GSC Advanced Research and Reviews

eISSN: 2582-4597 CODEN (USA): GARRC2

Cross Ref DOI: 10.30574/gscarr

Journal homepage: https://gsconlinepress.com/journals/gscarr/

(RESEARCH ARTICLE)

Check for updates

Evaluation of environmental microorganism's anti-bioresistance from sewage collectors from Abobo to Vridi, in the District of Abidjan, Côte d'Ivoire

Ahua René KOFFI [1, *], Affoué Carole KOFFI [2], Marcelle Sandrine N'GUESSAN [3], Ahou Charlotte Nadège KOUADIO-N'GBESSO [4] and Adjéhi DADIE [5]

[1] *Ecology Research Center (CRE, Laboratory of Microbiology and Biotechnology, NANGUI ABROGOUA University Abidjan, Côte d'Ivoire.*

[2] *Training and Research Unit for Agronomic, Forestry and Environmental Engineering; Pedagogical Unit Biochemistry-Microbiology of the University of Man. Bp20 Man, Côte d'Ivoire.*

[3] *Bacteriology and mycology laboratory in Diagnosis and research on AIDS and other diseases center from Treichville University Hospital, Faculty of Biological Sciences, University of FELIX HOUPHOUËT BOIGNY, Abidjan Cocody, Côte d'Ivoire.*

[4] *Oceanological Research Center; microbiology laboratory Abidjan, Côte d'Ivoire.*

[5] *University of NANGUI ABROGOUA, Department of Food Science and Technology; Microbiology and Biotechnology Laboratory, Côte d'Ivoire.*

GSC Advanced Research and Reviews, 2022, 12(03), 081–086

Publication history: Received on 14 March 2022; revised on 16 July 2022; accepted on 18 July 2022

Article DOI: https://doi.org/10.30574/gscarr.2022.12.3.0056

Abstract

Objectives: The spread of bacterial resistance to antibiotics in the environment remains a major public health problem. Health centers, effluent treatment plants, livestock farms, households and collection centers are considered platforms for the transit and exchange of multi-resistant bacteria.

Methods: This study was conducted to evaluate the level of antibiotic resistance of microorganisms in the environment. It consisted in collecting samples of wastewater from evacuation and sanitation channels in the city of Abidjan. A total of 99 samples of 2970 mL of wastewater were collected and standard microbiology methods were implemented.

Results: At the end of the analysis, 215 coliforms of faecal or non-faecal, aquatic and telluric origin were identified. These microorganisms showed 55.81% resistance to β-lactams, mainly to amoxicillin, 7.67% to aminoglycosides, 6.05% to tetracycline and 3.49% to quinolones.

Conclusion: Environmental bacterial strains are more and more resistant to the usual antibiotics.

Keywords: Antimicrobial resistance; Coliforms; Environment; Public health; Wastewater

1. Introduction

Antibiotic resistance knows no boundaries or species barriers, whether bacterial or animal [1]. This is a particularly complex problem [2] because of the diversity of the actors involved (manufacturers, producers, consumers, bacteria, fungi and viruses), multiple influences that are exerted, including environment, to contribute to the expansion of the phenomena of resistance currently observed.

* Corresponding author: Ahua René Koffi

Ecology Research Center (CRE), Laboratory of Microbiology and Biotechnology, Nangui Abrogoua University Abidjan (Côte d'Ivoire);.

In fact, antibiotics not only act on the bacterium responsible for the infection to be treated, but also, for the majority of them, on the useful and non-pathogenic bacteria of our organism and the environment [3], [4]. All bacteria are thus likely to acquire new mechanisms of resistance to antibiotics, in addition to those that some of them naturally possess [5].

Antibiotic treatment, repeated or occasional, may lead to the emergence of resistant bacteria that may lead to treatments with subsequent less effective antibiotics, for patients, but also for the community when they diffuse into the environment and transmit to other patients [6].

Through this study, the aim was to assess the level of antimicrobial resistance of certain bacterial strains of the environment, isolated from the sewage disposal channels.

2. Methods

2.1. Wastewater treatment

Samples were taken at various sewage treatment in the sanitation section in Abidjan. These were the S7 Stations, at Abobo "SAMAKE", PK9 at Riviera 3, Aquarium at the Indénié in Adjamé, at the station of Pont De Gaulle in Plateau, at MU 8, 7J1, RPB1, MU 7, KS 1, in Koumassi and at Vridi-cité station (these names depend on the geographical location of the stations).

2.2. Sample size

Three samples of wastewater 30 mL were taken per station and per month over three (3) months; a total volume of wastewater were 2970 mL.

2.3. Sampling technique and transport

The wastewater samples were collected using a device with a 20 meter long rope attached to a sterile, glass container specific to each station. The samples were then transferred to 50 mL sterile plastic tubes, placed in a cooler containing with carboglasses and sent to the laboratory within two hours for analysis.

2.4. Determination of the concentration of microorganisms in a sample

The determination of the concentration of microorganisms on a culture medium was carried out by counting from a colony counter and the figure obtained is related to the volume of water analyzed according to the following steps:

- A colony culture in a petri dish is placed on the agar side, against a light source;
- Any colonies grown on the agar are counted;
- The number of colony obtained is related to the volume of water analyzed;

Thus for 5 ml of water analyzed after 24 hours, x colonies are obtained on the agar in question. The result of the enumeration performed is equal to x / 5 colonies per mL and expressed per colony forming unit per mL (CFU / mL).

2.5. Bacteriological analysis

The analysis of these samples was done in four stages for Enterobacteria: pre-enrichment, enrichment, isolation and identification.

2.5.1. Pre-enrichment

Each sample is homogenized, then 25 mL is aseptically removed and added to 225 mL of buffered peptone water (EPT) This 1/10 dilution of each sample is prepared by aseptically adding, manually, 225 mL of EPT to the sample previously measured unit of analysis. For each series of samples, a positive control and a blank are added. The positive control consists of a culture of *Salmonella Typhimurium* (for this bacteria) seeded in 225 mL of EPT. The negative control consists of 225 mL of EPS.

The samples are incubated at (35 ± 1) ° C for 18 to 24 hours.

2.5.2. Enrichment

Using a sterile Pasteur pipette, 0.1 mL of each pre-enrichment broth is transferred into 10 mL of Vassiliadis Soyapapad Rappaport (RVS), and 0.1 mL of the pre-enrichment culture in 9 mL of Tetrathionate supplemented with Bright Green (TBG); the whole is incubated at (42 ± 0.5) ° C for 24 ± 2 hours.

2.5.3. Isolation

SS selective agars, Bile Esculin Agar, Potassium Tellurite, Methyl Blue Eosin and Hecktoen are used. They are seeded by depletion streak technique from the same enrichment broth and are incubated for 24 hours in an oven at 37 ° C.

2.5.4. Identification

The identification of isolated strains is based on the biochemical characters displayed by each crop. For this purpose, a reduced rack of Le Minor which is incubated at 37 ° C for 24 hours.

2.5.5. Antibiotic resistance

The antimicrobial resistance of the microorganisms identified was carried out according to the Mueller-Hinton Agar agar diffusion method, for 16 antibiotic discs (BIORAD), according to the recommendations of the Committee of the antibiogram of the French microbiology company (CA- SFM, 2014).

3. Results and discussion

The enterobacteria isolated in this study number 172 and are listed in the table 1.

Table 1 Isolated enterobacteria

	Microorganisms	**Enterobacteria**	
		Isolated workforce (N=172)	**%**
1	*Citrobacter diversus*	5	2.91
2	*Citrobacter freundii*	18	10.47
3	*Enterobacter cloacae*	31	18.02
4	*Escherichia coli*	53	30.81
5	*Klebsiella ocytoca*	28	16.28
6	*Klebsiella pneumoniae*	13	7.56
7	*Klebsiella terrigina*	5	2.91
8	*Salmonella spp.*	14	8.14
9	*Serratia fonticola*	3	1.74
10	*Serratia odorifera*	2	1.16

Table 2 Non-Enterobacteria isolated

N°	**Microorganisms**	**Non-Enterobacteria**	
		Isolated workforce (N= 43)	**%**
1	*Enterococcus faecalis*	9	20.93
2	*Enterococcus* spp.	13	30.23
3	*Pseudomonas aeruginosa*	12	27.91
4	*Staphylococcus aureus*	9	20.93

The work carried out has shown non-enterobacterial microorganisms, they are microorganisms naturally present in the environment Table 2.

The microorganisms isolated in this study fall into two groups: enterobacteria and non-enterobacteria. *E coli* strains belonging to the enterobacteria group are not naturally present in the environment; they are microorganisms of the digestive tract of humans and warm-blooded animals [7]. The results of our study show a remarkable presence (53%) of these microorganisms in samples taken from the environment. This is the same observation made by [8], and it showed that the contamination of the urban environment by *E coli* resulted from the presence mainly of wastewater and domestic waste. This could mean that the presence of a source responsible for faecal contamination of the sewage channels. At the level of treatment plants, the treatments applied to eliminate microorganisms have an impact on the survival of certain microorganisms including *E. coli* [9]. *E coli* strains can be detected up to 13 months in wastewater and in soils near sewage treatment plants [10]. The presence of *E. coli* remains an important indicator of water quality analysis.

The level of resistance expressed by the microorganisms of the family of β-lactams, aminoglycosides, quinolones sulfonamides, diaminopyrimidine, polypeptides and tetracycline are recorded in the table 3.

Table 3 Characteristics of microorganism's vis-à-vis the usual antibiotics

Family	Antibiotics	% Enterobacteriaceae (N= 172)	% Non-Enterobacteriaceae (N=43)	% Total number (N= 215)
β-lactamines	Amoxicilline	53.49	65.12	55.81
	Amoxicilline + acide clavulanique	44.77	48.84	45.58
	Ticarcilline	43.60	46.51	44.19
	Cefalotine	39.53	41.86	40.00
	Cefoxitne	31.98	53.49	36.28
	Cefuroxine	37.79	44.19	39.07
	Ceftriazone	12.21	25.58	14.88
	Aztreonam	8.72	32.56	13.49
	Imipenem	14.53	30.23	17.67
Aminosides	Kanamycine	5.23	16.28	7.44
	Gentamycine	6.40	11.63	7.44
	Tobramycine	7.56	4.65	6.98
	Netilmycine	5.81	20.93	8.84
Quinolones	Acide Nalidixique	1.74	4.65	2.33
	Ciprofloxacine	4.07	6.98	4.65
Sulfamindes et diaminopyrimidine	Cotrimoxazole	2.33	4.65	2.79
Polypeptides	Colistine	1.16	2.33	1.40
Tetracycline	Tetracycline	5.23	9.30	6.05

A part from *E. coli*, several other germs in the Enterobacteriaceae group have been isolated from environmental samples.

The remarkable presence of *P aeruginosa* (27.91%) in the environmental samples is a sign of the proximity of our study sites to hospitals or the expression of the discharge of hospital effluents into the collection pipes. Wastewater. Because

P aeruginosa although present in the environment, is much more of hospital origin. The analysis of the environmental samples also revealed the presence of *Enterococcus spp* (30. 23%) strains of which 20. 93% are *E faecalis*. This presence of enterococci is a sign of faecal contamination of wastewater; better the drainage channels of these waters could be connected to septic tanks or latrines.

Different microorganisms from the non-enterobacteria group were isolated during this study.

The overall level of resistance of microorganisms isolated from wastewater is presented and expresses a specificity related to a level of resistance to β-lactams. These antibiotics, which according to [11] are widely used in human therapies, have within their chemical structure a β-lactam nucleus that enables them to act on the bacterial wall by blocking the transpeptidation process. Unfortunately the abuse and bad practices of antibiotic therapy have led microorganisms to develop resistance to these molecules.

4. Conclusion

The wastewaters collected in the evacuation channels of the District of Abidjan, contain countless microorganisms, some of which have been identified as carriers of resistance; thus confirming the importance of the level of resistance of these microorganisms in the environment. This represents a public health risk to populations. In addition, these microorganisms that were previously sensitive to antibiotics such as β-lactams, aminoglycosides and quinolones used in human therapy have acquired forms of high and disturbing resistance.

This is the place to challenge the competent health authorities on arrangements to guarantee a rational use in order to preserve the quality of the environment.

Compliance with ethical standards

Acknowledgments

Our thanks to the Ivorian Company of Water Distribution (SODECI), to NANGUI ABROGOUA University (UNA) and the Center for Diagnosis and Research on AIDS and other diseases (CeDReS/University Hospital of Treichville).

Disclosure of conflict of interest

No conflict of interest.

References

[1] Pénali L, Guessend N, Dosso M. Surveillance of bacterial resistance to antimicrobials. Pasteur Institute experience in Côte d'Ivoire AFRICA CDC MEETING. 2017; 30.

[2] ANSES (National Agency for Food Safety Safety, work environment). Antibiotic consumption and antibiotic resistance in France: Need for a determined and sustainable mobilization. 2016; 30.

[3] Toitot C, Adebiotech MVL. Symposium Report: Antibiotic Resistance: An Integrated Approach from the Environment to Man. Biocitech, 16-17 mars, Romainville. 2016; 12.

[4] Ouédraogo AS, Jean-Pierre H, Banuls AL, Ouédraogo R, Godreuil S. Emergence and diffusion of antibiotic resistance in West Africa: contributing factors and threat assessment. Medicine and Tropical Health. 2017; 27: 147-154.

[5] Bouchaud O, Blanchy S, Delmont J, Gay-andrieu F. Antibiotic resistance: a global problem. Society of Exotic Pathology (SPE); SPILF Conference. 2013; 13.

[6] Carle S. Antibiotic resistance: an important public health issue! Pharmactuel. 2009; 42(2): 16.

[7] CEAEQ (Center of Expertise in Environmental Analysis of Quebec). Simultaneous search and enumeration of total coliforms and *Escherichia coli* in drinking water with MI culture medium; Membrane filtration method, MA.700-Ecctmi 1.0, Rev.3, Ministry of Sustainable Development, Environment and Climate Change, 2015; 19.

[8] Balière C. *Escherichia coli* potentially pathogenic in the coastal environment: the case of STEC and EPEC. Microbiology and Parasitology. University of Western Brittany - Brest, 2016.

[9] Marin I, Goni P, Lasheras AM, Ormad MP. Efficiency of a Spanish wastewater treatment plant for removal potentially pathogens: Characterization of bacteria and protozoa along water and sludge treatment lines. Ecological Engineering. 2015; 74: 28-32.

[10] Yang K, Pagaling E, Yan T. Estimating the Prevalence of Potential Enteropathogenic *Escherichia coli* and Intimin Gene Diversity in a Human Community by Monitoring Sanitary Sewage. *Applied and Environmental Microbiology.* 2014; 80(1): 119-127.

[11] Mainardi J-L, Villet R, Bugg TD, Mayer C, Arthur M. Evolution of peptidoglycan biosynthesis under Gram-positive antibiotics bacteria. *FEMS Microbiology Reviews.* 2008; 32: 386–408.