Comparative Microbiological and Physicochemical analysis of Domestic and Industrial wastewater in Ado-Odo Ota LGA, Ogun State, Nigeria

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Abstract

Wastewater from industrial and domestic sources flows into major bodies of water where people drink and bathe, resulting in a high incidence of severe water-borne infections. This study aims to compare the microbiological and physicochemical parameters of domestic and industrial wastewater in Ado-Odo LGA, Ogun State, Nigeria. Microorganisms were isolated from collected wastewater samples and were phenotypically identified using standard microbiological methods. Antimicrobial Sensitivity testing was carried out using Kirby Bauer`s disk diffusion technique. The physicochemical analysis of the water samples was also determined using standard analytical methods. A total of nineteen microorganisms were isolated from the samples collected with the predominant organism isolated from the two sources being *Escherichia coli*. The microorganisms isolated from the industrial wastewater sample were *Citrobacter freundii* (37.5%), *Escherichia coli* (37.5%), and *Proteus vulgaris* (25.0%) while those isolated from the domestic wastewater sample were *Klebsiella oxytoca* (18.18%), *Escherichia coli* (45.45%), *Proteus vulgaris* (9.09%), *Staphylococcus saprophyticus* (9.09%) and *Staphylococcus aureus* (18.18%). The physicochemical analysis conducted indicated that there were lower levels of impurities in industrial wastewater pH (4.1 ± 0.14), Electrical conductivity (479 ± 1.41), Total Dissolved Solids (223.5 ± 3.53), and Chloride content (168.12) compared to the domestic wastewater with pH (8.05 ± 0.07), Electrical Conductivity (1928 ± 4.24), Total Dissolved Solids (765.5 ± 0.70) and Chloride content (238.224) which suggests that industrial wastewater has been subjected to series of chemical and biological processes. The presence of *Escherichia coli* in higher levels in domestic wastewater indicates fecal contamination which is a major public health challenge since this wastewater leaks into various water bodies.

Keywords: Domestic wastewater, Microbiological analysis, Industrial wastewater, Physicochemical analysis

1. Introduction

Wastewater has been a major source of waterborne pathogens and is classified as sewage wastewater and non-sewage wastewater [1]. Sewage wastewater includes discharge from domestic activities such as schools, restaurants, hospitals, houses, hotels, and public toilets containing body wastes (urine and feces). Non-sewage wastewater is referred to as the other types of wastewater generated from flooding, stormwater, and commercial activities such as from industrial plants and factories. According to Munter (2003), industrial wastewater contains dissolved and suspended substances discharged from processes such as manufacturing and cleaning. The nature of contaminants found in industrial wastewater depends on what is being produced in such an industry. Examples of industries that produce wastewater include the mining industry, power plants, industrial laundries, steel/iron production plants, metal finishers, oil and gas fracking plants, and the food/beverage industry. The commonly found contaminants in industrial wastewater outlets are chemicals, pesticides, heavy metals, silt, oils, pharmaceuticals, and other industrial by-products [2]. Direct release of untreated or improperly treated hazardous effluents in the sewerage drains eventually pollutes the groundwater as well as other major water bodies such as the oceans, seas, rivers, and even streams causing detrimental health effects on humans who drink, feed on the aquatic animals or bathe in/with such water sources, animals that live in them as well as the aquatic life. The discharge of effluent that are not adequately treated into the environment could lead to different types of environmental pollution. Casual disposal of industrial wastewater can also impact agriculture negatively in the area of crop irrigation by leading to poor quality and poisonous crop yields which can also reach the food chain [3]. The pollution of water bodies by these effluents could result into waterborne diseases such as diarrhea, typhoid, gastroenteritis, amoebic dysentery, giardiasis, cholera, hepatitis among many others [4] jaundice [5], and cancer [6]. Waste water management and treatment is therefore very crucial and necessary because daily human activities are majorly water dependent.

Domestic wastewater can be described as the water produced by human household activities which are generally of two main sources: toilet waste, i.e. the liquid released from laundry/sanitary or bathing facilities, and the wastewater generated from all other household activities such as cooking[7] Depending on the source, domestic wastewater is classified into three: black, grey and yellow wastewater [8]

Black wastewater is majorly wastewater from toilets. It is largely composed of organic compounds which makes decomposition fast and readily available to microorganisms [9] Grey water is wastewater generated from other domestic processes such as laundry, washing of dishes, and bathing [10] The name Grey wastewater was generated from its cloudy appearance and its nature as being neither fresh (whitewater) from groundwater or potable water nor heavily polluted (blackwater) Though this used water may contain impurities or contaminants such as food particles, grease, hair, and myriads of other impurities which may still be suitable for reuse [11]. Wastewater collected from processes in commercial building welling units, and institutions of the community can also be referred to as grey water. It is spent water from the building water supply which has been added to the waste effluent of bathrooms, kitchens, and laundry [12]

Yellow water is specifically composed of urine without any other contaminants of blackwater and greywater. In yellowwater no toilet paper, fecal matter, chemicals, or even any food particles is present.

Wastewater especially from domestic and industrial sources has been a major health concern regarding waterborne pathogens which leak into rivers and ocean bodies where people drink from, cook with and even bathe with thus leading to a high occurrence of water-related infections. Some of these organisms could also get into the environment where they are inhaled and could cause severe infections such as respiratory tract infections, especially in individuals with suppressed immune systems. Therefore, this research was carried out to comparatively assess the microbiological and physicochemical properties of industrial and domestic wastewater.

2.0 Materials and methods

2.1 Study location

The study was carried out in Sango Ota and Igbesa in the Ado-Odo Ota Local Government of Ogun state. Ado-Odo Ota is a veritable industrial Local Government that has the largest industrial area and the highest number of industries in the state.

2.2 Sample collection

Sampling was done during morning hours and all water samples were collected in sterile polyethylene terephthalate bottles. The bottles were dipped in the effluent to enable easy flow into the bottles. Samples were collected from three different points at the depth of 10 cm.

2.3 Isolation of bacteria from samples

One milliliter of each wastewater sample was serially diluted to 10^5 dilution. Then one milliliter of the serially diluted sample was inoculated into sterilized Nutrient agar, Eosin methylene blue (EMB) agar, Maconkey agar, and Mannitol salt agar and incubated aerobically at 37^0C for 18-24 h.

2.4 The phenotypic identification of the bacterial isolates

Smears were made using normal saline from 18-24 hours culture plates and heat-fixed by passing through the flame 3-4 times. Dried smears were first stained with crystal violet solution for one minute, then the smear is washed under slow-running tap water, then Lugol's iodine was added for one minute and was washed under slow-running tap water. The smear was then decolorized with acetone and the slides were washed immediately. The slides were then counterstained with safranin for one minute. Slides were arranged on the rack and allowed to dry. Dried stained slides were then examined under the microscope using x100 objectives (oil immersion)

Bacteria isolates from industrial and domestic samples were identified according to the methods of Cowan and Steel [13] Preliminary identification was based on colonial morphological characteristics of isolated organisms. Further identification was done based on their Gram's reactions and was classified into Gram positives and Gram-negatives according to their cell wall components and morphology.

Gram-positive bacteria were characterized by a biochemical test (catalase test). Catalase negative Gram positive cocci (GPC) were grouped as *Streptococci*. Catalase-positive organisms were further characterized by the coagulase test. Coagulase-positive Gram-positive cocci were identified as *Staphylococcus aureus* and Coagulase Negative Gram-positive cocci were identified as Coagulase Negative Staphylococci (CoNS). Gram-positive bacteria. Novobiocin test was carried out on Coagulase-negative *Staphylococcus*.

2.4 MICROBACT identification of the bacteria isolates

The Oxoid MICROBACT Gram-negative system was used to identify aerobic and facultative anaerobic Gram-negative bacteria (Enterobacteriaceae and miscellaneous GNB). This system is a standardized micro substrate system designed to simulate conventional biochemical substrate used for the identification of Enterobacteriaceae and miscellaneous Gram-Negative Bacilli (MGNB). Identification is based on pH change and substrate utilization). The Microbact 12A strip was used alone for the identification of oxidase-negative, nitrate-positive glucose fermenters. The inoculated strips were incubated for 24 hours and interpreted using the octal coding system adopted for Microbact.

2.5 Antimicrobial sensitivity tests

Antimicrobial sensitivity tests were carried out using the Kirby–Bauer disc diffusion method. Results were interpreted according to the Clinical and Laboratory Standards Institute (CLSI) guidelines [14A 0.5 McFarland standard inoculum was prepared and inoculated on the Mueller–Hinton Agar plate. The appropriate antibiotic disc containing antibiotic discs including Tetracycline (30 µg), Ciprofloxacin (5 µg), Cefotaxime (30 µg), Chlorpromazine (30 µg), Cotrimoxazole (5 µg), Cefuroxime (30 µg), Chloramphenicol (30 µg), Meropenem (10 µg), Ceftriaxone (30 µg), Gentamicin (10 µg), Amikacin (30 µg). were placed onto the media and incubated at 37 °C for 16–18 h. Zones of inhibition were read and resistance rates to respective antibiotics were determined as per the CLSI guidelines

2.6 Determination of physicochemical parameters in the samples

The physical parameters of the collected water samples that were measured in this study were pH and TDS. The pH and TDS of the water samples were determined using the Hach pH meter (ModelEC1O) and total dissolved solids were measured in situ using the Hach conductivity meter (ModelCO150) respectively.

The chemical analysis of the collected water samples measured in this study was chloride content and electrical conductivity through the titration (Argentino) method of Standard Methods for the Examination of Water and Wastewater American Public Health Association [15].

2.7 Statistical Analysis

All data were entered into a secure database and imported into excel for data cleaning and analysis.

3.0 Results and discussion

3.1 Results

A total of 19 organisms were isolated from the samples collected out of which (84.21%) were Gram negatives and (15.79%) were Gram positives.

The organisms isolated were: *Escherichia coli* (42.10%), *Citrobacter freundii* (15.79%), *Proteus vulgaris* (15.79%), *Staphylococcus aureus* (10.53%), *Staphylococcus saprophyticus* (5.23%), *Klebsiella oxytoca* (10.53%) shown in Figure 1

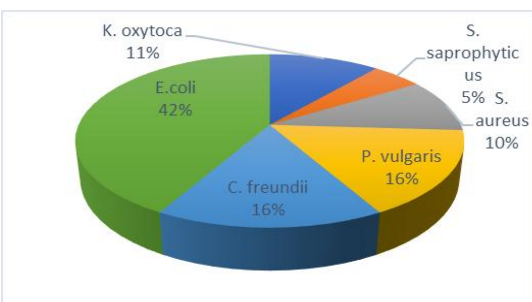

Fig. 1: Percentage distribution of bacterial isolates in the wastewater samples

The prevalence of microorganisms in the industrial and domestic wastewater samples is presented as shown in Table 1. The organisms isolated in industrial wastewater samples were *Citrobacter freundii* (37.5%) *Escherichia coli* (37.5%), and *Proteus vulgaris* (25.0%) while those isolated from domestic wastewater samples were *Klebsiella oxytoca* (18.18%), *Escherichia coli* (45.45%), *Proteus vulgaris* 1(9.09%), *Staphylococcus saprophyticus* (9.09%) and *Staphylococcus aureus* (18.18%)

Table 1: Percentage prevalence of microorganisms in industrial and domestic wastewater

Sample	Bacterial isolates	Percentage (%)
Industrial	*Citrobacter freundii*	37.5%
	Escherichia coli	37.5%
	Proteus vulgaris	25.0%
Domestic	*Klebsiella oxytoca*	18.18%
	Escherichia coli	45.45%
	Proteus vulgaris	9.09%
	Staphylococcus saprophyticus	9.09%
	Staphylococcus aureus	18.18%

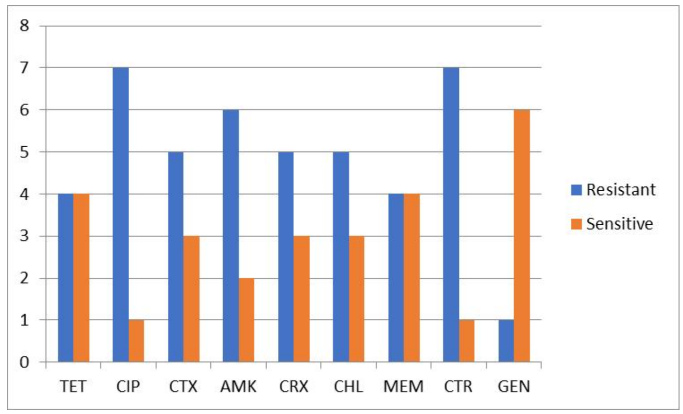

Figure 2: Antibiogram pattern of *E. coli*
Key: TET-Tetracycline, CIP-Ciprofloxacin, CTX-Cefotaxime, AMK-Amikacin, CRX-Cefuroxime, CHL-Chloramphenicol, MEM-Meropenem, CTR- Ceftriaxone, GEN-Gentamicin

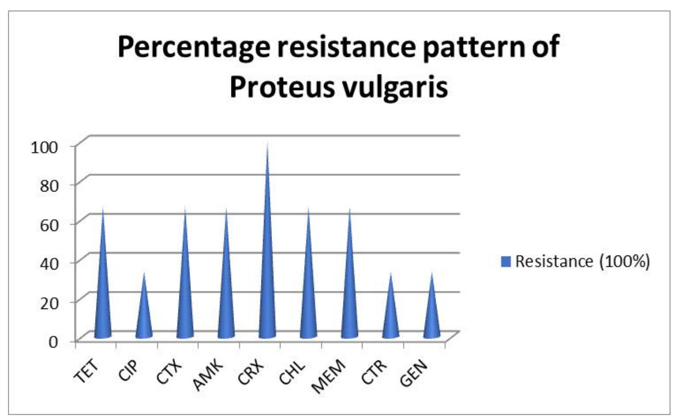

Figure 3: Percentage resistance pattern of *Proteus Vulgaris*
Key: TET-Tetracycline, CIP-Ciprofloxacin, CTX-Cefotaxime, AMK-Amikacin, CRX-Cefuroxime, CHL-Chloramphenicol, MEM-Meropenem, CTR- Ceftriaxone, GEN-Gentamicin

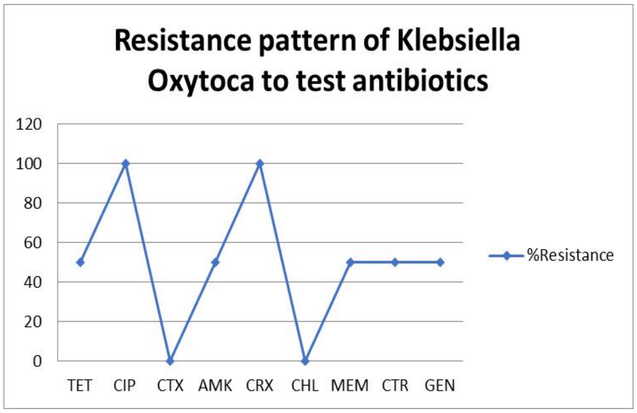

Figure 4: Resistance pattern of *Klebsiella oxytoca* to test antibiotics

Key: TET-Tetracycline, CIP-Ciprofloxacin, CTX-Cefotaxime, AMK-Amikacin, CRX-Cefuroxime, CHL-Chloramphenicol, MEM-Meropenem, CTR- Ceftriaxone, GEN-Gentamicin

Table 2. Determination of physicochemical parameters

Source	pH	EC	TDS	CC (ppm)
Industrial	4.1 ± 0.14	479 ±1.41	223.5 ±3.53	168.12
Cafeteria	7.1 ± 01.4	1590 ±14.14	880.5 ±27.57	130.45
Bathroom	8.05 ±0.07	1928 ± 4.24	765.5 ± 0.70	238.224
Standard (WHO)	6.5-8.5	300	500	250

Key: pH**,** EC- Electrical conductivity, TDS-Total dissolved Solids CC-Chloride content

3.2 Discussion of Results

Domestic and industrial wastewater is one of the major contributors to wastewater generated from human and industrial activities in Nigeria. From the results generated, the organism with the highest prevalence is *E.coli* (42.1%) and the least prevalent is *S. saprophyticus* (5.3%). These bacteria have previously been reported as wastewater pathogens. Some of the isolated microorganisms from this research have been reported to be among the six globally leading antimicrobial-resistant bacteria[16]. Microbial contamination may pose a mild to severe life threatening challenge to health if untreated wastewater comes into contact with humans. The isolated microorganisms may release toxins from the effluents into the environment which could be dangerous.

Discharge of these organisms, *Citrobacter freundii*, *Klebsiella oxytoca,* and *E. coli* which belong to the Enterobacteriaceae family into wells could be potentially dangerous for people who drink from such contaminated well water.

The predominant organism detected in the bathroom wastewater was *Escherichia coli* which usually is indicative of fecal contamination but could in this case be suggestive of a high level of urinary tract infection in people who urinate in the bathroom during bathing. *S.saprophyticus* is also associated with urinary tract infections.

The presence of *Staphylococcus aureus* could be from human beings since it is part of the normal flora of the skin, nose, throat, mucus membranes, and gut, and the escape of the organism into the industrial and cafeteria wastewater samples and could pose a high risk of foodborne diseases when ingested in contaminated food.

The electrical conductivity from domestic wastewater recorded a higher value when compared with industrial wastewater. The significant difference in the electrical conductivity values maybe is attributed to the total number of dissolved ions in the detergents used for different cleaning activities. This is in congruence with the research carried out by [12]

The pH of the domestic wastewater was neutral (7.0-8.1), which plays a role in determining both the qualitative and quantitative abundance of microorganisms in the wastewater. This is in line with the limit for wastewater discharge of the Federal Environmental Protection Agency, Nigeria for domestic wastewater[17].

The total dissolved solids of the cafeteria wastewater sample were higher than that of the bathroom wastewater. While the chloride levels were higher in the bathroom sample which may be a result of the type of detergent or soap used. It has been shown that most commercially available detergents or soaps are currently manufactured using various types and quantities of sodium salt.

The bacterial isolates showed a high rate of resistance to most of the test antibiotics which is a major concern to the health sector.

4. Conclusion

Wastewater is fast becoming a source of antibiotic-resistant organisms which is a major public health challenge. In terms of microbial risks, Industrial wastewater produces more isolates of resistant organisms than domestic wastewater and this could be due to the high levels of organic and inorganic matter present.

References

[1]. Chahal, C., van den Akker, B., Young, F., Franco, C., Blackbeard, J. and Monis, P. (2016). Pathogen and Particle Associations in Wastewater: Significance and Implications for Treatment and Disinfection Processes. Advances in applied microbiology, 97, 63–119. https://doi.org/10.1016/bs.aambs.2016.08.001

[2]. Azimi, A, Azari, A, Rezakazemi, M and Ansarpour, M.(2017). Removal of heavy metals from industrial wastewaters: a review, ChemBio Eng Reviews 4, 37 -59

[3]. Iqbal, M., Nauman, S., Ghafari, M., Parnianifard, A., Gomes, A. and Gomes, C. (2021). Treatment of Wastewater for Agricultural Applications in Regions of Water Scarcity. Biointerface Research in Applied Chemistry, 12, 6336-6360. 10.33263/BRIAC125.63366360

[4].Wang, Q. and Yang, Z. (2016) Industrial water pollution, water environment treatment, and health risks in China, Environmental Pollution 218, 358 -365

[5]. Rathour, R. K., Sakhuja, D., Bhatt, A. K., and Bhatia, R. K. (2022). Municipal Wastewater Connection for Water Crisis and Jaundice Outbreaks in Shimla City: Present Findings and Future Solutions. International journal of environmental research and public health, 19(18), 11266 https://doi.org/10.3390/ijerph191811266

[6]. Lin, L., Yang, Haoran and Xu, X.(2022). Effects of Water Pollution on Human Health and Disease Heterogeneity: A Review, Frontiers in Environmental Science.10 https://www.frontiersin.org/articles/10.3389/fenvs.2022.880246

[7]. Widyarani, Wulan, D.R., Hamidah, U. Komarulzaman, A., Rosmalina, R. T., and Sintawardaniet, N. (2022). Domestic wastewater in Indonesia: generation, characteristics and treatment. Environ Sci Pollut Res 29, 32397–32414 https://doi.org/10.1007/s11356-022-19057-6

[8].Friedler, E., Butler, D and Alfiya, Y. (2013). Wastewater composition, Source Separation and Decentralization for Wastewater Management , IWA Publishing, London, UK, page. 241–257

[9]. Ghaly, A. Mahmoud, N., Ibrahim, M., Mostafa, E., Rahman, E., Hassanien, R., Kassem, M. and Hatem, M. (2021). Greywater Sources, Characteristics, Utilization and Management Guidelines: a review. 4. 128-145

[10]. Oteng-Peprah, M., Acheampong, M. A., and deVries, N. K. (2018). Greywater Characteristics, Treatment Systems, Reuse Strategies and User Perception-a Review. Water, air, and soil pollution, 229(8), 255 https://doi.org/10.1007/s11270-018-3909-8

[11]. Dwumfour-Asare, B., Nyarko, K.B., Essandoh, Esi Awuah, M.K., Kofi, K. A. Anim, A. Q. (2018). Greywater in the drains of a sewered community in Ghana. Water Practice and Technology 1 December 13 (4), 965–979. doi: https://doi.org/10.2166/wpt.2018.103

[12].Eze, V. C., Onwuakor, C.E. and Mgbeokwere, E.U. (2015). Comparative Analysis of the Microbiological and Physicochemical Characteristics of Greywater Sources in Off-Campus Hostels at Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria, Int. J. Curr.Microbiol.App.Sci 4(8), 196-205

[13]. Gunathilaka, N., Ranasinghe, K., Amarasinghe, D., Rodrigo, W., Mallawarachchi, H., and Chandrasena, N. (2020). Molecular Characterization of Culturable Aerobic Bacteria in the Midgut of Field-Caught Culex tritaeniorhynchus, Culex gelidus, and Mansonia annulifera Mosquitoes in the Gampaha District of Sri Lanka. BioMed research international, 2020, 8732473. https://doi.org/10.1155/2020/8732473

[14]. CLSI. (2016). Performance Standards for Antimicrobial Susceptibility Testing; TwentyFourth Informational Supplement. CLSI document M100-S24. Wayne, PA: Clinical and Laboratory Standards Institute

[15].American Public Health Association (APHA) (2005). Standard Methods for the Examination of Water and Wastewater 21st ed., Washington DC: American Public Health Association

[16]. Muray, C. J. L., Ikuta, K. S., Sharara, F., Swetschinski, L., and Collaborators, A. R. (2022). Global burden of bacterial antimicrobial resistance in 2019 : A systematic analysis. Lancet 399, 629–655. doi: 10.1016/S0140-6736(21)02724-0

[17]. Kayode, O.F., Luethi, C. and Rene, E.R. (2018). Management Recommendations for Improving Decentralized Wastewater Treatment by the Food and Beverage Industries in Nigeria. Environments, 5, 41. https://doi.org/10.3390/environments5030041