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Tracking Criminal Actions on Drug Consumption on Campus with Wastewater-Based

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Abstract. Wastewater based epidemiology is applied to estimate the crime of narcotics consumption, twelve narcotics on campus. Seven consecutive 24-hour composite raw wastewater samples (n=10) were obtained once a month from sampling locations that captured > 95% of campus wastewater. The samples were analyzed for indicators of consumption of *morphine, codeine, oxycodone, heroin, fentanyl, methadone, buprenorphine, amphetamines, methylphenidate, alprazolam, cocaine,* and *MDMA* using LC-MS/MS. Eleven indicator compounds (*oxycodone, codeine, norcodeine, 6-acetylmorphine, EDDP, amphetamine, alprazolam, alpha hydroxyalprazolam, cocaine, benzoylecgonine, and MDMA*) occurred at a detection frequency of 100% across studies, followed by *morphine-3-glucuronide* (98%), noroxycodone (95%), *methylphenidate* (90%), *heroin* (7%), *norfentanyl* (7%), and *fentanyl* (5%). Estimates of average drug consumption are classified as follows in mg/day/1000 persons: *heroin* (474 ± 32), *cocaine* (551 ± 49), *amphetamines* (256 ± 12), *methylphenidate* (236 ± 28), *methadone* (72 ± 8), *oxycodone* (80 ± 6), *alprazolam* (60 ± 2), *MDMA* (88 ± 35), *codeine* (50 ± 4), and *morphine* (18 ± 3). This study of criminal acts against narcotics trafficking in campus resulted in basic data on 12 narcotics for campus and demonstrated for the first time the feasibility of detecting metabolites.

Keywords. Crime, Narcotics Consumption, Wastewater-based Epidemiology

1. Introduction

Wastewater where the contributing population is mostly college-age adults and assesses and measures the potential for prescription consumption which college-age adults (ages 18-22) throughout have targeted and illicit drugs in the campus population. has historically been associated with the highest percentage of drug abuse offenses of all age groups, with 24-28% of respondents in the 2019 survey admitting to using drugs in the past 30 days. While 48% of high school respondents to the same survey reported having tried at least one illegal drug in their lifetime, the prevalence of drug use was shown to be higher in those aged 18-29 years. People aged 15-24 have seen some of the lowest overdose death rates (4-10 deaths per 100,000 people) across all age categories from 2000 to 2019, but the next age group (25-34 years) has been identified with the number of drug overdose deaths. highest in 2019 (35+ deaths per 100,000 people).

This observation can partly be explained through drug-related associative learning, in which drug-seeking habits are maintained later in life, whereas the subjective e-effect that initially promotes drug use is reduced. Continued neurological development in the early 20s

coupled with changes in brain chemistry due to drug use may have a marked effect on this demographic group. Addressing crime and substance abuse in college-age adults should be seen as a major task – but understanding the level of abuse in this age category is met with a significant no-no. Current data analysis involves a combination of population surveys, crime statistics, medical records, and narcotics seizure data, but this analysis provides data on previous years and may not adequately capture the current state of drug use. Expensive and complicated procedures can also inject into studies unwanted bias through misrepresentation in self-reporting surveys. First proposed in 2001, wastewater-based epidemiology has proven to be a viable alternative to current data collection methods on drug use and non-criminal use and abuse. This tool has been applied worldwide to obtain near-narcotics abuse statistics real-time for various population sizes. The concept of tracking chemical consumption and fate through diagnostics rooted in urban process flow analysis has been further expanded under the umbrella and moniker of urban metabolic metrology, which examines multiple environmental matrices to estimate health statistics for a population or area of interest. Time analysis and low-weight composite wastewater samples can provide unique wastewater-based epidemiological insights into consumption statistics, with: flow-weighted estimates provide statistically more favorable results, and can theoretically be obtained for a broad spectrum of chemical products consumed and discharged by a population. Sampling for the purpose of wastewater epidemiological analysis generally focuses on the main work of wastewater treatment plants but this technique has also been applied to obtain equivalent information for smaller population sizes, such as college campuses or prisons. Wastewater-based epidemiology has seen limited application in. To the authors of knowledge four studies have applied the technology at sampling points local to campuses to obtain drug use statistics. Two studies were primarily interested in measuring attention deficit hyperactivity disorder of prescription drug use, while the other two were screened for a wider range of illicit and prescription drugs including: *amphetamine, opioids, cocaine, cannabinoids, and lysergics*. None of these studies screened the potent synthetic opioid fentanyl, despite its known association with still-rising overdose rates and death from drug abuse. These studies typically focus on a single university and are thus limited in their generalizability due to social, economic, and state factors that cause variations in drug use around the world.

Therefore, the main objective of this study is to (i) apply the liquid chromatography tandem mass spectrometry method to detect 12 drugs of abuse, including some of their metabolites which are known in the university environment; (ii) obtain the first data on the incidence of fentanyl on campus.

2. Materials and methods

2.1 Study sites and wastewater sampling methods

Seven 24 consecutive hours of flow-weighted wastewater samples were collected using an automated sampler for a period of one week per month from August 2019 to December 2019. Sampling site 1 accounted for approximately 95% of the total wastewater carried away campus while sampling location 2 accounted for about 5% of the total ouch. The population of sewer contributors for both sites ranged from about 15,000 to 60,000 people depending on the day of sampling. The mean age (26.5 years) of the catchment population was estimated by comparing the mean age of the population in years with available age data. About 53.6% of students were male and 46.4% female, with 81.8% pursuing a bachelor's degree and 18.2% pursuing a bachelor's degree. The ethnic demographics of undergraduates are as follows: white: 50.5%.

Population demographics were obtained from publicly available records from participating universities. The campus has a sewer system designed to separate municipal

wastewater from rainwater inlet. The ambient temperature during the study period ranged from 3.4 to 42.8 °C. The average travel distance of waste within the study catchment area is estimated to be 2700m. Waste retention time in the catchment system is estimated to average around 50 minutes but can range from 10-110 minutes depending on travel distance and sewer flow conditions. Sampling was carried out one week per month during the study period h through e. shared between study researchers and city personnel. Samples are stored in vials for transport and storage and immediately processed through solid phase extraction upon receipt by laboratory personnel. The remaining samples and concentrated sample extracts were stored at 20 °C until analysis.

2.2 Target

Ten parental prescriptions and illegal narcotics and nine metabolites monitored in raw wastewater collected from two sampling sites on the university campus that constitute the majority of campus wastewater The drug under investigation is *morphine's* main metabolite *morphine-3-glucuronide codeine (COD),* a metabolite main metabolite *norcodeine, oxycodone*, its main metabolite *noroxycodone*, *fentanyl*, its main metabolite *norfentanyl*, *heroin*, its minor but exclusive metabolite *6-acetylmorphine*, *methadone's* main metabolite *2-ethylidene-1,5-dimethyl-3,3-diphenylpyrrolidine, buprenorphine,* its metabolite *norbuprenorphine, amphetamine, methylphenidate, alprazolam (ALP),* its metabolite *alpha-hydroxyalprazolam, cocaine,* its metabolite *benzoylecgonine,* and *3,4-methylenediox methamphetamine*. High purity standard (> 97%) of the target compound. Eighteen deuterated compounds, one for each of the parent opioid target compounds were also purchased from cerilliant for use as internal standards for quantity.

2.3 Isotope dilution liquid chromatography

Mass spectrometric analysis was performed on an instrument coupled to a Shimadzu HPLC. Chromatographic separation was achieved with a Symmetry C18 3.5µm by 6.4mm by 75 mm analytical column preceded by a protective column of the same material. The analyte was fed into a mass spectrometer using an electrospray ionization probe operating in positive mode. Multiple reaction monitoring was used for qualitative analysis. Further information on isotopic dilutions and mass spectrometric analysis is provided in the supporting information.

2.4 Analysis concentration in raw wastewater

All filtered analyzes were considered as potential indicators of drug consumption during the sampling campaign. Potential loss of opioids and metabolites from wastewater during sample extraction was corrected through internally labeled standards and isotope dilution methods. The mass loading of narcotics was calculated from the analyte concentration in raw wastewater (in units of ng/L) for daily.

$$\text{Beban Massa}\left(\frac{mg}{hari}\right) = \text{Konsentrasi Mentah}\left(\frac{ng}{L}\right) \text{Mengalir}\left(\frac{L}{D}\right) \left(\frac{1\,mg}{1.000.000\,ng}\right)$$

2.5 Estimated mass per capita consumption of narcotics

Estimates of drug consumption were obtained by normalizing the mass burden of narcotics with estimates of the contributing population and then subjected to a correction factor that took into account the metabolic excretion of the compound, and the molar mass ratio of the

indicator compound to the parent opioid. Population estimates of analyte concentrations assumed 5.1mg/day/person for *caffeine* [37,38], 13.8mg/day/person for *paraxanthine*, 0.125mg/day/smokers for *nicotine* [39], and 14% smoking prevalence in the population [40]. Per the National Center for Drug Intelligence's report on Heroin Consumption the average daily mass use of pure heroin is assumed to be equal to 50mg/day per user. Estimates of mean doses of *cocaine* (50mg/dose) and *MDMA* (100mg/dose) were obtained from relevant human pharmacokinetic studies. The remainder of the prescription dose information is obtained from the prescribing guide. Further explanation of the estimated mass per capita is available in the supporting information document.

2.6 Estimated black market value and other assumptions

The black market value of heroin and cocaine is calculated by comparing the estimated average mass of the narcotic compound consumed with the "average street value and drug purity. Further assumptions are that: (i) there is no waste water loss due to leaks or pipe damage; (ii) no transformation or degradation in the sewer; and (iii) no direct discharge of drugs into the sewer system.

2.7 Statistical analysis Statistical

analysis of the data was performed with the product combination, software Analyst 1.5 The normality of the data set was determined through two analyzes run on IBM SPSS 25; (1) analysis of slope and kurtosisz-value, and (2) Shapiro-Wilk test for normality. Following the epidemiological statistical testing of wastewater described previously, a two-tailed T-test was used for comparison of weekend vs weekday mass load observations.

3. Results and Discussion

3.1 Method performance Method

Detection limits (MDLs) for various parent narcotics and their metabolites ranged from 0.2 to 1.7 ng/L with the exception of *buprenorphine* (140 ng/L) and *norbuprenorphine* (120 ng/L), data which are in line with previous [20-22.24]. All MDLs were determined according to the EPA guidelines described in 40 CFR 136, Appendix B in the materials supplementary. Potential loss of narcotics and metabolites from wastewater during sample extraction was corrected using internal standards labeled and isotope dilution methods. Recovery from 10 trials of sample matrix spikes for various analytes was averaged 119%. The accuracy of the analysis was expressed as the percent relative difference (RPD) for duplicate samples of composite wastewater, an average of 7.4%. A detailed description of the procedure for determining the rate of recovery experimentally is provided in the supporting information (Section S-1.4).

3.2 Concentrations of Narcotics and Metabolites in Raw Wastewater

Concentrations in raw wastewater (ng/L) for all analytes of interest were screened for each sampling site for seven consecutive days.

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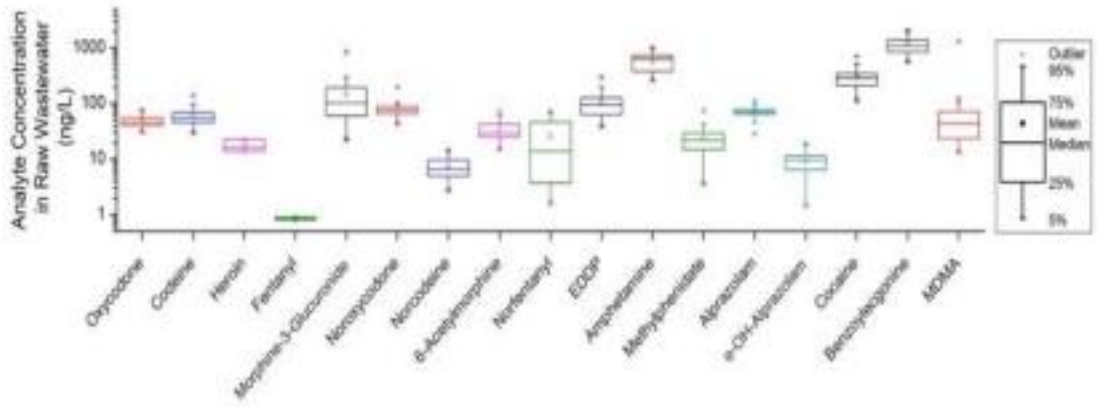

Figure 1. Box plot identifying analyte concentrations in raw wastewater (ng/L)

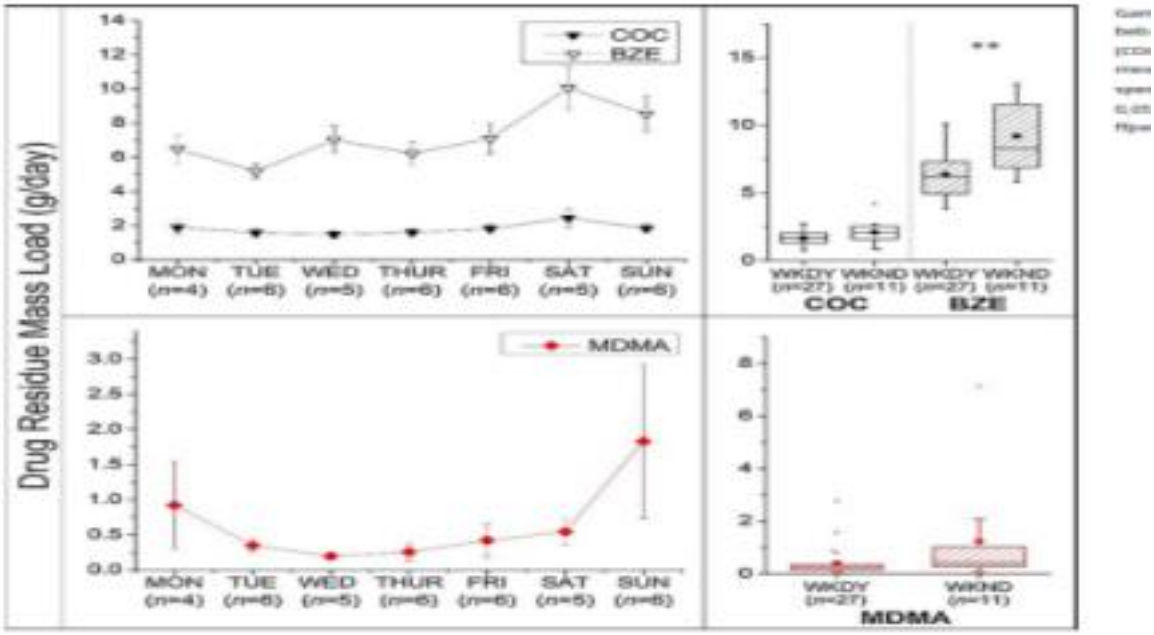

Figure 2. Average drug mass load per day and comparison between weekend and weekday mass loads.

At both sampling sites with the exception of 6-acetylmorphine (DF = 30%) and *alpha-hydroxyalprazolam* (DF = 33%) at one sampling site. *Fentanyl* and *norfentanyl* analytes were detected sporadically during the sampling campaign. Due to the relatively low prescription rate of fentanyl compared to other commonly prescribed *opioids*, the infrequent detection of this analyte may suggest infrequent non-medical use of *fentanyl*. No other narcotics considered in this analysis were identified with a sporadic pattern. Following normalization of the data by wastewater flow, the resulting data were analyzed to determine trends and patterns of weekly consumption. The data were log transformed for normality and tested with 2 T-tests to examine statistical differences in drug use weekdays and weekends. Mass load did not vary significantly between weekend and weekday use ($p > 0.05$) for any of the monitored opioids, as indicated by values for parent and corresponding metabolites. This finding is similar to the trend in the use of *opioids* reported previously derived from an analysis of expenses masses *amphetamines* on weekdays was statistically higher ($p < 0.001$ compared to bulk load the weekend, but this trend was not observed for *methylphenidate* ($p = 0303$).

While the burden of the masses may represent the most robust data that can be obtained through epidemiological analysis of wastewater, it is also important to consider the statistical

differences of the normalized mass load population – despite the known increase in error associated with population estimation via chemical biomarkers. While most of the statistical analyzes mentioned above was confirmed when two T-tests were applied to the normalized values of the transformed population of weekday and weekend drug consumption - some differences were observed. The statistical differences between weekdays and weekends differences were confirmed for oxycodone (p=0.032), amphetamines (p=0.009), coca in (p=0.044), *benzoylegonine* (p=0.0002), and MDMA (p=0.025), while there was no statistical difference.

While analyte concentrations in raw wastewater are required for further data modeling, they provide little insight beyond analysis of long-term trends. This was demonstrated when comparing amphetamine concentrations in raw wastewater at sampling site 1 (mean AMP = 574 ± 10) with sampling sites.

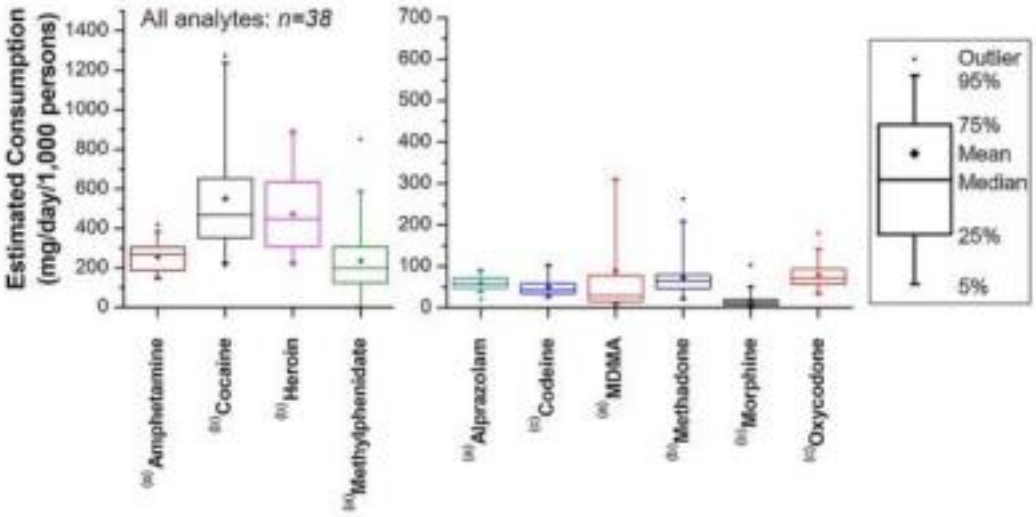

Figure 3. Estimated drug consumption of campus residents.

While one might perceive higher amphetamine use in the second location, in water use and in rainwater fluxes could also encourage such a finding. The computational mass flux of the concentration data eliminates the potential for bias caused by dilution of the effluent. This suggests that comparisons of analyte concentrations in raw wastewater may not yield comparable results and this is an example.

3.3 Estimates of substance consumption

Population narcotic analyte mass loads were normalized and corrected using a co-kinetic pharma correction factor to provide estimates of narcotic consumption. The normalized and corrected population data were then compared with the dosage guidelines to obtain an estimate of the ingested dose of

3.4 Number of drug users and estimated black market value

Estimates of the number of drug users presented in units of users/1000 people yield values between 0.5 ± 0.1 (*MDMA*) to 14.9 ± 0.6 (*alprazolam*) (Table S8), with significant values were observed for *oxycodone* (4 ± 0.26), *heroin* (7.9 ± 0.6), *amphetamines* (4.3 ± 0.2), *methylphenidate* (3.9 ± 0.47), *alprazolam* (14 .9 ± 0.6), and *cocaine* (5.8 ± 0.5). The number of calculated *heroin* users exceeds the national average of 0.21% to four times but is below the national average lifetime *heroin* use of 1.6%.

3.5 Effects of elimination half-life

After ingestion of the narcotic parent compound and its metabolites are stored in the body and excreted over time. This excretion, generally expressed as the elimination half-life of the narcotic, is governed by the average dose of the drug, route of administration, duration of use, metabolic rate, and chemical properties. Consideration of the elimination half-life in the development of the chance to return method. analytical techniques and increase the value of the data collected. The drug analytes used in this study displaying estimated elimination half-lives ranging from 0.6 to 39.5 hours were collected from the currently available pharmacokinetic and pharmacodynamic literature [56,57,70-80], which may influence the presented results. The elimination half-life of *6-acetylmorphine* (0.6 hours) results in the elimination of 99.9% of the compound from the body in a 24-hour period, so the detectable presence of *6-acetylmorphine* can be associated with heroin consumption in the 24-hour sampling period. The long elimination half-life of *EDDP* (39.5 hours) results in an excretion period exceeding 16 days from a single dose of methadone making estimates of daily consumption in FFI cultus. It is also important to consider the route of administration of narcotics when considering the elimination half-life as this will have a marked effect on the excretion rate.

3.6 Ethics in Monitoring

Researchers, governments and professionals working in wastewater-based epidemiology are expected to conduct their work ethically and transparently – especially when working with sensitive catchment areas such as university campuses. Prior to starting the work described in this manuscript, a public forum was held that allowed individuals to voice their concerns regarding the epidemiological monitoring of campus-based wastewater. The public sensitivity of the data must also be considered alongside the potential benefits and drawbacks of obtaining data on specific drug use.

With regard to the campus population, it is also important to note that many people contribute to sewerage on a daily basis, and thus many reasons can influence the occurrence of narcotic indicator compounds in campus wastewater. While it is desirable for privacy reasons to obtain samples from areas with a significant number of contributing individuals, for example, hundreds or ideally above 1000 individuals, monitoring such large populations can have the disadvantage of losing some signals due to diluents. the value of population size used to obtain the best possible data while protecting the privacy of the monitored sub-populations is an ethical question that cannot be answered by the researcher alone. As was done through the workshops leading up to this study, ideally all stake-holders community should be invited and have the opportunity to have their unique perspectives heard, valued and considered.

3.7 Limitations

Analytical concentrations in raw wastewater and narcotic mass loads can be considered the most robust data that can be collected from the procedure because errors mostly stem from deviations in sample collection, preparation, analyte loss, population variance, and instrument error. These errors can be quantified through sample replication and the use of appropriate controls. Estimates of drug consumption can provide a more realistic analysis of the data but also include errors in the analysis. The variation in daily mass consumption of narcotics between individuals within a population, the specific prevalence of narcotic abuse within certain limits, areas of uncertainty in pharmacokinetic metabolism and excretion rates, uncertainty in the number of contributing populations, potential for sample degradation during a 24-hour sampling procedure, and the rate of degradation of narcotics in sewers and drains. /or biotic transformations can distort results, potentially by an order of magnitude. Comparison of the

ratios of metabolites and parental narcotics can provide some insight into the magnitude but cannot fully quantify the impact on data quality. Specific analysis of narcotics with various limiting factors such as low proxies for urinary and faecal excretion or rapid degradation may present additional challenges for the quantity of certain narcotics in wastewater. Furthermore, the relatively large percentage differences observed for some samples during the sampling campaign can be explained in part by errors in sampling, preparation, and analytical procedures, as well as the hydrophobicity of the target analyte. The lack of detection of buprenorphine and its metabolite norbuprenorphine in campus samples is likely due to the sensitivity of the method to the compound but could also return.

4. Conclusion

The concentration and detection frequency of narcotics examined in this study exceeds the values presented in previously published campus-related literature. Estimates of consumption values vary by narcotic compared to estimates but are mostly higher than international estimates from the city-based international literature. the findings correlate with the observed higher drug consumption of college-aged young adults and may suggest that variations in drug use can be tracked and compared between different geographically via a first analysis of fentanyl and its metabolites in campus wastewater may also suggest illegal non-medical consumption in the campus population. Certain design factors such as reduced retention time waste and consideration of elimination half-life in analyte selection were chosen to improve the study design. This may explain the high concentrations observed for some narcotic analytes but further examination into the degradation and metabolism of target analytes across appropriate demographics is needed to understand these observations. These results have shown that application in a university setting can provide useful temporal information related to the use of various narcotics in the near future and should be adopted by institutions that have an interest in the welfare of society. college population.

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