8).

Table 7. Sensitivity of *E. faecalis* strains to antibiotics.

Disc of antibiotics	<i>E. faecalis</i> (n=10)	
	Sensitive (%)	Resistant (%)
Ampicillin	10 (100.0)	0 (0.0)
Imipenem	10 (100.0)	0 (0.0)
Gentamicin	10 (100.0)	0 (0.0)
Co-trimoxazole	0 (0.0)	10 (100.0)
Doxycycline	5 (50.0)	5 (50.0)
Erythromycin	8 (80.0)	2 (20.0)
Vancomycin	10 (100.0)	0 (0.0)
Streptomycin	10 (100.0)	0 (0.0)

Table 8. Distribution of resistance phenotypes to beta-lactams.

Bacteria	Phenotype	Frequency	Percentage (%)
<i>E. coli</i>	Wild strain	11	42.3
	Low-level Penicillinase	8	30.8
	Others	5	19.2
	IRT*	1	3.8
	ESBL**	1	3.8
	Total	26	100
<i>P. aeruginosa</i>	High-level Penicillinase	7	70.0
	Impermeability	3	30.0
	Total	10	100.0

\*IRT: Inhibitor Resistant TEM, \*\*ESBL: Extended-Spectrum Beta-Lactamase

### 3.2. Discussion

Coastal cities with their wonderful sea beaches, lagoon as well as river and lakes waters are places of entertainment attracting many people (different classes of the population) including tourists and local people living along or around these water bodies to engage in recreational activities such as walking, swimming, laundry. It's known that those located particularly in major cities may be subject to various anthropogenic pressures including the discharge of wastewater from human and/or animal origin and thus harboured bacteria, some of which may induce serious pathologies in users. Therefore, it is important for these waters used in recreational activities to show an acceptable sanitary quality for the preservation of population's health.

Thus, the present study consisted of the search for bacteria, including some indicators of faecal contamination, in various recreational surface waters in three cities of southern Côte d'Ivoire over a period of thirteen months and to determine their profile of resistance to antibiotics commonly used in therapeutics. To do this, water samples were taken at two rivers and one lake.

Microbiological counts in these waters showed high levels of thermo-tolerant coliforms, *E. coli* and *E. faecalis*. In Banco waters, mean levels were 6364 CFU/100 mL for thermo-tolerant coliforms, 5518 CFU/100 mL for *E. coli* and 9115 UFC/100 mL for *E. faecalis*; at the Moutcho River, the average levels of thermo-tolerant coliforms and *E. coli* were respectively 4885 CFU/100 mL and 4315 CFU/100 mL and 4548 CFU/100 mL for *E. faecalis*. These mean microbial levels were lower at the Lac Jacquerville level and were 1616 CFU/100 mL for thermo-tolerant coliforms, 1543 CFU/100 mL for *E. coli* and 1048 CFU/100 mL for *E. faecalis*, respectively. *P. aeruginosa*, present at low levels in Banco waters (158 CFU/100 mL), was almost absent at the Moutcho River and lake. These results are consistent with those of many authors who have found the presence of these indicator germs faecal contamination of human or faecal origin [10, 14, 19, 20].

European Directive 2006/7/EC on the management of the quality of bathing water, recreational waters stipulates that water is of insufficient quality if the concentrations of *E. coli* are higher than 500 CFU per 100 mL and the concentrations enterococci greater than 200 CFU per 100 mL, based on a 95th

percentile. As a result, compared with the *E. coli* concentrations of the waters analyzed above, the latter as a whole can be considered of "insufficient quality" and unsuitable for bathing because they are contaminated with bacteria indicating faecal contamination at low levels above the European standard. One of the direct consequences would be the risk of infection for users of these waters. This observation disagrees with the results of Mwanamokid *and al* in a study conducted in the DRC in 2014 on the evaluation of pathogenic bacteria in water and sediments of a water reservoir under tropical conditions (Lac Ma Vallée). In fact, they had shown that the lake water had an acceptable microbiological quality because it was moderately contaminated by the indicator bacteria of faecal contamination. However, even if these are in low concentrations in the water samples, their presence suggests the possible presence of pathogenic bacteria of intestinal origin [19, 21].

The bacterial isolation from the water samples revealed enterobacteria in which *E. coli* was the predominantly isolated species with a rate of 44.0%, strains of *P. aeruginosa* and *E. faecalis*. These results are noticeably similar to those generally observed in other studies, which, in addition to the Gram-negative bacilli commonly found in human pathology, have also isolated other bacteria such as Staphylococci and *Aeromonas* [10, 19, 22-24]. In two studies carried out in Côte d'Ivoire on hospital effluents in the city of Abidjan, the authors showed a strong predominance of bacteria of the family Enterobacteriaceae and Pseudomonadaceae [12, 13, 25]. The antibiotic susceptibility tests of these strains were made with different molecules presented in Tables 1, 2 and 3, in order to be able to identify the different acquired resistance phenotypes present in the latter. Note that antibiotics are a class of drugs widely used in human therapy, prevention and treatment in animals, for plant infection and other activities. Approximately 30-90% of the administered dose of most antibiotics to humans and animals is excreted via urine and faeces into the environment via domestic effluents [26-28]. Antibiotic susceptibility testing results in this study, dominated by 62.9% enterobacterial isolates, showed high susceptibility to second- and third-generation cephalosporins and aminoglycosides and fluoroquinolones. In addition, average resistance was noted for some antibiotics such as cotrimoxazole (23.1%) and nalidixic acid (23.1%). In contrast, this resistance was high for aminopenicillins

including amoxicillin (50.0%) and ticarcilin (38.5%). These observed results on bacteria isolated from surface waters for recreational use in southern Côte d'Ivoire are contrary to the results obtained by other authors who worked on surface water. These authors had for the most part observed a high resistance of the germs to antibiotics tested. Thus, in its phenotypic study of the resistance of bacteria isolated from lakes and rivers of the city of Franceville to third-generation cephalosporins, Yala *and al.* in 2017 showed a variable sensitivity towards third-generation cephalosporins, contrary to our study in which all cephalosporins tested had very low or even less than 5% resistance [10]. Enterobacterial strains resistance to aminopenicillin, nalidixic acid, and co-trimoxazole could be explained either by the selection pressure exerted by these antibiotics often prescribed in first line against some infections, or due to others molecules with antibacterial activity from hospital effluents, industrial, agricultural and domestic wastes. Indeed, various antibiotics and other molecules with antibacterial activity are often evacuated directly or after treatment in the environment especially in surface waters through sewers thus favoring emergence of multidrug-resistant bacteria [10, 29-31]. Of all isolated *E. coli* strains, one was a broad-spectrum beta-lactamase producer and eight low-level penicillinase producers. These results show that on a sanitary level, the contamination of recreational waters by hospital strains is possible and therefore the risk of contamination during swimming is likely. Guessennnd *and al.* in the study of hospital effluents in the city of Abidjan in 2013, showed the presence of extended spectrum beta-lactamase (ESBL) in all isolated enterobacterial strains [12]. In 2013, Cablan *and al.* in their study on the microbiological control of various effluents from the University Hospital of Treichville, found that ninety-three percent of enterobacteria were producing ESBL [13]. In a study conducted in Ireland, the ability of antimicrobial resistant *E. coli* to survive the wastewater treatment process of a modern secondary treatment facility was also shown, demonstrating its importance in the propagation of resistance genes antibiotics [11, 28]. This resistance could be correlated with the spread of antibiotics by hospital effluents and domestic wastewater over long distances in surface water. The exchange of genetic material that is well known in enterobacteria could promote the spread of this resistance to other germs.

Moreover, in this study, the presence of opportunistic pathogens such as *P. aeruginosa*, reveals the survival capacity of pathogens in the analyzed waters [19]. Of all *P. aeruginosa* strains, 100% were resistant to ticarcillin and 30.0% to aztreonam as some strains isolated from hospital effluents [12]. This population of *P. aeruginosa* consisted mainly of 70.0% high-level penicillinase-producing strains and 30.0% exhibiting a phenotype of impermeability. Aboulfotou Hashish *and al.* in 2017 in a study conducted in Egypt to evaluate the bacteriological quality, appearance and antimicrobial resistance of *P. aeruginosa* in swimming pools (competition and recreation), showed that three of the 26 isolates were sensitive to all antibiotics used and nine (34.6%) were multidrug-resistant *P. aeruginosa* strains. In

the study by Cablan *and al.* in 2015, all isolated strains of *P. aeruginosa* exhibited CASE HN, Imipenemase, PEF R and CIP R phenotypes. This indicates that *P. aeruginosa* strains in recreational waters can be multi-resistant, especially for those who bathe there [22, 32]. Enterococci are bacteria intrinsically resistant to a range of antibiotics which has always limited the choice of antibiotics used against these microorganisms; however, in this study, all strains of *E. faecalis* isolated were sensitive to ampicillin, vancomycin, gentamicin and streptomycin (Table 7). These results are close to those obtained by Nadia O. *and al.* in 2012. The absence of ampicillin resistance has been reported by M Teresa Tejedor *and al.* in 2001 for all enterococci isolated from the analyzed water samples; According to ANSES (2010), the enterococcal strains isolated from the different pathways showed low to no resistance to vancomycin and ampicillin. Clinically, the low level of resistance to gentamicin, ampicillin and vancomycin appears to be favorable, because resistance to these molecules significantly reduces therapeutic treatments in enterococcal infections [33]. The percentages of resistance against erythromycin and doxycycline were respectively 20.0% and 50.0%, which is close to the percentages of resistance reported by Paulo Martins *and al.* in 2006 found that 34.6% of enterococci were resistant to tetracycline and 24.8% to erythromycin in raw, purified water and sludge samples.

## 4. Conclusion

The present study performed on surface waters with recreational activity in the south of Cote d'Ivoire, showed the presence of faecal contamination indicator bacteria from human faecal material in these waters. This shows the unfit nature of these waters and the infection risk for users, especially since these waters are widely used for recreational activities.

The presence of opportunistic bacteria and the high resistance to certain antibiotics suggest a probable contamination by domestic or hospital wastewater effluents of these waters. This could contribute to the further spread of antimicrobial resistance and could lead to public health problems. All this shows the importance of monitoring the microbiological contamination of surface waters in our country and also determining the resistance levels of bacteria isolated from these waters.

It would be important to add to that, the monitoring of the sediments of rivers and lakes as well as the sands of recreational beaches. Indeed, some studies have shown that bacterial concentrations are higher in these areas than in the water and there for they constitute a reservoir of bacteria.

## Acknowledgements

The Laboratory is grateful for Mrs Alphonse N'gbakou and Paul Akié for their technical contributions.

## Conflict of Interest

There is no conflict of interest.

## References

- [1] Pond K. Water recreation and disease: plausibility of associated infections: acute effects, sequelae, and mortality. World Health Organization; 2005.
- [2] George I, Servais P. Sources and dynamics of coliforms in the Seine basin: Summary report. 2002.
- [3] Chee-Sanford JC, Aminov RI, Krapac I, Garrigues-Jeanjean N, Mackie RI. Occurrence and diversity of tetracycline resistance genes in lagoons and groundwater underlying two swine production facilities. *Appl Environ Microbiol.* 2001; 67 (4): 1494502.
- [4] Volkmann H, Schwartz T, Bischoff P, Kirchen S, Obst U. Detection of clinically relevant antibiotic-resistance genes in municipal wastewater using real-time PCR (TaqMan). *Journal of microbiological methods.* 2004; 56 (2): 27786.
- [5] Sayah RS, Kaneene JB, Johnson Y, Miller R. Patterns of antimicrobial resistance observed in *Escherichia coli* isolates obtained from domestic-and wild-animal fecal samples, human septage, and surface water. *Appl Environ Microbiol.* 2005; 71 (3): 1394404.
- [6] Antunes P, Machado J, Peixe L. Characterization of antimicrobial resistance and class 1 and 2 integrons in *Salmonella enterica* isolates from different sources in Portugal. *Journal of Antimicrobial Chemotherapy.* 2006; 58 (2): 297304.
- [7] Messi P, Guerrieri E, De Niederhausern S, Sabia C, Bondi M. Vancomycin-resistant enterococci (VRE) in meat and environmental samples. *International journal of food microbiology.* 2006; 107 (2): 21822.
- [8] Hamelin K, Bruant G, El-Shaarawi A, Hill S, Edge TA, Fairbrother J, Harel J, Maynard C, Masson L, Brousseau R. Occurrence of virulence and antimicrobial resistance genes in *Escherichia coli* isolates from different aquatic ecosystems within the St. Clair River and Detroit River areas. *Appl Environ Microbiol.* 2007; 73 (2): 47784.
- [9] Sidrach-Cardona R, Hijosa-Valsero M, Marti E, Balcázar JL, Becares E. Prevalence of antibiotic-resistant fecal bacteria in a river impacted by both an antibiotic production plant and urban treated discharges. *Science of the Total Environment.* 2014; 488: 2207.
- [10] Yala J-F, Mabika RM, Boucka DB, Lepengue AN, Souza A. Phenotypic study of the resistance of bacteria isolated from lakes and rivers of the city of Franceville to third-generation cephalosporins. *Journal of Applied Biosciences.* 2017; 119 (1).
- [11] Galvin S, Boyle F, Hickey P, Vellinga A, Morris D, Cormican M. Enumeration and characterization of antimicrobial-resistant *Escherichia coli* bacteria in effluent from municipal, hospital, and secondary treatment facility sources. *Appl Environ Microbiol.* 2010; 76 (14): 47729.
- [12] Guessennd N, Ouattara M, Ouattara N, Nevry R, Gbonon V, Tiekoura K, Dosso M, Ger B. Study of Multiresistant Bacteria in Hospital Effluents of a Hospital and University Center (CHU) in the City of Abidjan (Côte d'Ivoire). *Journal of Applied Biosciences.* 2013; 69: 545664.
- [13] Cablan MA, Nanga YZ, Kouassi-Agbessi T, Loukou YG, Kacou-N'douba A, Dosso M. Microbiological control of different effluents from the University Hospital Center of Treichville. [SPB]: FHB; 2015.
- [14] Ahoussi KE, Koffi YB, Kouassi AM, Soro G, Soro N, Biémi J. Study of the chemical and microbiological characteristics of the water resources of the N'zi watershed: case of the commune of N'zianouan (southern Côte d'Ivoire). *International Journal of Biological and Chemical Sciences.* 2012; 6 (4): 185473.
- [15] Sadat A, N'goran EBZ, Siaka S, Parinet B. Interest of the multidimensional analysis for the evaluation of the physicochemical quality of the water of a tropical lacustrine system: case of the Yamoussoukro lakes (Côte d'Ivoire). *Journal of Applied Biosciences.* 2011; 38: 257385.
- [16] Coulibaly A, Mondé S, Wognin V, Aka K. State of anthropic pollution in the estuary of Ebrié lagoon (Côte d'Ivoire) by analysis of the metal elements traces. *European Journal of Scientific Research.* 2008; 19 (2): 37290.
- [17] Amin NC, Agbessi T, Gbassi K, Dosso V, Kpaibe SP, Kouadio L. Quality control of the bathing water of the Cascade de Man in Ivory Coast. *Journal of Scientific Research of the University of Lome.* 2017; 19 (4): 8590.
- [18] APHA (2005) Standard Methods for the Examination of Water and Wastewater. 21st Edition, American Public Health Association/American Water Works Association/Water Environment Federation, Washington DC.-References-Scientific Research Publishing [Internet]. [cité 26 sept 2019]. Disponible sur: [https://www.scirp.org/\(czech2tfqyw2orz553klw0r45\)\)/reference/ReferencesPapers.aspx?ReferenceID=1870039](https://www.scirp.org/(czech2tfqyw2orz553klw0r45))/reference/ReferencesPapers.aspx?ReferenceID=1870039).
- [19] Mwanamoki PM, Devarajan N, Thevenon F, Atibu EK, Tshibanda JB, Ngelinkoto P, Mpiana PT, Prabakar K, Mubedi JI, Kabele CG. Assessment of pathogenic bacteria in water and sediment from a water reservoir under tropical conditions (Lake Ma Vallée), Kinshasa Democratic Republic of Congo. *Environmental monitoring and assessment.* 2014; 186 (10): 682130.
- [20] Adingra A, Sako A, Kouassi A. Bacteriological quality of surface waters of Banco River, Abidjan, Côte d'Ivoire. 2010.
- [21] Haller L, Poté J, Loizeau J-L, Wildi W. Distribution and survival of faecal indicator bacteria in the sediments of the Bay of Vidy, Lake Geneva, Switzerland. *Ecological indicators.* 2009; 9 (3): 5407.
- [22] Tiroidimos I, Christoforidou EP, Nikolaidou S, Arvanitidou M. Bacteriological quality of swimming pool and spa water in northern Greece during 2011–2016: is it time for *Pseudomonas aeruginosa* to be included in Greek regulation? *Water Science and Technology: Water Supply.* 2018; 18 (6): 193745.
- [23] Jaiani E, Kokashvili T, Mitaishvili N, Elbakidze T, Janelidze N, Lashkhi N, Kalandadze R, Mikashvidze E, Natroshvili G, Whitehouse CA. Microbial water quality of recreational lakes near Tbilisi, Georgia. *Journal of water and health.* 2013; 11 (2): 33345.
- [24] Schwartz T, Kohnen W, Jansen B, Obst U. Detection of antibiotic-resistant bacteria and their resistance genes in wastewater, surface water, and drinking water biofilms. *FEMS microbiology ecology.* 2003; 43 (3): 32535.

- [25] Munoz-Aguayo J, Lang KS, LaPara TM, Gonzalez G, Singer RS. Evaluating the Effects of Chlortetracycline on the Proliferation of Antibiotic-Resistant Bacteria in a Simulated River Water Ecosystem. *Applied and Environmental Microbiology*. 1 sept 2007; 73 (17): 54215.
- [26] Kumarasamy KK, Toleman MA, Walsh TR, Bagaria J, Butt F, Balakrishnan R, Chaudhary U, Doumith M, Giske CG, Irfan S, Krishnan P, Kumar AV, Maharjan S, Mushtaq S, Noorie T, Paterson DL, Pearson A, Perry C, Pike R, Rao B, Ray U, Sarma JB, Sharma M, Sheridan E, Thirunarayan MA, Turton J, Upadhyay S, Warner M, Welfare W, Livermore DM, Woodford N. Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular, biological, and epidemiological study. *Lancet Infect Dis*. sept 2010; 10 (9): 597602.
- [27] Pharmacology/H. P. Rang [and others] ; illustrations by Peter Lamb.-Version details-Trove [Internet]. [cité 26 sept 2019]. Disponible sur: <https://trove.nla.gov.au/work/25740167?q&versionId=45535332>.
- [28] Densities and antimicrobial resistance of *Escherichia coli* isolated from marine waters and beach sands.-PubMed-NCBI [Internet]. [cité 26 sept 2019]. Disponible sur: <https://www.ncbi.nlm.nih.gov/pubmed/25963763>.
- [29] Weiss K. Bacterial resistance: the new cold war. *The Doctor of Quebec* 37 (3). 2002; 419.
- [30] Drieux-Rouzet L, Jarlier V. Multiresistant bacteria in water: models of enterobacteria producing extended spectrum beta-lactamase. *Francophone Journal of Laboratories*. 2014; 2014 (460): 759.
- [31] Harris S, Morris C, Morris D, Cormican M, Cummins E. Antimicrobial resistant *Escherichia coli* in the municipal wastewater system: effect of hospital effluent and environmental fate. *Science of the Total Environment*. 2014; 468: 107885.
- [32] Aboulfotoh Hashish NM, Gawad Abbass AA, Khamis Amine AE. *Pseudomonas aeruginosa* in swimming pools. *Cogent Environmental Science*. 2017; 3 (1): 1328841.
- [33] Klare I, Konstabel C, Badstübner D, Werner G, Witte W. Occurrence and spread of antibiotic resistances in *Enterococcus faecium*. *International journal of food microbiology*. 2003; 88 (23): 26990.