

Bacteria with dual resistance to elevated concentrations of heavy metals and antibiotics in Nigerian contaminated systems

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Abstract Samples of soil, water, and sediments from industrial estates in Lagos were collected and analyzed for heavy metals and physicochemical composition. Bacteria that are resistant to elevated concentrations of metals (Cd^{2+} , Co^{2+} , Ni^{2+} , Cr^{6+} , and Hg^{2+}) were isolated from the samples, and they were further screened for antibiotic sensitivity. The minimum tolerance concentrations (MTCs) of the isolates with dual resistance to the metals were determined. The physicochemistry of all the samples indicated were heavily polluted. Twenty-two of the 270 bacterial strains isolated showed dual resistances to antibiotics and heavy metals. The MTCs of isolates to the metals were 14 mM for Cd^{2+} , 15 mM for Co^{2+} and Ni^{2+} , 17 mM for Cr^{6+} , and 10 mM for Hg^{2+} . Five strains (*Pseudomonas aeruginosa*, *Actinomyces turicensis*, *Acinetobacter junni*, *Nocardia* sp., and *Micrococcus* sp.) resisted all the 18 antibiotics tested. Whereas *Rhodococcus* sp. and *Micrococcus* sp.

resisted 15 mM Ni^{2+} , *P. aeruginosa* resisted 10 mM Co^{2+} . To our knowledge, there has not been any report of bacterial strains resisting such high doses of metals coupled with wide range of antibiotics. Therefore, dual expressions of antibiotics and heavy-metal resistance make the isolates, potential seeds for decommissioning of sites polluted with industrial effluents rich in heavy metals, since the bacteria will be able to withstand *in situ* antibiosis that may prevail in such ecosystems.

Keywords Antibiotics · Biotransformation · Heavy metal · Industrial effluent · Resistance

Introduction

Heavy metals are those elements with a molecular weight greater than 53, a density greater than 6 g cm^{-3} , and an atomic number greater than 20. They occur naturally in rocks and soils, but concentrations are frequently elevated as a result of pollution. They are called trace elements, which are toxic to living organisms at excessive concentrations, but some including Fe, Zn, Cu, Mn, Mo, and so on, at low but critical concentrations are micronutrients used in the redox processes, regulation of the osmotic pressure, and also enzyme components which are essential for the normal healthy growth and reproduction by living

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organisms. At high concentrations, micronutrients damage DNA and membrane, as well as loss of functions of enzyme. However, heavy metals, like Hg, Cd, and Pb, of no known biological importance directly cause oxidative stress, lipid peroxidation, carcinogenesis, mutagenesis, and neurotoxicity on humans, animals, and plants at low concentrations (Howlett and Avery 1997).

Elevated concentration of heavy metals are introduced into the environment through metal-liferous mining, metal smelting, activities of metallurgical industries, waste disposals, corrosion of metals in use, agriculture, and petroleum exploration among others. The discharge of effluents containing heavy metals mounts pressures on the ecosystem and consequently causing health hazards to plants, animals, aquatic life, and humans (Pazirandeh et al. 1998). Upon surface contamination, the toxic metals are transported to groundwater and/or bioaccumulated (Stephen et al. 1999). Introduction of certain concentrations of heavy metals into the environment kills the majority of the microflora, thereby selecting for a few cells that would have evolved resistance mechanisms to the metals. The emergence of resistant strains from the indigenous community participates in various self-recovery processes which occur in such polluted habitat. Some microorganisms have been reported to have evolved mechanisms to detoxify heavy metals, and some even use them for respiration, thereby, becoming resistant to such metals (Clausen 2000). The resistance mechanisms used by microorganisms to tolerate heavy metal stress include permeability barriers, intra- and extracellular sequestration, efflux pumps, enzymatic detoxification, and reduction (Nies 1999). Living organisms absolve sublethal concentrations of heavy metals upon pollution by diffusion of ions or complexes, mediated transport, and/or endocytosis of particulate metal and pinocytosis of organo-metallic aggregates (Otchere 2003).

The toxicity of heavy metals to microbial community in the soil has been reported to also inhibit biodegradation of organic pollutants in co-contaminated sites (Said and Lewis 1991; Sandrin et al. 2000). In some cases, resistance to metal ions has been reported to be plasmid-mediated and observed to be encoded by genes in close

proximity to antibiotic-resistance genes. A correlation between heavy metal tolerance/resistance and antibiotic resistance in *Escherichia coli* (Spain 2003) and *Staphylococcus* sp. (Groves et al. 1975) has been reported. Alonso et al. (2000) implicated a cluster of genes to be involved in antibiotic and heavy-metal resistance of a clinical isolate of Gram-negative bacterium, *Stenotrophomonas (Xanthomonas) maltophilia*. Therefore, metal resistance and antibiotic resistance may sometimes be transferred together in the environment.

The use of microorganisms to sequester, precipitate, or alter the oxidation state of various heavy metals (Gadd and White 1993; Rittle et al. 1995) through reduction, accumulation, mobilization, and immobilization (Lovley 1994; Avery 1995; Valentine et al. 1996) has been studied in America and Europe but not in Africa and specifically in Nigeria. Notable among the reported bacteria include *E. coli*, phytopathogenic *Pseudomonas* and *Xanthomonas* species which resist copper; *Bacillus* spp. (Valentine et al. 1996; Clausen 2000), *Alcaligenes* spp. (Dressler et al. 1991; Clausen 2000), *Klebsiella* spp. (Clausen 2000), *Acinetobacter* spp. (Clausen 2000) which tolerate some levels of concentration of various metal ions and strains of *Streptomyces* which are highly resistant to mercury (Ravel et al. 1998).

Industrial estates in Lagos, Nigeria are laced with industries manufacturing textile, allied chemicals, electroplating, batteries, paints, plastics, and petrochemicals to mention a few. These industries empty their effluents, without any known metal-removing treatment before discharge, into the surrounding river which eventually empties into the lagoon. These receiving environments in Lagos (Fakayode and Onianwa 2002; Oyeyiola et al. 2006) and other parts of Nigeria (Olajire 1998; Akpan et al. 2002; Aremu et al. 2002; Osuji and Onojake 2004) have been reported to contain high concentrations of various heavy metals. Notable toxic metals found in Lagos, due to activities of most allied chemical industries in the city, are cadmium, chromium, arsenic, cobalt, nickel, copper, mercury, and lead among others (Fakayode and Onianwa 2002; Oyeyiola et al. 2006). Osuji and Onojake (2004) also reported that high concentration of heavy metal persists in sites polluted with crude oil even after degradation of the pollutant.

The continual discharge of effluents laden with heavy metals alters the ecological status of the affected environment with the evolution of (1) microorganisms which can reduce heavy metals and detoxify them, (2) microorganisms which are resistant to the heavy metals and can perform their metabolic activity in the presence of the metals, and (3) microorganisms which are heavy-metal-sensitive but can survive the presence of such metals in the soil and perform their metabolic activity once the metals have been reduced (Turpeinen et al. 2004; Nakatsu et al. 2005). Isolation and characterization of competent microorganisms able to resist elevated concentrations of heavy metals as well as showing multiple resistances to antibiotics are critical to the development of an effective bioremediation strategy in such polluted sites. More importantly, an in-depth knowledge on the environmental distribution of heavy metals is of great interest as such information will help determine the process conditions that should be used in practical treatment systems and, hence, be very valuable in developing effective remediation strategies for the cleanup of contaminated sites. Therefore, in this study, we evaluated the physicochemistry as well as heavy

metal concentrations of water, sediment, and soil samples with long history of exposure to industrial effluents in Lagos and subsequently, isolated bacteria that showed resistance to elevated concentrations of selected heavy metals. The isolates were screened for multiple resistances to antibiotics. The organisms with combination of antibiotic and heavy-metal resistance would be useful for bioremediation of environments polluted with metals and also help to overcome metabolic bottlenecks still existing in the biodegradation processes such as the inhibition that heavy metals can exert on the biodegradation of organic pollutants. Such organisms would be able to compete well with antibiotic-producing flora in the polluted environment.

Materials and methods

Sample collection

Soil samples were collected from Isolo Industrial Estate, water samples from Alaro River, and effluent sediment from Ikeja Industrial Estate, all in Lagos, Nigeria (Fig. 1). The sampling points

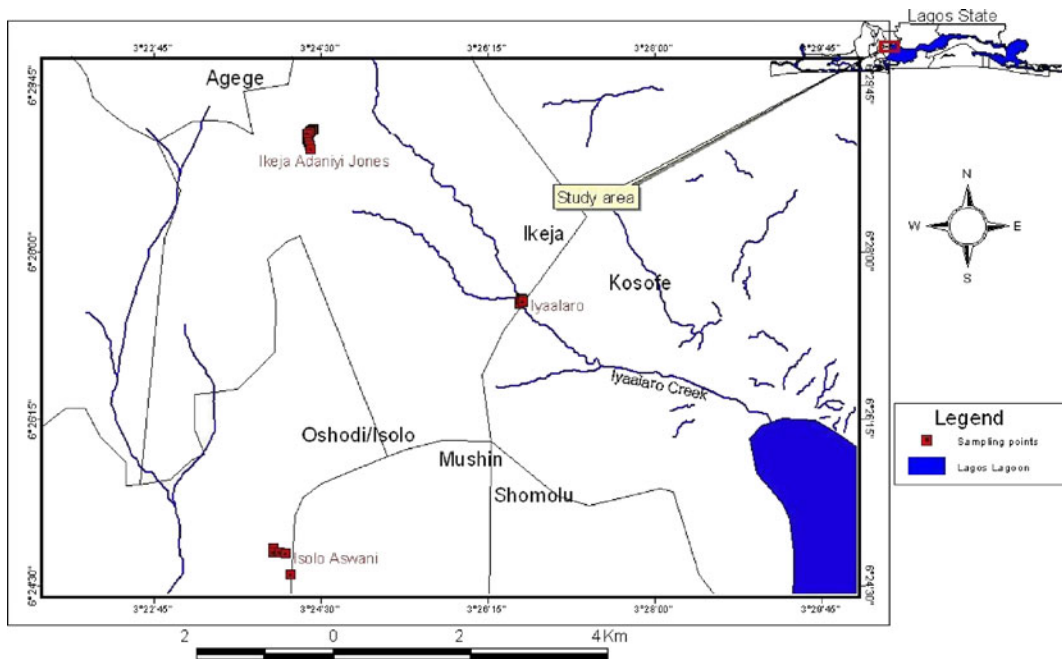


Fig. 1 Map of study area showing the sampling points

at Isolo were abandoned industries of textile, battery, and pharmaceuticals while that of Ikeja were functional allied chemical and battery. Alaro River is the central collection point of all effluents emanating from Ikeja industrial area before it eventually empties into the Lagos lagoon. Samples were collected in sterile screw cap glass specimen bottles. In the case of liquid samples, the bottles were held from the bottom and then plunged into the water downward, setting the mouth against the direction of water flow. Composite samples (ten replicates) were collected randomly at each location and were processed immediately.

Physicochemical analysis

Physicochemistry of the samples was determined by standard methods described by Eaton et al. (1995), Nelson and Sommers (1982), and AOAC (1990). Parameters assayed were pH, total solids, total dissolved solids, acidity, alkalinity, hardness, chlorides, chemical oxygen demand (COD), and biochemical oxygen demand (BOD).

Analytical procedure for heavy metals

Samples were filtered through No. 1 (Whatman Inc. NJ, USA) filter paper, then through 0.45 μm Millipore membrane filters (Millipore Corp. MA, USA), and acidified with nitric acid (pH 2.0). Heavy metal analyses of the samples were determined using flame atomic absorption spectrophotometry.

Chemicals and stock solutions

The following metal salts namely, HgCl_2 , CdCl_2 , CoCl_2 , NiCl_2 , and CrO_3 were purchased from Sigma–Aldrich Corp. (St. Louis, MO, USA). All other chemicals were of analytical reagent grade. Stock solutions (1 M) were made in distilled water, sterilized by filtration through 0.22- μm membrane filters (Nucleopore Corp., Pleasanton, CA, USA), and stored in sterile flasks in the dark at 4°C for no longer than 1 day.

Isolation of heavy-metal-resistant bacteria

The samples were inoculated into Luria Bertani (LB) broth amended with 1% metal solutions (0.05 mM, HgCl_2 [Hg^{2+}]; 0.1 mM, CdCl_2 [Cd^{2+}]; 0.5 mM, CoCl_2 [Co^{2+}]; 0.5 mM, NiCl_2 [Ni^{2+}]; 1.0 mM, CrO_3 [Cr^{6+}]) after which appropriate dilutions of the 48-h-old cultures were plated onto LB agar supplemented with 0.2% metal solutions. Pure cultures were initially screened for heavy-metal resistance using sterile filter paper disk impregnated with varying concentrations of metals. The discs were placed on LB agar already seeded with the isolates and incubated at $29 \pm 2^\circ\text{C}$ for 48 h. Resistant colonies were selected, subjected to Gram, spore, and ZN-staining procedures, and observed morphologically under the microscope. Isolates were tentatively identified on the basis of their biochemical techniques following the taxonomic schemes of Bergey's Manual of Determinative Bacteriology (Holt et al. 1994) and Cowan and Steel (1994).

Metal-resistance assay

Isolates were grown in LB broth for 18 h at room temperature ($29 \pm 2^\circ\text{C}$). Cells were harvested by centrifugation (7,000 $\times g$, 10 min), washed twice with sterile buffered phosphate solution, and re-suspended in the same buffer solution. The cell concentration (dry weight) of bacterial suspensions was determined by measuring the optical density (OD) of the samples at 600 nm and relating the value to a calibration curve (10^{10} cfu l^{-1} = 1 OD unit).

Dilution to various concentrations of Cd^{2+} (1.0–20.0 mM), Co^{2+} (6.0–20.0 mM), Hg^{2+} (1.0–15.0 mM), Ni^{2+} (6.0–20.0 mM), and Cr^{6+} (1.0–20.0 mM) were made from the stock into LB broth. The media were subsequently dispensed in 10-ml aliquots into test tubes and inoculated with 50 μl inoculum. Growth media without heavy metals and inoculation served as controls. Growth of the inocula was measured by absorbance at 600 nm and occasional viable count assay. Resistance was tested as maximum tolerance concentrations (MTCs) for the isolates following incubation for 7 days at room temperature. The

MTC was defined as the highest concentration of metal which do not affect the viable counts of organisms. In all experiments, each metal concentration was tested in three replicates and each experiment was repeated three times.

Antibiotics sensitivity

The paper disc method was adopted for determining the antibiotics sensitivity patterns of isolates. Standard antibiotic disks of septrin 30 µg (SXT), chloramphenicol 30 µg (CH), sparfloxacin 10 µg (SP), ciprofloxacin 10 µg (CPX), amoxicillin 30 µg (AM), augmentin 30 µg (AU), gentamycin 10 µg (CN), pefloxacin 30 µg (PEF), Tarivid 10 µg (OFX), streptomycin 30 µg (S), ampiclox 30 µg (APX), zinnacef 20 µg (Z), rocephin 25 µg (R), erythromycin 10 µg (E), metronidazole 30 µg (MTZ), cefuroxime 30 µg (CXM), amoxycillin 30 µg (AMC), and ceftriaxone 30 µg (CRO) were placed on Mueller–Hinton agar already seeded with 0.1 ml inoculum of isolates. Plates were incubated at 30°C for 48 h and observed for zones of inhibition.

Results and discussion

The physicochemical and heavy metal compositions of the samples are presented in Tables 1 and 2, respectively. The results obtained revealed that the samples were heavily contaminated with

heavy metals. Samples obtained from Alaro River were strikingly acidic with a pH value of 3.92, while sediment and soil collected from Ikeja and Isolo, respectively, were weakly acidic (pH 6.45–6.51). It is noteworthy that virtually all the parameters determined were far above the limits set by the regulatory body in Nigeria. In addition, the samples were found to be purple, brown, and green in color compared to colorless that is recommended by FEPA. This must be due to the presence of dyes used for fabrics and other industrial operations, which would doubtlessly have adverse effects on the ecosystem as color impacted in the receiving water body might affect penetration of light for photosynthetic organisms. According to the report of Wong and Yu (1999), color is usually the first contaminant to be recognized in wastewaters, and this affects the esthetics, water transparency, and gas solubility.

From the standpoint of the samples’ physicochemistry, it is apparent that they lack adequate treatment processes, hence, are not fit for discharge into the environment. Previously, Odokuma and Okpokwasili (1993) reported that a BOD value of 2.5 mg/l indicates fairly polluted water while a value of 4 or 5 mg/l and above indicates polluted water. The astronomical values of COD and BOD of the samples might result in a drift of available oxygen for chemical and biochemical reactions in the system away from biomass sustainability in the ecosystem. This will definitely set survival pressure in the receiving ecosystem, thus, wiping out many vulnerable

Table 1 Physico-chemical properties of samples

Parameter ^a	Wemco	Wash Tank	Alaro River	Ikeja sediment	Isolo soil	WHO recommended limit	FEPA maximum allowed limit
pH	8.84	8.81	3.92	6.48	6.51	7.0–8.9	6.0–9.0
Total solids (ppm)	16190	16590	480	1595	6400	500	500
Total dissolved solids (ppm)	15395	15435	460	325	405	NA	30
Acidity (ppm)	2	2	260	40	40	NA	30–500
Alkalinity (ppm)	40	40	0	20	20.5	100	30–500
Hardness (ppm)	300	308	76	84	48	100	100
Chlorides (ppm)	ND	ND	649.8	59.9	39.9	200	100
COD (ppm)	3000	3200	820	780	800	NA	80
BOD (ppm)	980	765	245	185	215	NA	10

NA not applicable

^aValues represent averages of three replicate determinations

Table 2 Heavy metal composition of samples

Metal (ppm) ^a	Alaro River	Ikeja sediment	Isolo soil	WHO recommended limit	FEPA maximum allowed limit
Chromium	0.02	5.54	7.18	0.05	< 0.1
Lead	0	2.46	1.34	0.01	0.05
Cadmium	0.04	0.35	14.18	0.003	< 1
Nickel	0.05	0.95	12.67	0.02	< .05

^aValues represent averages of three replicate determinations

strict aerobic organisms with the emergence of more anaerobes, micro-aerophiles, and facultative anaerobes.

Apart from lead that was not detected in the water sample from Alaro River, the concentrations of the heavy metals obtained for the samples generally exceeded maximum allowed limits (Table 2). However, contrary to the data summarized in Table 1, Ikeja wastewater sediment and Isolo soil exhibited higher concentrations of all metals assayed. Of significance is the finding that soil samples from Isolo industrial estate contained high concentrations of chromium (7.18 ppm), lead (1.34 ppm), cadmium (14.18 ppm), and nickel (12.67 ppm) being predominant metals associated with textile and battery industries, many of which are sited in this locality. The presence of high concentration of these metals is undesirable as the long-term effect may be injurious to human health. The concentration buildup of the metals must be due to decades of production and indiscriminate discharge of these toxic metals into the environment. The age-long exposure of the industrial environment to heavy metal contamination could have affected the ecosystem, wiping out sensitive fauna and flora, thus, giving rise to evolution of some heavy-metal-resistant organisms. Several mechanisms have been reported through which this kind of evolution could take place.

Over 270 species of bacteria were initially isolated from the samples by selective enrichment protocols. Most of the isolates were taken from soil sample obtained from Ikeja industrial estate (Fig. 2). Of this, 22 strains showed resistance to elevated concentrations of a broad range of metals (Fig. 3), but the thresholds are different (Table 3). The metal-resistant profiles ranged significantly from 1 to 14 mM for cadmium, 6 to 15 mM for cobalt and nickel, 5 to 17 mM for chromium, and 1 to 10 mM for mercury

(Table 3). Strains that showed resistance to metals as determined by the plate assay were also resistant to the same metals in broth cultures. The antibiotic resistance patterns of the isolates are shown in Fig. 4. These strains combine ability to resist metals with antibiotic resistance as illustrated in Table 3 and were also able to utilize a wide range of xenobiotic compounds (data not shown). Ampiclox, zinnacef, and metronidazole were by far the most common antibiotics resisted among the isolates where ampiclox and zinnacef were the most resisted, while sparfloxacin, pefloxacin, pefloxacin, and tarivid were the most active against the isolates (Fig. 4). Among the 22 isolates, only five strains, namely, CA207Ni (*Pseudomonas aeruginosa*), AL36Cd (*Actinomyces turicensis*), CA92Cd (*Nocardia* sp.), CA151Cd (*Nocardia* sp.), and AL06Ni (*Micrococcus* sp.) exhibited resistance to all the 18 antibiotics tested. Strain AL05Ni (*Rhodococcus* sp.) also showed equal level of resistance with the exception of tarivid. These six isolates were

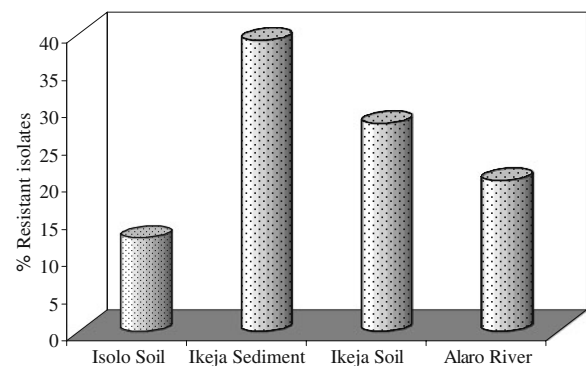


Fig. 2 Occurrence of heavy-metal-resistant bacterial isolates from sampling sites. Data presented were averages of three replicate samples obtained from each site. Heavy metal assayed were Cd²⁺, Hg²⁺, Co²⁺, Ni²⁺, and Cr⁶⁺

Table 3 Heavy-metal resistance dynamics of isolates

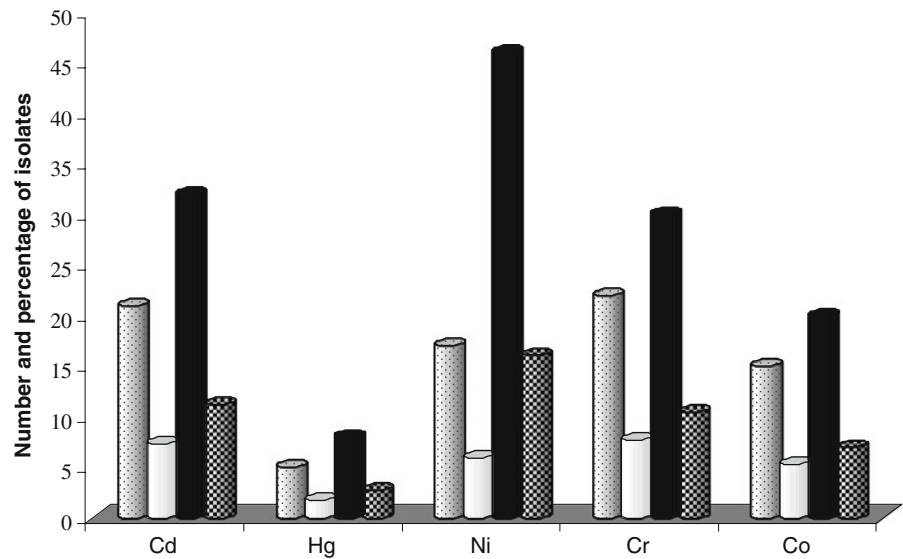
Isolate	Origin	Maximum tolerance concentration (mM)					No. of antibiotic resisted	Putative identification
		Cd ²⁺	Hg ²⁺	Co ²⁺	Ni ²⁺	Cr ⁶⁺		
AL251Cr	Alaro	1	<1.0	<6.0	<6.0	ND	8	<i>Burkholderia</i> sp.
CA114Cr	Ikeja	3	1	6	15	12	9	<i>Nocardia</i> sp.
AL252Cr	Alaro	3	1	6	6	5	11	<i>Streptomyces</i> sp.
CA109Cr	Ikeja	1	1	6	6	9	3	<i>Acinetobacter junni</i>
AL94Co	Alaro	10	1	14	6	8	1	<i>Micrococcus</i> sp.
AL95Co	Alaro	1	1	15	6	7	1	<i>Flavobacterium</i> sp.
AL96Co	Alaro	7	10	15	15	6	9	<i>Burkholderia cepacia</i>
CA56Co	Ikeja	2	1	15	15	10	5	<i>Corynebacterium ulcerans</i>
CA60Co	Ikeja	10	1	7	6	10	3	<i>Rhodococcus</i> sp.
FL108Hg	Isolo	10	10	15	15	17	11	<i>Corynebacterium kutscheri</i>
AL05Ni	Alaro	2	1	6	15	5	17	<i>Rhodococcus</i> sp.
CA207Ni	Ikeja	10	1	10	15	12	18	<i>Pseudomonas aeruginosa</i>
AL36Cd	Alaro	10	1	6	6	7	18	<i>Actinomyces turicensis</i>
CA92Cd	Ikeja	10	1	6	6	6	18	<i>Nocardia</i> sp.
CA151Cd	Ikeja	10	1	7	6	7	18	<i>Nocardia</i> sp.
AL06Ni	Ikeja	6	1	6	15	6	18	<i>Micrococcus</i> sp.
CA215Hg	Ikeja	10	7	12	15	12	3	<i>Alcaligenes</i> sp.
CA202Ni	Ikeja	6	1	6	7	5	3	<i>Corynebacterium</i> sp.
AL80Ni	Ikeja	2	1	9	15	4	12	<i>Pseudomonas</i> sp.
AL32Cd	Ikeja	14	1	6	15	10	4	<i>Rhodococcus</i> sp.
AL03Ni	Ikeja	10	10	6	15	10	11	<i>Rhodococcus</i> sp.
CA57Co	Ikeja	2	1	15	15	4	5	<i>Corynebacterium</i> sp. group G.

particularly resistant to higher doses of cadmium (2–10 mM) and chromium (6–17 mM) while strains CA207Ni (*P. aeruginosa*) and AL06Ni were particularly resistant to over 15 mM of nickel with the former exhibiting resistance to 10 mM of cobalt (Table 3). In a relatively recent study, Kimiran-Erdem et al. (2007) demonstrated dual resistance of 100 species of environmental enterococci to zinc, iron, cadmium, cobalt, chromium, and some antibiotics. However, the MICs recorded for these metals were much lower compared to over 100-fold concentrations that our isolates resisted. Similarly, Ben Said et al. (2007) documented multiple polycyclic aromatic hydrocarbon-degrading bacterial strains isolated from Bizerte lagoon sediments, in Tunisia with capacity to resist some heavy metals and antibiotics. Unlike our isolates and those of Kimiran-Erdem et al. (2007), the authors failed to show the concentrations of metals resisted, but this could be several orders-of-magnitude lower than we have reported (Table 3).

Many researchers believed that combination of antibiotic and metal resistance may not be a

fortuitous phenomenon (Nakahara et al. 1977; Calomiris et al. 1984), rather bacterial resistance against heavy metals appeared to be directly related to the presence of these elements as environmental contaminants (De Vicente et al. 1990; Silva and Hofer 1993; Raja et al. 2006). Interestingly, the same metals which our isolates showed striking resistance to, are elements of concern to the environment at concentrations which have never been reported to the best of our knowledge. Perhaps, the uniqueness of this study is the discovery of four of our isolates which unusually resisted up to 10 mM of mercury. It should be noted, however, that mercury is reputed as the sixth most toxic in a universe of six million substances (Nascimento and Chartone-Souza 2003) and has been found to be toxic at extremely low concentrations to most microorganisms. These isolates were subsequently phenotypically characterized and identified as *P. aeruginosa*, (CA207Ni); *Burkholderia cepacia* (AL96Co); *Corynebacterium* group G (CA57Co); *Corynebacterium kutscheri* (FL108Hg), and *Micrococcus* sp. (AL03Ni). This resistant metal

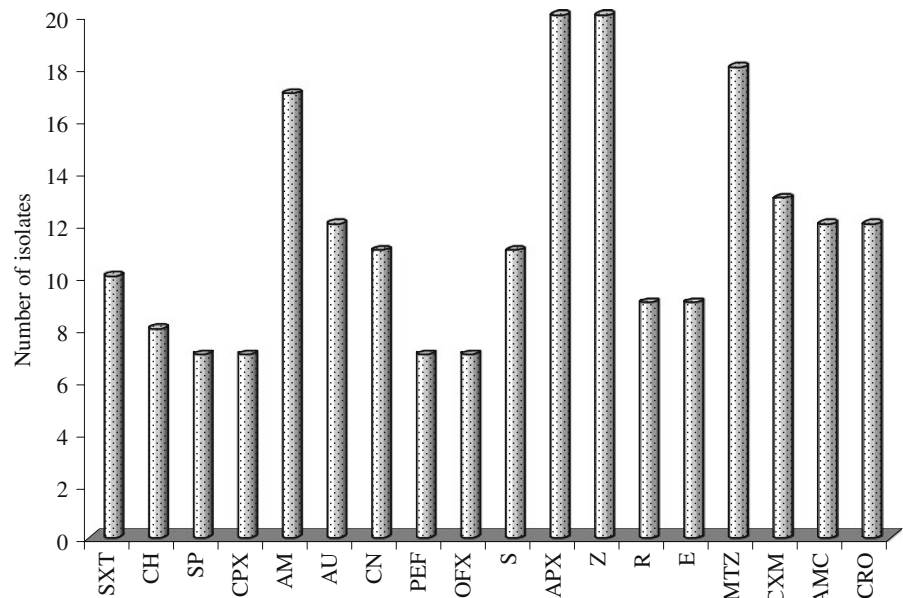
Fig. 3 Resistant patterns of bacterial isolates. Number (*dotted bar*) and proportion (*empty bar*) of isolates resistant to elevated concentrations metals; total number (*filled bar*) and proportion (*checkered bar*) of isolates resistant to metals. Values presented are means of triplicate determinations



dosage is remarkable and previously unreported even though their antibiotic-resistance patterns are limited, except for strains *C. kutscheri* and *Micrococcus* sp. (AL03Ni). For instance, Sprocati et al. (2006) reported resistant concentrations of 0.025 and 0.05 mM of mercury. It is noteworthy that among the five strains which showed marked resistance to mercury, only two, *C. kutscheri* (FL108Hg) and *Alcaligenes* sp. (CA215Hg), were isolated on nutrient-rich medium fortified

with differential doses of mercury, while others had their medium supplemented with either cobalt or nickel contrary to previous isolation techniques. Mercury pollution can contribute to increased antibiotic resistance, and it has been demonstrated that the combined expression of antibiotic resistance and mercury may be caused by selection, as a consequence of the mercury present in the environment (Sant'ana et al. 1989). It is, therefore, not surprising that those strains

Fig. 4 Antibiotic resistance pattern. SXT septrin; CH chloramphenicol; SP sparfloracin; CPX ciprofloxacin; AM amoxicillin; AU augmentin; CN gentamycin; PEF pefloxacin; OFX Tarivid; S streptomycin; APX ampiclox; Z, zinnacef; R, rocephin; E, erythromycin; MTZ metronidazole; CXM cefuroxime; AMC amoxicillin; CRO ceftriaxone



which exhibited resistances to high concentrations of metals were particularly resistant to most of the antibiotics tested.

Conclusion

The industrial use of mercury, cadmium, nickel, and other heavy metals have led to the pollution of the environment. Consequently, removal of these bad metals is a challenge for environmental management. Biological processes have been employed in bioremediation, including metal recovery, and are potentially low cost. Therefore, heavy-metal-resistant organisms may help play a major role in natural decontamination. The isolation of these strains from effluents and contaminated systems has considerable ecological advantage. The metabolic active system of metal resistance by our isolates is novel and represents a point of interest for possible environmental applications by promoting mobilization and loss of toxic metals and potentially dangerous organic components from the environment. They can help to overcome metabolic bottlenecks still existing in the biodegradation processes such as the inhibition that heavy metals can exert on the biodegradation of organic pollutants most especially in matrices where they occur as co-contaminants. However, it seems that this ecological advantage carries with it the possibility of an increase in the gene pool for antibiotic resistance. This would be a survival strategy for these organisms to proliferate ahead of others in the environment, since most possible antibiosis that could exist in such sites would not affect them. This could be a very important feature going by the report of Sarmah et al. (2006) that the antibiotic concentrations in some environments have been found higher enough to inhibit many bacterial species.

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