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Quality Assessment of Wastewater: Physicochemical and Bacteriological Evidence from Dutse Abattoir, North-West Nigeria

Garba Barde Bate¹, Adeniyi Olarewaju Adeleye^{1*}, Emmanuel Madu Ijanu¹, Esther Oluwafunmilola Olalere¹, Afeez Oladeji Amoo¹, Catherine Iyabo Asaju¹, Peter Gimba Shiaka², Mohammed Bello Yerima³

¹Department of Environmental Sciences, Federal University Dutse, Dutse, Jigawa State, Nigeria. ²Department of Microbiology and Biotechnology, Federal University Dutse, Dutse, Jigawa State, Nigeria.

³Department of Microbiology, Sokoto State University, Sokoto, Sokoto State, Nigeria.

ABSTRACT

This study was conducted to determine the physicochemical properties of the wastewater as well as isolate and identify heterotrophic bacterial populations in the wastewater while comparing results obtained with permissible standards of the World Health Organisation (WHO) and the National Environmental Standards and Regulations Enforcement Agency (NESREA). Grab sampling method was used to collect 12 wastewater samples over four weeks. Temperature, pH, turbidity, electrical conductivity (EC), nitrates, sulfate, phosphate, dissolved oxygen (DO), chemical oxygen demand (COD), biochemical oxygen demand (BOD), and total heterotrophic bacterial count (THBC) were analyzed using standardized methods. Results obtained show that the overall mean values of pH (6.91), EC (2.6 μ S/cm), sulfate (81.7 mg/L), DO (7.05 mg/L), BOD (1310 mg/L) and COD (3543 mg/L) conformed with allowable limits. Nitrate (821.4 mg/L) and phosphate (231.7 mg/L) were found to be above the permissible standards. Mean BOD values were not significantly different (p > 0.05) from each other during sampling. THBC for all the samples exceeded the WHO limit, as it ranged between 1.9×10^4 and 5.6×10^2 CFU/mL. Results obtained during the conduct of this study have clearly shown that the wastewater generated from the Dutse abattoir was unfit for discharge into the environment.

Keywords: Abattoir, Biochemical oxygen demand, Chemical oxygen demand, Total heterotrophic bacterial count, Wastewater

Corresponding author: Adeniyi Olarewaju Adeleye e-mail ⊠ adeniyi.adeleye@fud.edu.ng Received: 02 June 2023 Accepted: 14 September 2023

INTRODUCTION

Wastewater is the antagonistic outcome of human influence against the aquatic environment, which can originate from homes and industrial settings (Edokpayi et al., 2017). The authors further submitted that wastewater can house loads of microorganisms, pharmaceutical products, heavy metals, nutrients, and other waste products that might have found their way into the aquatic environment. However, the composition of all wastewater is continuously fluctuating and extremely inconstant, which is why it is so problematic to find a stable description of it (Tuser, 2020; Kirilmaz, 2022). Wastewater is a multifaceted medium comprising substantial concentrations of total solids that range between 350 and 1200 mg/L, chemical oxygen demand (COD) that ranges between 250 and 1000 mg/L, microorganisms that can add up to 109 number/mL, some nutrients, micro-pollutants and considerable heavy metals (Kundu & Dutta, 2018; Almulhim et al., 2022; Alshammari, 2022).

Canneries, milk dairies, sugar factories, breweries and distilleries, the beverage and meat industries, the manufacture

of fertilizer, pulp and paper, tanneries, and yeast manufacturing are the main industrial sources of wastewater (Micronics Engineered Filtration Group, 2023). When people consume more meat, the meat industry contributes significantly to the volume of wastewater produced. This means that wastewater containing a lot of bacteria is continuously produced, which has an impact on the receiving environment in several ways, including the buildup of toxic chemicals in the soil, the contamination of surface and groundwater, the release of greenhouse gases that cause global warming, and many other effects (Lapygina *et al.*, 2002). Failure to adequately treat wastewater before final disposal into the environment normally leads to the growth of pathogenic microorganisms uncontrollably in water bodies (Adeleye *et al.*, 2021; Shetgaonkar, 2022).

Most industrial processes necessitate a tremendous amount of water, which almost equally discharges wastewater (Negi & Suthar, 2018). These authors reported further that industries dependent on freshwater for their production have consistently contributed to the pollution of the environment. Untreated wastewater originating from both industrial and residential sources frequently contains a sizable number of contaminants, nutrients, and biological agents (Manasa *et al.*, 2020). Owing to the increasing number of people who consume meat products, meat industries are categorized among those industries capable

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of generating high volumes of wastewater (Bustillo-Lecompte & Mehrvar, 2017). The slaughterhouse wastewater has harmful levels of nitrogen, phosphorus, and organic carbon that are primarily contributed by animal dung (Matheyarasu et al., 2015; Pushkin, 2023). These authors further reiterated that the highest organic load concentration is seen in the wastewater stream from slaughterhouses, which also has higher COD (8000 mg/L), proteins (70%), and suspended particles (15-30 mg/L) concentrations than other wastewater sources. The continuous drive to increase meat production for the protein needs of the ever-increasing world population can equally be attributed to the pollution problems attached (Milman, 2021; Terela & Strilets, 2023). This author reiterated further that in many countries, pollution arises from activities in meat production as a result of failure to adhere to Good Manufacturing Practices (GMP) and Good Hygiene Practices (GHP). Consideration is hardly given to safety practices during animal transport to the abattoir, during slaughter, and dressing. Also, during meat production, wastewater is produced in large quality and then stored in septic tanks for a long period and then evacuated.

In the Dutse abattoir, wastewater generated daily is not subjected to any form of treatment before disposal into the environment. When this wastewater that is hardly treated gets discharged into the environment indiscriminately, it subsequently flows through the public drainage, thereby polluting the environment. A considerable percentage of abattoir wastewater is employed for irrigating agricultural farmland and as feed for fish farming. However, owing to the enormous microbial load inherently found in wastewater, if it is discharged into the environment untreated, it can result in contamination of soil and water, percolation of soil, methane emission, and many more. This study was conducted to determine the physicochemical properties of the wastewater as well as isolate and identify heterotrophic bacterial populations in the wastewater while comparing results obtained with permissible standards of the World Health Organisation (WHO) and the National Environmental Standards and Regulations Enforcement Agency (NESREA).

MATERIALS AND METHODS

Study area

Dutse abattoir is located in the capital of Jigawa state, northern Nigeria. It lies north of the road between Kano City and Birnin Kudu. The undulating relief of the area is covered by Sudan's savannah. Dutse is a city located in northwestern Nigeria. It is the capital city of Jigawa state. It is home to Federal University Dutse, which opened in November 2011. The abattoir is situated on latitude 11°43'10" N and longitude 9°21'38" E. The number of animals slaughtered per day ranges between 5 and 6 cows, 10 to 16 goats, and sheep. During the dry season, 2 to 4 camels are also slaughtered. Waste materials ranging from horns, hooves, bones, dung, undigested feeds, and wastewater are produced every day in the abattoir.

Collection of wastewater samples

During the peak of the activities (8:00 am) that normally take place in the abattoir, the grab sampling method was used to collect three wastewater samples weekly over a month. Homogenized samples were taken in the butchering section, rinsing section, and stream using sterilized plastic bottles, as reported by Dankaka *et al.* (2018) during each wastewater sampling.

Determination of physical properties of wastewater

Determination of pH, temperature, total dissolved solids (TDS), and electrical conductivity (EC) was done using a hand-held Extik EC 600 multimeter. Measurement was carried out both *insitu* and *ex-situ* to determine if there was any significant change while samples were transported to the Laboratory.

Determination of nitrate

Nitrate was estimated in the sampled wastewater through the employment of the procedure reported by the American Water Works Association (AWWA) (2017). This was done as follows: Nitrate nitrogen standard solution was added to the sample to turn red and neutralize ammonia to nitrite and then nitrate. The sample was poured into the sample cell test tube and inserted into the sample cell of the spectrophotometer. The cover was then placed over the sample cell. The absorbance or concentration of this sample was read and recorded. The absorbance value was then converted to the equivalent concentration of nitrate by using a standard curve.

Determination of sulphate

The method reported by Kia et al. (2017) was utilized to determine the amount of sulfate present in the wastewater samples. A 250 mL Erlenmeyer flask was filled with a part of the wastewater samples. The conditioning reagent was then measured out to be 5 mL, added, and mixed using a magnetic stirrer. Timing started as soon as a teaspoon of barium chloride crystals was inserted. It was whisked for exactly one minute at a steady speed. Following agitation, a portion of the mixture was transferred into the photometer's absorption cell, and the turbidity was assessed every 30 seconds for four minutes. A calibration curve was created. The standards were created in the 0-40 mg/L Sulphate range at 5 mg/L increments, and their absorbance or turbidity was measured. A curve was produced by plotting the absorbance against sulfate concentration. With the use of the calibration curve, the absorbance for a particular sample allowed for the determination of the sulfate concentration in the solution.

Determination of phosphate

Phosphate was estimated in the sampled wastewater through the employment of the procedure reported by Phesatcha *et al.* (2016). The wastewater was poured into the cuvette of the spectrophotometer. The cuvette was wiped down with a microfiber to remove fingerprints. The cuvette was placed into the spectrophotometer, and the meter was switched on. A drop of reagent (stannous chloride) was added and then waited for 30 seconds to 5 minutes based on when the reagents fully reacted with the phosphate in the sample. When it fully reacted, it produced a blue color. The spectrophotometer button was pressed and held after adding the reagent. The reading of the phosphate concentration that was displayed on the machine was then recorded.

Determination of biochemical oxygen demand

Utilizing specific 300 mL biochemical oxygen demand (BOD) bottles that were intended to be filled without any air spaces and to provide an airtight seal, the BOD was measured (Water

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Science School, 2018). To represent varying dilutions, different amounts of wastewater samples were added to the bottles containing the sample and oxygenated water (distilled). As a control or "blank," dilution water alone was used to fill at least one bottle. Each bottle's initial dissolved oxygen (DO) concentration (mg/L) was determined using a DO meter. After that, each bottle was kept in a dark, 20 °C incubator for five days. The DO meter was used once more to determine the ultimate DO content (mg/L) after five days. After deducting the final DO measurement from the initial DO reading, the BOD concentration (mg/L) was obtained. We multiplied the result by the serial dilution factor.

Determination of chemical oxygen demand

The commonly used test to determine the amount of organic matter in wastewater samples instead of BOD is COD. The 5-day BOD test lacks the significant advantage that the COD test offers, as it may be finished in a matter of hours (Pharmaguideline, 2013). The titration method outlined by Ogbu et al. (2016) was used to calculate COD. After pipetting 10 mL of the samples into a conical flask, 5 mL of 0.025 N potassium dichromate (K₂Cr₂0₇) was added. Additionally, 40 mL of distilled water was added, and 15 mL of concentrated tetraoxosulphate (VI) acid (H₂SO₄) was diluted. Subsequently, the phenanthroline ferrous sulfate indicator was applied in seven drops. The flask became hot as a result of the effervescent that was produced. As a result, the combination was allowed to cool, changing color to a faint shade of blue-green. For the blank (i.e., the solution devoid of the water sample), the process was repeated. The spectrophotometer was used to read the results.

Determination of total bacterial count

The bacterial population was estimated through the employment of the procedure reported by Amalfitano *et al.* (2018).

Preparation of media

As directed by the manufacturers, MacConkey agar (MA), nutrient agar (NA), and eosin methylene blue (EMB) were made. Weighing the amount of agar needed for the analysis, a sensitive weighing balance was employed. The preparation and dissolution of the culture media followed the manufacturer's recommendations. As reported by Adeleye *et al.* (2019), the agar was sterilized in an autoclaving machine for 15 minutes at 121 °C to truncate the growth of any organisms and avoid compositional changes.

Inoculation and incubation of bacterial isolates

To identify and enumerate bacteria in the wastewater, 1 mL of the wastewater was diluted serially with 9 mL of distilled water (Cheesbrough, 2006). The solution of the serially diluted sample was then inoculated into NA and MA with 1.0 mL of diluted wastewater. The plate was gently swirled on the bench top to mix the culture and the medium thoroughly. The agar plate was allowed to completely gel without disturbing it, which took approximately 10 minutes. Each of the agar plates was sealed and incubated at 37 °C for 24 hours. After incubation, distinct colonies were subsequently sub-cultured on sterilized EMB for preliminary identification of the bacterial isolates.

Gram staining of bacterial isolates

Gram staining was used to color bacterial colonies that had been sub-cultured on NA, as described by Olutiola *et al.* (2000). On a spotless glass slide with no grease, a thin stain was created. After being air-dried, heated, and put on a rack, it was rinsed with distilled water after a minute of crystal violet coverage. Subsequently, 70% ethanol was used to decolorize the smear, and it was rinsed with distilled water after a minute of Lugol iodine treatment. After soaking in safranin for a minute, the smear was cleaned with distilled water. After using cotton wool to wipe the back of the slide, it was set on a rack to drain. The gram staining smear was examined microscopically using an x100 oil immersion objective lens, as reported by Cheesbrough (2006).

Biochemical tests for the identification of bacterial isolates

Some biochemical tests were carried out to further identify the various bacterial isolates present in the sampled wastewater. The catalase test was conducted as described by Ochei and Kolhatkar (2008), the coagulase test was conducted following the method described by Sapkota *et al.* (2020), the Indole test was done following the procedure highlighted by Cheesbrough (2006), the Methyl-red test and Voges-Prokaeur test were conducted as described by Ochei and Kolhatkar (2008) while citrate utilization test was done according to the method documented by Maina *et al.* (2021).

Data analyses

Descriptive statistics was employed to ingeminate the results of the physicochemical properties coupled with the identified heterotrophic bacterial population in the sampled wastewater. Results generated from all the parameters were compared with WHO and NESREA permissible standards. Data generated from the physical and chemical parameters of the sampled wastewater were subjected to One-way analysis of variance (ANOVA) using SAS software (version 9.1). Significant means were subsequently separated using Duncan's Multiple Range Test at p < 0.05.

RESULTS AND DISCUSSION

Table 1 shows the results of the physicochemical analyses of the wastewater samples. The temperature of the samples ranged between 28 and 30 °C in weeks 1 and 2. The sampled wastewater samples had the highest temperatures of 30 and 29.8 °C, respectively. However, wastewater sampled in weeks 3 and 4 recorded the lowest temperatures of 28.8 and 28.1 °C, respectively (**Table 1**). The highest pH (7.16) recorded during sampling was in week 1.

Table 1. Physicochemical parameters of the sampled wastewater

Parameters	Week 1	Week 2	Week 3	Week 4	Mean	WHO	NESREA
Temperature (°C)	30	29.8	28.8	28.1	29.18	12-25	40.0
рН	7.16	6.59	7.11	6.76	6.91	6.5-8.5	6 - 9
Turbidity (NTU)	122.0	120.5	105.6	100.6	112.18	1250	NS

Electrical conductivity (µS/cm)	4,540	3,080	1,670	1,282	2,643	1000	1000
Nitrate (mg/L)	980.7	978.4	711.2	615.3	821.4	45	20
Phosphate (mg/L)	246.75	220.74	225.45	233.75	231.7	No Standard	5-10
Sulphate (mg/L)	94.8	84.47	75.1	72.3	81.7	No Standard	500-1000
Dissolved oxygen (mg/L)	8.4	8.2	6.1	5.5	7.05	7.5	Unspecified
Biochemical Oxygen Demand (mg/L)	14.3	11.9	13.4	12.7	13.1	40	30
Chemical Oxygen Demand (mg/L)	48.2	35.0	30.9	29.3	35.9	80	60

The overall mean pH recorded for the sampled wastewater across the board was 6.91 **(Table 1)**. The DO of the samples ranged between 5.5 and 8.4 mg/L **(Table 1)**. The BOD of the wastewater samples ranged between 11.9 and 14.3 mg/L. Meanwhile, the COD of the wastewater samples ranged between 29.3 and 48.2 mg/L. The concentrations of phosphate recorded in the wastewater samples ranged between 220.74 and 246.75 mg/L, while nitrate concentrations ranged between 615.3 and 980.7 mg/L **(Table 1)**. The mean temperature (29.18 °C) of the wastewater samples recorded in this study was below the permissible limit of NESREA while it was above the WHO

allowable limit **(Table 1)**. However, during the sampling period, it can be observed the temperatures recorded were not significantly (p > 0.05) different from each other **(Table 2)**. A temperature range (29-31 °C) similar to the one obtained in this study has been reported by Dankaka *et al.* (2018) in their study conducted on old Sokoto abattoir wastewater. Also, a similar temperature range (29.5-35 °C) was reported by Odeyemi *et al.* (2011) in their study conducted on the wastewater assessment of abattoirs in Ado Ekiti. Furthermore, Ogunnusi and Dahunsi (2014) reported a temperature range (31-33.3 °C) in their study on abattoir effluent in Oyo, Oyo state, Nigeria.

	Temperature (°C)	рН	Turbidity (NTU)	Electrical Conductivity (μS/cm)	Nitrate (mg/L)	Phosphate (mg/L)	Stulphate (mg/L)	Dissolved Oxygen (mg/L)	Biochemical Oxygen Demand (mg/L)	Chemical Oxygen Demand (mg/L)
Week 1	30	7.16	122.0 ^a	4540 ^a	980.7ª	246.75ª	94.8ª	8.4ª	14.3ª	48.2ª
Week 2	29.8	6.59	120.5 ^b	3080 ^b	978.4 ^b	220.74 ^d	84.47 ^b	8.2ª	11.9 ^d	35.0 ^b
Week 3	28.8	7.11	105.6 ^c	1670 ^c	711.2°	225.45°	75.1°	6.1 ^b	13.4 ^b	30.9°
Week 4	28.1	6.76	100.6 ^d	1282 ^d	615.3 ^d	233.75 ^b	72.3 ^d	5.5°	12.7c	29.3 ^d
Level of significance	Not significant	Not significant	Significant	Significant	Significant	Significant	Significant	Significant	Significant	Significant

pH is known as the most important factor that determines the corrosive nature of any water body. It has been established in the literature that the lower the pH value, the higher the corrosive nature of the water. In this study, the pH of the sampled wastewater, which ranged between 6.59 and 7.16, fell within the permissible limits set by WHO and NESREA. The recorded wastewater pH in this study was near neutral, which might have played a role in the abundance of microorganisms recorded in the wastewater during sampling. It can be noted that mean pH values during wastewater sampling in this study were not significantly (p > 0.05) different from each other **(Table 2)**.

The EC recorded in the collected wastewater samples ranged between 1,282 and 4,540 μ S/cm, which was above the allowable limits set by WHO and NESREA **(Table 1)**. This finding is similar to the finding of Rabah *et al.* (2008), who reported EC ranging between 5.97 and 4448 μ S/cm in Sokoto abattoir wastewater. The murky and cloudy appearance of the wastewater sampled in this study can be attributed to the presence of organic matter. The wastewater sampled in this study recorded a mean value (112.18 NTU) of turbidity, which is way below the permissible limit (1250 NTU) set by WHO. This finding is in concord with a turbidity range of 388.00 ± 117.00 NTU reported by Ogbu *et al.* (2016) in their study. However, it can be observed that the

weekly mean values of EC recorded during this study were significantly (p < 0.05) different from each other (**Table 2**).

It can be seen in **Table 1** that the nitrate concentration in the sampled wastewater, which ranged between 615.3 and 908.7 mg/L, was higher than the permissible limits recommended by NESREA (20 mg/L) and WHO standard (45 mg/L). Weekly mean nitrate concentrations can be observed to be significantly (p < 0.05) different from each other **(Table 2)**. The presence of nitrate and phosphorus in the sampled wastewater can be attributed to the feed consumed by the cows that are slaughtered in the sampled wastewater can be attributed in the sampled wastewater can be attributed to the high concentration of organic matter content, which might have resulted from the decomposition of protein and nitrogenous compounds. This, upon breakdown, gives rise to simpler substances, including ammonia.

The results obtained in this study indicate that the mean phosphate concentration (231.7 mg/L) was far above the allowable limit of NESREA **(Table 1)**. Again, weekly mean phosphate concentrations can be observed to be significantly (p < 0.05) different from each other **(Table 2)**. The high phosphate concentration recorded in this study could pose a serious threat to the aquatic ecosystem as it has been implicated in causing eutrophication and cyanobacterial bloom. However, Kroiss *et al.*

(2011) reported that the main sources of phosphorus obtainable in wastewater range from the excreta of humans to trade wastes, industrial-based wastes, and household detergents containing phosphorus. These authors further reiterated that it is only when there is the presence of combined sewers that precipitation runoff adds to the load of phosphorous in wastewater.

The overall mean value (81.7 mg/L) of sulfate recorded in this study was below the permissible limits set by NESREA (**Table 1**). However, weekly mean sulfate concentrations can be observed to be significantly (p < 0.05) different from each other (**Table 2**). It has been reported by Gupta and Germida (2021) and Obeta *et al.* (2023) that the presence of sulfate in wastewater allows sulfate-reducing bacteria to initiate the oxidation of organic compounds and hydrogen to sulfate reduction.

Dissolved oxygen is the amount of oxygen that is present in water. Water bodies receive oxygen from the atmosphere and aquatic plants. DO is defined as the concentration of oxygen in any fluidic substance (Fondriest Environmental, 2022). Low DO primarily results from excessive algae growth caused by phosphorus. Nitrogen is another nutrient that can contribute to algal growth. Just as low dissolved oxygen can cause problems, so too can high concentrations. The mean DO (7.05 mg/L) recorded in this study was slightly below the allowable limit (7.50 mg/L) set by WHO (Table 1). Similar results have been reported by Dankaka et al. (2018) in their physicochemical analyses of Sokoto Abattoir wastewater, which ranged between 4.2 and 8.5 mg/L. However, it can be observed that the mean DO concentrations in the sampled wastewater in the first and second weeks were not significantly (p > 0.05) different from each other but were significantly (p < 0.05) different from those recorded in the third and fourth weeks of sampling (Table 2). Biochemical oxygen demand directly affects the amount of dissolved oxygen in rivers and streams. Results obtained in this study (Table 2) indicate that the mean BOD (1310 mg/L) was way below the allowable limits set by WHO (40 mg/L) and NESREA (30 mg/L). Similar results have been reported by

Dankaka *et al.* (2018). However, the documented weekly mean BOD values were not significantly different (p > 0.05) from each other during sampling **(Table 2)**. Higher BODs contrary to the one obtained in this study have been reported by Adesemoye and Adedire (2005) in a study conducted in Agege abattoir (35 ± 1.5 mg/L) and Odo abattoir (30 ± 2.0 mg/L) while Rabah *et al.* (2008) reported a BOD range between 22.40 and 31.40 mg/L in Sokoto abattoir wastewater. Generally, the higher the concentration of BOD, the more swiftly oxygen is exhausted in any body of water. According to the United States Environmental Protection Agency (USEPA) (2012), the main penalty of high concentration of BOD in water bodies is ultimate depletion of dissolved oxygen, which will invariably snowball into extinction of aquatic life therein.

Chemical oxygen demand is the amount of oxygen required to chemically break down pollutants. Specifically, it measures the equivalent amount of oxygen required to chemically oxidize organic compounds in water, thus removing the pollution. From the results obtained in this study and depicted in Table 1, the overall mean COD (3593 mg/L) was above the permissible limits set by both WHO (80 mg/L) and NESREA (60 mg/L). However, the weekly mean COD values recorded were not significantly different (p > 0.05) from each other during sampling (Table 2). A similar result was reported by Egwuonwu et al. (2012) in their study conducted on the wastewater generated by Nigerian Breweries in Enugu, Nigeria. The total heterotrophic bacterial counts (THBC) in the sampled wastewater are presented in Table 2. It can be observed that the highest (5.6×10² CFU/mL) THBC, which was counted on MA, was recorded in week 1, while the lowest (1.9×104 CFU/mL), which counted on NA, was recorded in week 4 (Table 3). The higher rate of contamination of the sampled wastewater with these organisms is an indication of the deplorable state and poor sanitary practices put in place in the management of the Dutse abattoir. Also, the abundance of microorganisms can be attributed to the abundance of nutrients in the wastewater, which are suitable for optimal microbial metabolism (Neboh et al., 2013; Darbar & Saha, 2023).

Table 3. Total heterotrophic bact	rial counts and co	olonial	characteristics of t	he ba	acterial iso	olates in t	he sampled	wastewater
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Samples on media	Number of colonies	CFU/mL	Colonial characteristics
Week 1 MA	280	5.6×10 ²	Pinkish colonies with heavy growth
NA	170	3.4×10 ²	Creamy white colonies with moderate growth
NA	158	3.1×10 ²	Creamy white colonies with moderate growth
Week 2 MA	196	3.9×10^{4}	Pink-red colonies with moderate growth
NA	164	3.2×10^{4}	Yellow colonies with moderate growth
NA	126	2.5×10^{4}	Yellow colonies with moderate growth
Week 3 MA	152	3.0×10 ²	Pink-red colonies with moderate growth
NA	108	2.1×10 ²	Greenish white colonies with moderate growth
NA	99	1.9×10 ²	Greenish white colonies with moderate growth
Week 4 MA	180	3.6×10 ⁴	Mucoid pink colonies with moderate growth
NA	102	2.0×10^{4}	Creamy yellow colonies with moderate growth
NA	96	1.9×10^{4}	Creamy yellow colonies with moderate growth

In a similar study conducted by Rabah *et al.* (2008), THBC ranging between 4.9×10^7 and 7.3×10^7 CFU/mL was recorded in Sokoto Abattoir wastewater. In the same way, Adesemoye and Adedire (2005) reported 3.32×10^7 CFU/g for the total number

of bacteria found in the soil of the Agege Abattoir in Lagos, while Ogbonna and Igbenijie (2006) reported 2.08×10³ CFU/mL for the total number of bacteria found in the wastewater collection sites in Port Harcourt, Nigeria. The outcome of indiscriminate discharge of untreated wastewater can be detrimental to the ecosystem balance of the aquatic environment and public health generally (Edokpayi *et al.*, 2017).

The colonial characteristics of the bacterial isolates from the sampled wastewater are presented in **Table 3**. Pinkish colonies with heavy growth were observed on the wastewater sample plated on MA in week 1, while creamy yellow colonies with moderate growth were seen on the wastewater plated on NA **(Table 3)**. **Table 3** equally shows the physical characteristics of the isolated bacteria. The bacterial isolates that emerged in pinkish, pink, red, yellow, greenish-white, and mucoid pink colonies may all be seen under the microscope to be gramnegative rod-shaped bacteria **(Table 4)**. Gram-positive cocci in clusters and gram-positive rod-shaped bacteria, respectively, were the bacterial isolates that emerged under the microscope in creamy white and creamy yellow colonies **(Table 4)**.

Biochemical tests are used to identify different species of bacteria based on changes in their various metabolic activities

(Aryal, 2019). This author also stated that detecting microorganisms has made considerable use of bacteria's ability to make organic chemicals through the metabolism of certain carbohydrates and other related molecules. Table 5 shows the outcomes of the tests conducted for biochemical characterization to identify the bacterial isolates. Escherichia coli, Citrobacter sp., Salmonella sp., Klebsiella sp., and Enterobacter sp. were shown to be the gram-negative rodshaped bacterial isolates (Table 5). Staphylococcus aureus and Bacillus spp. were the biochemically identified bacterial isolates that showed up as gram-positive cocci in clusters and grampositive rods, respectively (Table 5). Escherichia coli (15.30%), Staphylococcus aureus (17.9%), Citrobacter sp. (10.7%), Salmonella sp. (15.8%), Klebsiella sp. (19.61%), Enterobacter sp. (9.8%), and Bacillus sp. (10.8%) were identified from the assayed wastewater (Figure 1).

Table 4. Morph	ological character	istics of the bacte	rial isolates

Colonial characteristics	Gram staining results
Pinkish colonies	Gram-negative in rod shape
Creamy white colonies	Gram-positive cocci in clusters
Pink-red colonies	Gram-negative in rod shape
Yellow colonies	Gram-negative in rod shape
Greenish white colonies	Gram-negative in rod shape
Mucoid pink colonies	Gram-negative in rod shape
Creamy yellow colonies	Gram-negative in rod shape

Table 5. Biochemical characterization tests conducted on the bacterial isolates

Gram reaction	Catalase	Coagulase	Indole	Methyl Red	Voges Proskauer	Citrate	Identity
Gram-negative in rod shape	+	-	+	+	-	-	Escherichia coli
Gram-positive cocci in clusters	+	+	-	+	+	+	Staphylococcus aureus
Gram-negative in rod shape	+	-	-	+	-	+	Citrobacter sp.
Gram-negative in rod shape	+	-	-	+	-	+	Salmonella sp.
Gram-negative in rod shape	+	-	-	-	+	+	Klebsiella sp.
Gram-negative in rod shape	+	-	-	-	+	+	Enterobacter sp.
Gram-negative in rod shape	+	-	-	-	+	+	Bacillus sp.

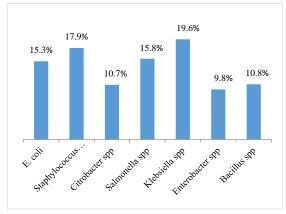


Figure 1. Isolated bacteria percentage of occurrence in the assayed wastewater

Different regions of Nigeria have recorded similar pathogenic bacteria that have been isolated from abattoirs' wastewater (Ogbomida et al., 2016). In particular, bacteria including Staphylococcus aureus, Escherichia coli, Proteus sp., Klebsiella sp., Penicillium sp., and Aspergillus niger were isolated and identified from abattoir effluents in Oyo, Oyo state, Nigeria by Ogunnusi and Dahunsi (2014). Neboh et al. (2013) also reported the presence of Escherichia coli, Penicillium sp., Aspergillus niger, Staphylococcus aureus, Enterobacter aerogenes, and Klebsiella pneumonia in the Ijegbu-Igbo abattoir effluent. Similarly, effluent from an abattoir in Lagos, Nigeria, was found to contain Bacillus sp., Aspergillus niger, Penicillium sp., and Mucor sp. (Adesemoye & Adedire, 2005). However, contrary to the bacteria isolated in this current study, Kacprzak et al. (2005) isolated Sporothrix schenkii and Rhodotorula sp. from untreated and treated wastewater in Poland.

The results generated from the total heterotrophic bacterial count isolated from the wastewater samples are presented in **Table 6**.

Table 6. Compa	Table 6. Comparison of total heterotrophic bacterial count with							
permissible standards								
Number of	THBC	WHO Standard	NESREA					
- -	(CEU/I)	(CEII / I)	Chan Jan J					

	weeks	(CFU/mL)	(CFU/mL)	Standard
	Week 1	12.1×10 ²		
_	Week 2	9.6×10 ⁴	-	
_	Week 3	7×10 ²	- 1×10 ²	No standard
_	Week 4	7.5×10 ⁴	- 1×102	NO Stanuaru
_	Total	36.2×10 ⁶	-	
	Mean	9.05×10 ⁶	-	

It can be seen that the mean value (9.05×10⁶ CFU/mL) was high, indicating that the wastewater ought to be treated before onward discharge into the environment. This result is contrary to the WHO allowable limit (1×10² CFU/mL) reported by Dankaka *et al.* (2018). Rabah *et al.* (2008) reported a mean (6.4×10⁷ CFU/mL) for the total viable count of bacteria isolated in Sokoto abattoir effluent, which is consistent with our investigation.

CONCLUSION

It can be concluded that the wastewater generated from the Dutse abattoir was unfit for discharge into the environment without subjecting it to any treatment during the conduct of this study. Due to the high concentration of organic materials in the wastewater, oxygen-consuming microbes may proliferate quickly and cause the water to become septic or eutrophic. Most of the physical and chemical parameters recorded in this study were beyond permissible limits, making the wastewater unfit for disposal without treatment. Also, the isolation of some pathogenic microorganisms in the wastewater sampled is an indication that it could serve as a reservoir of pathogenic bacteria that can ultimately cause a public health crisis if the wastewater is released indiscriminately into the surrounding environment. Therefore, good hygienic and sanitary practices should be established and implemented in the abattoir. Wastewater generated from the abattoir should be treated before it is discharged into the public drainage system. Also, strict legislation coupled with standard practices should be ensured to prevent the currently witnessed uncritical disposal of raw effluent into the environment.

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REFERENCES

Adeleye, A. O., Amoo, A. O., Bate, G. B., Sadiq, I. S., & Ugba, S. (2021). Physicochemical and bacteriological assessment of rice mill wastewater discharged into river Benue, Nigeria. *Biological Environment and Pollution*, 1(1), 1-10.

- Adeleye, A. O., Yalwaji, B., Shiaka, G. P., Amoo, A. O., & Udochukwu, C. C. (2019). Bacteriological quality of freshlycut and ready-to-eat vegetables sold in Dutse market north west, Nigeria. *FUW Trends in Science & Technology Journal*, 4(1), 054-057.
- Adesemoye, A. O., & Adedire, C. O. (2005). Use of cereals as basal medium for the formulation of alternative culture media for fungi. World Journal of Microbiology and Biotechnology, 21, 329-336.
- Almulhim, F. A., Alshahrani, M. M. A., Hakami, A. M., Shammaa, A. M., Aljehaiman, T. A., Alsaihati, A. M., Alqarni, A. A. S., Alotaibi, H. B. B., Alotaibi, M. N., & Hawsawi, A. O. A. (2022). Review on Pneumothorax Diagnostic and Management Approach in Emergency Department. *International Journal* of Pharmaceutical Research and Allied Sciences, 11(1), 35-39.
- Alshammari, S. T. M., Turkistani, H. A., Almatar, Y. I., Alhuraish, A. M. A., Hefni, S. T., Bagabir, R. A., Alghamdi, Y. A., Alareefi, J. A., Alyamani, M. N., & Alfari, A. Y. (2022). An overview on endodontic irrigation solution role in management. *International Journal of Pharmaceutical Research and Allied Sciences*, 11(1), 17-20. doi:10.51847/hQTG6i8ZFI
- Amalfitano, S., Levantesi, C., Garrelly, L., Giacosa, D., Bersani, F., & Rossetti, S. (2018). Water quality and total microbial load: A double-threshold identification procedure intended for space applications. *Frontiers in Microbiology*, 9, 2903. doi:10.3389/fmicb.2018.02903
- Aryal, S. (2019). Water quality analysis by most probable number (MPN). Available from: https://microbe notes.com/water-quality-analysis-by-most-probablenumber-mpn/.
- AWWA (American Water Works Association) (2017). Standard methods for the examination of water and wastewater. American Public Health Association, American Water Works Association, Water Environment Federation.
- Bustillo-Lecompte, C., & Mehrvar, M. (2017). Slaughterhouse wastewater: Treatment, management and resource recovery. *Physico-Chemical Wastewater Treatment and Resource Recovery*, 153-174. doi:10.5772/65499
- Cheesbrough, M. (2006). *District laboratory practice in tropical countries. Part 2*. Low Price Edition. Cambridge University Press, London.
- Dankaka, S. M., Farouq, A. A., Bagega, A. I., & Abubakar, U. (2018). Microbiological and physicochemical analysis of old Sokoto abattoir wastewater. *Bioremediation Science and Technology Research Journal*, 6(2), 9-13.
- Darbar, S., & Saha, S. (2023). Novel multi-herbal formulation (SS-IM-21) and its sanative effect in CCl₄ induced experimental animals. *International Journal of Pharmaceutical and Phytopharmacological Research*, 13(2), 30-36. doi:10.51847/ZFscjZHxtU
- Edokpayi, J. N., Odiyo, J. O., & Durowoju, O. S. (2017). Impact of wastewater on surface water quality in developing countries: A case study of South Africa. *Water Quality*, *10*(66561), 10-5772. Available from: https://cd n.intechopen.com/pdfs/53194.pdf.
- Egwuonwu, C. C., Uzoije, A. P., Okafor, V. C., Ezeanya, N. C., & Nwachukwu, M. U. (2012). Evaluation of the effects of industrial wastewater discharge on surface water (a case

study of Nigeria Breweries Plc Enugu). *Greener Journal of Physical Sciences*, 2(3), 56-63.

- Fondriest Environmental (2022). Dissolved Oxygen. Available from: https://www.fondriest.com/environmentalmeasurements/parameters/water -quality/dissolvedoxygen/.
- Gupta, V. V. S. R., & Germida, J. J. (2021). Microbial transformations of sulfur in soil. In *Principles and Applications of Soil Microbiology* (pp. 489-522). Elsevier.
- Kacprzak, M., Neczaj, E., & Okoniewska, E. (2005). The comparative mycological analysis of wastewater and sewage sludges from selected wastewater treatment plants. *Desalination*, 185(1-3), 363-370.
- Kia, A., Rastegary, M., Dara, A., Banki, M. S., Rezaei, E., Ebrahimi, F., & Alizade, M. (2017). Experimental study of Siri-Island refinery wastewater treatment using a photocatalytic process. *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects*, 39(2), 232-239. doi:10.1080/15567 036.2016.1185482
- Kirilmaz, S. K. (2022). Mediating role of positive psychological capital in the effect of perceived organizational support on work engagement. *Journal of Organizational Behavior Research*, 7(1), 72-85.
- Kroiss, H., Rechberger, H., & Egle, L. (2011). Phosphorus in water quality and waste management. In *Integrated Waste Management-Volume II* (pp. 181-214). IntechOpen. doi:10.5772/18482
- Kundu, P. P., & Dutta, K. (2018). Progress and recent trends in microbial fuel cells. Elsevier.
- Lapygina, E. V., Lysak, L. V., & ZVyagintSev, D. G. (2002). Tolerance of soil bacterial complexes to salt shock. *Microbiology*, 71, 143-147. doi:10.1023/A:10 15181717601
- Maina, J., Mathara, J. M., Kikuvi, G. M., & Wafula, E. (2021). Isolation and identification of bacteriocin-producing *Bacillus* spp. From *Rastrineobola argentea* (Omena) with activity against bovine mastitis bacterial pathogens. *Journal of Food Security*, 9, 62-75.
- Manasa, P., Lei, Z. J., & Ran, F. F. (2020). Biomass waste derived low cost activated carbon from *Corchorus olitorius* (Jute Fiber) as sustainable and novel electrode material. *Journal* of Energy Storage, 30, 101494. doi:10.1016/j.est.202 0.101494
- Matheyarasu, R., Seshadri, B., Bolan, N. S., & RaviNaidu, R. (2015). Impact of abattoir wastewater irrigation on soil fertility and productivity. In (Ed.), Irrigation and drainage-Sustainable strategies and systems. IntechOpen. doi:10.5772/59312
- Micronics Engineered Filtration Group (2023). Industrial Wastewater. Available from: https://www.micronicsinc.com/filtration-news/what-isindustrial-wastewater/
- Milman, O. (2021). Meat accounts for nearly 60% of all greenhouse gases from food production, study finds. *The Guardian*, 13. Available from: https://www.the guardian.com/environment/2021/sep/13/meat greenhouses-gases-food-production-study.
- Neboh, H. A., Ilusanya, O. A., Ezekoye, C. C., & Orji, F. A. (2013). Assessment of Ijebu-Igbo abattoir effluent and its impact on the ecology of receiving soil and river. *IOSR Journal of Environmental Science and Food Toxicology*, 7(5), 61-67.

- Negi, R., & Suthar, S. (2018). Degradation of paper mill wastewater sludge and cow dung by brown-rot fungi Oligoporus placenta and earthworm (*Eisenia fetida*) during vermicomposting. *Journal of Cleaner Production*, 201, 842-852.
- Obeta, U. M., Jaryum, P. L., Ejinaka, O. R., & Utibe, E. (2023). Turmeric is medicinal and cosmetic in nature, and it is used in the production of obeturmeric powder and cream. *International Journal of Pharmaceutical and Phytopharmacological Research*, 13(2), 18-24. doi:10.51847/IrnOzra23V
- Ochei, J. O., & Kolhatkar, A. A. (2008). *Medical laboratory science: Theory and practice.* Tata McGraw Publishing Company Limited. Seventh Edition. pp. 637-745.
- Odeyemi, A. T., Dada, A. C., Akinjogunla, O. J., & Agunbiade, O. R. (2011). Bacteriological, physicochemical, and mineral analysis of water use in abattoirs in Ado-Ekiti, Southwest Nigeria. *Journal of Microbiology and Biotechnology Research*, 1(2), 14-20.
- Ogbomida, E. T., Kubeyinje, B., & Ezemonye, L. I. (2016). Evaluation of bacterial profile and biodegradation potential of abattoir wastewater. *African Journal of Environmental Science and Technology*, 10(2), 50-57.
- Ogbonna, D. N., & Igbenijie, M. (2006). Characteristics of microorganisms associated with waste collection sites in Port Harcourt City, Nigeria. *Nigerian Journal of Microbiology*, 20(3), 1427-1434.
- Ogbu, K. C., Ebenebe, C. I., & Abajue, M. C. (2016). Physicochemical characteristics of Ama brewery effluent and its receiving Ajali River in Udi, Enugu State, Nigeria. *Animal Research International*, 13(2), 2392-2399.
- Ogunnusi, T. A., & Dahunsi, O. V. (2014). Isolation and identification of microorganisms from abattoir effluents from Oyo, Oyo State, Nigeria. *Asian Journal Applied Science*, 2(2), 218-222.
- Olutiola, P. O., Famurewa, O., & Sonntag, H. G. (2000). Introduction to microbiology, (2nd edn), Heidelberg: Nigeria, pp. 267.
- Pharmaguideline (2013). Determination of chemical oxygen demand of wastewater. Available from: https://www.pharmaguideline.com/2013 /06/COD-testwaste-water-organic-poll ution-determination.html.
- Phesatcha, T., Tukkeeree, S., & Rohrer, J. (2016). Determination of total phosphorus in wastewater using Caro's reagent and lon chromatography. Available from: https://assets.Thermofis her.com/TFS-Assets/CMD/Application-Notes/AN-254-IC-Total-Phosphorus-Wastewater-AN71350-EN.pdf.
- Pushkin, S. V., Chistohina, O. I., Turalieva, A. I., Subaeva, S. R., Karapetyan, D. A., Romanova, A. A., & Zhatko, Y. A. (2023). The use of microcomputed tomography to study the anatomical features of the body. *International Journal of Pharmaceutical Research and Allied Sciences*, 12(2), 89-98. doi:10.51847/HHwaBnfPZu
- Rabah, A., Ijah, U. J. J., Manga, S. B., & Ibrahim, M. L. (2008). Assessment of physicochemical qualities of abattoir wastewater in Sokoto, Nigeria. *Nigerian Journal of Basic* and Applied Science, 16(2), 149-154.
- Sapkota, A., Thapa, A., Budhathoki, A., Sainju, M., Shrestha, P., & Aryal, S. (2020). Isolation, characterization and screening of antimicrobial-producing actinomycetes from soil

samples. International Journal of Microbiology, 2020, 2716584. doi:10.1155/2020/2716584

- Shetgaonkar, K. A., Suragimath, G., Varma, S., & Zope, S. (2022). Two way relationship between diabetes and periodontitis: A cross-sectional survey of knowledge, awareness, and attitude. *International Journal of Pharmaceutical Research* and Allied Sciences, 11(2), 1-7. doi:10.51847/tobUt0H7E0
- Terela, H., & Strilets, V. (2023). Supervision and control over compliance with labor law in Ukraine during the martial law. *Journal of Organizational Behavior Research*, 8(1), 39-51.
- Tuser, C. (2020). What is wastewater. Water & Wastes Digest. Available from: https://www.wwdmag.com/wastewatertreatment/what-wastewater.
- USEPA (United States Environmental Protection Agency) (2012). Dissolved oxygen and biochemical oxygen demand. Available from: https://archive.epa.gov/water/archive/web/html/vms5 2.html.
- Water Science School (2018). Biological oxygen demand (BOD) and water. Available from: https://www. usgs.gov/special-topic/water-science-

school/science/biological-oxygen-demand-bod-andwater?qt-sciencecenterobjects=0#qtsciencecenterobjects