Physicochemical and Microbiological Characterization of Effluents from the Mohammed VI University Hospital in Marrakech and Study of Their Impact on the Environment

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ABSTRACT
Water pollution from sewage has become a subject of considerable public and scientific concern, given its demonstrated extreme toxicity to human health and biological ecosystems. Hospital wastewater poses a serious health risk to staff, the public, and the environment. This study was conducted to determine the physical, chemical and biological parameters of hospital wastewater from Mohammed VI University Hospital in Marrakech, which is discharged into the municipal sewage system, where it mixes with sewage from urban areas without prior treatment. This study was conducted between October 2022 and January 2023 with a sampling frequency of 15 days each. Hospital wastewater samples were collected at the main collector of all manholes in the hospital. Physicochemical characterization of the effluents from each department showed that they are heavily polluted with organic matter, with average values in the range of 750 mg/L COD, 512.14 mg/L BOD5 and 879.86 mg/L TSS. These values are higher than those recommended in the Moroccan discharge standards. Total phosphorus, sulfates, nitrates and nitrites are 5.92 mg/L, 427.97 mg/L, 7.39 mg/L and 0.60 mg/L, respectively. Bacteriological characterization shows that fecal coliform bacteria reach $12 \times 10^6$ CFU/100 mL. These effluents also contain pathogens responsible for nosocomial infections, especially Staphylococcus aureus. The ratio COD / BOD5 is about 1.46, a value indicating that hospital wastewater is readily biodegradable. Therefore, proper treatment of these hospital effluents is essential due to their physicochemical and pathogenic bacterial load, which can negatively affect public health and carry the risk of spreading epidemic diseases.

Keywords: bacteriological characterization, hospital wastewater, physicochemical characterization, population health.

INTRODUCTION
The security of urban water supply in Morocco is threatened by major challenges such as population growth, urbanization, economic expansion, and climate change. Urban water demand in most major cities is expected to increase by 60% to 100% by 2050 (WBG 2017).

Marrakech, like the other cities in the country, is experiencing a population explosion and a development in the tourism sector, which affects the surface and underground water resources, both in quantitative terms (lowering of the water table due to overexploitation) and in qualitative terms (the domestic and industrial effluents contain substances that degrade the water quality).

The Mohammed VI University Hospital in Marrakech, with a capacity of about 1139 beds, is one of the most important public health facilities in the entire Marrakech-Safi region and one of the largest health facilities in Morocco.

Hospitals discharge a large amount of wastewater with different physicochemical compositions, including chemical and biological products,
toxic drugs, radioactive elements, and pathogenic microorganisms (Gautam et al. 2017; Bouha et al. 2014; Dahhou et al. 2016). Wastewater from Mohammed VI University Hospital in Marrakech is discharged into the municipal sewage network, where it mixes with municipal wastewater without prior treatment. Previous studies have shown that hospitals are an undeniable source of many compounds that enter the environment through the activities of medical laboratories (Alabi et al. 2012). In addition to other works, it has been shown that the overall physicochemical parameters of hospital wastewater (temperature, pH, salinity, conductivity, dissolved oxygen, biological oxygen demand, chemical oxygen demand, nitrate, nitrite, phosphate, and suspended solids) Hospital wastewater is more polluted than municipal wastewater (El Morhit et al. 2015).

Other studies have shown that hospital wastewater is a complex mixture of pathogenic microorganisms. Although the microbial quality and organic content of wastewater are significantly reduced after treatment steps, the remaining microorganisms, including multidrug-resistant bacteria such as Staphylococcus aureus, are sufficient to cause public health and environmental problems (Berto et al. 2009).

This study addresses the management of hospital liquid waste at Mohammed VI University Hospital in Marrakech. It evaluates the pollution level of this hospital based on physicochemical and bacteriological analyzes of the wastewater at the level of the sewers of the internal sewerage network before their discharge into the municipal sewerage network.

**MATERIALS AND METHODS**

This study was conducted between October 2022 and January 2023 with a sampling frequency of 15 days each. Hospital wastewater samples were collected at the main collector of all manholes in the hospital (Fig. 1).

After sampling, the samples were stored according to the General Guide for Storage and Handling of Samples as per ISO 5667-3 (1994) and the Good Practice Guide of the National Office for Drinking Water (ONEP 1999).

Water samples were collected in plastic bottles and stored at 4 °C before being transported to the laboratory in electric cooler (Fig. 2) within two hours. Temperature, pH, and conductivity were measured using a multiparameter sensor (Fig. 3). Other chemical parameters were analyzed in the laboratory according to the standard methods of the French Association for Normalization (AFNOR 1996) and Rodier et al. (2009).

Samples for microbiological analyzes were collected in a 250 mL glass bottle, sterilized in an autoclave (120 °C) for 15 minutes, and immediately transported in electric cooler to the laboratory, where they were subjected to bacteriological analysis in less than an hour. The microbiological analysis methods and culture media used are listed in Table 1.
RESULTS AND DISCUSSION

Physicochemical characterization of hospital wastewater

The analysis of the results shows that the temperature of the samples taken varies between 19.5 and 25.6 °C, with an average value of 23.21 °C (Table 2). These values are below the limits allowed for direct and indirect discharges (below 30 °C) (ME 2002).

The pH value shows that the effluents are slightly alkaline. The only acidic pH value recorded is 6.9 (Table 2). The values are within the allowable limits for direct and indirect releases (ME 2002).

Conductivity can be used to assess the degree of mineralization of hospital wastewater. On average, these effluents have a fairly high degree of mineralization, with values of electrical conductivity (EC) ranging from 1,314 to 2,750 μS/cm (Table 2), placing these effluents in Class 3 of the water quality network (ONEP 1995); thus, with these values, these effluents are at the EC limit allowed for direct discharges (less than 2,700 μS/cm). This high value of EC is due to the high load of inorganic and organic substances generated by the hospital’s effluents.

The values of COD measured in the different samples range from 600 to 925 mg/L (Table 2) and are significantly higher (p < 0.01) than the value reported by Boillot Boillot (2008) for hospital wastewater (43–270 mg/L) and also higher than the average value reported by Verlicchi et al. (2010)(500 mg/L) or compared to the values of wastewater from a conventional community (300 and 1000 mg/L) (Metcalf and Eddy 2003).

The average BOD$_5$/COD ratio (0.68) reflects the low biodegradability of the substances contained in these effluents. The average COD / BOD$_5$ ratio is also low (1.46), indicating that the organic matter in the effluents is readily biodegradable according to Henze et al. (1997).

The total suspended solids (TSS) measured in the effluents of the different samples vary from 124 to 3353 mg/L with an average value of 879.86 mg/L (Table 2), which is far above the indirect discharge limits of the Moroccan standard (600 mg/L). These values are very high compared to those of municipal wastewater (150–500 mg/L) (Metcalf and Eddy 2003) and compared to the wastewater of the provincial hospital in the city of Sidi Kacem with an average value of 165.99 mg/L (Elouakfaoui et al. 2022).

These results show that these effluents have a high concentration of minerals and/or organic matter, with a good correlation ($R^2 = 0.87$) between BOD$_5$, COD and TSS. Several authors have pointed out the presence of high concentrations of organic compounds in hospital wastewaters (Gartiser et al. 1996; Leprat 1998), which have been associated with the presence of iodine-containing contrast agents used in

<table>
<thead>
<tr>
<th>Microorganisms</th>
<th>Incubation</th>
<th>Growth medium</th>
<th>Analysis method</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total coliforms</td>
<td>37 °C for 24 hours</td>
<td>Lactose agar with TTC and Tergitol</td>
<td>Enumeration by incorporation in agar</td>
</tr>
<tr>
<td>Fecal coliforms</td>
<td>44 °C for 18 to 24 hours</td>
<td>Lactose agar with TTC and Tergitol</td>
<td>Enumeration by incorporation in agar</td>
</tr>
<tr>
<td>Fecal streptococci</td>
<td>37 °C for 24 hours</td>
<td>BEA medium (Bile Aesculin Agar)</td>
<td>Enumeration by incorporation in agar</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>37 °C for 24-48h</td>
<td>Baird–Parker medium</td>
<td>Enumeration by incorporation in agar</td>
</tr>
</tbody>
</table>

Table 1. Incubation conditions, growth medium and analysis method for the germs sought
radiography, certain drugs and their metabolites that may contain organohalogen elements, the use of disinfectants and chlorinated solvents, and other substances from laboratories (Kuemmerer et al. 1998; Sprehe et al. 2001; Emmanuel et al. 2004).

The average nitrate concentration is 7.39 mg/L and nitrite concentration is 0.60 mg/L (Table 2). This concentration is much higher than the values obtained by Endamana et al. (2002) for the wastewater of Yaoundé (Cameroon) and Faruqui et al. (2004) for that of Dakar (Senegal).

The average total phosphorus concentration is 5.92 mg/L with extreme maximum and minimum values of 8.13 and 3.88 mg/L, respectively (Table 2). The different concentrations measured are below 10 mg/L, which is considered an acceptable limit according to the Moroccan standard for indirect discharges to the receiving environment (ME 2002).

For sulfates, SO_4^{2-} values ranged from 260 to 770.6 mg/L (Table 2) with an average of 427.97 mg/L. These values are high and above the limit allowed by the Moroccan standard for indirect discharges to receiving waters (400 mg/L).

### Microbiological characterization of hospital wastewater

In our study, Total Coliforms (TC) varied from 6 × 10^6 to 15 × 10^6 CFU/100 ml, the average load in TC is 9.57 × 10^6 CFU/100 ml (Table 3), was higher than that of Al Ghassani Hospital of Hassan II University Hospital in Fez (Tahiri et al. 2009). However, higher average concentrations of TC were found in the wastewater of Mohammed V Hospital in Meknes (Ameziane and Benabidate 2014).

The average concentration of Fecal Coliforms (FC) found in the main sewer of the Mohammed VI University Hospital in Marrakesh (7.57 × 10^6 CFU/100 mL) (Table 3), is very significant compared to the concentration reported in wastewater from a hospital in a large city in southeastern France (2 × 10^6 CFU/100 mL) (Emmanuel et al., 2014). The concentration of fecal coliforms in hospital wastewater can tell us the level of ecotoxicity of this effluent. Indeed, Emmanuel (2002) used the concentration of fecal coliforms as an indicator of water contamination by fecal bacteria. Moreover, consider it as an indirect indicator of the massive presence of antibiotics and/or disinfectants.

In our study, the values of Fecal Streptococci (FS) ranged from 1 × 10^6 to 4 × 10^6 CFU/100 ml, with an average load of 2.29 × 10^6 CFU/100 ml (Table 3), which is higher than that observed in Al Ghassani Hospital of Hassan II University Hospital in Fez (Tahiri et al. 2009), but this value is comparable to that observed in hospital effluents of Mohammed V in Meknes (Ameziane and Benabidate 2014).

The average concentration of Staphylococcus Aureus (SA) in the wastewater of the Mohammed VI University Hospital is about 2.71 × 10^6 CFU/100 ml (Table 3), which is higher than the values reported by Boillot (2008) and Chitnis et al. (2004) of 1.2 × 10^5 CFU/100 ml and 6.08 × 10^2 CFU/100 ml, respectively. This could be explained by the low consumption of water, cleaning agents, disinfectants and detergents in the hospital.

### Table 3. Results of microbiological analyzes of hospital wastewater

<table>
<thead>
<tr>
<th>Settings</th>
<th>TC (CFU/100 ml)</th>
<th>CF (CFU/100 ml)</th>
<th>FS (CFU/100 ml)</th>
<th>SA (CFU/100 ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Max.</td>
<td>15 × 10^6</td>
<td>12 × 10^6</td>
<td>4 × 10^6</td>
<td>5 × 10^6</td>
</tr>
<tr>
<td>Min.</td>
<td>6 × 10^6</td>
<td>3 × 10^6</td>
<td>1 × 10^6</td>
<td>1 × 10^6</td>
</tr>
<tr>
<td>Avg.</td>
<td>9.57 × 10^6</td>
<td>7.57 × 10^6</td>
<td>2.29 × 10^6</td>
<td>2.71 × 10^6</td>
</tr>
<tr>
<td>Standard deviation</td>
<td>3.10 × 10^4</td>
<td>3.10 × 10^4</td>
<td>1.11 × 10^4</td>
<td>1.50 × 10^4</td>
</tr>
</tbody>
</table>
CONCLUSION

According to our study, the wastewater from the Mohammed VI University Hospital was discharged into the sewerage system without any treatment, except for the liquid waste from the laboratory and the operating room, which was collected by a specialized company and processed at the city of Casablanca.

The results of the analyzes performed showed that the hospital wastewater had a high level of physicochemical and bacteriological pollution. In fact, the results of these analyzes showed that the values of the three physicochemical parameters (TSS, COD and $\text{BOD}_5$) were far above the values established in the standards for direct and indirect discharges.

Moreover, the fecal contamination of these effluents is higher compared to other hospital effluents mentioned in the bibliography. In addition, significant concentrations of specific control bacteria responsible for nosocomial infections and polyresistant to antibiotics, such as staphylococci, are present in these hospital effluents, which is a potential danger that should not be overlooked.

Considering all these data, we cannot ignore the harmful effects of these pollutants and micro-organisms on public health and the environment, which is why a clear and comprehensive regulatory framework for the treatment of these effluents must be developed. In addition, knowing the characteristics of these hospital wastewaters will help ensure proper pretreatment before they are discharged into the public sewage system, and we can take advantage of previous studies conducted worldwide in the field of hospital wastewater treatment and select the appropriate processes.

Acknowledgments

The authors would like to thank all those who contributed to this study. A.B. Ayoub, the corresponding author, is acknowledged for his contributions in collecting and analyzing the wastewater and preparing the manuscript. H. Abdessamad is acknowledged for his contributions in wastewater analysis and interpretation of results. E. El Habib and L. Hayat are acknowledged for their participation in interpreting the results and preparing the manuscript.

The authors also express their sincere thanks to L. Mandi, Director of the National Center for Research and Study on Water and Energy, for providing the necessary tools for the analysis. The authors are grateful for the support and resources provided by the Center.

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