

ORIGINAL

Rivers and pools in Tungurahua- Ecuador, are contaminated with superbugs: an alarming reality

Ríos y piscinas de Tungurahua-Ecuador contaminados con superbacterias, una realidad alarmante

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ABSTRACT

Introduction: the pollution of both stagnant and flowing water bodies has emerged as a pressing threat to public health and ecosystems. Freshwater sources are increasingly degraded by unchecked biological, industrial, and agricultural waste, fostering the spread of multidrug-resistant bacteria—colloquially termed “superbugs.” Among these, extended-spectrum beta-lactamase (ESBL)-producing strains are particularly alarming. Their resistance to first-line antibiotics complicates treatment of gastrointestinal, urinary, respiratory, and skin infections, driving rising morbidity and mortality rates across Tungurahua Province. This study aimed to detect superbugs in rivers and swimming pools throughout Tungurahua and evaluate their antimicrobial resistance patterns.

Method: we conducted a descriptive field study combining qualitative and quantitative analyses. Water samples were collected from five rivers and twenty-five swimming pools using non-probabilistic sampling.

Results: while fecal coliform levels in rivers complied with limits set by Ecuador’s TULSMA regulations, swimming pools consistently exceeded thresholds. ESBL-producing *Klebsiella* and *E. coli* were isolated from rivers, whereas pools harbored a broader range of resistant pathogens, including *Escherichia coli*, *Klebsiella*, *Acinetobacter*, and *Pseudomonas*. These bacteria demonstrated resistance to cephalosporins and aztreonam but retained susceptibility to carbapenems and aminoglycosides—a profile that underscores their public health risk.

Conclusions: our findings reveal severe ESBL-driven contamination in the Cutuchi, Ambato, Ulba, and Pachanlica Rivers, rendering them unsafe for recreation or aquatic life. Only the Verde River remained uncontaminated, supporting its continued use for human activities. Swimming pools, however, function as reservoirs for multidrug-resistant bacteria, threatening both public health and local tourism. Urgent interventions—including enhanced water quality monitoring and targeted public health policies—are needed to curb the spread of these pathogens and reduce infection rates.

Keywords: Water Pollution; Drug - Resistant Bacteria; Beta-Lactamases; Drug Resistance Microbial; Freshwater.

RESUMEN

Introducción: la contaminación de las aguas estancadas y en movimiento se ha convertido en un problema creciente para la salud pública y el medio ambiente, el agua dulce está siendo afectada por la contaminación progresiva debido al mal manejo de residuos biológicos, industriales, agrícolas, desencadenando la aparición de bacterias multirresistentes o “superbacterias” en ríos y piscinas, especialmente productoras de betalactamasas de espectro extendido (BLEE), las mismas que por su cualidad de resistencia frente a los antibióticos convencionales, producen graves enfermedades difíciles de tratar como infecciones gastrointestinales, urinarias, respiratorias y de la piel, incrementado la morbilidad y mortalidad en la provincia. El objetivo del estudio fue reconocer la presencia de superbacterias en ríos y piscinas en la provincia de Tungurahua y analizar su resistencia antimicrobiana.

Método: estudio de campo descriptivo, de enfoque cuali-cuantitativo realizado mediante un muestreo no probabilístico con la recolección de muestras de cinco ríos y veinticinco piscinas.

Resultados: los coliformes fecales en los ríos se encontraron dentro del límite permisibles según el TULSMA y en las piscinas, superaron el límite. En los ríos se aislaron bacterias como *Klebsiella* y *E. coli* BLEE, mientras que en las piscinas se aislaron *Escherichia coli*, *Klebsiella*, *Acinetobacter* y *Pseudomonas* BLEE, resistentes a antibióticos como las cefalosporinas y aztreonam, y sensibles a carbapenémicos y aminoglucósidos, lo cual constituye un factor de riesgo para la salud pública.

Conclusión: se evidencia una contaminación severa por bacterias productoras de betalactamasas de espectro extendido en los ríos Cutuchi, Ambato, Ulba, Pachanlica, convirtiéndolos en no aptos para la vida ni la práctica recreativa, en tanto que el Río Verde al no presentar contaminación, garantiza mayor tranquilidad para la realización de actividades recreativas y de consumo humano. Las piscinas de Tungurahua se han constituido en un reservorio de bacterias multirresistentes siendo un riesgo para los usuarios y el turismo, por lo que es necesario implementar medidas de salud pública enfocadas en la protección de la calidad del agua y una vigilancia continua para disminuir el incremento de las infecciones en la población.

Palabras clave: Contaminación del Agua; Farmacorresistencia Bacteriana Múltiple; Beta-Lactamasas; Agua Dulce.

INTRODUCTION

Water is the most vital natural resource for the survival of living beings and the environment; it is utilized in industrial, agricultural, and domestic processes.⁽¹⁾ Seventy percent of the Earth's surface is made up of water, and only 2,5 percent of it is freshwater, which is why it is essential to establish preventive measures for the care of this resource. The evolution of human beings has left an indelible mark on the Earth, generating a negative environmental impact, including global warming, loss of natural resources, and water pollution, the latter being a threat to the water cycle.^(2,3)

The primary sources of pollution in moving and standing water are due to the mismanagement of biological and industrial waste resulting from social and industrial development, which spills chemicals, fertilizers, pesticides, and bacteria into nearby water sources.^(2,4) Areas corresponding to rivers and pools have been affected by microbiological contamination by pathogens that cause infectious diseases in humans such as typhoid fever, paratyphoid, acute gastroenteritis, acute diarrhoeal disease, urinary tract infections, meningitis, parasitosis, and mycosis.^(5,6,7) The pathogens involved are *Salmonella typhi*, paratyphi A and B, *Shigella*, *Vibrio cholerae*, *Escherichia coli* ET, *Campylobacter*, *Yersinia enterocolitica*, *Salmonella* sp, *Shigella* sp, viruses, parasites and fungi.⁽⁵⁾

In 1980, Ambler classified beta-lactamases structurally into four categories: A, B, C, and D, of which the presence of serine, a non-essential amino acid that favors hydrolysis, characterizes categories A, C, and D. On the other hand, zinc acts on the carbonyl and amide groups of the beta-lactams, characterizing category B. However, after several studies, their classification has evolved to a molecular and functional character, focusing on isoelectric points, molecular weight, and their susceptibility to certain antibiotics, such as clavulanic acid and tazobactam. In recent years, multi-resistant bacteria, also known as ‘superbugs,’ especially those producing extended-spectrum beta-lactamases (ESBLs), have come under scrutiny in rivers, ponds, and swimming pools due to the increased morbidity and mortality of people living in nearby communities or using these areas for recreational purposes.⁽⁸⁾

The main extended-spectrum beta-lactamases (ESBLs) are produced by the Enterobacteriaceae group, including TEM, SHV, and CTX-M, whose primary mechanism is the degradation of the bacterial cell wall by binding to penicillin-binding proteins (PBPs).⁽⁸⁾ These enzymes are encoded by genes organized extrachromosomally through plasmids or by mobile molecules such as transposons and integrons, which facilitate the horizontal

transfer of genetic material between different bacterial species.⁽⁹⁾ They are also capable of encoding membrane proteins, called porins, especially OmpK35 and OmpK36, which are hydrophilic channels responsible for transporting antibiotics into the intracellular environment. Still, mutations affect the expression of these channels, decreasing permeability to these drugs. Efflux pumps and membrane proteins, such as AcrB, play a similar transport role, the difference being that they transport from the intracellular to the extracellular medium in a non-specific manner to decrease the concentration of the antibiotic. Finally, modification of the antibiotic binding site may occur, as has been demonstrated with methicillin-resistant *S. aureus*, where its affinity and, consequently, its action decrease.⁽¹⁰⁾

The leading cause of contamination in rivers and pools is the presence of fecal coliforms, which are gram-negative, anaerobic, or facultative aerobic bacteria originating from the waste of the intestinal tract of mammals, including humans, as well as from soil, vegetation, estuarine waters, and wastewater management systems.^(11,12) These include *E. coli*, *Klebsiella*, *Enterococcus*, *Citrobacter*, and *Serratia*, whose permissible limit according to the TULSMA for recreational waters is 200 amp/100 ml and for secondary contact recreational waters 1000 amp/100 ml.^(13,14)

BLEE are enzymes produced by certain bacteria and are intended to break down various types of antibiotics, making them resistant, especially to penicillins and cephalosporins, including third and fourth generation, and can be inhibited by clavulanic acid, sulbactam, and tazobactam; due to this resistance to treatment, stronger antibiotics and higher doses are required.⁽⁸⁾ The resistance exerted by these BLEE bacteria makes it necessary for antibiotics to increase the minimum inhibitory concentration (ug/ml) to inhibit the growth and proliferation of these microorganisms and thus be able to eradicate infections.^(11,12,15)

In Ecuador, the presence of enterobacteria that are pathogenic for humans has been observed, specifically *E. coli*, producer of β -lactamases of extended-spectrum (BLEE), in the rivers Chimbo, located in the province of Bolivar and in the Machangara situated in the province of Pichincha, which has generated a growing concern among the inhabitants of the region, who use this resource for recreational purposes; that is why the purpose of this research is to demonstrate the existence of resistant pathogens in rivers and swimming pools in the province of Tungurahua.^(16,17)

METHOD

This is a descriptive field study employing a qualitative-quantitative approach and non-probabilistic sampling to determine the presence of coliforms and BLEE-producing enterobacteria, as well as to assess their sensitivity and resistance to various antibiotics, in five rivers and twenty-five freshwater pools for recreational use in the province of Tungurahua, Ecuador, from July to December 2023.

Sampling

Five rivers belonging to the province of Tungurahua were considered: Ambato, Pachanlica, Cutuchi, Ulba, and Verde rivers; and twenty-five swimming pools considering the affluence of bathers, the distance from the riverbanks and the hour of highest demand; the authorization of participation for the collection of the samples from the pools was made by informed consent, and the samples were anonymized using an alphanumeric code, to preserve their prestige; The samples were collected in 250 ml amber bottles, sterilized using moist heat, and once the sample was obtained, the physical analysis was carried out in situ, obtaining the environmental humidity and water temperature.

In the laboratory, physical-chemical analysis was carried out to determine the residual chlorine and pH using a calibrated potentiometer. The determination of the concentration of colony forming units (CFU) was carried out using two techniques: the most probable number and the count in Petrifilm; in the samples where growth was obtained, Gram staining was performed, and they were reseeded in MacConkey agar, being able to perform a visual and microscopic differentiation of the colonies to differentiate species, each different colony was reseeded in CHROMagar for gram-negative antibiotics resistant to beta-lactams, the colonies that grew in the test were diluted in sterile water until obtaining a value of 0,5 on the McFarland scale, 30 μ L of this solution was incubated in Mueller Hinton Broth. Subsequently, 50 μ L of this new solution was deposited in each of the 96 wells of the Sensititre™ GNX2F plate and incubated for 24 hours at 37°C.

RESULTS

Table 1. Physical-chemical analysis of freshwater from rivers in the province of Tungurahua

N°	Samples	River	pH	Chlorine residual (mg/L)	Water temp. (°C)	Ambient Temp (°C)	Hum. Environmental (%)
1	RV-A	River Green	6,8	0,01	14	22	67
2	RV-B	River Green	6,9	0	14	24,6	63
3	RU-A	River Ulba	7,1	0,03	16,5	32,6	39

4	RU4-A	River Ulba	6,8	0,04	15,7	31,3	36
5	RC	River Cutuchi	7,2	0,01	17,6	28,9	44
6	RP	River Pachanlica	6,5	0,02	18,2	25	50
7	RA	River Ambato	6,7	0	16,4	27,5	43

Table 1 presents the physicochemical analysis of the water in the rivers of Tungurahua province. Parameters such as pH ranged between 6,5 and 7,2. The residual chlorine had the highest value of 0,04 mg/L in the Ulba River. The water temperature fluctuated between 14 and 18,2 °C, and the ambient temperature ranged between 22 and 32,6 °C. On the other hand, the ambient humidity was between 36 % and 67 %.

Table 2. Physical-chemical analysis of fresh water from pools in the province of Tungurahua

N°	Samples	Swimming pools	pH	Chlorine residual (mg/L)	Water temp. (°C)	Ambient Temp. (°C)	Hum. Environmental (%)
1	M	Swimming pools M	7,2	0,229	28,1	12,8	80
2	N	Swimming pools N	7,6	0,166	31,9	29,2	44
3	O	Swimming pools O	6,5	0,178	23,8	13,5	84
4	P	Swimming pools P	6,2	0,1	22,6	18	86
5	Q	Swimming pools Q	6,7	0,12	26	36,2	40,5
6	R	Swimming pools R	6,8	0,176	32,1	32,7	41,5
7	A	Swimming pools A	6,9	0,08	29,7	32,5	39,7
8	S	Swimming pools S	6,5	0,299	32	32,6	40,8
9	T	Swimming pools T	6,1	0,09	32,2	32,6	42
10	B	Swimming pools B	6,8	0,04	25	20	88
11	U	Swimming pools U	6,7	0,124	28	24	64
12	C	Swimming pools C	6,4	0,031	31,1	16	63,6
13	D	Swimming pools D	6,2	0,02	33	16	63,6
14	E	Swimming pools E	6,8	0,03	33,5	30,6	56,7
15	F	Swimming pools F	6,6	0,09	25,3	31,8	34,6
16	V	Swimming pools V	6,1	0,198	31,5	29,7	45,4
17	W	Swimming pools W	6,8	0,045	25,8	27,8	64,6
18	X	Swimming pools X	7,7	0,14	24,5	30,1	32,9
19	G	Swimming pools G	6,4	0,096	32,4	32,4	24,5
20	H	Swimming pools H	6,1	0,118	27,7	32	70
21	I	Swimming pools I	6,6	0,07	29,9	31,6	88,7
22	Y	Swimming pools Y	7,1	0,137	21,3	28	50
23	J	Swimming pools J	6,8	0,05	22,2	34,4	59
24	K	Swimming pools K	6,5	0,06	27,3	29,6	31
25	L	Swimming pools L	7	0,09	26,8	33,3	58

Table 2 presents the physicochemical analysis of the pools in the province of Tungurahua. The pH values ranged from 6,1 to 7,7, the concentration of residual chlorine ranged from 0,02 in pool D to 0,299 mg/L in pool S, the water temperature of the pools ranged from 21,3 °C in pool Y to 33,5 °C in pool E, with an ambient temperature between 12,8 °C pool M to 36,2 °C pool Q. On the other hand, the ambient humidity was found to range from 24,5 % in Pool G to 88,7 % in Pool I.

Table 3. Analysis of fecal coliforms in fresh water from rivers in the province of Tungurahua

N°	Samples	Rivers	UFC/100 mL	Permissible limit faecal coliforms (CFU/100 mL)	Permissible limit total coliforms (CFU/100 mL)	Complies with faecal coliform limit	Complies with the limit for total coliforms
1	RV	Green River	0	200	1000	yes	yes
2	RU4-A	River Ulba	2,55	200	1000	yes	yes
3	RC	River Cutuchi	12,78	200	1000	yes	yes

4	RP	River Pachanlica	6	200	1000	yes	yes
5	RA	River Ambato	16	200	1000	yes	yes

Table 3 shows the number of colony forming units in the rivers of the province of Tungurahua. The Ulba river has 2,55 CFU/100mL, the Pachanlica 6 CFU/100mL, the Cutuchi 12,78 CFU/100mL, the Ambato river 16 CFU/100mL and the Verde river 0 CFU/100mL, complying with the permissible limit of fecal coliforms established by the TULSMA.

Table 4. Analysis of faecal coliforms in fresh water from pools in the province of Tungurahua

N°	Samples	Swimming pools	UFC/100 mL	Permissible limit faecal coliforms (CFU/100 mL)	Permissible limit total coliforms (CFU/100 mL)	Complies with faecal coliform limit	Complies with the limit for total coliforms
1	M	Swimming pools M	0	200	1000	yes	yes
2	N	Swimming pools N	0	200	1000	yes	yes
3	O	Swimming pools O	0	200	1000	yes	yes
4	P	Swimming pools P	0	200	1000	yes	yes
5	Q	Swimming pools Q	0	200	1000	yes	yes
6	R	Swimming pools R	9,45	200	1000	yes	yes
7	A	Swimming pools A	2,55	200	1000	yes	yes
8	S	Swimming pools S	2,55	200	1000	yes	yes
9	T	Swimming pools T	0	200	1000	yes	yes
10	B	Swimming pools B	129,55	200	1000	yes	yes
11	U	Swimming pools U	0	200	1000	yes	yes
12	C	Swimming pools C	5133,3	200	1000	No	No
13	D	Swimming pools D	14053,3	200	1000	No	No
14	E	Swimming pools E	69,33	200	1000	yes	yes
15	F	Swimming pools F	10	200	1000	yes	yes
16	V	Swimming pools V	0	200	1000	yes	yes
17	W	Swimming pools W	0	200	1000	yes	yes
18	X	Swimming pools X	0	200	1000	yes	yes
19	G	Swimming pools G	2173,33	200	1000	No	No
20	H	Swimming pools H	2,55	200	1000	yes	yes
21	I	Swimming pools I	254,7	200	1000	No	yes
22	Y	Swimming pools Y	0	200	1000	yes	yes
23	J	Swimming pools J	10,8	200	1000	yes	yes
24	K	Swimming pools K	15,8	200	1000	yes	yes
25	L	Swimming pools L	19	200	1000	yes	Yes

Table 4 shows the number of colony-forming units in the pools in the province of Tungurahua. Pool C presents 5133,3 CFU/100mL, Pool D, 14053,3 CFU/100mL, Pool G, 2173,33 CFU/100mL, and Pool I, 254,7 CFU/100mL, values that exceed the permissible limit of total coliforms according to the value established by the TULSMA.

In the samples obtained from the Cutuchi, Pachanlica, and Ambato rivers and cultivated in CHROMagar, the visual and microscopic differentiation showed colonies with red pigmentation corresponding to *E. coli* BLEE. In the case of the Ulba River, the colonies were pigmented blue, corresponding to *Klebsiella* BLEE. When the samples obtained from the pools were cultured on CHROMagar, visual and microscopic characteristics were distinguished; cream-colored colonies were observed, corresponding to *Acinetobacter* BLEE, translucent *Pseudomonas* BLEE, blue *Klebsiella* BLEE, and red *E. coli* BLEE.

For the analysis of the resistance of β -lactamase-producing bacteria, a heat map was used, a visual tool that displays numerical data using colors. In this case, it represents bacterial resistance to antibiotics, where the rows indicate the drugs and the columns indicate the bacterial strains isolated from different rivers. The color indicates the minimum inhibitory concentration (MIC), with the more intense red indicating greater resistance. White cells indicate no resistance. This visualization facilitates rapid identification of resistance patterns, including multi-resistant strains or antibiotics with lower efficacy, helping to guide control

measures, surveillance, and choice of the appropriate antibiotic for clinical treatment.

The sensitivity for *Escherichia coli* BLEE in the Cutuchi River is 16 µg/mL for ceftazidime, in the Pachanlica River 16 µg/mL for doxycycline, and in the Ambato River 32 µg/mL for ceftazidime, on the other hand, in the Ulba river, for *Klebsiella* BLEE 8 µg/mL of ceftazidime is needed (figure 1).

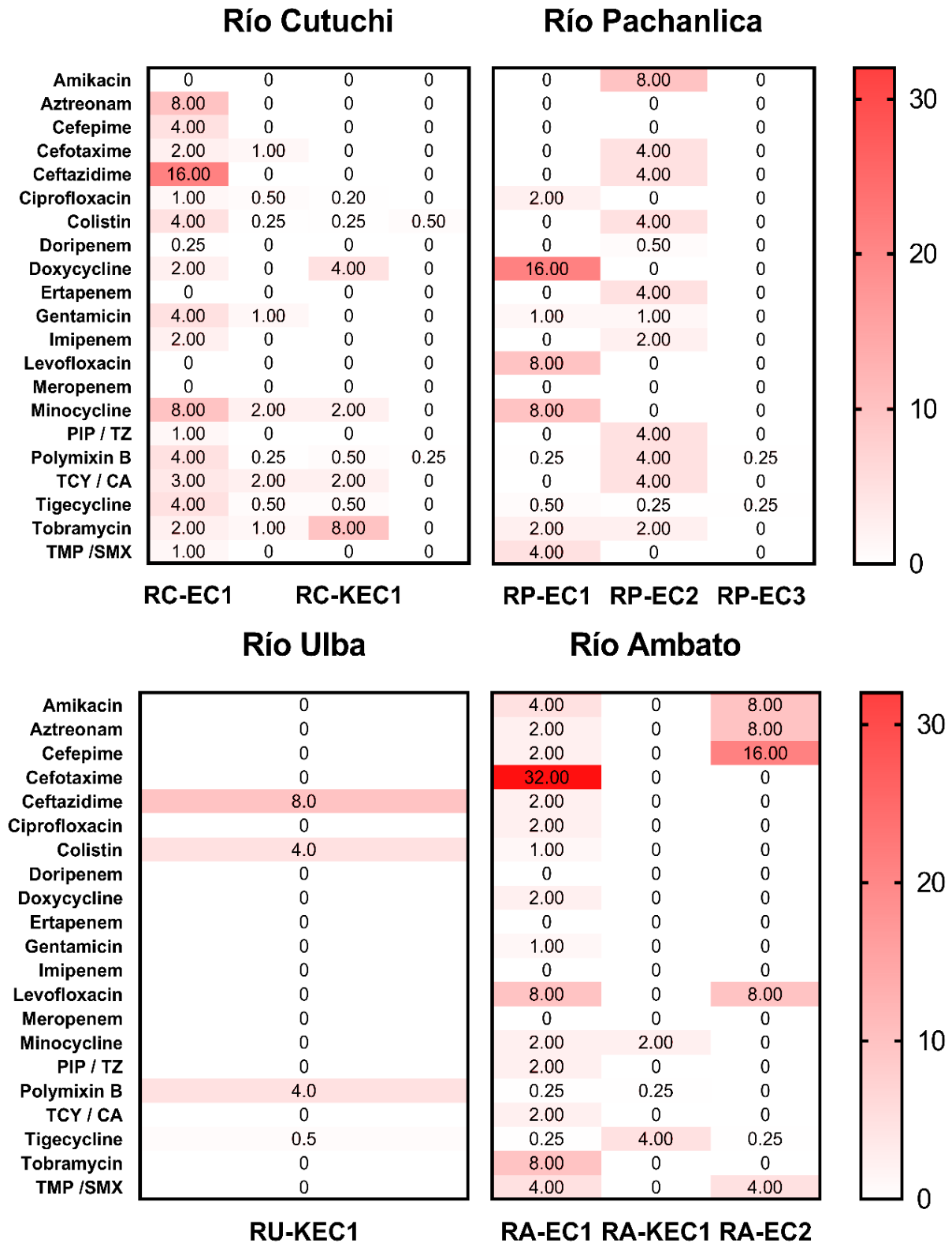


Figure 1. Heat map of the minimum inhibitory concentrations for BLEE bacteria in the rivers of Tungurahua

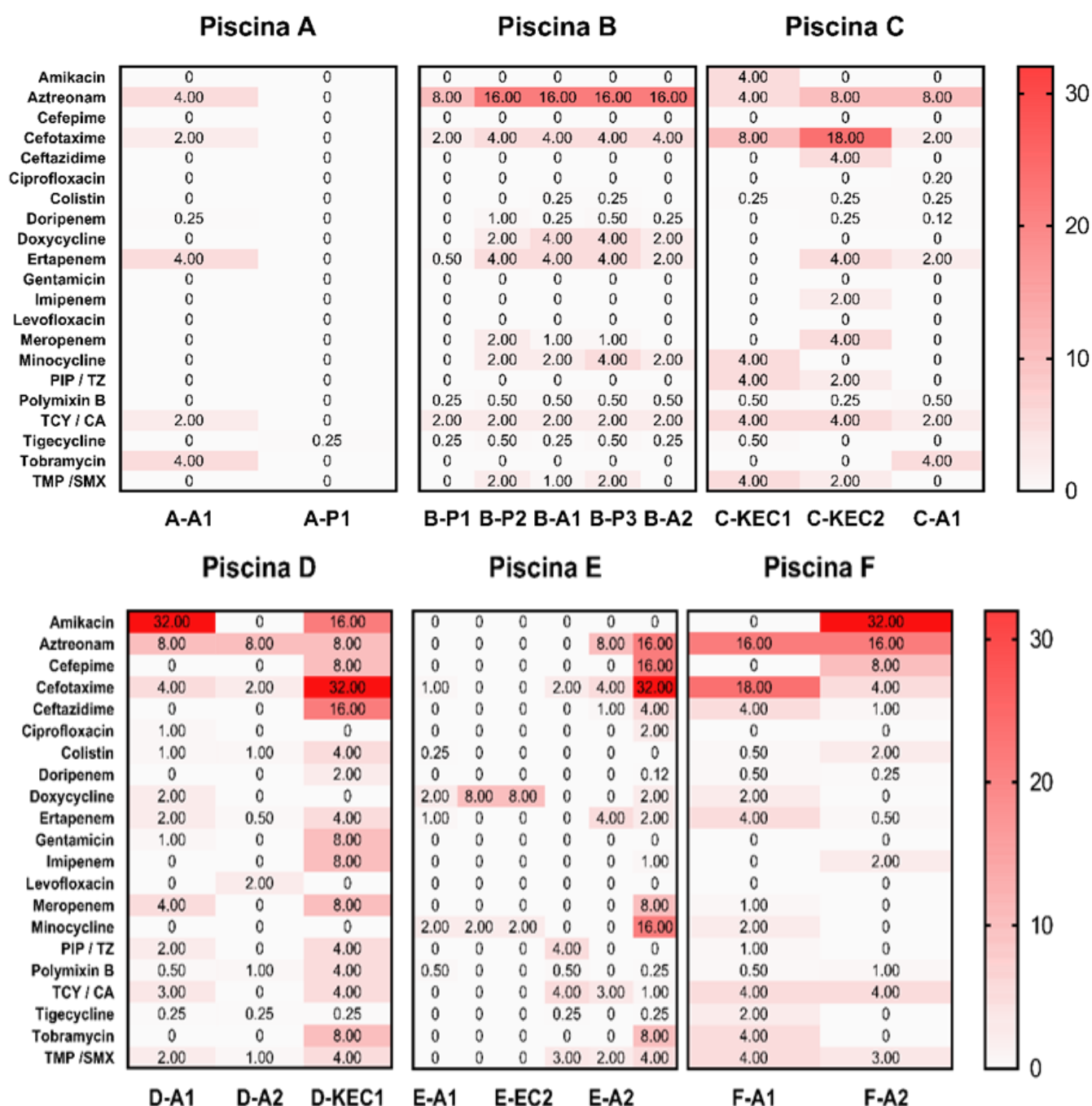


Figure 2. Heat map of minimum inhibitory concentrations for BLEE bacteria in Tungurahua pools

In heat map 2 (figure 2). In pool A, for *Acinetobacter* BLEE, four ug/mL of aztreonam is needed; in pool B, for *Pseudomonas* BLEE, 16 ug/mL of aztreonam; in pool C, for *Klebsiella* BLEE, 18 ug/mL of cefotaxime, in pool D, for *Acinetobacter* and *Klebsiella* BLEE 32 ug/mL amikacin is required, in pool E, for *Acinetobacter* and *Escherichia coli* BLEE 32 ug/mL cefotaxime is needed and finally in pool F, for *Acinetobacter* 32 ug/mL amikacin is required.

DISCUSSION

Bacterial contamination in freshwater bodies, both standing and moving, is a serious public health problem in the province of Tungurahua, Ecuador.

A study of urban wastewater from three socio-spatially distinct districts of the Ruhr metropolis (Germany) revealed a high prevalence of BLEE-producing *Escherichia coli* in waste generated from densely populated areas and industrial discharges.⁽¹⁸⁾ Beta-lactam-resistant (BLEE) strains were found at three sites in the Szreniawa River (Poland), related to household wastewater discharge and agricultural runoff; double-disk synergy tests and polymerase chain reaction (PCR) analysis were used to identify BLEE-producing bacteria, finding resistance to

amoxicillin/clavulanate (90 %) and ampicillin (36 %).⁽¹⁹⁾ In this study carried out in the province of Tungurahua, in the Ambato, Cutuchi, Pachanlica, and Ulba rivers, *Escherichia coli* and *Klebsiella* BLEE were isolated by CHROMagar from human, agricultural, and industrial discharges in the province of Tungurahua. In contrast, no bacterial strains were isolated in the Verde River, which is a living river suitable for human consumption and recreational use.

The determination of the pH of water is considered fundamental in evaluating its quality, as it directly influences the survival, proliferation, and resistance of the microorganisms present.⁽²⁰⁾ The physicochemical and microbiological characteristics of water in swimming pools belonging to two recreational centers located in the western area of the city of Maracaibo, Zulia State (Venezuela) were evaluated, finding average values of residual chlorine and pH outside the limits established by health recommendations, with the normal pH for swimming pools being 7,2 and 7,6.⁽²¹⁾ In the present study, the pH ranged between 6,1 and 7,7, which indicates that there is a slight variation, a situation that could favor bacterial proliferation since a low pH can compromise the effectiveness of chlorine as a disinfectant. At the same time, high values alter the natural balance and generate unfavorable conditions for beneficial organisms but are ideal for resistant pathogens. The normal value of free residual chlorine in swimming pools is 0,5 to 2 mg/L. In our study, the rivers showed a maximum value of 0,04 mg/L. In the swimming pools, the residual chlorine value ranged from 0,02 to 0,29 mg/L, indicating inadequate chlorination to fulfill its disinfectant activity and thus promoting the proliferation of microorganisms. In a physicochemical and bacteriological analysis of the most frequented thermal pools in the parish of Baños, Ecuador, it was determined that the temperature of the pools was highly variable, which favored the growth of pathogens.⁽²²⁾ In our study, the temperature of the pools was found to be between 21,3 °C and 33,5 °C, which constitutes a favorable environment for the proliferation of microorganisms.

In the Rimac River (Peru), *E. coli* BLEE strains resistant to third-generation cephalosporins, such as cefotaxime, ceftazidime, and aztreonam, and sensitive to gentamicin, ciprofloxacin, and carbapenems, including imipenem and meropenem, were isolated.⁽²³⁾ In this study, samples from the Ambato, Cutuchi, and Pachanlica rivers, highly influenced by human activities and agricultural dumping, showed growth of *E. coli* and *Klebsiella* BLEE resistant to cefotaxime, ceftazidime, amikacin, levofloxacin and piperacillin/tazobactam. However, sensitivity to carbapenems, such as meropenem, doripenem, and imipenem, as well as gentamicin, was observed, indicating that the strains isolated in our rivers are less resistant than those found in Peru.

In Bavaria (Germany), bacteria resistant to antibiotics were found in eleven therapy pools, related to the lack of adequate disinfection, which could cause illnesses in bathers, especially in immunocompromised patients, mostly non-fermenting Gram-negative bacilli were isolated, such as *Pseudomonas* spp, *S. maltophilia* and *S. paucimobilis*, resistant to commonly used antibiotics such as imipenem, ertapenem, ciprofloxacin and levofloxacin. However, they were sensitive to piperacillin, ceftazidime and cefepime.⁽²⁴⁾ On the other hand, in the hot springs in Baños (Ecuador), *Staphylococcus aureus* was found in 77 %, Coliforms in 8 %, *Escherichia coli* in 2 %, fungi in 1 % and yeasts in 8 %, concluding that *Staphylococcus aureus* does not constitute a significant threat to people who frequent these recreational areas, since it is a microorganism that usually inhabits the human body naturally as part of its normal microbiota.⁽²²⁾ In our study, using CHROMagar culture media from swimming pool water, we found a bacterial landscape dominated by enterobacteria producing extended-spectrum β -lactamase (ESBL) such as *Escherichia Coli*, *Klebsiella*, *Pseudomonas* and *Acinetobacter* BLEE resistant to conventional antibiotics and sensitive to carbapenems (meropenem, doripenem, ertapenem, imipenem), these results indicate that our strains are still sensitive to carbapenems and resistant to few antibiotics in contrast to the colonies found in the German pools. This situation is related to non-compliance with TULSMA regulations, lack of cleanliness, and poor control of pH and chlorine levels, resulting in potential risks to human and ecosystem health.

The minimum inhibitory concentration (MIC) is the lowest concentration of an antibiotic that prevents the visible growth of a specific bacterial strain, measured in micrograms per milliliter ($\mu\text{g/mL}$). This parameter is essential for determining the efficacy of an antibiotic against a particular microorganism and is classified as sensitive (S), intermediate (I), and resistant (R).⁽²⁵⁾ In the Cutuchi River, the *E. Coli* BLEE requires an MIC of 16 $\mu\text{g/mL}$ of Ceftazidime when the MIC value is $\leq 0,12 \mu\text{g/mL}$, indicating that a higher dose of the drug is needed to inhibit its proliferation. In the case of pool D, *Acinetobacter* BLEE requires an MIC of 32 $\mu\text{g/mL}$ of Amikacin when the MIC value is $\leq 2 \mu\text{g/mL}$.⁽²¹⁾ For Other antibiotics, such as amikacin and levofloxacin in infections caused by BLEE in rivers and pools, the MIC is zero $\mu\text{g/mL}$, indicating that they have good sensitivity and can be used successfully at therapeutic doses.

CONCLUSIONS

The study revealed that freshwater recreational sources in Tungurahua, Ecuador, both stagnant and flowing, are contaminated with multidrug-resistant bacteria that produce extended-spectrum beta-lactamases (ESBLs), including *E. coli*, *Klebsiella*, *Acinetobacter*, and *Pseudomonas*. Although the rivers complied with the permissible limit for coliforms, some pools exceeded the parameters, representing a health risk. On the other hand, these

bacteria showed sensitivity to carbapenems and aminoglycosides but resistance to other common antimicrobials. Current regulations in Ecuador do not include specific microbiological criteria for these multidrug-resistant bacteria, which highlights the need to strengthen environmental surveillance and expand research into the genetic mechanisms of resistance and their impact on public health.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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