FENTANYL DEGRADATION IN SYRINGES OBTAINED FROM IV DRUG USERS IN WASHINGTON D.C.

by

Shayla Montalvo Vargo

A Research Project
Submitted to the
Forensic Science Forensic Research Committee
of
George Mason University
in Partial Fulfillment of
The Requirements for the Degree
of
Master of Science
Forensic Science

Primary Research Advisor
Morgan Levitas, MFS
Supervisory Chemist
D.C. Department of Forensic Sciences

Secondary Research Advisor
Lucy Nguyen, MFS
Chemist
D.C. Department of Forensic Sciences

GMU Graduate Research Coordinator
Dr. Joseph A DiZinno
Associate Professor
GMU Forensic Science Program

Spring Semester 2022
George Mason University
Fairfax, VA

ACKNOWLEDGEMENTS

First and foremost, I would like to thank my research advisors Morgan Levitas, Lucy Nguyen and Dr. DiZinno for helping throughout this tedious research project and guiding me to produce original work that can be useful in the Forensic Chemistry Unit. Thank you to my coworkers in FCU and CTU for helping me obtain data for my research using syringes from the syringe exchange program. Thank you to my friends and family for cheering me on and keeping me grounded.

DEDICATION

I would like to dedicate this research project to my dad, mom and Tori. Without your support and encouragement, I would not be where I'm at today. Thank you for always pushing me to challenge myself and continue my education.

And lastly, to my husband, Sergio. There are no words that can fully convey how grateful I am for all the encouragement and support you have given me from the start of my program. You make me a better human being and my life has so much meaning because you. This one is for you. Te quiero mucho. 1000/10

Table of Contents

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Considering the continued opioid epidemic, it is important to understand the local drug trends to support public health initiatives. In 2020, the Public Health Laboratory (PHL) within the D.C. Department of Forensic Sciences (DFS) established a needle-exchange program to monitor local intravenous (IV) drug trends. The syringes are collected anonymously from various programs throughout the District and are analyzed in the lab for the presence of controlled dangerous substances (CDS). In addition to identifying the CDS qualitatively, there is also forensic interest in determining the degradation of specific drugs in syringes over time. Presented in this study is a timeline of fentanyl stability and other various adulterants such as heroin, etizolam and xylazine that are commonly found in combination with fentanyl in syringes from the D.C. needle-exchange program. Polypropylene syringes were conditioned to mimic used syringes among intravenous drug users in D.C. and subsequently analyzed for drug residue after 28 days. The method followed in this study consisted in the detection of unknown quantities of the target drugs via gas chromatography-mass spectrometry. This proved to be successful in the identification of degraded compounds as well as the quantification of the specific drugs over a tracked period of time. Data gathered from this study supported the efforts of the D.C. PHL Forensic Chemistry Unit (FCU) in the needle-exchange program by providing an accurate timeline for storage protocols and the optimal timeframe for drug analysis.

Key Words: Degradation, Syringes, Fentanyl

BACKGROUND

The Public Health Laboratory (PHL) within the D.C. Department of Forensic Sciences established a needle-exchange surveillance program in 2020 to monitor local intravenous (IV) drug trends. The program included analyzing syringes collected from four harm reduction facilities located in Washington, D.C., where needle-exchange services were conducted (Evans et al., 2021). The facilities offer a service where IV users can exchange their used syringe for a new, clean syringe. This program supports the community in three primary ways by 1) reducing the spread of blood-borne pathogens such as Hepatitis B and C, 2) facilitating safe disposal of used syringes, and 3) spreading awareness of available treatment programs for those who are battling drug abuse. In partnership, the syringes are collected anonymously and are packaged in a syringe tube sealed with biohazard tape for transportation to the laboratory weekly for drug residue analysis.

Analyzing the residual content captures current drug abuse trends within the IV user population. The analysis is performed by chemists in the Forensic Chemistry Unit (FCU) within the PHL via a chloroform/methanol extraction process. The residue is screened for all drugs – controlled and non-controlled dangerous substances. By surveilling for all controlled dangerous substances (CDS), the syringe program can serve as an "early warning system for dangerous emerging substances" and help monitor any shifts in drug consumption within the District (Evans et al., 2021). In addition to detecting drugs qualitatively, there is forensic interest in determining the degradation of drugs in used syringes over time as well as the amount of drug recovered from the residue.

INTRODUCTION

Results obtained from the needle-exchange program have contributed to understanding drug trends in Washington, D.C. and can help the community reduce opioid related deaths (Evans et al., 2021). As of March 31, 2022, the syringe surveillance program has processed 2585 total syringes. The top five drugs commonly seen in the District are fentanyl, cocaine, 4-ANPP, 6-monoacetylmorphine (6-MAM), and heroin. Out of all syringes processed to date, 1005 syringes (39%) contained at least fentanyl, with 345 syringes (13%) containing fentanyl and heroin, 266 syringes (10%) containing fentanyl and xylazine, and 19 (0.7%) containing fentanyl and etizolam (**Figure 1**). The remainder of the syringes (61%) did not contain fentanyl, but instead other CDS. The most recent data illustrates the high prevalence of fentanyl in used syringes in Washington, D.C. As shown in the numbers, adulteration of fentanyl is not uncommon with heroin, xylazine, and etizolam being adulterants often observed.

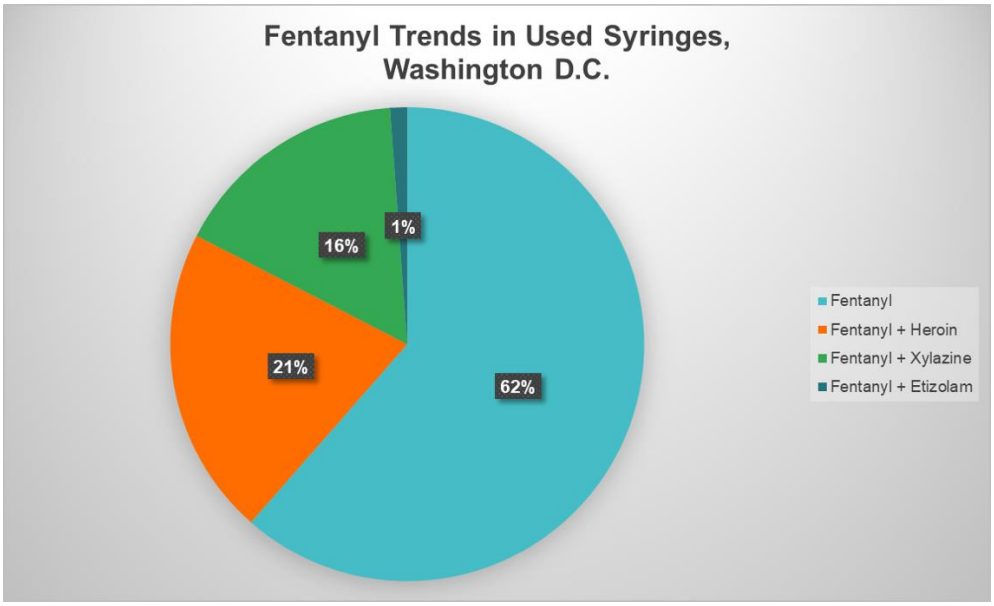

Figure 1. *2585 syringes were processed in the Forensic Chemistry Unit and 1005 contained fentanyl.*

As the syringe program continued to gather more data, the question of whether drugs degrade in the syringe became a topic of interest and in turn the subject of this study. Understanding how drugs degrade is important because it can help create a timeline for drug stability. It can also identify degradant products to see if there are any toxic chemicals that potentially are consumed when using drugs. From an analyst's viewpoint, this means that impurities or precursors may be detected instead of the actual parent drug, which may impact how current drug trends are being reported. To investigate this, the following drug targets were monitored to further understand the impact of degradation in IV syringes over a set period of time: fentanyl, heroin, xylazine and etizolam.

Types of Degradation

Degradation is the process of degrading or declining to a lower state, condition, or quality. In the case of drugs, this means that impurities or byproducts may develop depending on the environment or chemicals it's exposed to. The loss or change in drugs can occur in many ways, however, this research will investigate chemical and physical degradation because they are the more relevant degradation pathways in injection drug users. Physical degradation is associated with the environmental conditions the drugs are exposed to in the syringe. Chemical degradation is associated with the reaction of the drugs with blood or other bodily fluids.

Physical degradation covers how drugs can change potency or size. An example of this is exposure to humidity which can cause drugs to absorb water, reduce its concentration, and change its effectiveness. A more relevant example is drug-plastic interaction between the polypropylene-type syringes that are common among IV drug users. Drug-plastic interaction has been previously

studied in the pharmaceutical industry, where absorption has been a major challenge when drugs are stored in plastic materials (Ekoja, 2017). Certain plastic containers are known to absorb individual components of the solvent stored in them or on the contrary, the solvent may leach several undesired components from the plastic causing the drug to alter (Hung et al., 1988). One study conditioned multiple syringes with different solvents to observe plasticizers and fatty acid profiling. Results from samples that use methanol as a solvent confirmed chromatograph peaks for stearic acid, palmitic acid, and silicon-based lubricants – siloxanes (Lee et al., 2015). Another study focused on calculating the abundance of palmitic acid and stearic acid; common contaminants found in syringes used in laboratory settings (Cheng & Yu, 2020). The results also showed high levels of the acids with plastic syringes, filters and pipettes. However, there was a significant decrease in these contaminants when the tools were switched to glassware. In this research, all syringes that were analyzed are polypropylene-type syringes. Therefore, the presence of free fatty acids and siloxane peaks are expected to be present in the chromatograph.

Chemical degradation is the process of changing the chemical structure of a drug. Hydrolysis, decarboxylation, and oxidation are some of the chemical degradations that drugs undergo in the body. The enzymes in the blood react with the drugs causing a change in the chemical structure of the parent drug and leading to precursors or metabolites. From a pharmaceutical standpoint, it is important to understand which functional groups of the drug structure are susceptible to metabolic reaction and which are stable (Snape et al., 2010). In a clinical setting, this information will improve the use of medication in patients or users. Any change in the original drug can cause the medication to lose its potency and alter its effect on the body. In a forensic setting, this information is useful for the study of drug detection and trends. It

can be assumed that chemical degradation of drug-blood interaction rarely occurs in the syringe barrel as the drug is injected intravenously. However, there is a common practice known as "booting and jacking ... in which an injection drug user (IDU) repeatedly draws blood from their vein into the drug filled syringe (booting) and partially flushes the mixture back into their vein (jacking)" (Ciccarone, 2013). The drug-blood interaction residue in the syringe contains markers in the form of chemical degradation. An example of a chemical change in a drug is the degradation of acetaminophen to its primary metabolite *p*-aminophenol via N-acetylation. One study observed the concentrations of both acetaminophen and *p*-aminophenol in the livers of rats and mice. It was found that acute hepatotoxicity occurs in mice but not rats (Song & Chen, 2001). Another study researched the degradation of morphine, codeine, apomorphine, and pseudomorphine in aqueous solutions kept in plastic syringes over a 12-week period (Hung et al., 1988). Several factors were observed such as exposure to light and different storage temperatures. Results reported that less than 3% of the drug was degraded when exposed to 25-watt light and stored at 22°C, mimicking normal lab setting conditions. Pseudomorphine was also noted as the major degradation product (Hung et al., 1988). This indicates that light exposure and mild temperature storage might not be critical sources of degradation. In fact, no major modifications to the lab conditions were planned for the research. However, the research will closely observe any precursors or contaminants that are linked to high exposure of light and high temperatures if observed.

As highlighted above, there are various factors that may contribute to the degradation of drugs in polypropylene-type syringes used among IV drug users. The focus of this research is to provide a qualitative and quantitative timeline of drug degradation in these syringes, which will have a positive impact on the detection capabilities of the D.C. Public Health Laboratory.

Target Drugs

As mentioned above, the target drugs in the research are fentanyl, heroin, xylazine and etizolam.

Fentanyl is a Schedule II controlled substance according to the Controlled Substance Act (CSA) created by the Drug Enforcement Administration (DEA). The drug can be administered appropriately to humans and for veterinary uses to treat chronic pain. Fentanyl is 50-100 times more potent than morphine, causing a very strong euphoric effect which is highly sought after by drug abusers (Drug Enforcement Administration, 2008). Due to fentanyl's high potency, users often have difficulty determining how much of the drug to take and often take a lethal quantity (Drug Enforcement Administration, 2008). Fentanyl converts into many fentanyl derivatives during metabolism, and therefore they should all be studied comprehensively to understand their synthesis. On average, 85% of fentanyl is excreted in urine and its major metabolite is norfentanyl, which is created by N-dealkylation at the piperidine nitrogen (Concheiro et al., 2018). Another fentanyl degradant is depropionylfentanyl, also known as 4-ANPP, found as a byproduct after fentanyl breaks down in the body. In addition, 4-ANPP is used as a precursor to make fentanyl-type drugs synthetically. As found in these studies, it is common to find fentanyl in its original chemical structure in significantly higher quantities compared to its metabolites in used syringes. For this reason, the research provides qualitative data of fentanyl and its metabolites but only focuses on a quantitively study on the amount of fentanyl found in the syringes. Fentanyl is often used as a substitute for heroin addicts, but it can induce severe respiratory depression depending on the amount administered.

Heroin is a Schedule I semi-synthetic opioid that also produces euphoric effects on the body and is only used illicitly since it does not have any approved medical uses. In 2005-2006, the DEA observed a nationwide increase of seized illicit fentanyl laced or spiked with heroin and it is commonly seen administered intravenously (Drug Enforcement Administration, 2008). In contrast to fentanyl, heroin metabolizes rapidly in the liver into three commonly seen metabolites: 6-monoacetaylmorphine (6-AM or 6-MAM), morphine and morphine 6-glucuronide (Dinis-Oliveira, 2019). Because of heroin's metabolic reaction, moderate concentrations of heroin compared to its metabolites are predicted to be found in the syringes used in this study. Observations of higher amounts of heroin might indicate that the drug was not exposed to the plasma from the blood of the IV user. In the research, primarily the quantity of heroin will be calculated to track its degradation.

Xylazine and etizolam are common adulterants that have been recently found present with fentanyl since the start of the syringe surveillance program in the PHL. Xylazine is a muscle relaxant drug explicitly for veterinary use only, however, there have been reports that show xylazine used as an adulterant (U.S. Drug Enforcement Administration DEA, 2021). Although there are no human medical uses for xylazine, it is not scheduled. Since there are very few studies of xylazine on human subjects, its metabolism is based on how different animals such as dogs, cattle, and horses breakdown the drug. When administered intravenously, xylazine is noted to have a rapid metabolism rather than a rapid excretion (EMEA, 2002). Therefore, small levels of xylazine should be seen in the syringes analyzed in this study. Etizolam is a benzodiazepine; a drug that depresses the central nervous system and used to treat insomnia and anxiety (U.S. Drug Enforcement Administration DEA, 2020). The drug is not scheduled in the United States because

there are no approved medical uses, but it is a prescribed medication in Japan, India and Italy (U.S. Drug Enforcement Administration DEA, 2020). The use of this drug has increased illicitly in the U.S., also as an adulterant, commonly seen with CDS to offset the effects. The data gathered from the syringes will show the presence and quantity of any contaminants seen with these adulterants in IV drug users.

Impact

In this study, the condition in which the syringes were exposed to before arriving to the lab is unknown. Therefore, the specific factors that cause drug degradation prior to collection are difficult to determine. For this reason, the subject of this study is to identify degradation trends after collection in normal lab settings at FCU. Ideally, the results of this study would provide a clear trend of drug degradation over time. However, variability in the results is expected due to the inherent variability in the conditions of the syringes collected. In an attempt to isolate the drug degradation solely on the lab conditions, polypropylene syringes were conditioned to mimic used syringes among IV users in D.C. and subsequently analyzed for drug residue after 28 days. These results are compared with similar analysis of “real” syringes collected from the needle-exchange facilities. The degradation study included measurements collected in three/four-day intervals for a total of 28 days.

The impact of this research will be multifold: 1) it will allow Forensic and Public Health laboratories to adopt preferred storage protocols for used IV syringes, 2) the degradation rate determined in this study will improve testing methods. For example, this research could serve as a tool to identify any interference or ion suppression matrix that can affect detection of residues

within syringes. In addition, 3) it will improve current extraction procedures for the identification of controlled dangerous substances in syringes within our laboratory.

METHODS

The methods of preparation and analysis in the research follows a standard operating procedure (SOP) written by FCU which was reviewed and approved by the laboratory.

The method consists of the preparation of mimicked syringes to estimated quantities of drugs in IV users' syringes and the analysis of drug degradation over time. The method relied on gas chromatography-mass spectroscopy (GC-MS) for the detection and quantity of the substances. The residue in the syringes needs to be extracted and prepared for GC-MS analysis. For this reason, each prepared syringe could only be evaluated once. Therefore, in order to study drug degradation over time, multiple mimicked syringes were prepared with standard concentrations to observe consistent trends. For example, on 3/2/22, 64 syringes were prepared with similar concentrations to evaluate drug degradation after different time intervals (1-day, 3-day, 7-day, etc.). In the following two sections, the sample preparation and analysis are explained in more detailed.

Sample Preparation

For the mimicked syringes, 1 mL of the following substances were drawn into individual 3 mL polypropylene syringes: fentanyl, heroin, xylazine, and etizolam each at a concentration of approximately 2 mg/mL. The drug solution consisted of a weighed amount of solid drug and deionized water (DI) which was vortexed until it was dissolved. **Table 1, 2** and **3** shows the calculations for the drug concentrations. Due to low supply on etizolam, not enough samples were prepared to evaluate degradation past a 7-day interval.

Table 1. *The drug solutions were prepared on 3/1/22 for the mimicked syringes. These samples were used to observe degradation in intervals of 0, 1, 3, 7, 17, 21, 24 and 28-days. Due to low supply on drugs, some concentrations were lower than 2 mg/mL.*

	Fentanyl	**Heroin**	**Xylazine**	**Etizolam**
Weight (g)	0.0305 g	0.0321 g	0.0321 g	0.0121 g
H_20 (mL)	16 mL	16 mL	16 mL	8 mL
Final Concentration	**1.91 mg/mL**	**2.01 mg/mL**	**2.00 mg/mL**	**1.51 mg/mL**

Table 2. *The drug solutions prepared on 3/2/22 for a 14-day interval period.*

	Fentanyl	**Heroin**	**Xylazine**
Weight (g)	0.00342 g	0.00400 g	0.00404 g
H_20 (mL)	2 mL	2 mL	2 mL
Final Concentration	**1.71 mg/mL**	**2.00 mg/mL**	**2.02 mg/mL**

Table 3. *The drug solutions prepared on 3/7/22 for a 10-day interval period.*

	Fentanyl	**Heroin**	**Xylazine**
Weight (g)	0.00403 g	0.00400 g	0.00424 g
H_20 (mL)	2mL	2 mL	2mL
Final Concentration	**2.02 mg/mL**	**2.00 mg/mL**	**2.12 mg/mL**

In the syringe preparation, the syringe barrel was coated with each drug solution for approximately 30 seconds, via plunging. The solution was then discarded, the needle was capped, and the syringes were stored in a cryovial box with no exposure to light in a room kept at 72°F. The syringes were then processed and analyzed following a timeline to track degradation versus time. Evidently, there was no sample preparation for the "real" syringes since they arrive to the lab from the syringe exchange facilities. Due to the unknown time frame of when the IV drug users used the syringes, the timeline of degradation was initiated when the syringes were received at the lab. These syringes were also properly stored in a cryovial box located in a 72°F room and processed after a recorded number of days. **Figure 2** depicts the 3 mL syringes used for the mimicked syringes (bottom) and the 1 mL syringes (top) that are distributed and collected at needle exchange locations. The syringes are concealed in a syringe tube and brought to the lab for drug residue analysis as part of the syringe surveillance program.

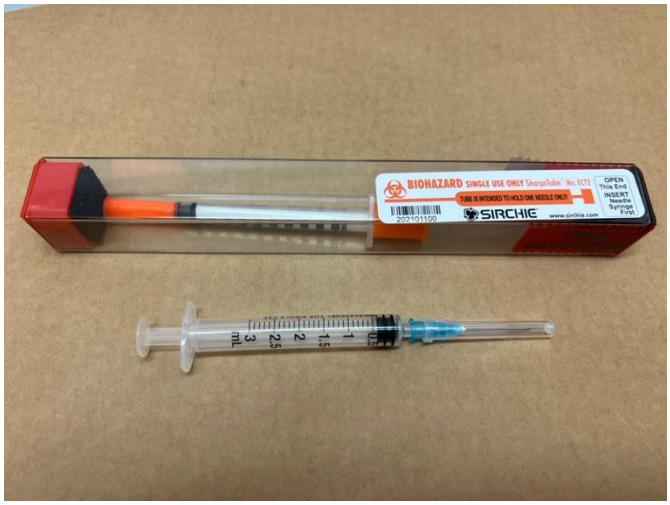

Figure 2. *Syringes obtained at the public health lab by needle-exchange facilities (TOP) and 3 mL needles used to mimic "real" syringes (BOTTOM).*

Analytical Method

After the designated amount of time, 1 mL of solvent with internal standard (0.025 mg/mL tetracosane in 9:1 $CHCl_3$:MeOH) was drawn into each mimicked syringe. To maximize drug residue extraction, the syringe was inverted several times for approximately 30 seconds before releasing the solvent into an autosampler vial for analysis. Due to possible contaminants that cannot be injected into the instrument, the "real" syringes had a similar but slightly altered extraction method. The drug residue was filtered through a wool pipette before releasing it into the vial (**Figure 3**). The drugs were identified using a GC-MS instrument, the Agilent 7890B GC System (Santa Clara, CA) coupled with an Agilent 5977B HES MSD (Santa Clara, CA) seen in Figure 2. 1 μL injections of the drug were carried out in split mode using a 20:1 ratio with Helium as a carrier gas with a constant flow of 1.2 mL/min. The samples were initially brought up from room temperature to 50℃ and increased to 180℃ at a rate of 30℃/min. Then it increased to 300℃ at a rate of 10℃/min with a 20 min hold time.

Figure 3. *Autosampler vials were loaded on the GC-MS starting with the point calibrators following the syringe extracts (LEFT). GC-MS used for the research (RIGHT).*

Each sample took approximately 36 minutes to run on the instrument: ~22 minutes for the drug method and ~14 minutes for the blank method. The mass spectra data was collected by running the mass spectrometer in full SCAN mode (50-500 m/z mass range). Each drug had a linear calibration curve established prior to the analysis of the syringes. The calibration standards used are shown in **Table 4** where the correlation value was >0.98. To confirm that the curve was still linear, a single point calibrator from each curve was processed with each batch of samples which serves as a quality control (QC).

Table 4. *Calibrators that were used to build a calibration curve for each drug of interest. The solvent used was 0.025 mg/mL tetracosane as the internal standard in a 9:1 $CHCl_3$:MeOH solvent.*

Calibrator	Fentanyl	Heroin	Etizolam	Xylazine
S7	50 µg/mL	-	-	-
S6	45 µg/mL	100 µg/mL	500 µg/mL	50 µg/mL
S5	40 µg/mL	75 µg/mL	400 µg/mL	40 µg/mL
S4	35 µg/mL	50 µg/mL	300 µg/mL	30 µg/mL
S3	30 µg/mL	25 µg/mL	200 µg/mL	20 µg/mL
S2	25 µg/mL	10 µg/mL	100 µg/mL	10 µg/mL
S1	20 µg/mL	5 µg/mL	50 µg/mL	5 µg/mL
R^2 Value	**0.987**	**0.990**	**0.987**	**0.994**

RESULTS AND DISCUSSION

Calibration Curve and Equations

After the raw data was collected from the GC-MS, a sequence of data processing and analytical calculation steps were performed to obtain the results presented in this paper.

With an automated process using Excel, the raw data from the GC-MS was organized into tables and charts for easier readability and statistical calculations. A total of 60 mimicked syringes were analyzed for fentanyl, heroin and xylazine; 20 syringes for each drug. As mentioned before, only eight mimicked syringes for etizolam were prepared due to low drug stock. For etizolam, drug degradation was only tracked up until a 7-day interval. There was an overall count of 68 "real" syringes that were analyzed for this project.

The linear calibration curve established for each drug was used to calculate its concentration. It uses the slope-intercept form, $y = mx + b$ to create a linear curve of known concentrations that plots instrumental signal vs. concentration. **Figure 4** is the calibration curve used for heroin. The *m* term corresponds to the linear term describing the slope of the curve for each drug. This value is constant across measurements of the same drug. The *b* term is associated with each calibration curve to fit the data and it is also constant across measurements of the same drug. The *y* is a parameter that is calculated from equation 1

$$y = \frac{Response\ Analyte}{Response\ Internal\ Standard\ (ISTD)} \quad \text{(Eq. 1)}$$

where the *Response Analyte* is the mass spectral response of the actual drug, and the *Response Internal Standard (ISTD)* is the mass spectral response of the internal standard with the solvent. The ratio between these two values yields the *y* term associated with the calibration curve. Finally,

the *x* term corresponds to the concentration ratio between the analyte and the internal standard, as shown in equation 2.

$$x = \frac{Concentration\ Analyte}{Concentration\ Internal\ Standard\ (ISTD)} \quad \text{(Eq. 2)}$$

Given the definition of the parameters *y*, *m*, *x*, and *b*, the concentration of the analyte, i.e. the drug of interest, can be calculated using equation 3, below.

$$Concentration\ of\ Analyte = \frac{\left(\left(\frac{Response\ Analyte}{Response\ ISTD}\right) - b\right)}{\frac{m}{concentration\ ISTD}} \quad \text{(Eq. 3)}$$

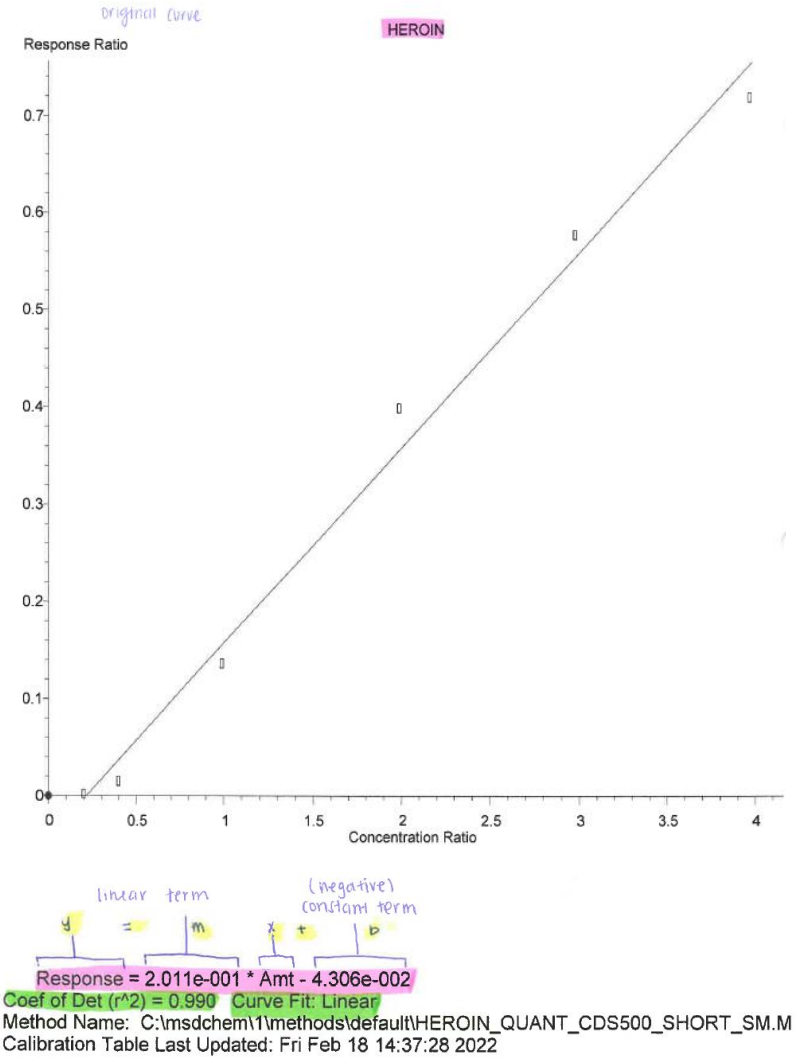

Figure 4. *Calibration curve for quantifying heroin using the y=mx+b formula.*

Results and Discussion: Mimicked Syringes

Table 5 depicts the data collected from each mimicked syringe with time intervals ranging from 0-day to 28-day periods. The table shows the individual variables required to calculate the amount of drug leftover in the syringe (concentration of analyte). **Figure 5** illustrates the data from **Table 5** in a concise and easy to read graph. As shown in this figure, two mimicked syringes were prepared identically for each time interval. Intuitively, a decrease in concentration overtime was

the predicted response in this study. However, alternative responses were not discarded from the predictions as other factors could influence the outcome.

The first result that is worth mentioning is the extraction quantities recovered from the mimicked syringes. With all original syringes prepared with approximately 2 mg/mL solutions, an average of 0.182 mg/mL for fentanyl, 0.050 mg/mL for heroin, 0.042 mg/mL for xylazine, and 0.125 mg/mL for etizolam were extracted, after removing clear outliers. This indicates that the amount extracted corresponds to less than 10% of the original prepared solution, indicating a similar recovery compared to "real" syringes. A probable explanation for the clear outliers in fentanyl data is cross contamination of the prepared syringes resulting in a significantly higher concentration (10x) of drug solution in these syringes.

As indicated in figure 5, the response of drug degradation does not match a decreasing trend. For fentanyl, the shorter time intervals (0-day to 3-day) contained a lower concentration of the drug compared to longer time intervals (7-day and 28-day). These two-time intervals could be considered as the main outliers of a more general trend. However, it is perhaps the case that a higher number of syringes (*n*) are needed to evaluate the distribution of drug concentration resulting from the artificial process of syringe mimicking. In other words, the 16 syringes prepared on 3/1/22 could have drug concentrations that are not evenly distributed. From this distribution it could have been possible that the syringes with the lower concentrations had been picked for the shorter time interval measurements. The human element during preparation of the drugs must be statistically quantified to obtain an accurate distribution of drug concentration prior to drug degradation analysis.

A similar reading from the data of heroin, xylazine and etizolam could be concluded as they also do not follow a decreasing concentration response over time. However, it can be argued

that the lab conditions did not heavily influence the degradation of the drugs in the time range of 28 days included in this study. In fact, if the degradation was indeed predominant under the conditions found in the lab, this would dominate the concentration response and a decreasing trend would have been observed.

Results and Discussion: "Real" Syringes

The benefit of using "real" syringes in this study is that it provided a data set that would corroborate some of the claims mentioned above. In particular, the "real" syringes were useful in determining the contribution of the lab setting to an overall effect to drug degradation. **Table 6** lists the syringe-exchange facilities where the "real" syringes were collected. There are two factors that must be acknowledged when reviewing the data: 1) the degradation timeline started when the syringes were first accessioned at the laboratory and 2) the timeframe of when the IV drug user utilized the syringe is unknown. In addition, the content of the syringes is also unknown prior to collection. Evaluating a consistent number of syringes for every time interval as in the mimicked syringe study was desired. Unfortunately, the number of syringes the laboratory received varied each day, as can be seen in **Table 7**. Given all these limitations, at least three syringes were set aside for each time interval, having five syringes being the normal amount. There was effort to set aside a new set of syringes for the days with no observed target drugs. As shown in **Table 7** there were many syringes that did not have target drugs and instead, qualitative data was provided to explain what was seen, such as the detection of other common CDS. All drugs were identified using drug libraries on mass spectrometer software from Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) and the National Institute of Standards and Technology (NIST).

Another benefit of using "real" syringes is that an evaluation of the fatty acids resulting from physical degradation and the metabolites related to chemical degradation were observed. From the total of 68 "real" syringes, 17 syringes contained fentanyl (**Table 7**). The lowest amount recovered was 0.02230 mg at 5-day interval, and the highest residue was 6.817 mg at 14-day interval, a significant amount as it can be considered a lethal dose for fentanyl. In contrast to our mimicked syringes an average extraction of residual fentanyl was 0.7681 mg/mL, a value considerably higher. From the 17 fentanyl syringes, eight syringes also contained 4-ANPP, a precursor and metabolite of fentanyl. 4-ANPP was noted to elute off the column before the fentanyl peak on the chromatograph. However, it is unknown if 4-ANPP was present in the syringe due to human metabolism or formed during gas chromatography with high injection temperatures. Similarly to the data displayed for heroin in mimicked syringes, the "real" syringes also had no distinct degradation trend. Eight syringes contained heroin with an overall average of 0.0565 mg. At least four of the syringes showed a peak identified as 6-MAM. Heroin is known to have a low-binding affinity and converts to 6-MAM when it encounters blood (Gottås et al., 2013). There was a total of 14 "real" syringes that contained xylazine and the average residue recovered was 0.0206 mg/mL. One noticeable trend was that there was no xylazine detected after the 14-day interval. This may indicate that syringes received at the lab should be processed within two weeks to identify all substances present before they are no longer detectable. Unfortunately, there were no "real" syringes that had residual etizolam, and therefore no degradation data was concluded for this drug.

In addition to identifying and quantifying the target drugs present in each syringe, **Table 7** also notes other substances that were identified in the "real" syringes. There were four other drugs that were commonly seen in combination with the target drugs: methamphetamine (27 total),

cocaine (11 total), caffeine (11 total) and diphenhydramine (nine total). Other noted drugs with less amounts were the following: 1) lidocaine, a local anesthetic, 2) ketamine, a hallucinogen, 3) tramadol, an opiate analgesic, 4) nicotine, a stimulant and depressant, and 5) codeine, a pain-relieving opioid. Data on other CDS is important because it provides a better understanding of how the detection of drugs contributes to drugs trends in Washington D.C. It is interesting to note that there were three "real" syringes that contained more than seven drugs which may indicate that there is a possibility the syringe was used more than once. Creating more calibration curves to include other common drugs can be implemented in future research which would provide detail as to the quantity of drug residue. Fatty acids such as palmitic, myristic, stearic, lauric, elaidic and oleic acid were also found in most, if not all, the "real" syringes that were analyzed. A clear trend exhibited palmitic acid with abundances between $1 \cdot 10^6$-$2 \cdot 10^6$ a.u., which is enough acid to generate a distinguishable peak on the chromatographs. Research highlights that palmitic acid accounts for "20-30% of total fatty acids in the human body" where high concentrations are found in our tissue (Carta et al., 2017). This is relevant to syringes because as the needle penetrates through the skin, the oils may contaminate and interact with the drugs in the barrel creating fatty acid peaks on the chromatograph.

Overall, the conclusion from the data obtained from the "real" syringes is that the FCU lab conditions are not a dominant factor in drug degradation over a 28-day period. As mentioned in the mimicked syringes section, if the conditions in the lab would be aggressive towards the stability of the drug, the response would be dominated by a decreasing trend and high concentrations wouldn't be observed after a 28-day time interval or even longer.

MIMICKED SYRINGES – DEGRADATION DATA

Table 5. *Raw data for each variable used to calculate the concentration of analyte in each syringe using equation 3.*

Drug	Response Analyte	Response ISTD	Conc. ISTD	m	b	Solve for Conc.	Conc.	Average
0-day - Fentanyl A	72415	4091729	0.02551	1.41E-02	-1.17E-02	5.32219E-02	0.0532219	0.0421291
0-day - Fentanyl B	18340	3379593	0.02551	1.41E-02	-1.17E-02	3.10363E-02	0.0310363	
0-day - Heroin A	300724	3304983	0.02551	2.01E-01	-4.31E-02	1.70047E-02	0.0170047	0.01818955
0-day - Heroin B	393703	3589832	0.02551	2.01E-01	-4.31E-02	1.93744E-02	0.0193744	
0-day - Xylazine A	689421	4080701	0.02551	5.31E-01	-3.93E-01	2.69572E-02	0.0269572	0.0843016
0-day - Xylazine B	11984532	4685058	0.02551	5.31E-01	-3.93E-01	1.41646E-01	0.141646	
0-day - Etizolam A	17577	3739949	0.02551	7.69E-02	-1.91E-01	6.47951E-02	0.0647951	0.0742121
0-day - Etizolam B	264657	4305633	0.02551	7.69E-02	-1.91E-01	8.36291E-02	0.0836291	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
1-day - Fentanyl A	206660	4442385	0.02551	1.41E-02	-1.17E-02	1.05331E-01	0.105331	0.0578342
1-day - Fentanyl B	183977	4048999	0.02551	1.41E-02	-1.17E-02	1.03374E-01	0.0103374	
1-day - Heroin A	161011	3492422	0.02551	2.01E-01	-4.31E-02	1.13105E-02	0.0113105	0.03164855
1-day - Heroin B	1280390	3491088	0.02551	2.01E-01	-4.31E-02	5.19866E-02	0.0519866	
1-day - Xylazine A	17386	2666412	0.02551	5.31E-01	-3.93E-01	1.91599E-02	0.0191599	0.02302605
1-day - Xylazine B	517640	3088689	0.02551	5.31E-01	-3.93E-01	2.68922E-02	0.0268922	
1-day - Etizolam A	2296	3173123	0.02551	7.69E-02	-1.91E-01	6.34759E-02	0.0634759	0.09267445
1-day - Etizolam B	624515	3533532	0.02551	7.69E-02	-1.91E-01	1.21873E-01	0.121873	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
3-day - Fentanyl A	16601	2506737	0.02551	1.41E-02	-1.17E-02	3.31984E-02	0.0331984	0.03963235

3-day - Fentanyl B	39372	2865498	0.02551	1.41E-02	-1.17E-02	4.60663E-02	0.0460663	
3-day - Heroin A	209763	3015347	0.02551	2.01E-01	-4.31E-02	1.42868E-02	0.0142868	0.0150268
3-day - Heroin B	219461	2701625	0.02551	2.01E-01	-4.31E-02	1.57668E-02	0.0157668	
3-day - Xylazine A	4702	2593106	0.02551	5.31E-01	-3.93E-01	1.89339E-02	0.0189339	0.01893445
3-day - Xylazine B	5127	2791369	0.02551	5.31E-01	-3.93E-01	1.89350E-02	0.018935	
3-day - Etizolam A	2885	2658812	0.02551	7.69E-02	-1.91E-01	6.35959E-02	0.0635959	0.07008415
3-day - Etizolam B	116024	2886328	0.02551	7.69E-02	-1.91E-01	7.65724E-02	0.0765724	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
7-day - Fentanyl A	11196634	7429841	0.02551	1.41E-02	-1.17E-02	2.74575E+00	2.74575	2.99381
7-day - Fentanyl B	13845149	7772101	0.02551	1.41E-02	-1.17E-02	3.24187E+00	3.24187	
7-day - Heroin A	11041027	7681719	0.02551	2.01E-01	-4.31E-02	1.87789E-01	0.187789	0.16153
7-day - Heroin B	7599427	7426334	0.02551	2.01E-01	-4.31E-02	1.35271E-01	0.135271	
7-day - Xylazine A	98967	8195213	0.02551	5.31E-01	-3.93E-01	1.94266E-02	0.0194266	0.02221025
7-day - Xylazine B	926374	7234556	0.02551	5.31E-01	-3.93E-01	2.49939E-02	0.0249939	
7-day - Etizolam A	2684318	7384473	0.02551	7.69E-02	-1.91E-01	1.83838E-01	0.183838	0.262974
7-day - Etizolam B	7050973	8388443	0.02551	7.69E-02	-1.91E-01	3.42110E-01	0.34211	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
10-day - Fentanyl A	490591	5912435	0.02551	1.41E-02	-1.17E-02	1.71241E-01	0.171241	0.2948555
10-day - Fentanyl B	1344620	6119618	0.02551	1.41E-02	-1.17E-02	4.18470E-01	0.41847	
10-day - Heroin A	1876870	5755142	0.02551	2.01E-01	-4.31E-02	4.68314E-02	0.0468314	0.0517494
10-day - Heroin B	2428220	6015514	0.02551	2.01E-01	-4.31E-02	5.66674E-02	0.0566674	
10-day - Xylazine A	4722	5597999	0.02551	5.31E-01	-3.93E-01	1.88874E-02	0.0188874	0.02354325
10-day - Xylazine B	1162917	5969301	0.02551	5.31E-01	-3.93E-01	2.81991E-02	0.0281991	

Drug	Response Analyte	Response ISTD	Conc. ISTD	m	b	Solve for Conc.	Conc.	Average
14-day - Fentanyl A	978948	7373055	0.02551	1.41E-02	-1.17E-02	2.61272E-01	0.261272	0.223053
14-day - Fentanyl B	685339	7573266	0.02551	1.41E-02	-1.17E-02	1.84834E-01	0.184834	
14-day - Heroin A	1978805	7332981	0.02551	2.01E-01	-4.31E-02	3.96934E-02	0.0396934	0.0401632
14-day - Heroin B	2241009	8082771	0.02551	2.01E-01	-4.31E-02	4.06330E-02	0.040633	
14-day - Xylazine A	744757	7469722	0.02551	5.31E-01	-3.93E-01	2.36332E-02	0.0236332	0.02655095
14-day - Xylazine B	1693827	7655268	0.02551	5.31E-01	-3.93E-01	2.94687E-02	0.0294687	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
17-day - Fentanyl A	1075835	5335068	0.02551	1.41E-02	-1.17E-02	3.85802E-01	0.385802	0.5828165
17-day - Fentanyl B	2342902	5583692	0.02551	1.41E-02	-1.17E-02	7.79831E-01	0.779831	
17-day - Heroin A	2126152	5333864	0.02551	2.01E-01	-4.31E-02	5.60273E-02	0.056027	0.0628415
17-day - Heroin B	2839448	5610988	0.02551	2.01E-01	-4.31E-02	6.96560E-02	0.069656	
17-day - Xylazine A	69049	5213241	0.02551	5.31E-01	-3.93E-01	1.94827E-02	0.019482	0.035339
17-day - Xylazine B	3870839	5744233	0.02551	5.31E-01	-3.93E-01	5.11960E-02	0.051196	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
21-day - Fentanyl A	556441	6123047	0.02551	1.41E-02	-1.17E-02	1.85524E-01	0.185524	0.1014955
21-day - Fentanyl B	543742	6406539	0.02551	1.41E-02	-1.17E-02	1.74670E-01	0.017467	
21-day - Heroin A	1064169	5998779	0.02551	2.01E-01	-4.31E-02	2.79656E-02	0.0279656	0.0253414
21-day - Heroin B	865545	6363184	0.02551	2.01E-01	-4.31E-02	2.27172E-02	0.0227172	
21-day - Xylazine A	9422426	6250711	0.02551	5.31E-01	-3.93E-01	9.12108E-02	0.0912108	0.0552119
21-day - Xylazine B	49440	6482209	0.02551	5.31E-01	-3.93E-01	1.92130E-02	0.019213	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
24-day - Fentanyl A	10433	6186042	0.02551	1.41E-02	-1.17E-02	2.42743E-02	0.0242743	0.11453115
24-day - Fentanyl B	338781	3336699	0.02551	1.41E-02	-1.17E-02	2.04788E-01	0.204788	

24-day - Heroin A	362951	5884874	0.02551	2.01E-01	-4.31E-02	1.32859E-02	0.0132859	0.0238114
24-day - Heroin B	1582730	6953268	0.02551	2.01E-01	-4.31E-02	3.43369E-02	0.0343369	
24-day - Xylazine A	2440	6502063	0.02551	5.31E-01	-3.93E-01	1.88649E-02	0.0188649	0.01943345
24-day - Xylazine B	165537	6879257	0.02551	5.31E-01	-3.93E-01	2.00020E-02	0.020002	
Drug	**Response Analyte**	**Response ISTD**	**Conc. ISTD**	**m**	**b**	**Solve for Conc.**	**Conc.**	**Average**
28-day - Fentanyl A	12537026	9489937	0.02551	1.41E-02	-1.17E-02	2.40967E+00	2.40967	1.938295
28-day - Fentanyl B	7866191	9837168	0.02551	1.41E-02	-1.17E-02	1.46692E+00	1.46692	
28-day - Heroin A	4639734	9660216	0.02551	2.01E-01	-4.31E-02	6.63885E-02	0.066388	0.07173435
28-day - Heroin B	4195363	7430928	0.02551	2.01E-01	-4.31E-02	7.70807E-02	0.0770807	
28-day - Xylazine A	25346007	7154073	0.02551	5.31E-01	-3.93E-01	1.88924E-01	0.188924	0.11472625
28-day - Xylazine B	4885244	10816412	0.02551	5.31E-01	-3.93E-01	4.05285E-02	0.0405285	

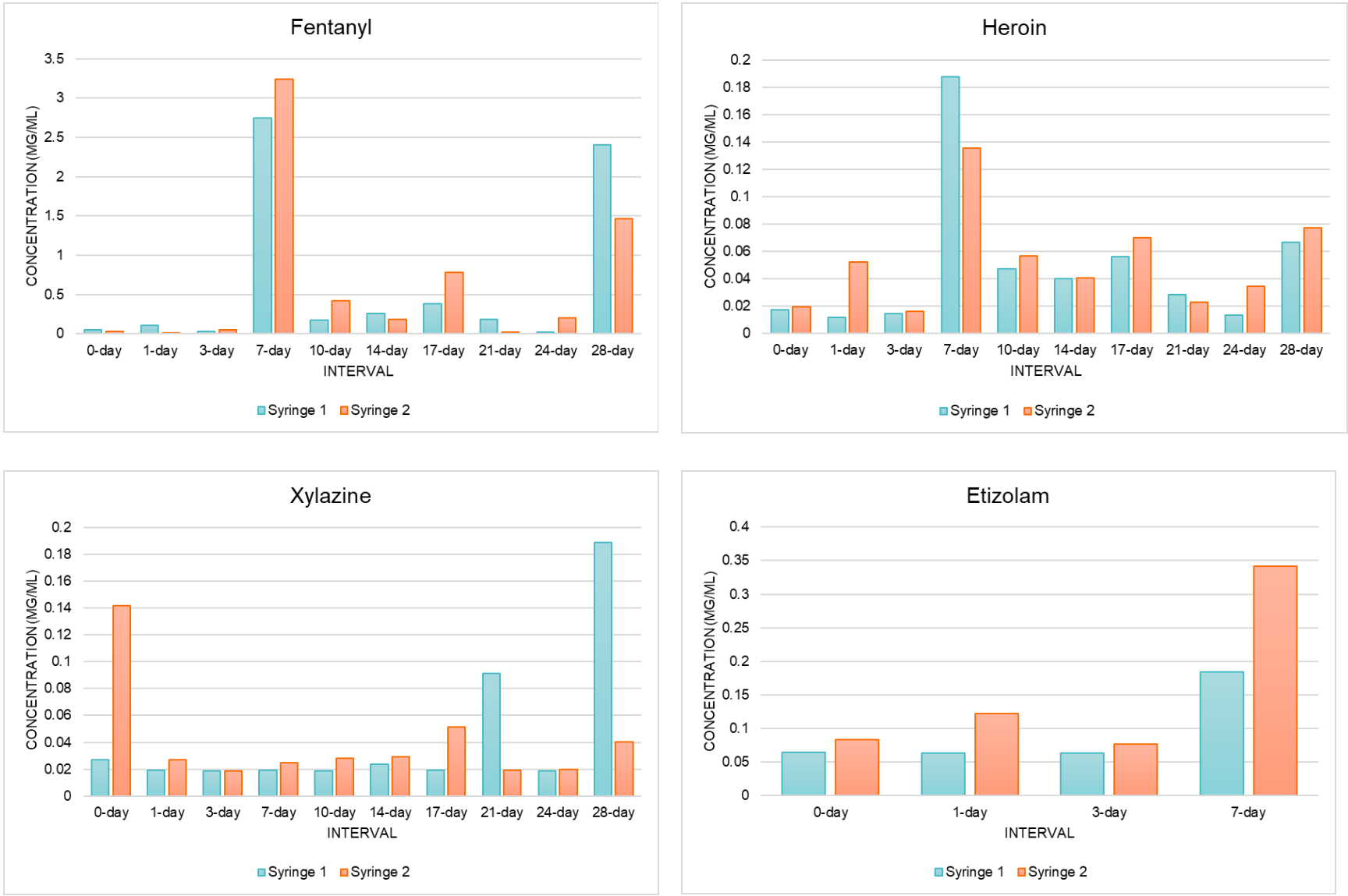

Figure 5. *Graphs containing data collected from two mimicked syringes prepared identically. As seen in the timeline response, the data suggest that the drug degradation does not follow a clear trend.*

REAL SYRINGES – DEGRADATION DATA

Table 6. *List of syringe-exchange facilities where the "real" syringes were collected.*

Facilities		
HIPS	13 total syringes	Honoring Individual Power & Strength
FMCS	20 total syringes	Family & Medical Counseling Services
BFTC	25 total syringes	Bread For The City
UHU	10 total syringes	Us Helping Us, People Into Living Inc.

Table 7. *List of detected substances for each syringe. Raw data was collected if a target drug was present and used in equation 3 to calculate the final concentration.*

Syringes	Substances Detected	Response Analyte	Drug	Conc. (mg)
1st Set				
Syringe 1 - 0-day	xylazine, cocaine		Xylazine	0.020708
Syringe 2 - 0-day	xylazine, diphenhydramine		Xylazine	0.0200265
Syringe 3 - 0-day	xylazine, heroin, caffeine, cocaine, diphenhydramine		Xylazine	0.023901
			Heroin	0.0066784
Syringe 4 - 0-day	xylazine, caffeine		Xylazine	0.030365
Syringe 5 - 0-day	xylazine		Xylazine	0.019598
2nd Set				
Syringe 1 - 0-day	methamphetamine	no target drug detected		
Syringe 2 - 0-day	caffeine	no target drug detected		
Syringe 3 - 0-day	caffeine, fentanyl		Fentanyl	0.0830403
Syringe 4 - 0-day	procaine, fentanyl, 4-ANPP		Fentanyl	0.221528
Syringe 5 - 0-day	6-MAM		Heroin	0.0055161
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
1st Set				
Syringe 1 - 1-day	methamphetamine	no target drug detected		
Syringe 2 - 1-day	methamphetamine, ketamine	no target drug detected		
Syringe 3 - 1-day	methamphetamine	no target drug detected		
Syringe 4 - 1-day	methamphetamine	no target drug detected		
Syringe 5 - 1-day	methamphetamine	no target drug detected		

2nd Set				
Syringe 1 - 1-day	methamphetamine	no target drug detected		
Syringe 2 - 1-day	methamphetamine	no target drug detected		
Syringe 3 - 1-day	methamphetamine	no target drug detected		
Syringe 4 - 1-day	methamphetamine	no target drug detected		
Syringe 5 - 1-day	methamphetamine	no target drug detected		
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
Syringe 1 - 3-day	methamphetamine, caffeine	no target drug detected		
Syringe 2 - 3-day	no controlled substances	no target drug detected		
Syringe 3 - 3-day	4-ANPP, 6-MAM, heroin, fentanyl		Heroin	0.010215
			Fentanyl	0.245205
Syringe 4 - 3-day	xylazine, fentanyl		Xylazine	0.0272404
			Fentanyl	0.0361268
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
Syringe 1 - 5-day	methamphetamine	no target drug detected		
Syringe 2 - 5-day	caffeine, fentanyl		Fentanyl	0.130761
Syringe 3 - 5-day	adrafinil, caffeine, 4-ANPP, fentanyl		Fentanyl	0.0250146
Syringe 4 - 5-day	methamphetamine	no target drug detected		
Syringe 5 - 5-day	methamphetamine	no target drug detected		
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
1st Set				
Syringe 1 - 7-day	Eutylone, cocaine	no target drug detected		
Syringe 2 - 7-day	no controlled substances	no target drug detected		
Syringe 3 - 7-day	anhydroecgonine methyl ester,ecgonine methyl ester, ecgonidine, lidocaine, tramadol, cocaine, benzoyl ecgonine	no target drug detected		
2nd Set				
Syringe 1 - 7-day	anhydroecgonine methyl ester,ecgonine methyl ester, diphenhydramine, xylazine, cocaine, 4-ANPP, fentanyl		Xylazine	0.02602
			Fentanyl	0.04154
Syringe 2 - 7-day	caffeine, diphenhydramine, xylazine, cocaine, fentanyl		Xylazine	0.04386
			Fentanyl	0.03171
Syringe 3 - 7-day	diphenhydramine, cocaine	no target drug detected		

Syringe 4 - 7-day	methamphetamine, anhydroecgonine methyl ester, ecgonine methyl ester, xylazine, cocaine, fentanyl		Xylazine	0.03845
			Fentanyl	0.03279
Syringe 5 - 7-day	acetaminophen, caffeine, diphenhydramine, xylazine, cocaine, 4-ANPP, fentanyl, heroin		Xylazine	0.02474
			Fentanyl	0.1171
			Heroin	0.03242
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
1st Set				
Syringe 1 - 10-day	no controlled substances	no target drug detected		
Syringe 2 - 10-day	caffeine, diphenhydramine, xylazine		Xylazine	0.0240609
Syringe 3 - 10-day	diphenhydramine, xylazine		Xylazine	0.021467
Syringe 4 - 10-day	diphenhydramine, xylazine		Xylazine	0.0191517
Syringe 5 - 10-day	no controlled substances	no target drug detected		
2nd Set				
Syringe 1 - 10-day	methamphetamine	no target drug detected		
Syringe 2 - 10-day	methamphetamine	no target drug detected		
Syringe 3 - 10-day	methamphetamine	no target drug detected		
Syringe 4 - 10-day	methamphetamine	no target drug detected		
Syringe 5 - 10-day	fentanyl		Fentanyl	0.0223
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
Syringe 1 - 14-day	methamphetamine	no target drug detected		
Syringe 2 - 14-day	heroin, fentanyl		Fentanyl	0.076283
			Heroin	0.00631207
Syringe 3 - 14-day	methamphetamine, papaverine	no target drug detected		
Syringe 4 - 14-day	nicotine, diphenhydramine, caffeine, lidocaine, tramadol, cocaine, codeine, 4-ANPP, cholesterol, xylazine, heroin, fentanyl		Xylazine	0.0193912
			Fentanyl	6.81725
			Heroin	0.151651
Syringe 5 - 14-day	methamphetamine	no target drug detected		
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
Syringe 1 - 17-day	fentanyl		Fentanyl	0.038655
Syringe 2 - 17-day	no controlled substances	no target drug detected		

Syringe 3 - 17-day	no controlled substances	no target drug detected		
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
Syringe 1 - 21-day	methamphetamine	no target drug detected		
Syringe 2 - 21-day	no controlled substances	no target drug detected		
Syringe 3 - 21-day	Lidocaine, cocaine	no target drug detected		
Syringe 4 - 21-day	no controlled substances	no target drug detected		
Syringe 5 - 21-day	methamphetamine, cocaine, fentanyl, 4-ANPP		Fentanyl	0.031713
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
Syringe 1 - 25-day	Diacetylcodeine, 6-MAM, heroin		Heroin	0.20115
Syringe 2 - 25-day	methamphetamine, benzophenone, flavone	no target drug detected		
Syringe 3 - 25-day	cetene	no target drug detected		
Syringe 4 - 25-day	methamphetamine	no target drug detected		
Syringe 5 - 25-day	1-octadecene, fentanyl		Fentanyl	0.150115
Syringes	**Substances Detected**	**Response Analyte**	**Drug**	**Conc. (mg)**
Syringe 1 - 28-day	nicotine, 6-MAM, acetaminophen, caffeine, diphenhydramine, lidocaine, tramadol, cocaine, 4-ANPP, codeine, acetylcodeine, heroin, fentanyl		Heroin	0.0381419
			Fentanyl	5.72396
Syringe 2 - 28-day	cocaine, benzoyl ecogonine	no target drug detected		
Syringe 3 - 28-day	5-octadecene	no target drug detected		

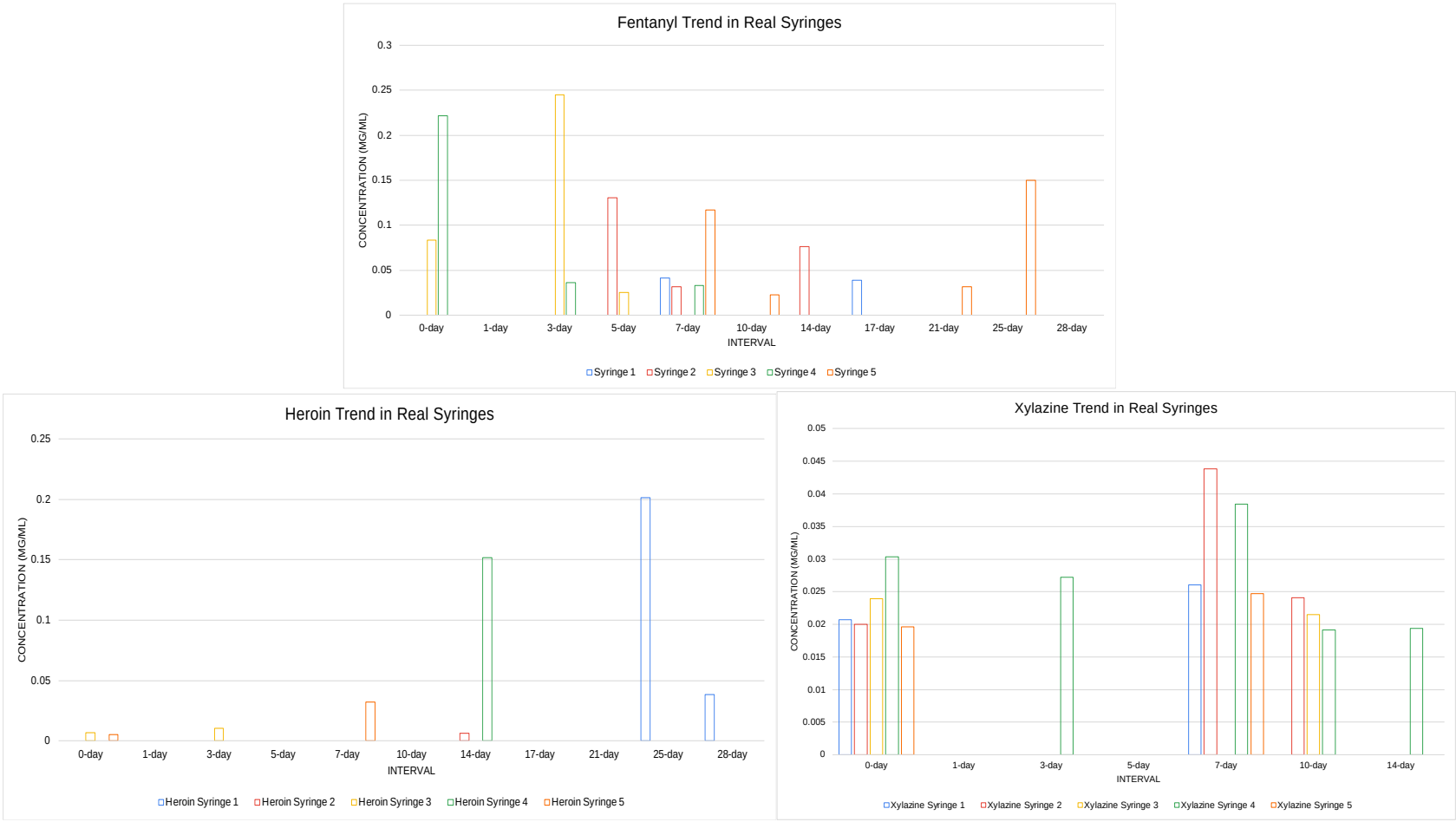

Figure 6. *Graphs indicating data collected for "real" syringes. Fentanyl trend is shown without the highest concentrations in Day 14 with 6.817 mg and 28-day with 5.724 mg for a better representation of the average residue (TOP). Heroin trend includes data from all days (BOTTOM LEFT). No decreasing trend is detectable in heroin. Xylazine trends includes data up to 14-day since there was no detection after this day (BOTTOM RIGHT). This may indicate that the syringes should be processed within two weeks upon arrival to the lab in order to detect xylazine and other possible fast degrading substances.*

CONCLUSION

In this study, we investigated the drug degradation trends in used IV drug user syringes over a 28-day time period. Before conducting the method and collecting the data, the predicted response was a clear decreasing trend of drug concentration extracted over time due to physical and chemical degradation. However, as discussed previously in this paper, the data collected in this study supports the conclusion that the environmental conditions in our lab do not substantially contribute to a noticeable drug degradation over the course of a 28-day period. That being said, it is important to note that a higher number of (mimicked) syringes would provide a more statistically rich study. The main reason the amount of overall processed syringes was not higher is because of funding and timeline limitation. Nevertheless, the findings in this research can serve as the foundation of future studies on drug degradation in syringes obtained from IV drug users in Washington D.C.

The main takeaways from this research are: 1) the method used to mimic IV drug user syringes yielded a comparable drug residue extraction to “real” syringes, and 2) normal laboratory environmental conditions did not contribute to drug degradation trends in polypropylene syringes.

The current operational time frame involved in the processing of syringes in laboratories like FCU is approximately 1-4 business days. Therefore, the findings in this paper suggest that these procedures are risk-free of significant drug degradation on fentanyl and heroin. For xylazine and etizolam, further research could help improve the current understanding as the data in this study was lower than for the other two drugs and our analysis suggests a larger degradation of these drugs.

All in all, with the amount of data collected in this study and the analysis performed and disseminated in this paper, the findings will help continue the efforts of monitoring drug trends in Washington D.C. and understand how drugs degrade in syringes.

REFERENCES

1. Evans, A., Krause, M., Leach, S., Levitas, M., Nguyen, L., & Short, L. C. (2021). Analysis of drug residue in needle-exchange syringes in Washington, D.C. *Forensic Science International*, *329*, 111083. https://doi.org/10.1016/j.forsciint.2021.111083
2. Ekoja, Ali. (2017). DEGRADATION PATHWAY OF PHARMACEUTICAL DOSAGE FORMS. 10.13140/RG.2.2.11814.73281.
3. Snape, T. J., Astles, A., & Davies, J. (2010, October 9). *Understanding the chemical basis of drug stability and degradation*. The Pharmaceutical Journal. https://pharmaceutical-journal.com/article/ld/understanding-the-chemical-basis-of-drug-stability-and-degradation
4. Hung, C. T., Young, M., & Gupta, P. K. (1988). Stability of morphine solutions in plastic syringes determined by reversed-phase ion-pair liquid chromatography. *Journal of Pharmaceutical Sciences*, *77*(8), 719–723. https://doi.org/10.1002/jps.2600770816
5. *Total Ion Chromatogram (TIC)*. (2022). Www.shimadzu.com. https://www.shimadzu.com/an/service-support/technical-support/analysis-basics/gcms/fundamentals/retention/total_ion.html
6. Drug Enforcement Administration. (2008). Control of Immediate Precursor Used in the Illicit Manufacture of Fentanyl as a Schedule II Controlled Substance [Review of *Control of Immediate Precursor Used in the Illicit Manufacture of Fentanyl as a Schedule II Controlled Substance*]. *Federal Register*, *73*(69), 19175–19179. federalregister.gov. https://www.federalregister.gov/documents/2008/04/09/E8-7391/control-of-immediate-precursor-used-in-the-illicit-manufacture-of-fentanyl-as-a-schedule-ii

7. U.S. Drug Enforcement Administration DEA. (2021). DEA PRB # 2021-15. United States. Retrieved from the Department of Justice. https://www.deadiversion.usdoj.gov/drug_chem_info/Xylazine.pdf
8. U.S. Drug Enforcement Administration DEA (2020). DEA PRB 03-24-20-11. United States. Retrieved from the Department of Justice. https://www.deadiversion.usdoj.gov/drug_chem_info/etizolam.pdf
9. Cheng, Y. Y., & Yu, J. Z. (2020). Minimizing Contamination from Plastic Labware in the Quantification of C16 and C18 Fatty Acids in Filter Samples of Atmospheric Particulate Matter and Their Utility in Apportioning Cooking Source Contribution to Urban PM2.5. *Atmosphere*, *11*(10), 1120. https://doi.org/10.3390/atmos11101120
10. Hung, C. T., Young, M., & Gupta, P. K. (1988). Stability of morphine solutions in plastic syringes determined by reversed-phase ion-pair liquid chromatography. *Journal of Pharmaceutical Sciences*, *77*(8), 719–723. https://doi.org/10.1002/jps.2600770816
11. Concheiro, M., Chesser, R., Pardi, J., & Cooper, G. (2018). Postmortem toxicology of new synthetic opioids. *Frontiers in Pharmacology*, *9*. https://doi.org/10.3389/fphar.2018.01210
12. Dinis-Oliveira, R. J. (2019). Metabolism and metabolomics of opiates: A long way of forensic implications to unravel. *Journal of Forensic and Legal Medicine,* 61, 128–140. https://doi.org/10.1016/j.jflm.2018.12.005
13. EMEA. (2002). Committee For Veterinary Medicinal Products - Xylazine Hydrochloride [Review of *Committee For Veterinary Medicinal Products - Xylazine Hydrochloride*]. *The European Agency for the Evaluation of Medicinal Products Veterinary Medicinal Evaluation Unit*. https://www.ema.europa.eu/en/documents/mrl-

report/xylazine-hydrochloride-summary-report-1-committee-veterinary-medicinal-products_en.pdf

14. Gottås, A., Øiestad, E. L., Boix, F., Vindenes, V., Ripel, Å., Thaulow, C. H., & Mørland, J. (2013). Levels of heroin and its metabolites in blood and brain extracellular fluid after i.v. heroin administration to freely moving rats. *British Journal of Pharmacology*, *170*(3), 546–556. https://doi.org/10.1111/bph.12305
15. Carta, G., Murru, E., Banni, S., & Manca, C. (2017). Palmitic acid: Physiological role, metabolism and nutritional implications. *Frontiers in Physiology*, *8*. https://doi.org/10.3389/fphys.2017.00902
16. Lee, Tet Woo, et al. "Chemicals Eluting from Disposable Plastic Syringes and Syringe Filters Alter Neurite Growth, Axogenesis and the Microtubule Cytoskeleton in Cultured Hippocampal Neurons." *Journal of Neurochemistry*, vol. 133, no. 1, 2015, pp. 53–65., https://doi.org/10.1111/jnc.13009.
17. Song, Hong, and Theresa S. Chen. "P-Aminophenol-Induced Liver Toxicity: Tentative Evidence of a Role for Acetaminophen." *Journal of Biochemical and Molecular Toxicology*, vol. 15, no. 1, 2001, pp. 34–40., https://doi.org/10.1002/1099-0461(2001)15:1<34::aid-jbt4>3.0.co;2-u.
18. Ciccarone D. (2013). Saying goodbye to high-dead-space syringes. *The International journal on drug policy*, 24(1), 15–16. https://doi.org/10.1016/j.drugpo.2012.09.011