Fentanyl Decontamination Studies

Evan Durnal, M.S.

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Challenging Problems. **Smart** Solutions.

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The Process

- ✓ Proof-of-concept Testing
- ✓ Fentanyl Operational testing
- ✓ Carfentanil Operational Testing
- ✓ Effluent Evaluation
- Optimization





Opiates - Physiology

• Attach to opioid receptors on nerve cells

Receptor location	Function	Opiate affect
Limbic System	Controls emotions	Creates feelings of pleasure, relaxation, and contentment
Brainstem	Controls automatic functions (breathing)	Slows breathing, reduce feelings of pain
Spinal cord	Receives sensations from body to send to brain	Decrease pain feelings



- Each analog elicits a different physiological response according to the sum of occupancies of the different receptor types and the associated binding kinetics
- Lethal dose of Heroin \approx 20-30mg
- Lethal dose of Fentanyl $\approx 2mg$
- Lethal dose of Carfentanil \approx 0.2mg



Image from www.dea.gov/.../fentanyl_briefing Guide...

"China White"

- First fentanyl illicit case 1979 in CA. Drug overdose tested negative for narcotics. In 1981 >100 over doses.
- "China White" refers to any of a number of clandestinely produced fentanyl analogues, especially α -methylfentanyl (AMF).
- Appeal of AMF: resultant drug relatively more resistant to metabolic degradation
- Other street names: Apache, China Girl, Dance Fever, Goodfella, Jackpot, Murder 8, King Ivory, Tango & Cash, Great Bear
- Pure heroin also known as China White
- Jan-March 2017, single toxicology lab¹ analyzed 293 heroin samples
 - 78% contained detectable amounts of fentanyl
- Advantages
 - Appearance: white powder; easy to mix with heroin or sugar
 - Potency: small batches and easy to hide from authorities
- Disadvantage
 - Potency: easy to overdose



1. Clark, Andrew. Director of Lab Operations – Bluewater Toxicology

Global Incidents & Emergence

- October 2002 Moscow theater hostage crisis. Russian security forces used a fentanyl cocktail to incapacitate
 - ~130/850 hostages dead
- April 2006 in Azusa, CA. Domestic fentanyl lab bust. Contained bulk fentanyl and counterfeit 80mg OxyContin tablets with fentanyl instead of oxycodone.
- June 2006, 945g of 83% pure fentanyl powder seized by Border Patrol California in a vehicle from Mexico.
- April 2016 PRINCE!!
- November 2016 Cottonwood Heights, UT. Counterfeit pills oxycodone (~70,000) and Xanax (>25,000). The accused owned a pill press and ordered fentanyl powder from China.
- Increased uses (and deaths) in Europe last 10 years as heroin supply is low due to Taliban/ISIS control on opiate production
- June 2017- 45 Kilos seized in New Jersey
- Countless others...



Potential for Dual-Use

- Wide variety of chemical and biological decontamination solutions available on the market
- Designed for field use and typically include multiple components
- Originally designed for chemical weapons decontamination
- Three (3) part mixture viable for 12-24 hours after mixing
- Shelf life of the individual components is >10 yrs





Considerations

- Standard?
- TTOP-08-2-061A decontamination verification studies
- Previous Studies
 - Garg et. al. 2010
 - Qi et. al. 2011
 - Peroxides & hypochlorites



Scheme 1. The degradation pathways of fentanyl in peracetic acid solution

Source: Qi et. al. Oxidative Degradation of Fentanyl in Aqueous Solutions of Peroxides and Hypochlorites. Defence Science Journal. 61 (1). 2011

Synthesis QA/QC

Purity analyses LC, NMR, Elemental





Analytical Methods – LC/MS/MS

 Waters Acquity I-Class ultra-performance liquid chromatograph (UPLC) coupled to a Waters Xevo TQ-XS tandem triple quadrupole mass spectrometer (MS/MS)

Analyte	MRM Ion Transitions	Polarity	Cone Voltage (V)	Collision energy (volts)
Carfentanil	395.5 > 246 395.5 > 363 395.5 > 335	Positive	15 15 15	19 10 15
Fentanyl	337 > 105 337 > 188 337 > 216	Positive	15 15 15	37 21 21

- Column: Phenomenex Kinetex PFP 1.7µm 50 × 2.1mm; S/N H15-146651
- Mobile Phase A: 0.1% Formic Acid in Water
- Mobile Phase B: 0.1% Formic Acid in Acetonitrile
- Column Temperature: 50°C
- Autosampler Temperature: 5°C
- Sample Solvent: MeOH
- Injection Volume: 0.5 μL
- Flow rate: 1.0 mL/min



Analytical Methods – GC/MS

- Agilent 7890 gas chromatograph (GC) coupled to an Agilent 5977B Inert Mass Spectrometer (MS)
- Column: Restek Rtx-5MS; 30 m × 0.25 mm × 0.1 μ m df
- Oven Program: 40°C initial, 10°C/min to 280°C hold 3 min
- Total Run Time 27 min
- Pressure: 16 psi
- Initial Flow: 2 mL/min
- Inlet: Splitless, 230°C
- Injection Volume: 1 μL
- Scan Range 40-400 m/z
- Solvent Delay 4 min
- MS Source: 230°C
- MS Quad: 150°C



Proof of Concept Objectives

- Determine 2 and 5 minute efficacy of Dalhgren Decon
 - Fentanyl Freebase and Carfentanil Oxalate
 - Two target:decontamination ratios
- Determine 2 and 5 minute efficacy of First Responder Dahlgren Decon
 - Fentanyl Freebase and Carfentanil Oxalate
 - Two target:decontamination ratios





- All test trials included negative control samples used to monitor system cleanliness each day
 - Solvent Method Blank: A volume of solvent equal to the volume of target solution used. The sample is analyzed to verify a non-detection at the start of each testing day and/or for each solvent.
 - **Decon Matrix Blank:** A volume of decon solution equal to the volume of decon solution used in test trials. The sample is analyzed to verify a non-detection at the start of each testing day and/or for each formulation.



Solvent/System Blank (left) vs Decon Matrix Blank (Right)



- All test trials included positive control samples used to verify the presence and quantity of the target compounds each day
 - **Continuing Calibration Verification (CCV):** A mid-level (usually C2-C4) calibration standard was periodically analyzed to verify system performance and recovery.
 - Sensitivity Verification Standard (SVS): The low level (C1) calibration standard was periodically analyzed to verify system sensitivity.
 - **Positive Control Spike:** A volume of target solution equal to the volume of target solution used is transferred into a container, a volume of solvent equal to the volume of decon solution used is transferred into the same container, mixed briefly, and analyzed to verify system performance and recovery.
 - **Positive Control Spike Time:** A volume of target solution equal to the volume of target solution used is transferred into a container, a volume of solvent equal to the volume of decon solution used is transferred into the same container, mixed for the appropriate time, and analyzed to verify system performance.





Fentanyl Control Spike (left) vs Sprayer 5 Minute 1:10 Decontaminated Sample (Right)



Quality Control Measurement	Frequency	Data Quality Objective
Average relative error from standard curve	Each analyte Daily	The average of the absolute values of the relative deviation across all calibration levels included in the curve must be less than 10%.
Regression Fit	Each analyte Daily	The R ² value associated with a calibration curve must be 0.98 at minimum. Values over 0.99 are preferred.
Single point relative error in curve	Each analyte Daily	No single calibration point can have a relative deviation greater than \pm 30%.
Low level standard sensitivity	Each analyte Daily	Signal-to-noise of at least 3:1 for each ion used for identification.
System Blanks	Not set	No indication of carryover of background contamination (i.e., less than 3:1 signal-to-noise for peak at analyte retention time.
Number of Calibration points	Each Curve	A minimum of four points must be used for linear regressions and five points for quadratic regressions. Removal of any point is allowable, when necessary, to meet the acceptance criteria or to improve linearity, provided the sample response remains bracketed by standards.
Quantitation Range	Each Sequence	It is acceptable to provide quantified results for samples within \pm 25% of the calibrated range. Samples outside the calibrated range but within the 25% must be caveated.





Analyte	Туре	Weighting	# points	R ²	Average % Dev.
Fentanyl	Linear	1/x	6	0.998	12.1
Carfentanil	Linear	1/x ²	6	0.994	8.1

Y



Initial Results



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Proof-of-Concept

- In-solution vs. Pure powder
- 5mg (remember LD₅₀ is ~ 2mg for 150lb person)
- Controlled laboratory pour vs. Tactical Sprayer



Operational

MRI

- Fentanyl HCl is not immediately soluble in the aqueous product
 - ~ 5 minutes for the 5mg Fentanyl to be visibly gone in the Dahlgren solution
 - Minimum reaction time tested was 5 minutes
 - Water solubility dramatically increased¹
 - ~ 1mg/mL at pH 7 to nearly 14mg/mL at pH 6
 - Dahlgren Decon pH was recorded at 6.51-7.15.

1. Samir D. Roy and Gordon L. Flynn. Solubility behavior of Narcotic Analgesics in Aqueous Media: Solubilities and Dissociation Constants of Morphine, Fentanyl, and Sufentanil. Pharmaceutical Research. 6. (2) 1989.



Bulk Results – Fentanyl HCI

Condition (Analyte Mass [mg]: Decon mass[mL])	Target mass (mg)	Decon volume (mL)	Sample pH	Dilution Volume(mL)	Expected Concentration (mg/mL)	² Observed Concentration (mg/mL)	Percent Recovery	¹ Percent Recovery (corrected for T ₀)	Decon Efficacy
DD Positive Control 1	4.61			25000	6.65E-06	6.23E-06	93.695%	100%	
DD 5:25 - 15 minutes A	4.94	25		10.00	1.78E-02	7.79E-06	0.044%	0.047%	99.95%
DD 5:25 - 15 minutes B	5.84	25		10.00	2.11E-02	6.92E-06	0.033%	0.035%	99.96%
DD 5:25 - 15 minutes C	4.60	25		10.00	1.66E-02	6.83E-06	0.041%	0.044%	99.96%
DD 5:25 - 15 minutes D	4.49	25		10.00	1.62E-02	6.93E-06	0.043%	0.046%	99.95%
DD 5:25 - 15 minutes E	4.45	25		10.00	1.61E-02	6.11E-06	0.038%	0.041%	99.96%
DD Negative Control 1		25	6.51	10.00					
DD Positive Control 2	5.92		16.0	25000	8.54E-06	8.35E-06	97.786%	100%	
DD 5:50 - 15 minutes A	5.74	50		100.00	1.04E-03	1.74E-06	0.168%	0.172%	99.83%
DD 5:50 - 15 minutes B	4.93	50		100.00	8.89E-04	1.54E-06	0.173%	0.177%	99.82%
DD 5:50 - 15 minutes C	6.24	50		100.00	1.13E-03	1.01E-06	0.090%	0.092%	99.91%
DD 5:50 - 15 minutes D	5.54	50		100.00	9.99E-04	9.94E-07	0.099%	0.102%	99.90%
DD 5:50 - 15 minutes E	6.14	50		100.00	1.11E-03	1.05E-06	0.095%	0.097%	99.90%
DD Negative Control 2		50		100.00					
DD Positive Control Spray	5.13			25000.00	7.40E-06	5.98E-06	80.752%	100%	
DD 5:25 - 5 minutes Spray A	5.74	25		10.00	2.07E-02	4.11E-06	0.020%	0.02%	99.98%
DD 5:25 - 5 minutes Spray B	5.17	25		10.00	1.87E-02	3.68E-06	0.020%	0.02%	99.98%
DD 5:25 - 5 minutes Spray C	5.85	25	7.15	10.00	2.11E-02	3.61E-06	0.017%	0.02%	99.98%
DD 5:25 - 5 minutes Spray D	5.14	25		10.00	1.85E-02	3.37E-06	0.018%	0.02%	99.98%
DD 5:25 - 5 minutes Spray E	5.45	25		10.00	1.97E-02	3.38E-06	0.017%	0.02%	99.98%
DD Negative Control 3 Spray		25		10.00					



Bulk Results – Fentanyl HCI

• >99.9% efficacy



n=5 each condition

- 15 liters of Dahlgren decon could potentially neutralize (to the LD₅₀ level) around 3 kilos of pure fentanyl
- Active fentanyl may still be present
 but at significantly less hazardous
 levels



Bulk Results – Fentanyl HCl





What about Carfentanil?

- ~ 10X stronger than Fentanyl
- Bulk (5mg) testing







Bulk Results – Carfentanil HCI

Condition (Analyte Mass [mg]: Decon mass[mL])	Target mass (mg)	Decon volume (mL)	Sample pH	Dilution Volume(mL)	Expected Concentration (mg/mL)	² Observed Concentration (mg/mL)	Percent Recovery	¹ Percent Recovery (corrected for T ₀)	Decon Efficacy
DD Positive Control 1 (T ₀)	5.79			25000.00	7.97E-06	7.53E-06	94.467%	100%	
DD 5:25 - 5 minutes A	5.70	25		10.00	1.96E-02	3.51E-07	0.0018%	0.0019%	99.998%
DD 5:25 - 5 minutes B	5.93	25		10.00	2.04E-02	5.21E-07	0.0026%	0.0027%	99.997%
DD 5:25 - 5 minutes C	5.47	25	7.14	10.00	1.88E-02	6.95E-07	0.0037%	0.0039%	99.996%
DD 5:25 - 5 minutes D	5.55	25		10.00	1.91E-02	7.40E-07	0.0039%	0.0041%	99.996%
DD 5:25 - 5 minutes E	5.67	25		10.00	1.95E-02	5.51E-07	0.0028%	0.0030%	99.997%
DD Negative Control 1		25		10.00					
DD Positive Control 1 (T ₀)	5.16			50000.00	3.55E-06	3.99E-06	112.468%	100%	
DD 5:50 - 5 minutes A	5.56	50		10.00	9.57E-03	2.92E-07	0.0031%	0.0027%	99.997%
DD 5:50 - 5 minutes B	5.77	50		10.00	9.93E-03	2.93E-07	0.0030%	0.0026%	99.997%
DD 5:50 - 5 minutes C	5.40	50	6.99	10.00	9.29E-03	2.43E-07	0.0026%	0.0023%	99.998%
DD 5:50 - 5 minutes D	5.41	50		10.00	9.31E-03	2.84E-07	0.0031%	0.0027%	99.997%
DD 5:50 - 5 minutes E	5.13	50		10.00	8.83E-03	2.35E-07	0.0027%	0.0024%	99.998%
DD Negative Control 1		50		10.00					
DD Positive Control 1 (T ₀)	5.50			25000.00	7.55E-06	8.78E-06	115.474%	100%	
DD 5:25 - 5 minutes Spray A	5.20	25		10	2.03E-02	1.59E-07	0.000781%	0.000676%	99.999%
DD 5:25 - 5 minutes Spray B	5.00	25		10	2.11E-02	1.97E-07	0.000932%	0.000807%	99.999%
DD 5:25 - 5 minutes Spray C	4.90	25	6.78	10	1.98E-02	1.43E-07	0.000723%	0.000626%	99.999%
DD 5:25 - 5 minutes Spray D	5.30	25		10	1.98E-02	1.11E-07	0.000560%	0.000485%	99.999%
DD 5:25 - 5 minutes Spray E	5.00	25		10	1.88E-02	1.80E-07	0.000958%	0.000830%	99.999%
DD Negative Control 1		25		10.00					



Bulk Results – Carfentanil HCI

• >99.99% efficacy



- The LD₅₀ of Carfentanil HCl not well characterized
- 5 liters of Dahlgren decon could potentially neutralize (to the LD₅₀ level) around 1 kilo of pure carfentanil
- Active carfentanil may still be present but at significantly less hazardous levels



- All test trials included negative control samples used to monitor system cleanliness each day
 - Solvent Method Blank: A volume of solvent equal to the volume of target solution used. The sample is analyzed to verify a non-detection at the start of each testing day and/or for each solvent.
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Solvent/System Blank (left) vs Decon Matrix Blank (Right)



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 - **Positive Control Spike Time:** A volume of target solution equal to the volume of target solution used is transferred into a container, a volume of solvent equal to the volume of decon solution used is transferred into the same container, mixed for the appropriate time, and analyzed to verify system performance.





Carfentanil Control Spike (left) vs 5 Minute 1:5 Decontaminated Sample (Right)



Effluent

- Creating a hazardous waste stream?
- Known metabolites/degradants/analogues
 - Norfentanyl
 - ANPP
 - Acetyl Fentanyl
- Unknown & Unintended components









- All test trials included positive control samples used to verify the presence and quantity of the target compounds each day
 - **Positive Control Spike:** A volume of target solution equal to the volume of target solution used is transferred into a container, a volume of solvent equal to the volume of decon solution used is transferred into the same container, mixed briefly, and analyzed to verify system performance and recovery.



Fentanyl (Blue trace) and Carfentanil (Red Trace) Locator Solutions Overlaid on MeOH blank (Black trace)



What's in the Waste?

- Six compounds entered as target chemicals and each sample chromatogram more fully interrogated via extracted ion chromatograms
- Definitively evaluate the presence/absence of the six compounds listed

Compound	CASRN	Retention Time (min)	Target lons	Present in Blank
Fentanyl	437-38-7	22.06	245, 146, 189	No
Carfentanil	59708-52-0	23.09	303, 304, 187	No
Norfentanyl	1609-66-1	15.81	83, 82, 120	No
N- Phenylpropanamide	620-71-3	10.17	93, 60, 149	No
Despropionyl fentanyl (ANPP)	21409-26-7	17.00	146, 189, 44	No
Acetyl fentanyl	3258-84-2	19.00	231, 146, 188	No



Fentanyl Effluent

		TIC: 20171212_011.D/data.ms TIC: 20171212_009.D/data.ms (*)				
48700 - 20+08 -	1	Compound	CASRN	Retention Time (min)	lons	Best Quality
	N,N-Dimet	nyloctylamine	7378-99-6	5.54	58, 157	86
3e+08 –	Long-chain	(>C10) aldehyde		6.18	43, 41, 57	93
	Tridecyl ac	etate		6.59	43, 69, 97	87
1	Long-chain	(>C10) aldehyde		6.81	43, 61, 82	92
e+08 -	Unknown H	lalogenated Ammonium salt		7.39	58, 50, 59	72
	Unknown L	ong-Chain (>C10) Amine		7.49	58, 59, 185	50
e+u8 -	Unknown A	Idehyde		7.66	41, 97, 55	38
e+08 -	Unknown A	cetate		7.92	43, 70, 55	30
	Unknown A	mine		8.27	58, 44	72
e+08 -	Unknown o	rganic Alcohol		9.22	44, 61, 83	38
e+08 -	N-Phenylpi	opanamide	620-71-3	10.142	93, 149	m
	Unknown L	ong-Chain (>C10) amine		10.591	58	53
e+08	Cetene		629-73-2	11.61	43, 41, 55	96
1	Unknown A	mine		12.53	72, 44	53
]	Unknown			12.84	58	64
+08 -	Unknown			13.03	112, 44	22
	Unknown A	Ikene		13.79	43, 41, 55	99
- 80+6	Unknown L	ong-Chain Amine		14.89	58	80
e+07	Unknown L	ong-Chain Amine		16.78	58	93
	Unknown			16.91	184	53
9+07 -	Unknown s	aturated alcohol		18.19	43, 61, 184	37
a+07	Unknown s	aturated alcohol		18.64	43, 61, 103	42
	💧 🕺 Unknown b	iphenyl		21.74	184, 92	46
1e+07	Fentanyl (r	n)	437-38-7	22.06	245, 146, 189	m
1 minut	m-manual	search, spectral subtraction, and/or	ion extraction required t	o identify		
ne-> 5.00	10.00	15.00	20.0	0	25.00	

Overlaid TICs of 25mL (Blue Trace) and 50mL (Black Trace) Fentanyl Effluent



Carfentanil Effluent

	Compound	CASRN	Retention Time (min)	lons	Best Quality	
bundance	N,N-Dimethyloctylamine	7378-99-6	5.58	58, 157	87	
.4e+08 -	Unknown Long-Chain (>C10) Amine		5.68	44, 61	50	
	Long-chain (>C10) aldehyde		6.14	43, 61, 103	53	
.2e+08 -	Unknown		6.27	43, 70, 103	42	
3e+08 -	Long-change (>C10) Alkene		6.59	43, 69, 97	64	
	Long-chain (>C10) aldehyde		6.82	43, 61, 82	92	
.86+08 -	Unknown Ammonium salt		7.32	58, 50, 59	64	
.6e+08 -	Unknown Long-Chain (>C10) Amine		7.49	58, 59, 185	52	
40+00	Unknown		7.70	41, 97, 55	35	
	Unknown Acetate		7.99	43, 70, 55	43	
.2e+08 -	Unknown Amine		8.31	58, 44	72	
2e+08	1-Tetradecene	1120-36-1	9.25	44, 61, 83	96	
	N-Phenylpropanamide	620-71-3	10.15	93, 149	m	
.8e+08 -	Unknown Long-Chain (>C10) amine		10.59	58	64	
.6e+08 -	Cetene	629-73-2	11.61	43, 41, 55	96	
	Unknown Amine/Amide		12.54	72, 44	53	
.4e+08 -	Unknown w/ Amine group		12.86	58	95	
.2e+08	TENTATIVE 2-(Hexamethyleneimino)ethanol	20603-00-3	13.03	112, 58, 44	m	
1e+08 -	Long Chain (>C10) Alkene		13.78	97. 83. 69	99	11
8e+07	Dimethyl palmitamine	112-69-6	14.89	58	74	anii
	Dimantine	124-28-7	16.77	58	93	FIC)
6e+07 -	TENTATIVE					
4e+07 -	(3S.5R.8aR)-3-Butvl-5-	96894-83-6	16.85	166, 180, 222	m	
	propyloctahydroindolizine			,		\sim
	1-Decanamine. N-decvl-N-methyl-	7396-58-9	16.92	184, 58	m	
ime-> 5.00	Unknown		19.58	210, 182	35	25.00
	Unknown		21.74	184, 58	38	
	Unknown		22.05	182, 184, 131	90	
	Carfentanil	59708-52-0	23.09	303, 212, 105	m	
	m manual coarch coactral subtraction or	ad/or ion avtracti	on required to id	optify		

m –manual search, spectral subtraction, and/or ion extraction required to identify



Effluent Summary

- N-Phenylpropanamide
- LD₅₀ Oral mouse of 1,100 mg/kg
- Not considered a dangerous good by IATA, the US DOT, or IMDG
- NFPA health hazard rating of one
- Automated Integration and Search
- Manual Search and Confirmation

Component	Decontaminated Fentanyl Effluent	Decontaminated Carfentanil Effluent
Fentanyl	\checkmark	X
Carfentanil	X	\checkmark
N-Phenylpropanamide	\checkmark	\checkmark
N,N-Dimethyloctylamine	\checkmark	\checkmark
Cetene	\checkmark	\checkmark
Diamantine	\checkmark	\checkmark
1,1'-Biphenyl, 4-methoxy-	\checkmark	X
1-Tetradecene	X	\checkmark
TENTATIVE 2-(Hexamethyleneimino)ethanol	x	\checkmark
Dimethyl palmitamine	X	\checkmark
1-Dodecanamine, N,N-dimethyl-	X	\checkmark
1-Tetradecanamine, N,N-dimethyl-	X	\checkmark

X = absent; 🗸 = present

Component	Decontaminated Fentanyl Effluent	Decontaminated Carfentanil Effluent
Acetyl Fentanyl	X	X
Norfentanyl	X	X
Propanil	X	X
Benzaldehyde	X	X
Benzonitrile	X	X
Despropionyl fentanyl (ANPP)	X	X

X = absent; \checkmark = present



In Conclusion....

• >99.9% effective



- Effluent is free of hazardous byproducts
- <u>NOT</u> final field guidance
 - Specific decon to threat ratio needed
 - Specific decon time needed
 - Efficacy on additional analogues



Questions?

Contact us:

MRIGlobal

816-326-5466

edurnal@mriglobal.org

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