Downstream Impacts of Data Collected from Drug Checking and Harm Reduction Initiatives

Exploring the Intersection of Forensic Testing and Public Health Collaboration for Overdose Prevention
APHL Webinar Series Part 2: Harm Reduction Initiatives – Wednesday August 2, 2023 – 1:00 to 2:30 PM

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DISCLOSURES

- I have no conflicts of interest to disclose.
- I am a scientist/employee of FRFF/CFSRE, a 501(c)(3) non-profit research & educational facility.
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  - The opinions, findings, conclusions and/or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect those of the CDC, the DOJ, or other federal, state, local, or private agencies.

WHAT IS DRUG CHECKING?

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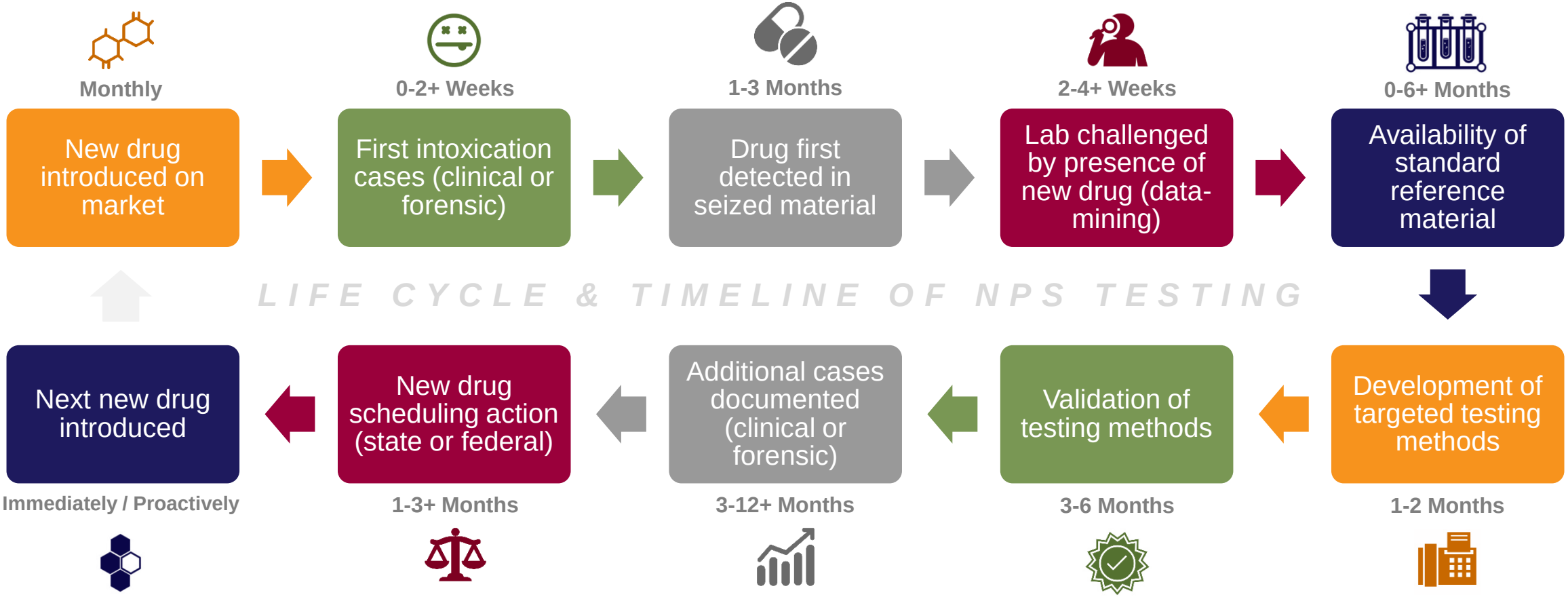
INTRODUCTION TO HARM REDUCTION

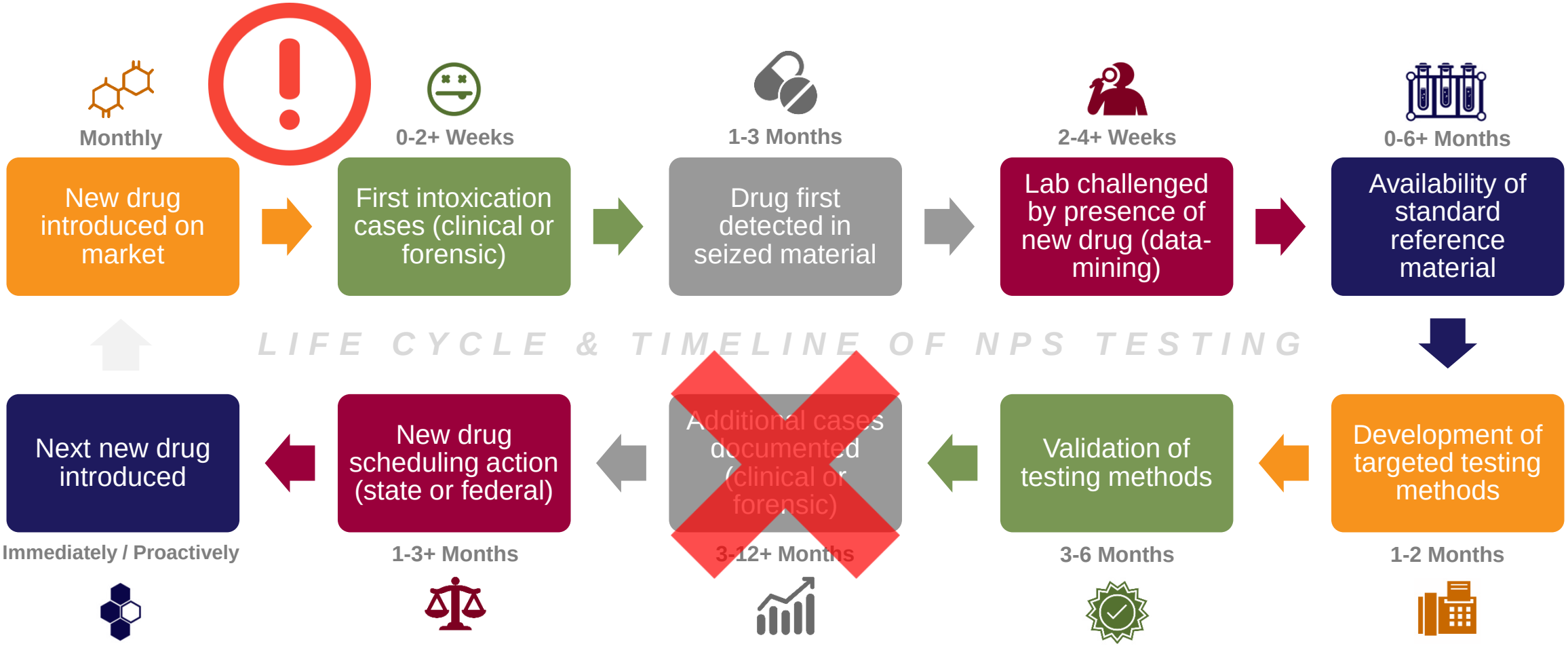
- **Public Health and Safety Strategies:**
  - Supply Reduction
  - Demand Reduction
  - **Harm Reduction**

- **Examples of Harm Reduction:**
  - Naloxone distribution
  - Needle exchange programs
  - **Drug checking***
  - Safe supply

LIFE CYCLE OF A DRUG



LIFE CYCLE OF A DRUG



TYPES OF DRUG CHECKING

THE OLD SAYING → QUICK, CHEAP, OR ACCURATE – PICK TWO

Types of Testing	Examples	Time	$$$	Accuracy	Ease of Use
"Field" Testing	Test Strips, Reagents	**5 mins**	**Low Cost**	**Low Accuracy**	**Easy**
Point-of-Use	FTIR, Raman, MS*	**5-10 mins**	**Mid Cost**	**Mid Accuracy**	**Medium**
Lab – Qual	GC-MS, LC-MS	**20+ mins**	**High Cost**	**High Accuracy**	**Hard**
Lab – Quant	GC-MS, LC-MS	**20+ mins**	**Higher Cost**	**High Accuracy**	**Extra Hard**

THE CFSRE & NPS DISCOVERY

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THE CFSRE & OUR LAB

- The Center for Forensic Science Research and Education (CFSRE)
  - 501(c)(3) non-profit research and educational facility
  - Home to *NPS Discovery* and other programs

Waters Xevo® G2-S LC-QTOF-MS

Sciex X500R LC-TOF-MS

Sciex TripleTOF® 5600+ LC-TOF-MS

Agilent 6495 LC-QQQ-MS

Agilent 6430 LC-QQQ-MS

Waters TQS LC-QQQ-MS

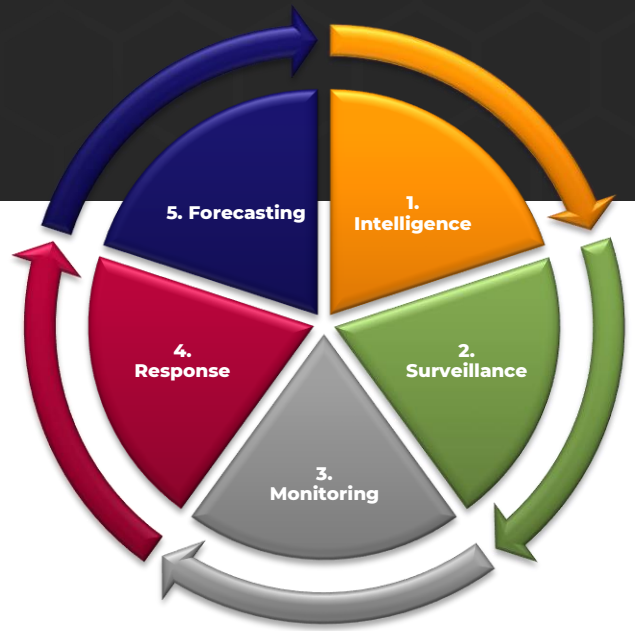
Waters TQD LC-QQQ-MS

Agilent 5975 GC-MS

Agilent 5975 GC-MS

NPS DISCOVERY – THE CFSRE'S EWS

- Open-access drug early warning system (EWS)
  - Combine aspects of research & authentic cases
  - Analyze samples and generate data in-house
  - Develop a panel of high impact reports
  - Disseminate results and reports widely to stakeholders



2016

U-47700 & Furanylfentanyl

Deadly outbreak investigation involving 20+ cases centralized in Midwestern states.

2017

***N*-Ethyl Pentylone**

Investigation of deaths and impaired driving cases involving new stimulant drug linked to Ecstasy and Molly use.

2018

Cyclopropylfentanyl & Methoxyacetylfentanyl

Postmortem investigations involving new fentanyl analogues linked to 40+ deaths in Midwestern states and Florida.

2019

Isotonitazene

First outbreak investigation in the U.S. involving 20+ deaths, primarily from Indiana and Illinois.

2020

MDMB-4en-PINACA

Investigation of 50+ cases involving deaths and hospitalizations from states in the South, Midwest, and Northeast.

2021

Metonitazene & Nitazene Analogues

Continued monitoring and investigations of 40+ deaths involving new nitazene analogues emerging in various regions across the U.S.

2022

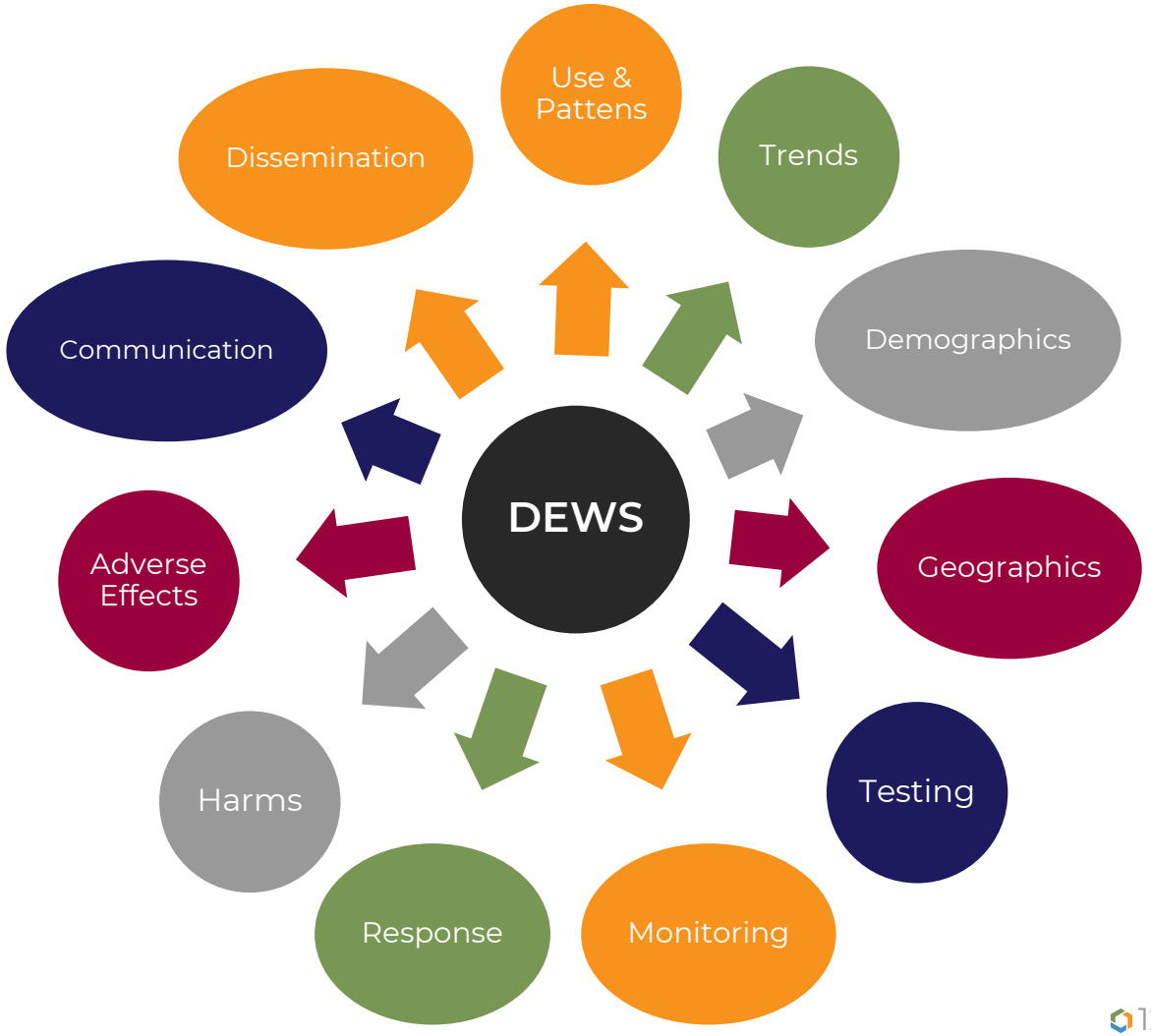
***N,N*-Dimethylpentylone**

First outbreak investigations involving a new stimulant drug linked to 50+ cases, including deaths, primarily originating from Florida and Northeastern states.

DRUG EARLY WARNING SYSTEM (DEWS)

- Four core components of an EWS:
  - **risk knowledge** → understanding drugs, drug use, drug use patterns, drug trends, demographics, geographics, etc.
  - **monitoring** → primarily analytical drug testing but can encompass other data collection techniques
  - **response** → series of calculated actions to reduce drug use harms and adverse effects (many approaches)
  - warning **communication** and actionable outcomes → reporting, dissemination, etc.

- … to **reduce harm** or loss
  - Primary goal for both public health and safety partners and stakeholders



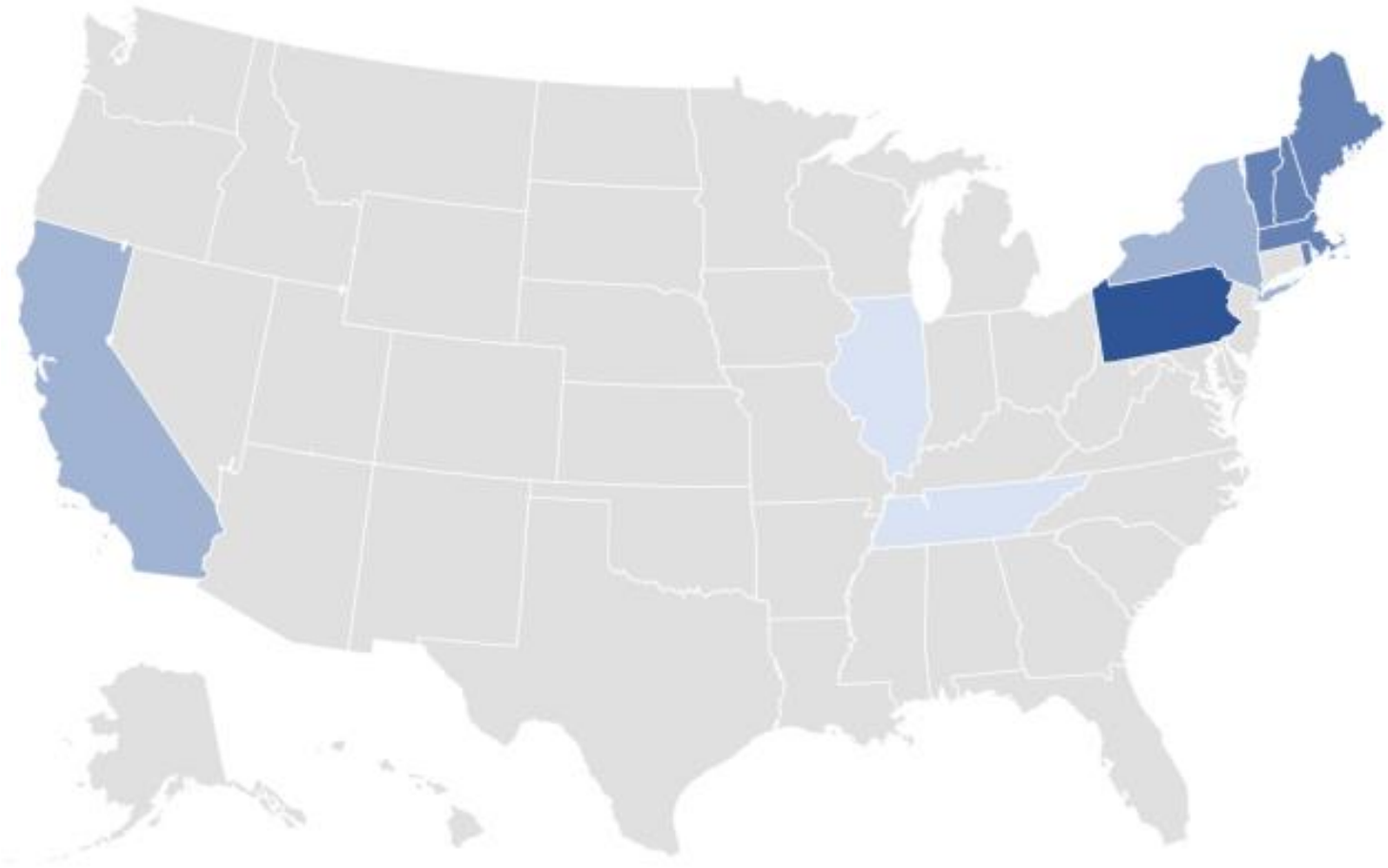
EXAMPLES OF SAMPLE "POPULATIONS"

- **Important → Right populations paired with good intelligence**
- **Toxicology Specimens:**
  - Collaborations with medical examiner and coroner offices, other toxicology labs, clinical partners, and other
  - Example: Initial toxicology testing negative but "suspected overdose"
- **Drug Materials:**
  - Collaborations with crime labs, law enforcement agencies, **public health partners**, and others
  - Routine analysis vs. chemical characterization (structural elucidation)
- **Intelligence & Surveillance:**
  - Monitor online surface web gray market sites, drug use forums, etc.
  - Some correlation between sites and drug markets but delayed

COLLABORATING DRUG CHECKING PROGRAMS

- Philadelphia, PA
- New England
- Providence, RI
- New York City, NY
- San Francisco, CA

- *Pending Developments*



COLLABORATING DRUG CHECKING PROGRAMS

Location	Test Strips	FTIR	Lab Confirmation	Program Details
Philadelphia, PA	Yes	Sometimes	Yes	City-Wide
New England	Yes	Yes	Yes	Community-Based
Providence, RI	Yes	N/A	Yes	Clinical Aspects
New York City, NY	Yes	Yes	Yes	OPS
San Francisco, CA	Yes	Yes	Yes	Mobile Van / Site

DRUG CHECKING SURVEILLANCE

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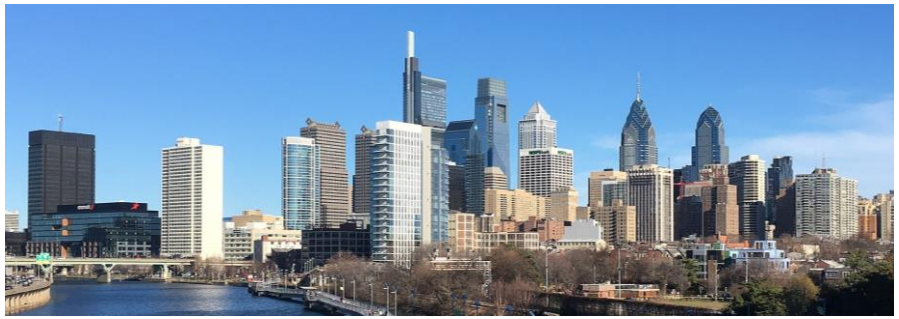
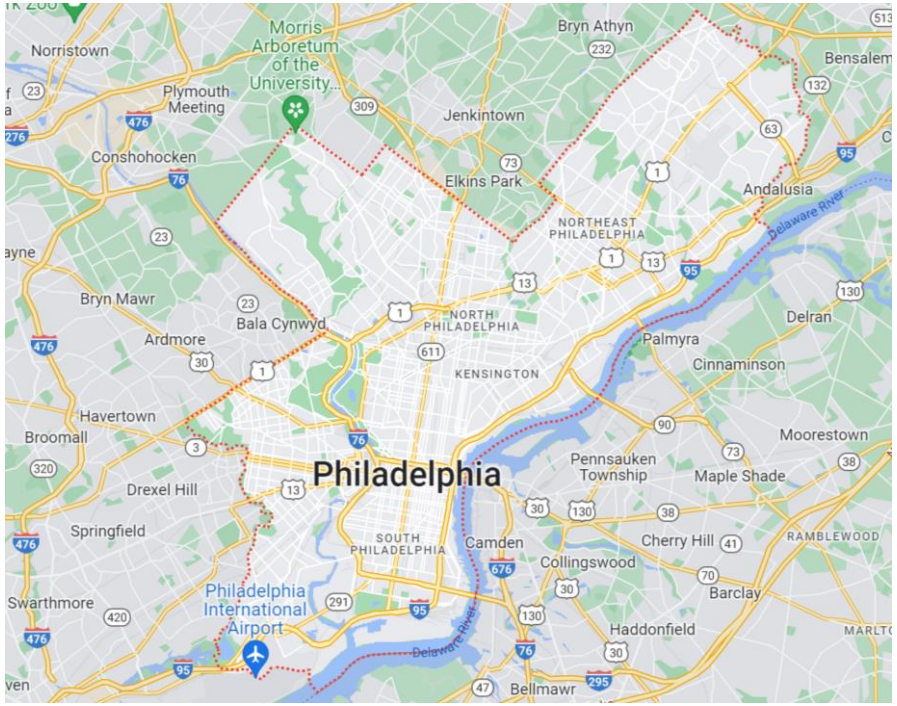
PHILADELPHIA & ITS DRUG SUPPLY

- Nestled in the center of the larger mid-Atlantic metropolitan region ("Northeast Corridor")
  - 6th largest city by population and 7th largest metro area
- **"Open air drug market"** (Kensington neighborhood)
- Drug markets → dope, crack/coke, meth, K2, etc.
- Continually changing and diverse drug environment
- Collaboration between the **CFSRE** and the **Philadelphia Department of Public Health (PDPH)**



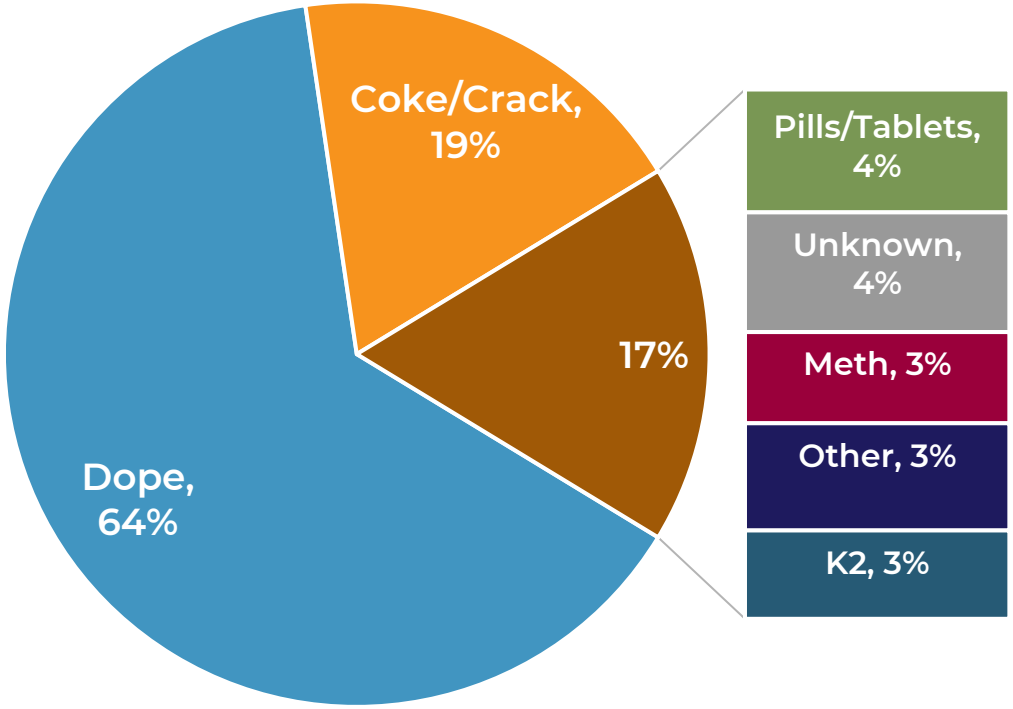


PDPH & CFSRE DRUG CHECKING COLLABORATION

- **2020** → Partnership formally launched

- **Sample Analyzed**
  - 1,000+ samples received since 2020
  - Variety of sample types (suspected contents) →
  - Paired FTIR and test strip results***

- **Key Findings**
  - "Dope": >99% contain fentanyl and >90% contain xylazine
  - Methamphetamine – rarely adulterated or substituted
  - Cocaine – "coke" samples sometimes test positive for trace fentanyl
  - K2 – revolving door of synthetic cannabinoids

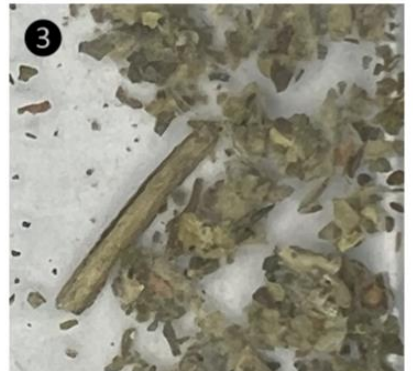


DRUG CHECKING RESULTS

			
Clonazolam	Etizolam	Etizolam	Desalkylflurazepam
		 MDA	
Fentanyl, Gabapentin	*para*-Fluorofentanyl, Gabapentin	Methamphetamine	
			
Cocaine, Lidocaine	Cocaine, Lidocaine	Methamphetamine, Caffeine	Methamphetamine

Methamphetamine

Methamphetamine



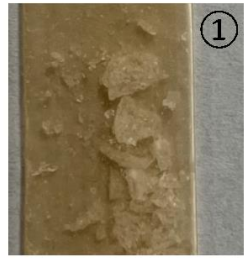
ADB-5'Br-IANCA

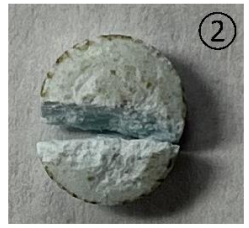
ADB-5'Br-IANCA

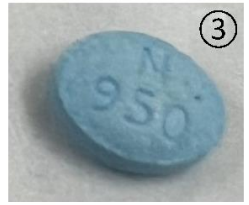
	
ADB-BINACA	ADB-BINACA

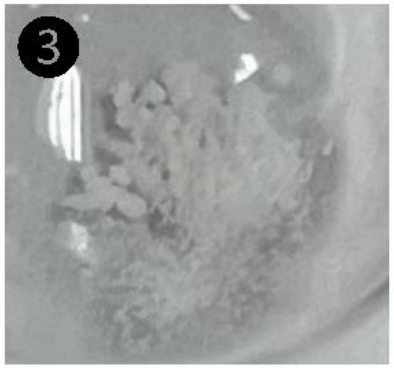
Date	Suspected	Drugs Identified
3/1/2022	Coke	Cocaine (1p), Lidocaine (0.3p), Levamisole (trace)
3/15/2022	Coke	Cocaine (1p), Fentanyl (0.1p), Lidocaine (0.3p), Xylazine (trace) → ①
3/15/2022	Coke	Cocaine (1p), Fentanyl (0.08p), Lidocaine (1.1p), Xylazine (0.1p) ← ❶
3/24/2022	Coke	Cocaine (1p), Lidocaine (0.9p), Dimethylsulfone (0.3p) ← ❷
3/31/2022	Coke	Cocaine (1p), Dimethylsulfone (>10p), Lidocaine (0.7p) → ②
3/31/2022	Coke	Cocaine (1p), Lidocaine (0.9p) ← ❸
3/31/2022	Coke	Cocaine (1p), Phenacetin (0.6p), Levamisole (0.3p)
3/1/2022	Crack	Cocaine → ③
3/15/2022	Crack	Cocaine
3/15/2022	Crack	Cocaine (1p), Lidocaine (0.4p) ← ❹
3/15/2022	Crack	Cocaine (1p), Lidocaine (0.9p), Dimethylsulfone (0.3p)
3/15/2022	Crack	Cocaine (1p), Levamisole (0.5p), Lidocaine (trace)
3/24/2022	Crack	Cocaine
3/24/2022	Crack	Cocaine, Levamisole (trace)
3/24/2022	Crack	Cocaine

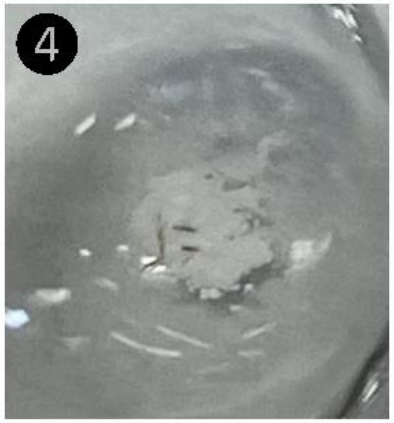
Date	Suspected	Drugs Identified
3/15/2022	Meth	Methamphetamine → ①
3/24/2022	Meth	Methamphetamine
3/31/2022	Meth	Methamphetamine
4/14/2022	Meth	Methamphetamine
3/1/2022	Methadone	Methadone
3/15/2022	Oxycodone	Fentanyl (1p), Acetaminophen (1.5p), Lidocaine (2p), Tramadol (1.3p), Xylazine (1p) *[Fentanyl Byproducts]* ← ❶
3/15/2022	Oxycodone	Fentanyl (1p), Acetaminophen (>10p) *[Fentanyl Byproducts]* → ②
3/24/2022	Adderall	Amphetamine → ③
3/15/2022	Weed	THC and Cannabinoids
3/15/2022	PCP	Tenocyclidine (1p), Nicotine (0.2p) ← ❷
3/31/2022	PCP	Tenocyclidine (1p), Nicotine (trace)



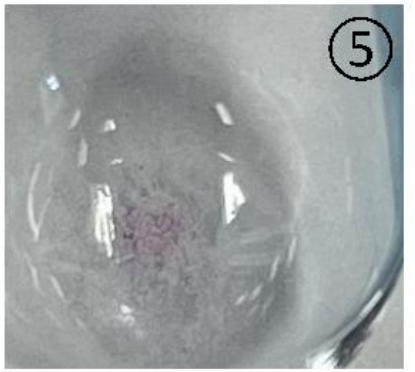








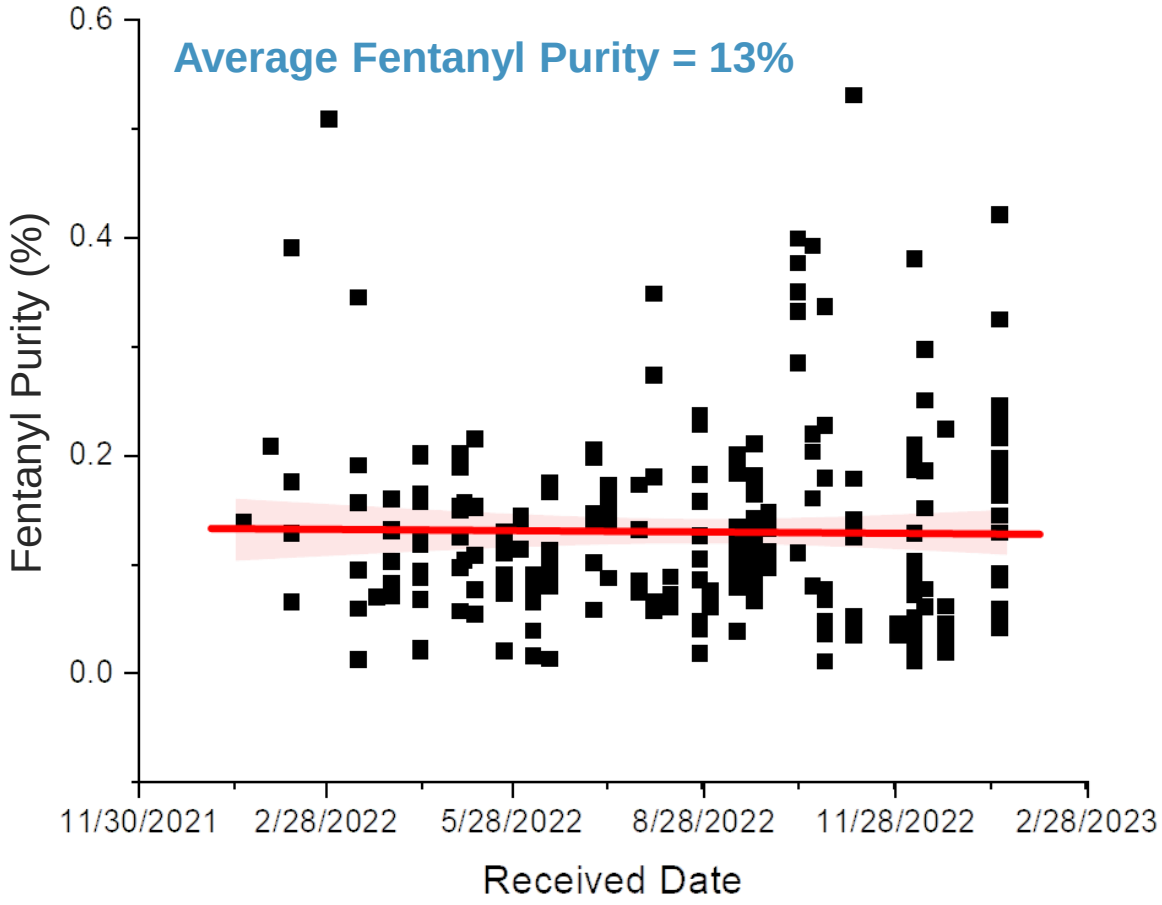
Date	Suspected	Drugs Identified
3/1/2022	Dope	Fentanyl (1p), Heroin (1p), Xylazine (0.3p) *[Fentanyl and Heroin Byproducts]*
3/1/2022	Dope	Fentanyl (1p), Caffeine (0.2p), Xylazine (trace) *[Fentanyl Byproducts]* → ④
3/1/2022	Dope	Fentanyl (1p), Xylazine (1p) *[Fentanyl Byproducts]* ← ❸
3/1/2022	Dope	*para*-Fluorofentanyl (1p), Fentanyl (0.7p), Xylazine (16p) *[Fentanyl and para-Fluorofentanyl Byproducts]*
3/1/2022	Dope	Fentanyl (1p), *para*-Fluorofentanyl (trace), Xylazine (15p), Lidocaine (2p), Caffeine (1p) *[Fentanyl Byproducts]*
3/1/2022	Dope	Fentanyl (1p), Caffeine (0.25p), Xylazine (0.2p) *[Fentanyl Byproducts]*
3/1/2022	Dope	Fentanyl (1p), Xylazine (2p) *[Fentanyl Byproducts]* → ⑤
3/1/2022	Dope	Fentanyl (1p), Caffeine (0.3p), Xylazine (trace) *[Fentanyl Byproducts]*
3/1/2022	Dope	Fentanyl (1p), Xylazine (1p), Quetiapine (0.1p) ← ❹
3/1/2022	Dope	Fentanyl (1p), *para*-Fluorofentanyl (0.6p), Xylazine (18p), Lidocaine (trace) *[para-Fluorofentanyl Byproducts]*
3/15/2022	Dope	Fentanyl (1p), *para*-Fluorofentanyl (0.7p), Xylazine (10p) *[Fentanyl and para-Fluorofentanyl Byproducts]*
3/15/2022	Dope	Fentanyl (1p), Xylazine (1p) *[Fentanyl Byproducts]*
3/15/2022	Dope	Fentanyl (1p), *para*-Fluorofentanyl (trace), Xylazine (19p) *[Fentanyl and para-Fluorofentanyl Byproducts]*
3/15/2022	Dope	*para*-Fluorofentanyl (1p), Fentanyl (0.5p), Xylazine (3p)
3/15/2022	Dope	Fentanyl (1p), Xylazine (0.3p), Caffeine (trace) *[Fentanyl Byproducts]* → ⑥
3/15/2022	Dope	Fentanyl (1p), Xylazine (1p) *[Fentanyl Byproducts]*
3/15/2022	Dope	Fentanyl (1p), Xylazine (1.3p) *[Fentanyl Byproducts]* ← ❺

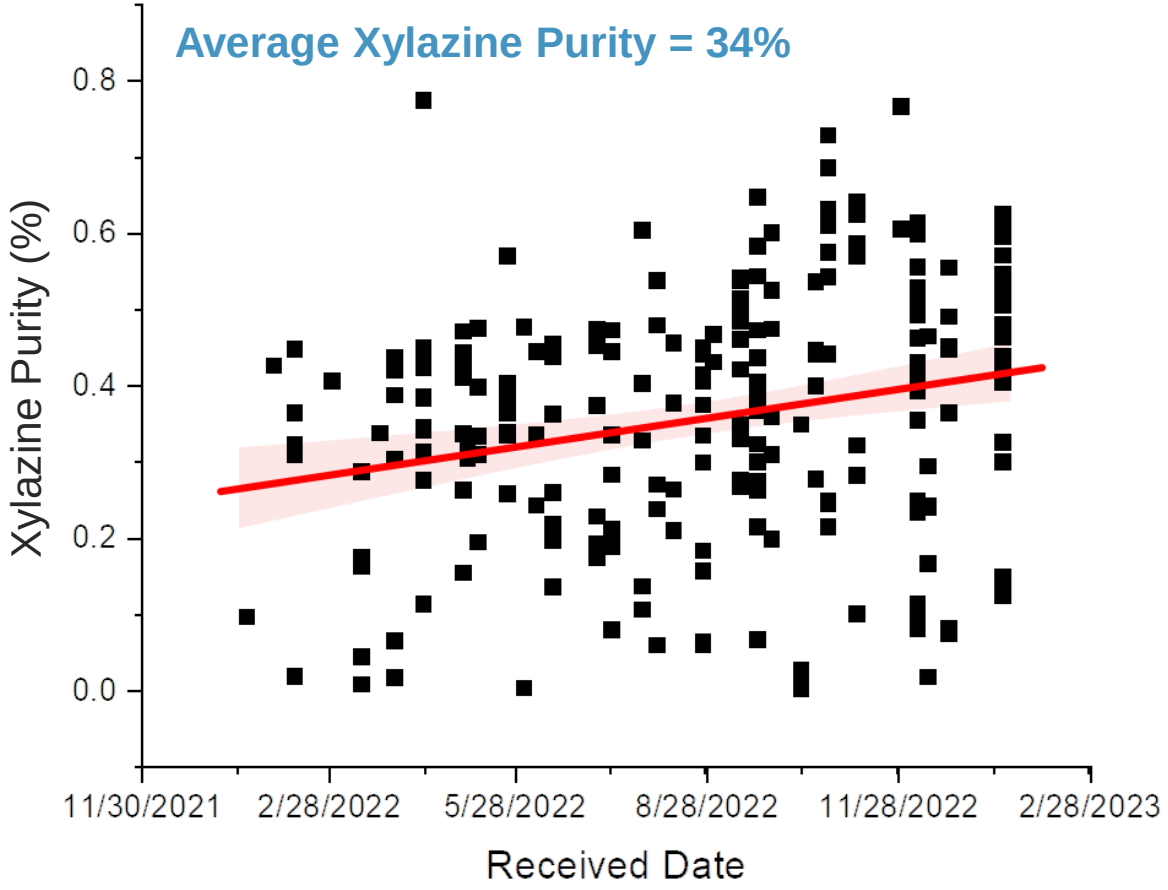


PHILLY DOPE – DRUG PRESENCE & PURITY

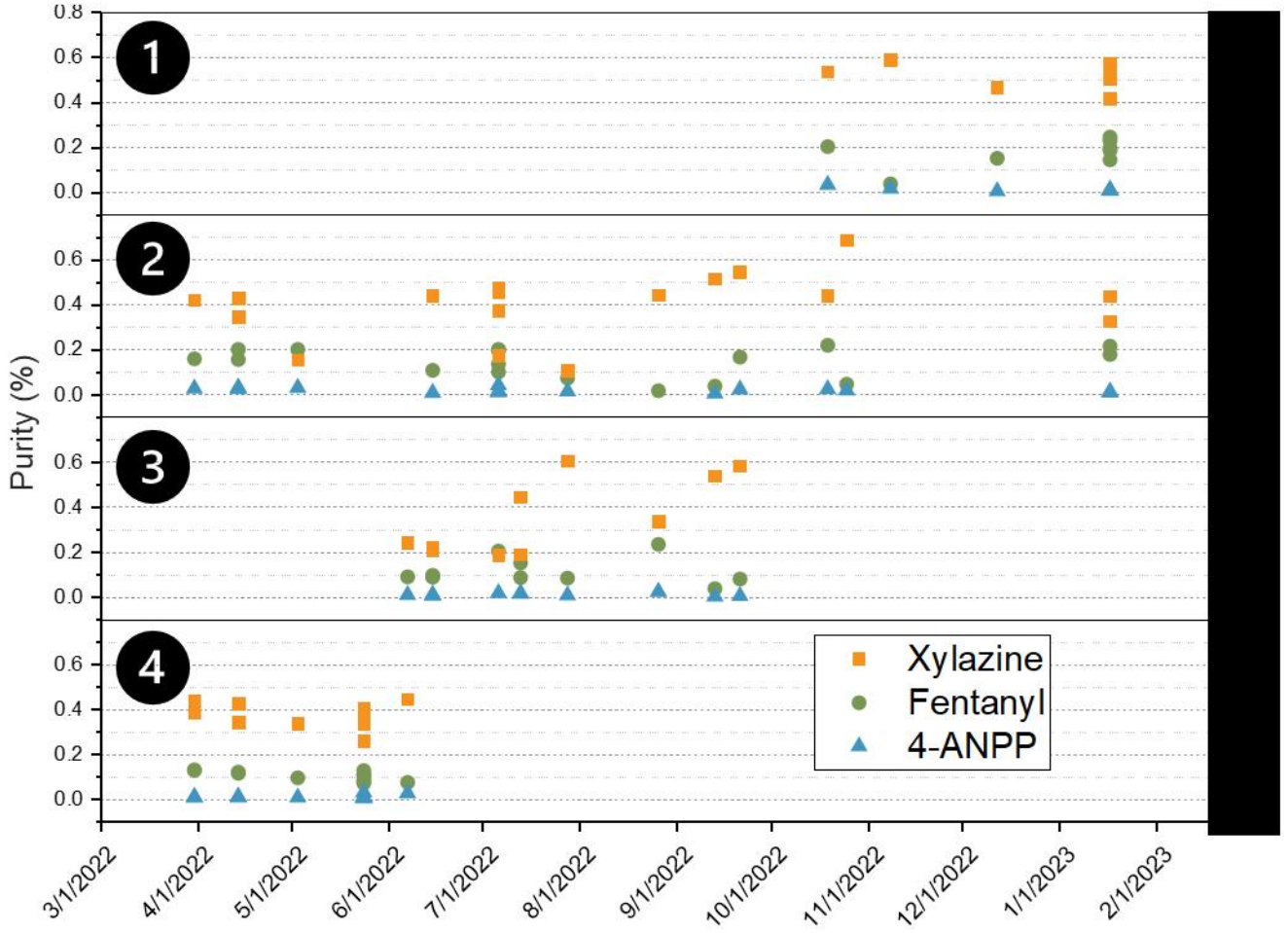
Year →	2019	2020	2021	2022	2023*
Total Samples →	47	46	199	306	156
Samples Containing Heroin (N)	12	4	26	27	20
Samples Containing Heroin (%)	26%	9%	13%	9%	13%
Avg. Purity of Heroin (%)	-	-	-	6.0%	1.8%
Samples Containing Fentanyl (N)	46	46	196	305	154
Samples Containing Fentanyl (%)	98%	100%	98%	100%	99%
Avg. Purity of Fentanyl (%)	-	-	-	12.6%	15.2%
Samples Containing Xylazine (N)	31	36	187	279	154
Samples Containing Xylazine (%)	66%	78%	94%	91%	99%
Avg. Purity of Xylazine (%)	-	-	-	34.8%	39.0%

TEMPORAL CHANGES IN PURITY (2022)





LONGITUDINAL ASSESSMENT OF DRUG PRODUCTS



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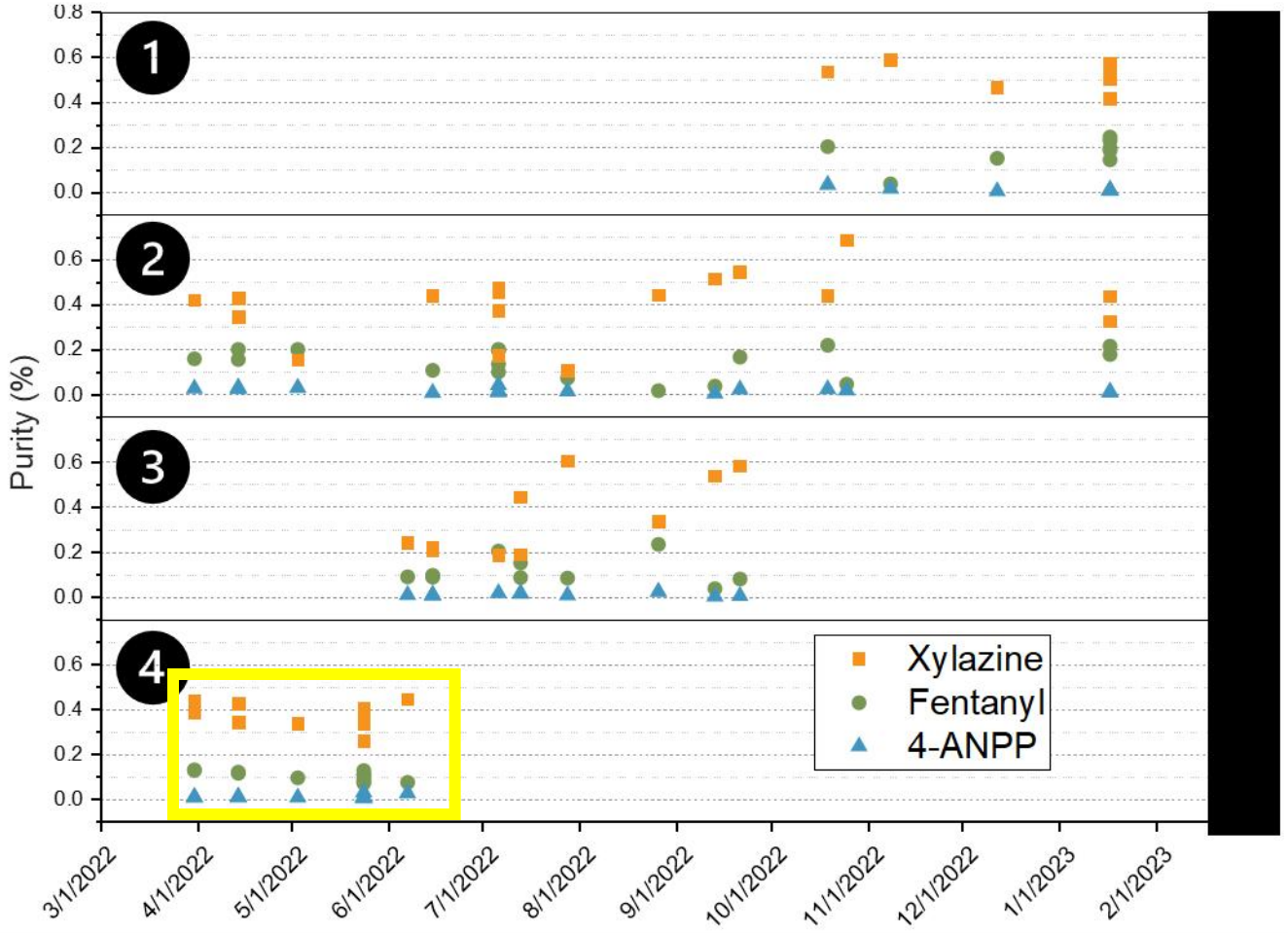

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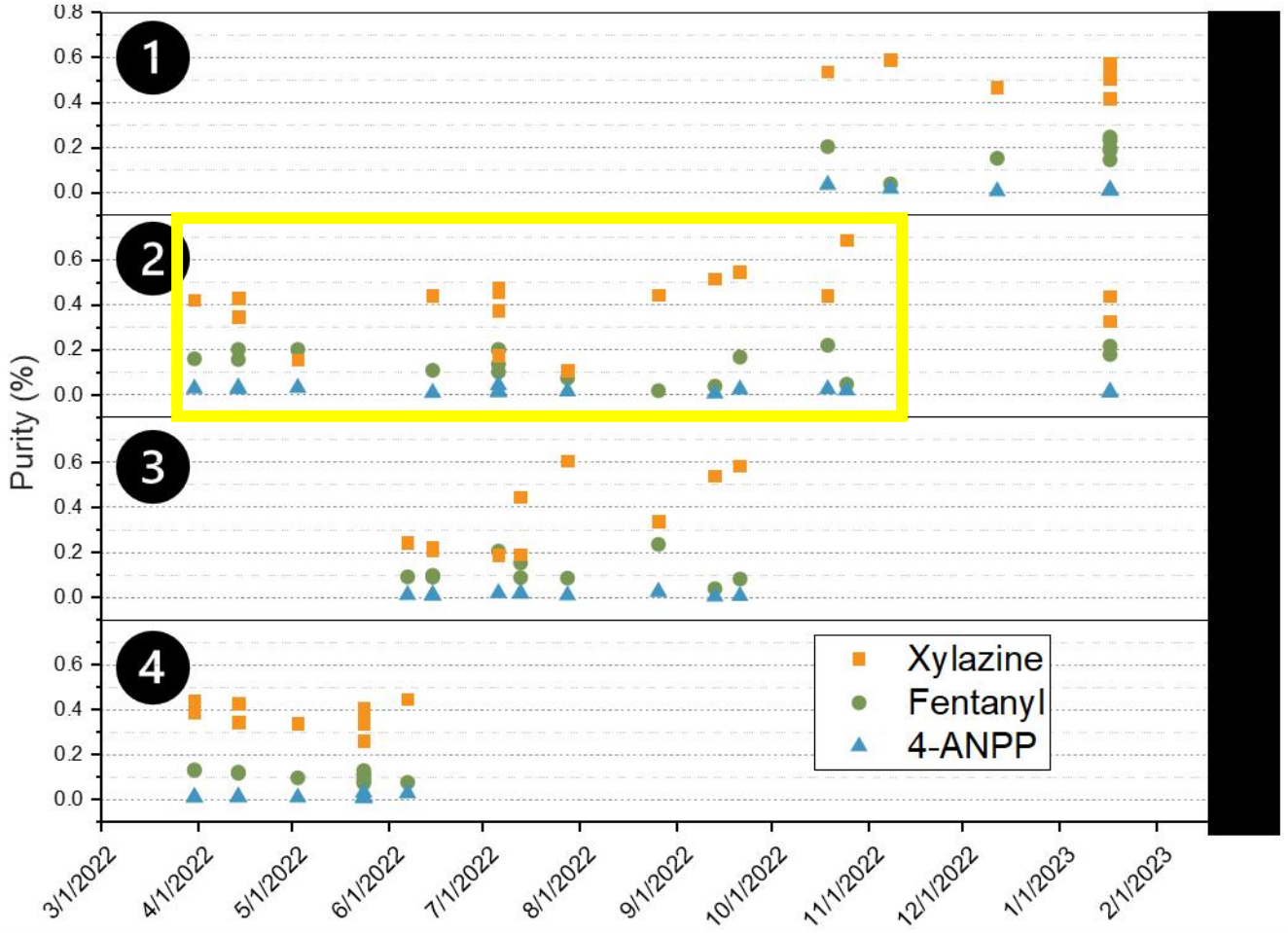

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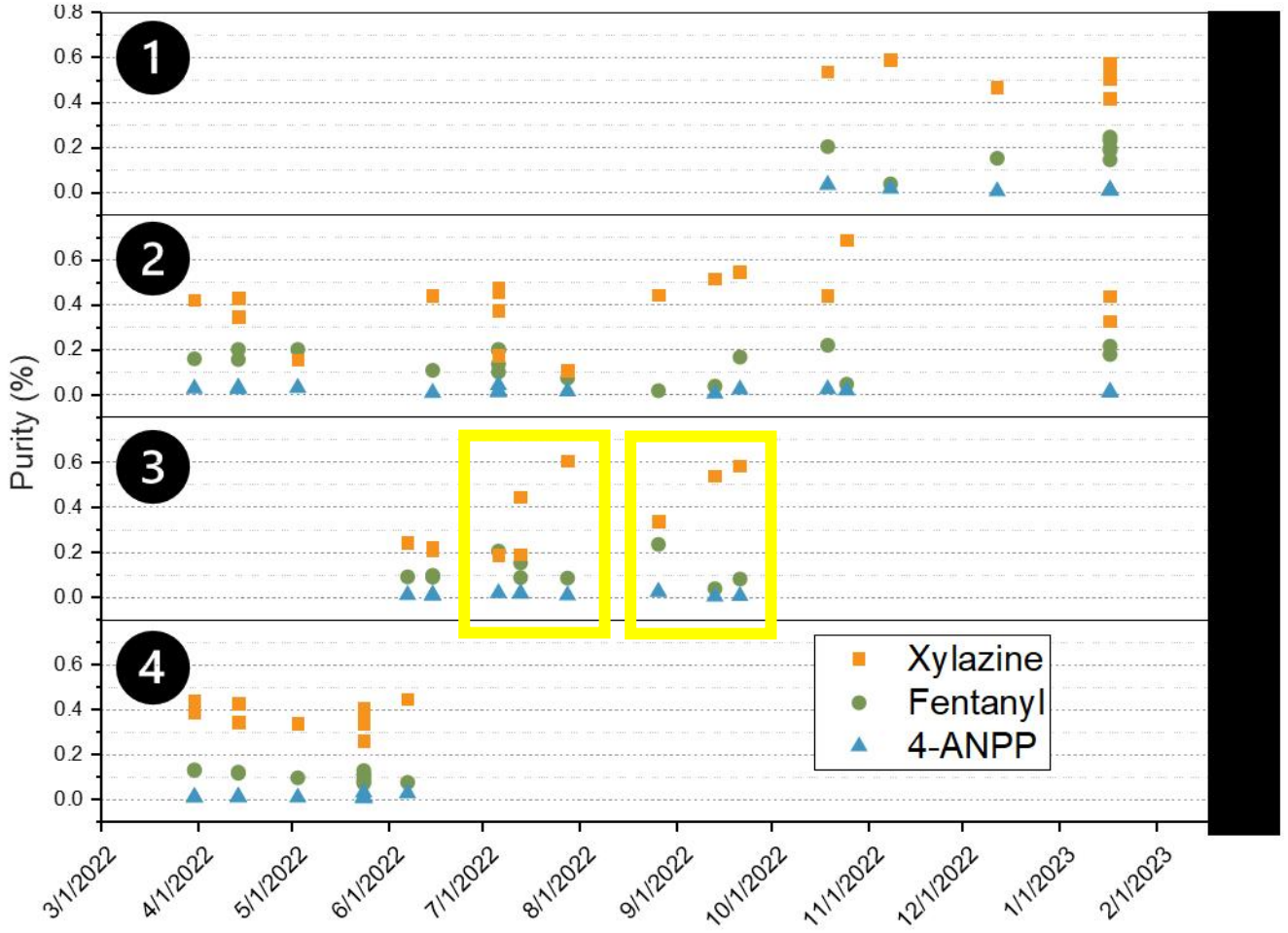
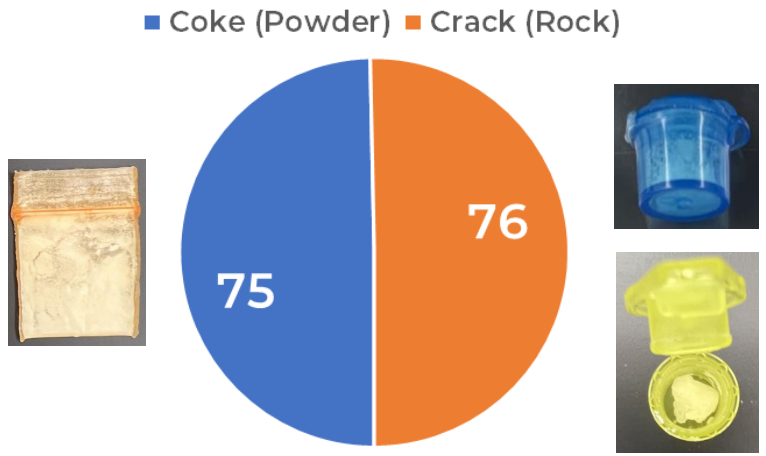
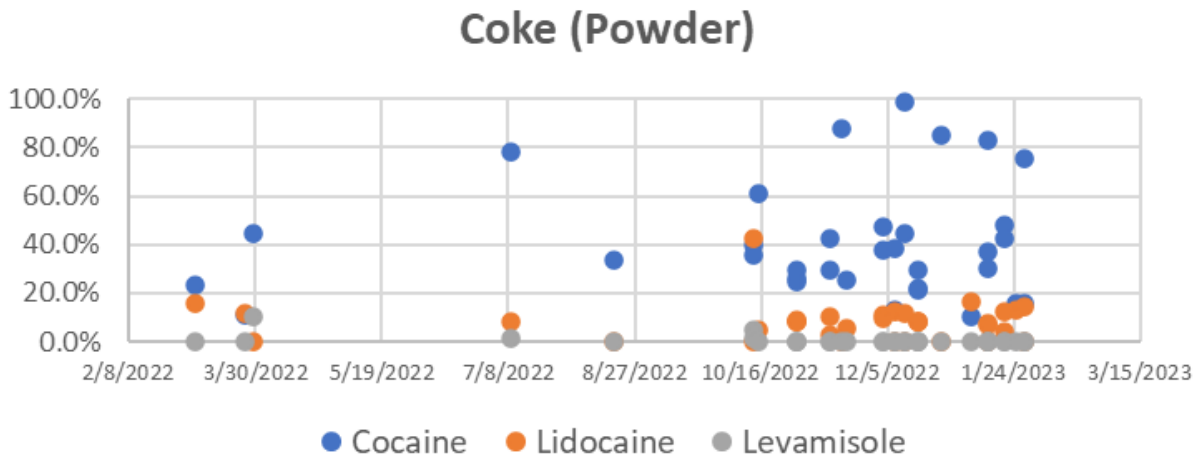
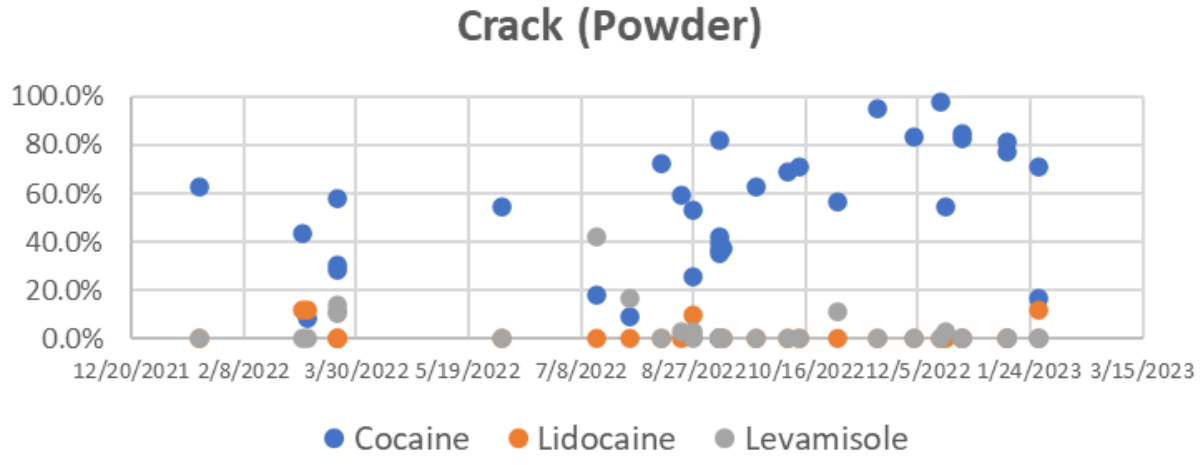


COCAINE PURITY (N=151 / 2020-2023)

Purity (%)	Mean (Std. Dev.)	Median	Range
Coke (n=34)	**40.9 ± 23.8**	36.6	10.3 - 99+
Crack (n=33)	**54.6 ± 24.8**	56.7	8.6 - 97.9







COCAINE ADULTERATION (N=151 / 2020-2023)

Adulterants	Coke (n=75)	Crack (n=76)
Lidocaine	45	10
Levamisole	14	21
Benzocaine	1	0
Dimethylsulfone	23	5
Phenacetin	7	4
Caffeine	4	1
Examples →		

Drugs	Coke (n=75)	Crack (n=76)
Fentanyl	12	1
Xylazine (all w/ Fent)	11	0
Methamphetamine	2	0

- *Five suspected coke samples contained primary drugs other than cocaine*
  - *Methamphetamine (3) and ketamine (2)*

- **Fentanyl Quants → 0.4 to 4.7%**
- **Xylazine not a concern in Philly cocaine or meth/amphetamine supplies currently**

OPIOID POTENCY INDEX (OPI)

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QUALITATIVE VS. QUANTITATIVE RESULTS

Why did the CFSRE need to move to quantitative testing to assist public health?

QUALITATIVE TESTING

- Sample A: **Fentanyl (1p)**, Xylazine (9.4p)
- Sample B: **Fentanyl (1p)**, Xylazine (18.9p)
- Sample C: **Fentanyl (1p)**, Xylazine (1.1p)
- Sample D: **Fentanyl (1p)**, Xylazine (0.3p)
- Sample E: **Fentanyl (1p)**, Xylazine (0.2p)

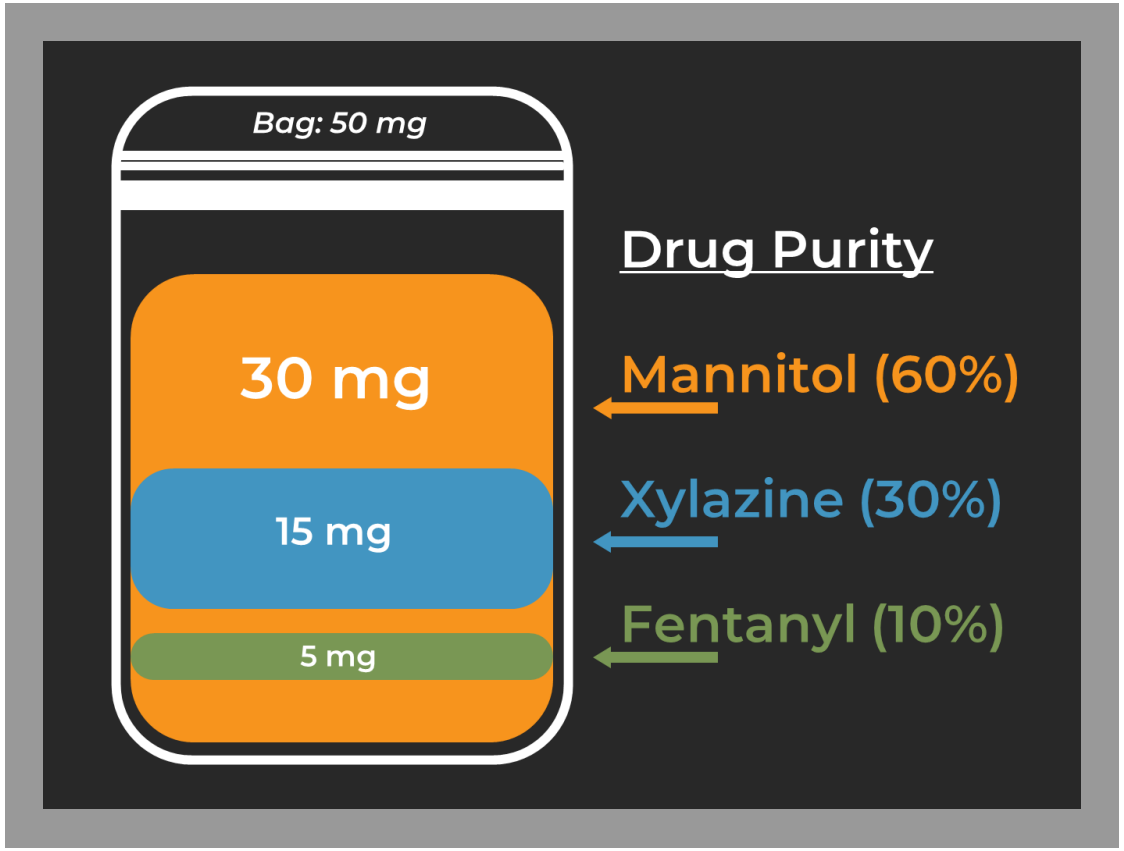
QUANTITATIVE TESTING

- Sample A: **Fentanyl (7.6%)**, Xylazine (50.6%)
- Sample B: **Fentanyl (3.8%)**, Xylazine (58.8%)
- Sample C: **Fentanyl (33.7%)**, Xylazine (35.5%)
- Sample D: **Fentanyl (7.0%)**, Xylazine (1.8%)
- Sample E: **Fentanyl (53.1%)**, Xylazine (10.1%)

PURITY VS. AMOUNT VS. DOSE

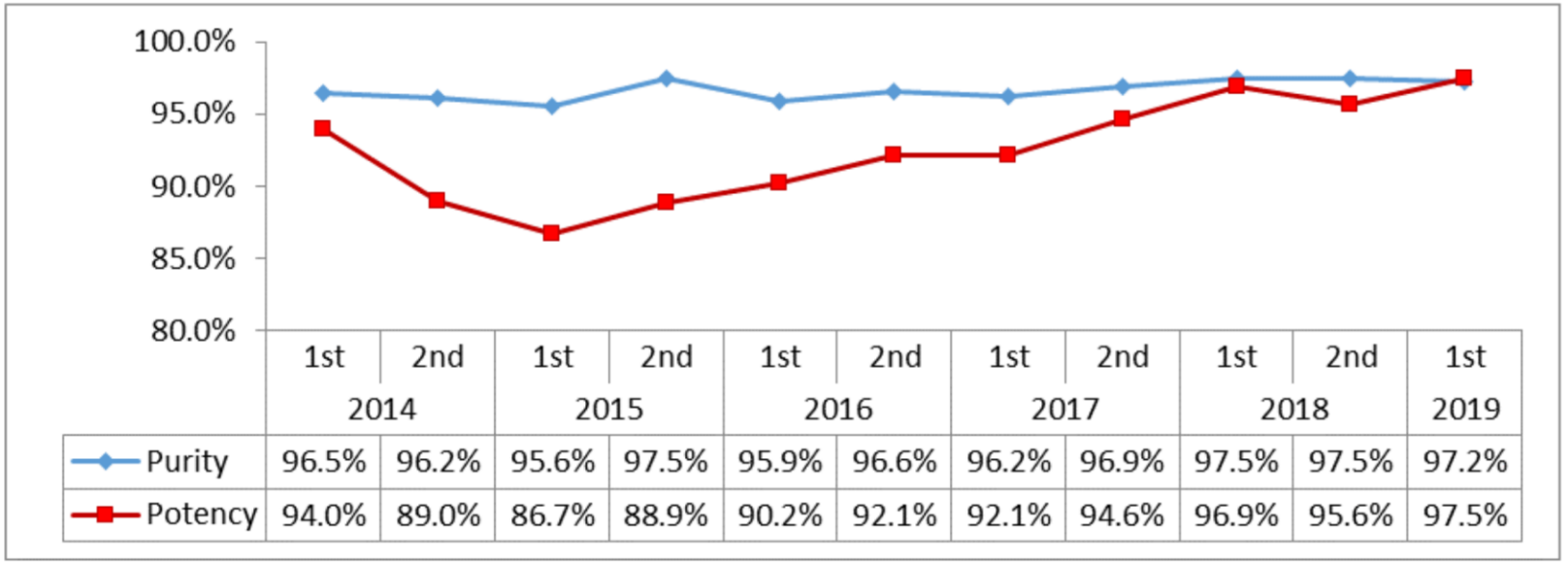
- **Purity** = proportion (%) of drug per weight
- **Amount** = weight (mg) of drug per sample
- **Dose** = total weight (mg) of drug consumed

- **Other Factors:**
  - Multiple drugs → potency index
  - Tolerance
  - Route of administration
  - Frequency of use



CONCEPTUALIZING THE APPROACH

- **Existing Concept** → Methamphetamine Purity vs. Potency
- **Purity** = amount (concentration, %) of methamphetamine
- **Potency** = amount of *d*-methamphetamine vs. *l*-methamphetamine



	1st 2014	2nd 2014	1st 2015	2nd 2015	1st 2016	2nd 2016	1st 2017	2nd 2017	1st 2018	2nd 2018	1st 2019
Purity	96.5%	96.2%	95.6%	97.5%	95.9%	96.6%	96.2%	96.9%	97.5%	97.5%	97.2%
Potency	94.0%	89.0%	86.7%	88.9%	90.2%	92.1%	92.1%	94.6%	96.9%	95.6%	97.5%

Source: DEA Methamphetamine Profiling Program

OPIOID POTENCY INDEX (OPI)

Fentanyl Purity, Potency, & Synthesis

Real-Time Testing of Opioid Drug Products in the United States

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WHAT IS FENTANYL?

Fentanyl is a synthetic opioid first synthesized in 1960. Fentanyl is used widely in medicine for the treatment of severe pain. Fentanyl is reported to be 50 to 100 times more potent than morphine. Like other opioids, fentanyl is a central nervous system depressant and in overdose scenarios can lead to a range of adverse effects, including sedation, respiratory depression, and in severe cases fatal overdose. The prevalence of fentanyl in the United States recreational drug supply has continued to increase since the mid-2000s, becoming the most frequently encountered opioid in the United States. Recreational fentanyl (sometimes referred to as illicitly manufactured fentanyl) is the primary synthetic opioid identified in fatal drug overdoses, although there are increasingly reports of fentanyl poly-drug occurrences (e.g., in combination with xylazine, benzodiazepine, stimulants). Recreational fentanyl is commonly ingested through various routes of administration, including injection, smoking, and ingestion. Fentanyl remains a drug of high public health concern among an increasingly volatile drug supply, and its prevalence has thus far only increased despite various countermeasures.

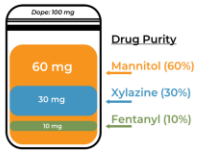

Figure 1: Illustration of drug purity.

WHAT ARE FENTANYL PRECURSORS, INTERMEDIATES, AND BYPRODUCTS?

Fentanyl is a synthetic drug produced via chemical processes and reactions between starting materials or precursors. Fentanyl can be chemically synthesized in different ways (Figures 5-9) using a variety of precursors. **Precursors** are defined as the starting molecules used for synthesis. During controlled pharmaceutical synthesis, careful selection of chemical reactions and clean-up processes ensure a correct pathway is followed, flowing through known **intermediates** to high purity final products (Figure 4). However, during clandestine synthesis, it is common that byproducts (or impurities) can appear. **Byproducts** are defined as unwanted molecules produced or left behind from chemical reactions of precursors or with intermediates. Byproducts are not the intended final drug product (e.g., fentanyl). Examples of precursors include 4-ANPD, 4-AP, benzylfentanyl, phenethylchloride, and NPP. Examples of intermediates include 4-ANPP, 4-ANBP, and benzylfentanyl. Examples of byproducts include 4-ANPP, phenethyl-4-ANPP, N-propionyl norfentanyl, and acetylfentanyl. To complicate matters, some molecules can be classified as a precursor and/or an intermediate and appear as a suspected byproduct (e.g., 4-ANPP) depending on the synthesis route. Based on currently available data or interpretive assessments, fentanyl precursors, intermediates, and byproducts are largely inactive or retain only very low opioid activity (with the exception of acetylfentanyl), making their presence in drug materials, especially in small quantities, of low pharmacological significance (although true toxicity of these chemicals remains unknown).

WHAT ARE PURITY AND POTENCY?

Purity is defined as the amount or quantity of a specific drug in a material or product. Purity is assessed on an individual drug basis and can be reported as a percent (%) or absolute weight (mg). For example, the purity of a 100 mg powder might be 10% (or 10 mg) for fentanyl, 30% (30 mg) for xylazine, and 60% (60 mg) for mannitol (Figure 1). **Potency** is defined by the strength of effects the drug product can have in humans. Potency relates the purity of all pharmacologically active constituents. **Purity vs. potency** is best described by methamphetamine quantitative testing where purity is the amount of methamphetamine in a product and potency is more specifically the amount of *d*-methamphetamine. Herein relating to fentanyl, purity is the amount of fentanyl in a product while the **Potency Index** (Figure 3) represents the combined effects of all opioids, including drugs like heroin and *para*-fluorofentanyl. *Potency Index* is calculated using relative pharmacological activity (EC_{50}), normalized, and expressed on a scale where a fentanyl powder of 10% purity represents a baseline unit of 1.

HOW IS PURITY DETERMINED?

Purity testing is determined through accurate quantitative analysis using **gas chromatography mass spectrometry (GC-MS)**. An external calibration model is developed using known quantities of drug reference materials to which the sample in question is compared. A defined weight (mg) of the drug product is measured followed by a series of specified dilutions in organic solvents and extraction of drug(s) from matrix (i.e., isolation from unwanted components). A final calculation is performed to determine purity as a percent (%) relative to the initial measured mass of an aliquot (or sub-sample).

HOW DOES PURITY TESTING SUPPORT HARM REDUCTION?

Qualitative fentanyl testing of drug products has been employed for many years in various forms for harm reduction purposes, from the employment of fentanyl test strips and FTIR in the field to GC-MS and LC-MS assays in the laboratory. The results of qualitative testing for fentanyl are useful in certain scenarios; however, it has been observed that qualitative testing alone cannot answer more complex questions about drug products and their impacts (e.g., overdose surges, unexpected adverse effects). It has long been hypothesized that comprehensive, population-level quantitative fentanyl testing would serve as a better assessment of the drug supply – an observation that continues to be assessed and validated in countries outside the United States. Having fentanyl purity and potency data allows public health officials to better understand and assess the drug supply and use outcomes. Our preliminary data show that drug purity can vary between samples marked identically over time (Figure 2).

Stamp "X"

- **June 2022** Fentanyl (9%), Xylazine (24%) Potency Index: 0.9
- **June 2022** Fentanyl (9%), Xylazine (22%) Potency Index: 0.9
- **June 2022** Fentanyl (10%), Xylazine (22%) Potency Index 1.0
- **June 2022** Fentanyl (10%), Xylazine (21%) Potency Index 1.0
- **July 2022** Fentanyl (21%), Xylazine (19%) Potency Index: 2.1
- **July 2022** Fentanyl (15%), Xylazine (19%) Potency Index 1.5
- **July 2022** Fentanyl (9%), Xylazine (45%) Potency Index: 0.9

Figure 2: Authentic quantitative data from drug products with identical markings (stamp) collected in Philadelphia, Pennsylvania, USA, showcasing fluctuation in purity and potency.

Objective: A partnership between the Center for Forensic Science Research and Education (CFSRE) and the Philadelphia Department of Public Health (PDPH) has been established to accurately assess the drug supply in Philadelphia, Pennsylvania, USA. This initiative was established as a comprehensive effort examining various drug materials and drug forms, in both qualitative and quantitative fashions. The information and results reported here represent a subset of the drug supply and not its entirety.

Acknowledgements: The authors acknowledge CFSRE and PDPH personnel for their contributions and involvements. This work was funded by a federal grant in partnership with the PDPH. The opinions, findings, conclusions, and/or recommendations expressed in this publication are those of the authors and do not necessarily reflect those of funding agencies and/or federal, state, local, or private agencies. For more information, please contact npsdiscovery@drexel.edu or visit www.npsdiscovery.org.

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Fentanyl Purity, Potency, & Synthesis (CONT.)

DRUG PURITY VS. POTENCY INDEX

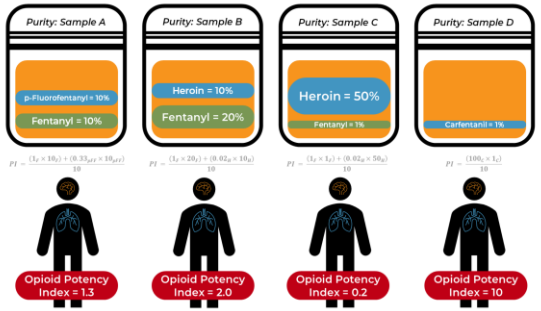
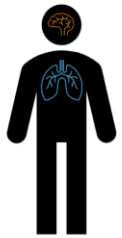


Figure 3: Illustration of drug purity (top) vs. potency index (bottom) with example calculations for opioids commonly encountered in the recreational drug supply.

$$Potency\ Index = \frac{(PF \times Purity) + (PF \times Purity)\ldots}{10} \quad (Eq.1)$$

CALCULATING POTENCY INDEX (PI)

The United States is in the midst of an overdose epidemic which is underlined by poly-drug use. Poly-drug combinations can be rooted in drug products containing, at times, more than one opioid. As quantitative drug purity data become available, scientists and public health officials need a comprehensive yet simple methodology to compare drug products. For this purpose, the **potency index** was developed. *Potency Index* is a numeric value that takes into account the amount of a drug present (i.e., purity) and its relative potency, or *potency factor* (e.g., compared to fentanyl). The calculation is the sum of all similar pharmacologically active drugs present and is normalized to a comparator (e.g., fentanyl at 10% purity) for ease of understanding and utility. Potency *Index* can be calculated for and applied to other drug classes, but herein is used as a representation of opioids.



Equation 1 can be applied to calculate Opioid Potency Index, where…

- ⇒ PF is the *Potency Factor* compared to fentanyl and is calculated as $EC_{50}^{naloxone}$/ EC_{50}^{test} at the mu opioid receptor.
- ⇒ Examples of PFs: Fentanyl = 1, Heroin = 0.02, *para*-Fluorofentanyl = 0.33, ortho-Fluorofentanyl = 3, Carfentanil = 100.
- ⇒ *Purity* is the amount of drug in a specified sample and is expressed as a percent (e.g., 10%, 23%).
- ⇒ […] indicates that the numerator should be continued for all drugs (in this case opioids) present in the sample.
- ⇒ The denominator is 10 — a *Normalizing Factor* applied so a sample of 10% fentanyl-only represents a *Potency Index* of 1.
- ⇒ *Potency Index* is reported to one decimal place (e.g., 0.9, 4.6, etc.) until the value eclipses 10.
- ⇒ Tolerance, dose, and other use factors are assumed to be constant at the individual level when assessing *Potency Index*; however, it should be understood that these factors will influence inter-individual outcomes.

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Fentanyl Purity, Potency, & Synthesis (CONT.)

Figure 4: Generic flow of chemicals during drug synthesis. Illustration shows when byproducts may be formed during the synthesis process.

FENTANYL SYNTHESIS PATHWAYS

FIGURE 5: JANSSEN (A)

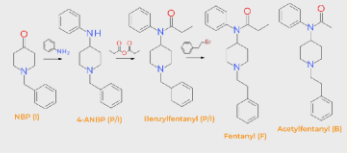


FIGURE 6: SIEGFRIED (B)

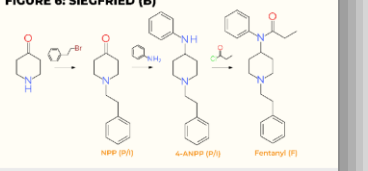


FIGURE 7: GUPTA 1 (C)

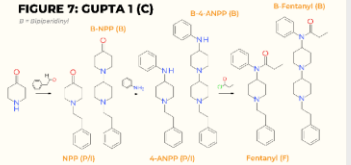


FIGURE 8: GUPTA 2 (D)

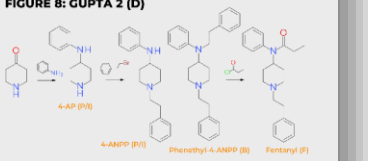
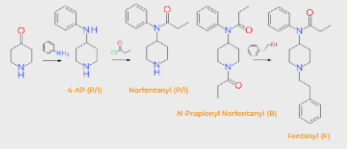


FIGURE 9: GUPTA 3 (E)



Molecule Name	Classification	Pathway Observed
4-ANBP	Precursor / Intermediate	A
4-ANPP	Precursor / Intermediate	B, C, D
4-AP	Precursor / Intermediate	D, E
Acetylfentanyl	Byproduct	A (Others possible)
B-4-ANPP	Byproduct	C
Benzylfentanyl	Precursor / Intermediate	A
B-Fentanyl	Byproduct	C
B-NPP	Byproduct	C
NBP	Precursor / Intermediate	A
Norfentanyl	Precursor / Intermediate	E (A not shown)
NPP	Precursor / Intermediate	B, C
N-Propionyl Norfentanyl	Byproduct	E
Phenethyl-4-ANPP	Byproduct	D

Note: All precursors, intermediates, and/or byproducts may not be listed for all pathways.

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OPIOID POTENCY INDEX (OPI)

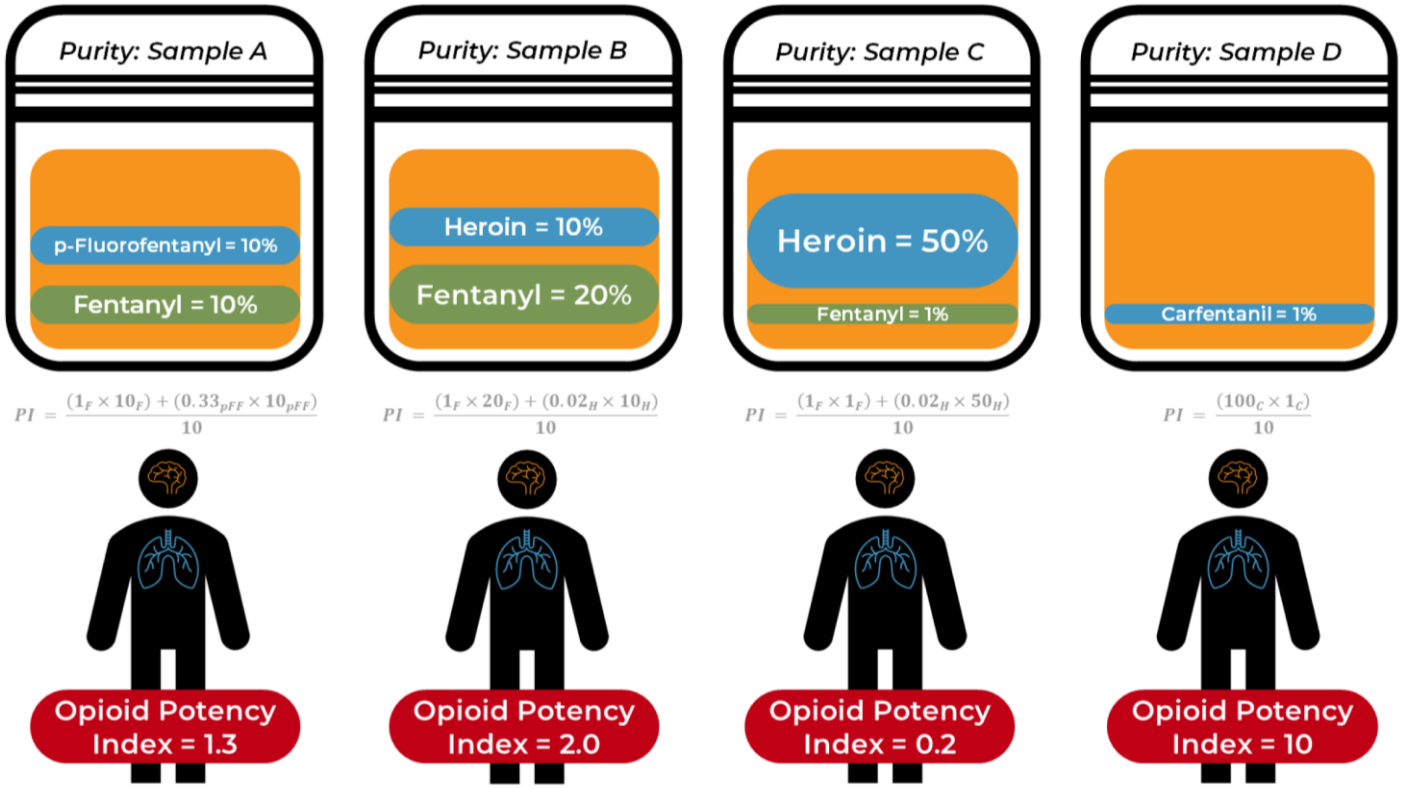

Figure 3: Illustration of drug purity (top) vs. potency index (bottom) with example calculations for opioids commonly encountered in the recreational drug supply.

Stamp "X"

June 2022
Fentanyl (9%), Xylazine (24%)
Potency Index: 0.9

June 2022
Fentanyl (9%), Xylazine (22%)
Potency Index: 0.9

June 2022
Fentanyl (10%), Xylazine (22%)
Potency Index: 1.0

June 2022
Fentanyl (10%), Xylazine (21%)
Potency Index: 1.0

July 2022
Fentanyl (21%), Xylazine (19%)
Potency Index: 2.1

July 2022
Fentanyl (15%), Xylazine (19%)
Potency Index: 1.5

July 2022
Fentanyl (9%), Xylazine (45%)
Potency Index: 0.9

Figure 2: Authentic quantitative data from drug products with identical markings (stamp) collected in Philadelphia, Pennsylvania, USA, showcasing fluctuation in purity and potency.

WHY IS OPI NEEDED?

Drugs (Purity)	Fent. Only PI	OPI
Heroin (~95%)	0	0.19
Fentanyl (2.4%), Xylazine (43.8%), **para-Fluorofentanyl (23.7%)**	0.24	1.03
Fentanyl (3.4%), Xylazine (8.2%), **para-Fluorofentanyl (3.5%)**	0.34	0.46
Fentanyl (3.5%), Xylazine (76.6%), 4-ANPP (0.4%), ***N*-Desethyl Isotonitazene (Approx. 0.05%)**, Bromazolam	0.35	0.47
Fentanyl (5.3%), Xylazine (3.2%), para-Fluorofentanyl (0.4%), 4-ANPP (0.6%), **Metonitazene (~0.5%)**	0.53	0.64
Fentanyl (9.5%), Xylazine (17.5%), **para-Fluorofentanyl (11%)**	0.95	1.31

ADDITIONAL EXAMPLES WITH OPI

Drugs (Purity)	OPI	Comments
Fentanyl (3.4%), Xylazine (8.2%), para-Fluorofentanyl (3.5%), Levamisole (1.3%)	**0.46**	N/A
Fentanyl (5.3%), Xylazine (28.3%), para-Fluorofentanyl (trace)	**0.53**	N/A
Fentanyl (4.6%), Xylazine (60.5%), para-Fluorofentanyl (0.5%), *N*-Desethyl Isotonitazene (Approx. 0.03%), Bromazolam (trace)	**0.55**	"Didn't get person well"
Fentanyl (8.8%), Xylazine (44.5%), Flubromazepam (trace), Bromazolam (trace)	**0.88**	Very "tranq" heavy
Fentanyl (8.8%), Xylazine (30.1%)	**0.88**	N/A
Fentanyl (20.2%), Xylazine (15.5%)	**2.02**	**Caused immediate OD**
Fentanyl (20.2%), Xylazine (45.3%), para-Fluorofentanyl (trace)	**2.02**	**Caused OD Surge**
Fentanyl (22%), Xylazine (44.1%)	**2.20**	**Caused 3 ODs**
Fentanyl (21.9%), Xylazine (30.1%), para-Fluorofentanyl (4.8%), Cocaine (6.0%)	**2.35**	**Involved with ODs**
Fentanyl (34.5%), Xylazine (28.8%)	**3.45**	**ODs**
Fentanyl (35%), Xylazine (2.1%), Heroin (Approx. 2%)	**3.51**	**Caused 5 ODs**

DOWNSTREAM IMPACTS

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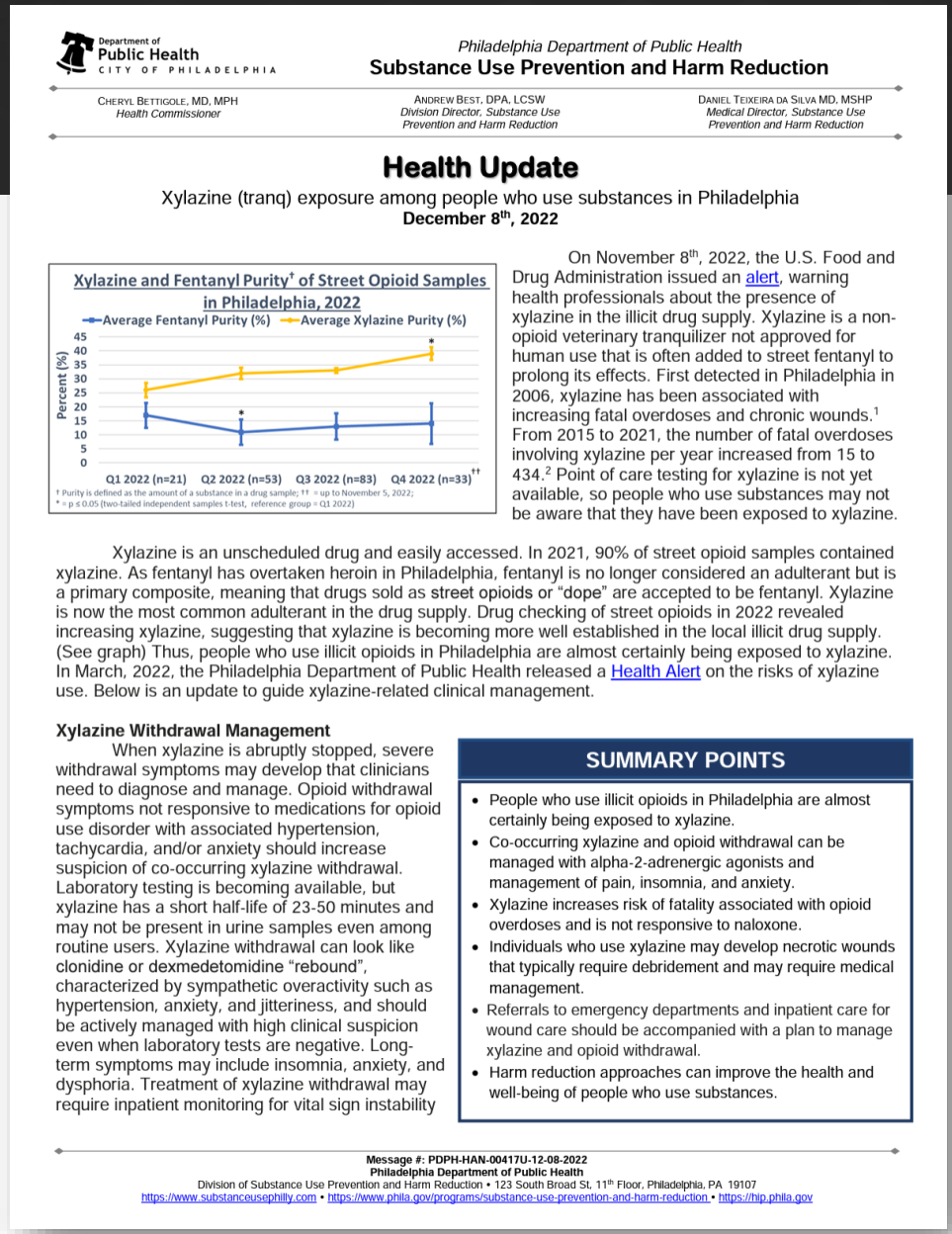
XYLAZINE PREVALENCE

- **Drug Checking →**
  - >90% of dope samples containing xylazine
  - Amount of xylazine per sample (or purity) increasing
- **Medical Examiner Data**
  - Increasing cases containing xylazine over time
- **Clinical Implications**
  - Skin infections and wound care
  - Increased sedation and adverse effects
- **Next Steps**
  - How to identify xylazine tainted dope?
    - Validation and distribution of xylazine test strips



Department of Public Health CITY OF PHILADELPHIA

Philadelphia Department of Public Health
Substance Use Prevention and Harm Reduction

CHERYL BETTIGOLE, MD, MPH — *Health Commissioner*
ANDREW BEST, DPA, LCSW — *Division Director, Substance Use Prevention and Harm Reduction*
DANIEL TEIXEIRA DA SILVA MD, MSHP — *Medical Director, Substance Use Prevention and Harm Reduction*

Health Update

Xylazine (tranq) exposure among people who use substances in Philadelphia
December 8th, 2022

Xylazine and Fentanyl Purity† of Street Opioid Samples in Philadelphia, 2022

Average Fentanyl Purity (%) — Average Xylazine Purity (%)

Percent (%)

Q1 2022 (n=21) Q2 2022 (n=53) Q3 2022 (n=83) Q4 2022 (n=33)††

† Purity is defined as the amount of a substance in a drug sample; †† = up to November 5, 2022;
* = p ≤ 0.05 (two-tailed independent samples t-test, reference group = Q1 2022)

On November 8th, 2022, the U.S. Food and Drug Administration issued an alert, warning health professionals about the presence of xylazine in the illicit drug supply. Xylazine is a non-opioid veterinary tranquilizer not approved for human use that is often added to street fentanyl to prolong its effects. First detected in Philadelphia in 2006, xylazine has been associated with increasing fatal overdoses and chronic wounds.[1] From 2015 to 2021, the number of fatal overdoses involving xylazine per year increased from 15 to 434.[2] Point of care testing for xylazine is not yet available, so people who use substances may not be aware that they have been exposed to xylazine.

Xylazine is an unscheduled drug and easily accessed. In 2021, 90% of street opioid samples contained xylazine. As fentanyl has overtaken heroin in Philadelphia, fentanyl is no longer considered an adulterant but is a primary composite, meaning that drugs sold as street opioids or "dope" are accepted to be fentanyl. Xylazine is now the most common adulterant in the drug supply. Drug checking of street opioids in 2022 revealed increasing xylazine, suggesting that xylazine is becoming more well established in the local illicit drug supply. (See graph) Thus, people who use illicit opioids in Philadelphia are almost certainly being exposed to xylazine. In March, 2022, the Philadelphia Department of Public Health released a Health Alert on the risks of xylazine use. Below is an update to guide xylazine-related clinical management.

Xylazine Withdrawal Management

When xylazine is abruptly stopped, severe withdrawal symptoms may develop that clinicians need to diagnose and manage. Opioid withdrawal symptoms not responsive to medications for opioid use disorder with associated hypertension, tachycardia, and/or anxiety should increase suspicion of co-occurring xylazine withdrawal. Laboratory testing is becoming available, but xylazine has a short half-life of 23-50 minutes and may not be present in urine samples even among routine users. Xylazine withdrawal can look like clonidine or dexmedetomidine "rebound", characterized by sympathetic overactivity such as hypertension, anxiety, and jitteriness, and should be actively managed with high clinical suspicion even when laboratory tests are negative. Long-term symptoms may include insomnia, anxiety, and dysphoria. Treatment of xylazine withdrawal may require inpatient monitoring for vital sign instability

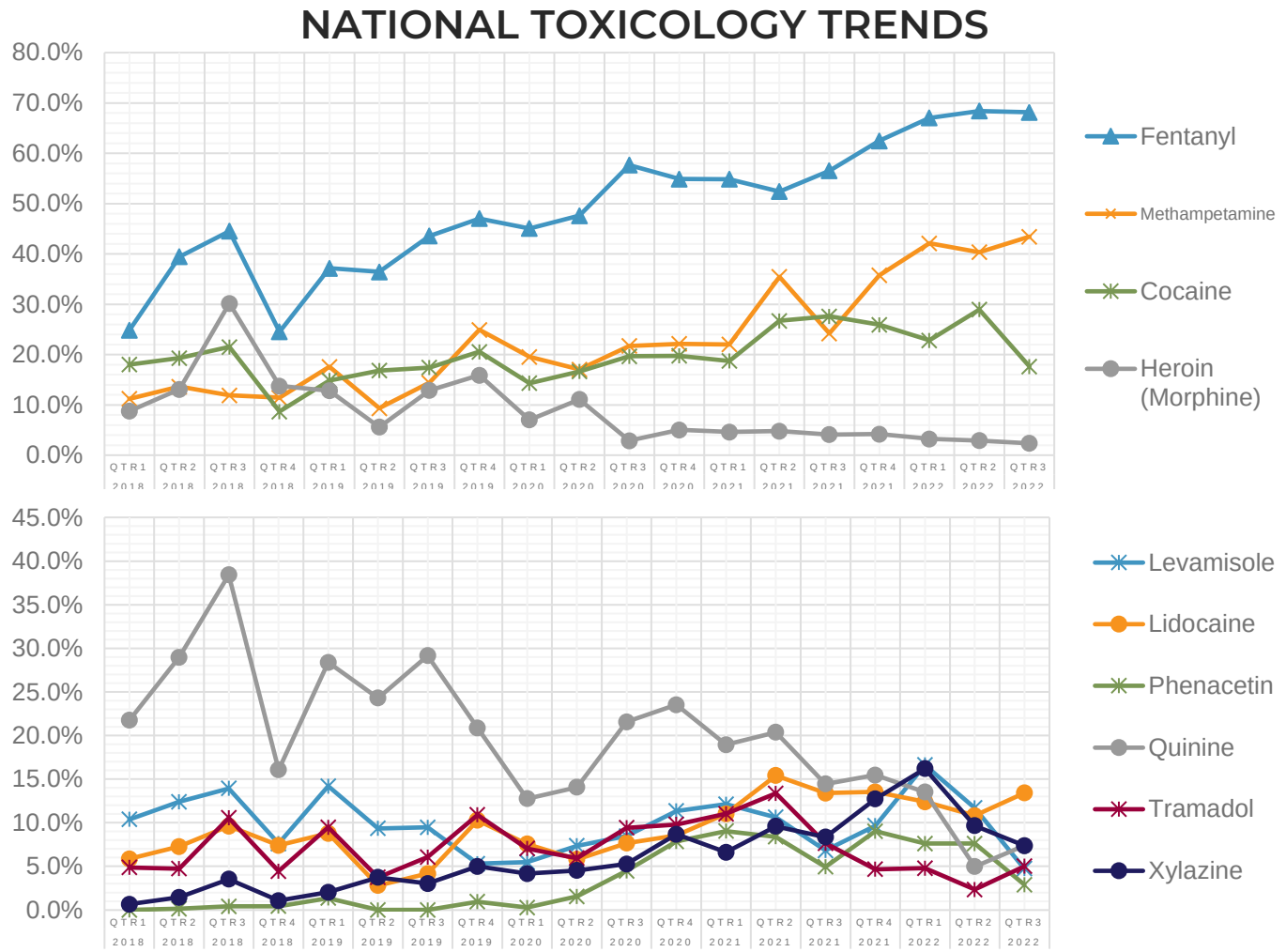
SUMMARY POINTS

- People who use illicit opioids in Philadelphia are almost certainly being exposed to xylazine.
- Co-occurring xylazine and opioid withdrawal can be managed with alpha-2-adrenergic agonists and management of pain, insomnia, and anxiety.
- Xylazine increases risk of fatality associated with opioid overdoses and is not responsive to naloxone.
- Individuals who use xylazine may develop necrotic wounds that typically require debridement and may require medical management.
- Referrals to emergency departments and inpatient care for wound care should be accompanied with a plan to manage xylazine and opioid withdrawal.
- Harm reduction approaches can improve the health and well-being of people who use substances.

Message #: PDPH-HAN-00417U-12-08-2022
Philadelphia Department of Public Health
Division of Substance Use Prevention and Harm Reduction • 123 South Broad St, 11th Floor, Philadelphia, PA 19107
https://www.substanceusephilly.com • https://www.phila.gov/programs/substance-use-prevention-and-harm-reduction • https://hip.phila.gov

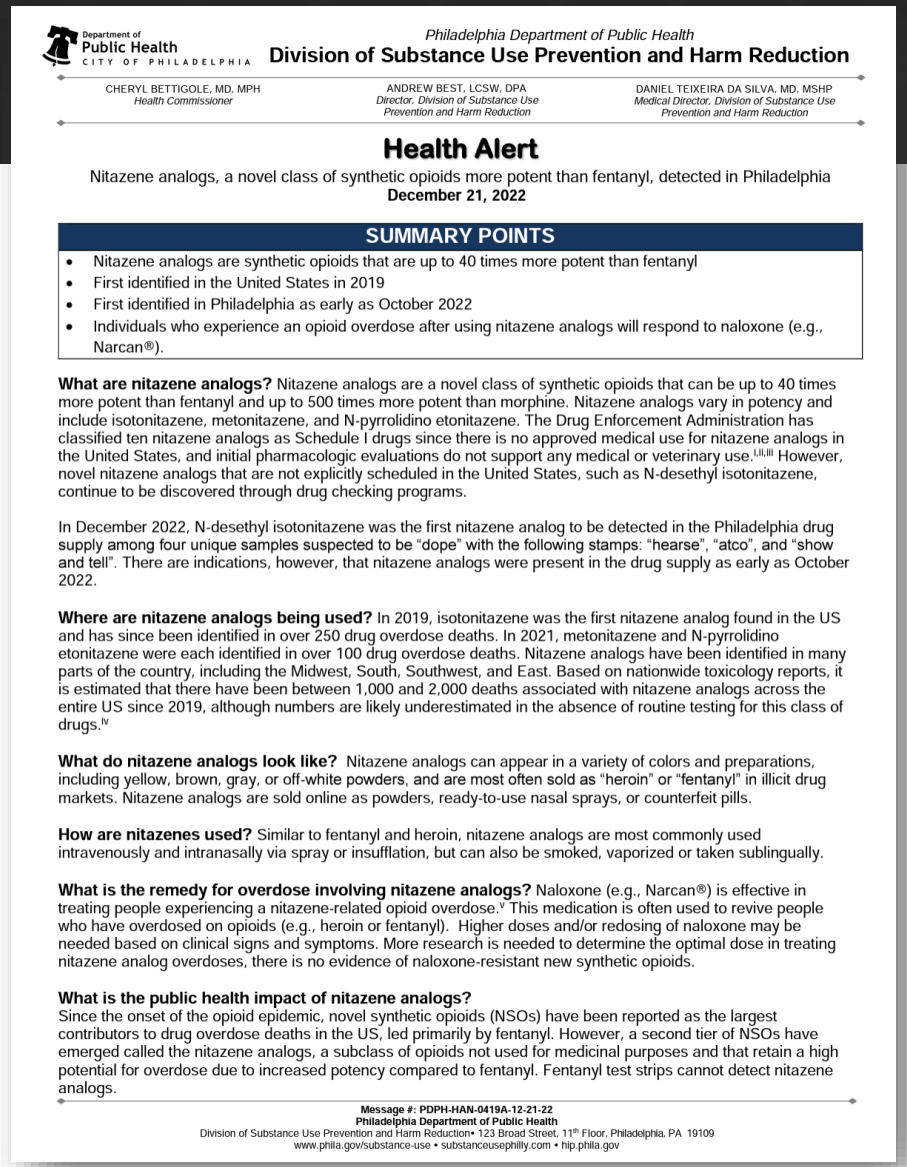
MONITORING & COMPARING TRENDS

- **Fentanyl**
  - >90% contained fentanyl
  - Rarely identifying heroin
- **Xylazine**
  - >90% contained xylazine
- **Other Adulterants**
  - Tramadol
    - 20% of dope in 2021 down to >1% in 2022
  - Lidocaine
    - ~30% of cocaine in 2022
  - Quetiapine
    - Increasing in positivity in 2022



EMERGENCE OF NEW OPIOID

- **Synopsis**
  - *N*-Desethyl Isotonitazene detected in drug supply
  - Ultra-potent novel synthetic opioid (20x more than fentanyl)
  - Alongside fentanyl, xylazine, bromazolam, and other drugs
- **Timeline**
  - First detection: September 2022 (urine)
  - Second detections: October 2022 (oral fluid)
  - Continued detections through end of 2022
- **Public Health Response →**
  - Medical examiner's office cases and testing
  - Messaging to people who use drugs
  - Naloxone dosing, administration, and monitoring



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Medical Director, Division of Substance Use Prevention and Harm Reduction

Health Alert

Nitazene analogs, a novel class of synthetic opioids more potent than fentanyl, detected in Philadelphia
December 21, 2022

SUMMARY POINTS

- Nitazene analogs are synthetic opioids that are up to 40 times more potent than fentanyl
- First identified in the United States in 2019
- First identified in Philadelphia as early as October 2022
- Individuals who experience an opioid overdose after using nitazene analogs will respond to naloxone (e.g., Narcan®).

What are nitazene analogs? Nitazene analogs are a novel class of synthetic opioids that can be up to 40 times more potent than fentanyl and up to 500 times more potent than morphine. Nitazene analogs vary in potency and include isotonitazene, metonitazene, and N-pyrrolidino etonitazene. The Drug Enforcement Administration has classified ten nitazene analogs as Schedule I drugs since there is no approved medical use for nitazene analogs in the United States, and initial pharmacologic evaluations do not support any medical or veterinary use.[i,ii,iii] However, novel nitazene analogs that are not explicitly scheduled in the United States, such as N-desethyl isotonitazene, continue to be discovered through drug checking programs.

In December 2022, N-desethyl isotonitazene was the first nitazene analog to be detected in the Philadelphia drug supply among four unique samples suspected to be "dope" with the following stamps: "hearse", "atco", and "show and tell". There are indications, however, that nitazene analogs were present in the drug supply as early as October 2022.

Where are nitazene analogs being used? In 2019, isotonitazene was the first nitazene analog found in the US and has since been identified in over 250 drug overdose deaths. In 2021, metonitazene and N-pyrrolidino etonitazene were each identified in over 100 drug overdose deaths. Nitazene analogs have been identified in many parts of the country, including the Midwest, South, Southwest, and East. Based on nationwide toxicology reports, it is estimated that there have been between 1,000 and 2,000 deaths associated with nitazene analogs across the entire US since 2019, although numbers are likely underestimated in the absence of routine testing for this class of drugs.[iv]

What do nitazene analogs look like? Nitazene analogs can appear in a variety of colors and preparations, including yellow, brown, gray, or off-white powders, and are most often sold as "heroin" or "fentanyl" in illicit drug markets. Nitazene analogs are sold online as powders, ready-to-use nasal sprays, or counterfeit pills.

How are nitazenes used? Similar to fentanyl and heroin, nitazene analogs are most commonly used intravenously and intranasally via spray or insufflation, but can also be smoked, vaporized or taken sublingually.

What is the remedy for overdose involving nitazene analogs? Naloxone (e.g., Narcan®) is effective in treating people experiencing a nitazene-related opioid overdose.[v] This medication is often used to revive people who have overdosed on opioids (e.g., heroin or fentanyl). Higher doses and/or redosing of naloxone may be needed based on clinical signs and symptoms. More research is needed to determine the optimal dose in treating nitazene analog overdoses, there is no evidence of naloxone-resistant new synthetic opioids.

What is the public health impact of nitazene analogs?
Since the onset of the opioid epidemic, novel synthetic opioids (NSOs) have been reported as the largest contributors to drug overdose deaths in the US, led primarily by fentanyl. However, a second tier of NSOs have emerged called the nitazene analogs, a subclass of opioids not used for medicinal purposes and that retain a high potential for overdose due to increased potency compared to fentanyl. Fentanyl test strips cannot detect nitazene analogs.

Message #: PDPH-HAN-0419A-12-21-22
Philadelphia Department of Public Health
Division of Substance Use Prevention and Harm Reduction• 123 Broad Street, 11th Floor, Philadelphia, PA 19109
www.phila.gov/substance-use • substanceusephilly.com • hip.phila.gov

MEDICOLEGAL DEATH INVESTIGATION

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PUBLIC ALERT: N-DESETHYL ISOTONITAZENE

New potent synthetic opioid proliferating among recreational drug supply in USA

- One of the latest nitazene analogues to emerge
- Approximately 20x more potent than fentanyl
- States: Florida, Pennsylvania, New Jersey, Colorado, etc.
- Various sample types: pills, powders, blood, oral fluid, etc.

"DOPE" SAMPLES CONTAINING *N*-DESETHYL ISOTONITAZENE

LOCATION: Philadelphia, PA, USA

NUMBER OF SAMPLES: 7+

CONTENTS (PURITY RANGE):

- Xylazine (49% to 76%)
- Fentanyl (1.1% to 5.1%)
- *N*-Desethyl Isotonitazene (0.05% to 0.4%)
- Bromazolam (trace to 2.5%)
- Flubromazepam (trace)
- *para*-Fluorofentanyl (trace)

TIMELINE — *N*-DESETHYL ISOTONITAZENE ...

Identified in urine samples from a drug treatment program (PA). — September 2022

Identified in oral fluid samples collected from people who use drugs (PA). — October

Identified in a counterfeit "A215" (oxycodone) round blue tablet (FL). — November

Identified in "dope" samples alongside fentanyl, xylazine, and bromazolam (PA). — December

Continues to be identified in "dope" samples among Philadelphia drug supply (PA). — January 2023

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CASE HISTORY

- Male in 20s found dead on friend's deck
- Suspected drug overdose
- Drug paraphernalia found on scene
  - White oval shaped "IP204" pill
- Reported history of polydrug abuse
- No additional information provided

FORENSIC LABORATORY TESTING

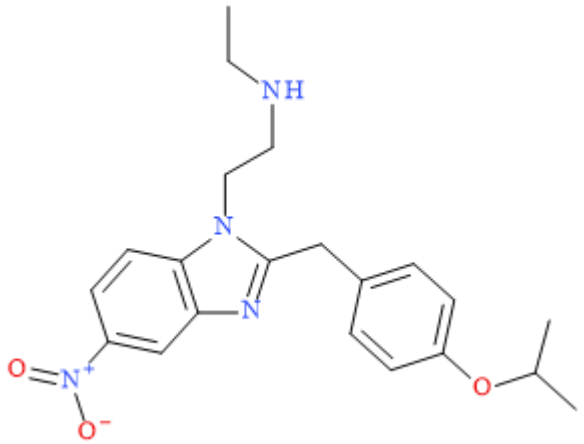
TOXICOLOGY RESULTS

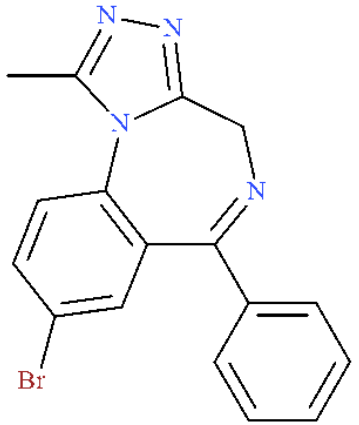
- **LC-QQQ-MS (Blood):**
  - *N*-Desethyl Isotonitazene – 5.0 ng/mL
  - Bromazolam – Positive (<5.0 ng/mL)
  - Oxycodone – Positive *(@ 41 ng/mL)*
  - Acetaminophen – Positive

- **LC-QQQ-MS (Urine):**
  - *N*-Desethyl Isotonitazene – 1.7 ng/mL
  - Bromazolam – Positive (<5.0 ng/mL)
  - Oxycodone – Positive
  - Noroxycodone – Positive
  - Acetaminophen – Positive

CHEMISTRY RESULTS

- **GC-MS (Pills):**
  - *N*-Desethyl Isotonitazene – Positive
  - Bromazolam – Identified
  - Acetaminophen – Positive
  - ***[Counterfeit oxycodone tablets]***





INTERPRETATION & DEATH CERTIFICATION

- **Toxicology Results:**
  - *N*-Desethyl Isotonitazene → novel opioid that is ~20 times more potent than fentanyl
  - Bromazolam → novel benzodiazepine suggested to be more potent than alprazolam
  - Polydrug use → Combined effects of opioids and benzodiazepines

- **Death Certification:**
  - Manner of Death:
    - Accident
  - Cause of Death:
    - Probable mixed drug intoxication (see toxicology)

Results and Conclusions:

Exhibit #	Analyte	Concentration
1 *(Blood)*	*N*-Desethyl Isotonitazene	5.0 ng/mL
1	Bromazolam	Positive (<5.0 ng/mL)
1	Oxycodone	Positive
1	Acetaminophen	Positive
2 *(Urine)*	*N*-Desethyl Isotonitazene	1.7 ng/mL
2	Bromazolam	Positive (<5.0 ng/mL)
2	Oxycodone	Positive
2	Noroxycodone	Positive
2	Acetaminophen	Positive
3 *(Pill)*	*N*-Desethyl Isotonitazene	Positive
3	Bromazolam	Identified
3	Acetaminophen	Positive

WRAP UP / CONCLUSIONS

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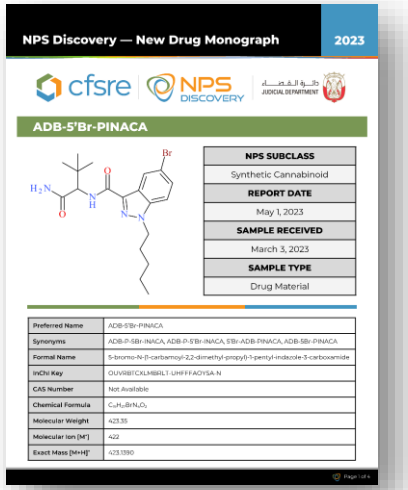
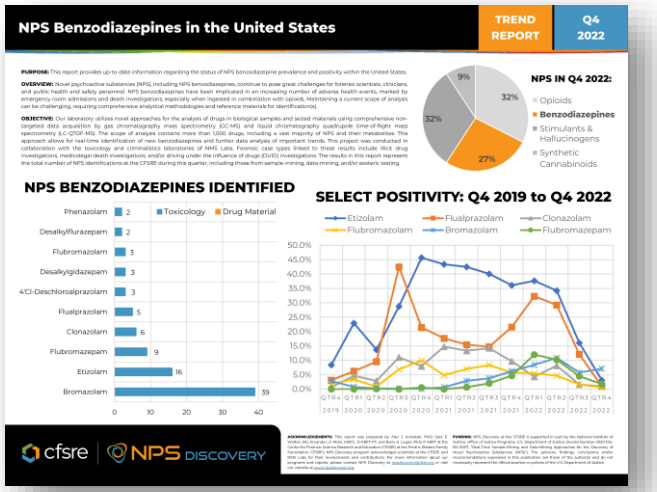
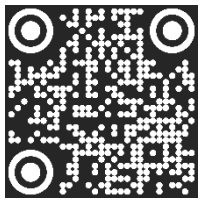
DRUG CHECKING & HARM REDUCTION INITIATIVES

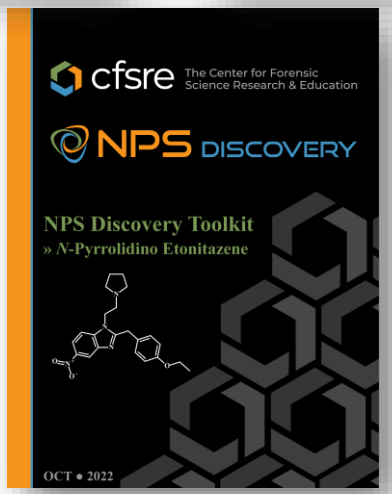
- **Vital components to public health**
  - Goal to reduce harms associate with drug consumption
  - Drug supply is ever-changing and increasing volatile
- **Drug checking can provide key information and have a positive impact on various practices**
  - Downstream impacts on PH and MDI community
  - Early warning for emerging drug trends
- **Responding can be key to achieving "success"**
  - Health alerts or public messaging
  - Collaboration continues to be superior

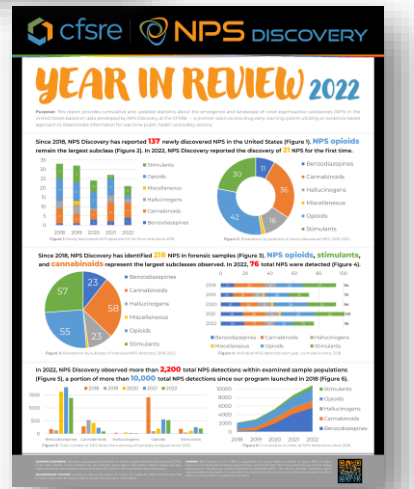
CFSRE'S NPS DISCOVERY REPORTS



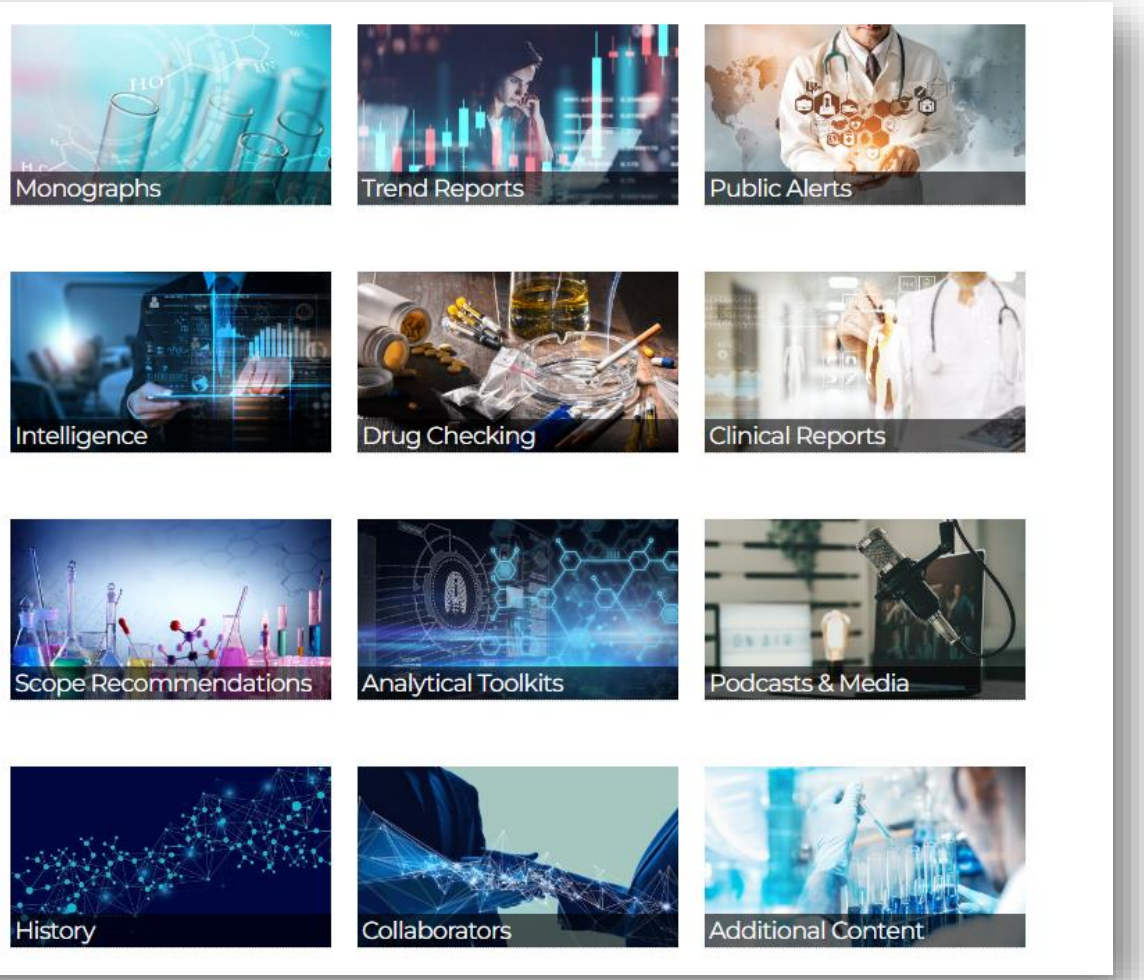








WEBSITE ▸ WWW.NPSDISCOVERY.ORG



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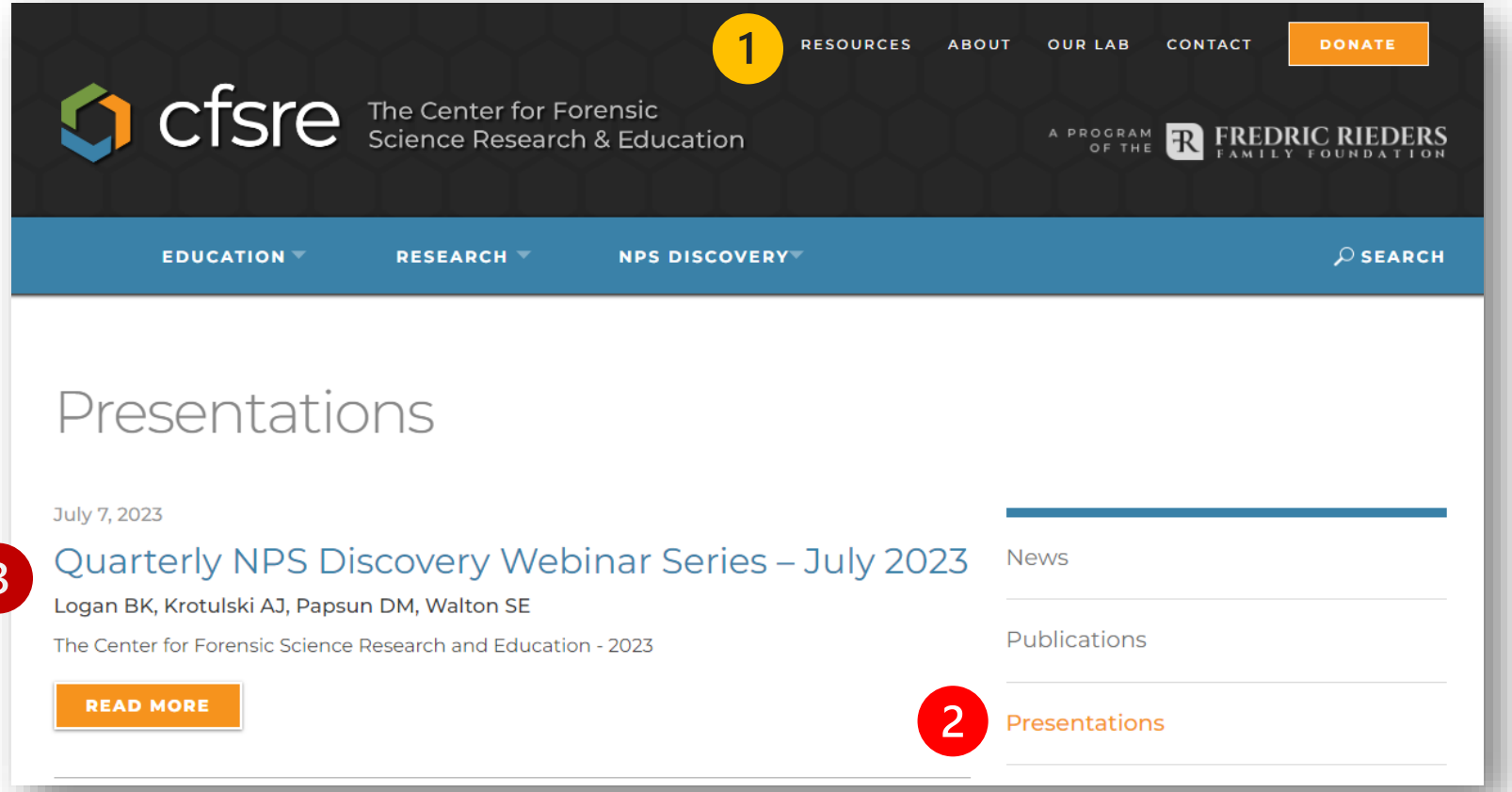

- Visit www.cfsre.org

1. Select → ***Resources***
2. Select → ***Presentations***
3. Browse & Download

SCAN ME

COLLABORATE WITH CFSRE & NPS DISCOVERY

- We accept toxicology samples and drug materials for NPS testing
- Contact Alex Krotulski for more information ▸ alex.krotulski@cfsre.org

BENEFITS OF TOXICOLOGY TESTING AT THE CFSRE:

- Perform routine testing for all NPS subclasses, including opioids, benzodiazepines, stimulants, hallucinogens, and cannabinoids.
- Assist medical examiners and coroners with determining cause of death when prior toxicology testing is negative or inconclusive.
- Analysis by state-of-the-art instrumentation and methodologies.
- Regularly updated, comprehensive in-house library database containing more than 1,000 drugs.
- Sample handling and analysis performed under chain of custody.
- Forensic quality data and individual reports generated per case.
- World-leading forensic toxicologists, chemists, and scientists.
- Laboratory follows forensic toxicology industry best practices.

TESTING CATALOG

NPS Opioids

Fentanyl Analogues, Nitazene Analogues, U-Series, AP-Series, Other Novel Opioids

NPS Benzodiazepines

Etizolam, Flualprazolam, Flubromazepam, Clonazolam, Bromazolam, Flubromazolam

NPS Stimulants

Empathogens, Cathinones, Amphetamines, Phenethylamines, Pyrrolidines

NPS Hallucinogens

Psychedelics, Dissociatives, PCP Analogues, Ketamine Analogues, LSD Analogues

Synthetic Cannabinoids

Classical, Indoles, Indazoles, Miscellaneous, Newly Emergent, & Many More!

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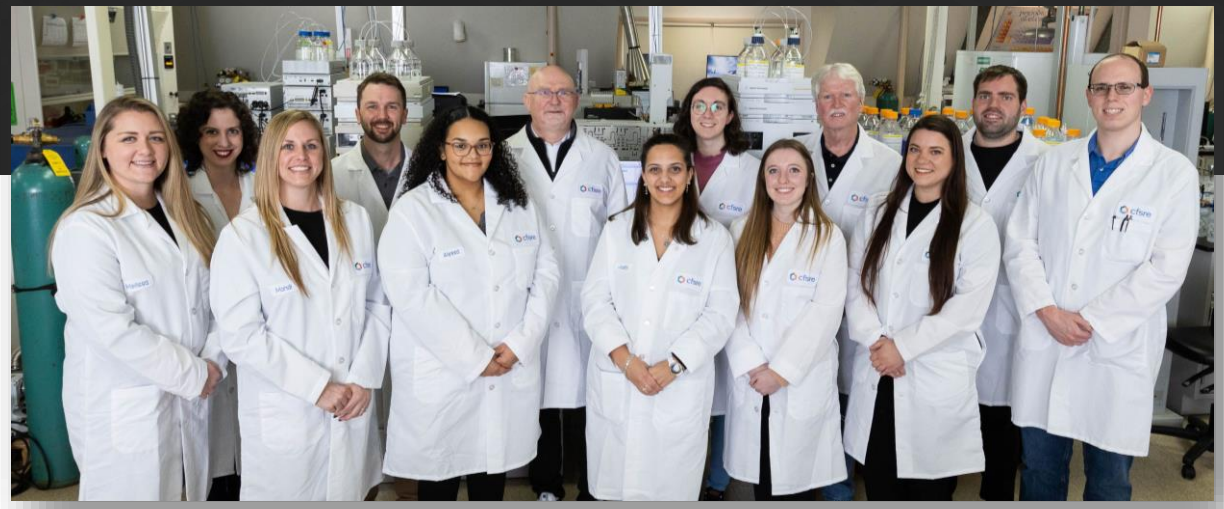
  - Clinical
  - Medical Examiners
  - Coroners
  - Crime Labs
  - Etc.



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THANK YOU! QUESTIONS?

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