In-Home Deactivation System for Psychoactive Drugs (SBIR Phase 2)

Final Report

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Verde Environmental Technologies
Minnetonka, MN

NIDA Study Contributors:

Verde

Andrew Korey Ph.D., Principal Investigator

William Fowler
Carter Anderson
Clay Anderson

Mercer University

Ajay K. Banga Ph.D. (Co-Investigator)
Professor and Department Chair
Department of Pharmaceutical Sciences
College of Pharmacy & Health Sciences
Mercer University, Atlanta GA.

Mahima Manian
Yang Song
Xinyi Gao
Pooja Bakshi
Yujin Kim
Sindhu Ganti
Behnam Bozorg

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Summary

When consumers became aware of the Deterra System, they were enthusiastic and used the product almost immediately. 91% of all respondents were motivated by concern for the environment, with 45% citing concern about drug abuse or diversion, and 37% citing concern about accidental poisoning.

Providers of Deterra were similarly enthusiastic about the System, and indicated high concern for the environment, though a significantly higher percentage of them also cited concern for abuse/diversion and/or accidental poisoning.

The activated carbon system was highly effective in adsorbing and deactivating all of the drugs tested, with an average of 89% of API adsorbed within the first 8 hours, and 99.6% deactivated at 28 days.

Activated carbon was highly effective for all formulations, and all chemical classes of drugs tested, and data from stability studies predict >10 years stability at room temperature.

The adsorbed pharmaceuticals were resistant to leaching by water, with only trace amounts detectable after an extensive washout.

The adsorbed pharmaceuticals were resistant to leaching by a washout procedure with 30% ethanol, with few exceptions. This demonstrated that the activated carbon was effective in rendering adsorbed pharmaceuticals unrecoverable by simple means.

No product complaints or adverse events were reported

Background Information: Verde has developed an inexpensive prescription medication disposal system that provides a simple means for patients or members of their households to safely render prescription drugs unusable and effectively contained in order to minimize the potential for diversion or accidental exposure to children or pets.

The disposal system consists of a 4" x 7" polyethylene-polyethylene terephthalate composite film pouch, with a zip-type water-tight seal. Inside the bag is a water-soluble polyvinyl alcohol wrapped pod that contains 15 grams activated carbon. The bag dimensions were established to contain an estimated unused amount of an individual prescription, i.e. up to 15 tablets and approximately 50 ml of water.

In the Phase I SBIR project, Verde demonstrated the feasibility, safety and marketability of the product. The product is now in deployable form for mass- distribution.

Phase II Technical Objectives described in the contract were for Verde to conduct assays, tests and surveys to assess:

1. The degree of adherence by prescribers, pharmacies and/or home-users
2. Whether adoption and usage under normal household conditions improves outcomes
3. The classes of chemical compositions and delivery systems for which the product is most effective
4. Adverse events from the use of the product
5. The durability or shelf life of the product
6. The development of ancillary materials to support adoption and consistent use

Technical Objectives 1 and 2: To determine the degree of adherence by prescribers, pharmacies and/or home-users, and (2) Whether adoption and usage under normal household conditions improves outcomes

Deterra System Survey Results In Brief…

1700	Number of user surveys distributed
32	Number of provider surveys distributed
1% to 15%	Typical response rate for external surveys
14%	Response rate of Deterra User Surveys
64%	Response rate of Deterra Provider Surveys
3	Number of pharmacies incentivized to distribute Deterra Surveys
20%	Response rate of pharmacy customers
91%	Consumers concerned with preventing damage to the environment
45%	Consumers concerned with preventing drug abuse or diversion
37%	Consumers concerned with preventing accidental poisoning
95%	Consumers who had no difficulty using the Deterra System pouch.
96%	Consumers who will use the Deterra System within 4 weeks
0	Number of providers who recommended product improvements
100%	Providers who will continue to provide Deterra System pouche
1	Rank of comment, “What do I do with the inside bag?”

Summary

Between April 25 and May 1, 2016, a total of 1665 Deterra System User Surveys were given to 6 different providers to distribute to users, along with free samples of the Deterra pouch. Each provider of User Surveys was given Provider Surveys for staff, and Provider Surveys were also sent to current Deterra System bulk users. To date (June 20, 2016), 233 User Surveys and 18 Provider Surveys have been sent back to Verde anonymously via mail. Location responses were tracked by a small code on each survey envelope.

Survey Type	Location Type	Location Name	Code	# Supplied	# Returned	Rate of Return
User	Pharmacy	Atlantic Apothecary	DSU1	350	94	27%
User	Pharmacy	Bayard Pharmacy	DSU2	350	9	3%
User	Pharmacy	Cape Pharmacy	DSU3	350	109	31%
User	Law Enforcement	Hennepin County Sheriff	DSU4	350	0	0%
User	Law Enforcement	Eden Prairie Police	DSU5	200	10	5%
User	Various	Other	DSU6	65	11	17%
TOTAL User				**1665**	**233**	**14%**
Provider	ALL	Various	DSP	**28**	**18**	**64%**

In general, the product was well received and people seemed eager to use it. Given some comments, they also seemed eager to have another chance to use it.

Providers of User Surveys

Pharmacies:

- Atlantic Apothecary
- Bayard Pharmacy
- Cape Pharmacy

Law enforcement agencies:

- Hennepin County Sheriff
- Eden Prairie Police

Various Sources of Completed User Surveys:

- Pharmacists who were unable to distribute surveys at their place of business but wanted to take the survey
- Members of the 3M First Response Team
- Other interested parties

Potential Respondents to Provider Surveys

- Atlantic Apothecary
- Bayard Pharmacy
- Cape Pharmacy
- Hennepin County Sheriff
- Eden Prairie Police
- Allina Pharmacy

Consumer Response to Deterra System

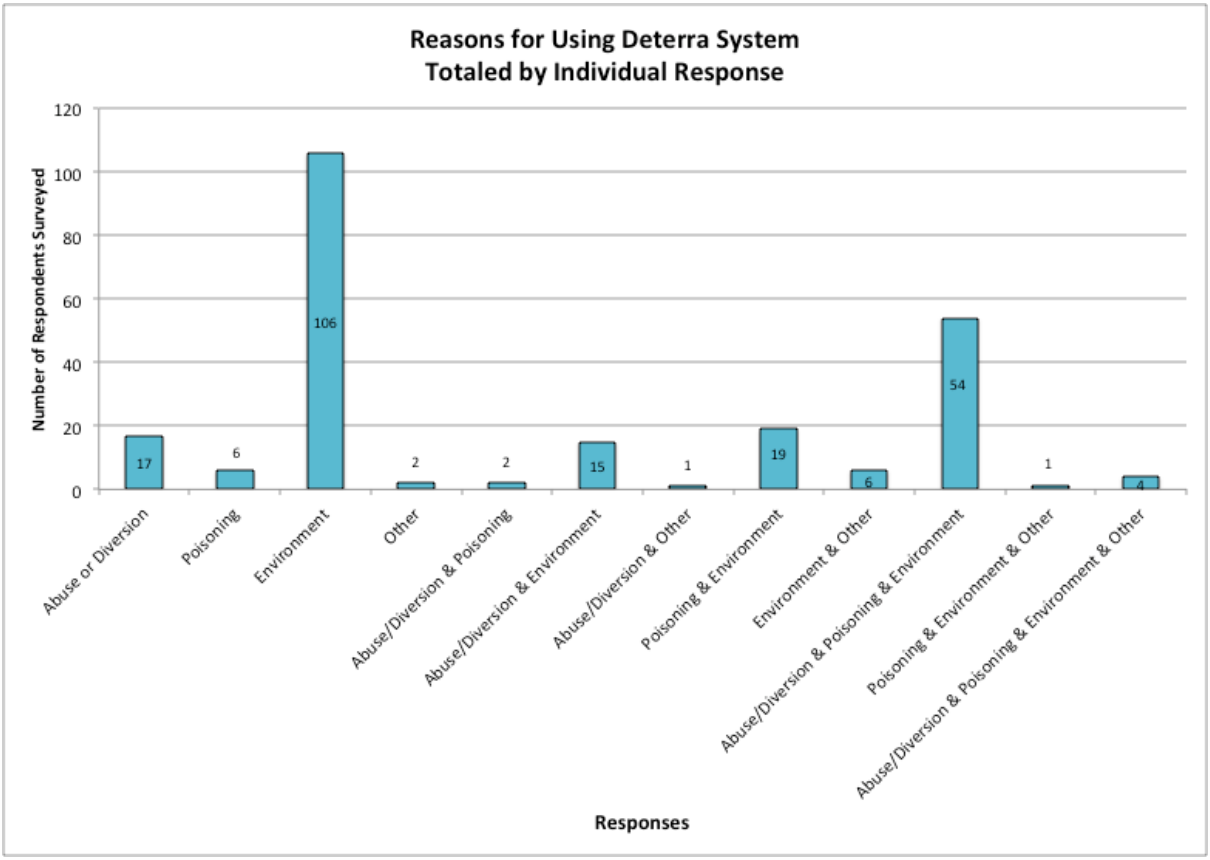
People who used the Deterra pouches to safely dispose of their unused or unwanted medications were largely enthusiastic about the product, most often citing environmental concerns as their reason for using the product and sometimes providing feedback on what would make the product better for them.

Why Did You Use This Product?

In Question 7, survey respondents were asked why they used the product and were offered 4 different answer options:

1. To lower the risk of abuse or diversion
2. To lower the risk of accidental poisoning
3. To remove prescription drugs from my home without causing environmental damage
4. Other (please specify)

The chart below shows the variety of response combinations received. The category that received more responses than any other is “Environment” (the desire to not cause environmental damage). The next most popular answer was a combination of the first three answers, including lowering the risk of abuse or diversion, lowering the risk of accidental poisoning, and preventing environmental damage.

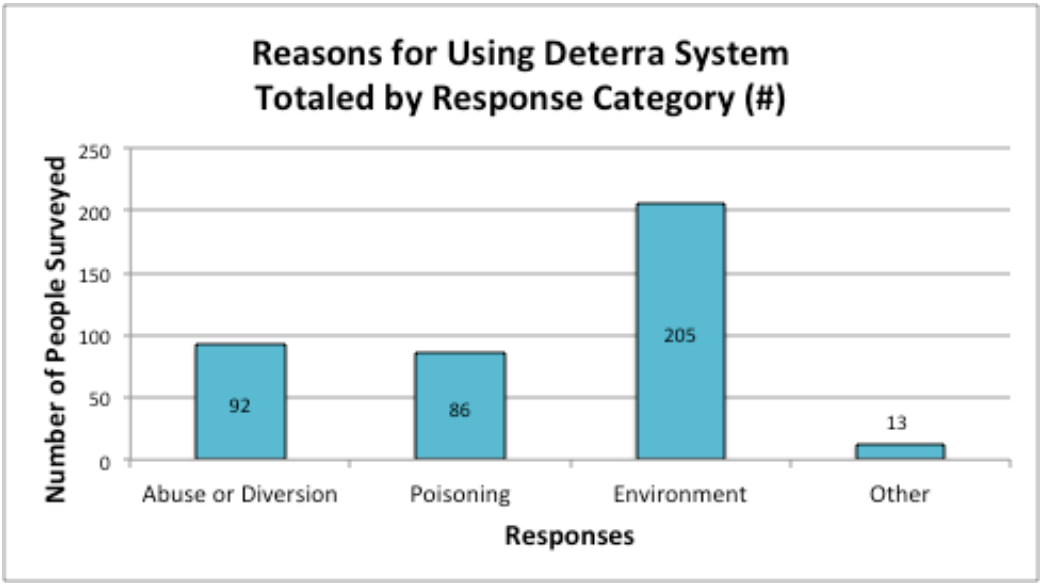


106 respondents—45%—used the product to remove drugs from home without causing environmental damage.

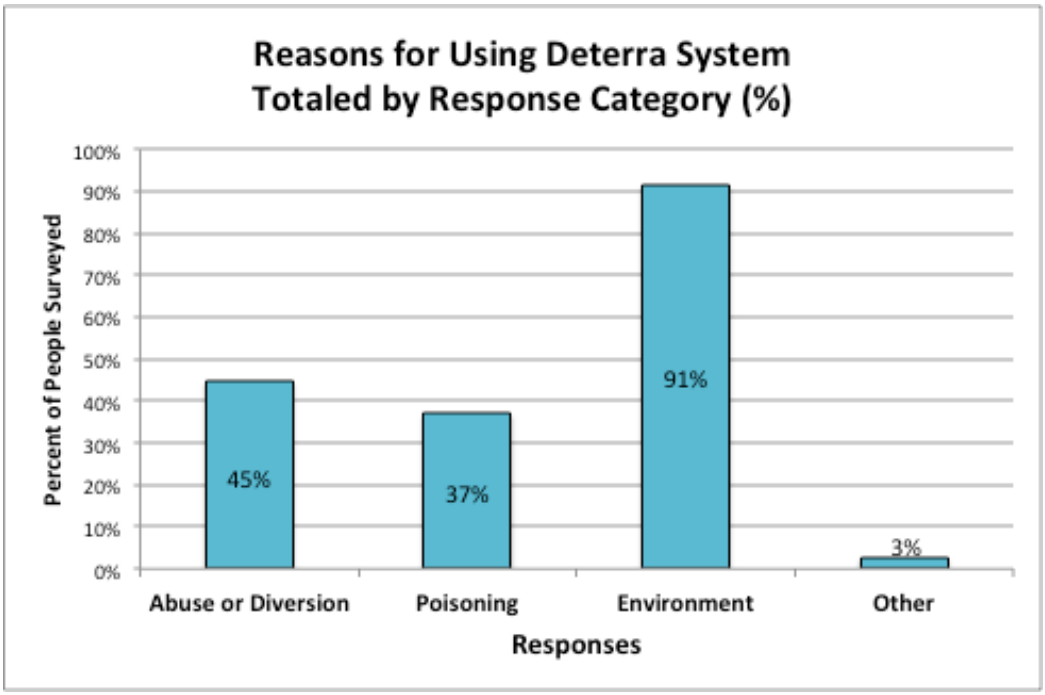
54 respondents—23%—used the product to 1) lower the risk of abuse or diversion, 2) lower the risk of accidental poisoning, **and** 3) remove drugs from home without causing environmental damage.

17 respondents received the product to lower the risk of abuse or diversion. 6 people received the product to lower the risk of accidental poisoning.

When responses are totaled by answer category rather than individual response, we can see (below) that twice as many respondents—205—cite concern for the environment over concern about abuse or diversion (92 respondents) or concern about accidental poisoning (86 respondents).



Sorting the same data by response category, we see that 91% of all respondents used Deterra out of concern for the environment, with 45% citing concern about abuse or diversion, and 37% citing concern about accidental poisoning.



Thirteen people used the product for other reasons and many made comments:

- "To make family aware to NOT put unwanted drugs in the garbage bin!"
- "Because of wife's various medical problems—her meds are constantly changing."
- "Expired medications disposal."
- "Easier than waiting for police times."
- "Convenient."
- "Easy."
- "Free!"
- "To keep meds out of ground water."
- "To help keep drinking water and air cleaner."
- "Closest disposal at county is far away."

Conclusions

1. Consumers are very much aware of the environmental impact of improper disposal of medications.
2. Some consumers are exploring options for drug disposal, including Take-Back programs.
3. Some consumers may face prescription confusion and want to weed out unwanted drugs.
4. There is much work to be done building awareness of the danger and likelihood of abuse, diversion or accidental poisoning.

Why Did You Purchase or Receive This Product?

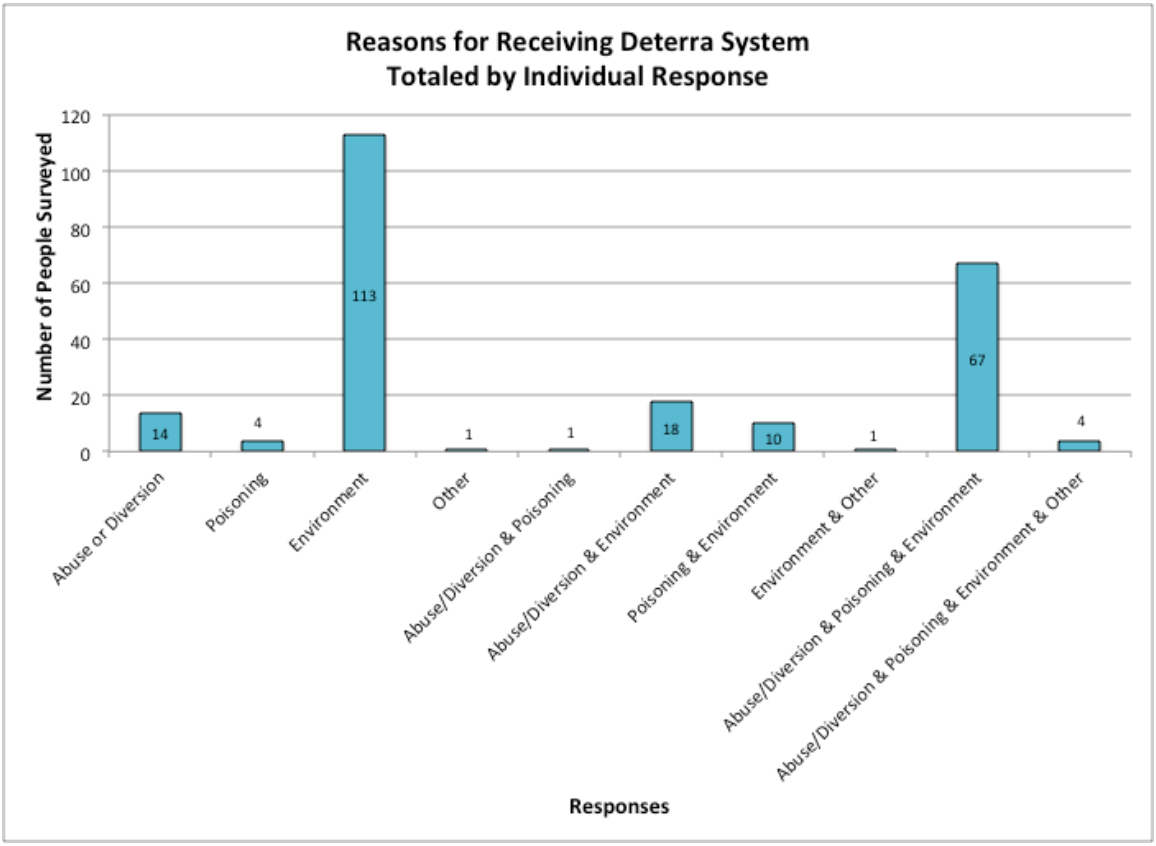
In Question 6, respondents were asked why they purchased or received the product. The phrasing of the question is a bit vague. If a consumer received the product at no charge, is the question then asking about the provider's values? (For instance, was this product offered to me because law enforcement cares about the accidental poisoning?) If so, what is the relevance? It is also unclear how the intent of the question is different from that of Question 7.

The same answer options were provided:

1. To lower the risk of abuse or diversion
2. To lower the risk of accidental poisoning
3. To remove prescription drugs from my home without causing environmental damage
4. Other (please specify)

Most consumers answered this question in the same way they answered Question 7, but there was a slight variance. When looking at the paper surveys themselves, most people simply duplicated their answers from one question to the next.

The chart below shows the variety of response combinations received. Similar to Question 7, the category that received more responses than any other is “Environment” (the desire not to cause environmental damage). Again, the next most popular answer was a combination of the first three answers, including lowering the risk of abuse or diversion, lowering the risk of accidental poisoning, and preventing environmental damage.

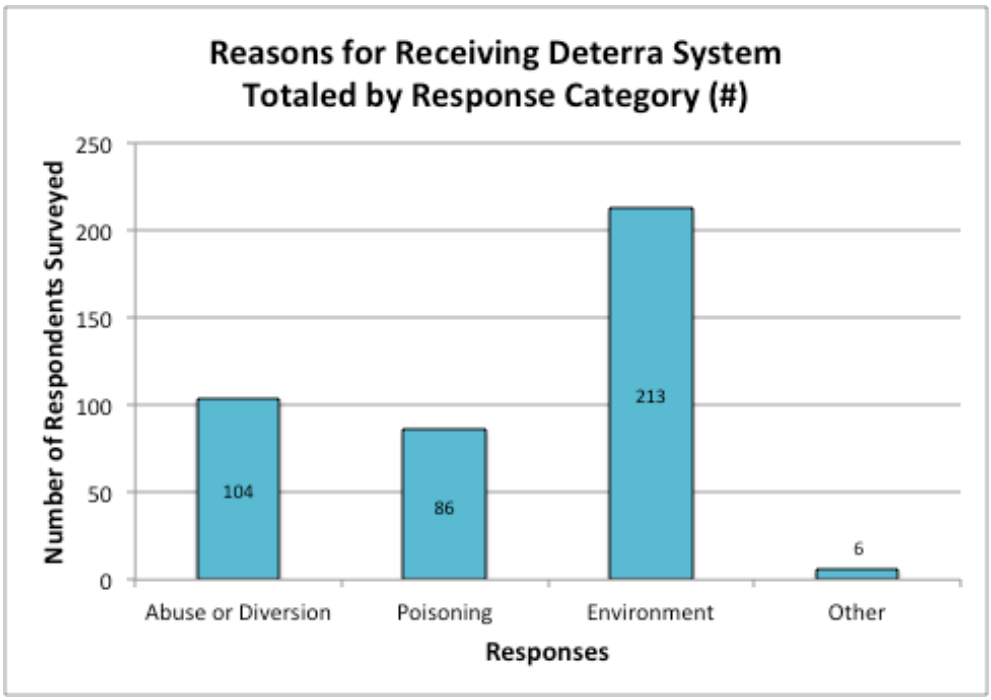


113 people - 48% of respondents - say they received the product for the sole purpose of removing drugs from home without causing environmental damage.

67 people—29% of respondents—say they received the product to 1) lower the risk of abuse or diversion, 2) lower the risk of accidental poisoning, **and** 3) remove drugs from home without causing environmental damage.

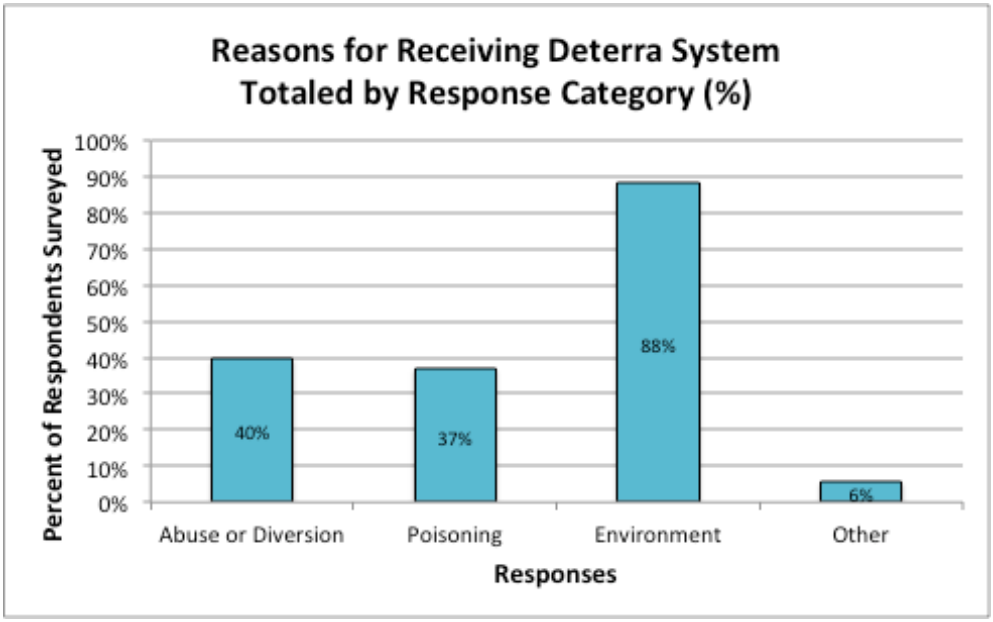
14 people say they received the product to lower the risk of abuse or diversion. 4 people received the product solely to lower the risk of accidental poisoning.

When responses were totaled by answer category rather than by individual response, we again see that 213 respondents believe they received Deterra out of concern for the environment.



Concern for the environment was two times more significant than concern about abuse or diversion (104 respondents) or concern about accidental poisoning (86 respondents).

The chart below shows response by category for the same data, but as a percent of all respondents. Of the survey respondents, 88% cited the environment, 40% cited concern for abuse or diversion, and 37% cited accidental poisoning as a reason they received the product.



Five people left comments in the "Other" category regarding their reasons for receiving the product:

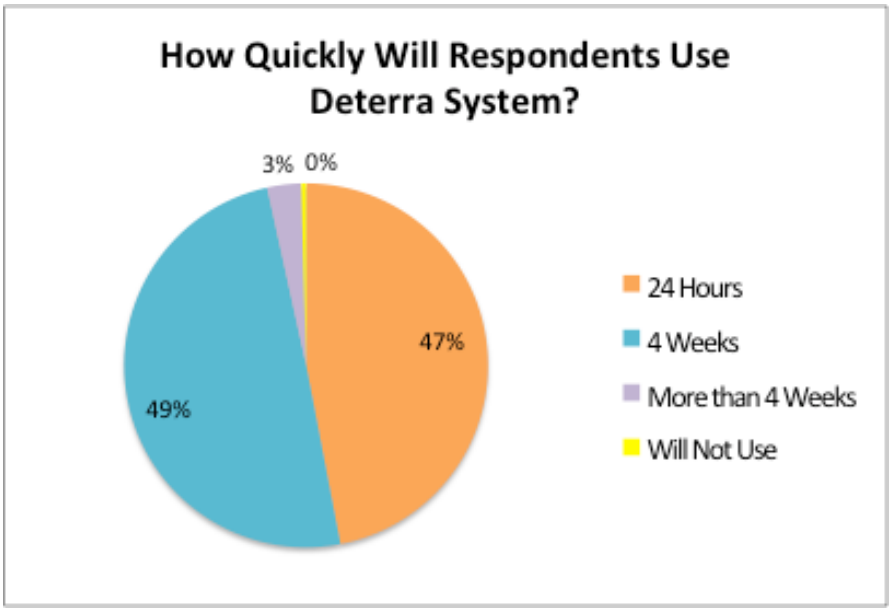
- "Facility is a pilot pharmacy."
- "To keep medications out of ground water."
- "To help keep drinking water and air cleaner."
- "I have witnessed drug prescription abuse."
- "It is difficult to dispose of medicine properly. This makes it easy!"
- "I want to try the product before introducing it to peers and clients."

Conclusions

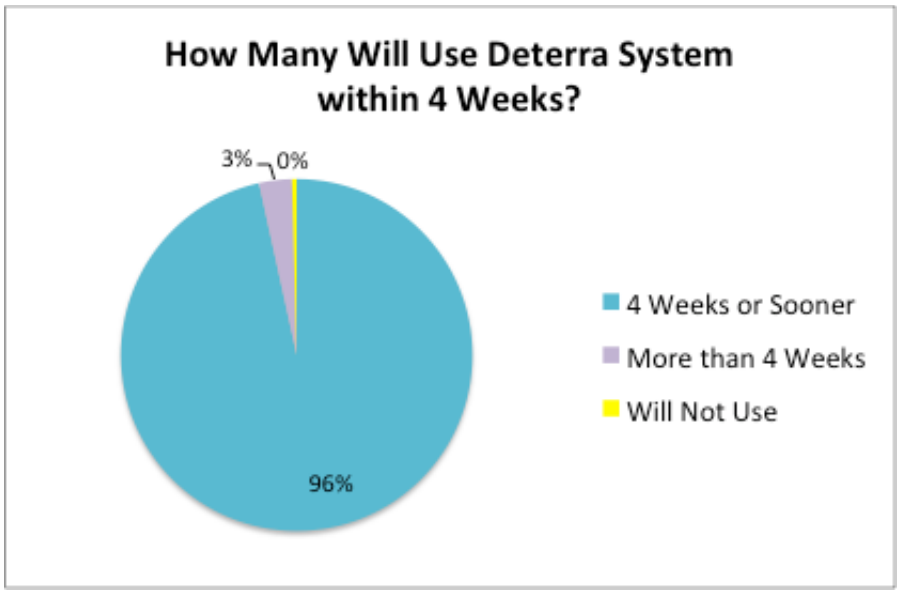
1. Consumers believe that organizations like law enforcement and businesses like pharmacies are providing products to help protect the environment.
2. Again, we see less awareness of the threat of drug abuse and diversion or accidental poisoning.

Upon Receipt of the Product, How Quickly Did You Use It?

Answers to Question 3—how soon did you use the Deterra System?—show us that 49% of consumers used the Deterra System within 24 hours of receiving it, and 47% used it within 4 weeks. A scant 3% will take longer than a month to use the product.



That means that 96% of consumers used the Deterra System in 4 weeks or less.



Only one person indicated that he/she would not use the product and instead preferred the DEA Take-Back program.

Conclusions

1. When consumers became aware of the Deterra System, they were enthusiastic and used the product almost immediately.

Using the Pouch

Question 4 asks consumers if the instructions were clear and easy to understand. Ninety-seven percent of respondents found the instructions clear and easy to understand. Three percent—8 people—did not. Their comments are about the small pouch of activated carbon within the larger pouch:

- "'Pouch' not clearly identified."
- "What do we do with the inside bag?"
- "You need to make it clear in large letters that you don't open the inside pocket. Does it dissolve or what?"
- "Didn't know what to do with the inside bag—finally noticed note."
- "Note about not opening the inside bag needs to be more prominent."
- "If you can get it to indicate that it was successful you would have a fantastic product!"[1]

[1] This comment was made on Question 7, but pertains to use of product.

Question 5 asks consumers if they had any trouble using the product. Nearly all respondents had no problems using the product. Of the 5%—11 people—who did, here's what they commented:

- "'Pouch' not clearly identified." (Confusion between larger pouch and inner pouch.)
- "A little difficult to see where to tear open package."
- "After tearing tab off, it was a little difficult to pull apart."
- "Tore unevenly and spilled."
- "Didn't read directions first."
- "Trouble sealing the bag—eventually did."
- "Need to make clear in large letters that you don't open inside pocket (does it dissolve or what?)"
- "What do we do with the inside bag?" (2 responses)
- "Some of the material did get on the seal, but I removed it so it would seal."
- "I think I added too much water—couldn't easily seal the pouch. Messy!"
- "Did not seal well, need to improve sealing mechanism."

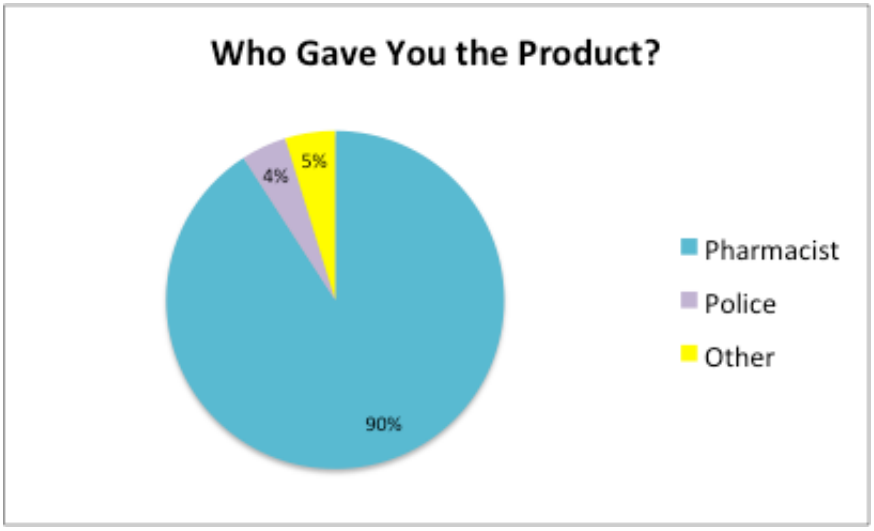
Conclusions

1. If 8 people did not immediately understand what to do with the smaller pouch within the larger pouch, it's quite likely that many more had or will have the same question. The language or pictograph on packaging needs to be more explicit or prominent.
2. Tearing the package open and resealing the bag were problematic for some. Again, this may well be true for many more consumers.
3. Online FAQ should include troubleshooting tips when consumers add too much water, spill the carbon, etc.

Who Gave You the Product?

Of the 233 User Surveys received, 90% were supplied by pharmacies[2], 4% by law enforcement agencies, and 5% by "other," typically co-workers or acquaintances. 3 respondents (1%) did not answer the question.

[2] On surveys from pharmacies where respondents selected "other" and then entered the words "pharmacy technician" or "cashier" or "store display," their answer was categorized in the general category of "pharmacy" and not "other."



Conclusions

None.

Did you purchase the product or was it given to you?

We know that all pouches were given free of charge, and yet 6 of 233 people marked that they had purchased the product. This result hopefully can be attributed to mis-reading of the question.

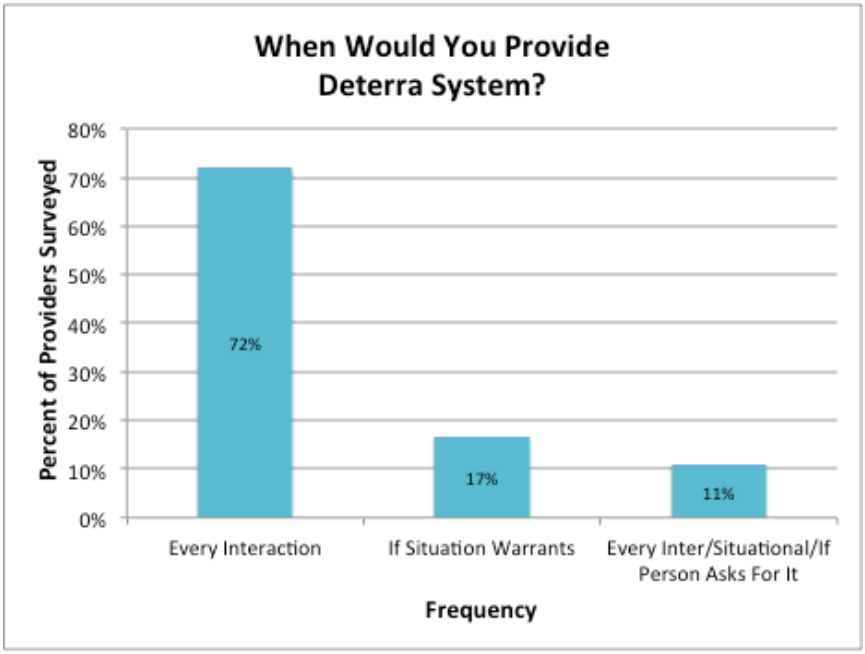
Conclusions

None.

Provider Response to Deterra System

1. When would you provide the product to client/customers/public?

Of the 18 respondents, 13 said they would provide it with every interaction. These responses may be from pharmacists who, for purposes of securing survey responses, were incentivized to provide the pouches with every interaction at no cost to themselves. Therefore, this data may be biased. One provider said he/she would provide it when there was a medication or dose change.



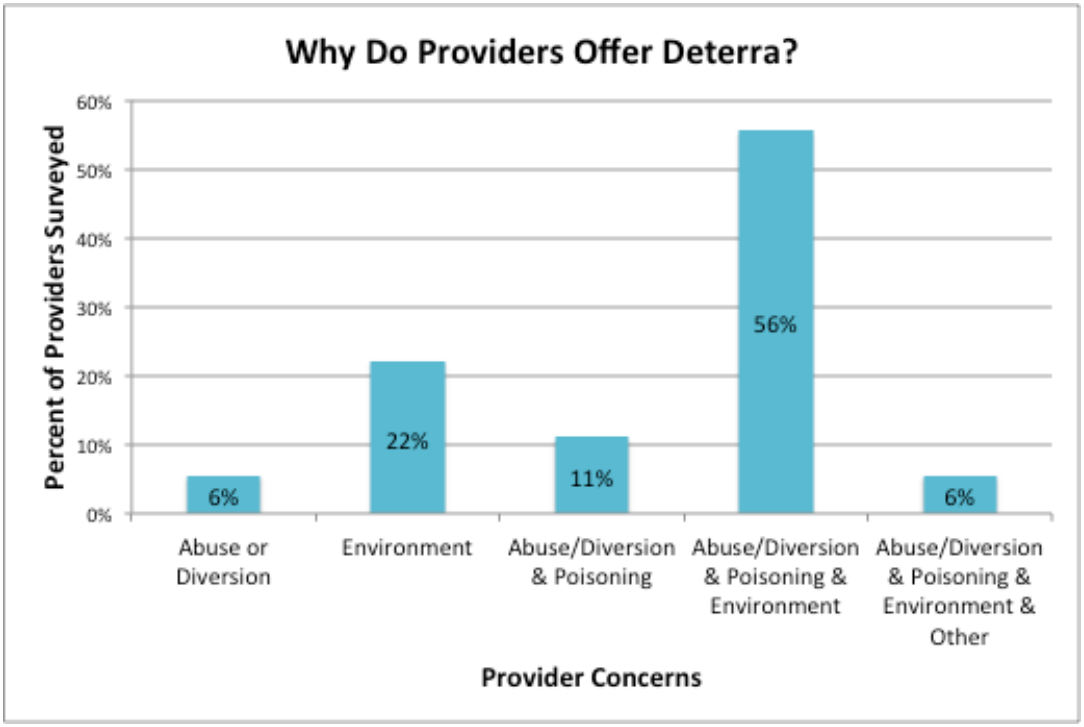
2. Do you have any suggestions for product improvement?

100% said no.[3]

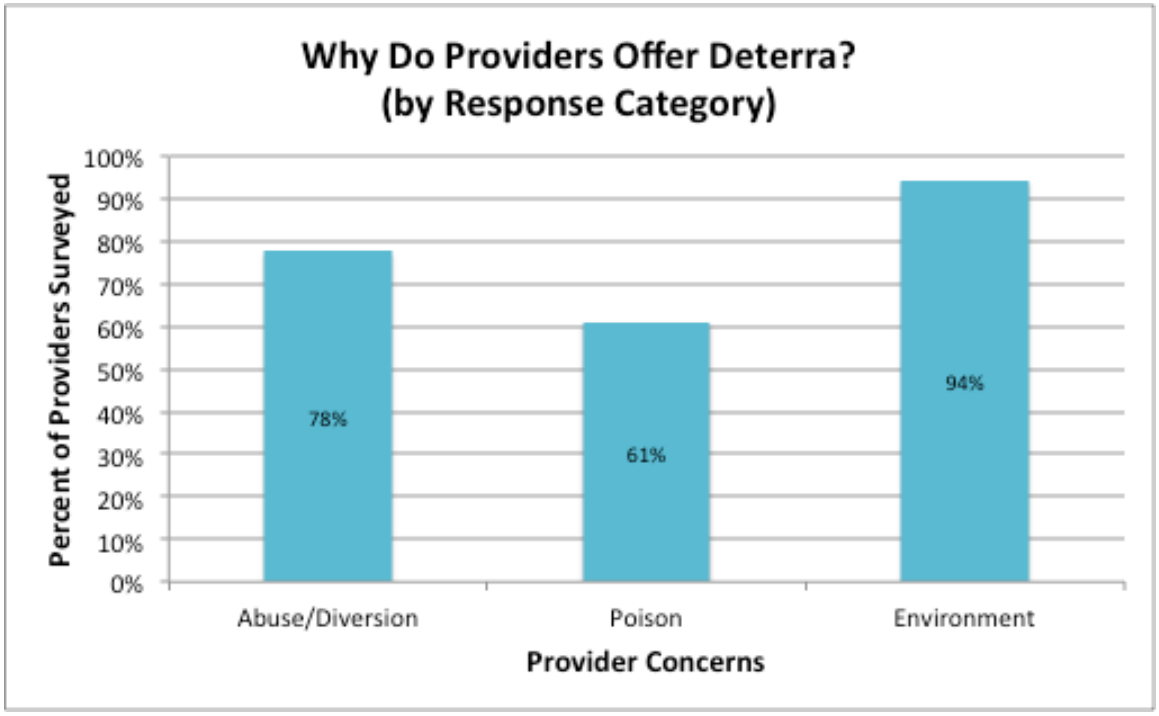
[3] Two respondents suggested, "the envelope be attached to the pouch," but since this refers to the survey and not the product, their answers were interpreted to mean that they did not have suggestions for product improvement.

3. Why did you provide this product?

56% of respondents indicated to lower the risk of abuse or diversion, to lower the risk of accidental poisoning, **and** to remove drugs from homes without causing environmental damage. 22% indicated concern primarily for the environment.



When responses were totaled by answer category rather than individual respondent, we saw that providers' concerns were somewhat more balanced between Abuse/Diversion (14 people), Poison (11 people), and Environment (17 people). Environment still led, but only by 16% over Abuse/Diversion and 33% over Poison.



We received one specific comment in the category of Other: "To prevent someone from trying to self-medicate for another illness, event/injury other than what it (the medication) was originally prescribed."

4. Do you plan to continue to provide this product?

100% said yes. One comment promised to carry the product if it were free of charge.

5. Are you aware of any complaints or adverse events that persons have experienced using this product?

94%—17 of 18 people—said no, and one person indicated a customer complained of having difficulty resealing the package.

Survey Distribution Strategy

The biggest uncertainty in survey distribution lay with the providers. We used a mix of pharmacy staff, law enforcement and a mishmash of other interested parties to help us reach consumers. We provided brief training and signage to help them better communicate.

Pharmacies. Because Deterra has strong support from Hooshang Shanehsaz, the Director of Pharmacy at Cardinal Health, and Mr. Shanehsaz offered to help in the survey effort, 950 Deterra surveys and pouch samples were distributed to 3 pharmacies in Delaware, Mr. Shanehsaz' home state. By the end of April, staff from the three pharmacies participated in brief training via conference call or pre-recorded training on Power Point. Every member of the pharmacy team from each store won a VISA gift card. The amount on the card was determined by which store delivered the most surveys by June 10, 2016. Specifically:

1st place Atlantic Apothecary: all members of pharmacy staff won $100

2nd place Cape Pharmacy: all members of pharmacy staff won $75

3rd place Bayard Pharmacy: all members of pharmacy staff won $50

Law Enforcement. Two of Deterra's largest customers and supporters are the Hennepin County Sheriff's Office and the Eden Prairie Police. Both organizations agreed to pass out User Surveys along with Deterra packets.

The Eden Prairie Police distributes Deterra pouches to people who come into the station seeking a place to dispose of their unused medications. They distribute surveys with the packets from their reception desk in their front entry.

The Hennepin County Sheriff office distributes Deterra pouches to people who attend their Town Hall meetings across Hennepin County.

Various. It seemed likely that we would come across more people who wanted to take the survey, so we created the 6th grouping of User Surveys, DSU6, or Various. We distributed 65 surveys to a variety of people, including pharmacists who were unable to distribute surveys at their place of business, members of the 3M First Response Team, and other interested parties.

Conclusions

1. Providers are generally enthusiastic about the Deterra System.

2. Similar to consumers, providers show the greatest concern for the environment, though a significantly higher percentage of them also cite concern for abuse/diversion and/or accidental poisoning.

3. When targeting new markets, it may be wise to understand the opportunities providers have for offering the Deterra System and the potential frequency. Specifically, how many touch points are there with consumers?

Technical Objective 3:

Summary Report Providing Effectiveness Results From Analysis Of A Range Of 20 Different Psychoactive Compounds

The effectiveness of the activated carbon system was tested for individual drugs according to a protocol to determine the degree of drug deactivation over a 28 day period. Intermittent samples were taken to plot the deactivation curves.

At the end of the 28 day period the carbon mixture was subjected to a washout procedure to determine the extent of leaching of drug from carbon by water, or ethanol. The Protocol procedures are shown below:

Deactivation & Desorption Protocol

Rate and Extent of Adsorption

- Placed 10 tablets, 10 suboxone sublingual films, 2 patches or 1 vial of liquid in disposal pouches
- Added 50 grams warm tap water (110° ± 10°F) to pouches containing tabs/films/patches. For liquid medications, brought final volume to 50 ml with warm tap water
- Following addition of water waited 30 seconds then seal pouches
- Pouches were shaken 5 seconds at rate of 1 shake per second
- Ensured medications were at bottom of pouches
- Stored upright, undisturbed at room temperature until tested
- Tested duplicate pouches at 8 hours, days 1, 2, 4, 7, 14, 21, 28

Washout (desorption)

- Used 28 day pouches
- Transferred pouch contents to 500 ml bottles
- Added 200 ml water (this water may be used to wash and transfer pouch contents to bottle)
- Rocked for 1 hour (~30 cycles per minute)
- Analyzed samples after an additional 23 hours exposure (total 24 hours)
- Replaced water with 250 ml of 30% (v/v) ethanol
- Rocked for 1 hour (~30 cycles per minute)
- Analyzed samples after an additional 23 hours exposure (total 24 hours)

Note on Results:
Verde and Mercer University independently evaluated the efficacy of the activated carbon system in drug deactivation, however Verde used UV/Vis determinations, while Mercer used specific validated HPLC assays for each active pharmaceutical ingredient (API). Both analytical methods showed a high degree of concurrence. The HPLC data is considered definitive, and is presented in this report. The Verde data will be made available on request. HPLC validation data are provided in Appendix A. Individual HPLC deactivation files are provided in Appendix B. A summary table of Verde UV/Vis results is provided in Appendix C.

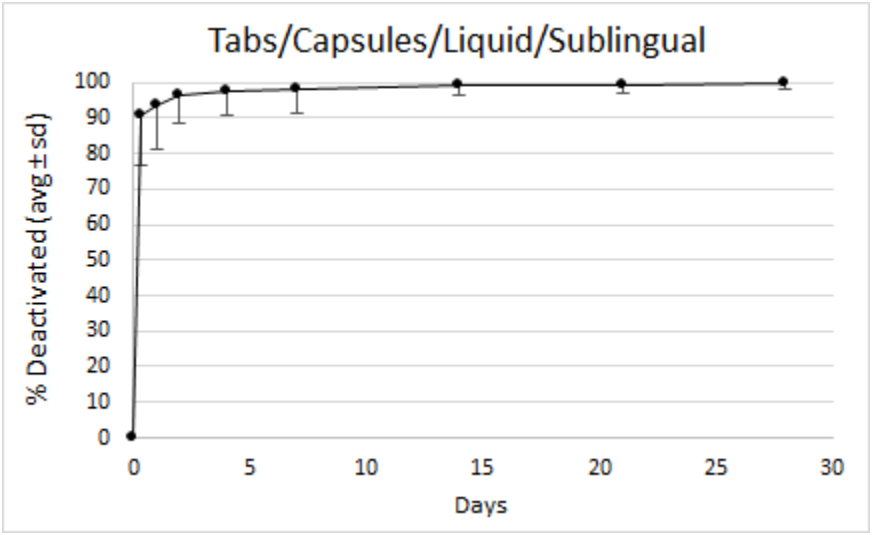
Summary Data:

HPLC Results For Drug Deactivation Over A 28 Day Period.

	Deactivation, Day									Desorption	
	0	0.33	1	2	4	7	14	21	28	Aqueous	EtOH
Alprazalom	0	100	100	100	100	100	100	100	100	0.0	0.0
Buprenorphine	0	96.9	96.0	99.1	99.8	99.9	100	100	100	0.0	0.1
Dextroamphetamine	0	99.9	100	100	100	100	100	100	100	0.0	9.4
Diazepam	0	46.3	50.8	72.1	73.4	74.2	90.0	94.9	99.3	1.0	1.6
Fentanyl	0	61.5	100	100	100	100	100	100	100	0.0	0.0
Fluoxetine	0	88.4	89.7	91.5	89.7	91.5	95.3	91.0	94.4	0.0	0.7
Hydromorphone	0	100	100	100	100	100	100	100	100	0.0	0.5
Ketamine	0	99.7	99.9	100	100	100	100	100	100	0.0	6.5
Lorazepam	0	70.7	79.1	87.5	96.2	99.8	99.9	99.9	100	0.3	0.3
Loxapine	0	96.9	96.5	99.6	99.6	99.6	100	100	100	0.0	0.0
Meperidine	0	97.9	99.6	99.8	99.9	99.9	100	100	100	0.0	1.4
Methadone	0	97.1	97.2	98.3	99.8	99.9	100	100	100	0.2	0.3
Methylphenidate	0	99.9	100	100	100	100	100	100	100	0.0	1.0
Morphine	0	99.8	99.9	100	100	100	100	100	100	0.0	1.6
Oxycodone*	0	84.3	85.0	86.7	92.1	97.5	100	100	100	0.1	1.6
OxyContin®	0	79.6	96.8	98.9	100	100	100	100	100	0.0	0.2
Quetiapine	0	82.9	84.0	86.4	94.4	97.5	99.9	99.3	99.6	0.0	1.2
Temazepam	0	98.5	98.5	98.5	98.7	98.8	98.6	98.9	99.3	0.0	0.0
Tramadol	0	96.3	99.0	100	100	100	100	100	100	0.0	4.8
Zolpidem	0	85.2	93.7	99.8	100	100	100	100	100	0.0	0.2
Average	0.0	89.1	93.3	95.9	97.2	97.9	99.2	99.2	99.6	0.1	1.6
SD	0	14.9	11.8	7.4	6.3	5.9	2.4	2.2	1.3	0.2	2.5

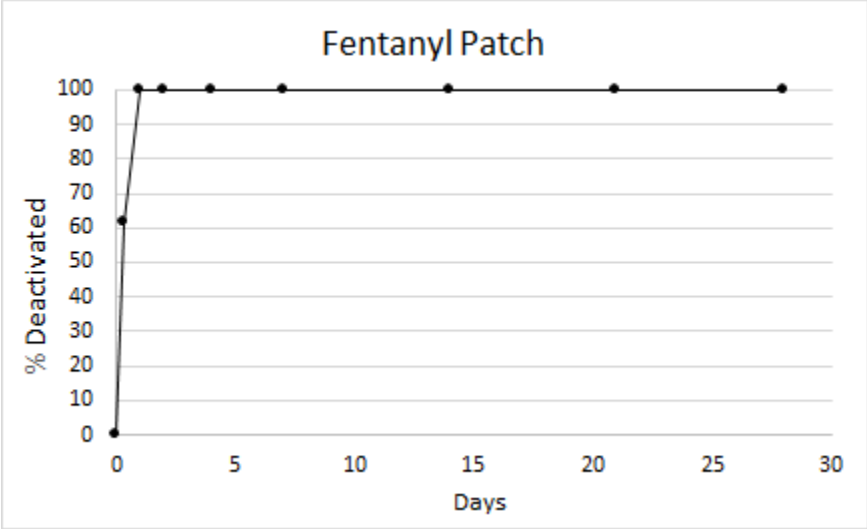
*Coformulated with acetaminophen.
Deactivation data are expressed as percent deactivated.
Washout (desorption) values are expressed as % recovered.

The Figure below illustrates the above data.



Fentanyl Patches were considered a special case, because the patches remained highly adhesive, and required careful handling in order to prevent the patch from sticking to the side of the pouch, or to itself, potentially affecting the deactivation process. The adhesive surface of the patch was covered with a KimWipe tissue, which adhered to the surface and allowed the free transfer of water.

Deactivation of fentanyl occurred rapidly upon release from transdermal patches:



Desorption Studies showed that water was ineffective in leaching out more than trace amounts of the API's tested.

With 30 % ethanol extraction, on average only 1.6% of the API's were recovered, however there were several agents where up to 9% of the starting dose could be recovered by this process. Considering the cost of ethanol and the time and equipment required, this method appears to be impractical for drug recovery procedures, particularly for unknown and potentially comingled drug contents.

Conclusions:

The activated carbon was highly effective in adsorbing all of the drugs tested. Data showed that an average of 89% of pharmaceuticals were adsorbed by activated carbon within the first 8 hours, and only trace amounts could be detected at 28 days.

Activated carbon was highly effective for all formulations tested, including the fentanyl transdermal patch.

Activated carbon was highly effective in deactivating all chemical classes of drugs tested over the test period. The mean deactivation of all products tested was 99.6% at day 28.

The rate of drug inactivation was relatively independent of the chemical compositions tested. Although some formulations such as liquids and sublingual dosage forms were inactivated very rapidly (>90% in 8 hours), some required several days to complete deactivation.

The adsorbed pharmaceuticals were resistant to leaching by water, with only trace amounts detectable after an extensive washout. This demonstrates the long-term effective adsorption and sequestration from the environment of the adsorbed and deactivated pharmaceutical.

The adsorbed pharmaceuticals were resistant to leaching by a washout procedure with 30% ethanol, with few exceptions. This demonstrated that the activated carbon was effective in rendering adsorbed pharmaceuticals unrecoverable by simple means.

Technical Objective 4:

Conduct Surveys to determine adverse events from the use of the product

During the reporting period several thousand pouches were distributed in the Survey, and used in deactivation studies at Verde and Mercer University, The product was also commercially available and distributed nationally.

There were no reports of product complaints and no adverse events have been reported from any source in the testing period.

Technical Objective 5:

Determine Durability Or Shelf Life Of The Product

Two studies were performed at Verde to examine stability of the product under accelerated aging conditions.

Study 1 utilized two medications examined under technical objective 3 in this contract: diazepam (10 mg tablets) and methylphenidate HCl (20 mg tablets). At each time point 10 tablets were placed in separate disposal pouches.

Study 2 utilized higher doses of over-the-counter medications in order to determine if carbon capacity diminishes over time. At each time point six tablets of acetaminophen (500 mg) or 16 tablets of naproxen (220 mg) were placed in separate disposal pouches. This resulted in a total dose of 3.5 grams API per pouch.

Accelerated Stability Protocol

- Pouches were stored at room temperature, 40°C and 50°C
- At each time point tablets were added to pouches (duplicate pouches for each medication and each temperature condition)

- 50 grams warm tap water (110° ± 10°F) was added to each pouch
- After 30 seconds pouches were sealed and placed on a rocker at room temperature
- Pouches were analyzed for drug content after 1-2 weeks

Summary Data:

Diazepam

Weeks	RT	40C	50C
0	99.9	NA	NA
8	99.7	100	100
18	100	100	100
28	100	100	100
52	100	100	100
78	100	100	100

Methylphenidate

Weeks	RT	40C	50C
0	100	NA	NA
8	100	100	100
18	100	100	100
28	100	100	100
52	100	100	100
78	100	100	100

Acetaminophen

Weeks	RT	40C	50C
0	99.2	NA	NA
8	99.6	99.7	99.7
18	99.5	99.6	99.6
28	99.6	99.5	99.7
52	99.7	99.7	99.7
78	99.7	99.9	99.9

Naproxen

Weeks	RT	40C	50C
0	99.0	NA	NA
8	100	100	99
18	99.5	99.3	99.5
28	100	100	100
52	99.4	99.3	99.5
78	99.4	99.4	99.3

Data are expressed as % deactivated

Conclusions:

Data predict a stable shelf life in excess of 10 years under room temperature condition with no deterioration in carbon capacity.

Technical Objective 6:

Development of ancillary materials to support adoption and consistent use.

Based on Survey results, product information and pouch labeling are being reviewed for revision for increased clarity of instruction. Website and product information will also be updated online.

Publications

Y. Song, M. Manian , W. Fowler, A. Korey , and A. K. Banga, Activated Carbon Based System for Disposal of Psychoactive Medications, submitted.

X. Gao, P. Bakshi, S. Ganti, M. Manian, W. Fowler, A. Korey , and A. K. Banga, Evaluation of a unique activated carbon based deactivation system for the disposal of highly abused opioids medications, manuscript in preparation

S. Ganti, X. Gao, W. Fowler, A. Korey , and A. K. Banga, Deactivation and disposal of immediate and extended release oxycodone oral dosage forms, manuscript in preparation

B. Bozorg, W. Fowler, A. Korey , and A. K. Banga, Deactivation of model CNS depressant medications using an activated carbon disposal system, manuscript in preparation.

Y. Kim, P. Bakshi, B. Bozorg, W. Fowler, A. Korey , and A. K. Banga, Activated carbon based disposal of model CNS active drugs, manuscript in preparation.

P. Bakshi, W. Fowler, . Korey , and A. K. Banga, Deactivation of fentanyl transdermal patches using activated carbon, manuscript in preparation.

P. Bakshi, W. Fowler, A. Korey , and A. K. Banga, Determination of loxapine and methylphenidate by reverse phase high performance liquid chromatography and their deactivation by activated carbon, manuscript in preparation.

Appendix A
Deactivation Data
(Mercer University, HPLC)

Alprazolam Deactivation Protocol

Alprazolam

Date of report: 01/05/16

Protocol:

1) 10 tablets of Alprazolam were placed in a pouch
2) 50ml warm water (43 °C) was added to the pouch and was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Washout Protocol:

1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken
3) Water was replaced with 250 mL of 30%(v/v) ethanol
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken

Filtration of samples:

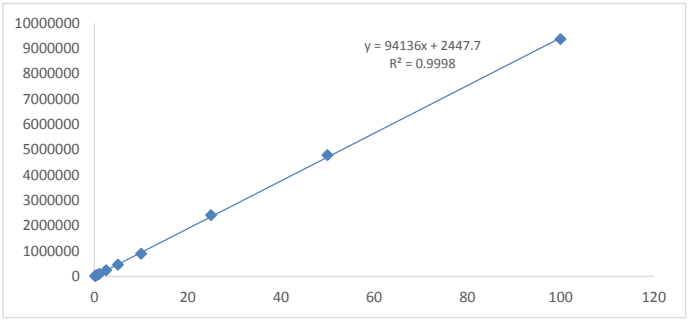
Pouch was shaken before taking the samples

1) 1ml of sample was taken by a syringe and centrifuged for 3 minutes 13400 rpm
2) The supernatant was filtered through 0.22 µm syringe filter
3) Filtered samples was analyzed using HPLC.

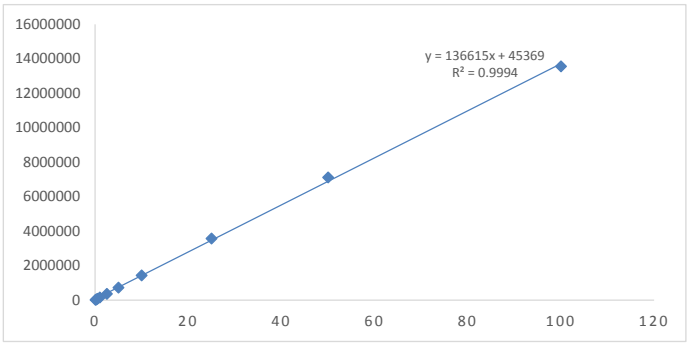
Chromatographic conditions: Aplrazolam
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN:Buffer (KH2PO4 0.01M pH 4.5) (40:60)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 4.7min
Column: (size and particle size): Phenomenex, Kinetex C18 100A, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 221 nm

Alprazolam Deactivation Standard Curve

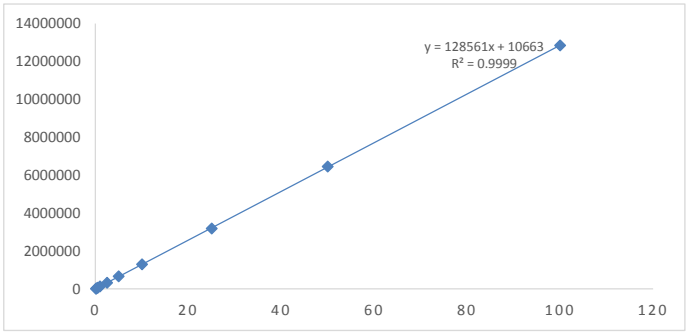
Standard curve for Alprazolam - 8hr to Day 14	
Concentration (µg/ml)	Area
0.1	8266
0.25	22398
0.5	44506
1	92452
2.5	239121
5	454905
10	890064
25	2411735
50	4786605
100	9369668



Standard curve for Alprazolam - Day 21	
Concentration (µg/ml)	Area
0.1	14025
0.25	34965
0.5	76251
1	144124
2.5	353056
5	716842
10	1422700
25	3564904
50	7116035
100	13561883



Standard curve for Alprazolam - Day 28 to washout studies	
Concentration (µg/ml)	Area
0.1	16021
0.25	35782
0.5	74492
1	135656
2.5	333558
5	672156
10	1305948
25	3198121
50	6459634
100	12861015



Alprazolam Deactivation/Desorption

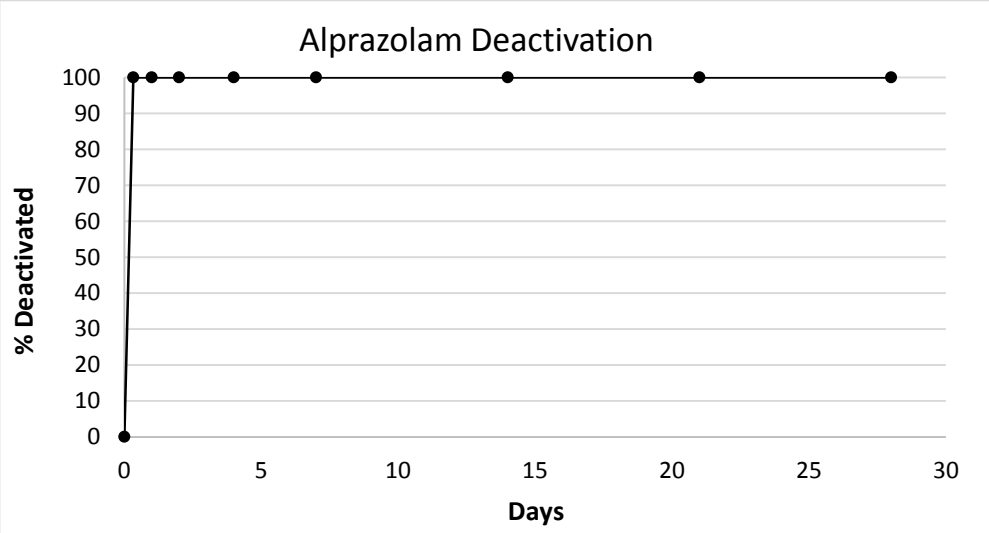
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
8hr	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 1	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 2	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 2	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 4	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 4	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 7	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 7	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 14	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 14	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 21	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 28	n-2	0.00	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (µg/ml)	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout- H2O	n-1	0.00	0.00	0.00	0.00	100.00	100.00
Day 29 Washout- H2O	n-2	0.00	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc (µg/ml)	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- ethanol	n-1	0.00	0.00	0.00	0.00	0.00	0.00
Day 30Washout- ethanol	n-2	0.00	0.00	0.00	0.00	0.00	

Total amount of drug added to the pouch-20mg

Alprazolam Deactivation - Mercer

2 mg Tablets



Days	% Deactivated
0	0
0.33	100
1	100
2	100
4	100
7	100
14	100
21	100
28	100

Buprenorphine Deactivation Protocol

Protocol

Buprenorphine adsorption Study

1) 10 Suboxone films were placed in pouch.
2) 50ml warm tap water (43 degree celcius) was added to the pouch and wait for 30 seconds.
3) Pouch was sealed and gently shaken from side to side
4) It was placed in upright position and take samples were taken at 8hour, 1, 2, 4, 7, 14, 21, 28days time points.

Filtration of Samples:

Pouch was shaken properly before taking samples.
1) Sample dilution
   a. 0.1ml sample was taken out from pouch and diluted to 16ml (8h), 5ml (1Day) with distilled water.
2) 500ul of sample was filtered using 0.22 micron syringe.
3) Samples were analyzed with HPLC.

Desorption Study

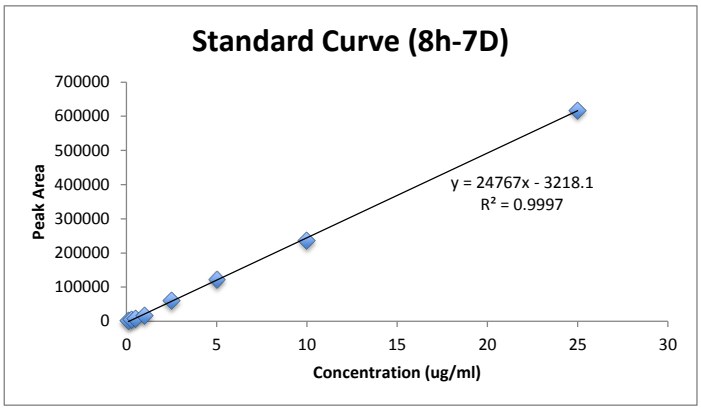
1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml DI water was added to 500ml bottles
4) Samples were rocked on shaker for 1 hour (50rpm)
5) Samples were analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50rpm)
8) Samples were analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

Chromatographic Conditions:

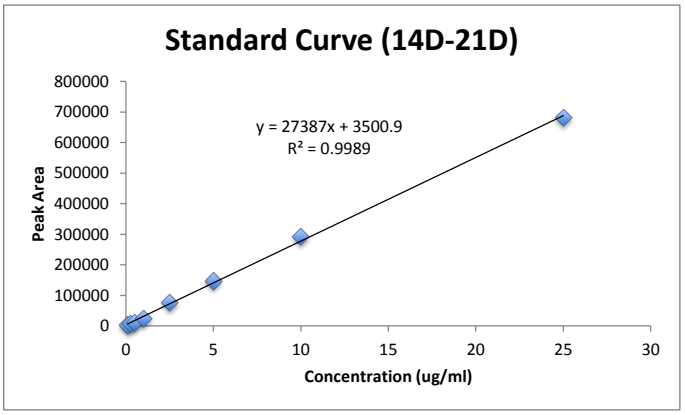
System: HPLC
Mobile Phase: ACN (83%), pH6 phosphate buffer (17%)
Flow rate: 1.0ml/min
Run time: 10min
Retention time: ~3.5min
Column: Kinetex 5u EVO C18 100A, New Column 150x4.6mm
Temperature: 30° C
Detection wavelength: 212nm

Buprenorphine Deactivation Standard Curve

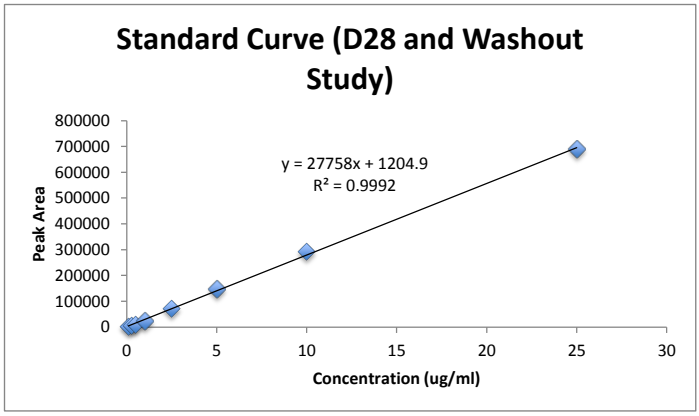
Standard Curve (8h-7D)	
Concentration(ug/ml)	Peak Area
0.1	1125
0.25	4928
0.5	9754
1	17959
2.5	60125
5	123080
10	237497
25	618219



Standard Curve (14D-21D)	
Concentration(ug/ml)	Peak Area
0.1	2721
0.25	6782
0.5	11946
1	24918
2.5	76298
5	147128
10	291800
25	681024



Standard Curve (28D and Desorption Study)	
Concentration(ug/ml)	Peak Area
0.1	2591
0.25	6590
0.5	10112
1	23102
2.5	70124
5	148119
10	291029
25	689025



Buprenorphine Deactivation

Time(days)	Pouch	Peak Area	Conc(ug/ml)	Dilution Factor	Original Conc(ug/ml)	Amount of drug in 50 ml(mg)	% Reacted	Avg %
0.33	1	4167	0.30	160	47.71	2.39	97.02	96.87
	2	4878	0.33	160	52.30	2.62	96.73	
1	1	27918	1.26	50	62.86	3.14	96.07	96.04
	2	28354	1.27	50	63.74	3.19	96.02	
2	1	4613	0.32	50	15.81	0.79	99.01	99.11
	2	3098	0.26	50	12.75	0.64	99.20	
4	1	1489	0.19	20	3.80	0.19	99.76	99.77
	2	1365	0.19	20	3.70	0.19	99.77	
7	1	1203	0.18	10	1.79	0.09	99.89	99.89
	2	1145	0.18	10	1.76	0.09	99.89	
14	1	23602	0.73	1	0.73	0.04	99.95	99.95
	2	26369	0.83	1	0.83	0.04	99.95	
21	1	12316	0.32	1	0.32	0.02	99.98	99.99
	2	7080	0.13	1	0.13	0.01	99.99	
28	1	3029	0.07	1	0.07	0.00	100.00	100.00
	2	2811	0.06	1	0.06	0.00	100.00	

Original amount of drug in the pouch=80mg

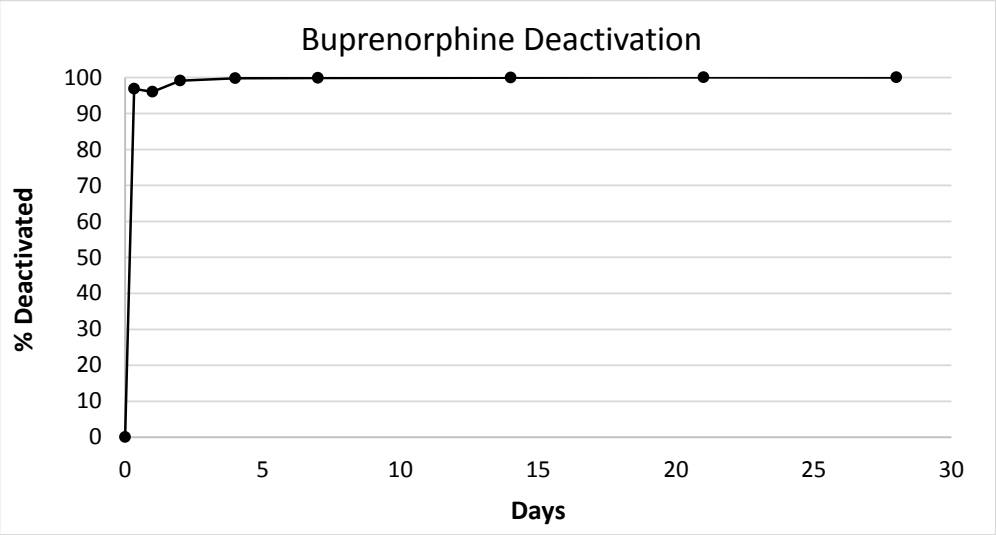
Buprenorphine Desorption

Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average
Day 29 Washout- H2O	1	3011.00	0.07	16.27	0.02	99.98	99.98
Day 29 Washout- H2O	2	3189.00	0.07	17.87	0.02	99.98	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- 30% ethanol	1	3390.00	0.08	19.68	0.08	0.08	0.09
Day 30 Washout- 30% ethanol	2	4091.00	0.10	25.99	0.10	0.10	

Medication	% leached in 30% ethanol
Buprenorphine	0.09

Buprenorphine Deactivation - Mercer

2 mg Sublingual Film (also contains 8 mg Naloxone)



Days	% Deactivated
0	0
0.33	96.9
1	96.0
2	99.1
4	99.8
7	99.9
14	100
21	100
28	100

Dextroamphetamine Deactivation Protocol

Dextroamphetamine Sulfate

Date: 12/19/15

Protocol:

1) 10 tablets of Dextroamphetamine 10 mg were placed in pouch
2) 50ml warm water (43 degree celcius) was added to the pouch and was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Washout Protocol:

1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken
3) Water was replaced with 250 mL of 30%(v/v) ethanol
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken

Filtration of samples:

Pouch was shaken before taking the samples

1) 1ml of sample was taken from syringe and filtered through 0.22 micron syringe filter
2) This filtered samples was analyzed by using HPLC.

Chromatographic conditions: Dextroamphetamine Sulfate

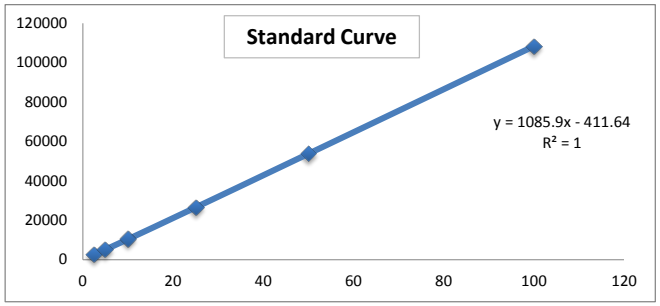
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Gradient
Mobile Phase A- ACN (0.05%TFA) B-Water (0.05%TFA)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 15min
Retention time: 4.18 min
Column: (size and particle size): Phenomenex, Prodigy 5µ ODS(2) 150 x 4.6 mm 5 micron
Column Temperature: 25°C
Detection wavelength: 258 nm

HPLC Gradient Method

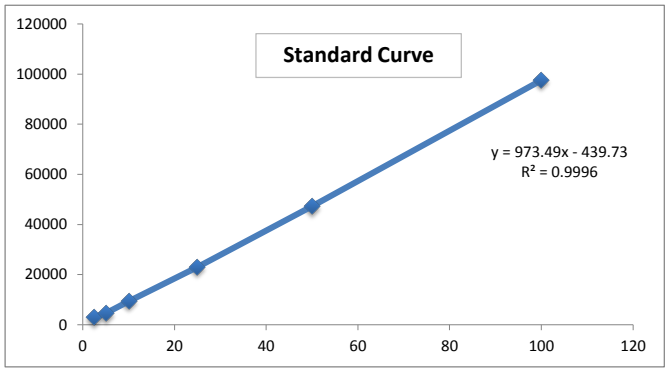
Time (min)	Flow (ml/min)	% A	% B
0	1	10	90
0.8	1	80	20
13	1	80	20
13.01	1	10	90
15	1	10	90

Dextroamphetamine Deactivation Standard Curve

Standard curve for Dextroamphetamine- 8hr to Day 14	
Concentration (µg/ml)	Area
2.5	2577
5	5125.00
10	10346.00
25	26482.00
50	53714.00
100	108330.00



Standard curve for Dextroamphetamine- Day21 to washout studies	
Concentration (µg/m	Area
2.5	2909
5	4606
10	9374
25	23035
50	47264
100	97571



Dextroamphetamine Deactivation/Desorption

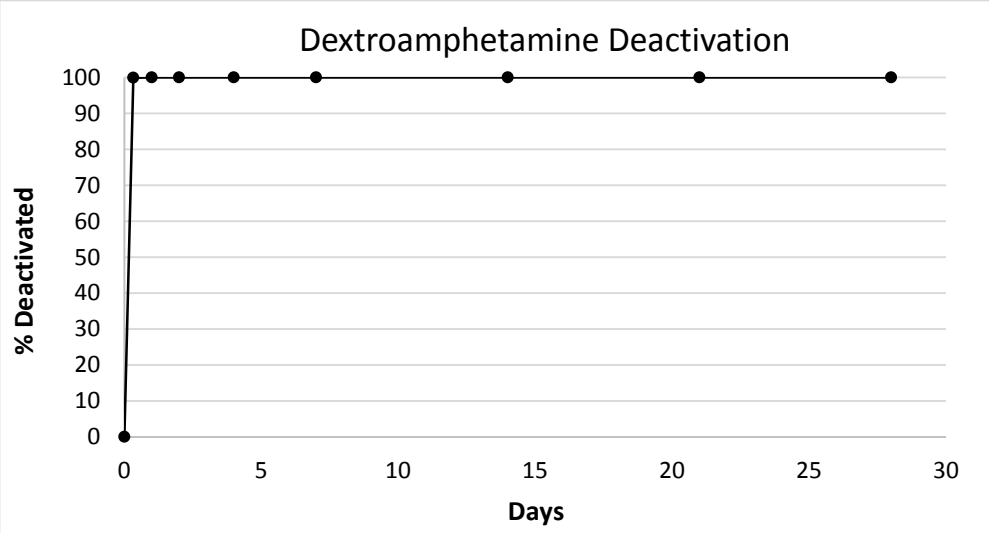
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	2210.00	2.41	0.00	120.71	0.12	99.88	99.87
8hr	n-2	2791.00	2.95	0.00	147.46	0.15	99.85	
Day 1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 1	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 2	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 2	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 4	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 4	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 7	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 7	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 14	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 14	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 21	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 28	n-2	0.00	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout- H2O	n-1	0.00	0.00	0.00	0.00	100.00	100.00
Day 29 Washout- H2O	n-2	0.00	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- ethanol	n-1	31018.00	32.31	8078.60	8.08	8.08	9.39
Day 30 Washout- ethanol	n-2	41258.00	42.83	10708.31	10.71	10.71	

Total amount of drug added to the pouch-100mg

Dextroamphetamine Deactivation - Mercer

10 mg Tablets



Days	% Deactivated
0	0
0.33	99.9
1	100
2	100
4	100
7	100
14	100
21	100
28	100

Diazepam Deactivation Protocol

Protocol

Diazepam Deadsorption Study

1) 10 tablets of diazepam were placed in pouch.
2) 50ml warm tap water (43 degree celcius) was added to the pouch and wait for 30 seconds.
3) Pouch was sealed and gently shaken from side to side
4) It was placed in upright position and take samples were taken at 8hour, 1, 2, 4, 7, 14, 21, 28days time points.

Filtration of Samples:

Pouch was shaken properly before taking samples.
1) Sample dilution
   a. 0.1ml sample was taken out from pouch (8h, 1day, 2day 4day and 7day pouches) and diluted to 16ml with distilled water (Dilution factor: 160X).
   b. 0.1ml sample was taken out from pouch (14day and 21day pouches) and diluted to 5ml with distilled water (Dilution factor: 50X)
2) 500ul of sample was filtered using 0.22 micron syringe.
3) Samples were analyzed with HPLC.

Desorption Study

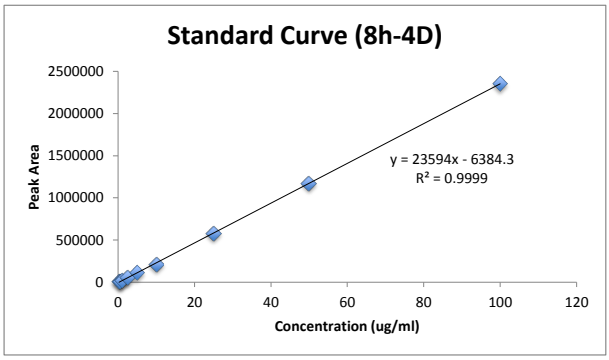
1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml DI water was added to 500ml bottles
4) Samples were rocked on shaker for 1 hour (50rpm)
5) Samples were analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50rpm)
8) Samples were analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

Chromatographic Conditions:

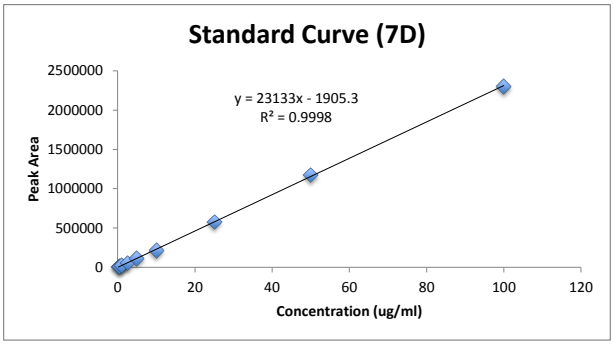
System: HPLC
Mobile Phase: Methanol (60%), pH2.5 phosphate buffer (40%)
Flow rate: 1.2ml/min
Run time: 10min
Retention time: ~5min
Column: Kinetex 5u EVO C18 100A, New Column 150x4.6mm
Temperature: 30° C
Detection wavelength: 230nm

Diazepam Standard Curve

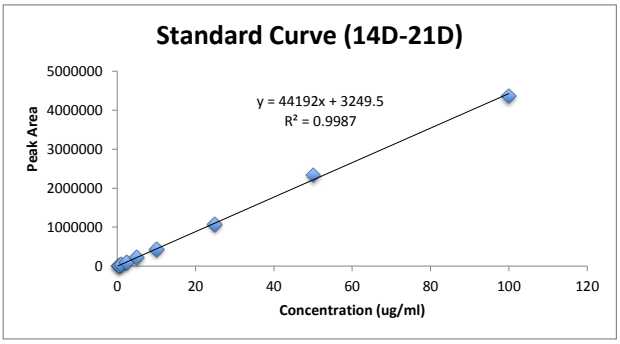
Standard Curve (8h-4D)	
Concentration(ug/ml)	Peak Area
0.25	5023
0.5	10469
1	18879
2.5	56256
5	112654
10	212763
25	581027
50	1173692
100	2354923



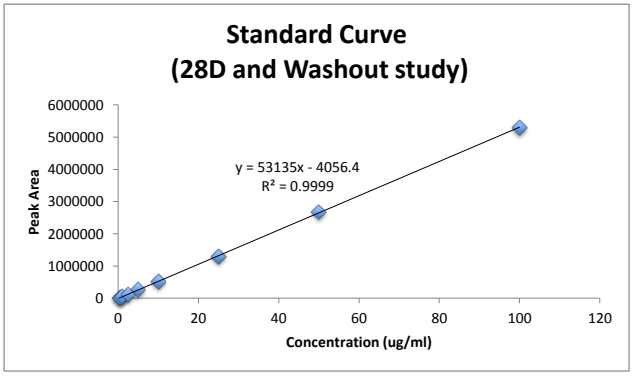
Standard Curve (7D)	
Concentration(ug/ml)	Peak Area
0.25	4844
0.5	9691
1	19524
2.5	56795
5	115279
10	213747
25	578945
50	1176041
100	2301660



Standard Curve (14D-21D)	
Concentration(ug/ml)	Peak Area
0.25	9059
0.5	20240
1	41650
2.5	101061
5	224239
10	429365
25	1075709
50	2346572
100	4365707



Standard Curve (28D and Desorption Study)	
Concentration(ug/ml)	Peak Area
0.25	10194
0.5	23781
1	51328
2.5	124956
5	263891
10	524957
25	1303911
50	2681720
100	5300192



Diazepam Deactivation

Time(days)	Pouch	Peak Area	Conc(ug/ml)	Dilution Factor	Original Conc(ug/ml)	Amount of drug in 50 ml(mg)	% Reacted	Avg %
0.33	1	175938	7.73	160	1236.40	61.82	38.18	46.25
	2	128330	5.71	160	913.55	45.68	54.32	
1	1	157786	6.96	160	1113.30	55.67	44.33	50.76
	2	119889	5.35	160	856.31	42.82	57.18	
2	1	79989	3.66	160	585.73	29.29	70.71	72.07
	2	71995	3.32	160	531.52	26.58	73.42	
4	1	69608	3.22	160	515.33	25.77	74.23	73.43
	2	74336	3.42	160	547.40	27.37	72.63	
7	1	75293	3.34	160	533.94	26.70	73.30	74.17
	2	70281	3.12	160	499.28	24.96	75.04	
14	1	188642	4.20	50	209.76	10.49	89.51	90.04
	2	170016	3.77	50	188.68	9.43	90.57	
21	1	74293	1.61	50	80.38	4.02	95.98	94.87
	2	113634	2.50	50	124.89	6.24	93.76	
28	1	23651	0.52	20	10.43	0.52	99.48	99.32
	2	40982	0.85	20	16.95	0.85	99.15	

Original amount of drug in the pouch=100mg

Diazepam Desorption

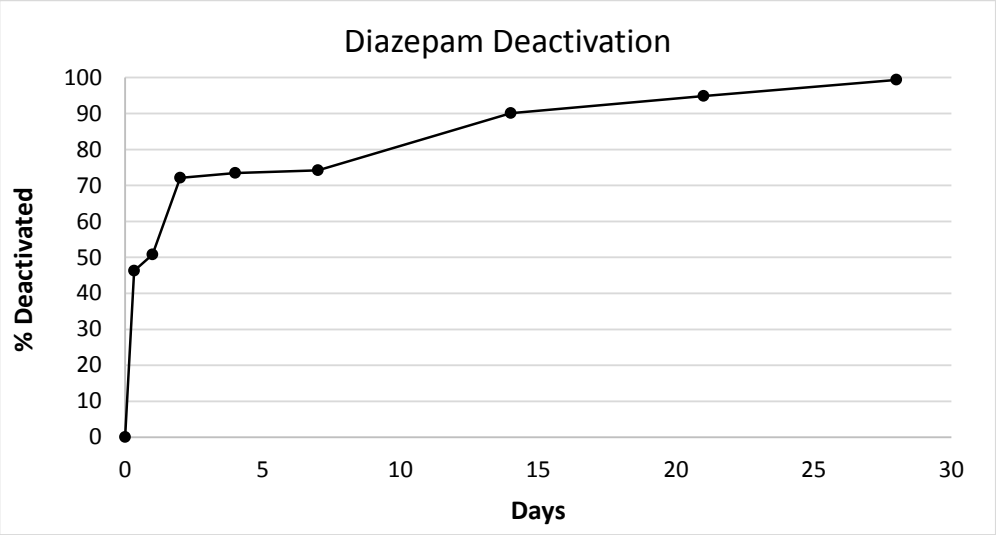
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average
Day 29 Washout- H2O	1	372542.00	7.09	1771.89	1.77	98.23	98.34
Day 29 Washout- H2O	2	324324.00	6.18	1545.03	1.55	98.45	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- 30% ethanol	1	306045.00	5.84	1459.03	1.46	1.48	1.61
Day 30 Washout- 30% ethanol	2	345728.00	6.85	1712.50	1.71	1.74	

Amount of drug reacted is 98.34mg

Medication	% leached in 30% ethanol
Diazepam	1.61

Diazepam Deactivation - Mercer

10 mg Tablets



Days	% Deactivated
0	0
0.33	46.3
1	51
2	72
4	73
7	74
14	90
21	95
28	99

Fentanyl Deactivation Protocol

Protocol: Amount of Fentanyl in the pouch

1) Attach a piece of Kimwipe to the adhesive portion of the fentanyl patch in order to prevent the patch from sticking to the walls of Medsaway pouches.

2) Place two fentanyl patches in each pouch - one on either side of carbon packet (Kimwipe facing carbon packet).

3) 50ml warm water (43 degree celcius) was added to the pouch and was kept open for 30 seconds
4) Pouch was sealed after 30 seconds and gently shaken from side to side.
5) They were then placed in a upright position at room temperature and samples were taken.
6) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Filtration of samples:

Pouch was shaken before taking the samples
1) 1ml of sample was taken from syringe and filtered through 0.22 micron syringe filter
2) These filtered samples were analyzed by using HPLC.

Desorption Protocol- Fentanyl

1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml water was added to 500ml bottles.
4) Samples were rocked on shaker for 1hr (50rpm)
5) Following this, samples were analyzed after additional 23 hr exposure. (Total 24 hrs)
6) Water was replaced with 250ml of 30% (V/V) ethanol.
7) Samples were rocked on shaker for 1hr (50rpm)
8) Samples were analyzed after additional 23 hr exposure. (Total 24hrs)

Filtration of samples:

Bottle was shaken before taking the samples
1) 1ml of sample was taken from syringe and filtered through 0.22 micron syringe filter
2) These filtered samples were analyzed by using HPLC.

Protocol: Amount of residual Fentanyl in the patch

1) After the samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time point, fentanyl patches were removed from the pouch.
2) To measure residual fentanyl in the patches, individual patches were placed on the bottom of glass jars (250 ml) using double-sided tape. **Note: Kimwipe will be easily removed from patch.**
3) 200ml water was added to the beaker containing these indivdual pouches.
4) Beakers were kept on a shaker (90rpm) in dark for 5 days (to get peak concentartions)

Filtration of samples:

1) 1ml of sample was taken from syringe and filtered through 0.22 micron syringe filter
2) This filtered samples was analyzed by using HPLC.

Chromatographic conditions: Drug- Fentanyl

System: Waters e2795

Detector: Waters 2988 photodiode array

Pump Mode: Isocratic

Mobile Phase: ACN (60%) : water (0.2% (v/v) Formic acid containing 10mM sodium -1-decane sulfonate) (40%)

Flow rate: 1ml/min

Injection Volume: 20µL

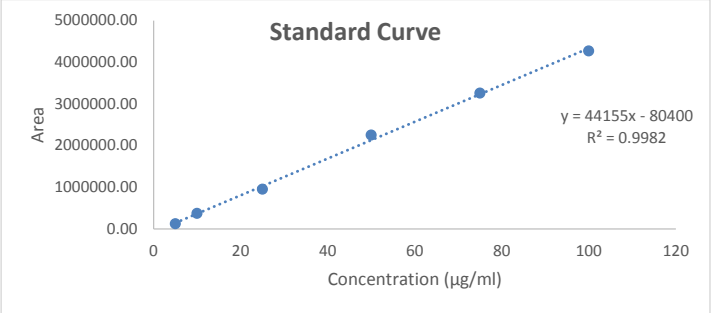
Run time: 10 min

Retention time: 4.8min

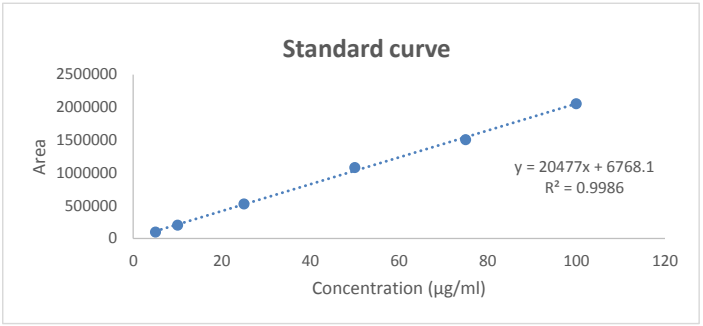
Column: (size and particle size): Gemini NX C18; 150x4.6 mm

Fentanyl Standard Curve

Standard curve for Fentanyl- 8hr to Day 14	
Concentration (µg/ml)	Area
5	117273.00
10	370570.00
25	950571.00
50	2251173.00
75	3256272.00
100	4272910.00



Standard curve for Fentanyl- Day21 to washout studies	
Concentration (µg/m	Area
5	94769
10	203167
25	524338
50	1082145
75	1507422
100	2055235



Fentanyl in Patch

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
8hr	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 1	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 2	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 2	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 4	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 4	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 7	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 7	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 14	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 14	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 21	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 28	n-2	0.00	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout- H2O	n-1	0.00	0.00	0.00	0.00	100.00	100.00
Day 29 Washout- H2O	n-2	0.00	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- ethanol	n-1	0.00	0.00	0.00	0.00	0.00	0.00
Day 30Washout- ethanol	n-2	0.00	0.00	0.00	0.00	0.00	

Total amount of drug added to the pouch-2.75mg

% Reacted

%= (Total amount of drug in pouch- Amount of drug in sample)/Total amount of drug in pouch)*100

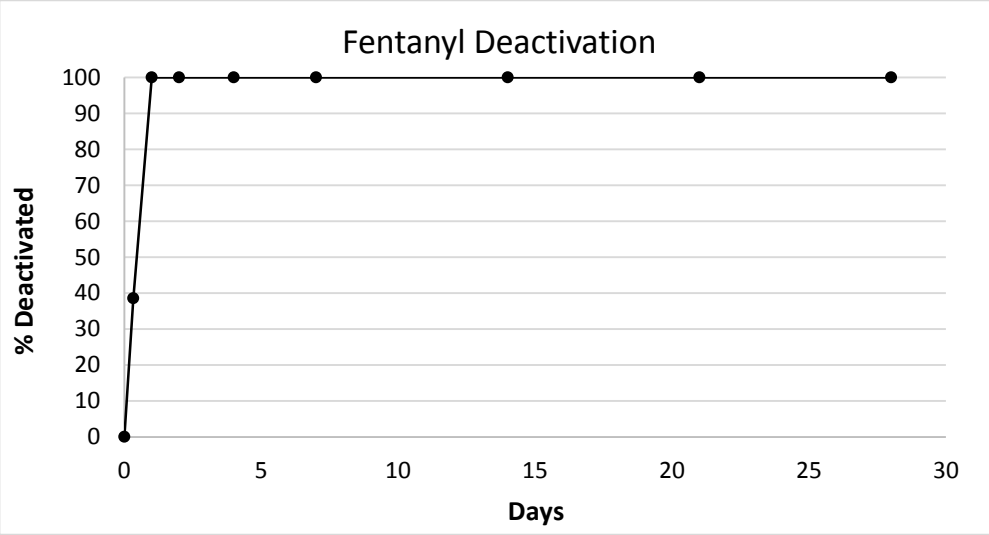
Residual Fentanyl

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 200ml (µg)	Amount of drug (mg)	% Reacted	Average (%)	% Fentanyl remaning in the patch
8hr N-1	n-1	217168.00	6.74	6.74	1348.00	1.35	50.98	38.55	61.45
8hr N-1	n-1	202522.00	6.41	6.41	1282.00	1.28	53.38		
8hr N-2	n-2	367467.00	10.14	10.14	2028.00	2.03	26.25		
8hr N-2	n-2	383494.00	10.51	10.51	2102.00	2.10	23.56		
Day 1 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00	0
Day 1 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00		
Day 1 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 1 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 2 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00	0
Day 2 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00		
Day 2 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 2 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 4 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00	0
Day 4 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00		
Day 4 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 4 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 7 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00	0
Day 7 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00		
Day 7 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 7 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 14 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00	0
Day 14 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00		
Day14 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 14 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 21 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00	0
Day 21 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00		
Day 21 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 21 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 28 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00	0
Day 28 N-1	n-1	0.00	0.00	0.00	0.00	0.00	100.00		
Day 28 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		
Day 28 N-2	n-2	0.00	0.00	0.00	0.00	0.00	100.00		

Total amount of drug added to the pouch-5.50mg

Fentanyl Deactivation - Mercer

2.75 mg (25 mcg/hr) transdermal patch



Days	% Deactivated
0	0
0.33	38.5
1	100
2	100
4	100
7	100
14	100
21	100
28	100

Fluoxetine Deactivation Protocol

Date: 12/30/14

Fluoxetine Protocol:

1) 10 tablets of Fluoxetine were placed in pouch
2) 50ml warm water (43 degree celcius) was added to the pouch and wait for 30 seconds
3) Seal the pouch and gently shake it from side to side
4) place it in upright position and take samples at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Filtration of samples:

Pouch was shaken before taking the samples

1) 1ml of sample was taken from syringe and filtered through 0.22 micron syringe filter
2) 500ul of the sample filtered was diluted 1:10 times. (100ul stock+900ul water)
3) This was further diluted 1:10 times.(100ul **(2)**+ 900ul water)
4) Diluted samples were ananlyzed by using HPLC.

Corrected Protocol for filtration of samples

Pouch was shaken before taking the samples

1) 0.1ml of sample was taken from pouch and diluted to 16ml with water.
2) 0.1ml of this diluted sample was taken and again diluted to 16ml with water.
3) 500ul of sample was filtered using 0.22 micron syringe.
3) The dilution factor was 25600X.
4) These samples were analyzed by using HPLC

Desorption Protocol

1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml water was added to 500ml bottles.
4) Samples were rocked on shaker for 1hr (50rpm)
5) Following this, samples were analyzed after additional 23 hr exposure. (Total 24 hrs)
6) Water was replaced with 250ml of 30% (V/V) ethanol.
7) Samples were rocked on shaker for 1hr (50rpm)
8) Samples were analyzed after additional 23 hr exposure. (Total 24hrs)

HPLC Analysis

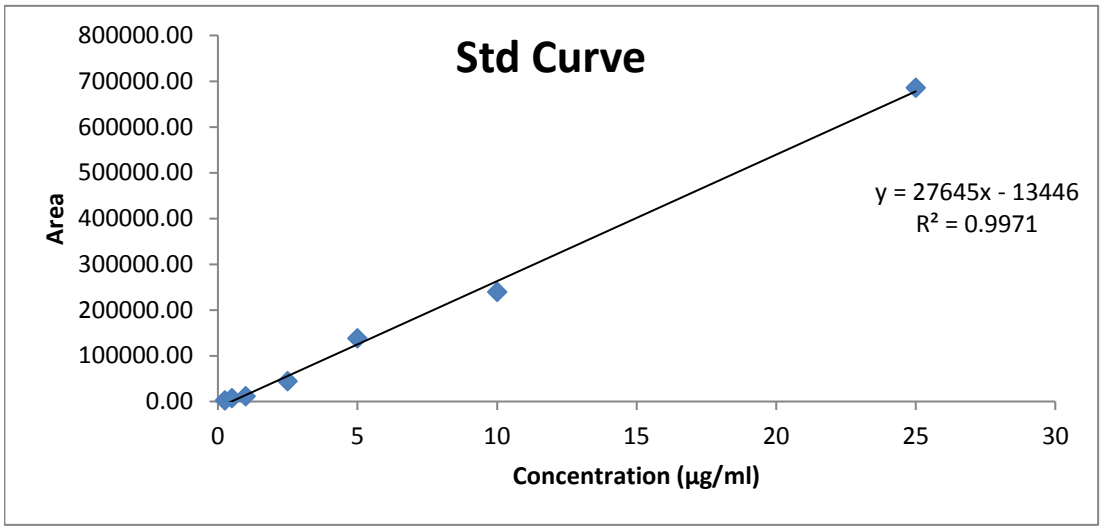
Chromatographic conditions:

System: Waters e2795
Mobile Phase: Acetonitrile: Potassium dihydrogen phosphate buffer(40:60)
Flow rate: 1ml/min
Run time: 10 min
Retention time: 3.4 min
Column: (size and particle size): C18 kinetex 5um, 250* 4.6mm
Column Temperature: 25°C
Detection wavelength: 226nm

Fluoxetine Standard Curve

Standard curve for Fluoxetine

Concentration (µg/ml)	Area
0.25	2539.00
0.5	7648.00
1	11656.33
2.5	44335.33
5	138226.67
10	239276.67
25	685465.33



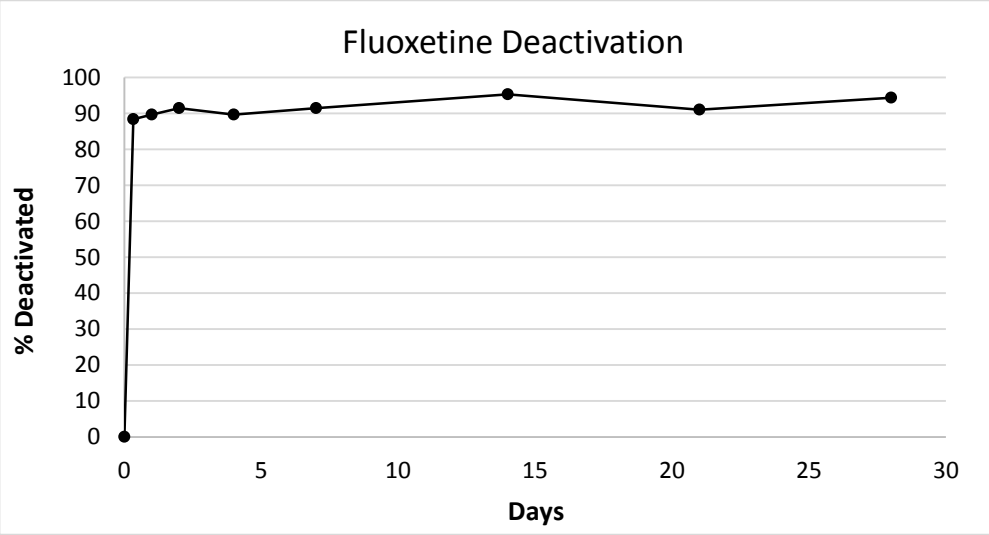
Fluoxetine Deactivation/Desorption

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (160X) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	99665.00	4.09	654.65	32732.43	32.73	83.63	84.46
8hr	n-2	88269.00	3.68	588.69	29434.62	29.43	85.28	
Day 1	n-1	73591.00	3.15	503.74	25187.05	25.19	87.41	87.37
Day 1	n-2	74128.00	3.17	506.85	25342.45	25.34	87.33	
Day 2	n-1	69411.00	3.00	479.55	23977.43	23.98	88.01	88.36
Day 2	n-2	64615.00	2.82	451.79	22589.55	22.59	88.71	
Day 4	n-1	58915.00	2.62	418.80	20940.06	20.94	89.53	89.68
Day 4	n-2	56887.00	2.54	407.06	20353.19	20.35	89.82	
Day 7	n-1	41877.00	2.00	320.19	16009.55	16.01	92.00	91.49
Day 7	n-2	48907.00	2.26	360.88	18043.91	18.04	90.98	
Day 14	n-1	21725.00	1.27	203.56	10177.90	10.18	94.91	95.30
Day 14	n-2	16332.00	1.08	172.35	8617.25	8.62	95.69	
Day 21	n-1	48575.00	2.24	358.96	17947.84	17.95	91.03	91.04
Day 21	n-2	48378.00	2.24	357.82	17890.83	17.89	91.05	
Day 28	n-1	24818.00	1.38	221.46	11072.96	11.07	94.46	94.37
Day 28	n-2	26155.33	1.43	229.20	11459.96	11.46	94.27	

Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout- H2O	n-1	60059.67	2.66	664.73	0.66	99.67	99.63
Day 29 Washout- H2O	n-2	75903.67	3.23	808.01	0.81	99.60	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- ethanol	n-1	110678.67	4.49	1122.49	1.12	0.56	0.71
Day 30Washout- ethanol	n-2	175900.33	6.85	1712.30	1.71	0.86	

Fluoxetine Deactivation - Mercer

20 mg Capsule



Days	% Deactivated
0	0
0.33	88.4
1	89.7
2	91.5
4	89.7
7	91.5
14	95.3
21	91.0
28	94.4

Hydromorphone Protocol

Protocol:

1) 10 tablets of Hydromorphone (4 mg) were placed in a pouch.
2) 50mL warm water (43 °C) was added to the pouch and it was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time points.

Washout Protocol:

1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle.
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.
3) Water was replaced with 250 mL of 30%(v/v) ethanol.
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.

Filtration of samples:

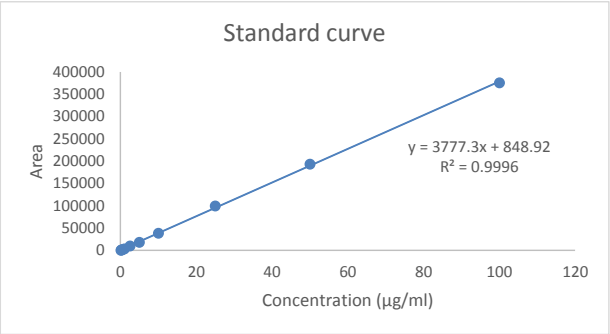
1) Pouch was shaken before taking the samples
2) 2mL of sample was taken by a syringe and centrifuged for 3 minutes at 13400 rpm
3) The supernatant was filtered through 0.45 µm syringe filter
4) Samples was analyzed using HPLC

Chromatographic conditions: Hydromorphone Hydrochloride

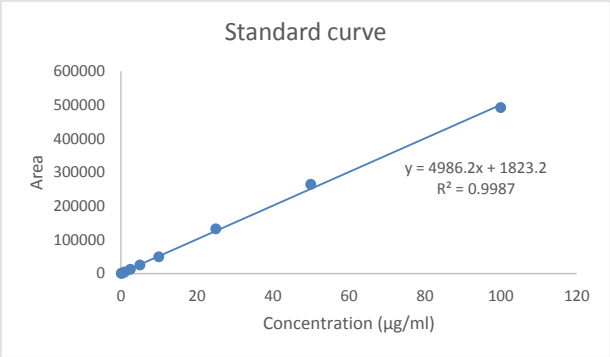
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN: Water (% 0.5 w/v Sodium dodecyl sulphate and 0.4% TFA) (35:65) (% v/v)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 5.36 min
Column: (size and particle size): Phenomenex, Kinetex EVO C18 5µm, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 282 nm
Sample Preparation: Standards were prepared in D.I Water

Hydromorphone Standard Curve

Standard curve - 8hr to Day 7	
Concentration (µg/ml)	Area
0.1	287
0.25	1059
0.5	2072
1	3572
2.5	9860
5	18287
10	38561
25	99779
50	193420
100	375701



Standard curve - Day 14 to washout studies	
Concentration (µg/ml)	Area
0.1	387
0.25	1472
0.5	2764
1	4868
2.5	12734
5	25218
10	50031
25	132721
50	264680
100	492420



Hydromorphone Deactivation/Desorption

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 1	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 2	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 4	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 7	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 14	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	

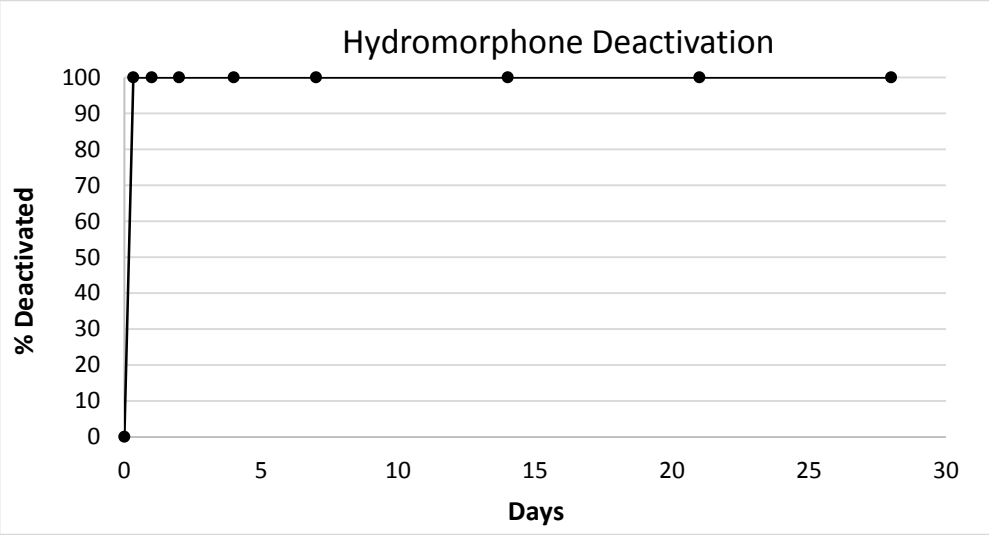
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout-H2O	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Leached out in ethanol	Average(%)
Day 30 Washout-Ethanol	n-1	4791	0.60	0.00	148.80	0.15	0.37	0.50
	n-2	6816	1.00	0.00	250.33	0.25	0.63	

Total amount of drug added to the pouch = 40 mg
Each tablet of hydromorphone = 4 mg

Hydromorphone Deactivation - Mercer

4 mg Tablet



Days	% Deactivated
0	0
0.33	100
1	100
2	100
4	100
7	100
14	100
21	100
28	100

Ketamine Protocol

Protocol:

1) contents of 1 vial of Ketamine hydrochloride inj sol (500mg/10mL) was transfered in a pouch.
2) 50mL warm water (43 °C) was added to the pouch and it was kept open for 30 seconds.
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time points.

Washout Protocol:

1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle.
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.
3) Water was replaced with 250 mL of 30%(v/v) ethanol.
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.

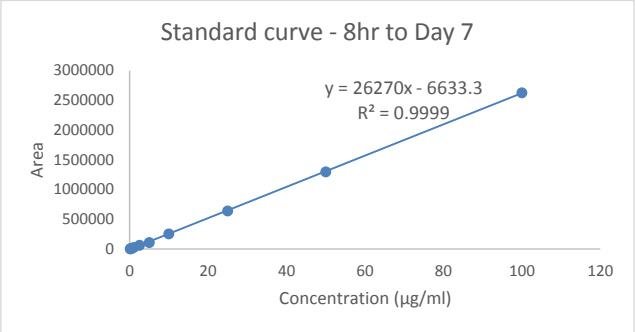
Filtration of samples:

1) Pouch was shaken before taking the samples
2) 2mL of sample was taken by a syringe and centrifuged for 3 minutes at 13400 rpm
3) The supernatant was filtered through 0.45 µm syringe filter
4) Ethanol wash out samples were diluted 2 times with DI water
5) Samples was analyzed using HPLC

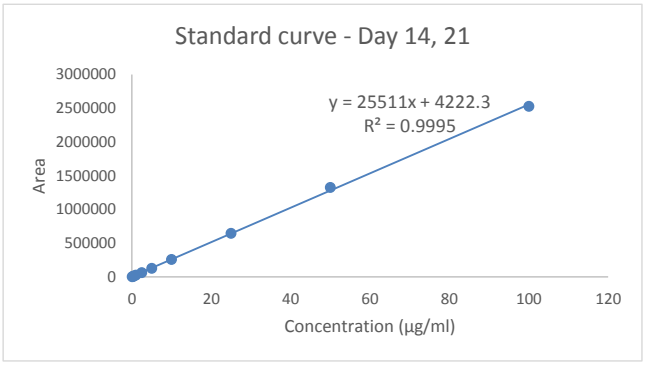
Chromatographic conditions: Ketamine hydrochloride
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: MeOH:Buffer (NaH_2PO_4 50mM pH=3) (30:70)
Flow rate: 1.2ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 6.1min
Column: Agilent ZORBAX Eclipse Plus C 18 (4.6×150mm 5µm)
Column Temperature: 25°C
Detection wavelength: 215 nm

Ketamine Standard Curve

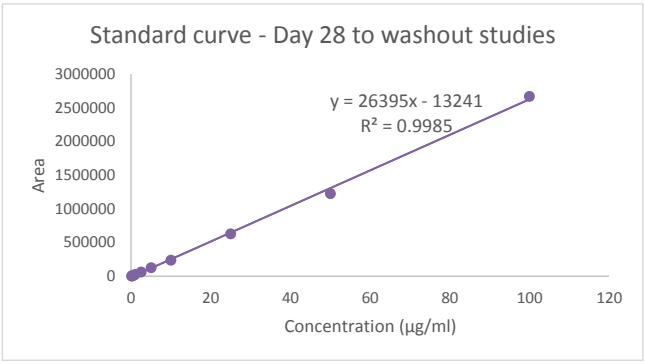
Standard curve - 8hr to Day 7	
Concentration (µg/ml)	Area
0.1	2561
0.25	5950
0.5	11707
1	25310
2.5	64385
5	108419
10	255020
25	641079
50	1296119
100	2628660



Standard curve - Day 14 & 21	
Concentration (µg/ml)	Area
0.1	2288
0.25	5711
0.5	11653
1	25225
2.5	62859
5	127783
10	259892
25	646520
50	1327845
100	2530529



Standard curve - Day 28 to washout studies	
Concentration (µg/ml)	Area
0.1	3431
0.25	6239
0.5	10631
1	24172
2.5	63554
5	126028
10	237618
25	627992
50	1226316
100	2671482



Ketamine Deactivation/Desorption

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	935419	35.86	0.00	1793.02	1.79	99.64	99.66
	n-2	852060	32.69	0.00	1634.36	1.63	99.67	
Day 1	n-1	125250	5.02	0.00	251.02	0.25	99.95	99.94
	n-2	176962	6.99	0.00	349.44	0.35	99.93	
Day 2	n-1	24970	1.20	0.00	60.15	0.06	99.99	99.99
	n-2	36997	1.66	0.00	83.04	0.08	99.98	
Day 4	n-1	14235	0.79	0.00	39.72	0.04	99.99	99.99
	n-2	16709	0.89	0.00	44.43	0.04	99.99	
Day 7	n-1	3819	0.40	0.00	19.89	0.02	100.00	100.00
	n-2	3596	0.39	0.00	19.47	0.02	100.00	
Day 14	n-1	3619	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	2157	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	3222	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	3580	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	2032	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	3354	0.00	0.00	0.00	0.00	100.00	

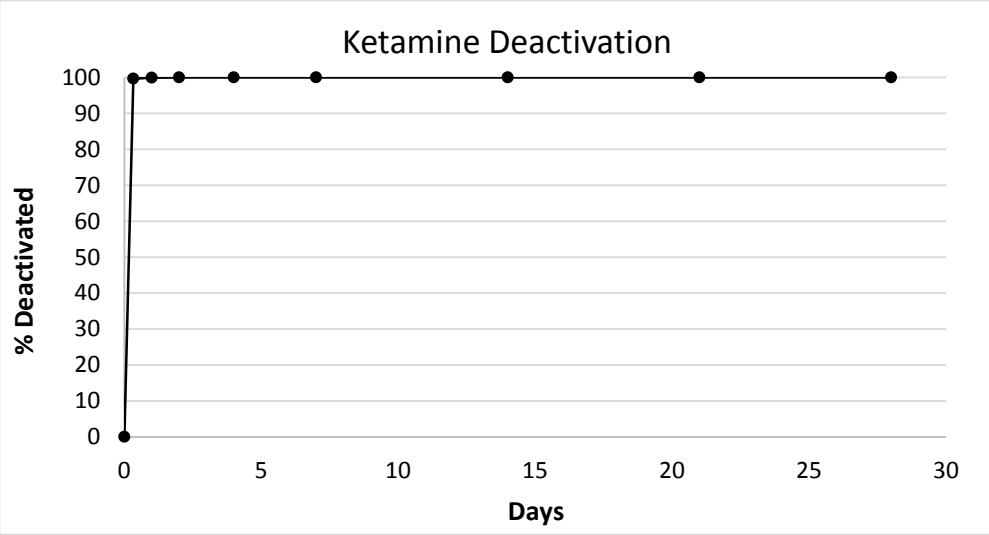
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout-H2O	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Leached out in ethanol	Average(%)
Day 30 Washout-Ethanol	n-1	1822028	69.53	2.00	34765.47	34.77	6.95	6.54
	n-2	1604180	61.28	2.00	30638.78	30.64	6.13	

Total amount of drug added to the pouch = 500 mg
Each vial contains 500mg Ketamine

Ketamine Deactivation - Mercer

Liquid. 10 ml of 50 mg/ml



Days	% Deactivated
0	0
0.33	99.7
1	99.9
2	100
4	100
7	100
14	100
21	100
28	100

Lorazepam Deactivation Protocol

Lorazepam Adsorption and Desorption Study Protocol

Lorazepam adsorption Study

1) 10 tablets of lorazepam were placed in pouch (20mg Lorazepam).
2) 50ml warm tap water (43 degree celcius) was added to the pouch and wait for 30 seconds.
3) Pouch was sealed and gently shaken from side to side
4) It was placed in upright position and take samples were taken at 8hour, 1, 2, 4, 7, 14, 21, 28days time points.

Filtration of Samples:

Pouch was shaken properly before taking samples.
1) Sample dilution
   a. 0.1ml sample was taken out from pouch (8h, 1day, 2day pouches) and diluted to 5ml with distilled water (Dilution factor: 50X).
   b. 0.1ml sample was taken out from 4day pouch and diluted to 1ml with distilled water (Dilution factor: 10X)
2)1mL of sample was filtered using 0.22 micron syringe.
3) Samples were analyzed with HPLC.

Desorption Study

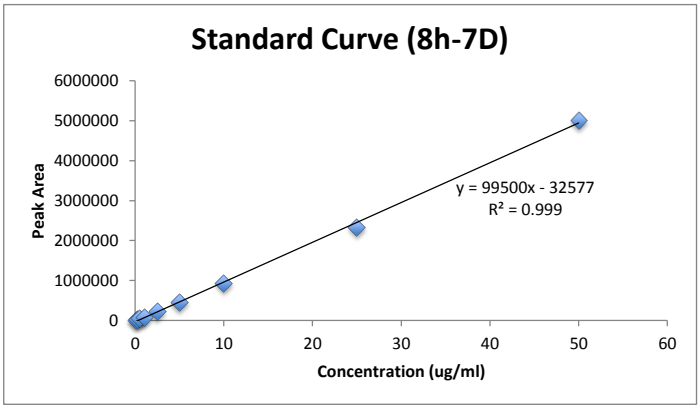
1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml DI water was added to 500ml bottles
4) Samples were rocked on shaker for 1 hour (50rpm)
5) Samples were analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50rpm)
8) Samples were analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

Chromatographic Conditions:

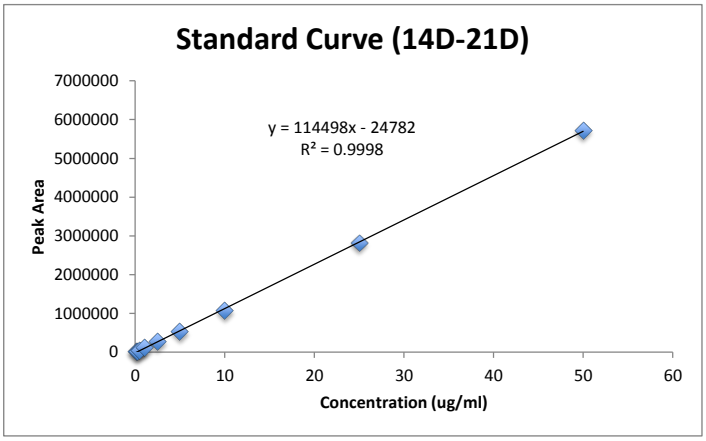
System: HPLC
Mobile Phase: Acetonitrile (0.05%TFA), Water (0.05% TFA)
Flow rate: 1.0ml/min
Run time: 8.5 min
Retention time: ~6.5min
Column: XBridge BEH Phenyl 4.6x50mm Column XP
Temperature: 30° C
Detection wavelength: 229nm

Lorazepam Standard Curve

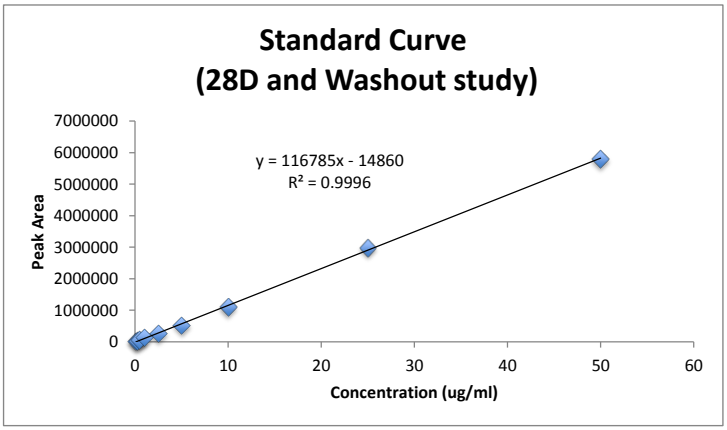
Standard Curve (8h-7D)	
Concentration(ug/ml)	Peak Area
0.1	6732
0.25	19234
0.5	42268
1	81993
2.5	214241
5	452560
10	933102
25	2336314
50	5008165



Standard Curve (14D-21D)	
Concentration(ug/ml)	Peak Area
0.1	10886
0.25	26842
0.5	49507
1	102970
2.5	262104
5	526164
10	1058583
25	2820015
50	5722819



Standard Curve (28D and Desorption Study)	
Concentration(ug/ml)	Peak Area
0.1	10194
0.25	23781
0.5	51328
1	124956
2.5	263891
5	524957
10	1103911
25	2981720
50	5800192



Lorazepam Deactivation

Time(days)	Pouch	Peak Area	Conc(ug/ml)	Dilution Factor	Original Conc(ug/ml)	Amount of drug in 50 ml(mg)	% Reacted	Avg %
0.33	1	189388	2.23	50	111.54	5.58	72.11	70.73
	2	211421	2.45	50	122.61	6.13	69.35	
1	1	118497	1.52	50	75.92	3.80	81.02	79.11
	2	148863	1.82	50	91.18	4.56	77.21	
2	1	67514	1.01	50	50.30	2.51	87.43	87.54
	2	65648	0.99	50	49.36	2.47	87.66	
4	1	125127	1.58	10	15.85	0.79	96.04	96.23
	2	110204	1.43	10	14.35	0.72	96.41	
7	1	38383	0.71	1	0.71	0.04	99.82	99.81
	2	48234	0.81	1	0.81	0.04	99.80	
14	1	18864	0.38	1	0.38	0.02	99.90	99.91
	2	17001	0.36	1	0.36	0.02	99.91	
21	1	7293	0.28	1	0.28	0.01	99.93	99.93
	2	8634	0.29	1	0.29	0.01	99.93	
28	1	6016	0.18	1	0.18	0.01	99.96	99.96
	2	5971	0.18	1	0.18	0.01	99.96	

Original amount of drug in the pouch=20mg

Lorazepam Desorption

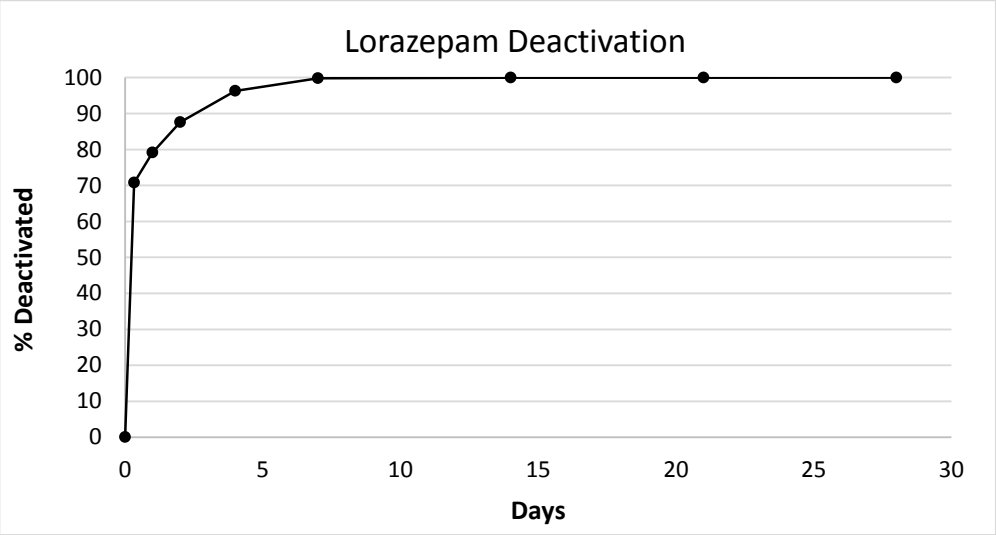
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average
Day 29 Washout- H2O	1	11699.00	0.23	56.85	0.06	99.72	99.70
Day 29 Washout- H2O	2	14802.00	0.25	63.50	0.06	99.68	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- 30% ethanol	1	8662.00	0.20	50.35	0.05	0.25	0.25
Day 30 Washout- 30% ethanol	2	8937.00	0.20	50.94	0.05	0.26	

Amount of drug reacted is 98.34mg

Medication	% leached in 30% ethanol
Lorazepam	0.25

Lorazepam Deactivation - Mercer

2 mg Tablet



Days	% Deactivated
0	0
0.33	70.7
1	79.1
2	87.5
4	96.2
7	99.8
14	99.9
21	99.9
28	100.0

Loxapine Protocol

Date: 1/5/16

Loxapine deactivation Protocol:

1) 10 Loxapine capsules were placed in pouch
2) 50ml warm water (43 degree celcius) was added to the pouch and wait for 30 seconds
3) Seal the pouch and gently shake it from side to side
4) place it in upright position and take samples at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Filtration of samples:

Pouch was shaken before taking the samples

1) Samples from 8hr and day 1 were diluted 1:1 dilution (2x) and then analyzed.
2) 1ml of sample was taken from syringe and filtered through 0.22 micron syringe filter
3) Filtered samples were ananlyzed by using HPLC.

Desorption Protocol

1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml water was added to 500ml bottles.
4) Samples were rocked on shaker for 1hr (50rpm)
5) Following this, samples were analyzed after additional 23 hr exposure. (Total 24 hrs)
6) Water was replaced with 250ml of 30% (V/V) ethanol.
7) Samples were rocked on shaker for 1hr (50rpm)
8) Samples were analyzed after additional 23 hr exposure. (Total 24hrs)

Filtration of samples:

Bottle was shaken before taking the samples

1) 1ml of sample was taken from syringe and filtered through 0.22 micron syringe filter
2) Filtered samples were ananlyzed by using HPLC.

Chromatographic conditions: Drug- Loxapine

System: Waters e2795

Detector: Waters 2988 photodiode array

Pump Mode: Isocratic

Mobile Phase: ACN (40%) : water (0.3% (v/v) Triethylamine pH-3 (60%)

Flow rate: 1ml/min

Injection Volume: 20µL

Run time: 10 min

Retention time: 4.8min

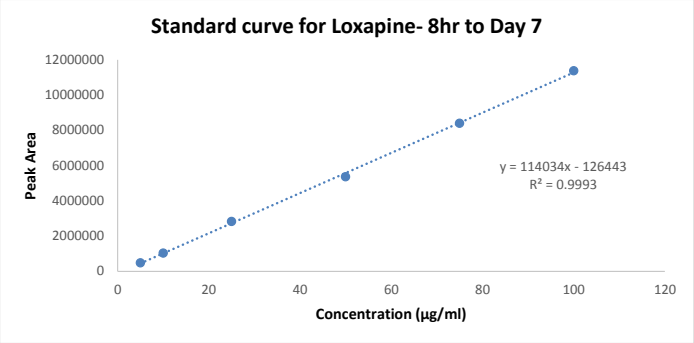
Column: (size and particle size): Phenomenex Luna 5µ C8 (2) 100A 250*4.60 mm 5µ

Column Temperature: 25°C

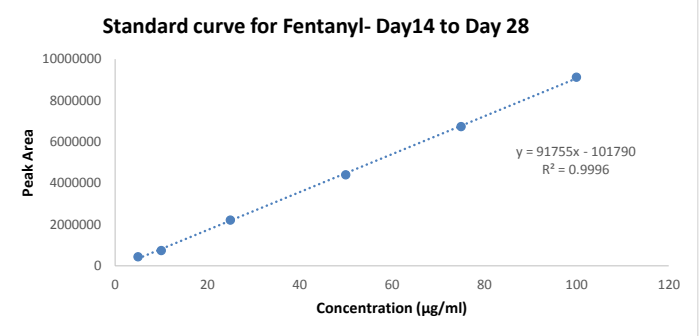
Detection wavelength: 190 nm

Loxapine Standard Curve

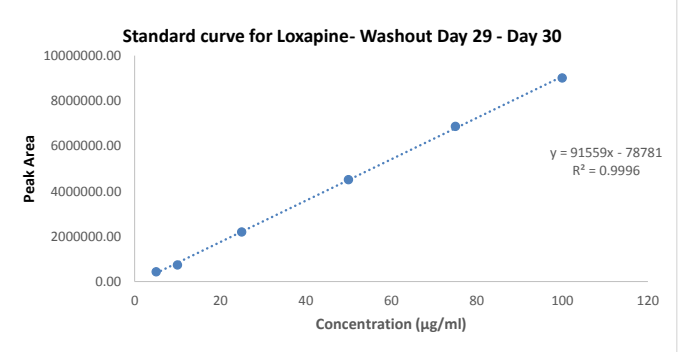
Standard curve for Loxapine- 8hr to Day 7	
Concentration (µg/ml)	Area
5	475481
10	1027112
25	2820079
50	5364589
75	8392174
100	11380906



Standard curve for Loxapine- Day14 to Day 28	
Concentration (µg/ml)	Area
5	444161
10	747811
25	2222508
50	4415896
75	6741966
100	9131928



Standard curve for Loxapine- Washout Day 29 - Day 30	
Concentration (µg/ml)	Area
5	437953.00
10	746034.00
25	2203163.00
50	4519873.00
75	6870597.00
100	9012783.00



Loxapine Deactivation/Desorption

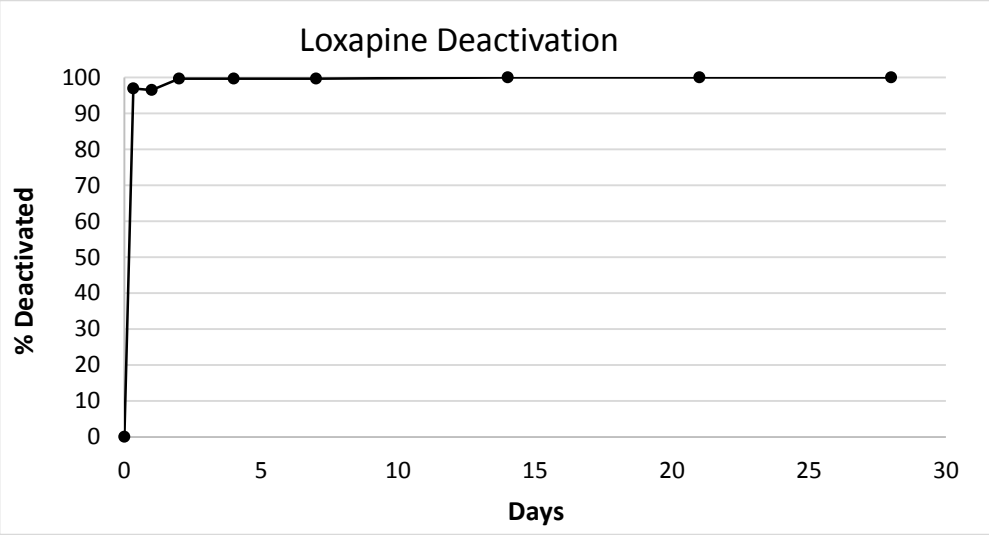
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	5959660	53.37	106.74	5337.10	5.34	97.33	96.94
8hr	n-2	7759435	69.15	138.31	6915.37	6.92	96.54	
Day 1	n-1	6427033	57.47	114.94	5746.95	5.75	97.13	96.51
Day 1	n-2	9233501	82.08	164.16	8208.03	8.21	95.90	
Day 2	n-1	1989136	18.55	18.83	941.60	0.94	99.53	99.62
Day 2	n-2	1157064	11.26	11.43	571.26	0.57	99.71	
Day 4	n-1	1466637	13.97	14.18	709.04	0.71	99.65	99.62
Day 4	n-2	1706022	16.07	16.31	815.59	0.82	99.59	
Day 7	n-1	1516687	14.41	14.63	731.32	0.73	99.63	99.62
Day 7	n-2	1664058	15.70	15.94	796.91	0.80	99.60	
Day 14	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 14	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 21	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 28	n-2	0.00	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout- H2O	n-1	0.00	0.00	0.00	0.00	100.00	100.00
Day 29 Washout- H2O	n-2	0.00	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- ethanol	n-1	0.00	0.00	0.00	0.00	0.00	0.00
Day 30Washout- ethanol	n-2	0.00	0.00	0.00	0.00	0.00	

Total amount of drug added in the pouch= 200mg

Loxapine Deactivation Plot

10 mg Tablets



Days	% Deactivated
0	0
0.33	96.9
1	96.5
2	99.6
4	100
7	100
14	100
21	100
28	100

Meperidine Protocol

Protocol

Meperidine Adsorption Study

1) 10 tablets of 50 mg meperidine were placed in pouch containing activated carbon
2) 50 ml warm water (43 degree celcius) was added to the pouch and wait for 30 seconds
3) Seal the pouch and gently shake it from side to side
4) Place it in upright position and take samples at 8hr, Days 1,2,4,7,14,21,28 time point.

Filtration of Samples

Pouch was shaken before taking the samples

1) Sample dilution:
   a) 0.1ml of sample was taken from pouch(8h,1D) and diluted to 1ml with distilled water.
   b) 0.1ml of sample was taken from pouch(2,4,7,14,21,28D).
2) 500ul of diluted or original sample was taken and filtered using 0.22 micron filter.
3) Samples were analyzed by HPLC

Desorption Study

1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml DI water was added to 500ml bottles
4) Samples were rocked on shaker for 1 hour (50rpm)
5) Samples were analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50rpm)
8) Samples were analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

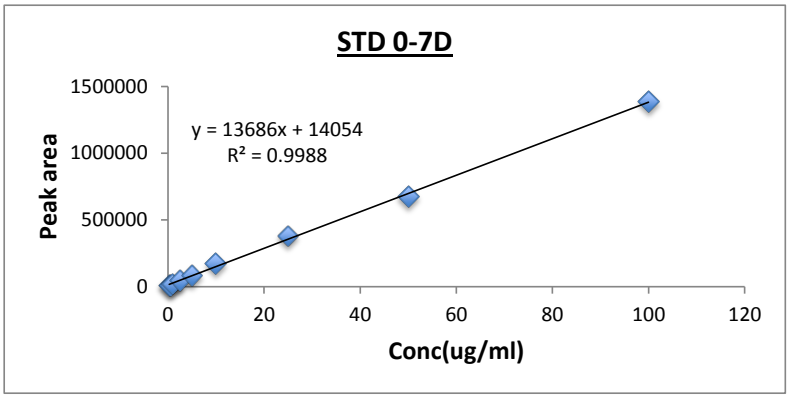
Chromatographic Conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector
Mobile Phase: ACN : NH4OAc Buffer(0.02M,pH 6.9)=35:65
Flow Rate: 1 ml/min
Run Time: 10 min
Retention Time: 5.5 min
Column: Phenomenex, Kinetex C18 5µm, 250x3.5mm
Injection Volume: 20 µl
Column Temperature: 40 °C
Detection Wavelength: 285 nm
Standards were made in distilled water.

Meperidine Standard Curve

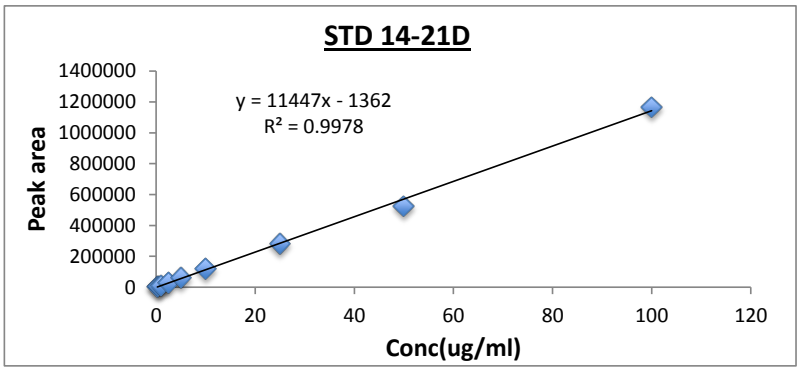
Day 0-7

Conc(ug/ml)	Peak Area
0.25	6795
0.5	10259
1	20856
2.5	47448
5	83572
10	174758
25	380664
50	674329
100	1386227



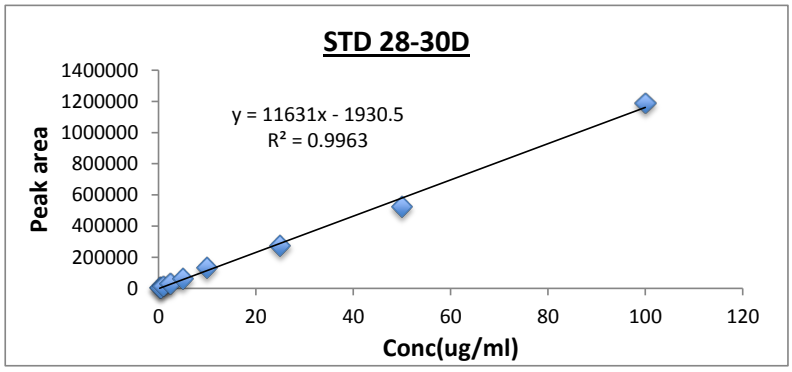
Day 14-21

Conc(ug/ml)	Peak Area
0.25	3353
0.5	6405
1	12331
2.5	31078
5	64061
10	120890
25	282063
50	525889
100	1165261



Day 28-30

Conc(ug/ml)	Peak Area
0.25	3579
0.5	6396
1	14456
2.5	31021
5	63124
10	133841
25	275619
50	524193
100	1189770



Meperidine Deactivation

Time(days)	Pouch	Peak Area	Conc(ug/ml)	Dilution Factor	Original Conc(ug/ml)	Amount of drug in 50 ml(mg)	% Reacted	Avg %
	1	281258	19.52	10	195.24	9.76	98.05	
0.33	2	314815	21.98	10	219.76	10.99	97.80	97.93
	1	69406	4.04	10	40.44	2.02	99.60	
1	2	73834	4.37	10	43.68	2.18	99.56	99.58
	1	226339	15.51	0	15.51	0.78	99.84	
2	2	225205	15.43	0	15.43	0.77	99.85	99.85
	1	163715	10.94	0	10.94	0.55	99.89	
4	2	176868	11.90	0	11.90	0.59	99.88	99.89
	1	84132	5.12	0	5.12	0.26	99.95	
7	2	89775	5.53	0	5.53	0.28	99.94	99.95
	1	13482	1.30	0	1.30	0.06	99.99	
14	2	14922	1.42	0	1.42	0.07	99.99	99.99
	1	8919	0.90	0	0.90	0.04	99.99	
21	2	10241	1.01	0	1.01	0.05	99.99	99.99
	1	2648	0.39	0	0.39	0.02	100.00	
28	2	2354	0.37	0	0.37	0.02	100.00	100.00

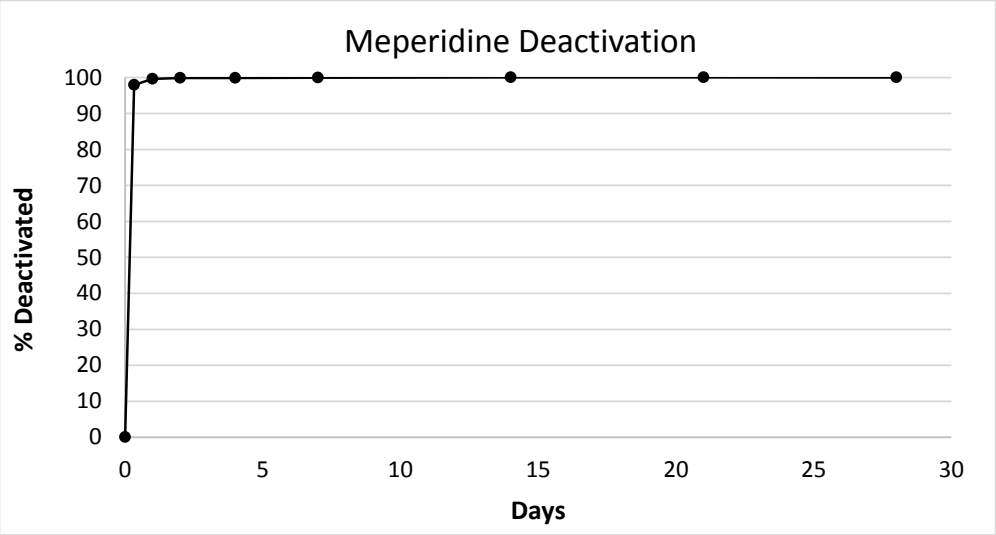
Original amount of drug in the pouch = 50*10 = 500mg

Meperidine Desorption

Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Reacted	Average
Day 29 Washout-H2O	1	0	0.00	0.00	0.00	100.00	100.00
Day 29 Washout-H2O	2	0	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Leached out in ethanol	Average
Day 30 Washout-30% Ethanol	1	292440	25.31	6327.28	6.33	1.27	1.40
Day 30 Washout-30% Ethanol	2	355372	30.72	7679.96	7.68	1.54	

Meperidine Deactivation - Mercer

50 mg Tablet



Days	% Deactivated
0	0
0.33	97.9
1	99.6
2	99.8
4	99.9
7	99.9
14	100
21	100
28	100

Methadone Protocol

Protocol

Methadone Adsorption Study

1) 10 tablets of Methadone Hydrochloride were placed in pouch
2) 50ml warm water (43 degree celcius) was added to the pouch and wait for 30 seconds
3) Seal the pouch and gently shake it from side to side
4) Place it in upright position and take samples at 8hr, Days 1, 2,4,7,14,21, 28 time point

Filtration of Samples

Pouch was shaken before taking the samples

1) Sample dilution:
   a) 0.1ml of sample was taken from pouch(8h, 1D) and diluted to 2ml with distilled water.
   b) 0.1ml of sample was taken from pouch(2D) and diluted to 1ml with distilled water.
   c) 1 ml of sample was taken from pouch(4,7,14,21,28D) .
2) 500ul of diluted sample was taken and filtered using 0.22 micron filter.
3) Samples were analyzed by HPLC

Desorption Study

1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml DI water was added to 500ml bottles
4) Samples were rocked on shaker for 1 hour (50rpm)
5) Samples were analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50rpm)
8) Samples were analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

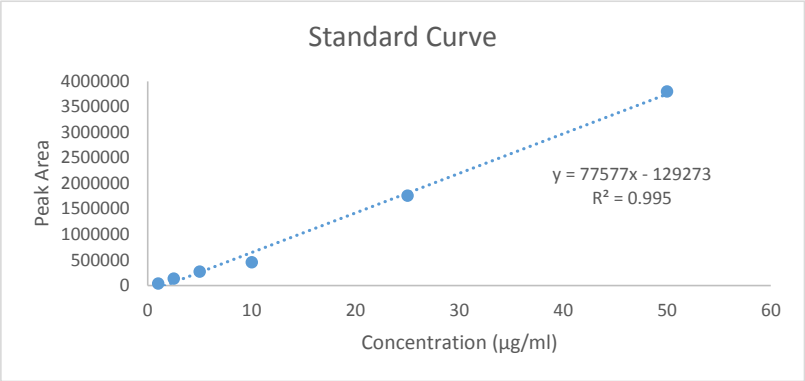
Chromatographic Conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector
Mobile Phase: ACN : Water=60:40(0.05% TFA)
Flow Rate: 1 ml/min
Run Time: 8 min
Retention Time: 5.7 min
Column: Kinetex C18 5µ column(250×4.6mm)
Injection Volume: 30 µl
Column Temperature: 30 °C
Detection Wavelength: 200 nm
Standards were made in distilled water.

Methadone Standard Curve

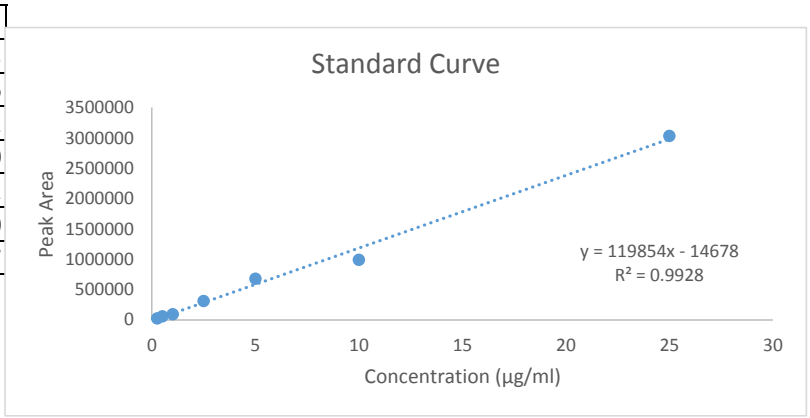
Day 0-7

Conc(µg/mL)	Peak Area
1	41642
2.5	136792
5	274167
10	458070
25	1763684
50	3803497



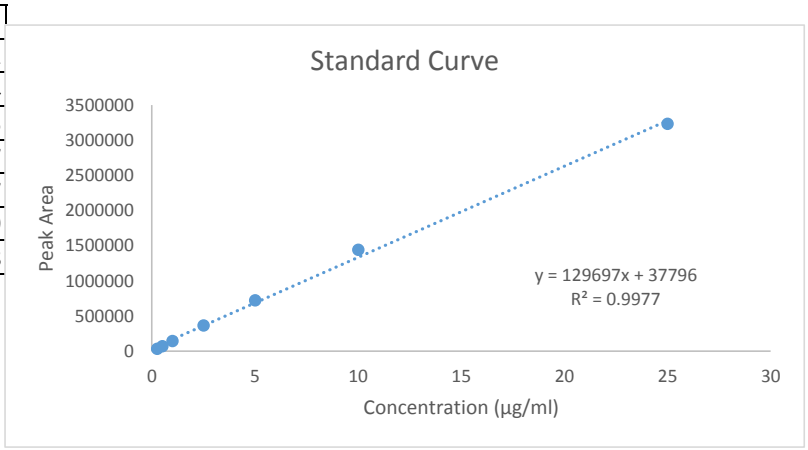
Day 14-21

Conc(µg/mL)	Peak Area
0.25	24393
0.5	59426
1	95981
2.5	311750
5	680251
10	992660
25	3036337



Day 28-30

Conc(µg/mL)	Peak Area
0.25	32341
0.5	68114
1	143645
2.5	364387
5	720977
10	1442049
25	3232168



Methadone Deactivation

Time(days)	Pouch	Peak Area	Conc(µg/ml)	Dilution Factor	Original Conc(µg/mL)	Amount of drug in 50 ml(mg)	% Reacted	Avg %
	1	99483	2.95	20	58.98	2.95	97.05	
0.33	2	89202	2.82	20	56.32	2.82	97.18	97.12
	1	87424	2.79	20	55.87	2.79	97.21	
1	2	92042	2.85	20	57.06	2.85	97.15	97.18
	1	107004	3.05	10	30.46	1.52	98.48	
2	2	153223	3.64	10	36.41	1.82	98.18	98.33
	1	129968	3.34	1	3.34	0.17	99.83	
4	2	103621	3.00	1	3.00	0.15	99.85	99.84
	1	111519	3.10	1	3.10	0.16	99.84	
7	2	94559	2.89	1	2.89	0.14	99.95	99.90
	1	145930	1.34	1	1.34	0.07	99.98	
14	2	176112	1.59	1	1.59	0.08	99.97	99.98
	1	158654	1.45	1	1.45	0.07	99.98	
21	2	169765	1.54	1	1.54	0.08	99.97	99.98
	1	144310	0.82	1	0.82	0.04	99.99	
28	2	168186	1.01	1	1.01	0.05	99.98	99.98

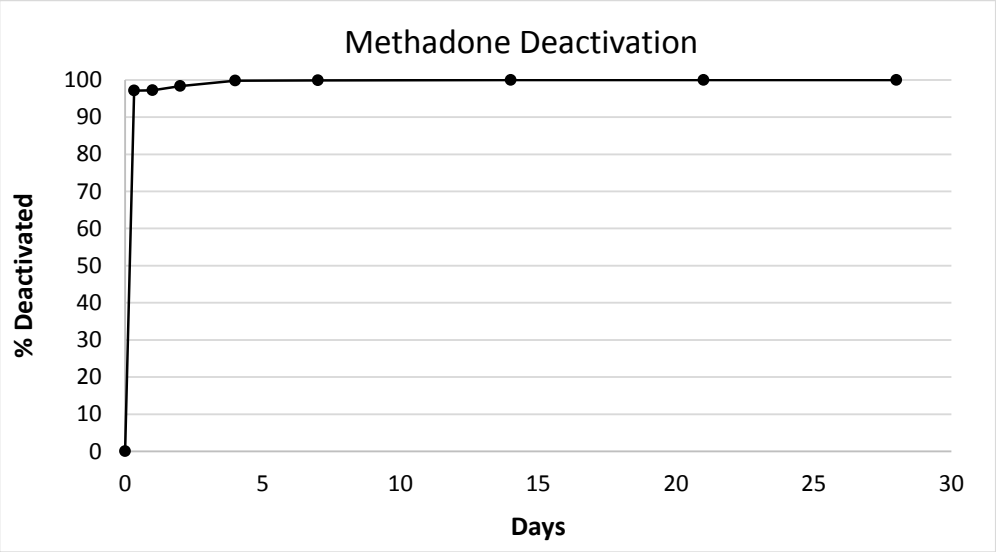
Original amount of drug in each pouch: 10* 10=100mg

Methadone Desorption

Time	Replicate	Peak Area	Conc(µg/ml)	Amount of drug in 250ml(µg)	Amount of drug(mg)	% Reacted	Average
Day 29 Washout-H2O	1	156278	0.91	228.38	0.23	99.77	99.80
Day 29 Washout-H2O	2	128150	0.70	174.16	0.17	99.83	
Time	Replicate	Peak Area	Conc(µg/ml)	Amount of drug in 250ml(µg)	Amount of drug(mg)	% Leached out in ethanol	Average
Day 30 Washout-30% Ethanol	1	148675	0.85	213.73	0.30	0.30	0.25
Day 30 Washout-30% Ethanol	2	140798	0.79	198.54	0.20	0.20	

Methadone Deactivation - Mercer

10 mg Tablet



Days	% Deactivated
0	0
0.33	97.1
1	97.2
2	98.3
4	99.8
7	99.9
14	100
21	100
28	100

Methylphenidate protocol

Methylphenidate

Date: 05/19/15

Protocol:

1) 10 tablets of Methylphenidate were placed in pouch
2) 50ml warm water (43 degree celcius) was added to the pouch and was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Filtration of samples:

Pouch was shaken before taking the samples

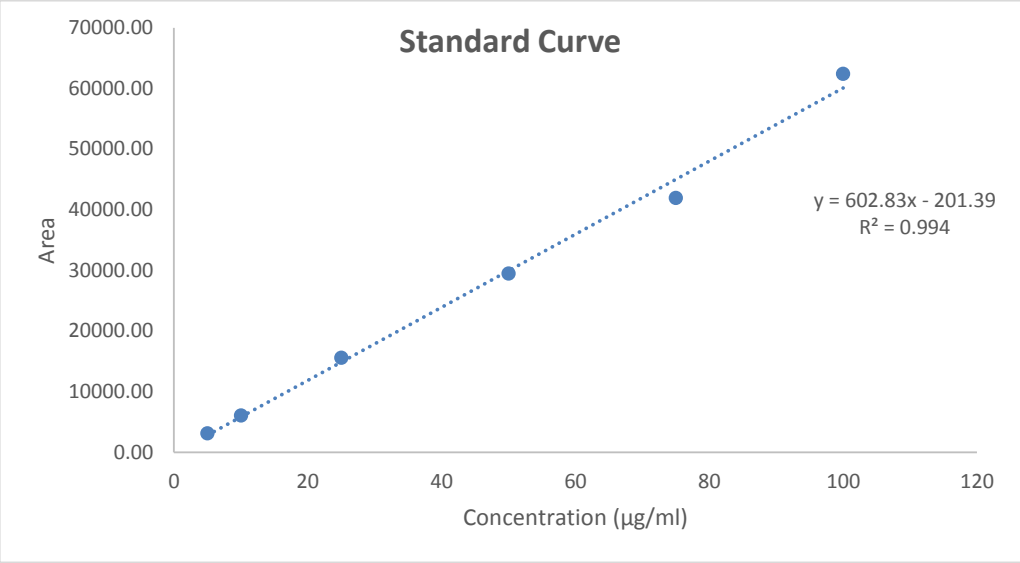
1) 1ml of sample was taken from syringe and filtered through 0.45 micron syringe filter
2) This filtered samples was analyzed by using HPLC.

Chromatographic conditions:

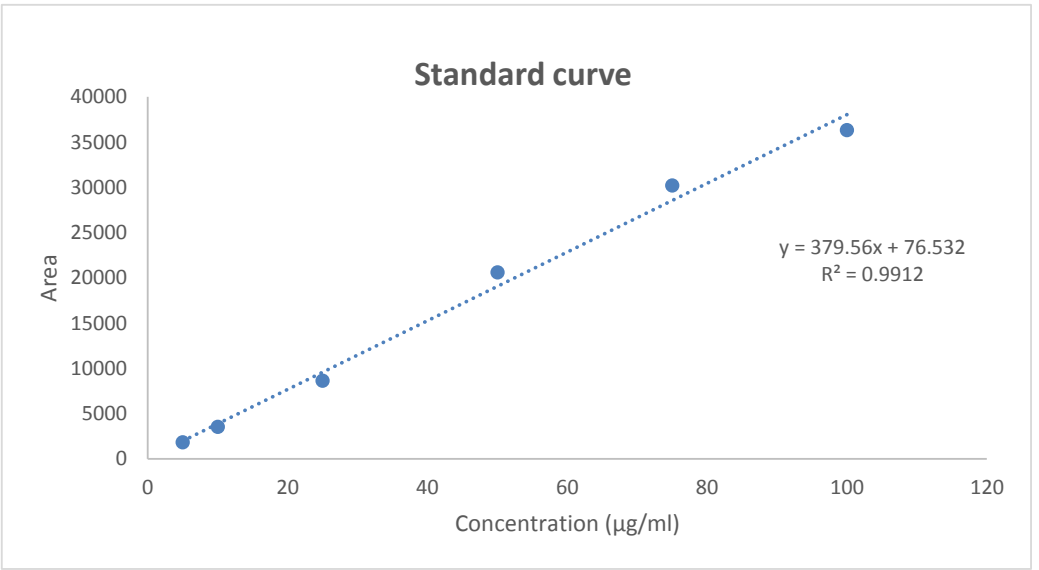
System: Waters e2795
Detector: Waters 2988 photodiode array
Pump Mode: Isocratic
Mobile Phase: Methanol (0.1% FA): water (0.1% FA, pH 6.8) (50:50)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 15 min
Retention time: 9.8min
Column: (size and particle size): kinetex 5u, Biphenyl 100A, 250* 4.6mm
Column Temperature: 25°C
Detection wavelength: 258nm
Sample Preparation: Standards were preapred in HPLC grade water

Methylphenidate Standard Curve

Standard curve for Methylphenidate- 8hr to Day 14	
Concentration (µg/ml)	Area
5	3112.00
10	6049.00
25	15565.00
50	29472.00
75	41924.00
100	62420.00



Standard curve for Methylphenidate- Day21 to washout studies	
Concentration (µg/ml)	Area
5	1800
10	3513
25	8637
50	20599
75	30196
100	36298



Methylphenidate Deactivation/Desorption

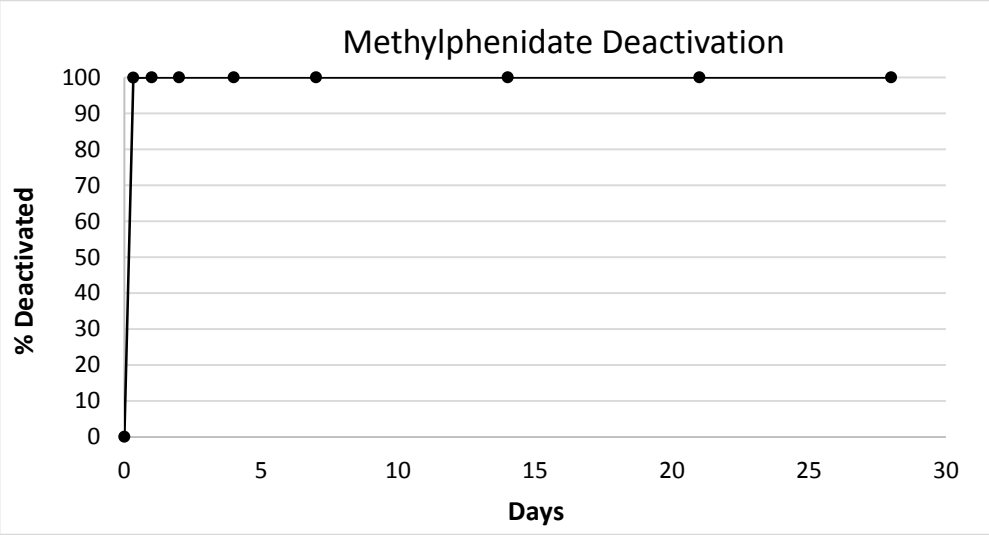
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	2757.00	4.96	4.96	248.00	0.25	99.88	99.87
8hr	n-2	2928.00	5.24	5.24	262.00	0.26	99.87	
Day 1	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 1	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 2	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 2	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 4	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 4	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 7	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 7	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 14	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 14	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 21	n-2	0.00	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0.00	0.00	0.00	0.00	0.00	100.00	100.00
Day 28	n-2	0.00	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout- H2O	n-1	0.00	0.00	0.00	0.00	100.00	100.00
Day 29 Washout- H2O	n-2	0.00	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc (ug/ml)	Amount of drug in 250ml (ug)	Amount of drug (mg)	% Leached out in ethanol	Average
Day 30 Washout- ethanol	n-1	3210.00	8.26	2063.88	2.06	1.03	1.01
Day 30Washout- ethanol	n-2	3051.00	7.84	1959.16	1.96	0.98	

Total amount of drug added to the pouch-200mg

Methylphenidate Deactivation - Mercer

20 mg Tablet



Days	% Deactivated
0	0
0.33	99.9
1	100
2	100
4	100
7	100
14	100
21	100
28	100

Morphine Protocol

Protocol

Morphine Adsorption Study

1) 20 ml morphine solution was placed in pouch
2) 30 ml warm water (43 degree celcius) was added to the pouch and wait for 30 seconds
3) Seal the pouch and gently shake it from side to side
4) Place it in upright position and take samples at 8hr, Days 1,2,4,7,14,21,28 time point.

Filtration of Samples

Pouch was shaken before taking the samples

1) Sample dilution:
   a) 0.1ml of sample was taken from pouch(8h,1D) and diluted to 2ml with distilled water.
   b) 0.1ml of sample was taken from pouch(2,4,7,14,21,28D).
2) 500ul of diluted or original sample was taken and filtered using 0.22 micron filter.
3) Samples were analyzed by HPLC

Desorption Study

1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml DI water was added to 500ml bottles
4) Samples were rocked on shaker for 1 hour (50rpm)
5) Samples were analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50rpm)
8) Samples were analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

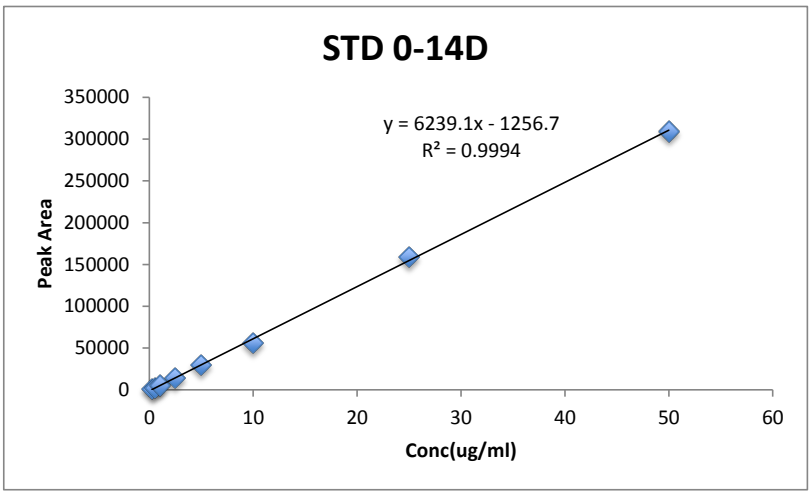
Chromatographic Conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector
Mobile Phase: ACN : NH4OAc Buffer(0.01M, PH5.5)=10:90
Flow Rate: 1 ml/min
Run Time: 10 min
Retention Time: 4 min
Column: Kinetex C18 5µ column(150 ×4.6mm)
Injection Volume: 30 µl
Column Temperature: 45 °C
Detection Wavelength: 285 nm
Standards were made in distilled water.

Morphine Standard Curve

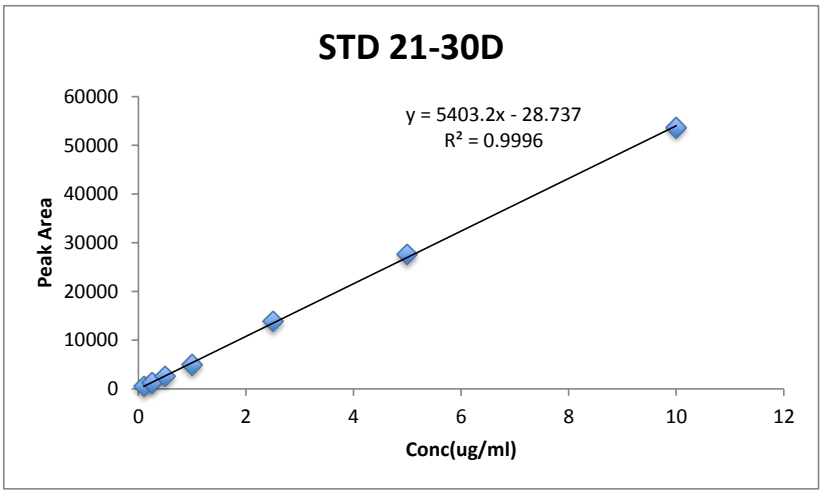
Day 0-14

Conc(ug/ml)	Peak Area
0.25	1383
0.5	2644
1	5311
2.5	14278
5	29672
10	56006
25	159188
50	309502



Day 21-30

Conc(ug/ml)	Peak Area
0.1	467
0.25	1201
0.5	2594
1	4973
2.5	13841
5	27641
10	53633



Morphine Deactivation

Time(days)	Pouch	Peak Area	Conc(ug/ml)	Dilution Factor	Original Conc(ug/ml)	Amount of drug in 50 ml(mg)	% Reacted	Avg %
	1	1779	0.49	20	9.73	0.49	99.84	
0.33	2	1572	0.45	20	9.07	0.45	99.85	99.84
	1	1500	0.44	20	8.84	0.44	99.85	
1	2	1738	0.48	20	9.60	0.48	99.84	99.85
	1	1658	0.47	1	0.47	0.02	99.99	
2	2	1029	0.37	1	0.37	0.02	99.99	99.99
	1	1816	0.49	1	0.49	0.02	99.99	
4	2	784	0.33	1	0.33	0.02	99.99	99.99
	1	901	0.35	1	0.35	0.02	99.99	
7	2	443	0.27	1	0.27	0.01	100.00	99.99
	1	367	0.26	1	0.26	0.01	100.00	
14	2	554	0.29	1	0.29	0.01	100.00	100.00
	1	0	0.00	1	0.00	0.00	100.00	
21	2	0	0.00	1	0.00	0.00	100.00	100.00
	1	0	0.00	1	0.00	0.00	100.00	
28	2	0	0.00	1	0.00	0.00	100.00	100.00

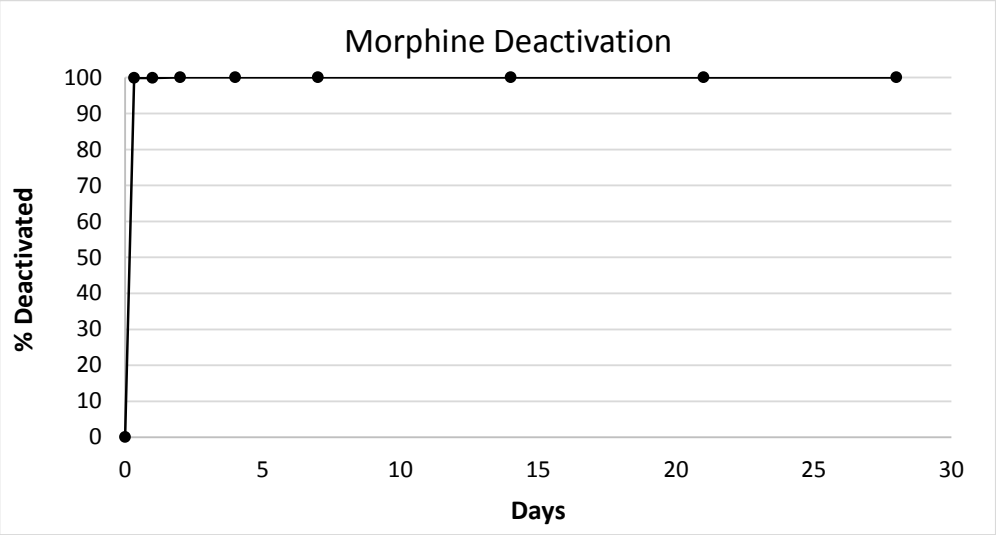
Original amount of drug in the pouch=15mg/ml*20ml=300mg

Morphine Desorption

Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Reacted	Average
Day 29 Washout-H2O	1	0	0.00	0.00	0.00	100.00	100.00
Day 29 Washout-H2O	2	0	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Leached out in ethanol	Average
Day 30 Washout-30% Ethanol	1	37120	6.88	1718.83	1.72	1.72	1.56
Day 30 Washout-30% Ethanol	2	30358	5.62	1405.96	1.41	1.41	

Morphine Deactivation - Mercer

Liquid. 22 ml of 15 mg/ml



Days	% Deactivated
0	0
0.33	99.8
1	99.9
2	100
4	100
7	100
14	100
21	100
28	100

Oxycodone Protocol

Protocol

Oxycodone Adsorption Study

1) 10 tablets of Endocet (10mg oxycodone hydrochloride/325mg acetaminophen) were placed in pouch
2) 50ml warm water (43 degree celcius) was added to the pouch and wait for 30 seconds
3) Seal the pouch and gently shake it from side to side
4) Place it in upright position and take samples at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Filtration of Samples

Pouch was shaken before taking the samples

1) Sample dilution:
   a) 0.1ml of sample was taken from pouch(8h, 1,2,4,7D) and diluted to 16ml with distilled water.
   b) 0.1ml of sample was taken from pouch(14,21,28D) and diluted to 2ml with distilled water.
2) 500ul of diluted sample was taken and filtered using 0.22 micron filter.
3) Samples were analyzed by HPLC

Desorption Study

1) 28th Day pouches were used.
2) Contents of the pouch were transferred to 500ml bottles.
3) 200ml DI water was added to 500ml bottles
4) Samples were rocked on shaker for 1 hour (50rpm)
5) Samples were analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50rpm)
8) Samples were analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

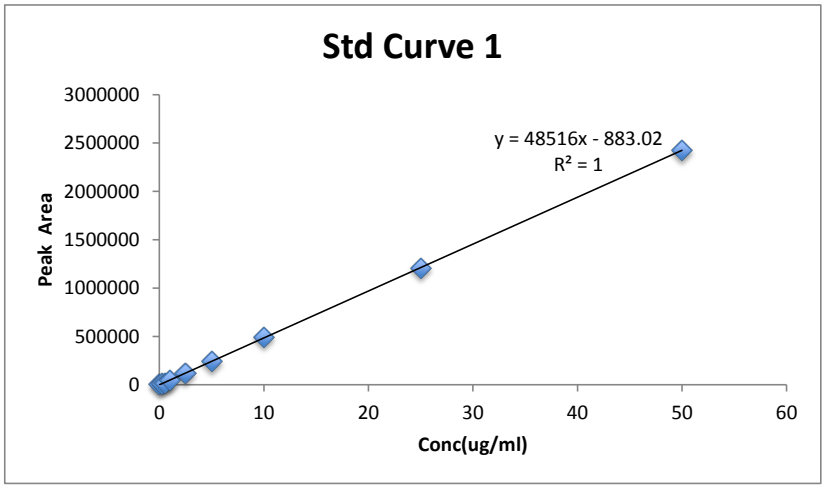
Chromatographic Conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector
Mobile Phase: ACN : NaH_2PO_4 Buffer(0.02M, PH 3.4)=8 : 92
Flow Rate: 1 ml/min
Run Time: 15 min
Retention Time: 9 min
Column: Kinetex C18 5µ column(250×4.6mm)
Injection Volume: 10 µl
Column Temperature: 25 °C
Detection Wavelength: 205 nm
Standards were made in distilled water.

Oxycodone Standard Curve

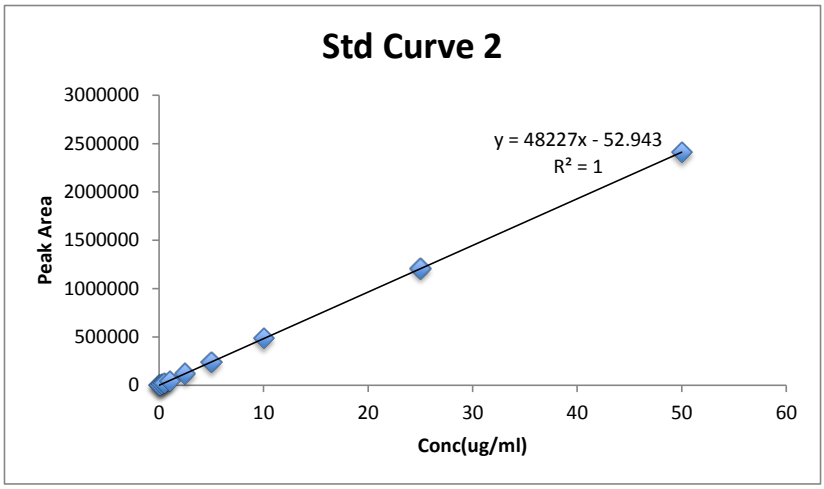
0-4 day

Conc(ug/ml)	Peak Area
0	0
0.25	11574
0.5	17653
1	43913
2.5	120054
5	242738
10	495496
25	1209087
50	2424206



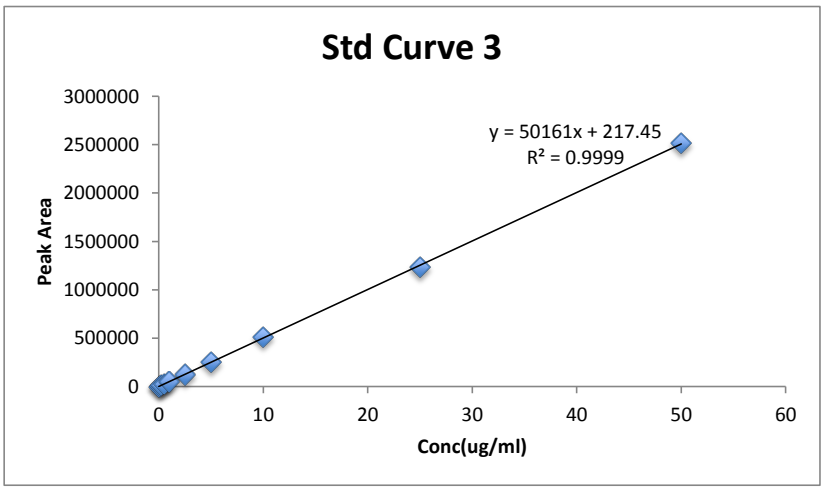
7 day

Conc(ug/ml)	Peak Area
0	0
0.1	4202
0.25	11480
0.5	23901
1	47852
2.5	120526
5	239807
10	484992
25	1206499
50	2410472



14-30 day

Conc(ug/ml)	Peak Area
0	0
0.1	5450
0.25	12257
0.5	25588
1	51349
2.5	123202
5	250547
10	515362
25	1236566
50	2514554



Oxycodone Deactivation

Time(days)	Pouch	Peak Area	Conc(ug/ml)	Dilution Factor	Original Conc(ug/ml)	Amount of drug in 50 ml(mg)	% Reacted	Avg %
	1	86491	1.80	160	288.15	14.41	85.59	
0.33	2	102611	2.13	160	341.31	17.07	82.93	84.26
	1	83462	1.74	160	278.16	13.91	86.09	
1	2	96725	2.01	160	321.90	16.09	83.91	85.00
	1	62899	1.31	160	210.35	10.52	89.48	
2	2	97041	2.02	160	322.94	16.15	83.85	86.67
	1	43150	0.91	160	145.22	7.26	92.74	
4	2	51542	1.08	160	172.89	8.64	91.36	92.05
	1	22054	0.46	160	73.34	3.67	96.33	
7	2	7827	0.16	160	26.14	1.31	98.69	97.51
	1	4910	0.09	20	1.87	0.09	99.91	
14	2	16721	0.33	20	6.58	0.33	99.67	99.79
	1	2795	0.05	20	1.03	0.05	99.95	
21	2	17612	0.35	20	6.94	0.35	99.65	99.80
	1	3146	0.06	20	1.17	0.06	99.94	
28	2	6611	0.13	20	2.55	0.13	99.87	99.91

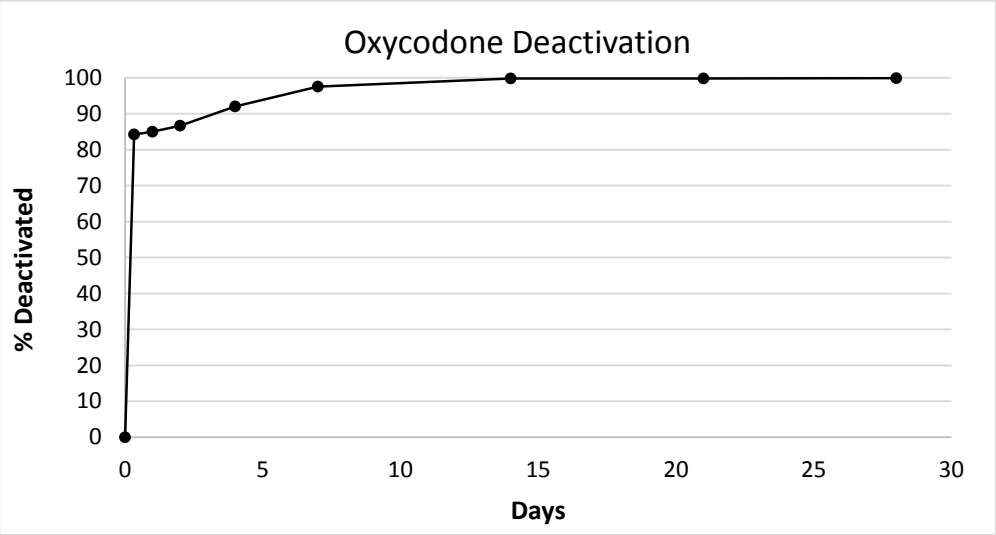
Total Amount in each pouch: 10*10= 100 mg

Oxycodone Desorption

Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Reacted	Average
Day 29 Washout-H2O	1	13714	0.27	67.27	0.07	99.93	99.93
Day 29 Washout-H2O	2	14237	0.28	69.87	0.07	99.93	
Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Leached out in ethanol	Average
Day 30 Washout-30% Ethanol	1	375253	7.48	1869.16	1.87	1.87	1.61
Day 30 Washout-30% Ethanol	2	272077	5.42	1354.93	1.35	1.36	

Oxycodone Deactivation - Mercer

10 mg Tablet (also contains 325 mg APAP)



Days	% Deactivated
0	0
0.33	84.3
1	85.0
2	86.7
4	92.1
7	97.5
14	100
21	100
28	100

Oxycontin Protocol

Controlled release oxycodone (oxycontin) Adsorption Study

1) 10 tablets of Oxycontin (10 mg oxycodone hydrochloride) were placed in pouch containing activated carbon
2) 50 ml warm water (43 degree celcius) was added to the pouch and wait for 30 seconds
3) Seal the pouch and gently shake it from side to side
4) Place it in upright position and take samples at 8hr, Days 1, 2,4,7,14,21, 28 time point.

Filtration of Samples

Pouch was shaken before taking the samples

1) Sample dilution:
   a) Contents from pouch (8h,1,2,4,7,14,21,28D) were decanted and diluted to 500 ml with distilled water. Followed by rocking overnight on the shaker at 120 rpm
2) 500 ul of diluted sample was taken and filtered using 0.22 micron filter.
3) Samples were analyzed by HPLC

Desorption Study

1) 28th Day pouches were used.
2) Carbon contents of the pouch were transferred to 500 ml bottles.
3) 250 ml of DI water was added to 500 ml bottles
4) Samples were rocked on shaker for 1 hour (50 rpm)
5) Samples were filtered and analyzed after additional 23 hours exposure. (Total 24 hours)
6) Water was replaced with 250 ml 30% (v/v) ethanol
7) Samples were rocked on shaker for 1 hour (50 rpm)
8) Samples were filtered and analyzed by HPLC after additional 23 hours exposure. (Total 24 hours)

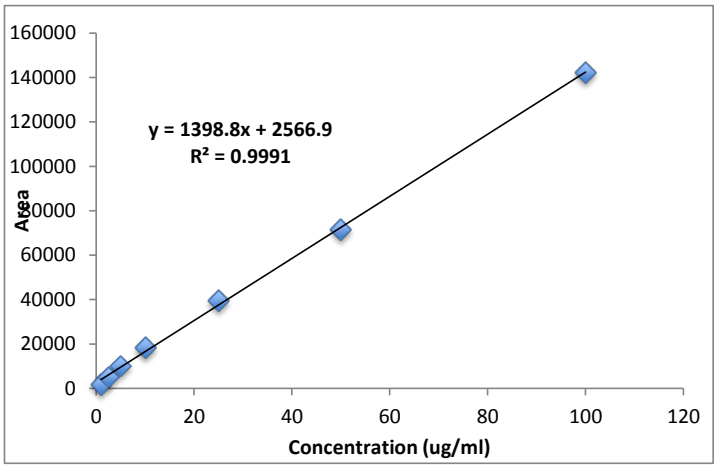
Chromatographic Conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector
Mobile Phase: ACN : Buffer (DI water with 0.1% TFA, PH 1.5) (15:85)
Flow Rate: 1.2 ml/min
Run Time: 10 min
Retention Time: 3.6 min
Column: Kinetex C18 5µ column(250×4.6mm)
Injection Volume: 10 µl
Column Temperature: 55 °C
Detection Wavelength: 285 nm
Standards were made in distilled water.

Oxycontin Standard Curve

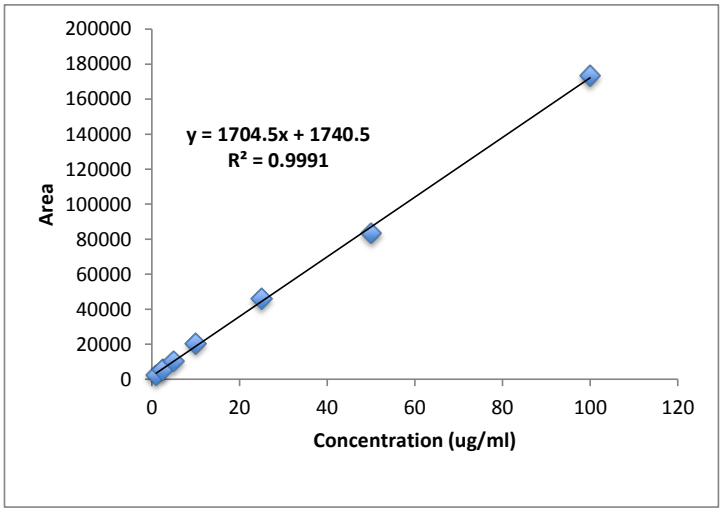
0-14 day

Conc(ug/ml)	Peak Area
1	1786
2.5	5019
5	10056
10	18377
25	39598
50	71553
100	142255



21-30 day

Conc(ug/ml)	Peak Area
1	2519
2.5	5679
5	10302
10	20561
25	46163
50	83444
100	173337



Oxycontin Deactivation

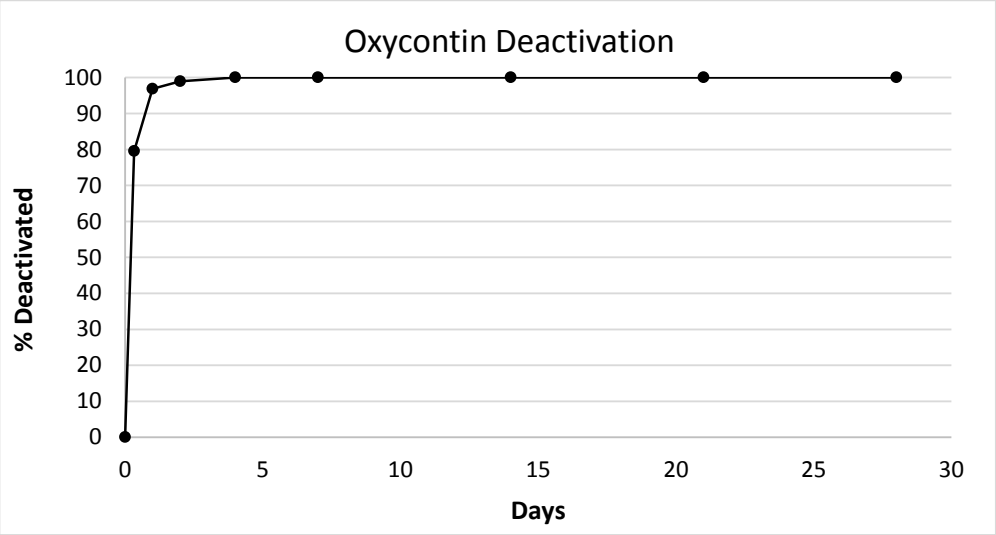
Time(days)	Pouch	Peak Area	Conc(ug/ml)	Dilution Factor	Original Conc(ug/ml)	Amount of drug in 50	% Reacted	Avg %
	1	60141	41.16	10.00	411.60	20.58	79.42	
0.33	2	59231	40.51	10.00	405.09	20.25	79.75	79.58
	1	10569	5.72	10.00	57.21	2.86	97.14	
1	2	12401	7.03	10.00	70.30	3.52	96.48	96.81
	1	5626	2.19	10.00	21.87	1.09	98.91	
2	2	5512	2.11	10.00	21.05	1.05	98.95	98.93
	1	0	0.00	10.00	0.00	0.00	100.00	
4	2	0	0.00	10.00	0.00	0.00	100.00	100.00
	1	0	0.00	10.00	0.00	0.00	100.00	
7	2	0	0.00	10.00	0.00	0.00	100.00	100.00
	1	0	0.00	10.00	0.00	0.00	100.00	
14	2	0	0.00	10.00	0.00	0.00	100.00	100.00
	1	0	0.00	10.00	0.00	0.00	100.00	
21	2	0	0.00	10.00	0.00	0.00	100.00	100.00
	1	0	0.00	10.00	0.00	0.00	100.00	
28	2	0	0.00	10.00	0.00	0.00	100.00	100.00

Oxycontin Desorption

Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Reacted	Average
Day 29 Washout-H2O	1	0	0.00	0.00	0.00	100.00	100.00
Day 29 Washout-H2O	2	0	0.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc(ug/ml)	Amount of drug in 250ml(ug)	Amount of drug(mg)	% Leached out in ethanol	Average
Day 30 Washout-30% Ethanol	1	3098	0.80	199.11	0.20	0.20	0.21
Day 30 Washout-30% Ethanol	2	3283	0.90	226.24	0.23	0.23	

Oxycontin Deactivation - Mercer

10 mg Tablet



Days	% Deactivated
0	0
0.33	79.6
1	96.8
2	98.9
4	100
7	100
14	100
21	100
28	100

Quetiapine Fumarate
Date of Report: June 05, 2016

Protocol:
1) 10 tablets of Quetiapine Fumarate (100 mg) were placed in a pouch
2) 50mL warm water (43 °C) was added to the pouch and it was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side
4) They were then placed in a upright position and samples were taken
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time points.

Washout Protocol:
1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken
3) Water was replaced with 250 mL of 30%(v/v) ethanol
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken

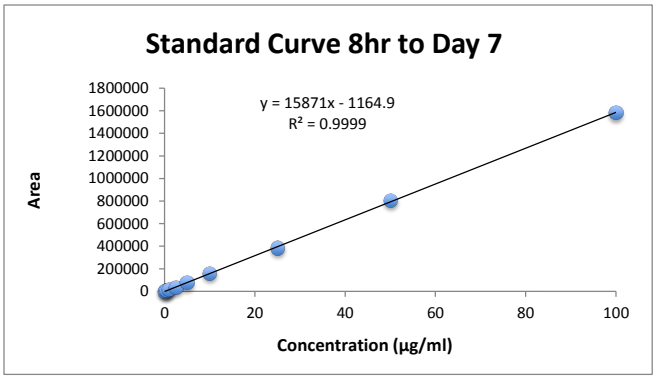
Filtration of samples:
1) Pouch was shaken before taking the samples
2) 2mL of sample was taken by a syringe and centrifuged for 5 minutes at 13400 rpm
3) The supernatant was filtered through 0.22 µm syringe filter

Chromatographic conditions: **Quetiapine Fumarate**

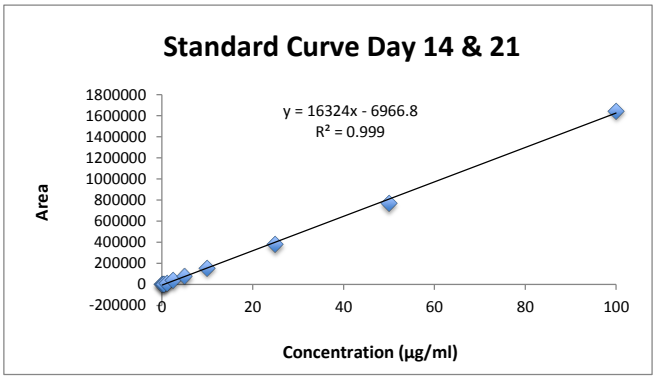
System: Waters 2695 Separations Module
Detector: Waters 2996 Photodiod Array Detector
Pump Mode: Isocratic
Mobile Phase: ACN:Water (0.1% O-phophoric acid (pH 3)) 35:65 (v/v)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 12min
Retention time: 5.9 min
Column: (size and particle size): Waters, XBridge C18, 3.5µm, 250×4.6mm
Column Temperature: 40°C
Wavelength 293nm
Sample Preparation: Standards were prepared in Acetonitrile:H2O 35:65 (v/v), All samples were diluted 100x in ACN:H2O 35:65 (v/v) except samples from day 14 n=1, day 28, and wash out studies with water and ethanol

Quetiapine Standard Curve

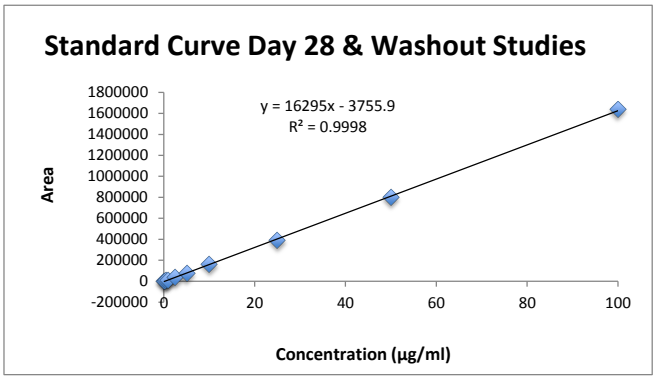
Standard curve - 8hr to Day 7	
Concentration (µg/ml)	Area
0.1	1303
0.25	4547
0.5	7566
1	15605
2.5	37320
5	77562
10	156809
25	384929
50	804660
100	1582597



Standard curve - Day 14 & 21	
Concentration (µg/ml)	Area
0.1	2051
0.25	3806
0.5	7386
1	15580
2.5	37035
5	75560
10	159892
25	381045
50	772098
100	1648355



Standard curve - Day 28 to washout studies	
Concentration (µg/ml)	Area
0.1	1724
0.25	4084
0.5	7906
1	15687
2.5	39308
5	78175
10	158597
25	389904
50	798838
100	1635202



Quetiapine Deactivation/Desorption

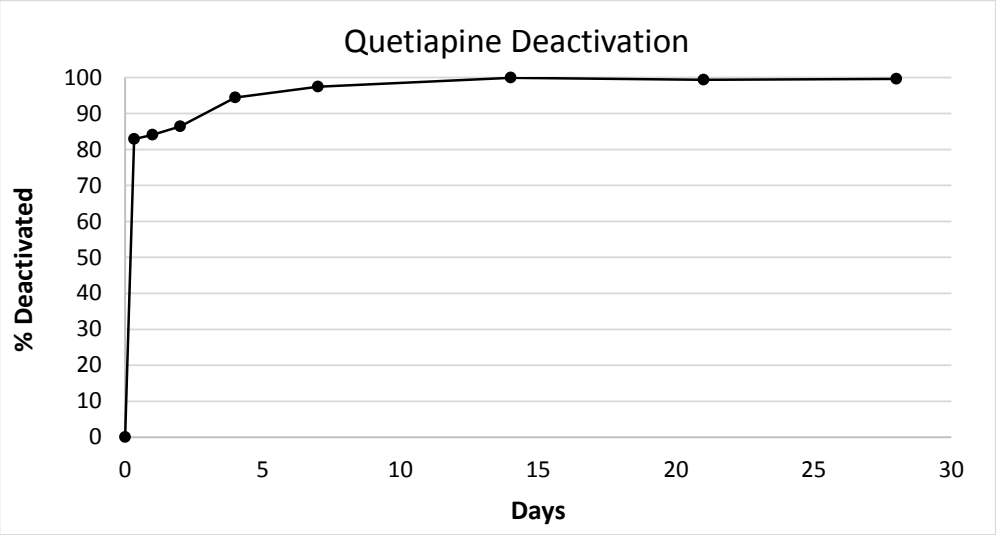
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (100)	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	546229	34.49	100.00	172450.98	172.45	82.75	82.85
	n-2	540209	34.11	100.00	170554.44	170.55	82.94	
Day 1	n-1	499814	31.57	100.00	157828.40	157.83	84.22	84.03
	n-2	511679	32.31	100.00	161566.35	161.57	83.84	
Day 2	n-1	445004	28.11	100.00	140561.05	140.56	85.94	86.41
	n-2	415581	26.26	100.00	131291.63	131.29	86.87	
Day 4	n-1	110657	7.05	100.00	35228.37	35.23	96.48	94.26
	n-2	251153	15.90	100.00	79490.23	79.49	92.05	
Day 7	n-1	109087	6.95	100.00	34733.76	34.73	96.53	97.45
	n-2	50547	3.26	100.00	16291.32	16.29	98.37	
Day 14	n-1	48724	3.41	1.00	170.58	0.17	99.98	99.70
	n-2	12268	1.18	100.00	5891.57	5.89	99.41	
Day 21	n-1	40592	2.91	100.00	14567.14	14.57	98.54	98.91
	n-2	16747	1.45	100.00	7263.48	7.26	99.27	
Day 28	n-1	12554	1.00	100.00	5004.57	5.00	99.50	99.41
	n-2	18270	1.35	100.00	6758.48	6.76	99.32	

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (100) µg/ml	Amount of drug in 200ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout-H2O	n-1	0.00	0.00	1.00	0.00	0.00	100.00	100.00
	n-2	0.00	0.00	1.00	0.00	0.00	100.00	
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (100) µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Leached out in ethanol	Average(%)
Day 30 Washout-Ethanol	n-1	940464	57.95	1.00	14486.34	14.49	1.45	1.17
	n-2	578857	35.75	1.00	8938.52	8.94	0.89	

Total amount of drug added to the pouch = 1000 mg

Quetiapine Deactivation - Mercer

100 mg Tablet



Days	% Deactivated
0	0
0.33	82.9
1	84.0
2	86.4
4	94.4
7	97.5
14	99.9
21	99.3
28	99.6

Temazepam Protocol

Date of report: 03/10/2016

Protocol:

1) 10 capsules of Temazepam (30 mg) were placed in a pouch
2) 50mL warm water (43 °C) was added to the pouch and it was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time points.

Washout Protocol:

1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle.
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.
3) Water was replaced with 250 mL of 30%(v/v) ethanol.
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.

Filtration of samples:

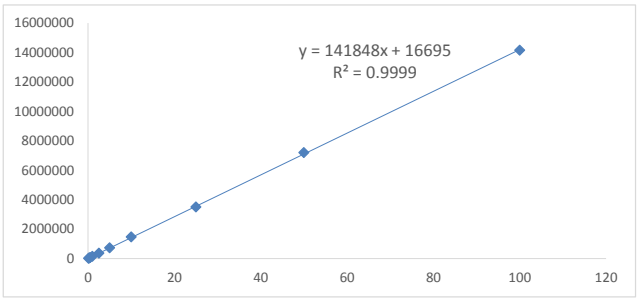
1) Pouch was shaken before taking the samples
2) 2mL of sample was taken by a syringe and centrifuged for 3 minutes at 13400 rpm
3) The supernatant was filtered through 0.45 µm syringe filter
4) Filtered samples was analyzed using HPLC.

Chromatographic conditions: Temazepam

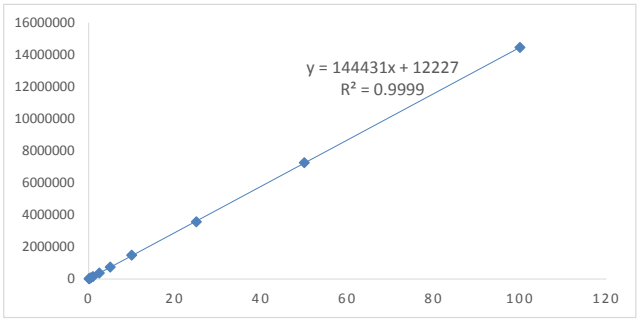
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN:Buffer (KH_2PO_4 0.02M pH=3) (45:55)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 4.8min
Column: (size and particle size): Phenomenex, Kinetex C18 5µm, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 230 nm

Temazepam Standard Curve

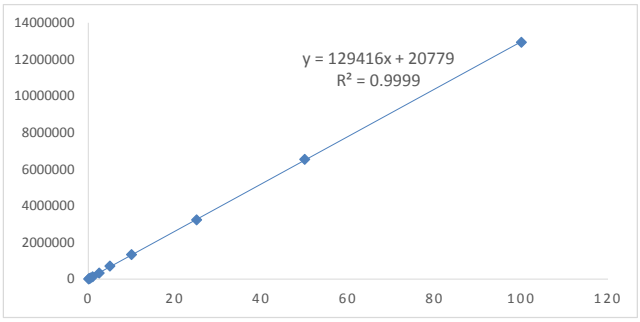
Standard curve - 8hr to Day 7	
Concentration (µg/ml)	Area
0.1	14709
0.25	37036
0.5	74888
1	152649
2.5	364536
5	737741
10	1470351
25	3515765
50	7206799
100	14160719



Standard curve - Day 14 & 21	
Concentration (µg/ml)	Area
0.1	17415
0.25	41240
0.5	77444
1	154271
2.5	374516
5	754384
10	1492017
25	3572111
50	7257299
100	14451908



Standard curve - Day 28 to washout studies	
Concentration (µg/ml)	Area
0.1	12788
0.25	32291
0.5	71216
1	135890
2.5	331757
5	711664
10	1344554
25	3238096
50	6546832
100	12934775



Temazepam Deactivation/Desorption

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	13118512	92.37	0.00	4618.26	4.62	98.46	98.46
	n-2	13141839	92.53	0.00	4626.48	4.63	98.46	
Day 1	n-1	13029863	91.74	0.00	4587.01	4.59	98.47	98.48
	n-2	12941162	91.11	0.00	4555.75	4.56	98.48	
Day 2	n-1	12980132	91.39	0.00	4569.48	4.57	98.48	98.48
	n-2	12928485	91.03	0.00	4551.28	4.55	98.48	
Day 4	n-1	11219187	78.98	0.00	3948.77	3.95	98.68	98.65
	n-2	11792039	83.01	0.00	4150.69	4.15	98.62	
Day 7	n-1	8673617	61.03	0.00	3051.48	3.05	98.98	98.75
	n-2	12554414	88.39	0.00	4419.42	4.42	98.53	
Day 14	n-1	9872375	68.27	0.00	3413.42	3.41	98.86	98.62
	n-2	14131012	97.75	0.00	4887.69	4.89	98.37	
Day 21	n-1	5790654	40.01	0.00	2000.40	2.00	99.33	98.94
	n-2	12526778	86.65	0.00	4332.33	4.33	98.56	
Day 28	n-1	6309678	48.59	0.00	2429.72	2.43	99.19	99.32
	n-2	4301196	33.07	0.00	1653.74	1.65	99.45	

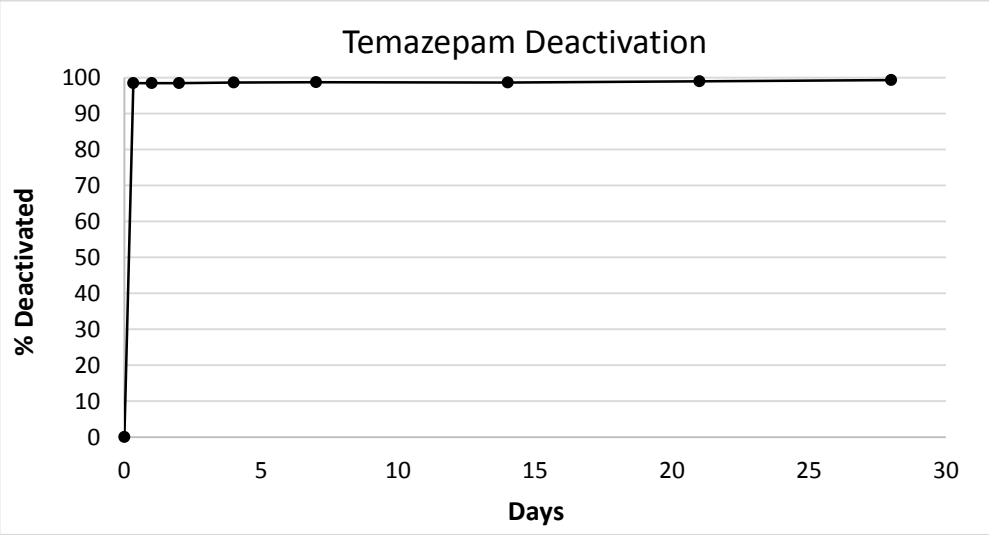
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout-H2O	n-1	129612	0.84	0.00	210.24	0.21	99.93	99.95
	n-2	56021	0.27	0.00	68.08	0.07	99.98	

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Leached out in ethanol	Average(%)
Day 30 Washout-Ethanol	n-1	97822	0.60	0.00	148.83	0.15	0.05	0.04
	n-2	76126	0.43	0.00	106.92	0.11	0.04	

Total amount of drug added to the pouch = 300mg

Temazepam Deactivation - Mercer

2 mg Tablet



Days	% Deactivated
0	0
0.33	98.5
1	98.5
2	98.5
4	98.7
7	98.8
14	98.6
21	98.9
28	99.3

Tramadol Protocol

Protocol:

1) 10 tablets of Tramadol (50 mg) were placed in a pouch.
2) 50mL warm water (43 °C) was added to the pouch and it was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time points.

Washout Protocol:

1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle.
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.
3) Water was replaced with 250 mL of 30%(v/v) ethanol.
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.

Filtration of samples:

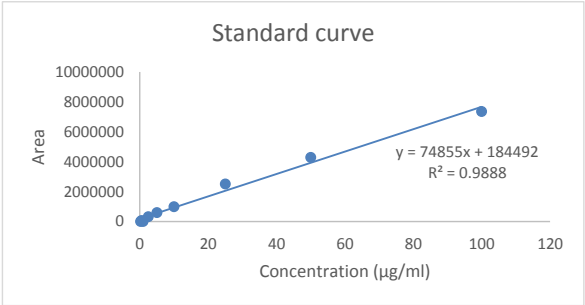
1) Pouch was shaken before taking the samples
2) 2mL of sample was taken by a syringe and centrifuged for 3 minutes at 13400 rpm
3) The supernatant was filtered through 0.45 µm syringe filter
4) 8-hour - Day 2 samples were diluted 10 times with DI water
5) Samples was analyzed using HPLC

Chromatographic conditions: Tramadol

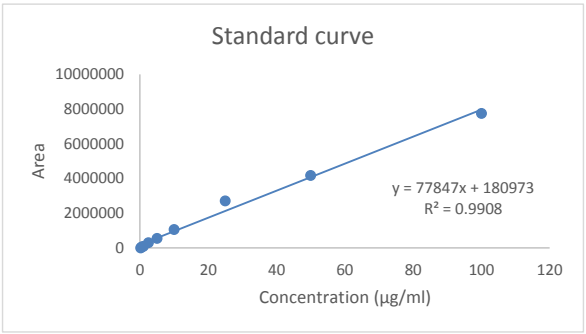
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN : Water (0.1% TFA) (30:70) (% v/v)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 3.4 min
Column: (size and particle size): Phenomenex, Kinetex EVO C18 5µm, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 200 nm
Sample Preparation: Standards were prepared in Water

Tramadol Standard Curve

Standard curve - 8hr to Day 7	
Concentration (µg/ml)	Area
0.25	8633
0.5	66075
1	10201
2.5	327560
5	609107
10	999411
25	2517261
50	4297563
100	7365164



Standard curve Day 14 to washout studies	
Concentration (µg/ml)	Area
0.25	8409
0.5	52763
1	98219
2.5	304334
5	557377
10	1070408
25	2717268
50	4185404
100	7756347



Tramadol Deactivation/Desorption

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	3189788	40.15	10.00	20074.12	20.07	95.99	96.32
	n-2	2694863	33.54	10.00	16768.23	16.77	96.65	
Day 1	n-1	906240	9.64	10.00	4820.97	4.82	99.04	99.00
	n-2	955283	10.30	10.00	5148.56	5.15	98.97	
Day 2	n-1	469360	3.81	0.00	190.28	0.19	99.96	99.97
	n-2	333868	2.00	0.00	99.78	0.10	99.98	
Day 4	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 7	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 14	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 21	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	

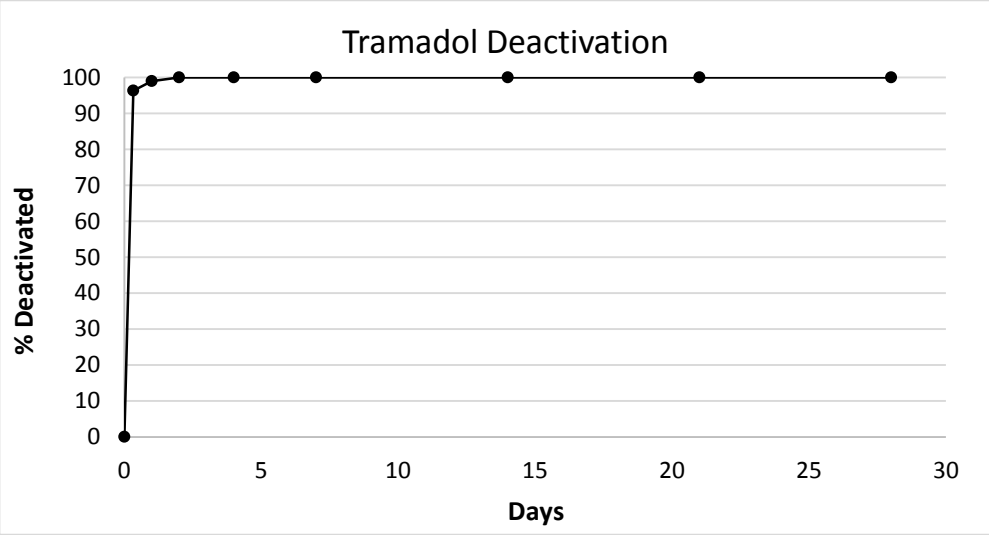
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout-H2O	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Leached out in ethanol	Average(%)
Day 30 Washout-Ethanol	n-1	7578454	95.03	0.00	23756.47	23.76	4.75	4.81
	n-2	7766819	97.45	0.00	24361.39	24.36	4.87	

Total amount of drug added to the pouch = 500mg
Each tablet contains 50mg Tramadol

Tramadol Deactivation - Mercer

50 mg Tablet



Days	% Deactivated
0	0
0.33	96.3
1	99.0
2	100
4	100
7	100
14	100
21	100
28	100

Zolpidem Protocol

Protocol:

1) 10 tablets of Zolpidem tartrate (5 mg) were placed in a pouch
2) 50mL warm water (43 °C) was added to the pouch and it was kept open for 30 seconds
3) Pouch was sealed after 30 seconds and gently shaken from side to side.
4) They were then placed in a upright position and samples were taken.
5) Samples were taken at 8hr, Days 1, 2,4,7,14,21, 28 time points.

Washout Protocol:

1) 28 day pouches' contents were transferred to two 500 ml bottles and 200 mL water was added to each bottle.
2) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.
3) Water was replaced with 250 mL of 30%(v/v) ethanol.
4) Bottles were rocked for 1 hour, and 23 hours after additional exposure samples were taken.

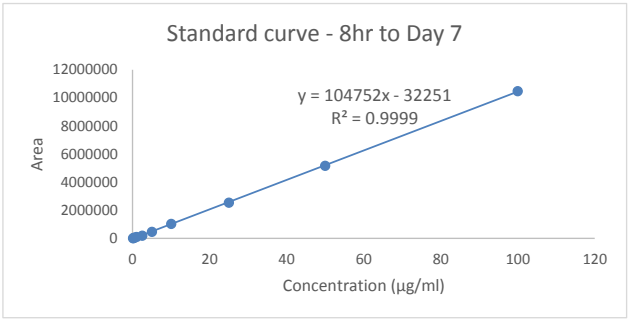
Filtration of samples:

1) Pouch was shaken before taking the samples
2) 2mL of sample was taken by a syringe and centrifuged for 3 minutes at 13400 rpm
3) The supernatant was filtered through 0.45 µm syringe filter
4) 8-hour samples were diluted 5 times with DI water
5) Samples was analyzed using HPLC

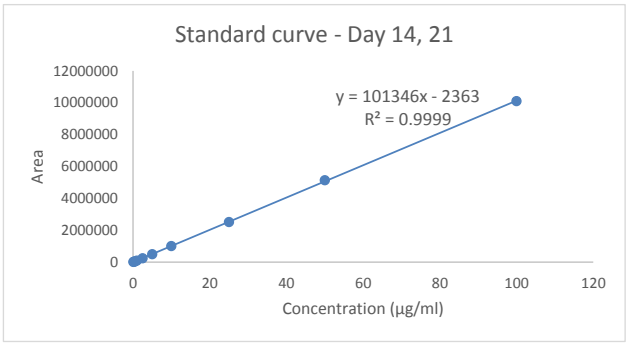
Chromatographic conditions: Zolpidem tartrate
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN:Buffer (KH_2PO_4 25mM pH=6) (40:60)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 5.1min
Column: Agilent ZORBAX Eclipse Plus C 18 (4.6×150mm 5µm)
Column Temperature: 25°C
Detection wavelength: 243 nm

Zolpidem Standard Curve

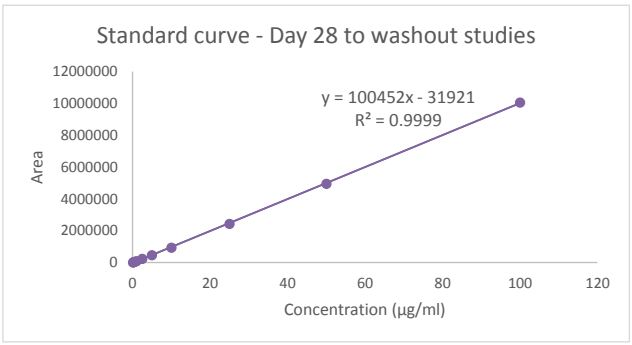
Standard curve - 8hr to Day 7	
Concentration (µg/ml)	Area
0.1	8581
0.25	18471
0.5	44295
1	98710
2.5	194509
5	462565
10	1024431
25	2541934
50	5169686
100	10472888



Standard curve - Day 14 & 21	
Concentration (µg/ml)	Area
0.1	8701
0.25	20242
0.5	46470
1	93437
2.5	243064
5	492330
10	1014586
25	2519811
50	5132454
100	10101868



Standard curve - Day 28 to washout studies	
Concentration (µg/ml)	Area
0.1	8647
0.25	19444
0.5	42801
1	89234
2.5	226347
5	461899
10	932445
25	2422113
50	4947899
100	10052851



Zolpidem Deactivation/Desorption

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 50ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
8hr	n-1	2569748	24.84	5.00	6209.90	6.21	87.58	85.17
	n-2	3580234	34.49	5.00	8621.52	8.62	82.76	
Day 1	n-1	6138782	58.91	0.00	2945.54	2.95	94.11	93.73
	n-2	6932825	66.49	0.00	3324.56	3.32	93.35	
Day 2	n-1	356370	3.71	0.00	185.50	0.19	99.63	99.80
	n-2	7716	0.38	0.00	19.08	0.02	99.96	
Day 4	n-1	28318	0.58	0.00	28.91	0.03	99.94	99.95
	n-2	4589	0.35	0.00	17.58	0.02	99.96	
Day 7	n-1	625	0.31	0.00	15.69	0.02	99.97	99.97
	n-2	488	0.31	0.00	15.63	0.02	99.97	
Day 14	n-1	1671	0.04	0.00	1.99	0.00	100.00	100.00
	n-2	823	0.03	0.00	1.57	0.00	100.00	
Day 21	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	
Day 28	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	

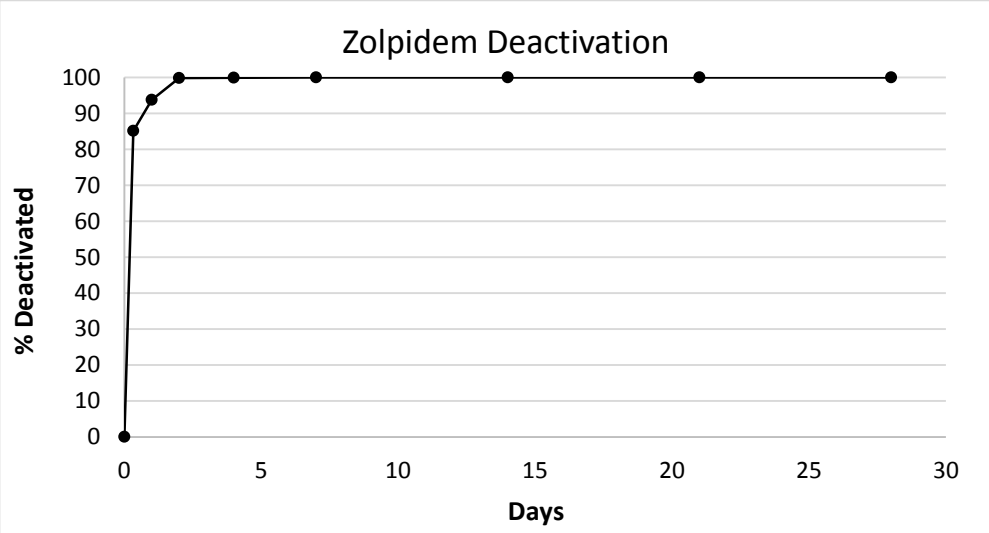
Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Reacted	Average (%)
Day 29 Washout-H2O	n-1	0	0.00	0.00	0.00	0.00	100.00	100.00
	n-2	0	0.00	0.00	0.00	0.00	100.00	

Time	Replicate	Peak Area	Conc (µg/ml)	Dilution Factor (0) µg/ml	Amount of drug in 250ml (µg)	Amount of drug (mg)	% Leached out in ethanol	Average(%)
Day 30 Washout-Ethanol	n-1	10992	0.43	0.00	106.80	0.11	0.21	0.21
	n-2	8713	0.40	0.00	101.13	0.10	0.20	

Total amount of drug added to the pouch = 50mg
Each tablet contains 5mg Zolpidem

Zolpidem Deactivation - Mercer

5 mg Tablet



Days	% Deactivated
0	0
0.33	85.2
1	93.7
2	99.8
4	100
7	100
14	100
21	100
28	100

Appendix B
HPLC Validation Data
(Mercer University)

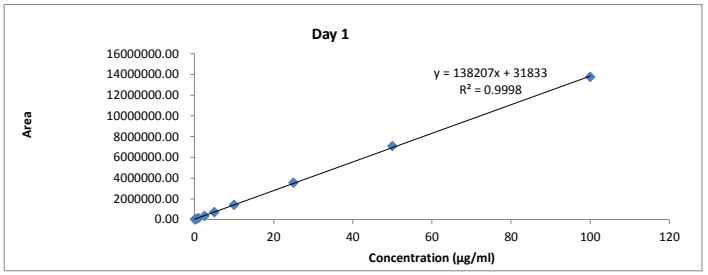
Alprazolam Validation HPLC Conditions

Chromatographic conditions: Aplrazolam

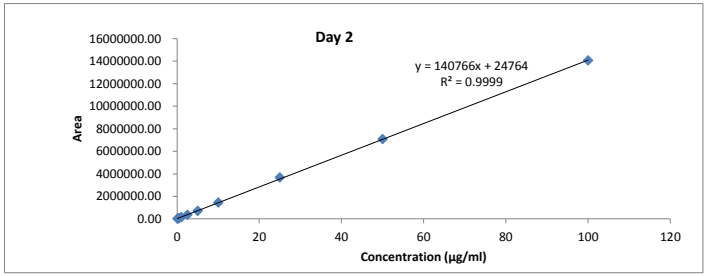
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN:Buffer (KH2PO4 0.01M pH 4.5) (40:60)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 4.7min
Column: (size and particle size): Phenomenex, Kinetex C18 100A, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 221 nm
Sample Preparation: Standards were prepared in ACN and Water (1:1)

Alprazolam Validation Std Curve

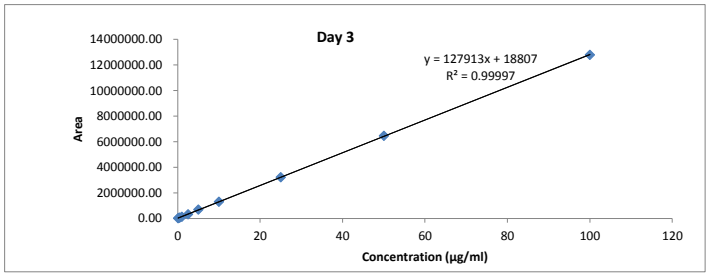
Day 1 (12/15/15)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	14025	16624	14317	14988.67	1423.75	9.50
0.25	34965	38882	38878	37575.00	2260.33	6.02
0.5	76251	74299	73594	74714.67	1376.41	1.84
1	144124	143930	142660	143571.33	795.18	0.55
2.5	353056	349173	355106	352445.00	3013.32	0.85
5	716842	722956	719642	719813.33	3060.60	0.43
10	1422700	1412591	1431429	1422240.00	9427.42	0.66
25	3564904	3558674	3573939	3565839.00	7675.33	0.22
50	7116035	7072805	7070922	7086587.33	25519.80	0.36
100	13561883	13545299	14175869	13761017.00	359368.05	2.61



Day 1 (12/22/15)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	15674	16317	17204	16398.33	768.24	4.68
0.25	36307	39143	37861	37770.33	1420.17	3.76
0.5	71014	67924	70464	69800.67	1648.34	2.36
1	143870	145302	141587	143586.33	1873.67	1.30
2.5	376827	363049	360055	366643.67	8945.18	2.44
5	690234	716061	721088	709127.67	16554.32	2.33
10	1448647	1463641	1417369	1443219.00	23608.72	1.64
25	3780133	3605760	3661847	3682580.00	89016.17	2.42
50	7102305	7077875	7045498	7075226.00	28495.99	0.40
100	14301168	14080883	13801669	14061240.00	250328.18	1.78



Day 3 (11/02/15)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	15802	14029	16021	15284.00	1092.36	7.15
0.25	39177	38203	35782	37720.67	1748.14	4.63
0.5	75185	74576	74492	74751.00	378.19	0.51
1	136988	135797	135656	136147.00	731.73	0.54
2.5	325319	327333	333558	328736.67	4295.11	1.31
5	721724	676191	672156	690023.67	27527.33	3.99
10	1302339	1298195	1305948	1302160.67	3879.58	0.30
25	3193561	3251939	3198121	3214540.33	32468.35	1.01
50	6452875	6481142	6459634	6464550.33	14760.88	0.23
100	12706171	12784701	12861015	12783962.33	77424.64	0.61



Alprazolam Validation LOD and LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	138297	31833
STD day 2	140766	24764
STD day 3	127913	18807
Aver.	135658.67	
SD		6520.91
LOD=3.3SD/Aver		**0.16**
LOQ=10SD/Aver		**0.48**

Alprazolam Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	1422700	10.06	10.05	0.09	100.48	0.92
		1431429	10.13				
		1432769	10.14				
		1412591	9.99				
		1409497	9.97				
		1407925	9.96				
	Day 2	1448647	10.12				
		1463641	10.22				
		1417369	9.89				
	Day 3	1302339	10.03				
		1298195	10.00				
		1305948	10.06				
25	Day 1	3611030	25.90	25.65	0.60	102.61	2.33
		3562031	25.54				
		3564904	25.56				
		3558874	25.52				
		3730425	26.76				
		3573939	25.63				
	Day 2	3780133	26.68				
		3605760	25.44				
		3661847	25.84				
	Day 3	3193561	24.82				
		3251939	25.28				
		3198121	24.86				
50	Day 1	7116035	51.26	50.67	0.49	101.34	0.96
		7072805	50.95				
		7070922	50.93				
		7109650	51.21				
		7077818	50.98				
		7117630	51.27				
	Day 2	7102305	50.28				
		7077875	50.11				
		7045498	49.88				
	Day 3	6452875	50.30				
		6481142	50.52				
		6459634	50.35				

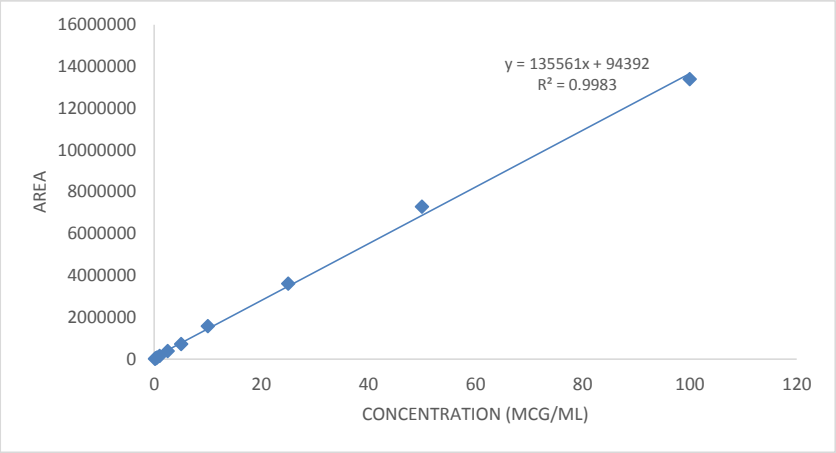
Alprazolam Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	1422700	10.06	10.04	0.08	100.40	0.80
	1431429	10.13				
	1432769	10.14				
	1412591	9.99				
	1409497	9.97				
	1407925	9.96				
25	3611030	25.90	25.82	0.48	103.28	1.87
	3562031	25.54				
	3564904	25.56				
	3558874	25.52				
	3730425	26.76				
	3573939	25.63				
50	7116035	51.26	51.10	0.16	102.20	0.32
	7072805	50.95				
	7070922	50.93				
	7109650	51.21				
	7077818	50.98				
	7117630	51.27				

Alprazolam Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	1578265	10.95	109.46
50	7293070	53.10	106.21
100	13400269	98.15	98.15

Conc	Peak Area
0.1	18570
0.25	43183
0.5	78748
1	150944
2.5	388831
5	727209
10	1578265
25	3611038
50	7293070
100	13400269

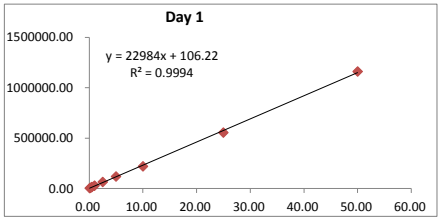


Buprenorphine Validation HPLC Conditions

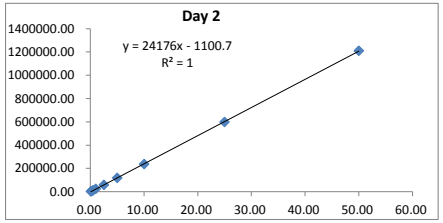
Chromatographic conditions:
System: HPLC
Mobile Phase: Acetonitrile (83%), 10mM phosphate buffer (17%)
Flow rate: 1ml/min
Run time: 10min
Retention time: ~3min
Column: (size and particle size): Kinetex 5u EVO C18 100A, New Column 150x4.6mm
Column Temperature: 35°C
Detection wavelength: 212nm

Buprenorphine Std Curve

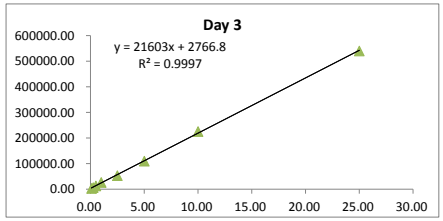
Day 1						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	2735.00	2612.00	2645.00	2664.00	63.66	2.39
0.25	6312.00	7014.00	6688.00	6671.33	351.30	5.27
0.50	14203.00	13887.00	14076.00	14055.33	159.01	1.13
1.00	27991.00	28034.00	28136.00	28053.67	74.47	0.27
2.50	64703.00	62234.00	68117.00	65018.00	2954.12	4.54
5.00	120041.00	119883.00	115572.00	118498.67	2535.80	2.14
10.00	229168.00	223786.00	203745.00	218899.67	13397.37	6.12
25.00	597783.00	573188.00	494780.00	555250.33	53793.35	9.69
50.00	1200340.00	1178564.00	1102347.00	1160417.00	51455.24	4.43



Day 2						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	2507.00	2494.00	2438.00	2479.67	36.67	1.48
0.25	6241.00	6057.00	6385.00	6227.67	164.41	2.64
0.50	12872.00	12463.00	11936.00	12423.67	469.24	3.78
1.00	23978.00	24165.00	24918.00	24353.67	497.59	2.04
2.50	58023.00	59251.00	58992.00	58755.33	647.31	1.10
5.00	118373.00	120363.00	118993.00	119243.00	1018.28	0.85
10.00	230194.00	239981.00	243517.00	237897.33	6901.58	2.90
25.00	601132.00	596283.00	601284.00	599566.33	2844.47	0.47
50.00	1228494.00	1203482.00	1198478.00	1210151.33	16081.05	1.33



Day 3						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	3526.00	3389.00	3654.00	3523.00	132.53	3.76
0.25	6721.00	6799.00	7021.00	6847.00	155.65	2.27
0.50	12736.00	13128.00	13995.00	13286.33	644.26	4.85
1.00	25283.00	27259.00	28013.00	26851.67	1409.85	5.25
2.50	50231.00	54291.00	55928.00	53483.33	2933.12	5.48
5.00	110294.00	102639.00	117394.00	110109.00	7379.24	6.70
10.00	227391.00	219471.00	230001.00	225621.00	5483.60	2.43
25.00	519381.00	539180.00	562910.00	540490.33	21794.06	4.03
50.00	1156294.00	1083910.00	1102818.00	1114340.67	37542.51	3.37



Concentration	Measured concentration
0.25	0.29
2.50	2.82
25.00	24.15

Buprenorphine LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	22984.00	106.22
STD day 2	24176.00	1100.70
STD day 3	22150.00	518.45
Aver.	23103.33	
SD		499.66
LOD=3.3SD/Aver		0.07
LOQ=10SD/Aver		0.22

Buprenorphine Interday Accuracy & Precision

Concentration (µg/mL)	Day	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	Day 1	6312.00	0.27	0.24	0.06	96.31	23.81
		7014.00	0.30				
		6688.00	0.29				
	Day 2	6241.00	0.30				
		6057.00	0.30				
		6385.00	0.31				
	Day 3	6721.00	0.18				
		6799.00	0.19				
		7021.00	0.20				
		7112.00	0.20				
		6782.00	0.19				
		6423.00	0.17				
2.5	Day 1	64703.00	2.81	2.53	0.21	101.36	8.15
		62234.00	2.70				
		68117.00	2.96				
	Day 2	58023.00	2.45				
		59251.00	2.50				
		58992.00	2.49				
	Day 3	50231.00	2.20				
		54291.00	2.39				
		55982.00	2.46				
		53817.00	2.36				
		57317.00	2.53				
		58365.00	2.57				
25	Day 1	597783.00	26.00	24.84	1.21	99.35	4.87
		573188.00	24.93				
		494780.00	21.52				
	Day 2	601132.00	24.91				
		596283.00	24.71				
		601284.00	24.92				
	Day 3	519381.00	23.91				
		539180.00	24.83				
		562910.00	25.93				
		562837.00	25.93				
		541819.00	24.95				
		553719.00	25.50				

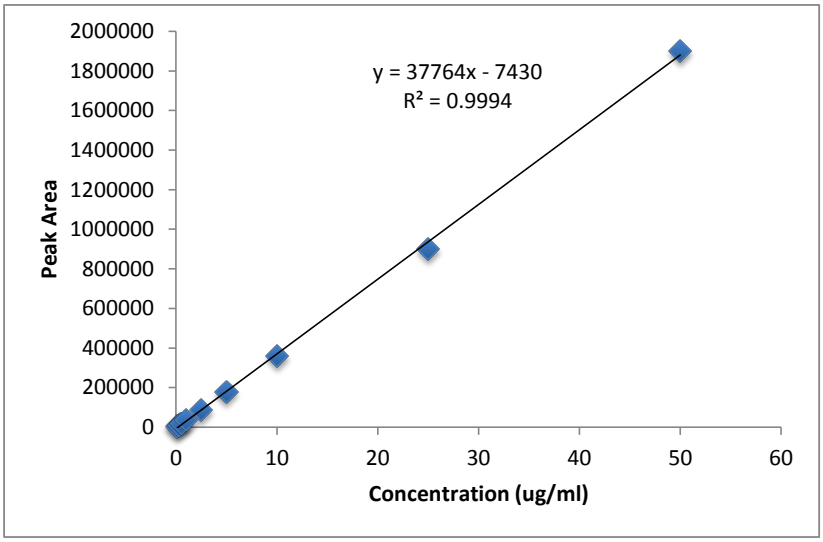
Buprenorphine Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	6721	0.18	0.19	0.01	74.86	6.00
0.25	6799	0.19				
0.25	7021	0.20				
0.25	7112	0.20				
0.25	6782	0.19				
0.25	6423	0.17				
2.5	50231	2.20	2.42	0.13	96.72	5.57
2.5	54291	2.39				
2.5	55982	2.46				
2.5	53817	2.36				
2.5	57317	2.53				
2.5	58365	2.57				
25	519381	23.91	25.18	0.77	100.70	3.08
25	539180	24.83				
25	562910	25.93				
25	562837	25.93				
25	541819	24.95				
25	553719	25.50				

Buprenorphine Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
0.25	8612	0.42	146.48
2.5	66105	1.95	69.05
25	650329	17.42	72.12

Standard Curve	
Concentration	Peak Area
0.1	3512
0.25	8930
0.5	18003
1	35733
2.5	89412
5	178549
10	359822
25	902175
50	1900053



Dextroamphetamine Validation HPLC Conditions

Chromatographic conditions: Dextroamphetamine Sulfate

System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Gradient
Mobile Phase A- ACN (0.05%TFA) B-Water (0.05%TFA)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 15min
Retention time: 4.18 min
Column: (size and particle size): Phenomenex, Prodigy 5µ ODS(2) 150 x 4.6 mm 5 micron
Column Temperature: 25°C
Detection wavelength: 258 nm

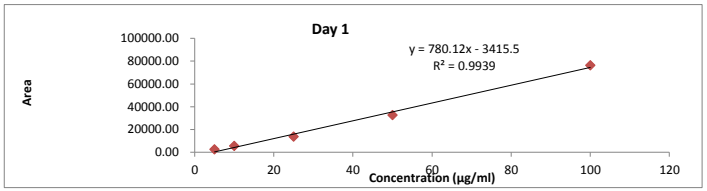
HPLC Gradient Method

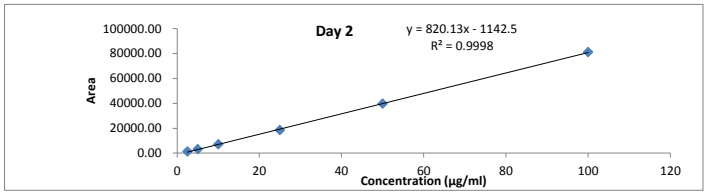
Time (min)	Flow (ml/min)	% A	% B
0	1	10	90
0.8	1	80	20
13	1	80	20
13.01	1	10	90
15	1	10	90

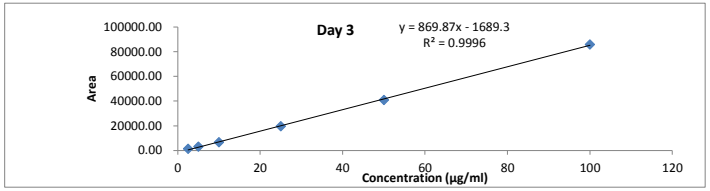
Dextroamphetamine Validation Std Curve

Day 1 (11/10/15)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
2.5	908	1048	828	928.00	111.36	12.00
5	2423	2787	2668	2626.00	185.60	7.07
10	5685	5463	5636	5594.67	116.63	2.08
25	13890	13915	13687	13830.67	125.05	0.90
50	32466	32730	33000	32732.00	267.01	0.82
100	76499	77133	75454	76362.00	847.84	1.11
Day 2 (11/11/15)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
2.5	1194	1537	1466	1399.00	181.05	12.94
5	3072	2931	3570	3191.00	335.71	10.52
10	7083	7026	7101	7070.00	39.15	0.55
25	18501	18530	18518	18516.33	14.57	0.08
50	39691	39747	39694	39710.67	31.50	0.08
100	81962	81102	80337	81133.67	812.96	1.00

Day 3 (11/12/15)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
2.5	1263	1312	1369	1314.67	53.05	4.04
5	3059	3061	3101	3073.67	23.69	0.77
10	6617	6457	6606	6560.00	89.37	1.36
25	19339	19670	19760	19589.67	221.70	1.13
50	40865	41036	40833	40911.33	109.14	0.27
100	85518	86080	85995	85864.33	302.93	0.35







Dextroamphetamine Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	780.12	3415.5
STD day 2	820.13	1142.5
STD day 3	869.87	1689.3
Aver.	823.37	
SD		1186.40
LOD=3.3SD/Aver		**4.75**
LOQ=10SD/Aver		**14.41**

Dextroamphetamine Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	5685	11.67	10.23	0.82	102.33	8.06
		5463	11.38				
		5636	11.60				
	Day 2	7083	10.03				
		7026	9.96				
		7101	10.05				
		6943	9.86				
		6891	9.80				
		7063	10.01				
	Day 3	6617	9.55				
		6457	9.36				
		6606	9.54				
25	Day 1	13890	22.18	23.57	0.94	94.27	4.01
		13915	22.22				
		13687	21.92				
	Day 2	18051	23.40				
		18530	23.99				
		18518	23.97				
		18577	24.04				
		18574	24.04				
		18259	23.66				
	Day 3	19339	24.17				
		19670	24.55				
		19760	24.66				
50	Day 1	32466	45.99	48.83	1.58	97.66	3.24
		32730	46.33				
		33000	46.68				
	Day 2	39691	49.79				
		39747	49.86				
		39694	49.79				
		40134	50.33				
		39985	50.15				
		39981	50.14				
	Day 3	40865	48.92				
		41036	49.12				
		40833	48.88				

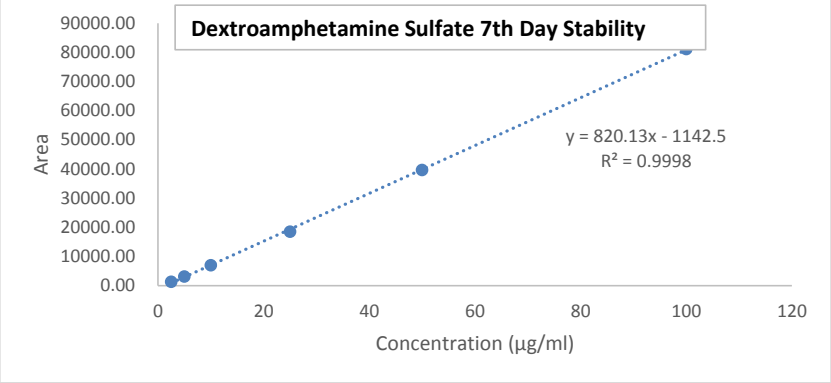
Dextroamphetamine Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	7083	10.03	9.95	0.10	99.50	1.02
	7026	9.96				
	7101	10.05				
	6943	9.86				
	6891	9.80				
	7063	10.01				
25	18051	23.40	23.85	0.26	95.40	1.10
	18530	23.99				
	18518	23.97				
	18577	24.04				
	18574	24.04				
	18259	23.66				
50	39691	49.79	50.01	0.23	100.02	0.45
	39747	49.86				
	39694	49.79				
	40134	50.33				
	39985	50.15				
	39981	50.14				

Dextroamphetamine Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	6653	9.51	95.05
50	41417	51.89	103.79
100	88190	108.92	108.92

Conc	Peak Area
2.5	1399.00
5	3191.00
10	7070.00
25	18516.33
50	39710.67
100	81133.67

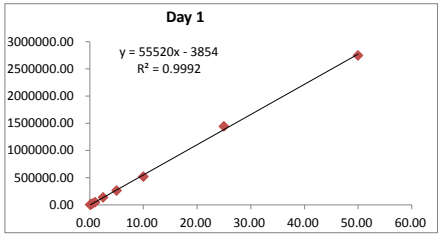


Diazepam Validation HPLC Conditions

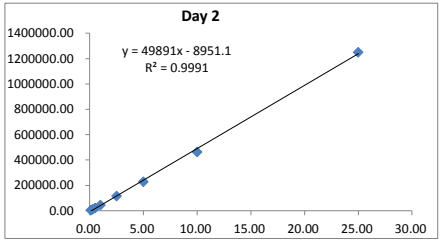
Chromatographic conditions:
System: HPLC
Mobile Phase: Methanol (60%), pH2.5 20mM phosphate buffer (40%)
Flow rate: 1.2ml/min
Run time: 10min
Retention time:~5min
Column: (size and particle size): Kinetex 5u EVO C18 100A, New Column 150X4.6mm
Column Temperature: 30° C
Detection wavelength: 230nm

Diazepam Validation Std Curve

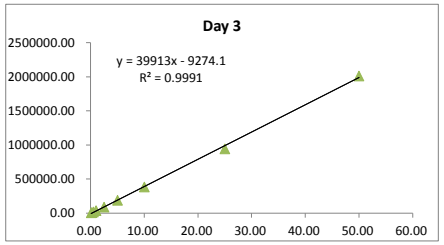
Day 1						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	4873.00	4340.00	4887.00	4700.00	311.85	6.64
0.25	12438.00	13320.00	11907.00	12555.00	713.73	5.68
0.50	23922.00	24012.00	23479.00	23804.33	285.32	1.20
1.00	48542.00	50212.00	49222.00	49325.33	839.78	1.70
2.50	136619.00	136312.00	137146.00	136692.33	421.81	0.31
5.00	267956.00	261168.00	259334.00	262819.33	4542.01	1.73
10.00	519710.00	520377.00	518674.00	519587.00	858.14	0.17
25.00	1434810.00	1439795.00	1460530.00	1445045.00	13640.07	0.94
50.00	2780630.00	2754916.00	2711605.00	2749050.33	34884.34	1.27



Day 2						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	4016.00	4432.00	4372.00	4273.33	224.87	5.26
0.25	10608.00	11538.00	10972.00	11039.33	468.64	4.25
0.50	19517.00	22627.00	22173.00	21439.00	1679.91	7.84
1.00	46342.00	44291.00	45296.00	45309.67	1025.57	2.26
2.50	110855.00	119385.00	116394.00	115544.67	4327.96	3.75
5.00	225420.00	230179.00	231628.00	229075.67	3247.74	1.42
10.00	457903.00	462812.00	470012.00	463575.67	6090.51	1.31
25.00	1178391.00	1283946.00	1290009.00	1250782.00	62765.70	5.02
50.00	2283910.00	2471839.00	2461824.00	2405857.67	########	4.39



Day 3						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	4380.00	4612.00	4677.00	4556.33	156.13	3.43
0.25	8755.00	9023.00	9436.00	9071.33	343.06	3.78
0.50	17834.00	18563.00	18984.00	18460.33	581.83	3.15
1.00	35276.00	37123.00	37254.00	36551.00	1106.12	3.03
2.50	88143.00	89534.00	87164.00	88280.33	1190.95	1.35
5.00	190002.00	185831.00	189426.00	188419.67	2260.27	1.20
10.00	384631.00	379260.00	389751.00	384547.33	5246.00	1.36
25.00	953792.00	938621.00	931765.00	941392.67	11272.04	1.20
50.00	2011889.00	2085174.00	1936193.00	2011085.33	74493.75	3.70



Concentration	Measured concentration
0.25	0.30
2.5	2.53
25	26.10

Diazepam Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	55520.00	3854.00
STD day 2	49891.00	8951.10
STD day 3	39913.00	9274.10
Aver.	48441.33	
SD		3040.35
LOD=3.3SD/Aver		0.21
LOQ=10SD/Aver		0.63

Diazepam Validation Interday Accuracy & Precision

Concentration (µg/mL)	Day	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	Day 1	12438.00	0.29	0.40	0.07	159.73	16.76
		13320.00	0.31				
		11907.00	0.28				
	Day 2	10608.00	0.39				
		11538.00	0.41				
		10972.00	0.40				
	Day 3	8755.00	0.45				
		9023.00	0.46				
		9436.00	0.47				
		8537.00	0.45				
		8383.00	0.44				
		8113.00	0.44				
2.5	Day 1	136619.00	2.53	2.49	0.05	99.60	2.17
		136312.00	2.52				
		137146.00	2.54				
	Day 2	110855.00	2.40				
		119385.00	2.57				
		116394.00	2.51				
	Day 3	88143.00	2.44				
		89534.00	2.48				
		87164.00	2.42				
		90271.00	2.49				
		88291.00	2.44				
		91638.00	2.53				
25	Day 1	1434810.00	25.91	24.81	1.11	99.26	4.48
		1439795.00	26.00				
		1460530.00	26.38				
	Day 2	1178391.00	23.80				
		1283946.00	25.91				
		1290009.00	26.04				
	Day 3	953792.00	24.13				
		938621.00	23.75				
		931765.00	23.58				
		962812.00	24.36				
		951483.00	24.07				
		942715.00	23.85				

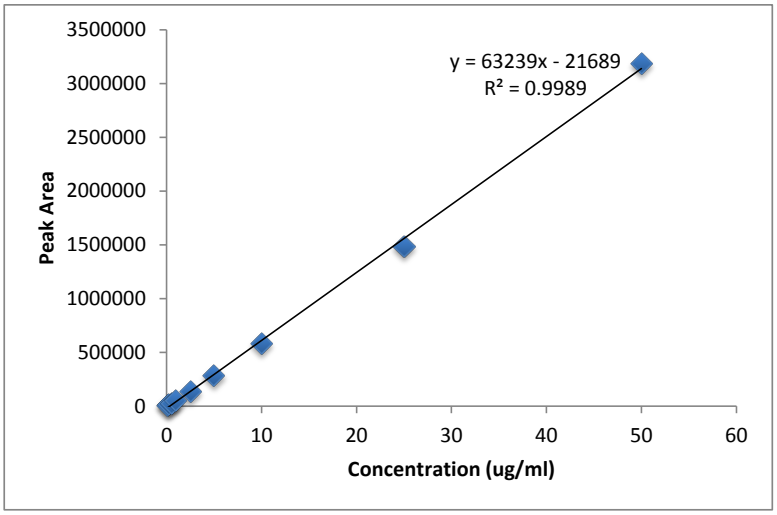
Diazepam Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	8755.00	0.45	0.45	0.01	180.21	2.63
0.25	9023.00	0.46				
0.25	9436.00	0.47				
0.25	8537.00	0.45				
0.25	8383.00	0.44				
0.25	8113.00	0.44				
2.50	88143.00	2.44	2.47	0.04	98.66	1.66
2.50	89534.00	2.48				
2.50	87164.00	2.42				
2.50	90271.00	2.49				
2.50	88291.00	2.44				
2.50	91638.00	2.53				
25.00	953792.00	24.13	23.96	0.28	95.82	1.18
25.00	938621.00	23.75				
25.00	931765.00	23.58				
25.00	962812.00	24.36				
25.00	951483.00	24.07				
25.00	942715.00	23.85				

Diazepam Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
0.25	9134	0.49	162.47
2.5	119670	2.24	88.35
25	1308521	21.03	80.59

Standard Curve	
Concentration (ug/ml)	Peak Area
0.1	4921
0.25	13728
0.5	25192
1	52019
2.5	140291
5	283910
10	583718
25	1483915
50	3183710



Fentanyl Validation HPLC Conditions

Chromatographic conditions: Drug- Fentanyl

System: Waters e2795

Detector: Waters 2988 photodiode array

Pump Mode: Isocratic

Mobile Phase: ACN (60%) : water (0.2% (v/v) Formic acid containing 10mM sodium -1-decane sulfonate) (40%)

Flow rate: 1ml/min

Injection Volume: 20µL

Run time: 10 min

Retention time: 4.8min

Column: (size and particle size): Gemini NX C18; 150x4.6 mm

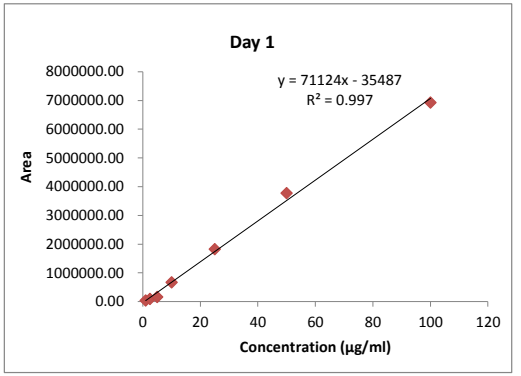
Column Temperature: 25°C

Detection wavelength: 192 nm

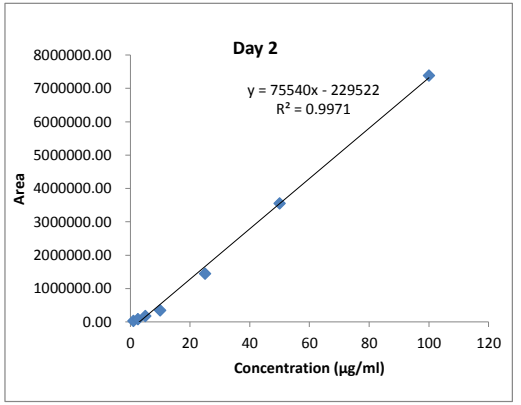
Sample Preparation: Standards were preapred in HPLC grade water

Fentanyl Validation Std Curve

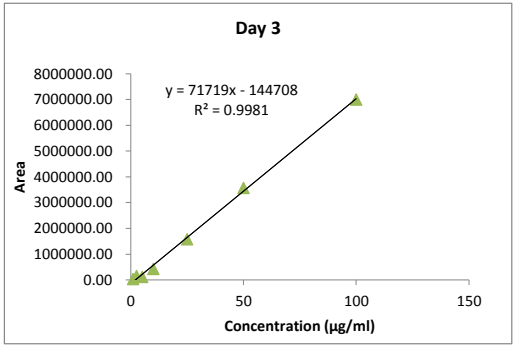
Day 1(08/10/15)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
1	45617	30834	37199	37883.33	7415.22	19.57
2.5	89978	105972	93366	96438.67	8428.11	8.74
5	177184	159082	154194	163486.67	12111.39	7.41
10	671261	641582	700030	670957.67	29225.18	4.36
25	1897793	1809936	1783518	1830415.67	59826.89	3.27
50	3749607	3758500	3832658	3780255.00	45599.64	1.21
100	6933682	6892595	6977487	6934588.00	42453.25127	0.6122



Day 2(08/11/15)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
1	28269	30199	23213	27227.00	3607.68	13.25
2.5	84844	82584	70296	79241.33	7828.87	9.88
5	170995	182014	169117	174042.00	6967.52	4.00
10	313194	367475	349058	343242.33	27603.86	8.04
25	1455933	1458718	1429161	1447937.33	16320.30	1.13
50	3486587	3598066	3568444	3551032.33	57743.11	1.63
100	7359045	7395088	7408789	7387640.667	25694.61937	0.34781



Day 3 (08/12/15)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
1	31610	28783	29726	30039.67	1439.37	4.79
2.5	165880	143699	146822	152133.67	12006.65	7.89
5	112861	120540	102673	112024.67	8962.81	8.00
10	427689	431487	425066	428080.67	3228.37	0.75
25	1565896	1582367	1583530	1577264.33	9862.42	0.63
50	3552971	3581543	3569496	3568003.33	14344.37	0.40
100	7023619	6984827	6982939	6997128.33	22961.00	0.33



Fentanyl Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	71124	35487
STD day 2	75540	229522
STD day 3	71719	144708
Aver.	72794.33	
SD		97273.00
LOD=3.3SD/Aver		**4.41**
LOQ=10SD/Aver		**13.36**

Fentanyl Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	671261	9.94	8.35	1.00	83.53	11.93
		641582	9.52				
		700030	10.34				
	Day 2	313194	7.18				
		367475	7.90				
		349058	7.66				
	Day 3	427689	7.98				
		431487	8.03				
		425066	7.94				
		408734	7.72				
		431141	8.03				
		428378	7.99				
25	Day 1	1897793	27.18	24.10	1.54	96.38	6.40
		1809936	25.95				
		1783518	25.58				
	Day 2	1455933	22.31				
		1458718	22.35				
		1429161	21.96				
	Day 3	1565896	23.85				
		1582367	24.08				
		1583530	24.10				
		1580733	24.06				
		1574705	23.97				
		1559506	23.76				
50	Day 1	3749607	53.22	51.80	1.40	103.59	2.71
		3758500	53.34				
		3832658	54.39				
	Day 2	3486587	49.19				
		3598066	50.67				
		3568444	50.28				
	Day 3	3552971	51.56				
		3581543	51.96				
		3569496	51.79				
		3581124	51.95				
		3546632	51.47				
		3567274	51.76				

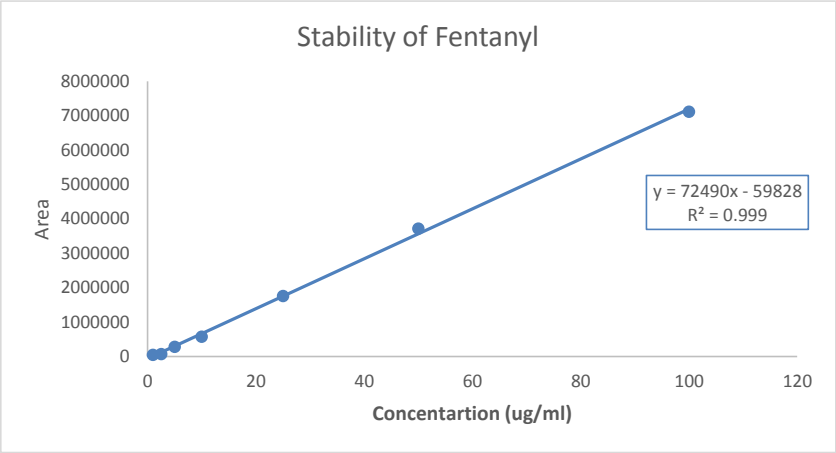
Fentanyl Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	427689	7.98	7.95	0.12	15.90	1.49
10	431487	8.03				
10	425066	7.94				
10	408734	7.72				
10	431141	8.03				
10	428378	7.99				
25	1565896	23.85	23.97	0.14	31.96	0.57
25	1582367	24.08				
25	1583530	24.10				
25	1580733	24.06				
25	1574705	23.97				
25	1559506	23.76				
50	3552971	51.56	51.75	0.20	51.75	0.39
50	3581543	51.96				
50	3569496	51.79				
50	3581124	51.95				
50	3546632	51.47				
50	3567274	51.76				

Fentanyl Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	584695	8.89	88.91
25	1761431	25.12	100.50
50	3720157	52.14	104.29

Conc	Peak Area
1	55862
2.5	76838
5	290528
10	584695
25	1761431
50	3720157
100	7118490



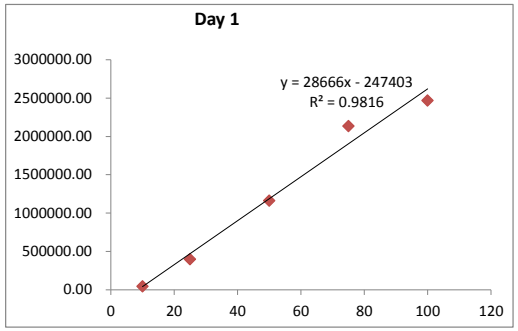
Fluoxetine Validation HPLC Conditions

Chromatographic conditions:

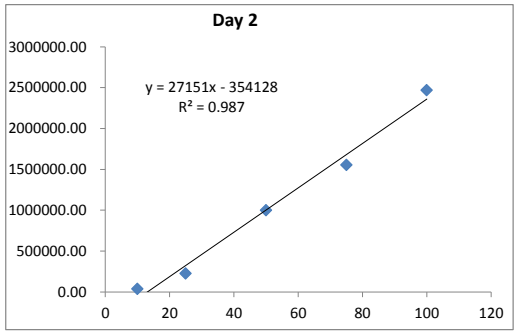
System: Waters e2795
Mobile Phase: Acetonitrile: Potassium dihydrogen phosphate buffer(40:60)
Flow rate: 1ml/min
Run time: 10 min
Retention time: 3.4
Column: (size and particle size): C18 kinetex 5um, 250* 4.6mm
Column Temperature: 25°C

Fluoxetine Validation Std Curve

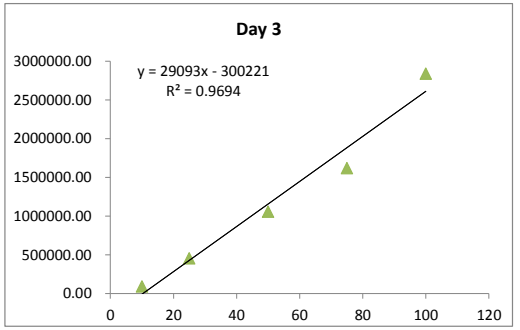
Day 1(12/16/14)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
10	41494	53008	39536	44679.33	7278.97	16.29
25	400001	401759	396562	399440.67	2643.42	0.66
50	1131052	1210945	1149163	1163720.00	41888.57	3.60
75	2247337	2075998	2087237	2136857.33	95843.08	4.49
100	2480295	2463515	2470689	2471499.67	8419.32	0.34



Day 2(12/17/14)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
10	29426	30564	48930	36306.67	10946.93	30.15
25	215770	233782	228984	226178.67	9327.94	4.12
50	979674	1023390	998650	1000571.33	21921.24	2.19
75	1577842	1545221	1542802	1555288.33	19569.46	1.26
100	2473891	2468362	2468879	2470377.33	3053.88	0.12



Day 3(12/18/14)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
10	91829	98768	89070	93222.33	4996.88	5.36
25	456181	467273	446322	456592.00	10481.55	2.30
50	1055984	1069391	1046639	1057338.00	11436.27	1.08
75	1630223	1611915	1612895	1618344.33	10298.89	0.64
100	2851024	2706374	2955122	2837506.67	124923.70	4.40



Fluoxetine Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	28666	247403
STD day 2	27151	354128
STD day 3	29093	300221
Aver.	28303.33	
SD		53363.43
LOD=3.3SD/Aver		**6.22**
LOQ=10SD/Aver		**18.85**

Fluoxetine Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
50	Day 1	1131052	48.09	48.39	1.52	96.77	3.14
		1210945	50.87				
		1149163	48.72				
	Day 2	979674	49.13				
		1023390	50.74				
		998650	49.82				
	Day 3	1055984	46.62				
		1069391	47.08				
		1046639	46.29				
		1096447	48.01				
		1073347	47.21				
		1098285	48.07				
75	Day 1	2247337	87.03	73.24	6.78	97.65	9.26
		2075998	81.05				
		2087237	81.44				
	Day 2	1577842	71.16				
		1545221	69.96				
		1542802	69.87				
	Day 3	1630223	66.35				
		1611915	65.72				
		1612895	65.76				
		1859986	74.25				
		1828875	73.18				
		1826819	73.11				
100	Day 1	2480295	95.15	98.96	7.94	98.96	8.03
		2463515	94.57				
		2470689	94.82				
	Day 2	2473891	104.16				
		2468362	103.96				
		2468879	103.97				
	Day 3	2851024	108.32				
		2706374	103.34				
		2955122	111.89				
		2304318	89.52				
		2262013	88.07				
		2311672	89.78				

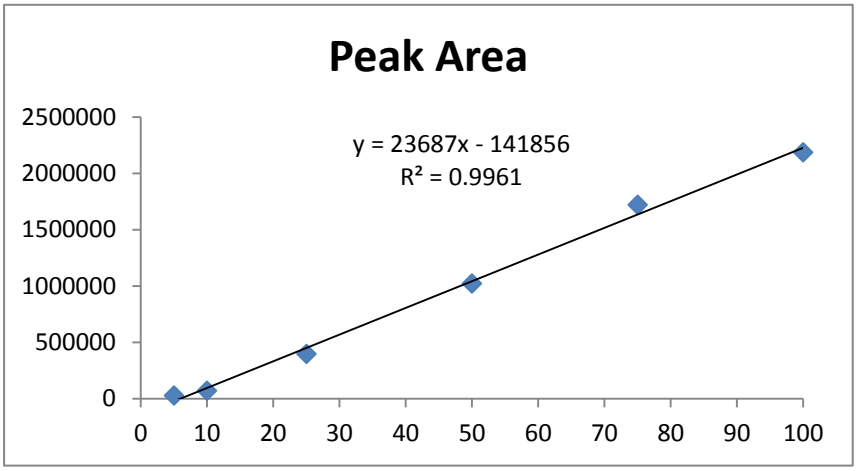
Fluoxetine Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
50	1055984	46.62	47.21	0.72	94.43	1.52
50	1069391	47.08				
50	1046639	46.29				
50	1096447	48.01				
50	1073347	47.21				
50	1098285	48.07				
75	1630223	66.35	69.73	4.17	92.97	5.98
75	1611915	65.72				
75	1612895	65.76				
75	1859986	74.25				
75	1828875	73.18				
75	1826819	73.11				
100	2851024	108.32	98.49	10.63	98.49	10.79
100	2706374	103.34				
100	2955122	111.89				
100	2304318	89.52				
100	2262013	88.07				
100	2311672	89.78				

Fluoxetine Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
50	1021990	49.13	98.27
75	1721031	78.65	104.86
100	2185467	98.25	98.25

Conc	Peak Area
5	29313
10	71360
25	396810
50	1021990
75	1721031
100	2185467



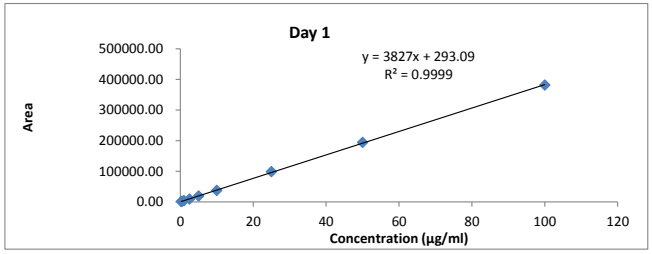
Hydromorphone Validation HPLC Conditions

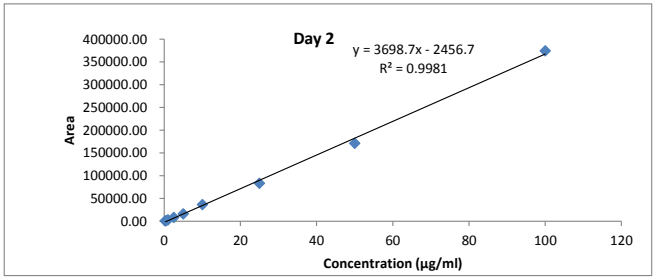
Chromatographic conditions: Hydromorphone Hydrochloride

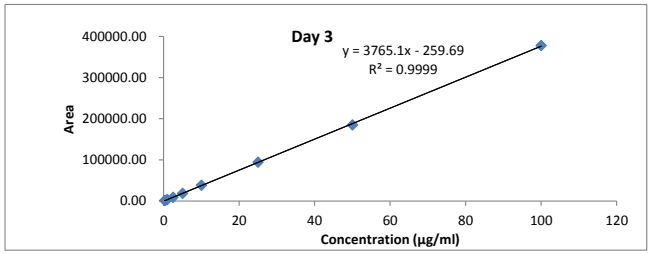
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN: Water (% 0.5 w/v Sodium dodecyl sulphate) (35:65) (% v/v)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 5.36 min
Column: (size and particle size): Phenomenex, Kinetex EVO C18 5µm, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 282 nm
Sample Preparation: Standards were prepared in D.I Water

Hydromorphone Validation Std Curve

Day 1						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.25	1052	799	959	936.67	127.97	13.66
0.5	2303	1604	2017	1974.67	351.42	17.80
1	4471	3908	3372	3917.00	549.56	14.03
2.5	8990	9845	8850	9228.33	538.62	5.84
5	18597	19120	19187	18968.00	323.04	1.70
10	36499	37485	37141	37041.67	500.45	1.35
25	96249	100401	98669	98439.67	2085.48	2.12
50	196169	194071	192576	194272.00	1804.91	0.93
100	376408	388862	378504	381258.00	6668.13	1.75
Day 2						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.25	702	960	498	720.00	231.53	32.16
0.5	1422	1356	1523	1433.67	84.11	5.87
1	3543	3514	3129	3395.33	231.11	6.81
2.5	8694	8500	8479	8557.67	118.53	1.39
5	16775	16889	15535	16399.67	750.99	4.58
10	37433	36514	36250	36732.33	620.99	1.69
25	82175	82892	85702	83589.67	1864.13	2.23
50	170793	169852	172925	171190.00	1574.50	0.92
100	381847	370502	370673	374340.67	6501.24	1.74
Day 3						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.25	752	1221	965	979.33	234.83	23.98
0.5	1547	1908	1557	1670.67	205.60	12.31
1	4018	3441	4097	3852.00	358.12	9.30
2.5	8848	9192	9271	9103.67	224.91	2.47
5	18005	17981	19637	18541.00	949.24	5.12
10	38222	37934	38002	38052.67	150.54	0.40
25	93366	95975	93994	94445.00	1361.72	1.44
50	183653	183225	187256	184711.33	2214.11	1.20
100	377713	380721	374619	377684.33	3051.10	0.81







Hydromorphone Validation LOD & LOQ

LOD and LOQ	**Slope**	**Y-intercept**
STD day 1	3827	293.09
STD day 2	3698.7	2456.7
STD day 3	3765.1	259.69
Aver.	3763.60	
SD		1258.91
LOD=3.3SD/Aver		**1.10**
LOQ=10SD/Aver		**3.34**

Hydromorphone Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
5	Day 1	1052	0.20	3.86	2.23	77.23	57.83
		799	0.13				
		959	0.17				
	Day 2	16775	5.20				
		16889	5.23				
		15535	4.86				
		16264	5.06				
		16955	5.25				
		16960	5.25				
	Day 3	18005	4.85				
		17981	4.84				
		19637	5.28				
10	Day 1	36499	9.46	10.19	0.40	101.90	3.91
		37485	9.72				
		37141	9.63				
	Day 2	37433	10.78				
		36514	10.54				
		36250	10.46				
		35879	10.36				
		35875	10.36				
		36136	10.43				
	Day 3	38222	10.22				
		37934	10.14				
		38002	10.16				
25	Day 1	96249	25.07	24.30	1.22	97.20	5.02
		100401	26.16				
		98669	25.71				
	Day 2	82175	22.88				
		82892	23.08				
		85702	23.84				
		83375	23.21				
		82245	22.90				
		83731	23.30				
	Day 3	93366	24.87				
		95975	25.56				
		93994	25.03				

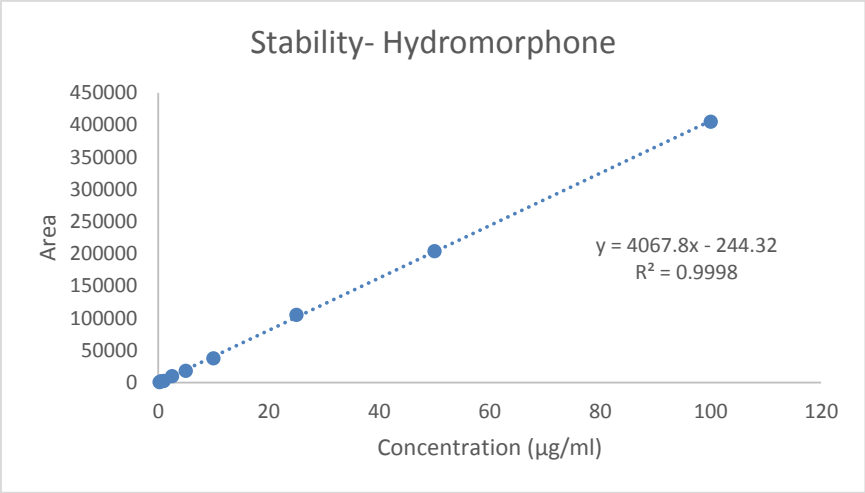
Hydromorphone Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
5	16775	5.20	5.14	0.15	102.85	2.98
	16889	5.23				
	15535	4.86				
	16264	5.06				
	16955	5.25				
	16960	5.25				
10	37433	10.78	10.49	0.16	104.91	1.50
	36514	10.54				
	36250	10.46				
	35879	10.36				
	35875	10.36				
	36136	10.43				
25	82175	22.88	23.20	0.35	92.80	1.52
	82892	23.08				
	85702	23.84				
	83375	23.21				
	82245	22.90				
	83731	23.30				

Hydromorphone Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
5	18577	3.45	69.03
10	37963	8.17	81.73
25	105547	24.63	98.53

Conc	Peak Area
0.25	842
0.5	2258
1	2851
2.5	10364
5	18577
10	37963
25	105547
50	204333
100	405239



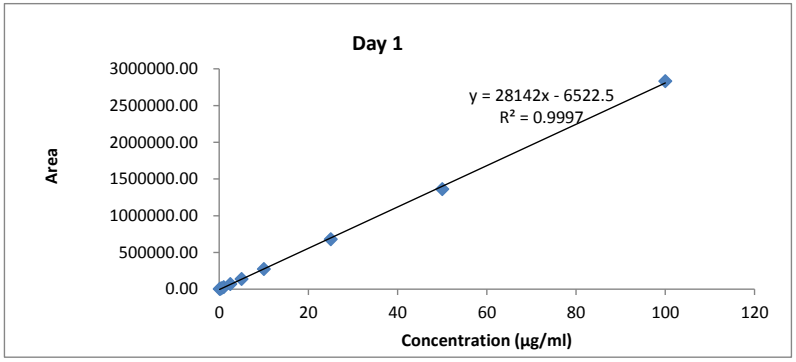
Ketamine Validation HPLC Conditions

Chromatographic conditions: Ketamine hydrochloride

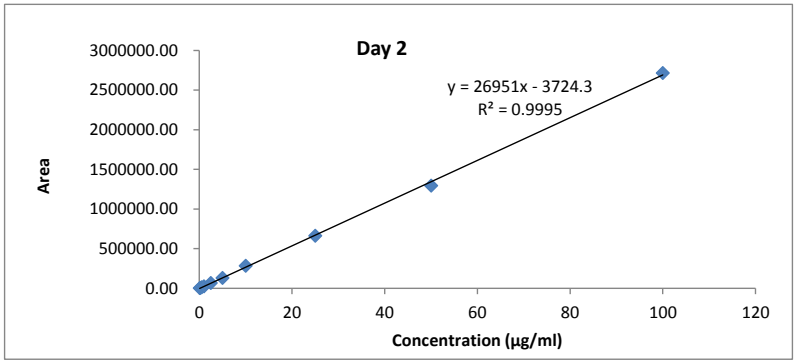
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: MeOH:Buffer (NaH2PO4 50mM pH=3) (30:70)
Flow rate: 1.2ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 6.1min
Column: Agilent ZORBAX Eclipse Plus C 18 (4.6×150mm 5µm)
Column Temperature: 25°C
Detection wavelength: 215 nm
Sample Preparation: Standards were prepared in water

Ketamine Validation Std Curve

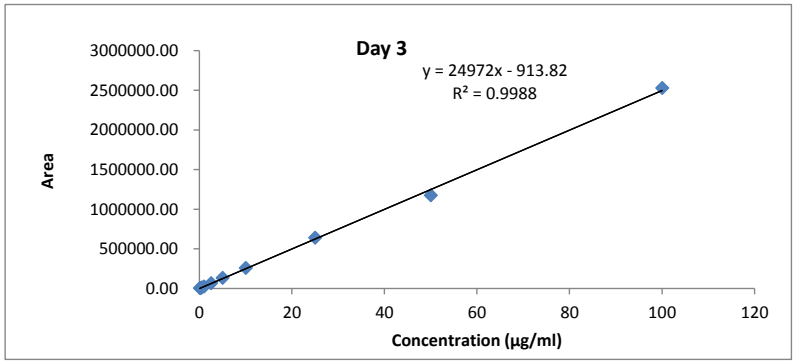
Day 1 (07/29/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	2353	2791	2419	2521.00	236.14	9.37
0.25	5419	5927	5734	5693.33	256.43	4.50
0.5	12334	13278	12614	12742.00	484.84	3.81
1	27608	27771	26642	27340.33	610.24	2.23
2.5	68109	69393	71188	69563.33	1546.55	2.22
5	137757	140896	139575	139409.33	1576.04	1.13
10	275210	272982	275597	274596.33	1411.38	0.51
25	681907	690813	667330	680016.67	11855.08	1.74
50	1327663	1368324	1387208	1361065.00	30428.96	2.24
100	2801156	2803072	2889301	2831176.33	50346.55	1.78



Day 2 (07/30/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	2524	2708	2897	2709.67	186.51	6.88
0.25	5593	5356	5649	5532.67	155.54	2.81
0.5	13913	13310	13466	13563.00	312.98	2.31
1	26550	27883	21698	25377.00	3255.07	12.83
2.5	68626	61584	66841	65683.67	3660.88	5.57
5	133882	130696	127432	130670.00	3225.08	2.47
10	280282	286255	284039	283525.33	3019.45	1.06
25	673187	648303	665717	662402.33	12768.85	1.93
50	1333862	1265184	1288346	1295797.33	34940.07	2.70
100	2662139	2799927	2684408	2715491.33	73966.30	2.72



Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	3127	3157	3419	3234.33	160.63	4.97
0.25	4986	5714	5321	5340.33	364.38	6.82
0.5	13382	11955	12826	12721.00	719.27	5.65
1	24722	25394	24335	24817.00	535.85	2.16
2.5	66453	63508	64988	64983.00	1472.51	2.27
5	135942	132615	130508	133021.67	2739.73	2.06
10	253650	272046	248374	258023.33	12427.21	4.82
25	644825	633642	644072	640846.33	6250.49	0.98
50	1146763	1197176	1177471	1173803.33	25405.84	2.16
100	2538648	2682760	2360785	2527397.67	161282.06	6.38



Ketamine Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	28142	6522.5
STD day 2	26951	3724.3
STD day 3	24972	913.82
Aver.	26688.33	
SD		2804.34
LOD=3.3SD/Aver		**0.35**
LOQ=10SD/Aver		**1.05**

Ketamine Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	275210	10.01	10.35	0.42	103.53	4.03
		272982	9.93				
		275597	10.02				
	Day 2	280282	10.54				
		286255	10.76				
		284039	10.68				
		271978	10.23				
		263090	9.90				
		294175	11.05				
	Day 3	253650	10.19				
		272046	10.93				
		248374	9.98				
25	Day 1	681907	24.46	24.53	0.96	98.13	3.93
		690813	24.78				
		667330	23.94				
	Day 2	673187	25.12				
		648303	24.19				
		665717	24.84				
		623759	23.28				
		641065	23.92				
		609691	22.76				
	Day 3	644825	25.86				
		633642	25.41				
		644072	25.83				
50	Day 1	1327663	47.41	47.77	1.24	95.54	2.60
		1368324	48.85				
		1387208	49.52				
	Day 2	1333862	49.63				
		1265184	47.08				
		1288346	47.94				
		1226308	45.64				
		1279317	47.61				
		1301062	48.41				
	Day 3	1146763	45.96				
		1197176	47.98				
		1177471	47.19				

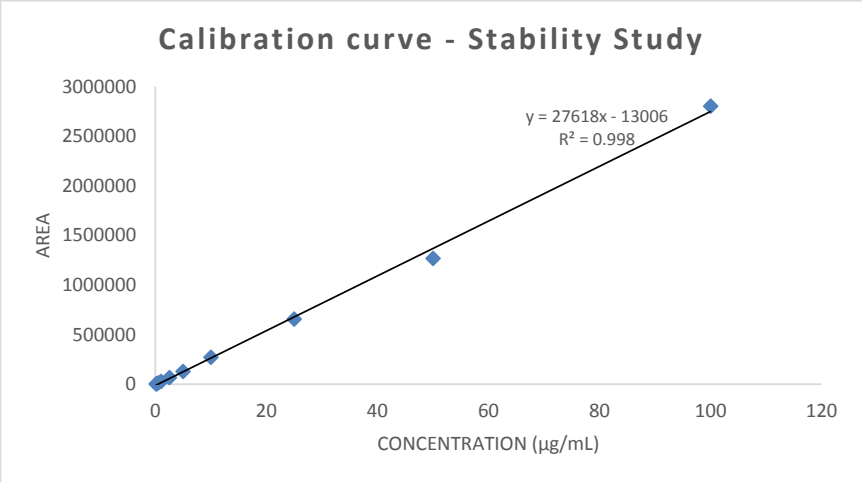
Ketamine Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	280282	10.54	10.53	0.41	105.26	3.88
	286255	10.76				
	284039	10.68				
	271978	10.23				
	263090	9.90				
	294175	11.05				
25	673187	25.12	24.02	0.90	96.08	3.74
	648303	24.19				
	665717	24.84				
	623759	23.28				
	641065	23.92				
	609691	22.76				
50	1333862	49.63	47.72	1.34	95.44	2.80
	1265184	47.08				
	1288346	47.94				
	1226308	45.64				
	1279317	47.61				
	1301062	48.41				

Ketamine Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	271564	10.30	103.04
50	1267341	46.36	92.72
100	2802995	101.96	101.96

Conc (µg/mL)	Peak Area
0.1	2495
0.25	5615
0.5	12719
1	25618
2.5	64971
5	128276
10	271564
25	655838
50	1267341
100	2802995

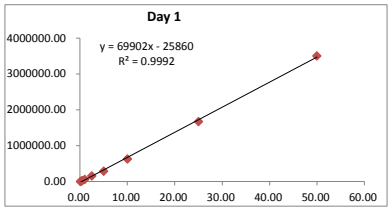


Lorazepam Validation HPLC Conditions

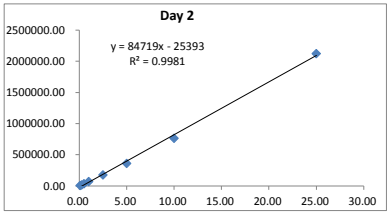
Chromatographic conditions:
System: HPLC
Mobile Phase (Gradient): ACN (0.05% TFA), Water (0.05% TFA)
Flow rate: 1.0ml/min
Run time: 8.5 min
Retention time: ~6.5min
Column: (size and particle size): Xbridge BEH Phenyl 2.5um 4.6x50mm Column XP
Column Temperature: 30° C
Detection wavelength: 229nm

Lorazepam Validation Std Curve

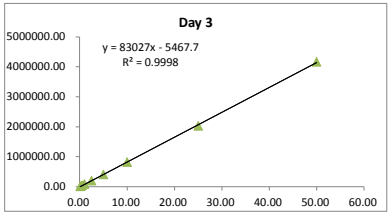
Day 1						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	4580.00	4660.00	4235.00	4491.67	225.85	5.03
0.25	14076.00	14088.00	13811.00	13991.67	156.58	1.12
0.50	27585.00	26740.00	26953.00	27092.67	439.47	1.62
1.00	58061.00	60184.00	62839.00	60361.33	2393.93	3.97
2.50	154267.00	168644.00	156197.00	159702.67	7803.32	4.89
5.00	292633.00	292598.00	285355.00	290195.33	4191.89	1.44
10.00	603697.00	637281.00	644645.00	628541.00	21828.32	3.47
25.00	1603476.00	1792033.00	1626493.00	1674000.67	########	6.14
50.00	3510306.00	3462918.00	3539281.00	3504168.33	38549.71	1.10



Day 2						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	5932.00	6320.00	5273.00	5841.67	529.31	9.06
0.25	16685.00	17416.00	17047.00	17049.33	365.51	2.14
0.50	33119.00	32372.00	35920.00	33803.67	1870.47	5.53
1.00	71249.00	70496.00	71160.00	70968.33	411.47	0.58
2.50	174092.00	178741.00	178832.00	177221.67	2710.75	1.53
5.00	359557.00	365813.00	359032.00	361467.33	3772.60	1.04
10.00	765916.00	766282.00	764480.00	765559.33	952.48	0.12
25.00	2060573.00	2144012.00	2162153.00	2122246.00	54175.12	2.55
50.00	4182739.00	4322187.00	4307090.00	4270672.00	76525.42	1.79



Day 3						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.10	7370.00	7316.00	7701.00	7462.33	208.45	2.79
0.25	19209.00	20728.00	22627.00	20854.67	1712.52	8.21
0.50	40463.00	42365.00	44164.00	42330.67	1850.74	4.37
1.00	87785.00	89371.00	90021.00	89059.00	1150.19	1.29
2.50	208925.00	198162.00	190599.00	199228.67	9209.45	4.62
5.00	406894.00	415225.00	406489.00	409536.00	4930.98	1.20
10.00	827863.00	827707.00	805311.00	820293.67	12975.60	1.58
25.00	1917921.00	1981920.00	2183610.00	2027817.00	########	6.84
50.00	4074792.00	4146836.00	4281790.00	4167806.00	########	2.52



Day 1	
Concentration	**Measured concentration**
0.25	0.57
2.5	2.65
25	24.32

Lorazepam Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	69902.00	25860.00
STD day 2	84719.00	25393.00
STD day 3	83027.00	5467.70
Aver.	79216.00	18906.90
SD	8110.40	11641.03
RSD	10.24	61.57
LOD=3.3SD/Aver	0.34	2.03
LOQ=10SD/Aver	1.02	6.16

Lorazepam Validation Interday Accuracy & Precision

Concentration (µg/mL)	Day	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	Day 1	14076.00	0.57	0.42	0.12	169.64	28.19
		14088.00	0.57				
		13811.00	0.57				
	Day 2	16685.00	0.50				
		17416.00	0.51				
		17047.00	0.50				
	Day 3	19209.00	0.30				
		20728.00	0.32				
		22627.00	0.34				
		20291.00	0.31				
		19920.00	0.31				
		20182.00	0.31				
2.5	Day 1	154267.00	2.58	2.47	0.13	98.96	5.37
		168644.00	2.78				
		156197.00	2.60				
	Day 2	174092.00	2.35				
		178741.00	2.41				
		178832.00	2.41				
	Day 3	208925.00	2.58				
		198162.00	2.45				
		190599.00	2.36				
		192833.00	2.39				
		193038.00	2.39				
		191638.00	2.37				
25	Day 1	1603476.00	23.31	24.49	1.15	97.96	4.70
		1792033.00	26.01				
		1626493.00	23.64				
	Day 2	2060573.00	24.62				
		2144012.00	25.61				
		2162153.00	25.82				
	Day 3	1917921.00	23.17				
		1981920.00	23.94				
		2183610.00	26.37				
		1962981.00	23.71				
		1954148.00	23.60				
		1994271.00	24.09				

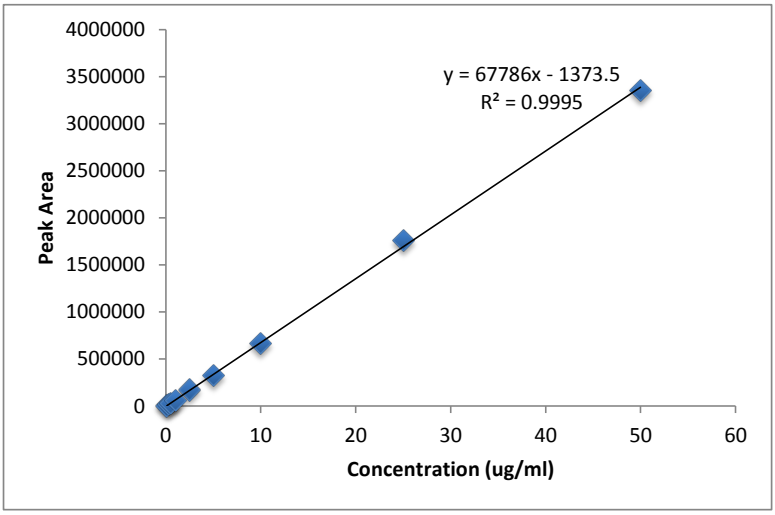
Lorazepam Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	19209.00	0.30	0.31	0.01	125.07	4.47
0.25	20728.00	0.32				
0.25	22627.00	0.34				
0.25	20291.00	0.31				
0.25	19920.00	0.31				
0.25	20182.00	0.31				
2.50	208925.00	2.58	2.42	0.08	97.00	3.43
2.50	198162.00	2.45				
2.50	190599.00	2.36				
2.50	192833.00	2.39				
2.50	193038.00	2.39				
2.50	191638.00	2.37				
25.00	1917921.00	23.17	24.14	1.13	96.58	4.69
25.00	1981920.00	23.94				
25.00	2183610.00	26.37				
25.00	1962981.00	23.71				
25.00	1954148.00	23.60				
25.00	1994271.00	24.09				

Lorazepam Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
0.25	19455	0.31	53.91
2.5	210069	3.12	117.71
25	2093707	30.91	127.09

Standard Curve	
Concentration (ug/ml)	Peak Area
0.1	5294
0.25	15950
0.5	30266
1	58614
2.5	171768
5	322111
10	661878
25	1756696
50	3360688



Loxapine Validation HPLC Conditions

Chromatographic conditions: Drug- Loxapine

System: Waters e2795

Detector: Waters 2988 photodiode array

Pump Mode: Isocratic

Mobile Phase: ACN (40%) : water (0.3% (v/v) Triethylamine pH-3 (60%)

Flow rate: 1ml/min

Injection Volume: 20µL

Run time: 10 min

Retention time: 4.8min

Column: (size and particle size): Phenomenex Luna 5µ C8 (2) 100A 250*4.60 mm 5µ

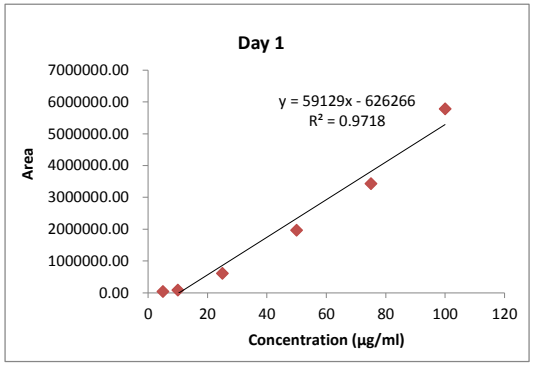
Column Temperature: 25°C

Detection wavelength: 190 nm

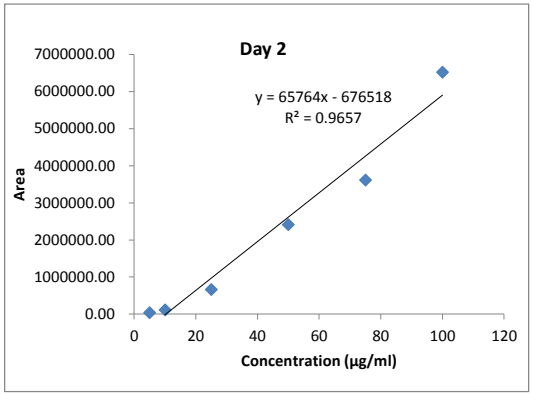
Sample Preparation: Standards were prepared in DI water

Loxapine Validation Std Curve

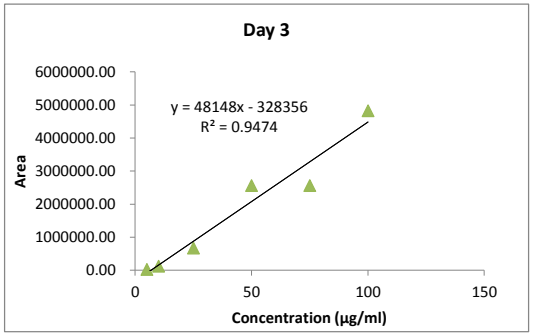
Day 1						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
5	40846	39247	35780	38624.33	2589.76	6.71
10	78115	87550	86269	83978.00	5117.75	6.09
25	619951	615174	589148	608091.00	16578.08	2.73
50	1991044	1974542	1942264	1969283.33	24811.54	1.26
75	3462569	3436021	3392924	3430504.67	35148.67	1.02
100	5804417	5766669	5772386	5781157.33	20345.27	0.35



Day 2						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
5	44142	36638	38407	39729.00	3922.79	9.87
10	108019	114704	115747	112823.33	4193.23	3.72
25	669933	668798	656811	665180.67	7270.53	1.09
50	2392860	2417947	2428873	2413226.67	18464.70	0.77
75	3628077	3630743	3581666	3613495.33	27597.22	0.76
100	6513821	6562023	6495711	6523851.667	34275.07901	0.525381



Day 3						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
5	27606	31807	29435	29616.00	2106.34	7.11
10	129427	129867	119690	126328.00	5752.88	4.55
25	655922	680115	687583	674540.00	16550.38	2.45
50	1206933	1220016	1255297	2564768.33	11527.81	0.45
75	2551786	2573806	2568713	2564768.33	11527.81	0.45
100	4745418	4835094	4906881	4829131.00	80896.50	1.68



Loxapine Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	59129	626266
STD day 2	65764	676518
STD day 3	48148	328856
Aver.	57680.33	
SD		187903.72
LOD=3.3SD/Aver		**10.75**
LOQ=10SD/Aver		**32.58**

Loxapine Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
25	Day 1	619951	21.08	20.52	0.43	82.09	2.11
		615174	21.00				
		589148	20.56				
	Day 2	669933	20.47				
		668798	20.46				
		656811	20.27				
		641783	20.05				
		624876	19.79				
		645687	20.11				
	Day 3	655922	20.44				
		680115	20.95				
		687583	21.10				
50	Day 1	1991044	44.26	42.27	6.12	84.54	14.49
		1974542	43.99				
		1942264	43.44				
	Day 2	2392860	46.67				
		2417947	47.05				
		2428873	47.22				
		2341758	45.90				
		2355026	46.10				
		2328417	45.69				
	Day 3	1206933	31.89				
		1220016	32.16				
		1255297	32.89				
75	Day 1	3462569	69.15	64.56	3.21	86.09	4.96
		3436021	68.70				
		3392924	67.97				
	Day 2	3628077	65.46				
		3630743	65.50				
		3581666	64.75				
		3501509	63.53				
		3559809	64.42				
		3600239	65.03				
	Day 3	2551786	59.82				
		2573806	60.28				
		2568713	60.17				

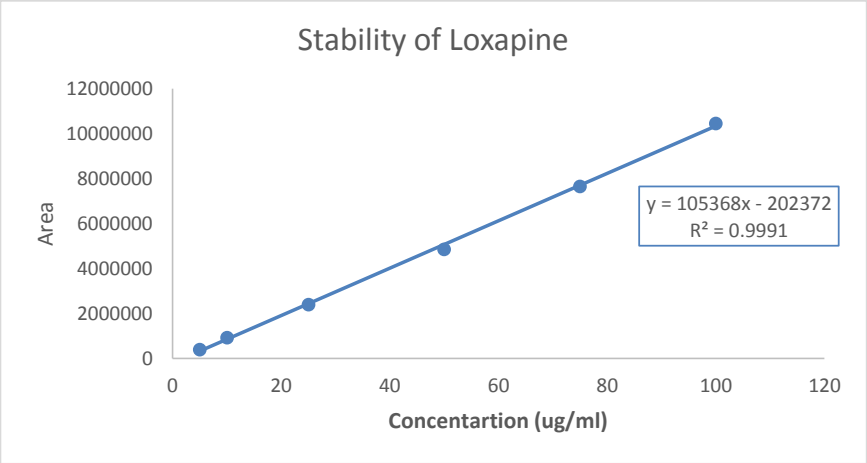
Loxapine Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
25	669933	20.47	20.19	0.26	80.76	1.31
	668798	20.46				
	656811	20.27				
	641783	20.05				
	624876	19.79				
	645687	20.11				
50	2392860	46.67	46.44	0.63	92.88	1.37
	2417947	47.05				
	2428873	47.22				
	2341758	45.90				
	2355026	46.10				
	2328417	45.69				
75	3628077	65.46	64.78	0.74	86.37	1.14
	3630743	65.50				
	3581666	64.75				
	3501509	63.53				
	3559809	64.42				
	3600239	65.03				

Loxapine Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	932486	10.77	107.70
25	2396502	24.66	98.66
50	4861746	48.06	96.12

Conc	Peak Area
5	392914
10	932486
25	2396502
50	4861746
75	7662345
100	10462215



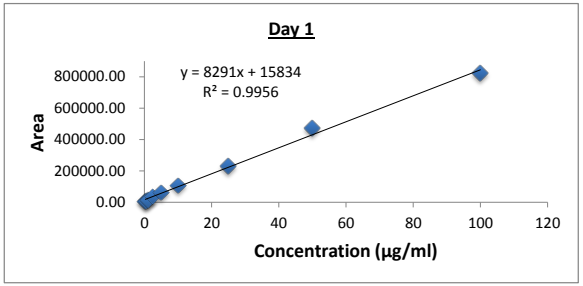
Meperidine Validation HPLC Conditions

Chromatographic conditions to detect: Meperidine

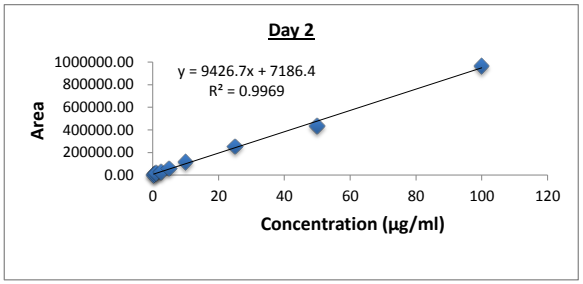
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: Acetonitrile:Buffer (0.02M ammonium acetate) (35:65)
Flow rate: 1ml/min
Injection volume: 20µL
Run time: 10min
Retention time: 5.5min
Column: (size and particle size): Phenomenex, Kinetex C18 5µm, 250×3.5mm
Column Temperature: 40 °C
Detection wavelength: 225 nm

Meperidine Validation Std Curve

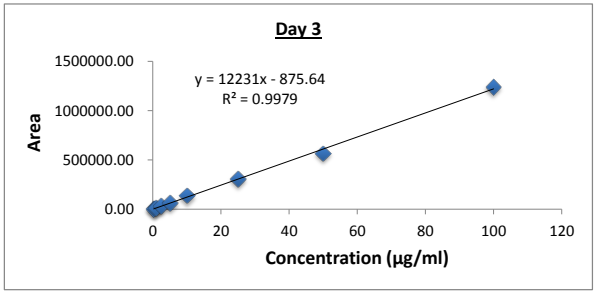
Day 1 (07/21/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.25	4943	4698	4411	4684.00	266.28	5.68
0.5	8427	8617	7900	8314.67	371.47	4.47
1	15562	14595	14617	14924.67	552.06	3.70
2.5	34821	35096	33862	34593.00	647.82	1.87
5	61227	61495	62412	61711.33	621.41	1.01
10	106299	106234	106631	106388.00	212.94	0.20
25	227586	229186	229030	228600.67	882.18	0.39
50	474009	470813	470061	471627.67	2096.29	0.44
100	814917	824864	826766	822182.33	6363.43	0.77



Day 2 (07/22/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.25	4022	4472	4386	4293.33	238.88	5.56
0.5	8154	7961	8699	8271.33	382.74	4.63
1	16905	15494	17481	16626.67	1022.32	6.15
2.5	29358	29657	29222	29412.33	222.53	0.76
5	58850	57612	58238	58233.33	619.01	1.06
10	120450	120244	120396	120363.33	106.81	0.09
25	256228	256407	256088	256241.00	159.90	0.06
50	437935	437090	436732	437252.33	617.71	0.14
100	965716	964629	965031	965125.33	549.61	0.06



Day 3 (07/25/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.25	3625	3890	3921	3812.00	162.69	4.27
0.5	7256	7244	7189	7229.67	35.73	0.49
1	14631	14426	14013	14356.67	314.78	2.19
2.5	30352	31898	30590	30946.67	832.43	2.69
5	60053	63040	58589	60560.67	2268.51	3.75
10	138329	138412	138566	138435.67	120.26	0.09
25	304602	304017	303835	304151.33	400.76	0.13
50	566167	565301	563937	565135.00	1124.23	0.20
100	1246502	1239392	1244464	1243452.67	3661.30	0.29



Meperidine Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	8291	15834
STD day 2	9426.7	7186.4
STD day 3	12231	875.64
Aver.	9982.90	
SD		7509.54
LOD=3.3SD/Aver		**2.48**
LOQ=10SD/Aver		**7.52**

Meperidine Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	106299	10.91	11.25	0.53	112.53	4.71
		106234	10.90				
		106631	10.95				
		102796	10.49				
		105446	10.81				
		105284	10.79				
	Day 2	120450	12.02				
		120244	11.99				
		120396	12.01				
	Day 3	138329	11.38				
		138412	11.39				
		138566	11.40				
25	Day 1	227586	25.54	25.62	0.56	102.46	2.20
		229186	25.73				
		229030	25.71				
		228535	25.65				
		225813	25.33				
		225995	25.35				
	Day 2	256228	26.42				
		256407	26.44				
		256088	26.40				
	Day 3	304602	24.98				
		304017	24.93				
		303835	24.91				
50	Day 1	474009	55.26	50.36	4.62	100.71	9.17
		470813	54.88				
		470061	54.79				
		470707	54.86				
		463097	53.95				
		470676	54.86				
	Day 2	437935	45.69				
		437090	45.60				
		436732	45.57				
	Day 3	566167	46.36				
		565301	46.29				
		563937	46.18				

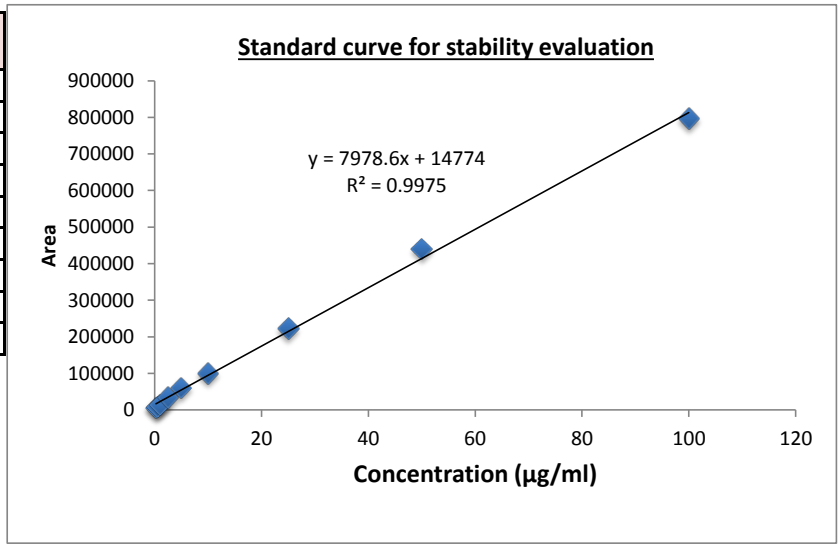
Meperidine Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	106299	10.91	10.81	0.17	108.09	1.56
	106234	10.90				
	106631	10.95				
	102796	10.49				
	105446	10.81				
	105284	10.79				
25	227586	25.54	25.55	0.18	102.21	0.71
	229186	25.73				
	229030	25.71				
	228535	25.65				
	225813	25.33				
	225995	25.35				
50	474009	55.26	54.77	0.44	109.53	0.80
	470813	54.88				
	470061	54.79				
	470707	54.86				
	463097	53.95				
	470676	54.86				

Meperidine Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	100930	10.80	107.98
50	439905	53.28	106.57
100	796690	98.00	98.00

Conc	Peak Area
0.25	4422
0.5	8175
1	14861
2.5	34001
5	61391
10	100930
25	222427
50	439905
100	796690



Methadone Validation HPLC Conditions

Chromatographic conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector

Mobile Phase: ACN: Water=60:40

Flow rate: 1ml/min

Run time: 8min

Retention time: 5.7 min

Column: Kinetex C18 5µ column(150×4.6mm)

Injection Volume: 30 µl

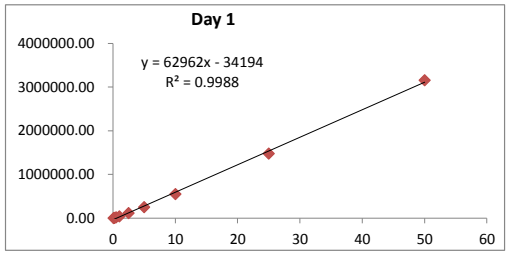
Column Temperature: 30 ℃

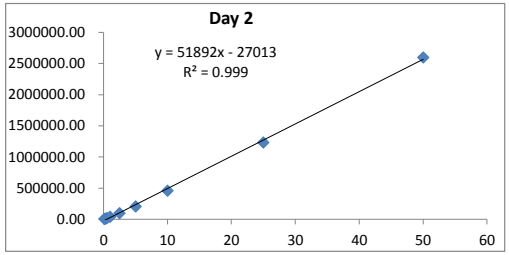
Detection wavelength: 210 nm

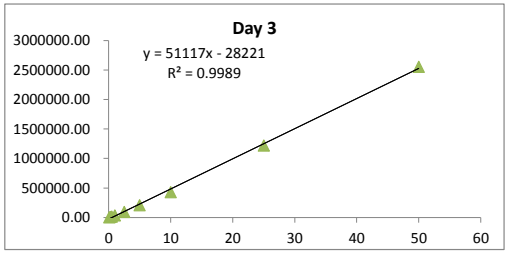
Standards were made in water

Methadone Validation Std Curve

Day 1 Aug 4						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.1	3252	3580	3351	3394.33	168.24	4.96
0.25	9253	9004	9197	9151.33	130.63	1.43
0.5	20641	19947	18881	19823.00	886.53	4.47
1	42657	42200	42987	42614.67	395.20	0.93
2.5	116539	115472	116078	116029.67	535.14	0.46
5	257654	256405	254407	256155.33	1637.83	0.64
10	548954	548889	559134	552325.67	5896.28	1.07
25	1478443	1472984	1480509	1477312.00	3887.90	0.26
50	3150072	3156676	3160890	3155879.33	5452.82	0.17
Day 2 Aug 7						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.1	3051	2906	2982	2979.67	72.53	2.43
0.25	8321	8306	8291	8306.00	15.00	0.18
0.5	17552	17337	17713	17534.00	188.65	1.08
1	37767	36776	37180	37241.00	498.31	1.34
2.5	97393	95368	100674	97811.67	2677.66	2.74
5	208405	205401	202844	205550.00	2783.49	1.35
10	454711	454818	459323	456284.00	2632.39	0.58
25	1219829	1235176	1234944	1229983.00	8794.39	0.72
50	2620703	2588120	2582779	2597200.67	20528.06	0.79
Day 3 Aug 8						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.1	2706	2853	2824	2794.33	77.86	2.79
0.25	7460	7741	7740	7647.00	161.95	2.12
0.5	15377	15954	16598	15976.33	610.81	3.82
1	36269	36560	35153	35994.00	742.72	2.06
2.5	94137	95321	93974	94477.33	735.17	0.78
5	205306	211396	204809	207170.33	3667.96	1.77
10	439627	421411	426913	429317.00	9342.92	2.18
25	1232251	1219451	1212192	1221298.00	10156.25	0.83
50	2568099	2546555	2547941	2554198.33	12058.26	0.47







Methadone Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	62692	34194
STD day 2	51892	27013
STD day 3	51117	28221
Aver.	55233.67	29809.33
SD	6470.72	3844.97
RSD	0.12	12.90
LOD=3.3SD/Aver	0.39	0.43
LOQ=10SD/Aver	1.17	1.29

Methadone Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	Day 1	9253	0.69	0.69	0.02	274.39	2.78
		9004	0.69				
		9197	0.69				
	Day 2	8321	0.71				
		8306	0.71				
		8291	0.71				
		7810	0.67				
		8048	0.68				
		7861	0.67				
	Day 3	7460	0.66				
		7741	0.67				
		7740	0.67				
2.5	Day 1	116539	2.39	2.39	0.05	95.75	2.22
		115472	2.38				
		116078	2.39				
	Day 2	97393	2.46				
		95368	2.42				
		100764	2.52				
		96676	2.38				
		96970	2.39				
		96079	2.37				
	Day 3	94137	2.33				
		95321	2.36				
		93974	2.33				
25	Day 1	1478443	24.02	24.12	0.39	96.48	1.63
		1472984	23.94				
		1480509	24.06				
	Day 2	1219829	24.42				
		1235176	24.72				
		1234944	24.71				
		1206815	23.78				
		1233537	24.29				
		1183493	23.33				
	Day 3	1232251	24.27				
		1219451	24.02				
		1212192	23.88				

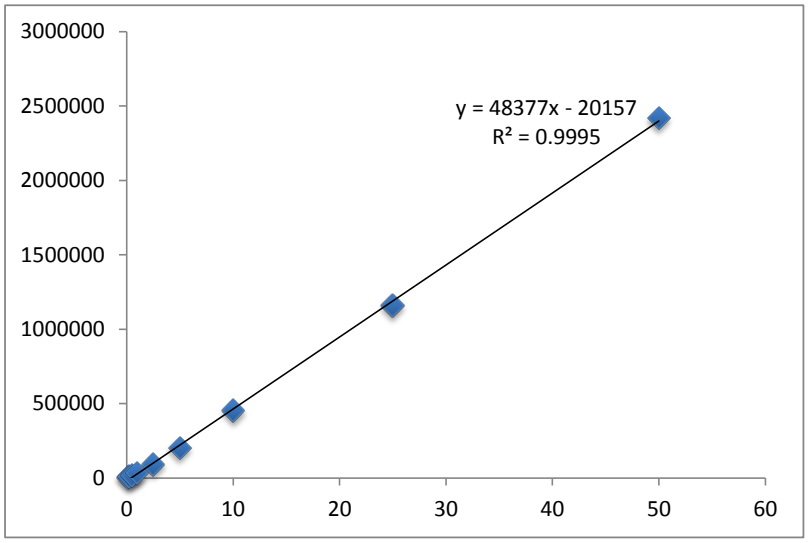
Methadone Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	8321	0.68	0.68	0.00	270.71	0.66
0.25	8306	0.68				
0.25	8291	0.68				
0.25	7810	0.67				
0.25	8048	0.68				
0.25	7861	0.67				
2.5	97393	2.40	2.39	0.04	95.74	1.49
2.5	95368	2.36				
2.5	100674	2.46				
2.5	96676	2.38				
2.5	96970	2.39				
2.5	96079	2.37				
25	1219829	24.03	24.01	0.40	96.04	1.66
25	1235176	24.32				
25	1234944	24.32				
25	1206815	23.78				
25	1233537	24.29				
25	1183493	23.33				

Methadone Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
0.25	7699	0.58	230.32
2.5	91554	2.31	92.37
25	1159253	24.38	97.52

Conc	Peak Area
0.1	2759
0.25	7699
0.5	15278
1	32575
2.5	91554
5	204284
10	451569
25	1159253
50	2418031



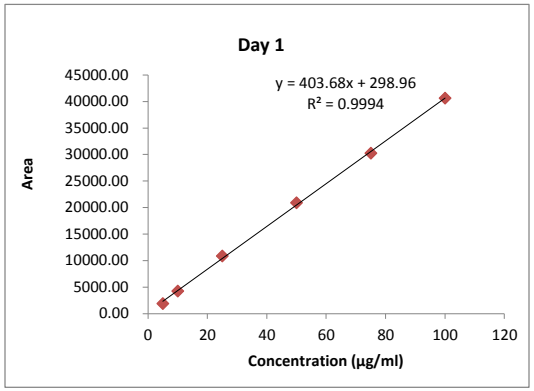
Methylphenidate Validation HPLC Conditions

Chromatographic conditions:

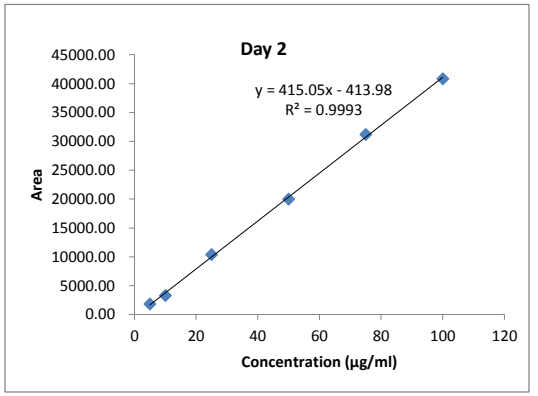
System: Waters e2795
Detector: Waters 2988 photodiode array
Pump Mode: Isocratic
Mobile Phase: Methanol (0.1% FA): water (0.1% FA, pH 6.8) (50:50)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 15 min
Retention time: 9.8min
Column: (size and particle size): kinetex 5u, Biphenyl 100A, 250* 4.6mm
Column Temperature: 25°C
Detection wavelength: 258nm
Sample Preparation: Standards were preapred in HPLC grade water

Methylphenidate Validation Std Curve

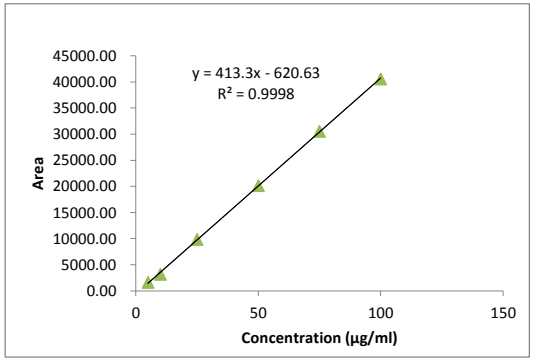
Day 1(05/18/15)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
5	2073	1894	1700	1889.00	186.55	9.88
10	4360	4408	3965	4244.33	243.10	5.73
25	10749	10620	11265	10878.00	341.30	3.14
50	20961	20926	20804	20897.00	82.42	0.39
75	30682	30727	29318	30242.33	800.81	2.65
100	40608	41055	40191	40618.00	432.09	1.06



Day 2(05/19/15)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
5	2361	1628	1493	1827.33	467.07	25.56
10	3363	3328	3144	3278.33	117.64	3.59
25	10993	9996	10171	10386.67	532.34	5.13
50	19955	20075	19881	19970.33	97.90	0.49
75	30359	32085	31145	31196.33	864.14	2.77
100	41451	41078	40007	40845.33	749.59	1.84



Day 3 (05/21/15)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
5	1355	1712	1743	1603.33	215.62	13.45
10	3446	2961	3045	3150.67	259.19	8.23
25	10239	9430	9846	9838.33	404.55	4.11
50	19735	20002	20690	20142.33	492.72	2.45
75	30789	30671	30057	30505.67	393.01	1.29
100	40062	41165	40455	40560.67	559.04	1.38



Methylphenidate Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	403.68	298.96
STD day 2	415.05	413.98
STD day 3	413.2	636.08
Aver.	410.64	
SD		171.37
LOD=3.3SD/Aver		**1.38**
LOQ=10SD/Aver		**4.17**

Methylphenidate Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
50	Day 1	20961	51.18	49.33	1.76	98.66	3.57
		20926	51.10				
		20804	50.80				
	Day 2	19955	49.08				
		20075	49.37				
		19881	48.90				
	Day 3	19735	49.25				
		20002	49.90				
		20690	51.56				
		19239	48.05				
		18851	47.11				
		18259	45.68				
75	Day 1	30682	75.27	74.11	2.68	98.81	3.61
		30727	75.38				
		29318	71.89				
	Day 2	30359	74.14				
		32085	78.30				
		31145	76.04				
	Day 3	30789	76.00				
		30671	75.71				
		30057	74.23				
		27583	68.24				
		28953	71.55				
		29365	72.55				
100	Day 1	40608	99.85	97.10	4.76	97.10	4.90
		41055	100.96				
		40191	98.82				
	Day 2	41451	100.87				
		41078	99.97				
		40007	97.39				
	Day 3	40062	98.43				
		41165	101.10				
		40455	99.38				
		36681	90.25				
		35700	87.88				
		36702	90.30				

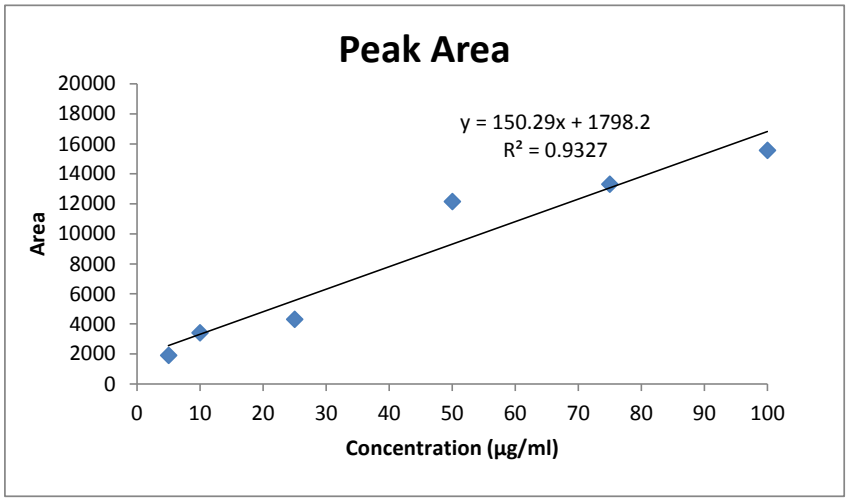
Methylphenidate Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
50	19735	49.25	48.59	2.09	97.19	4.31
50	20002	49.90				
50	20690	51.56				
50	19239	48.05				
50	18851	47.11				
50	18259	45.68				
75	30789	76.00	73.05	2.92	97.40	4.00
75	30671	75.71				
75	30057	74.23				
75	27583	68.24				
75	28953	71.55				
75	29365	72.55				
100	40062	98.43	94.56	5.70	94.56	6.03
100	41165	101.10				
100	40455	99.38				
100	36681	90.25				
100	35700	87.88				
100	36702	90.30				

Methylphenidate Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
50	12148	62.27	124.54
75	13304	68.76	91.68
100	15570	81.49	81.49

Conc	Peak Area
5	1893
10	3399
25	4301
50	12148
75	13304
100	15570



Morphine Validation HPLC Conditions

Chromatographic conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector

Mobile Phase: MeOH: NH4OAc Buffer(0.01M, PH5.5)=10:90

Flow rate: 1ml/min

Run time: 6 min

Retention time: 3min

Column: Kinetex C18 5µ column(150×4.6mm)

Injection Volume: 30 µl

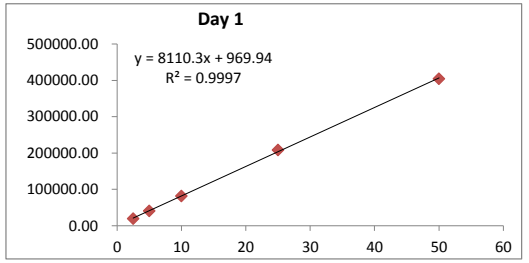
Column Temperature: 45 ℃

Detection wavelength: 285nm

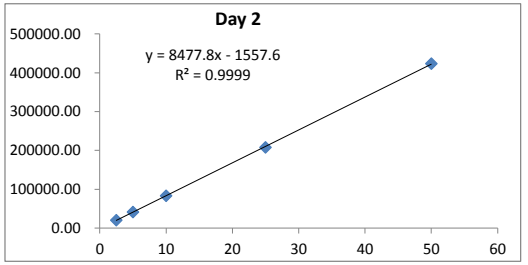
Standards were made in water

Morphine Validation Std Curve

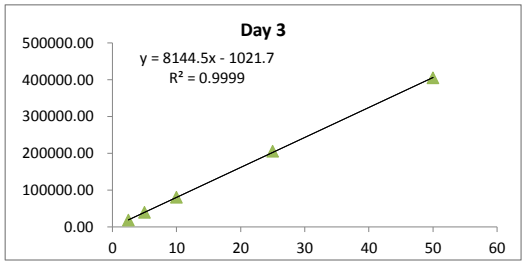
Day 1						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
2.5	18117	20703	20110	19643.33	1354.69	6.90
5	45029	39803	37956	40929.33	3668.56	8.96
10	78868	81345	84966	81726.33	3066.83	3.75
25	208880	210324	205972	208392.00	2216.66	1.06
50	399892	405956	407235	404361.00	3922.75	0.97



Day 2						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
2.5	20625	19439	21126	20396.67	866.37	4.25
5	41628	42430	40118	41392.00	1173.93	2.84
10	84347	82809	82147	83101.00	1128.69	1.36
25	204967	210659	208746	208124.00	2896.53	1.39
50	419414	419664	431098	423392.00	6674.76	1.58



Day 3						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
2.5	18477	18051	18815	18447.67	382.84	2.08
5	35613	41644	39996	39084.33	3117.14	7.98
10	81262	81703	78141	80368.67	1941.78	2.42
25	202020	210780	203642	205480.67	4660.46	2.27
50	407311	401432	405878	404873.67	3065.48	0.76



Morphine Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	8110.3	969.94
STD day 2	8477.8	1557.6
STD day 3	8144.5	1021.7
Aver.	8244.20	
SD		325.37
LOD=3.3SD/Aver		0.13
LOQ=10SD/Aver		0.39

Morphine Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL	SD	Accuracy (%)	Precision (%)
2.5	Day 1	18117	2.11	2.39	0.17	95.67	6.95
		20703	2.43				
		20110	2.36				
	Day 2	20625	2.62				
		19439	2.48				
		21126	2.68				
	Day 3	18477	2.39				
		18051	2.34				
		16281	2.12				
		18815	2.44				
		17706	2.30				
		18764	2.43				
10	Day 1	78868	9.60	9.95	0.21	99.53	2.14
		81345	9.91				
		84966	10.36				
	Day 2	84347	10.13				
		82809	9.95				
		82147	9.87				
	Day 3	81262	10.10				
		81703	10.16				
		78141	9.72				
		78338	9.74				
		80508	10.01				
		79358	9.87				
25	Day 1	208880	25.64	25.19	0.45	100.78	1.78
		210324	25.81				
		205972	25.28				
	Day 2	204967	24.36				
		210659	25.03				
		208746	24.81				
	Day 3	202020	24.93				
		210780	26.01				
		203642	25.13				
		203202	25.08				
		203383	25.10				
		203982	25.17				

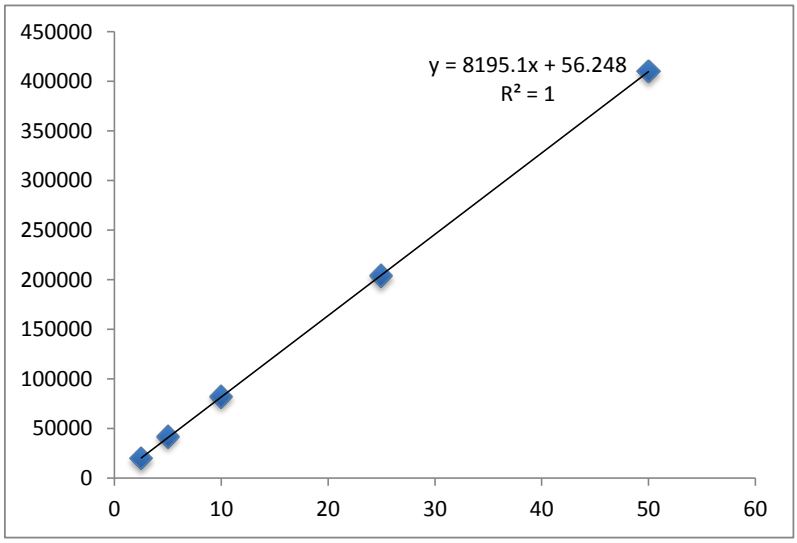
Morphine Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
2.5	18477	2.39	2.34	0.12	93.50	4.99
2.5	18051	2.34				
2.5	16281	2.12				
2.5	18815	2.44				
2.5	17706	2.30				
2.5	18764	2.43				
10	81262	10.10	9.93	0.18	99.34	1.86
10	81703	10.16				
10	78141	9.72				
10	78338	9.74				
10	80508	10.01				
10	79358	9.87				
25	202020	24.93	25.23	0.39	100.94	1.53
25	210780	26.01				
25	203642	25.13				
25	203202	25.08				
25	203383	25.10				
25	203982	25.17				

Morphine Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
2.5	19598	2.38	95.38
10	81638	9.95	99.55
25	202114	24.66	98.62

Conc	Peak Area
2.5	20059
5	41682
10	82393
25	204067
50	410126



Oxycodone Validation_1 HPLC Conditions

Chromatographic conditions:

System: Alliance HPLC Waters 2695 with Waters 2996 PDA detector

Mobile Phase: ACN: NaH_2PO_4 Buffer(0.02M, PH 3.4)=10:90

Flow rate: 1ml/min

Run time: 15 min

Retention time: 9 min

Column: Kinetex C18 5μ column(250×4.6mm)

Injection Volume: 10 μl

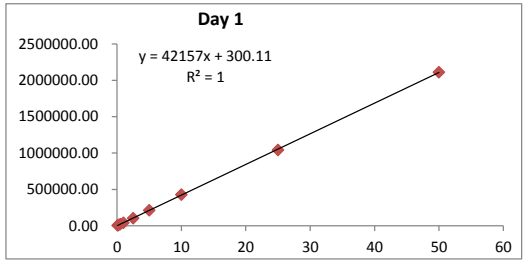
Column Temperature: 25 ℃

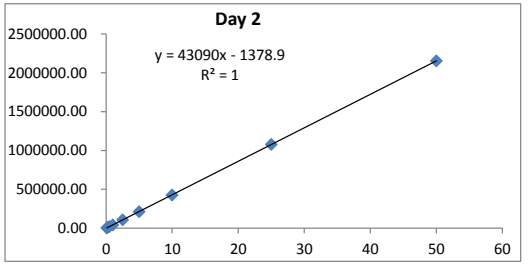
Detection wavelength: 205nm

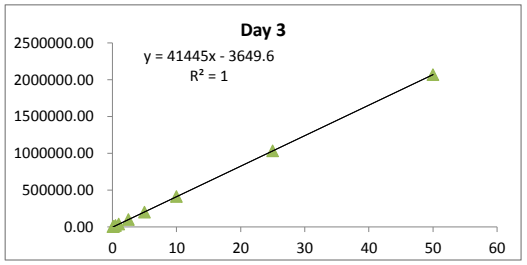
Standards were made in water

Oxycodone Validation_1 Std Curve

Day 1 Nov 26						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.1	4127	4162	4331	4206.67	109.09	2.59
0.25	10566	9664	10550	10260.00	516.21	5.03
0.5	22252	21874	23180	22435.33	672.02	3.00
1	43039	40993	41455	41829.00	1073.05	2.57
2.5	105299	103909	105030	104746.00	737.24	0.70
5	214824	212325	212475	213208.00	1401.51	0.66
10	429605	426550	428864	428339.67	1593.57	0.37
25	1039017	1041169	1048035	1042740.33	4709.87	0.45
50	2105952	2110374	2120926	2112417.33	7693.28	0.36
Day 2 Nov 27						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.1	3769	3872	3399	3680.00	248.74	6.76
0.25	9150	9923	10010	9694.33	473.41	4.88
0.5	19954	20205	20938	20365.67	511.30	2.51
1	41307	41529	41077	41304.33	226.01	0.55
2.5	108830	108801	107624	108418.33	688.07	0.63
5	213790	213367	211450	212869.00	1246.96	0.59
10	429504	424726	426188	426806.00	2448.22	0.57
25	1086479	1071194	1072565	1076746.00	8456.85	0.79
50	2160086	2143998	2155648	2153244.00	8309.05	0.39
Day 3 Nov 28						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.1	3272	3161	3356	3263.00	97.81	3.00
0.25	8457	8284	8695	8478.67	206.35	2.43
0.5	18618	19023	18926	18855.67	211.46	1.12
1	37560	36190	35547	36432.33	1028.15	2.82
2.5	97205	98103	99306	98204.67	1054.18	1.07
5	198989	198864	197502	198451.67	824.81	0.42
10	408826	415263	412894	412327.67	3255.66	0.79
25	1037149	1031560	1029422	1032710.33	3989.87	0.39
50	2084520	2062666	2059183	2068789.67	13733.73	0.66







Oxycodone Validation_1 LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	42157	300.11
STD day 2	43090	1378.9
STD day 3	41445	3649.6
Aver.	42230.67	
SD		1709.72
LOD=3.3SD/Aver		0.13
LOQ=10SD/Aver		0.40

Oxycodone Validation_1 Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	Day 1	10566	0.24	0.26	0.02	103.73	9.21
		9664	0.22				
		10550	0.24				
	Day 3	8457	0.29				
		8284	0.29				
		8695	0.30				
	Day 2	9923	0.26				
		9150	0.24				
		9357	0.25				
		10010	0.26				
		8788	0.24				
		10218	0.27				
2.5	Day 1	105299	2.49	2.50	0.04	99.88	1.55
		103909	2.46				
		105030	2.48				
	Day 3	97205	2.43				
		98103	2.46				
		99306	2.48				
	Day 2	108830	2.56				
		108801	2.56				
		107624	2.53				
		106475	2.50				
		107119	2.52				
		106125	2.49				
25	Day 1	1039017	24.64	24.86	0.20	99.45	0.80
		1041169	24.69				
		1048035	24.85				
	Day 3	1037149	25.11				
		1031560	24.98				
		1029422	24.93				
	Day 2	1086479	25.25				
		1071194	24.89				
		1072565	24.92				
		1066434	24.78				
		1056222	24.54				
		1065562	24.76				

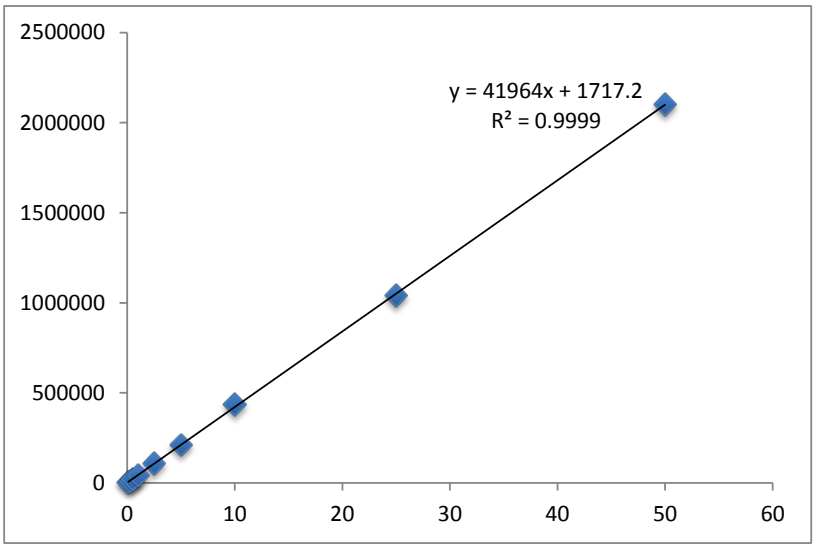
Oxycodone Validation_1 Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
0.25	9923	0.26	0.25	0.01	101.68	5.12
0.25	9150	0.24				
0.25	9357	0.25				
0.25	10010	0.26				
0.25	8788	0.24				
0.25	10218	0.27				
2.5	108830	2.56	2.53	0.03	101.07	1.05
2.5	108801	2.56				
2.5	107624	2.53				
2.5	106475	2.50				
2.5	107119	2.52				
2.5	106125	2.49				
25	1086479	25.25	24.86	0.23	99.43	0.93
25	1071194	24.89				
25	1072565	24.92				
25	1066434	24.78				
25	1056222	24.54				
25	1065562	24.76				

Oxycodone Validation_1 Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
0.25	9707	0.19	76.16
2.5	104105	2.44	97.60
25	1052923	25.05	100.20

Conc	Peak Area
0.1	4585
0.25	9910
0.5	20820
1	42337
2.5	106746
5	211891
10	434358
25	1042760
50	2101393



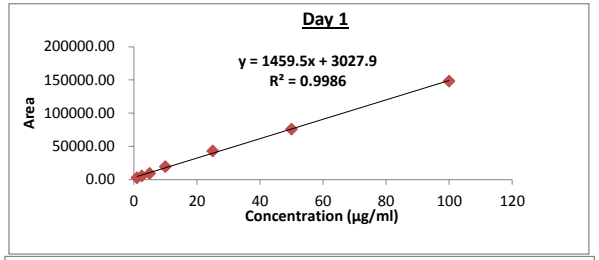
Oxycodone Validation_2 HPLC Conditions

Chromatographic conditions: Oxycodone HCL

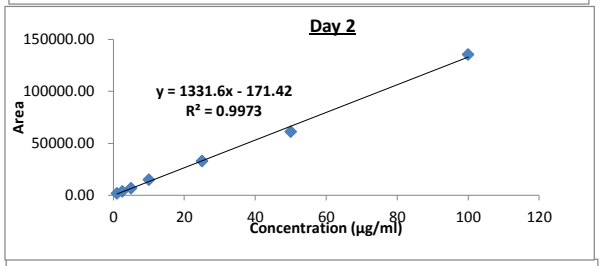
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN:Buffer (DI water with 0.1% TFA pH=1.5) (15:85)
Flow rate: 1.2 ml/min
Injection Volume: 10 µL
Run time: 10min
Retention time: 3.6 min
Column: (size and particle size): Phenomenex, Kinetex C18 5µm, 250×4.6mm
Column Temperature: 55°C
Detection wavelength: 285 nm
Sample Preparation: Standards were prepared in DI water

Oxycodone Validation_2 Std Curve

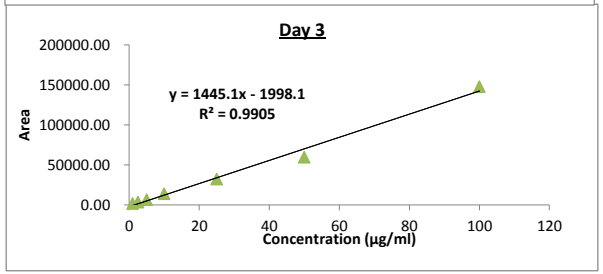
Day 1 (08.12.16)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
1	2516	2598	2608	2574.00	50.48	1.96
2.5	5284	5079	5436	5266.33	179.15	3.40
5	9279	9402	9469	9383.33	96.37	1.03
10	19197	19503	19152	19284.00	190.99	0.99
25	43266	43109	43055	43143.33	109.61	0.25
50	75730	75845	76122	75899.00	201.50	0.27
100	147821	148095	148258	148058.00	220.84	0.15



Day 2 (08.15.16)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
1	1755	1812	1753	1773.33	33.50	1.89
2.5	3396	3602	3678	3558.67	145.91	4.10
5	6670	6624	6661	6651.67	24.38	0.37
10	14971	14751	15043	14921.67	152.12	1.02
25	32625	32903	33287	32938.33	332.41	1.01
50	61328	61042	61138	61169.33	145.55	0.24
100	135589	135463	135298	135450.00	145.93	0.11



Day 3 (08.16.16)						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
1	1707	1748	1715	1723.33	21.73	1.26
2.5	3493	3483	3473	3483.00	10.00	0.29
5	6692	6396	6529	6539.00	148.25	2.27
10	13761	13907	13939	13869.00	94.89	0.68
25	32436	32627	32170	32411.00	229.52	0.71
50	59427	59699	59818	59648.00	200.43	0.34
100	147832	148250	147843	147975.00	238.22	0.16



Oxycodone Validation_2 LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	1459.5	3027.9
STD day 2	1331.6	171.42
STD day 3	1445.1	1998.1
Aver.	1412.07	
SD		1446.65
LOD=3.3SD/Aver		3.38
LOQ=10SD/Aver		10.24

Oxycodone Validation_2 Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	19197	11.08	11.15	0.16	111.49	1.47
		19503	11.29				
		19152	11.05				
		19082	11.00				
		19476	11.27				
		19320	11.16				
	Day 2	14971	11.37				
		14751	11.21				
		15043	11.43				
	Day 3	13761	10.91				
		13907	11.01				
		13939	11.03				
25	Day 1	43266	27.57	25.87	1.65	103.48	6.40
		43109	27.46				
		43055	27.43				
		43069	27.43				
		42721	27.20				
		42922	27.33				
	Day 2	32625	24.63				
		32903	24.84				
		33287	25.13				
	Day 3	32436	23.83				
		32627	23.96				
		32170	23.64				
50	Day 1	75730	49.81	47.11	3.14	94.22	6.66
		75845	49.89				
		76122	50.08				
		75996	50.00				
		75750	49.83				
		75302	49.52				
	Day 2	61328	46.18				
		61042	45.97				
		61138	46.04				
	Day 3	59427	42.51				
		59699	42.69				
		59818	42.78				

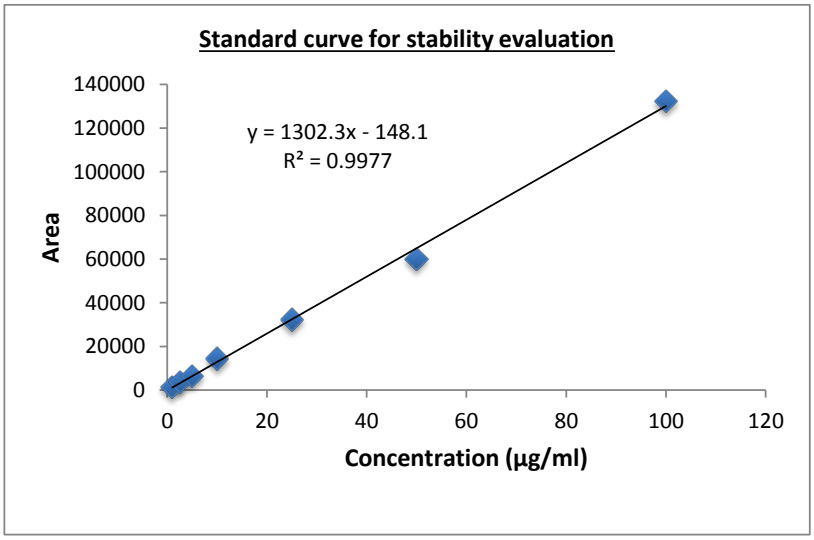
Oxycodone Validation_2 Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	19197	11.08	11.14	0.12	111.41	1.07
10	19503	11.29				
10	19152	11.05				
10	19082	11.00				
10	19476	11.27				
10	19320	11.16				
25	43266	27.57	27.40	0.13	109.61	0.46
25	43109	27.46				
25	43055	27.43				
25	43069	27.43				
25	42721	27.20				
25	42922	27.33				
50	75730	49.81	49.85	0.19	99.71	0.39
50	75845	49.89				
50	76122	50.08				
50	75996	50.00				
50	75750	49.83				
50	75302	49.52				

Oxycodone Validation_2 Stability

Intial conc. (µg/mL)	Storage at 4°C after 14 days (08.29.16)		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	14445	11.21	112.06
25	32269	24.89	99.57
50	60166	46.31	92.63

Conc	Peak Area
1	1598
2.5	3664
5	6477
10	14445
25	32269
50	60166
100	132330



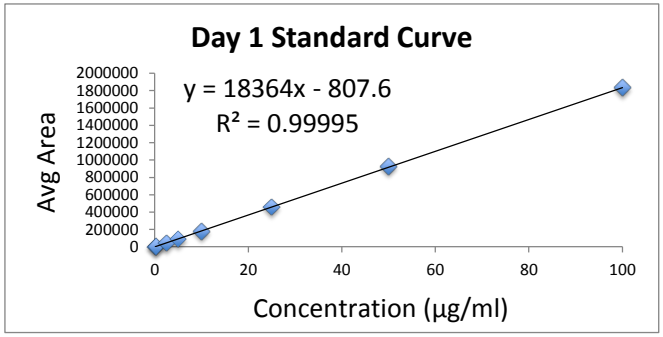
Quetiapine Validation HPLC Conditions

Chromatographic conditions: **Quetiapine Fumarate**
Date: April 25, 2016

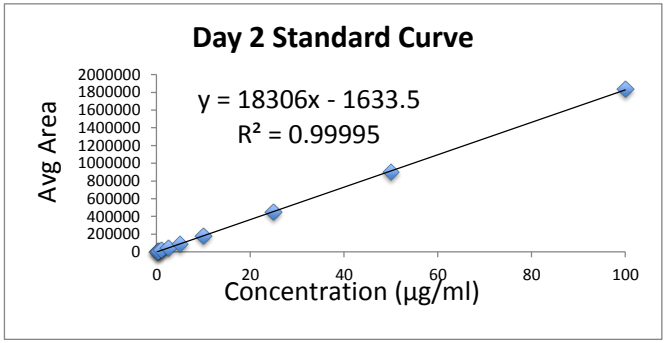
System: Waters 2695 Separations Module
Detector: Waters 2996 Photodiod Array Detector
Pump Mode: Isocratic
Mobile Phase: ACN:Water (0.1% O-phophoric acid (pH 3)) 35:65 (v/v)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 12min
Retention time: 5.9 min
Column: (size and particle size): Waters, XBridge C18, 3.5µm, 250×4.6mm
Column Temperature: 40°C
Wavelength 294nm
Sample Preparation: Standards were prepared in Acetonitrile:H2O 35:65 (v/v)

Quetiapine Validation Std Curve

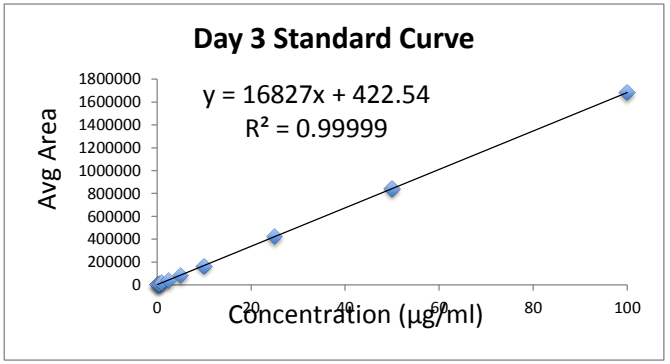
Day 1(04/25/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	2029	2134	2022	2062	62.7	3.0
0.25	4218	4955	4808	4660	390.1	8.4
2.5	44644	44313	44032	44330	306.3	0.7
5	91572	91559	91424	91518	82.0	0.1
10	178333	176882	177393	177536	736.0	0.4
25	455824	455935	455918	455892	59.8	0.0
50	926371	927479	927916	927255	796.4	0.1
100	1831660	1831933	1831741	1831778	140.2	0.0



Day 2(04/26/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	2238	2148	1565	1984	365.4	18.4
0.25	4417	4561	3938	4305	326.2	7.6
0.5	8011	8502	8350	8288	251.4	3.0
1	17997	18535	18371	18301	275.7	1.5
2.5	45438	44998	45110	45182	228.7	0.5
5	87048	88036	88175	87753	614.5	0.7
10	183402	182938	182979	183106	256.9	0.1
25	453806	454840	455420	454689	817.6	0.2
50	901512	904177	905100	903596	1863.1	0.2
100	1832658	1834415	1835520	1834198	1443.3	0.1



Day 3(04/27/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	1431	1767	1727	1642	183.5	11.2
0.25	4394	4164	4065	4208	168.8	4.0
0.5	8579	8365	8335	8426	133.1	1.6
1	16652	15983	17045	16560	536.9	3.2
2.5	42108	41923	41927	41986	105.7	0.3
5	84190	84665	84263	84373	255.8	0.3
10	167016	167484	168493	167664	754.8	0.5
25	426651	425044	424553	425416	1097.4	0.3
50	842343	842508	842280	842377	117.7	0.0
100	1683943	1681255	1680266	1681821	1902.8	0.1



Quetipine Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	18364	807.6
STD day 2	18306	1633.5
STD day 3	16827	422.54
Aver.	17832.33	
SD		618.71
LOD=3.3SD/Aver		**0.11**
LOQ=10SD/Aver		**0.35**

Quetiapine Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
2.5	Day 1	44644	2.5	2.5	0.0	99.6	1.7
		44313	2.5				
		44032	2.4				
	Day 2	45438	2.6				
		44998	2.5				
		45110	2.6				
	Day 3	42108	2.5				
		41923	2.5				
		41927	2.5				
		42490	2.5				
		42098	2.5				
		41844	2.5				
10	Day 1	178333	9.8	9.9	0.1	99.2	1.5
		176882	9.7				
		177393	9.7				
	Day 2	183402	10.1				
		182938	10.1				
		182979	10.1				
	Day 3	167016	9.9				
		167484	9.9				
		168493	10.0				
		167776	9.9				
		167999	10.0				
		167474	9.9				
50	Day 1	926371	50.5	50.0	0.4	100.1	0.8
		927479	50.5				
		927916	50.6				
	Day 2	901512	49.3				
		904177	49.5				
		905100	49.5				
	Day 3	842343	50.0				
		842508	50.0				
		842280	50.0				
		842637	50.1				
		844676	50.2				
		843953	50.1				

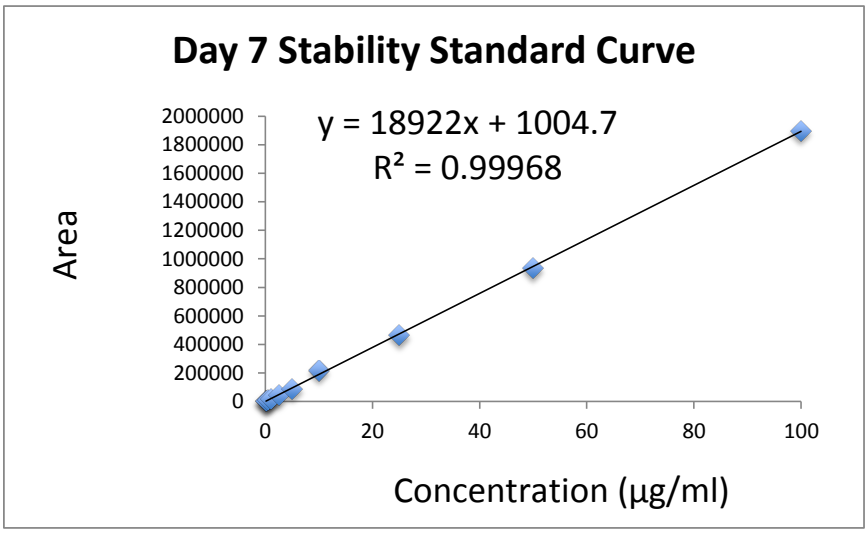
Quetiapine Validation Intraday Accuracy & Precision

Intraday Accuracy and Precision (Day 3)						
Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
2.5	42108	2.5	2.5	0.0	99.0	0.6
2.5	41923	2.5				
2.5	41927	2.5				
2.5	42490	2.5				
2.5	42098	2.5				
2.5	41844	2.5				
10	167016	9.9	9.9	0.0	99.4	0.3
10	167484	9.9				
10	168493	10.0				
10	167776	9.9				
10	167999	10.0				
10	167474	9.9				
50	842343	50.0	50.1	0.1	100.2	0.1
50	842508	50.0				
50	842280	50.0				
50	842637	50.1				
50	844676	50.2				
50	843953	50.1				

Quetiapine Validation Stability

Day 7 Stability (5/3/16)			
Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
2.5	46267	2	96
10	218296	11	115
50	935595	49	99

Concentration (µg/ml)	Peak Area
0.1	2182
0.25	4559
0.5	8272
1	18898
2.5	46267
5	90242
10	218296
25	464256
50	935595
100	1898891



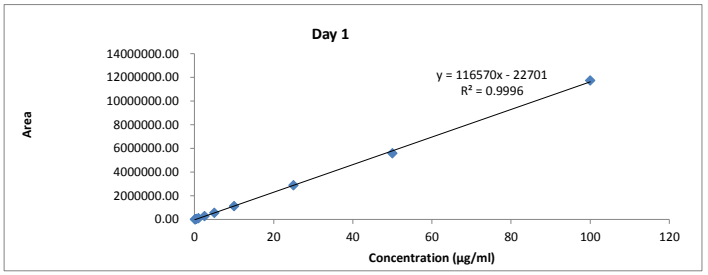
Temazepam Validation HPLC Conditions

Chromatographic conditions: Temazepam

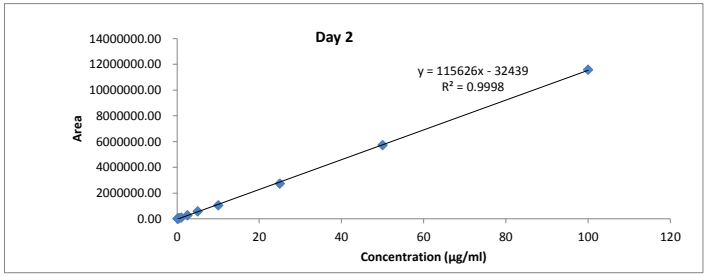
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN:Buffer (KH2PO4 0.02M pH=3) (45:55)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 4.8min
Column: (size and particle size): Phenomenex, Kinetex C18 5µm, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 230 nm
Sample Preparation: Standards were prepared in Methanol and Water (90:10)

Temazepam Validation Std Curve

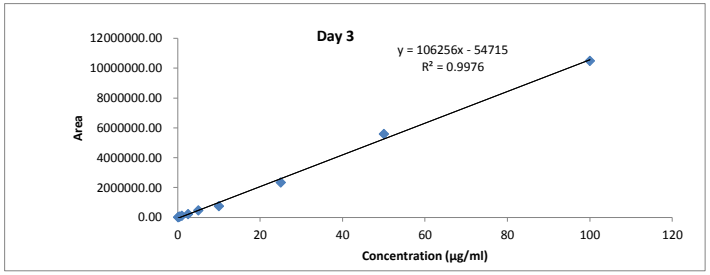
Day 1 (01/19/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	10952	10983	10897	10944.00	43.55	0.40
0.25	28882	28853	28383	28706.00	280.10	0.98
0.5	54726	53447	53959	54044.00	643.72	1.19
1	111270	111497	112284	111683.67	532.15	0.48
2.5	291129	288806	291489	290474.67	1456.27	0.50
5	561286	560675	564387	562116.00	1990.33	0.35
10	1135118	1129653	1143100	1135957.00	6762.65	0.60
25	2873783	2911616	2911012	2898803.67	21670.64	0.75
50	5565078	5585945	5652062	5601028.33	45411.28	0.81
100	11575645	11790743	11837184	11734524.00	139538.86	1.19



Day 2 (01/20/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	9868	9876	10134	9959.33	151.32	1.52
0.25	27393	26617	26581	26863.67	458.77	1.71
0.5	57093	57257	57061	57137.00	105.15	0.18
1	104338	104652	105112	104700.67	389.29	0.37
2.5	280508	278248	274455	277737.00	3058.68	1.10
5	581180	575620	577880	578226.67	2796.16	0.48
10	1056712	1057643	1050959	1055104.67	3620.30	0.34
25	2720511	2735561	2757474	2737848.67	18587.39	0.68
50	5727751	5671320	5767101	5722057.33	48143.67	0.84
100	11538214	11539542	11655943	11577899.67	67590.77	0.58



Day 3 (01/21/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	12789	12604	12603	12665.33	107.10	0.85
0.25	31920	31885	31591	31798.67	180.69	0.57
0.5	62753	56543	47385	55560.33	7730.98	13.91
1	109786	121757	121360	117634.33	6799.75	5.78
2.5	232082	214240	225784	224035.33	9048.63	4.04
5	509641	470841	447582	476021.33	31352.14	6.59
10	849003	721549	715914	762155.33	75265.04	9.88
25	2408346	2148609	2465335	2340763.33	168832.47	7.21
50	5763142	5768794	5237044	5589660.00	305387.49	5.46
100	11117022	10229025	10134477	10493508.00	542044.38	5.17



Temazepam Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	116570	22701
STD day 2	115626	32439
STD day 3	106256	54715
Aver.	112817.33	
SD		16411.10
LOD=3.3SD/Aver		**0.48**
LOQ=10SD/Aver		**1.45**

Temazepam Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	1135118	9.93	9.23	1.01	92.34	10.89
		1129653	9.89				
		1143100	10.00				
		1124663	9.84				
		1133093	9.92				
		1137730	9.95				
	Day 2	1056712	9.42				
		1057643	9.43				
		1050959	9.37				
	Day 3	849003	8.51				
		721549	7.31				
		715914	7.25				
25	Day 1	2935233	25.37	24.25	1.36	97.00	5.61
		2943035	25.44				
		2873783	24.85				
		2911616	25.17				
		2911012	25.17				
		2947296	25.48				
	Day 2	2720511	23.81				
		2735561	23.94				
		2757474	24.13				
	Day 3	2408346	23.18				
		2148609	20.74				
		2465335	23.72				
50	Day 1	5534519	47.67	49.68	2.57	99.35	5.17
		5565078	47.93				
		5585945	48.11				
		5565610	47.94				
		5467342	47.10				
		5652026	48.68				
	Day 2	5727751	49.82				
		5671320	49.33				
		5767101	50.16				
	Day 3	5763142	54.75				
		5768794	54.81				
		5237044	49.80				

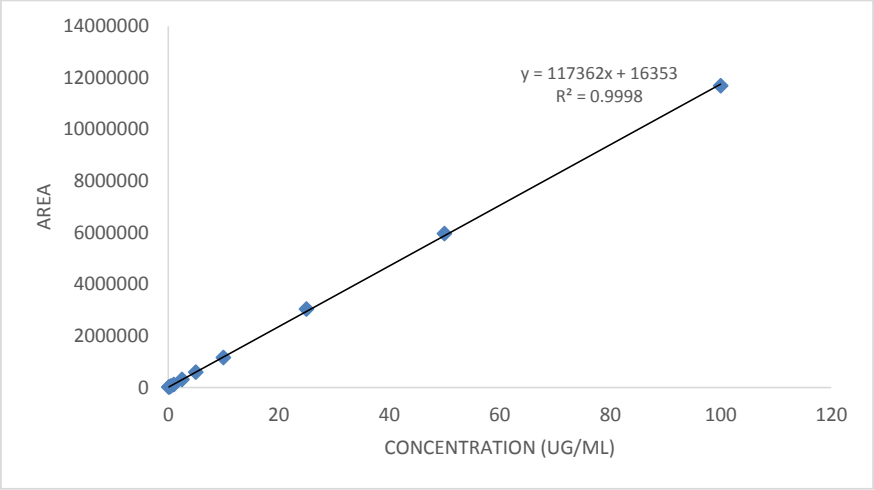
Temazepam Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	1135118	9.93	9.92	0.05	99.22	0.55
	1129653	9.89				
	1143100	10.00				
	1124663	9.84				
	1133093	9.92				
	1137730	9.95				
25	2935233	25.37	25.25	0.24	100.99	0.94
	2943035	25.44				
	2873783	24.85				
	2911616	25.17				
	2911012	25.17				
	2947296	25.48				
50	5534519	47.67	47.91	0.52	95.81	1.09
	5565078	47.93				
	5585945	48.11				
	5565610	47.94				
	5467342	47.10				
	5652026	48.68				

Temazepam Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	1156863	9.72	97.18
50	5966132	50.70	101.39
100	11693631	99.50	99.50

Conc	Peak Area
0.1	11224
0.25	29799
0.5	55842
1	111903
2.5	313470
5	595420
10	1156863
25	3038496
50	5966132
100	11693631



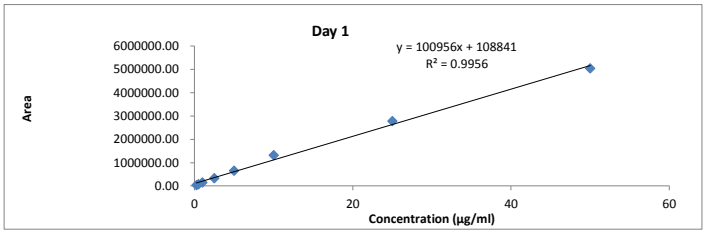
Tramadol Validation HPLC Conditions

Chromatographic conditions: Tramadol

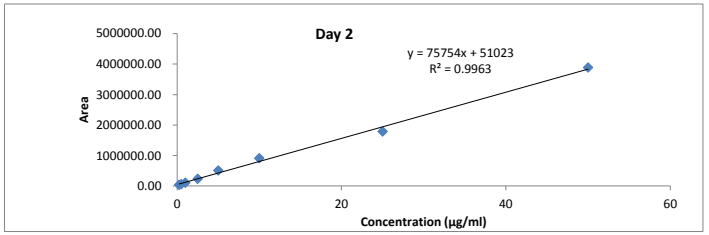
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN: Water (0.2% TFA) (30:70) (% v/v)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 3.4 min
Column: (size and particle size): Phenomenex, Kinetex EVO C18 5µm, 250×4.6mm
Column Temperature: 25°C
Detection wavelength: 200 nm
Sample Preparation: Standards were prepared in Water

Tramadol Validation Std Curve

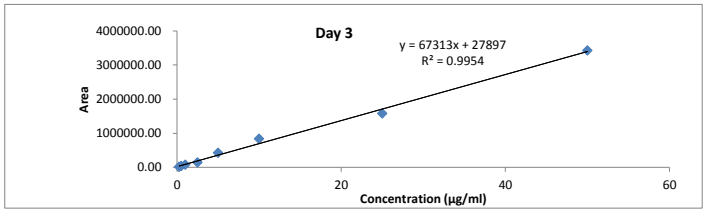
Day 1						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.25	33485	38393	33854	35244.00	2733.35	7.76
0.5	92513	57019	78409	75980.33	17871.20	23.52
1	159371	145450	148641	151154.00	7292.80	4.82
2.5	330801	322457	339033	330763.67	8288.06	2.51
5	640092	649567	668376	652678.33	14396.40	2.21
10	1340524	1321503	1282065	1314697.33	29817.80	2.27
25	2773735	2798805	2776124	2782888.00	13836.18	0.50
50	5028300	5013330	5085650	5042426.67	38173.52	0.76



Day 2						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.25	46941	34103	32381	37808.33	7955.85	21.04
0.5	65716	52171	68510	62132.33	8739.15	14.07
1	120285	108339	101301	109975.00	9597.16	8.73
2.5	245184	246569	224715	238822.67	12237.21	5.12
5	508294	511918	508479	509563.67	2041.01	0.40
10	910948	900501	924131	911860.00	11841.37	1.30
25	1801380	1785979	1779416	1788925.00	11274.46	0.63
50	3976822	3688958	4000880	3888886.67	173560.65	4.46



Day 3						
Concentration (µg/ml)	**Inj.1**	**Inj.2**	**Inj.3**	**Avg Area**	**SD**	**RSD**
0.25	17944	17180	16845	17323.00	563.28	3.25
0.5	41057	38623	45508	41729.33	3491.39	8.37
1	88272	79027	73421	80240.00	7499.44	9.35
2.5	151832	147091	150621	149848.00	2463.21	1.64
5	441067	424530	418954	428183.67	11500.35	2.69
10	845174	861517	816414	841035.00	22834.59	2.72
25	1556510	1603030	1589953	1583164.33	23991.50	1.52
50	3589090	3460950	3227679	3425906.33	183236.25	5.35



Tramadol Validation LOD & LOQ

LOD and LOQ	**Slope**	**Y-intercept**
STD day 1	100956	108841
STD day 2	75754	51023
STD day 3	67313	27897
Aver.	81341.00	
SD		41692.65
LOD=3.3SD/Aver		**1.69**
LOQ=10SD/Aver		**5.13**

Tramadol Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
5	Day 1	640092	5.26	6.04	0.53	120.73	8.72
		649567	5.36				
		668376	5.54				
	Day 2	508294	6.04				
		511918	6.08				
		494608	5.86				
		508479	6.04				
		501601	5.95				
		504438	5.99				
	Day 3	441067	6.97				
		424530	6.72				
		418954	6.64				
10	Day 1	1340524	12.20	11.81	0.78	118.07	6.59
		1321503	12.01				
		1282065	11.62				
	Day 2	910948	11.35				
		888412	11.05				
		887053	11.04				
		900501	11.21				
		879585	10.94				
		924131	11.53				
	Day 3	845174	12.97				
		861517	13.21				
		816414	12.54				
25	Day 1	2773735	26.40	23.55	2.12	94.20	9.01
		2798805	26.64				
		2776124	26.42				
	Day 2	1801380	20.55				
		1785979	22.90				
		1627278	20.81				
		1779416	22.82				
		1790157	22.96				
		1664938	21.30				
	Day 3	1556510	23.54				
		1603030	24.23				
		1589953	24.03				

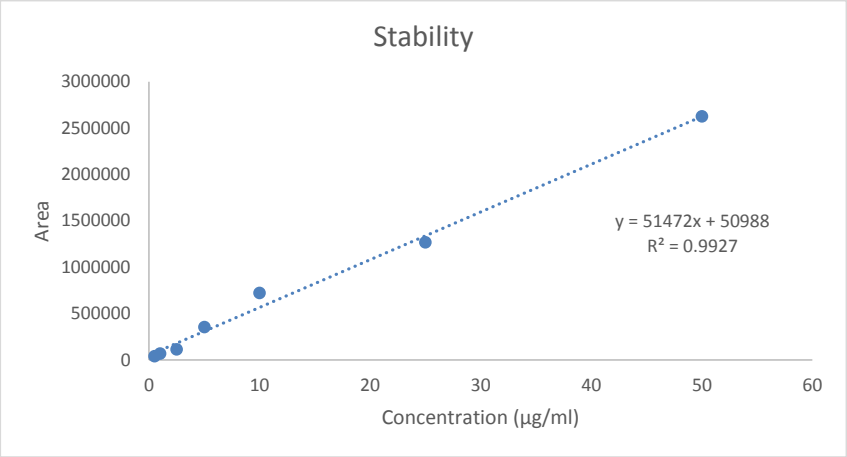
Tramadol Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
5	508294	6.04	5.99	0.08	119.83	1.36
	511918	6.08				
	494608	5.86				
	508479	6.04				
	501601	5.95				
	504438	5.99				
10	910948	11.35	11.19	0.22	111.86	1.98
	888412	11.05				
	887053	11.04				
	900501	11.21				
	879585	10.94				
	924131	11.53				
25	1801380	23.11	22.32	0.99	89.26	4.45
	1785979	22.90				
	1627278	20.81				
	1779416	22.82				
	1790157	22.96				
	1664938	21.30				

Tramadol Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
5	356069	5.93	118.54
10	722525	13.05	130.46
25	1267434	23.63	94.53

Conc	Peak Area
0.5	40328
1	67793
2.5	113125
5	356069
10	722525
25	1267434
50	2628025



Note:
Peak for 0.25 µg/ml concentration was not found after 7 days

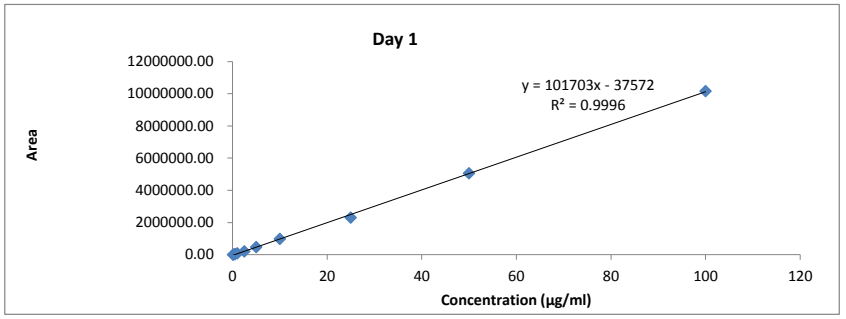
Zolpidem Validation HPLC Conditions

Chromatographic conditions: Zolpidem tartrate

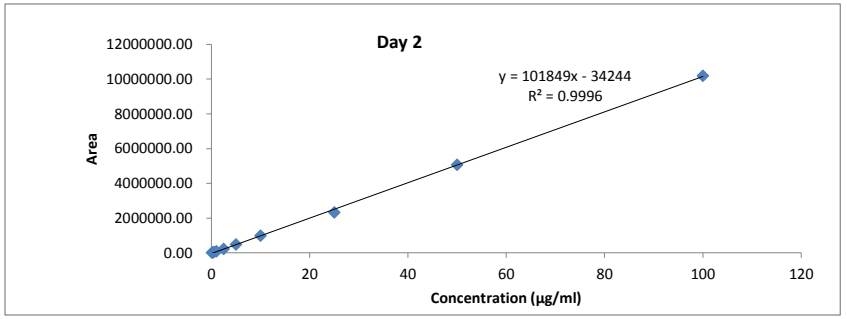
System: Waters e2795
Detector: Waters 2988 Photodiode Array
Pump Mode: Isocratic
Mobile Phase: ACN:Buffer (KH2PO4 25mM pH=6) (40:60)
Flow rate: 1ml/min
Injection Volume: 20µL
Run time: 10min
Retention time: 5.1min
Column: Agilent ZORBAX Eclipse Plus C 18 (4.6×150mm 5µm)
Guard Column: Agilent ZORBAX Eclipse Plus C 18 (4.6×12.5mm 5µm)
Column Temperature: 25°C
Detection wavelength: 243 nm
Sample Preparation: Standards were prepared in water

Zolpidem Validation Std Curve

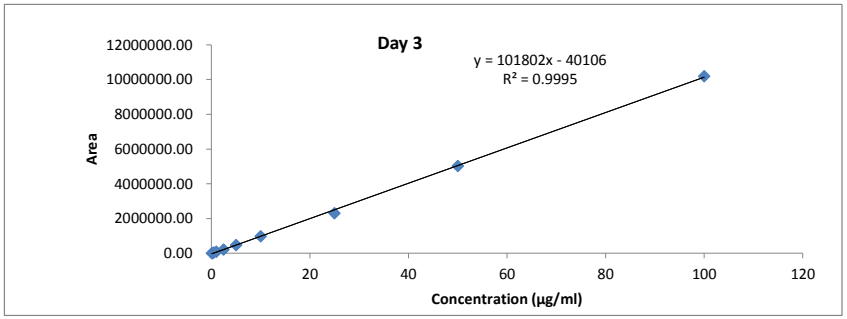
Day 1 (07/11/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	8712	8654	8682	8682.67	29.01	0.33
0.25	20050	19886	19577	19837.67	240.18	1.21
0.5	45281	41554	45449	44094.67	2201.88	4.99
1	93007	92815	93368	93063.33	280.77	0.30
2.5	213763	215856	216424	215347.67	1401.44	0.65
5	474805	475606	476595	475668.67	896.64	0.19
10	986262	979104	983644	983003.33	3621.75	0.37
25	2313038	2314984	2310040	2312687.33	2490.58	0.11
50	5062548	5068343	5075633	5068841.33	6556.72	0.13
100	10156967	10193645	10156741	10169117.67	21241.59	0.21



Day 2 (07/12/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	8459	8682	8993	8711.33	268.21	3.08
0.25	20023	20157	20279	20153.00	128.05	0.64
0.5	46253	46208	45763	46074.67	270.85	0.59
1	94006	93994	93592	93864.00	235.64	0.25
2.5	220724	220304	221217	220748.33	456.99	0.21
5	480789	486677	480058	482508.00	3628.91	0.75
10	996879	991499	997707	995361.67	3370.69	0.34
25	2335157	2334796	2308357	2326103.33	15369.84	0.66
50	5095762	5041086	5068046	5068298.00	27338.87	0.54
100	10189655	10211042	10169363	10190020.00	20841.90	0.20



Day 3 (07/13/16)						
Concentration (µg/ml)	Inj.1	Inj.2	Inj.3	Avg Area	SD	RSD
0.1	8792	8549	8486	8609.00	161.58	1.88
0.25	20506	20266	20193	20321.67	163.76	0.81
0.5	45391	45018	45412	45273.67	221.66	0.49
1	93038	93238	93027	93101.00	118.77	0.13
2.5	218023	219447	219120	218863.33	745.89	0.34
5	479035	476687	477395	477705.67	1204.43	0.25
10	975056	996302	979459	983605.67	11213.57	1.14
25	2281625	2294597	2349382	2308534.67	35964.52	1.56
50	4990194	5067213	5036695	5031367.33	38784.91	0.77
100	10103109	10232011	10255216	10196778.67	81945.85	0.80



Zolpidem Validation LOD & LOQ

LOD and LOQ	Slope	Y-intercept
STD day 1	101703	37572
STD day 2	101849	34244
STD day 3	101802	40106
Aver.	101784.67	
SD		2939.95
LOD=3.3SD/Aver		**0.10**
LOQ=10SD/Aver		**0.29**

Zolpidem Validation Interday Accuracy & Precision

Concentration (µg/mL)	Time	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	Day 1	986262	10.07	10.08	0.07	100.78	0.66
		979104	10.00				
		983644	10.04				
		985664	10.06				
		997270	10.18				
		989136	10.10				
	Day 2	996879	10.12				
		991499	10.07				
		997707	10.13				
	Day 3	975056	9.97				
		996302	10.18				
		979459	10.02				
25	Day 1	2297454	22.96	23.12	0.18	92.46	0.77
		2313038	23.11				
		2314984	23.13				
		2310040	23.08				
		2323766	23.22				
		2316214	23.14				
	Day 2	2335157	23.26				
		2334796	23.26				
		2308357	23.00				
	Day 3	2281625	22.81				
		2294597	22.93				
		2349382	23.47				
50	Day 1	4993055	49.46	49.98	0.30	99.95	0.60
		5047116	50.00				
		5062548	50.15				
		5068343	50.20				
		5075633	50.28				
		5036882	49.89				
	Day 2	5095762	50.37				
		5041086	49.83				
		5068046	50.10				
	Day 3	4990194	49.41				
		5067213	50.17				
		5036695	49.87				

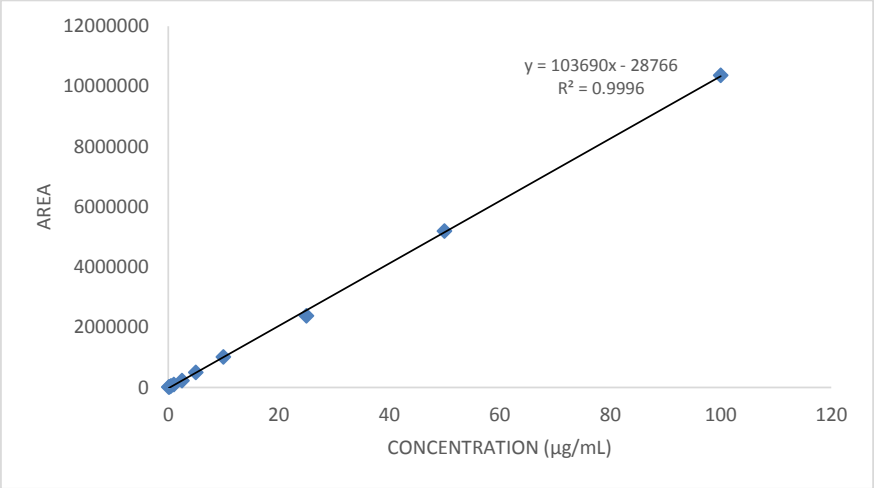
Zolpidem Validation Intraday Accuracy & Precision

Concentration (µg/mL)	Peak Area	Measured concentration (µg/mL)	Average Measured conc. (µg/mL)	SD	Accuracy (%)	Precision (%)
10	986262	10.07	10.07	0.06	100.73	0.60
	979104	10.00				
	983644	10.04				
	985664	10.06				
	997270	10.18				
	989136	10.10				
25	2297454	22.96	23.11	0.09	92.43	0.37
	2313038	23.11				
	2314984	23.13				
	2310040	23.08				
	2323766	23.22				
	2316214	23.14				
50	4993055	49.46	50.00	0.30	99.99	0.59
	5047116	50.00				
	5062548	50.15				
	5068343	50.20				
	5075633	50.28				
	5036882	49.89				

Zolpidem Validation Stability

Intial conc. (µg/mL)	Storage at 4°C after 7 days		
	Peak Area	Determined mean (µg/mL)	Mean % accuracy
10	1013853	10.06	100.55
50	5193442	50.36	100.73
100	10364538	100.23	100.23

Conc	Peak Area
0.1	9302
0.25	21593
0.5	47379
1	97265
2.5	231409
5	501498
10	1013853
25	2384125
50	5193442
100	10364538



Appendix C
Verde Data Summary (UV/ViS)

*Data is available on Request, and is not included within the report.

Verde Deactivation/Desorption Results

	Deactivation, Day									Desorption	
	0	0.33	1	2	4	7	14	21	28	Aqueous	EtOH
Alprazalom	0	76.4	91.5	98.2	100	100	100	100	100	0.0	0.0
Buprenorphine	0	97.8	99.8	100	100	100	100	NP	NP	0.3	NP
Dextroamphetamine	0	93.2	95.7	95.9	99.3	99.3	99.3	99.5	98.8	0.6	6.2
Diazepam	0	60.3	70.2	77.3	79.4	81.2	98.4	NP	NP	0.0	NP
Fentanyl	0	34.1	70.2	89.7	98.0	99.4	99.8	100	100	0.0	0.0
Fluoxetine	0	57.6	63.3	73.9	80.5	86.3	93.5	NP	NP	0.0	NP
Hydromorphone	0	99.4	97.3	99.0	100	100	96.5	99.9	100	0.1	2.7
Lorazepam	0	59.4	82.8	86.8	94.6	96.7	99.5	99.9	99.6	0.0	0.9
Loxapine	0	95.4	97.7	98.7	99.5	99.3	99.5	99.6	99.8	0.1	0.0
Meperidine	0	95.6	99.2	99.4	99.9	100	99.9	99.1	99.7	0.3	10.8
Methadone	0	100	100	100	100	100	100	100	100	0.0	0.0
Methylphenidate	0	99.6	99.6	100	100	100	100	NP	NP	0.0	NP
Morphine	0	99.4	99.8	100	100	100	100	NP	NP	0.0	NP
Quetiapine	0	43.7	57.1	59.6	76.6	88.5	86.9	98.5	98.2	0.0	1.0
Temazepam	0	80.1	84.2	87.5	96.8	96.0	100	100	99.5	0.0	0.0
Tramadol	0	91.6	98.1	99.7	99.9	100	99.5	96.7	98.4	0.7	9.1
Zolpidem	0	72.0	89.6	96.5	98.0	99.8	99.9	100	99.4	0.1	0.0
Average	0.0	79.7	88.0	91.9	95.4	96.8	98.4	99.4	99.4	0.1	2.6
SD	0	21.6	14.3	11.7	8.1	5.8	3.4	1.0	0.6	0.2	3.9

NP = Not Performed as part of Phase 1
All deactivation values expressed as % deactivated
Washout values expressed as % recovered