WASTEWATER EPIDEMIOLOGY TO ASSESS ILLICIT DRUG CONSUMPTION DURING CARNIVAL IN BELO HORIZONTE/MG: ANALYSIS OF CHEMICAL TOILET EFFLUENT SAMPLES

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Highlights:

- Wastewater based epidemiology as a fast and effective instrument for quantification of analytes in samples;
- Quantitative and qualitative analysis of illicit substances consumed during the 2024 carnival in Belo Horizonte;
- Use of Liquid Chromatography coupled with Mass Spectrometry for analysis in complex matrices (raw effluents).

Keywords: Wastewater Epidemiology; Illicit Substances; LC-MS/MS.

INTRODUCTION

Wastewater based epidemiology (WBE) aims to identify substances excreted by the population in wastewater. Such substances include chemicals, viruses, hormones, and others. This mechanism is widely used to identify consumption patterns and diseases within a society since sewage contains traces of these elements. A recent example is the surveillance of Covid-19 in the wastewater from Belo Horizonte by researchers from the Federal University of Minas Gerais. Data was cross-referenced with governmental data to estimate the number of people infected with the virus. Similarly, some international studies apply wastewater epidemiology to investigate the consumption of illicit substances.

During festive periods, it is expected that the consumption of alcohol and illicit drugs increases. The largest popular street festival in the world is the Brazilian Carnival, which occurs yearly in February. The increasing consumption of recreational and illicit drugs during festive periods, has become a significant concern for both national and international organizations. This trend involves substances like cannabis, cocaine, ecstasy, amphetamines, and even pharmaceuticals like Viagra.

Belo Horizonte (BH) is the capital of the State of Minas Gerais and is home to almost 3 million people. For the past 10 years, BH has attracted people from all over Brazil to enjoy the carnival. In 2024, for example, 262 thousand tourists were reported to be in the city, reaching a record number. Carnival in BH is known for its "street parades", which take place at previously scheduled locations and times organized by the City Hall. With the rapid increase in population in the city, it is necessary to allocate

temporary solutions to provide appropriate sanitation conditions during the festival. One alternative is the installation of chemical portable toilets (CPT) in strategic regions of the city. These toilets are mainly used by those attending the Carnival Street parades and can be used for the application of WBE by accessing the occurrence of recreational drugs and their metabolites, as well as for gathering information on drug use within a specific population and in developing consumption scenarios/profiles for the population. Therefore, this proposal aims to assess the occurrence of recreational and illicit drugs in CPT samples collected during the 2024 carnival season in the metropolitan region of Belo Horizonte.

METHODOLOGY

Samplings were conducted before the effluent from CPTs were discharged into the wastewater treatment system as the trucks carrying the effluent arrived at the plant between 1:00 AM and 6:00 AM. One liter of the effluent collected by each truck was sampled during the three days of the festival (February 11, 12, and 13/ 2024), totaling 14 samples. Samples were preserved prior to physicochemical characterization for pH, Color, Total solids, Turbidity, Chemical Oxygen Demand (COD), and Biochemical Oxygen Demand (BOD). For solid-phase extraction (SPE), samples were thawed at room temperature and filtered through a glass fiber filter (0.7 μm). The pH of all samples was adjusted to 3 and cartridges (Strata-X 33 μm, 500 mg/6) were conditioned as follows: 5 mL of MeOH, 5 mL of ultrapure water, and 5 mL of ultrapure water (pH = 3). Samples were percolated (200 mL) through each cartridge. Clean-up was performed using 10 mL of ultrapure water immediately after sample percolation. Then, cartridges were left to dry under vacuum for 15 minutes. During elution, 6 mL of MeOH were processed through all cartridges under gravity flow and extracted samples were deposited into falcon tubes. After this process, each sample was filtered by PVDF syringe membranes (0.22μm) and stored in a vial. The targets for WBE were cocaine and its metabolites and MDMA. The SPE extracted samples were quantified by Liquid chromatography–tandem mass spectrometry (LC-MS/MS).

RESULTS AND CONCLUSIONS

The characterization of CPT effluents is described in Table 1 which reveals the complex characteristics of CPT effluents, including an alkaline pH and elevated levels of Color, Turbidity, Total Solids, COD, and BOD. These parameters are indicative of organic and inorganic pollutants present in the effluents. Moreover, as samples represent different regions of the metropolitan region of BH (Belo Horizonte), they provide a comprehensive overview of the issue. The observed COD/BOD ratio indicates that the CPT effluent is non-biodegradable, meaning it contains substances that cannot be easily broken down by biological processes. This characteristic can also pose challenges for sewage treatment systems, especially smaller ones, which may struggle to effectively treat such effluents. Given the estimated population of 5 million people observed during the study period, nearly double of the population of Belo Horizonte, it is evident that effective management of effluents from the CPT is crucial for protecting the environment and public health.

Table 1 – Physicochemical characterization of portable toilets effluents sampled during 2024 BH carnival

Samples	pH	Color (mg/L Pt Co)	Turbidity (NTU)	Total Solids (mg/L)	COD (mg/L)	BOD (mg/L)	COD/BOD
1.1	8,43	182	365	8755000	1076	192	5,6
1.2	9,04	270	9072	49340000	2582	1374	1,9
1.3	8,94	376	815	28030000	2110	1152,9	1,8
1.4	8,98	253	6955	27230000	8142,3	1720	4,7
2.1	8,71	456	5000	49110000	11890	1899,5	6,3
2.2	8,91	429	824	33940000	5686,2	1206	4,7
2.3	8,92	447	4448	33715000	6899,7	1530	4,5
2.4	8,92	384	1172	39375000	5162,1	1122	4,6
3.1	9,02	460	826	38915000	5972,4	1140	5,2
3.2	8,82	450	394	43235000	7090,5	1087,5	6,5
3.3	9,12	258	996	46835000	7793,5	1925	4,0
3.4	9,03	155	752	39550000	6976,0	2595,1	2,7
3.5	9,15	163	536	36210000	7439,3	2075,6	3,6
3.6	9,07	253	2504	1545000	1076	192	5,6

Samples 1.1 – 1.4 (02/11/2024), 2.1 -2.4 (02/12/2024) and 3.1 -3.6 (02/13/2024)

The analysis of samples from CPTs which were located in 5 different regions in Belo Horizonte were analyzed by LC-MS/MS and indicated the occurrence of both cocaine and MDMA. The concentration of these illicit drugs is reported in parts-per-billion (ppb) (Table 2) and exceed the initially expected concentrations as they surpass concentrations detected in other studies. Drug consumption profile per person during Carnival in BH may be estimated based on these values. The highest concentrations of cocaine and MDMA were detected in the central region of the city (samples 1.3, 3.1, and 3.3), where most carnival groups parade. Followed by sample 2.1, located in the northwest region of the city near the Pampulha lagoon, is almost 10 km from the city center. Finally, point 1.1 is outside the metropolitan region of Belo Horizonte (20 km away) presented the lowest concentrations of drugs evaluated. The increase in recreational and illicit drug use during festive periods poses a challenge that requires strategies from sanitation companies, health authorities and policymakers. Furthermore, results demonstrate the effectiveness of wastewater-based epidemiology to estimate the consumption of illicit drugs by the population.

Table 2 – Concentrations of cocaine and MDMA in effluents from portable toilets in use in Belo Horizonte during the 2024 carnival

Samples	Cocaine concentration (µg/L)	MDMA concentration (µg/L)	Truck CPT volume (m³)
1.1	0,2	1	10,5
1.3	23	681,3	7

2.1	8,5	262,5	4,5
3.1	10,2	1414,5	6
3.3	5,3	501,7	5

Samples 1.1; 1.3 (02/11/2024), 2.1 (02/12/2024) and 3.1; 3.3 (02/13/2024)

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