Table of Contents

Note: Hyperlink and attachment maintenance (removal, addition, or correction of hyperlinks and attachments) shall not constitute a new document revision.

DR-01 Cannabis Analysis

Purpose:

Samples of a vegetative nature, green leafy substance, green leafy plants, ashes or any other submittal which is suspected to be marijuana, hashish, hashish oil, THC, or THCA-A will be subjected to the examinations set forth in this protocol. The materials shall be analyzed using a series of tests: stereomicroscopic examination, Fourier transform infrared spectroscopy (FTIR), thin layer chromatography (TLC), gas chromatograph/flame ionization detector (GC/FID) and gas chromatography/mass spectrometry (GC/MS). The identification of marijuana and hashish require a positive microscopic examination, TLC, GC/FID and GC/MS examination and a CBD:THC ratio less than 10.00.

Associated Protocol(s):

DR-01 Attachment 1 1% Threshold Testing for THC
DR-01 Attachment 2 Flowcharts
DR-30 Gas Chromatography Analysis (Flame Ionization Detector)
DR-60 Drug Analysis by FTIR
DR-70 Gas Chromatograph / Mass Spectrometer Methods for Drug Analysis

Specimen(s) Required (Sample handling and preservation):

The sample size used for analysis may vary from residue to approximately 0.5 grams depending on the size of the submitted exhibit. The portion removed for analysis should be representative of the entire exhibit. Samples can be found in different matrices including plant material, oils, waxes, gummies, chocolate, etc.

Reagents:

1. Hexanes
2. Methanol
3. Chloroform
4. Ethyl Ether
5. Petroleum Ether
6. BSTFA
7. Sodium Hydroxide
8. Deionized Water
9. Acetone
10. Fast Blue BB Salt

Standards:

1. delta 9 - Tetrahydrocannabinol (THC)
2. Cannabidiol (CBD)
3. delta 8 - Tetrahydrocannabinol
4. Cannabinol (CBN)
5. Tetrahydrocannabinolic Acid A (THCA-A)

Apparatus and Materials:

1. Stereomicroscope
2. Gas Chromatograph/Flame Ionization Detector
3. Gas Chromatograph/Mass Spectrometer
4. Thin Layer Chromatography
5. Fourier transform infrared spectroscopy

Individual Steps of Protocol:

Green Leafy Substances:

1. Microscopic Examination (Visual)
   Note: Photographs of the marijuana plant, leaves, seeds, reproductive structures and cystolithic hairs can be found in "Marijuana, Its Identification" (Reference #4).
   a. The sample is viewed at any magnification 10X or above which will allow for a good visual inspection of the sample.
   b. The sample can be observed for morphological characteristics of the marijuana plant, such as leaf structure, leaf venation, covering hairs, cystolithic hairs, glandular hairs, and seeds.
   c. The minimum requirement for a positive microscopic examination is the observation of cystolithic hairs. Note that other plants also have cystolithic hairs.
   d. Record results of visual exam in the Matrix. *A negative visual exam does not need to go in the rejection panel.*

2. Thin Layer Chromatography (TLC)
   a. Extract sample with a minimal amount of petroleum ether or methanol.
   b. Spot a small portion of the extract at the origin of a TLC plate with a capillary tube.
   c. Spot a THC standard at the origin on the same TLC plate next to or near the question sample for purposes of comparison.
   d. Spot an extraction blank at the origin on the same TLC plate.
   e. Develop the plate using 4:1 Hexanes:Ethyl Ether or 3:1 Hexanes:Acetone in a developing chamber.
   f. Allow the plate to dry and spray it with fast blue BB salt in water solution.
   g. A positive test is indicated by the sample developing as the same color and traveling equal distances as the standard.
   h. Record the results in the matrix. The results of the sample, controls, and number of TLC plates used must be recorded. *A record of a negative sample is not a rejection of a test result.*
   i. Record the date the TLC plate was run in the "Date of Analysis" field in the matrix. If plates are run over multiple days document the items and dates in the "Notes" section.

3. Instrumental analysis will be performed following DR-30 and DR-70.
   Note: Instrumental analysis and TLC can be performed in any order.

4. Special Considerations:
   a. Pipes/Smoking Devices
      If the pipe/smoking device potentially has a weighable amount of a green and/or burnt leafy substance, the analyst should scrape the contents into a weigh boat for weighing. Any weighable amount should also receive a visual exam for possible cystolithic hairs and be analyzed using TLC.

      Pipes/smoking devices which do not contain a potentially weighable amount of substance may be rinsed or swabbed for analysis on TLC, GC, and GC/MS. Visual exam is not required.

Suspected Cannabis Edible Products

1. Put a small portion of the sample in test tube or sample vial.
2. Add approximately 0.5 mL BSTFA.
3. Allow the samples to derivatize in BSTFA overnight.
4. Instrumental analysis will be performed following DR-30 and DR-70.
5. Derivatized THC and/or THCA-A standards will need to be ran on the GC for comparison.

Other Suspected Cannabis Products

1. Analyze the sample on the FTIR following DR-60.
   a. If the sample matches the delta 9 THC standard, then analyze the sample on the GC or GC and GC/MS to confirm.
   b. If the FTIR indicates a mixture, then the sample will need to be derivatized with BSTFA and analyzed on the GC and GC/MS to confirm.
   c. If the sample matches the THCA-A standard then the sample will need to be derivatized with BSTFA and analyzed on the GC and GC/MS.
2. Derivatize the sample in approximately 0.5 ml BSTFA and analyze on the GC and GC/MS.
   a. Allow the samples to derivatize in BSTFA overnight.
   b. Derivatized THC and/or THCA-A standards will need to be ran on the GC for comparison.

Protocol Notes:

1. Analysts must be aware of the possibility that a green leafy substance sample or any plant material may contain controlled substances other than marijuana. Indicators that a controlled substance may be present other than marijuana include:
   a. Perfume
   b. Foil wrapping
   c. Very small amounts of vegetable material in plastic packets
   d. Chemical odors
   e. Unusual colors, i.e., bright green, dark brown
   f. Information provided by the officer
   g. Pieces of white substance or powders mixed with the vegetable material.
2. Unless otherwise noted in the matrix, the date of the visual exam will be the date the analyst assigns the case to themselves in the BEAST.

If testing suspected marijuana samples occur on multiple dates: record the dates, item numbers, and visual exam in the "Notes" section. *This multiple date information only needs to be recorded one time in a case, i.e. in the Notes of the first item tested.*

Example

Notes:
07-24-17, Items 1A-1H, Visual Exam
07-26-17, Items 2A-2D, Visual Exam

3. When the results of a plant material are Cannabis (Marijuana containing Tetrahydrocannabinol), Schedule I or Tetrahydrocannabinols, Schedule III the following statement needs to be added to the report: "Quantitation was not performed."

4. When a sample consists entirely of seed(s), their identity alone is not sufficient to bring them within the purview of the law, which requires them to be fertile. If a case requires seeds to be grown for cultivation charges see the Technical Manager for guidance and a deviation.

5. delta 8-Tetrahydrocannabinol does not derivatize in BSTFA.

6. "CBD\THC Peak Ratio" can be found on the delta 9-Tetrahydrocannabinol PDF.

Recommended Report Wording / Interpretation of Test Data:

Interpretation	Report as	Additional Information for Analysts
Positive for Marijuana	Cannabis (Marijuana containing Tetrahydrocannabinol), Schedule I	- Cystolithic hairs observed - TLC positive for delta 9-THC - GC and GC/MS positive for delta 9-THC - CBD:THC Ratio is less than 10.00
DR-01 Attachment 1		- Cystolithic hairs observed - GC and GC/MS positive for delta 9-THC - CBD:THC Ratio is greater than or equal to 10.00
Positive for a THC Isomer	Specific THC Isomer ex. Cannabidiol. or Tetrahydrocannabinols, Schedule III	- Microscopic exam negative or is not performed. - If GC and GC/MS is positive for delta 9-THC or delta 8-THC then report THC, Schedule III. - If GC and GC/MS is positive for a specific THC isomer and delta 9-THC or delta 8-THC are not present then report that specific isomer.
THC	Tetrahydrocannabinols, Schedule III	- FTIR and GC and GC/MS and/or derivatization with BSTFA confirm delta 9-THC
THCA-A	Tetrahydrocannabinolic Acid.	- FTIR and GC and GC/MS and/or derivatization with BSTFA confirmed THCA-A
THC and THCA-A	Tetrahydrocannabinols, Schedule III and Tetrahydrocannabinolic Acid.	- Derivatization with BSTFA confirmed THC and THCA-A

Not enough sample for DR-01 Attachment 1	Test results are inconclusive for the presence of marijuana. There was insufficient sample for further testing.	- Cystolithic hairs observed - TLC positive for delta 9-THC - GC and GC/MS positive for delta 9-THC - CBD:THC Ratio is greater than or equal to 10.00 - There is less than 0.10 grams of GLS for testing
Indication	Examinations indicate the presence of a controlled substance however the laboratory's reporting criteria have not been met.	There is an indication of a CDS but it does not meet all the requirements to confirm.
Negative	No Controlled Dangerous Substances Identified.	Perform additional examinations prior to reporting if other controlled substance may be present.

References:

1. Oklahoma Statutes - Section 2 - 101 of Title 63.
2. Nakamura GR. Forensic aspects of cystolith hairs of cannabis and other plants. Journal of the Association of Official Analytical Chemists, Vol. 52, No. 1, 1969; 5-16.
3. Mechoulam R. Marihuana chemistry. Science, Vol. 168, No. 3936, June 5, 1970.
4. Marihuana, Its Identification. United States Government Printing Office: 1948.
5. Methods of Analysis. Internal Revenue Service Publication No. 341, Rev: 6-67.
6. Hughes RB, Warner VJ. Microgram, Vol. IX, No. 7; 94-101, July 1976.
7. Lowry WT, Garriott JC. 1975, J. Forensic Science, 20, 624.
8. Basic Training Program for Forensic Drug Chemists, Marihuana and THC, Lesson Plan #7, May 1972; 146-61.
9. Bailey FL, Rothblatt HB. Handling narcotic and drug cases. The Lawyers Co-operative Publishing Co., 1972; 235-39.
10. Analysis of Drugs. DEA Analytical Manual, U.S. Department of Justice, 165-68.
11. Coutts, Jones. A comparative analysis of cannabis material. Journal of Forensic Sciences, Vol. 24, No. 2, April 1979; 291-302.
12. Lowry, Garriott. On the legality of cannabis: The responsibility of the expert witness. Journal of Forensic Sciences, Vol. 20, No. 4, Oct 1975; 624-29.
13. Small E. The forensic taxonomic debate on cannabis: semantic hokum. Journal of Forensic Sciences, Vol. 21, No. 2, April 1976; 239-51.
14. Small E. American law and the species problem in cannabis. Microgram, November, 1974; 131-32.
15. Nakamura GR, Thornton JI. The Forensic Identification of Marihuana: some questions and answers. Journal of Police Science and Administration, Vol. I, No. I, 1973; 102-12.
16. Hughes RB, Warner VJ, Jr. A study of false positives in the chemical identification of marihuana, Journal of Forensic Sciences, Vol. 23, No. 2, April 1978; 304-10.
17. Michigan State Police Forensic Science Division. Controlled Substances Procedure Manual 6 Marihuana, 2022, Revision # 11.

DR-01 Attachment 1 – 1% Threshold Testing for Total THC

Purpose:

The 2018 Farm Bill was written into law to allow for industrial hemp to be defined separately from marijuana. This procedure will be used to determine if a green leafy substance is above or below a 1% threshold of total tetrahydrocannabinol by comparing the results of the green leafy substance to a known 0.1 mg/mL delta 9-Tetrahydrocannabinol (THC) with internal standard (ISTD) positive control. This procedure is not a quantitation, as it does not determine the exact concentration of THC in a sample.

Associated Protocol(s):

DR-30 Gas Chromatography Analysis (Flame Ionization Detector)
DR-70 Gas Chromatograph / Mass Spectrometer Methods for Drug Analysis

Specimen(s) Required (Sample handling and preservation):

This protocol is used for plant materials. It does not apply to liquids, oils, edibles, or other manufactured products.
A minimum of 0.10 grams of green leafy substance (not including seeds or stems) is needed to perform this analysis. The sample size used for analysis is approximately 0.05 grams of a green leafy substance that has a "CBD:THC Peak Ratio" greater than or equal to 10.00.

Reagents:

1. Methanol
2. Chloroform

Standards:

1. delta 9-Tetrahydrocannabinol (THC)
2. 4-Androsten-3,17-dione (AND)

Controls:

1. Negative Control: ISTD 4-Androsten-3,17-dione
2. Positive Control: 0.1 mg/mL solution of THC in ISTD
    a. The acceptance criteria for the positive control is THC:AND ratio > 1.
    b. The controls must be stored in the refrigerator. They should be used within 8 weeks from when they were made or until the response (THC:AND ratio) does not meet the acceptance criteria (whichever comes first).
3. Solvent Blank: Methanol:Chloroform (9:1 ratio)

Frequency and Tolerance of Controls and Corrective Action for Exceeded Tolerances:

1. If the positive control fails on the GC/MS:
    a. Run again to verify.
    b. Remake the stock solution and positive control or use a different approved GC/MS.
    c. If any samples were run after the failed positive control they will need to be rejected and rerun after a passing positive control.

Apparatus and Materials:

1. Gas Chromatograph/Flame Ionization Detector
2. Gas Chromatograph/Mass Spectrometer
3. Pipettes
4. Analytical Balance

Preparation of Reagents:

0.05 mg/mL Internal Standard Solution

1. Combine 450 mL of Methanol and 50 mL of Chloroform.
2. Weigh out 0.0250 g of 4-Androsten-3,17-dione, using the analytical balance, and dissolve in the Methanol:Chloroform solution.
   Note: Total volume may be adjusted as necessary but the final concentration must remain the same.

Stock Solution and Positive Control

1. To make the stock solution add 0.04 mL THC Standard (50 mg/mL) to 1.96 mL of ISTD using calibrated pipettes.
2. To make the positive control add 1.00 mL of the THC stock solution to 10.00 mL of ISTD using calibrated pipettes.
   Note: A 50 mg/mL liquid THC standard must be used. If another concentration of the standard is used then the concentration of ISTD in the positive control and the sample will need to be recalculated. Inform the Technical Manager if a THC standard with a different concentration will need to be used.

Individual Steps of Protocol:

1. Crumble and weigh 0.0500 grams +/- 0.0100 grams of the green leafy substance on the analytical balance. Transfer to test tube.
2. Add 5.00 mL of ISTD to the test tube using a calibrated pipette. Extract for approximately 10 minutes, vortexing for 10-15 seconds at least twice during the extraction.
3. Filter sample and then transfer to an autosampler vial.
4. Prepare a solvent blank the same way the sample was prepared.
5. Analyze the sample on the GC using the Drug1 method per DR-30.
6. Analyze on the GC/MS in the following order (per DR-70):
   a. Analyze the positive control using the Drug10 or Drug25 method prior to each sequence.
   b. Analyze a solvent blank using the Drug10 or Drug25 method for each case and at least every sixth injection.
   c. Analyze the negative control using the Drug10 or Drug25 method immediately before each sample to verify there is no carry over.
   d. Analyze the sample using the using Drug10 or Drug25 method.

Protocol Notes:

1. For a mass spectrometer to be used for this method a precision test and the positive control must be run. The steps to the precision test can be found in the validation plan at the following location: \\pm-fsc16482s\QA\Lab-System_Records\Management_System_Docs\Chemistry - Drugs\Validations\2024\1%_Comparison. Consult with the Technical Manager before starting the precision test.
2. The ratios of "THC/AND Peak Ratio" can be found on the 4-Androsten-3,17-dione pdf.
3. All reports where the 1% threshold testing was performed will have the following statement: "Quantitation was not performed."
4. The case file will need to include the positive control, negative control(s), solvent blank, and the sample data.

Recommended Report Wording / Interpretation of Test Data:

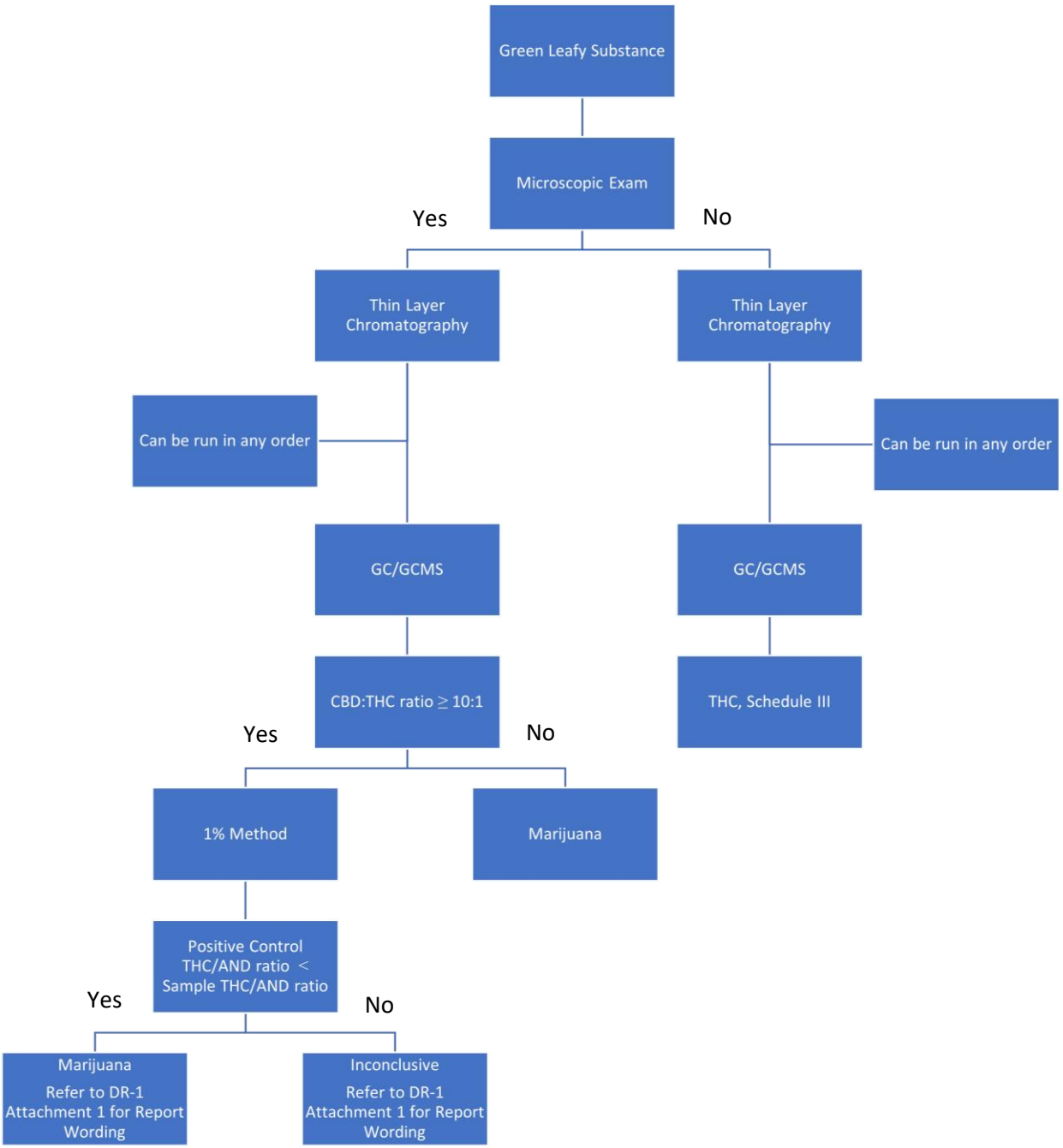
Results Obtained	Interpretation
- Cystolithic hairs observed - TLC positive for delta 9-THC - Positive Control THC/AND ratio is less than the Sample THC/AND ratio	Positive for Marijuana
- Positive Control THC/AND ratio is greater than or equal to the Sample THC/AND ratio	Inconclusive for Marijuana

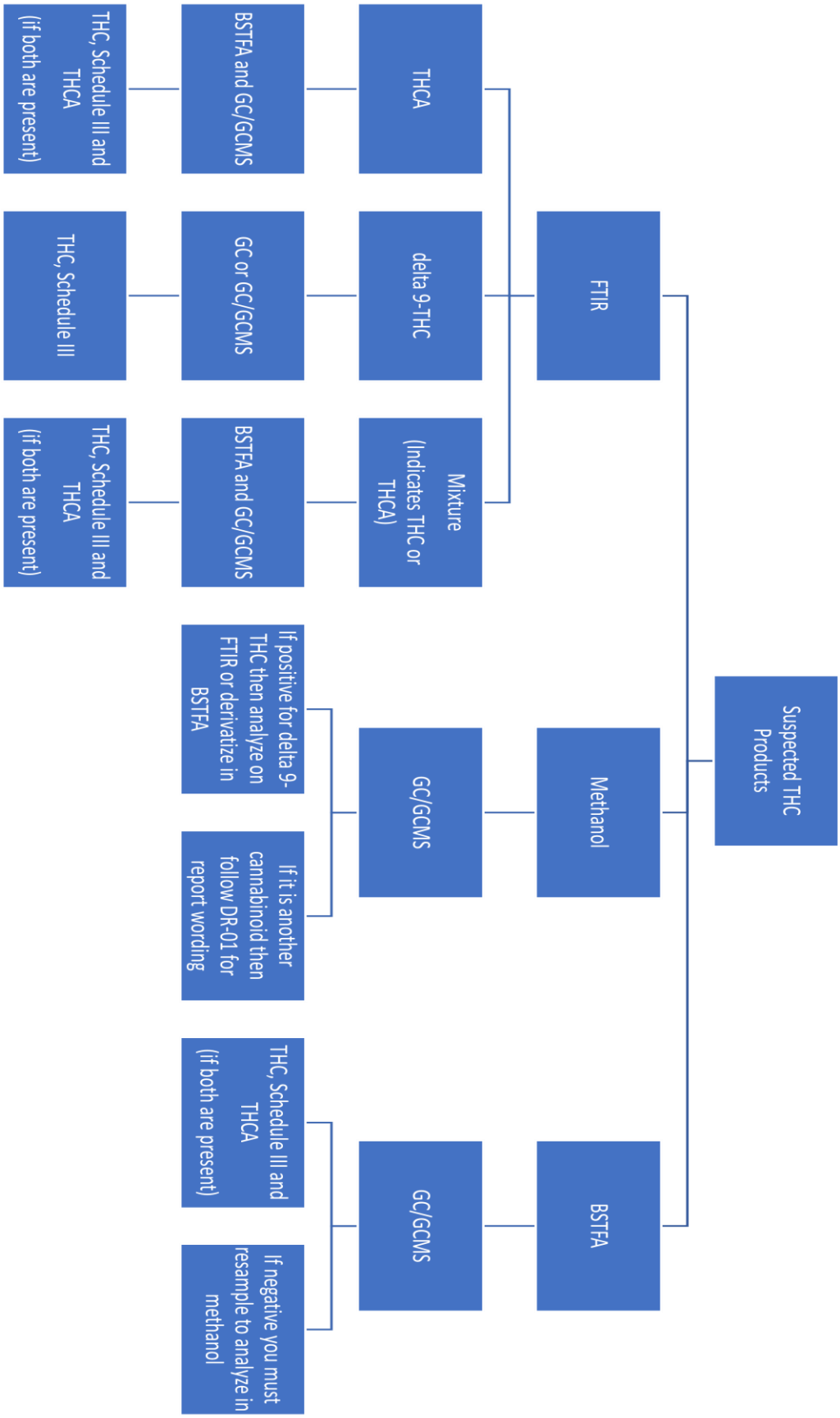
Interpretation	Report as	Additional Information for Analysts
Positive for Marijuana	Cannabis (Marijuana containing Tetrahydrocannabinol), Schedule I.	Marijuana is identified based upon an evaluation of the amount of delta 9-THC and THCA-A (measured as total THC) and comparison with a reporting limit of 1% Total THC.
Inconclusive (delta 9 THC ≤ 1%)	Test results are inconclusive as to the presence of marijuana. The concentration of delta 9-THC did not meet the laboratory's reporting limit.	Do not report out Tetrahydrocannabinols, Schedule III or Cannabidiol if the results are inconclusive.

References:

1. Oklahoma Statutes - Section 2 - 101 of Title 63.
2. Michigan State Police Forensic Science Division. Controlled Substances Procedure Manual 6 Marihuana, 2022, Revision # 11.
3. Tennessee Bureau of Investigation. Standard Operating Procedures, 2019, Version #3.0.
4. Kohl, A. (2019). Method Validation Report for the Identification and Quantitative Threshold Testing of Tetrahydrocannabinol (THC) in Cannabis.

DR-01 Attachment 2 Flowcharts




Suspected THC Products
Methanol
FTIR
delta 9-THC
GC or GC/GCMS
THC, Schedule III
Mixture (Indicates THC or THCA)
BSTFA and GC/GCMS
THC, Schedule III and THCA (if both are present)
THCA
BSTFA and GC/GCMS
THC, Schedule III and THCA (if both are present)
GC/GCMS
If positive for delta 9-THC then analyze on FTIR or derivatize in BSTFA
If it is another cannabinoid then follow DR-01 for report wording
BSTFA
GC/GCMS
THC, Schedule III and THCA (if both are present)
If negative you must resample to analyze in methanol

DR-02 Collection of a Portion of Evidence from Bundles Prior to Submission

Purpose:

This protocol outlines the proper procedures for collecting, handling and reporting of samples from bundles that are brought in by law enforcement agencies. The bundles will typically contain a suspected green leafy substance. If other forms are encountered, the items need to be submitted for analysis without taking samples.

Per Oklahoma State Statute §63-2-204.C any material, compound, mixture, or preparation which contains any quantity of marijuana, the whole sample shall be considered as marijuana. As a result of this allowance by state law, this protocol should not be considered a sampling plan.

Associated Protocols:

DR-01 Cannabis Analysis
DR-01 Attachment 1 – 1% Threshold Testing for Total THC
DR-01 Attachment 2 – Flowcharts
DR-03 Weights and Measures Utilized in Drug and Marijuana Reports
DR-04 Balance/Scale Calibration and Uncertainty

Specimen(s) Required (Sample handling and preservation):

Evidence such as bundles or packages containing a suspected green leafy substance. The sample taken from each bundle must be at least 0.20 grams.

Apparatus and Materials:

1. Scalpel (disposable or with replaceable blades) or Razor blades
2. Permanent marking pens
3. Plastic bags
4. 2” Tape
5. Labels
6. Brown/white rolled butcher paper
7. Tub for large capacity scale
8. Large capacity scale
9. Evidence envelope

Individual Steps of the Protocol:

1. The evidence/bundles that require sampling will be taken to the lab where weighing and sampling will occur. The evidence is to remain in the custody of the submitting officer and the officer must remain with the evidence at all times.

2. Remove and weigh a bundle. Document the weight type, weight of the bundle, any officer numbering, a packaging description and an item number; this is the item number that will be used in the BEAST.
    a. Documentation can be made on paper or in Excel. After the case has been assigned, the document will need to be uploaded into the OSBI lab casefile.

b. Multiple bundles may have the same packaging description and can be documented once in the notes made on paper or in Excel.
   Example: Items 1A through 1F are tape wrapped bundles.

3. Label the bundle with the OSBI case number, along with the item number, date and the analyst initials.

4. Open the packaging and remove a sample of the evidence and place it in a plastic bag. The amount of sample taken should be enough for OSBI instrumental analysis, while leaving enough for further testing if needed/requested.

5. Label each plastic bag with the following:
   a. laboratory case number
   b. item number
   c. officer numbering (if present)
   d. date
   e. analyst initials
   f. weight of bundle (noting if gross or net weight)

6. Place 2" tape over any markings made and the opening where the sample was removed. Initial the tape covering the opening.

7. Before moving on to the next bundle, ensure the bundle and plastic bag are properly marked and the bundle information, including item number, officer numbering, description and weight has been documented.

8. Check the scale with weights that bracket the weights of the bundles and document the weights. These weights will need to be added to the case information when analysis is started.

Protocol Notes:

1. During sample collection, only one bundle will be open at a time.

2. The statement "The results apply only to the sample(s) received" must be removed from the report of the cases being sampled prior to submission.

3. The analyst collecting the samples will have the case assigned to themselves for analysis.

4. After all bundles have been weighed and sampled (if necessary), the analyst should verify the number of bundles, plastic bags containing samples and documentation notes all agree in number. Example: 36 bundle, 36 plastic bags with green leafy samples and documentation for 36 samples.
   a. There may be cases where there are more bundles weighed than sampled and this is acceptable. The total weight should be documented. Example: 50 bundles, 36 weighed and sampled, the remaining 14 are weighed as one gross weight; documentation should reflect 37 items weighed.

b. Any bundles weighed but not sampled will be included in the narrative on the OSBI Criminalistics Examination Report. These bundles do not receive an item number, as they are not being sampled or submitted.

5. The weight reported will be the weight of the bundle, either net or gross weight. The plastic bag containing the sample does not have to be weighed.
6. If others assist in the collection of samples, notes of who assisted and what they did will be documented in the case notes.

7. A case number will be provided before sampling the case so the evidence can be labeled appropriately while collecting the samples.

8. A sample may be taken for analysis at the same time a sample is collected to be retained for further analysis.

Recommended Report Wording:

1. A Case Narrative will be included in the OSBI Criminalistics Examination Report (above the Description of Evidence) reflecting what OSBI analysts did. The Case Narrative will be typed into the matrix Narrative Panel and generated on the report.
   The following are examples of the narratives that need to be included on the report:

   On April 3rd, 2022, at OSBI FSC, [Criminalist's Name], OSBI, weighed and collected samples from bundles for OHP. The bundles were in the custody of OHP and [Officer's Name] was present during the sample collection. Plastic bags containing samples from each bundle were given to [Officer's Name] for submission to the OSBI.

   or

   On April 3rd, 2022, at OSBI NERL, [Criminalist's Name], OSBI, weighed and collected samples from 36 bundles for OHP. The bundles were in the custody of OHP and [Officer's Name] was present during the sample collection. Plastic bags containing samples from each bundle were given to [Officer's Name] for submission to OSBI. There were also 14 bundles that were only weighed; the total weight: of the 14 bundles was 36.43 pounds.

2. The description of the items will include the information for the bundle. Report using the following example:

   Item # One plastic bag containing a green leafy substance, sampled from one plastic-wrapped bundle "#4" containing a green leafy substance, gross weight: 14.40 pounds

3. The results will include the total weight recorded for the sampled bundles and will include the uncertainty of measurement; refer to DR-03 for reporting requirements and examples.

DR-03 Weights and Measures Utilized in Drug and Marijuana Reports

Purpose:

Establish uniform guidelines in the determination and reporting of weights and volumes of substances submitted for analysis.

Associated Protocol(s):

DR-04 Balance/Scale Calibration and Uncertainty

Specimen(s) Required (Sample handling and preservation):

Sample of sufficient mass to weigh or volume to measure.

Special Supplies:

1. Weigh container
2. Weigh paper/butcher paper
3. Plastic bags
4. Small plastic tub(s) for electronic scale
5. Large plastic tub(s) for large capacity scale

Apparatus and Materials:

1. Analytical balance(s)
2. Electronic balance(s)
3. Large capacity scale(s)

Individual Steps of Protocol:

Using weigh containers, weigh paper or appropriate plastic tubs, case samples will be weighed or measured using the following terminology and definitions.

1. **Net Weight** – Weight of the sample submitted for analysis, excluding packaging materials, limited by the ability to remove the entire sample from the packaging.

2. **Gross Weight** – Weight of the sample and the packaging medium. Routine use of gross weight is not encouraged, but can be utilized when packaging makes determining of net weight difficult or the sample is in powder form.

   Unless otherwise noted, a gross weight contains everything included in the description for the item.

   Examples:

   1A One plastic bag containing <u>one plastic bag containing a crystal-like substance</u>, gross weight: 2.34 grams (Note: gw inner most bag and contents only)

   1A <u>One plastic bag containing one plastic bag containing a crystal-like substance</u>, gross weight: 2.34 grams (No note is needed)

One plastic bag containing:

1A One plastic bag containing a crystal-like substance, gross weight 2.34 grams (No note is needed)

3. **Approximate Volume** – An estimation of the quantity of a liquid based upon calculation ($V=\pi r2h$), labeling, or size of the container.

4. **Residue** – A small quantity of substance relative to the size of the packaging. Sample manipulation and acquiring an accurate net weight is difficult or not possible due to a sample size or physical characteristics.

Recommended Report Wording / Interpretation of Test Data:

1. When determined on an electronic balance, weights in grams will be recorded and reported as follows:

0.00 to 0.09 gram	Report as "less than 0.10 gram." Record in BEAST matrix the actual weight including all digits to the right of the decimal point. *Example:* net weight: less than 0.10 gram
0.10 gram to Scale Capacity	Record and report the two digits to the right of the decimal point. *Example:* 0.11 grams *Example:* 10.68 grams *Example:* 1454.54 grams

2. When determined on an analytical balance, weights in grams will be recorded and reported as follows:

0.0000 to 0.0009 gram	Report as "less than 0.0010 gram." Record in BEAST matrix the actual weight including all digits to the right of the decimal point. *Example:* net weight: less than 0.0010 gram

0.0010 gram to Scale Capacity	Report to two significant digits to the right of the decimal point. The complete weight must be recorded in the UoM spreadsheet/PDF that is uploaded into the casefile. Document in the Matrix which analytical scale was used.

(The rules for significant figures can be found in DR-04 Attachment 1.)

Example: 0.0110 grams
Example: 10.0068 grams

3. When determined on a large capacity scale, weights in pounds will be recorded and reported as follows:

0.00 to 0.09 pound	Report as "less than 0.10 pound." Record actual weight in BEAST matrix, including all digits to the right of the decimal point. *Example:* net weight: less than 0.10 pound.
0.10 pound to Scale Capacity	Record and report the 2 digits to the right of the decimal point. *Example:* 6.60 pounds, 26.65 pounds

4. No values will be averaged or rounded up by the analyst.

5. Cases involving trafficking levels according to Title 63, O.S. 2-415, will be reported in the corresponding units when possible. In rare instances when it is not possible to report weights in the corresponding units, a note to the reason why should be included in the analyst notes for the item(s).

6. All approximate volumes will be reported in milliliters, ounces, liters, or gallons. Designations will be determined by the analyst based on sample packaging and the need for clarity in the OSBI Criminalistics Examination Report. In the case of containers of unknown size, such as containers at clandestine laboratories, the analyst may report approximate volumes relative to the container.

   Example: One, one gallon jar, approximately 3/4 full of a liquid

7. If safely obtainable, all liquids and solids will have the weight or approximate volume units reflected on the report, including items not analyzed. Items of similar description, which are not analyzed, can be grouped together and a gross weight reported. Weights of items not analyzed are not to be included in the calculations for uncertainty of measurement. "Empty" items and items with residues in/on containers, including smoking devices, do not require a reported weight.

   Example: Four plastic bags, each containing a crystal-like substance, gross weight: 4.23 grams

Example: One plastic bag containing six hand-rolled cigarettes, gross weight: 2.34 grams
Example: Five glass vials each containing approximately one milliliter of a clear liquid, approximate volume: 5 milliliters.
Example: Six empty plastic bags

8. Weights or approximate volumes will be listed in the Description of Evidence section of the report following the individual item description.

   Example: One plastic bag containing a green leafy substance, net weight: 17.24 grams

   Example: One plastic bottle containing a pink liquid, approximate volume: 12.00 milliliters

9. If the sample is a residue, the sample should be described as such.

   Example: One (1) plastic packet containing a residue

10. Total weights of similar samples may be listed in the Results and Interpretations section of the report, following the analysis results for those items.

    Example:

Item #	Analysis
1A through 1D	Methamphetamine, Schedule II, total net weight: 18.00 grams

11. For cases involving trafficking levels according to Title 63, O.S. 2-415:

    A. The total weights and expanded uncertainties of similar samples will be listed in the Results and Interpretations section of the report, following the analysis results for those items. For items weighed on the bench scales, items weighing less than 0.10 gram will not be included the calculation of trafficking levels and are to be reported separately. For items weighed on the large capacity scales, items weighing less than 0.10 pound will not be included in the calculation of trafficking levels and are to be reported separately.

    Examples:

Item #	Analysis
1A through 1J	Cocaine, Schedule II, total net weight: 28.42 grams +/- 0.60 gram
1K through 1O	Cannabis (Marijuana containing Tetrahydrocannabinol), Schedule I, total gross weight: 50.10 pounds +/- 1.14 pound
1P and 1Q	Cocaine, Schedule II
1R	Cannabis (Marijuana containing Tetrahydrocannabinol), Schedule I

B. If the analytical scale is used to weigh samples, use the rules for Significant Digits and Rounding, located in DR-04 Attachment 1 to report total weights and uncertainty to two significant digits to the right of the decimal.

Examples:

Item #	Analysis
1A through 1D	Cocaine, Schedule II, total net weight: 28.42 grams +/- 0.017 gram

C. If there are samples that are described as a residue or liquids that are reported out in milliliters, and not grams, those items need to be listed on a different line in the Results and Interpretations section.

D. If there are multiple samples that have been identified as containing the same compound, i.e., methamphetamine, the weights need to be combined and reported on one line in the Results and Interpretations section.

E. If there are sample(s) that contain a mixture, i.e., methamphetamine and marijuana, those items need to be on a separate line in the results section, with uncertainty for those measurements.

Examples:

One sealed bucket containing:
1A: One plastic bag containing a crystal-like substance, net weight: 5.00 grams
1B: One plastic bag containing a crystal-like residue
1C: One plastic bag containing a crystal-like substance, net weight: 5.00 grams
1D: One plastic bag containing a crystal-like substance, net weight: 5.00 grams
1E: One sample vial containing a liquid, approximate volume: 20.00 milliliters
1F: One plastic bag containing a white powder with a green leafy substance, net weight: 5.00 grams

Item#	Analysis
1A, 1C, 1D	Methamphetamine, Schedule II, total net weight: 15.00 grams +/- 0.24 gram
1B, 1E	Methamphetamine, Schedule II
1F	Methamphetamine, Schedule II and Cannabis (Marijuana containing Tetrahydrocannabinol), Schedule I, total net weight: 5.00 grams +/- 0.08 gram

13. When the expanded uncertainty is reported, the coverage probability for the expanded uncertainty will be added to the report directly under the results. Use the following wording:

"The expanded uncertainty for the above results was calculated using a coverage probability of 99.73%"

14. When a weight is obtained after sampling has occurred, the analyst will add the following wording in the Results and Interpretations section:

    "Reported weight for Item X was obtained after sampling."

15. When samples are reweighed after analysis and no further instrumental analysis is performed on the reweighed samples, a secondary supplemental report will be issued. In the report, underline the new weights in the Description of Evidence and use the following wording in the Results and Interpretations section:

    Items reweighed, but not analyzed will be listed in the results as

    "Reported weights were obtained after sampling and no further instrumental analysis was performed."

    Items not reweighed or analyzed will be listed in the results as

    "No further analysis."

References:

1. Latz, JL. Pharmaceutical Calculations, Second Edition. John Wiley and Sons, New York.
2. ASCLD/LAB. ASCLD/LAB Policy on Measurement Uncertainty. 2013, ASCLD/LAB Document Control Number: AL-PD-3060 Ver 1.1.

DR-04 Balance/Scale Calibration and Uncertainty

Purpose:

Establish uniform guidelines for assuring accuracy of weights reported in Criminalistics' Controlled Substances Examination Reports, and establish a procedure for documenting balance/scale verification, calibration, and uncertainty.

Associated Protocol(s):

DR-3 - Weights and Measures

Standards:

All standards will be stored and transported in cases specifically designed to hold the weight/weight set.

1. NIST Class F, ANSI/ASTM Class 6 or better weight set consisting of at least the following:
    a. 1- 50 mg (Analytical balance only)
    b. 1- 100 mg
    c. 1- 500 mg
    d. 1- 1 gram
    e. 1- 5 gram
    f. 1- 20 gram
    g. 1- 100 gram
    h. 1- 500 gram
    i. 1- 4 kilogram

2. NIST Class F, ANSI/ASTM Class 6 or better weights of the following units:
    a. 1 - 0.02 pound
    b. 1 - 0.10 pound
    c. 1 - 1.00 pound
    d. 1 - 5.00 pound
    e. 1 - 10.00 pound
    f. 1 - 25.00 pound

Equipment:

1. Analytical balance: Mettler Toledo XSR64 (0 to 61 grams)
2. Electronic balance: Mettler Toledo MS4002S (0 to 4200 grams)
3. Electronic balance: Mettler Toledo MS4002TS (0 to 4200 grams)
4. Large capacity scale: Ohaus T31P (0 to 150 pounds)

Forms:

Forms for each balance are in Excel Spreadsheet form and will be kept in the lab specific folder on the OSBI Server vm-fsc-files\ContSub\Balance. The forms (DR4-1, DR4-2 and DR4-3) in these folders will be updated for each scale in use by the assigned user of the balance or a designee.

The user's manual and calibration reports for each scale should be embedded in the Excel Spreadsheet for each scale. The originals can be found on the OSBI Server vm-fsc-files\ContSub\Balance.

Individual Steps of Protocol:

The following steps are performed monthly or if the balance/scale has been moved within the lab.

Analytical Balance & Electronic Balance

1. Using gloves, KimWipes or clean tweezers, place individual weight on clean, tared weigh pan (no weigh dish or paper).
2. Record results on OSBI Form DR4-1 (electronic balance) or DR4-3 (analytical balance). Record all digits displayed on scale.
3. Using gloves, KimWipes or clean tweezers return weight to storage box.
4. Repeat until all designated weights within the range of the balance have been checked and balance readings have been documented.
5. Check area immediately around balance to ensure it is clean, and record on OSBI Form DR4-1 (electronic balance) or DR4-3 (analytical balance).

Large Capacity Scale

1. Using gloves, place individual weight on clean, tared scale pan (no weigh dish or paper).
2. Record results on OSBI Form DR4-2. Record all digits displayed on scale.
3. Repeat steps 1 & 2 until all designated weights within the range of the scale have been checked and scale readings have been documented.
4. Check area immediately around balance to ensure it is clean, and record on OSBI Form DR4-2.

Frequency and Tolerance of Controls and Corrective Action to Be Taken If Tolerances Are Exceeded:

All balances (Mettler Toledo MS4002S, MS4002TS, and XSR64) and scales (Ohaus T31P) used to report measurements in laboratory casework will be calibrated annually by an outside vendor that maintains a quality system that meets or exceeds the requirements set forth in ISO/IEC 17025. This calibration will include the determination of uncertainty for each balance. The calibration documentation for each balance will be maintained in the appropriate folders at \\VM-FSC-FILES\ContSub\Balance.

Laboratory balances and scales will be verified monthly when in service; if a scale is in service, but not used for casework for 30 or more days the scale must be verified before being used for casework. All verifications and reasons for not performing a monthly verification (i.e. "no casework performed in April") must be documented on the appropriate OSBI DR4 form. When the balances and scales have been calibrated by an outside vendor, they must be verified and the verification documented on the appropriate OSBI DR4 form before casework can be resumed. The electronic balances and analytical balances will be verified using the weights listed in the Standards Section 1 of this protocol; while the large capacity scales will be verified using the weights listed in Section 2 of Standards.

All electronic benchtop balances, Mettler Toledo MS4002S and MS4002TS, have an internal calibration process (FACT) that is automatically executed when conditions are warranted. The MS4002S and MS4002TS benchtop balances will be verified daily before casework is performed, using a NIST traceable 500 gram weight and 1 gram weight; the serial number of both weights and the scale readout for the weight will be documented on OSBI Form DR4-1.

Balances that weigh within plus or minus 1% of all verification weights will be considered acceptable for service. In the event a balance fails to perform within the acceptable range, first the analyst will attempt to adjust the internal calibration. Press the function key and select "ADJ.INT" and the scale will perform an internal adjustment of the calibration using an internal weight. Check the balance again with the 500 gram and 1 gram weights. If the balance falls outside the 1% range a second time, remove the balance from service until it has been repaired and calibration checked by an outside vendor that is accredited to ISO/IEC 17025:2005 by an accrediting body that is a signatory to the ILAC Mutual Recognition Arrangement, with the calibration to be performed listed in the scope of accreditation.

Any balance that is not in service or removed from service will be labeled "Not in Service" and documented on the appropriate OSBI form DR4 as to the date the balance was removed from service and why, if applicable. When a balance is returned to service, reflect on the appropriate OSBI form DR4 the date the balance was returned to service and any repairs that were made. Perform a verification of the balance and document the verification; the electronic balances and analytical balances will be verified using the weights listed in the Standards Section 1 of this protocol; while the large capacity scales will be verified using the weights listed in Section 2 of Standards.

New analysts will participate in a Measurement Assurance Study before beginning casework. The study will be incorporated in OSBI Forms DR4-1, DR4-2 and DR4-3. The Technical Manager will monitor the study for any anomalies in balance or analyst's performance. If anomalies are identified, the Technical Manager or designee will ascertain the cause and determine if additional training is needed.

Reference OSBI DR4 Attachment 2, Controlled Substances Scale Scenarios, to determine when a scale might need calibration and/or a Measurement Assurance Study, outside the annual calibration. Calibration or re-calibration of any balance used to report measurements in laboratory casework will only be performed by an outside vendor that is accredited to ISO/IEC 17025:2005 by an accrediting body that is a signatory to the ILAC Mutual Recognition Arrangement, with the calibration to be performed listed in the scope of accreditation

Large capacity scales and analytical balances must be checked for accuracy whenever they are utilized for casework involving "Trafficking" charges. Calibration weights bracketing the sample weight(s) need to be utilized. Results will be documented in the Matrix for that case.

Example: Multiple marijuana samples individually weighing 2-3 pounds total 30.75 pounds. The balance accuracy will be checked using the 1 pound and 5 pound weights.

Uncertainty of Measurement:

Reference OSBI CSD QP22, Estimating Uncertainty of Measurement, to determine when it is necessary to report Uncertainty of Measurement.

Reference OSBI DR4 Attachment 1, OSBI Budget for Calculating Uncertainty of Measurement. This attachment will be reviewed annually at a minimum and updated with changes as needed.

The Uncertainty of Measurement will be reported as the expanded uncertainty with the coverage probability. The coverage probability will be added as a footnote in the report.

The units of measurement for uncertainty will be the same as the units of measurement for the compound being weighed. For example, weights measured on the bench scales will have the uncertainty reported in grams. Weights measured on the large capacity scales will be reported in pounds, as will the uncertainty. Weights reported from the bench scales will be reported separately from weights measured on the large capacity scales, and the uncertainties cannot be combined.

Example:

1A: One plastic bag containing a green leafy substance,
net weight: 4.13 grams +/- 0.09 gram

1B: One plastic wrapped bundle containing a green leafy substance,
net weight: 26.10 pounds +/- 0.05 pound

When the uncertainty is being reported for a single item, the calculated expanded uncertainty is the uncertainty for that measurement.

When multiple weights are added together for a total weight, the uncertainties for each weight are added to create a total expanded uncertainty. If the total expanded uncertainty exceeds two significant figures, the total reported weight and total expanded uncertainty shall be reported to the same level of significance.

Example:

The contents of four plastic bags are identified as methamphetamine. The total weight and uncertainty are:

	Net weight:	Uncertainty:
	8.50 grams	+/- 0.09 gram
	2.50 grams	+/- 0.09 gram
	3.50 grams	+/- 0.09 gram
	12.50 grams	+/- 0.09 gram
Totals:	27.00 grams	+/- 0.36 gram

Recommended Report Wording/Interpretation of Test Data:

Reference OSBI Protocol DR-3, Weights and Measures Utilized in Drug and Marijuana Reports.

Associated Forms:

DR4-1 Monthly Bench Scale Log
DR4-2 Monthly Large Capacity Scale Log
DR4-3 Monthly Analytical Scale Log

References:

1. Manufacturer's instructions and operating manual for balance/scale being used.
2. SWGDRUG. Supplemental Document SD-3 for Part IVC - Quality Assurance/Uncertainty, Measurement Uncertainty for Weight Determinations in Seized Drug Analysis. 2011.
3. ASCLD/LAB. Policy on Measurement Uncertainty. 2013, ASCLD/LAB Document Control Number: AL-PD-3060 Ver 1.1.
4. ASCLD/LAB. Guidance on the Estimation of Measurement Uncertainty – ANNEX A. 2013, ASCLD/LAB Document Control Number: AL-PD-3062 Ver 1.0.
5. ASCLD/LAB. Guidance on the Estimation of Measurement Uncertainty –ANNEX B. 2013, ASCLD/LAB Document Control Number: AL-PD-3063 Ver 1.0.
6. ASCLD/LAB. Guidance on Measurement Traceability. 2013, ASCLD/LAB Document Control Number: AL-PD-3058 Ver 1.0.
7. ASCLD/LAB. Guidance on Measurement Traceability-Measurement Assurance. 2013, ASCLD/LAB Document Control Number: AL-PD-3059 Ver 1.0.

DR-04 Attachment 1 Budget for Calculating Uncertainty of Measurement

Introduction:

Purpose

Laboratories performing testing that meet ISO/IEC 17025 must report uncertainties in conformance with the ISO Guide to the Expression of Uncertainty in Measurement (hereafter called the GUM).

Prerequisites

- Calibration certificates with valid uncertainties must be available for all calibrated equipment, reference standards and reference materials. These are supplied by the vendor.
- Statistical data regarding the calibration measurement process must be available; preferably from measurement control program (i.e. historical data) and available from the laboratories in-house measurement control process.
- Knowledge of the technical basis for the measurement is critical for completeness in uncertainty evaluation. This can be obtained through reference papers, reference procedures, experimentation, inter-laboratory comparisons and training.

Summary of Method:

Scope and Precision

Each measurement made has a corresponding uncertainty assigned to the measured value. The uncertainty is directly related to the measurement parameter (scope), range of the measurement, the equipment or measurement process being used (affecting precision), and the standards available with associated uncertainties.

Summary

This uncertainty analysis process uses the following eight steps:

1. Specify the measurement process.
2. Identify and characterize uncertainty components.
3. Quantify uncertainty components in applicable measurement units.
4. Convert uncertainty components to standard uncertainties in units of the measurement result.
5. Calculate the combined uncertainty.
6. Expand the combined uncertainty using an appropriate coverage factor.
7. Evaluate the expanded uncertainty against appropriate tolerances, user requirements, and laboratory capabilities.
8. Report correctly rounded uncertainties with associated measurement results.

The Process of Measurement Uncertainty Estimation

Step 1. Specify the process.

The measurement process consists of weighing evidence on electronic balances or large capacity scales. The samples could be in various forms: liquid, solid, powder, plant material, etc. The samples will either be net weighed or gross weighed. The containers for weighing could include: weigh paper, butcher paper, plastic bags, weigh boats, designated plastic tubs, or for gross weights: the containers the samples are in. The analyst has the options of:

1. Taring the scale and then placing the container and sample directly onto the scale (gross weights only).
2. Placing the container on the scale, taring the scale and then pouring/placing the sample into the container while on the scale.
3. Placing the container on the scale, taring the scale, removing the container from the scale, pouring/placing the sample into the container and then returning container/sample to the scale.

To calculate uncertainty begins by defining the measurand by inputting the following information into OSBI Form DR4-1, DR4-2 or DR4-3:

1. Measurement (e.g., Weight of sample)
2. Range of measurement values (e.g., 0 to 4200 grams)
3. Procedure Name and Revision (e.g., DR4, Revision 10)
4. Estimation Prepared by (e.g., John Doe)
5. Date Prepared

Step 2. Identify and characterize uncertainty sources.

Identify and compile a list of the possible uncertainty components that will have an influence on the measurement process, see list of possible sources below.

- Identify all possible sources of uncertainty in a comprehensive list, characterizing them based on the evaluation method that will be used to quantify them (Type A, statistical methods or Type B, scientific judgment) and transfer the sources of uncertainty to the column labeled "Uncertainty Component" on the "Measurement Uncertainty Budget Form," located on OSBI Forms DR4-1, DR4-2 and DR4-3.

- The uncertainty components can be grouped into two categories. Characterize the components based on the evaluation method (Type A, statistical methods or Type B, non-statistical methods). Enter "A" or "B" for the type of method into the column labeled "Type" in the budget.

Potential Sources of Uncertainty for Balances

Balance Repeatability

The electronic bench-top balance(s) are checked each day before casework and monthly using NIST Class F, ANSI/ASTM Class 6 or better, mass standard weights. The daily check consists of placing a 500-gram weight and 1-gram weight on the balance; the monthly function test consists of putting a pre-determined range of weights on the balance. Large capacity scales are function

tested monthly with a pre-determined range of weights. If the scales are not used routinely, they must be checked prior to use. The measurements obtained from each check include a range of masses which are documented on the appropriate OSBI Form DR4 (DR4-1, DR4-2 or DR4-3). Balances that weigh within plus or minus 1% of all test weights are considered acceptable for service. These daily and monthly measurement checks may be used to calculate the standard deviations used in determining the uncertainty of measurement value. This would be a Type A evaluation, a method of evaluation of uncertainty by the statistical analysis of a series of observations.

Balance Linearity

The linearity of the balance is obtained from the manufacturer specification. The linearity of the balance becomes a potential source of uncertainty. This value may be used in determining the uncertainty of measurement value. This is a Type B evaluation, a method of evaluation by means other than the statistical analysis of a series of observations.

Balance Readability/Resolution

The last digit in the readability of the balance poses a potential source of uncertainty as the true value of the measurement could be slightly more or less than the balances reads. This number is obtained from the manufacturer specifications for the individual scale. The resolution value may also be used in determining the uncertainty of measurement value. This is a Type B evaluation.

Operator Uncertainty

All scientists generating case data while utilizing balances have undergone a successful training program and possess experience in the proper use of equipment. New scientists will participate in a Measurement Assurance Study, to demonstrate proficiency in the operation of the balances/scales. The operator uncertainty may be considered in calculating uncertainty. If it is considered, it will be a Type A evaluation, using the standard deviation of the study data.

Temperature Coefficient/ Environmental Conditions

All balances are operated within an environment consistent with manufacturers' guidelines. The temperature coefficient/sensitivity temperature drift may be obtained from manufacturer specifications. If included in the calculation of uncertainty of measurement, a 15°C temperature variation is assumed. This temperature variation is larger than any fluctuation seen in an OSBI lab. This value may be used in determining the uncertainty of measurement value. If included in the calculation, this is a Type B evaluation.

If the scale is equipped to make internal adjustments for temperature, the scale will be checked before casework samples are weighed on the balance/scale. This check of calibration status will provide objective evidence that the internal adjustment has had minimal impact on the performance of the scale. The check will be made using a NIST Class F, ANSI/ASTM Class 6 or better, 500-gram weight and 1-gram weight that has been calibrated by an outside vendor that maintains a quality system that meets or exceeds the requirements set forth in ISO/IEC 17025. Record of this check will be made on the appropriate DR-4 form. The uncertainty due to environmental conditions may be considered in calculating uncertainty. If it is considered, it will be a Type A

evaluation, using the standard deviation of the generated data.

Calibration Procedures

All balances/scales and weights are calibrated annually by an ISO/IEC 17025 accredited outside vendor. The uncertainty calculated by the outside vendors may also be used in determining the uncertainty of measurement value. These are Type B evaluations.

Corner Loading

The possibility of obtaining different values of a mass from different areas of the weighing surface poses a potential source of uncertainty. This uncertainty, however, is not considered in the overall uncertainty of measurement as all staff have been properly trained in the operation of the balance.

Air Buoyancy

The difference between the density of the mass standard and the density of the test sample could cause a potential source of uncertainty. It is considered insignificant as the level of uncertainty is expected to be below the readability of the balance. If it is in fact measurable, it would result in a lower reported weight and is therefore, not considered in the overall uncertainty of measurement.

Sampling

Reported weights are typically that of net weights of test materials. Determination of the net weight generally requires emptying contents out of its packaging (i.e. plastic bag or plastic wrapping). Any residual material left in the packaging is not weighed and therefore introduces a potential source of uncertainty. This source is not significant in the overall uncertainty of measurement as it is considered minimal and would never result in a reported weight greater than the true weight of the test material. Sampling will not be considered in the overall uncertainty of measurement.

Step 3. Quantify uncertainty estimates.

After the initial evaluation of potential sources of uncertainty, it was determined the balance repeatability, linearity, readability, temperature coefficient (if available) and/or environmental conditions, uncertainty from balance and weights calibration reports from outside vendor and will be considered in the estimate of uncertainty. During subsequent review of this protocol other sources of uncertainty may be included, if deemed necessary by the Technical Manager. Any change to the budget requires a recalculation of the combined and expanded uncertainties.

To consider an uncertainty component significant, it should cause a change in the value of the second most significant digit, leading zeros excluded, when included in the uncertainty calculations. For example, if the expanded uncertainty value is currently 0.052 g and including the value of an uncertainty contributor causes the new value to be 0.053 g, that contributor is considered significant. If no change results in the second significant digit, the ‘2’, the contributor is not considered significant.

The balance repeatability, operator uncertainty, and environmental conditions check (on scales equipped to make internal adjustments for temperature) are represented by the standard deviation

of multiple measurements of a mass standard. These standard deviations are considered a normal distribution; thus, the standard deviation "s" is defined as:

$$s = \sqrt{\frac{\sum_i (x_i - \bar{x})^2}{n - 1}}$$

n= the number of individual measurements x_i;

$\bar{x}$ = the arithmetic mean of the individual measurements x_i

A minimum of thirty-one measurements will be taken for each balance to determine standard deviation.

The readability, linearity, and temperature coefficient/sensitivity temperature drift (from manufacturer, if available) of each balance was obtained from the manufacturer.
The calculation for including the largest impact of the temperature coefficient is:

Temp. coeff * temp variation * upper limit of balance = Largest impact of Temp Coeff

Example:

Sensitivity temperature drift = 3 ppm / °C → 0.000003 / °C
Widest temperature variation = 15°C = +/- 7.5°C;
Upper limit of electronic balance = 4200 g

Largest impact of Temp Coefficient = 0.000003/°C x 7.5 °C x 4200g =0.0945 g

The values for percent relative uncertainty will be expressed in the form of grams therefore place a "g" in the column labeled "Units" on the budget form.

Identify the probability distribution for 'Type A' and 'Type B' uncertainty components and enter the type into the column labeled "Distribution" on the budget form. In general, most measurement applications are the normal, rectangular, and Student's t distributions. Some recommendations for selecting the appropriate distribution are as follows:

1. Normal distribution ('Type A') should be applied when a collection of repeat measurements of a quantity of interest are presented such as historical positive control data.

2. Normal distribution ('Type B') should be applied when calibration certificates contain the "expanded uncertainties" and is divided by the coverage factor (k) to obtain the standard uncertainty. See Table 1.

Table 1. Typical coverage factors

Coverage factor (k)	Level of confidence (%)	Divisor
1	68.27	1
1.960	95	1.960
2	95.45	2
3	99.73	3

3. Rectangular distribution should be applied if limits of ± are given without a confidence level (i.e., A 10 ml Grade A volumetric flask is certified to within ±0.2 ml. The standard uncertainty is $0.2/\sqrt{3} = 0.11$ ml).

4. Student's t distribution typically arises when the measurement process lacks sufficient number of measurements (n) i.e., historical data. A sample size of 30 at 29 degrees of freedom or higher, the t-distribution begins to approximate the normal (Gaussian) distribution. Apply the student's t distribution when measurements are $n < 30$. Therefore, analysis lacking historical data, a corrected coverage factor is used based on the Student's t table, see Appendix A.

   For example, for an analysis with no historical control data, a control is analyzed 15 times (degrees of freedom or df = n-1, or 14 in this example).

   Using the Student's t table, k_{corr} value of 2.20 would be used to calculate the expanded uncertainty at 2σ or 95.45% confidence limit.

5. If the probability distribution cannot be determined then treat it as a rectangular distribution.

The "Divisor" is determined from the probability distribution that was entered into the column labeled "Distribution" and documented in the column labeled "Divisor".

Table 2. Distribution and Divisors

Distribution	Divisor
Type A – Normal	Use Appendix A and degrees of freedom
Type B - Normal	See Table 1 and coverage factor from Cal. Cert.
Type B - Rectangular	$\sqrt{3}$
Student t when n<30	Use Appendix A and degrees of freedom

Degrees of freedom are used as an indication of the reliability of the uncertainty value. When the Student's t table is used then enter the numerical value for degrees of freedom into the column labeled "Degrees Freedom (n-1)" on the budget form.

Once the "Value", "Distribution", and "Divisor" for each source of uncertainty has been determined then the standard uncertainties for all of the sources can be calculated.

Step 4. Convert all factors to standard uncertainties.

Convert the standard uncertainties by dividing the relative uncertainty inputted into the "Value" column by its respective "Divisor" and enter that numerical value into the column labeled "Standard Uncertainty" on the budget form.

The "Measurement Uncertainty Budget Form" will calculate this numerical value.

Step 5. Calculate the combined uncertainty.

After calculating the standard uncertainties, the combined standard uncertainty can be calculated. Determine the combined standard uncertainty uc(y), simply by calculating the square root of the sum of each standard uncertainty squared (RSS). The "Measurement Uncertainty Budget Form" will calculate this numerical value.

Step 6. Calculate the expanded uncertainty.

The combined standard uncertainty is then multiplied by a coverage factor, k, based on the degrees of freedom, to provide a level of confidence of 95.45 % or 99.73 %, respectively. The equation used to determine the expanded uncertainty is as follows:

The coverage factor must be determined from a statistical table such as provided in the Guide to the Expression of Uncertainty in Measurement or NIST Technical Note 1297 (See Appendix A).

After the expanded uncertainty, U, has been determined, the numerical value will be rounded up to two significant digits.

Example

k = 2.000 U = 0.036 * 2.000 = 0.072 → 0.072

k = 3.000 U = 0.036 * 3.000 = 0.108 → 0.10

Step 7. Evaluate the expanded uncertainty.

The expanded uncertainty may be evaluated against established criteria such as tolerance limits, customer requirements, and/or calibration and measurement capabilities on the laboratory scope. For example, the expanded uncertainty must not exceed 20% of the prior year's calculation.

Step 8. Report the uncertainty.

When reporting measurement uncertainty, the value shall be reported in the Criminalistics Examination Report and shall be expressed as an expanded uncertainty and include the coverage probability.

This measurement result shall include the measured quantity value, y, along with the associated expanded uncertainty, U, and this measurement result shall be reported as y ± U where U is consistent with the units of y.

The measurement result and the rounded expanded uncertainty shall be reported in the same units and to the same level of significance.

Refer to OSBI Protocol DR-3, Weights and Measures Utilized in Drug and Marijuana Reports, for report wording.

Significant Digits and Rounding:

Numerical values of expanded uncertainties shall be reported to at most two significant figures, unless the following applies: when multiple weights are added together for a total weight and the total

expanded uncertainty exceeds two significant figures, the total reported weight and total expanded uncertainty shall be reported to the same level of significance.

The following rules apply for significant figures:

1. ALL non-zero numbers (1,2,3,4,5,6,7,8,9) are ALWAYS significant.
2. ALL zeroes between non-zero numbers are ALWAYS significant.
3. ALL zeroes which are SIMULTANEOUSLY to the right of the decimal point AND at the end of the number are ALWAYS significant.
4. ALL zeroes which are to the left of a written decimal point and are in a number >= 10 are ALWAYS significant.

Examples:

Number	# Significant Figures	Rule(s)
48,923	5	1
3.967	4	1
900.06	5	1,2,4
0.0004	1	1,4
8.1000	5	1,3

The following applies for rounding:

1. The numerical value of the measurement result shall be rounded to the least significant figure in the value of the expanded uncertainty, U.
2. When rounding, examine the digit following (i.e., to the right of) the digit that is to be the last digit in the rounded off number. The digit you are examining is the first digit to be dropped.
   a. If the digit immediately to the right of the last significant figure is LESS than 5, the last significant figure is unchanged.
   b. If the digit immediately to the right of the last significant figure is GREATER than 5, you round up.
   c. If the digit immediately to the right of the last significant figure is EQUAL to 5, you round up if the last significant figure is odd. You round down if the last significant figure is even.

Calculations:

Historical control data is a 'Type A' component requiring these calculations.

- For n measurements of the historical data x, the mean value is: $\overline{x} = \frac{1}{n} \sum_i x_i$
- The **standard deviation** is found by averaging the squares of the deviations, and then taking the square root:

$$s = \sqrt{\frac{\sum_{i=1}^{n}(x_i - \overline{x})^2}{n-1}}$$

- The mathematical expression for **relative standard deviation** (RSD):

$$\mathrm{RSD} = \frac{s}{|\bar{x}|}$$

- The mathematical expression for relative standard deviation of the mean:

$$\mathrm{RSD}_{\mathrm{mean}} = \frac{s}{\sqrt{n}}$$

When 'Type B' components have been identified and the uncertainty is given, it should be used. Typically, uncertainties given on calibration reports will be "expanded uncertainties" usually using a "coverage factor of k = 2." An "expanded uncertainty" is the standard deviation (or "standard uncertainty") which has been multiplied by a number called the "coverage factor".

E.g., A calibration certificate for a mass standard states:

1. "The uncertainty in the reported mass is ±26 mg at a level of confidence of 95% assuming a normal distribution." The standard uncertainty is …±26 mg ÷ 1.960 = ±13 mg (1.960 from Appendix A).
2. "The expanded uncertainty in the reported mass is ±26 mg with a coverage factor, k, of 2." The standard uncertainty is …±26 mg ÷ 2 = ±13 mg.

Laboratory Personnel Responsible for Oversight of Plan:

Any adjustments, modifications or changes to the plan outlined above are submitted to the Controlled Substances Technical Manager for advisement. The Technical Manager will make the final approval or disposition.

Implementation:

The method of reporting uncertainty of measurements is currently in place and documented in OSBI Criminalistics policies.

References:

1. "Uncertainty Analysis in Forensic Science." World of Forensic Science. 2005. Retrieved July 06, 2010 from Encyclopedia.com: http://www.encyclopedia.com/doc/1G2-3448300579.html
2. SWG-Drug Recommendations, 5th edition, January 29, 2010
3. AFQAM 1st Mid-year Workshop, Salt Lake Community College, Salt Lake City, Utah, June 10-11, 2008, Presenter: Mark Reufenacht, Heusser Neweigh
4. Taylor, B. N. & Kuyatt, C. E. "Guidelines for Evaluating and Expressing the Uncertainty of NIST Measurement Results." NIST Technical Note 1297, 1994
5. Miller, Val, "Recommended Guide for Determining and Reporting Uncertainties for Balances and Scales." NISTIR 6919, January 2002
6. "Significant Figures – Rules." Retrieved October 2, 2013 from http://www.usca.edu/chemistry/genchem/sigfig.htm
7. International Organization for Standardization, ISO/IEC 17025:2005 - General Requirements for the Competence of Testing and Calibration Laboratories
8. International Organization for Standardization, ISO/IEC Guide 98-3: 2008 - Guide to the Expression of Uncertainty in Measurement

Appendix A: Student T Table

Value of $tp(v)$ from the t-distribution for degrees of freedom v that defines an interval $tp(v)$ to $+tp(v)$ that encompasses the fraction p of the distribution.
Note: Table is taken from NIST Technical Note 1297.

Degrees of freedom v	Fraction p in percent					
	68.27(a)	90	95	95.45(a)	99	99.73(a)
1	1.84	6.31	12.71	13.97	63.66	235.80
2	1.32	2.92	4.30	4.53	9.92	19.21
3	1.20	2.35	3.18	3.31	5.84	9.22
4	1.14	2.13	2.78	2.87	4.60	6.62
5	1.11	2.02	2.57	2.65	4.03	5.51
6	1.09	1.94	2.45	2.52	3.71	4.90
7	1.08	1.89	2.36	2.43	3.50	4.53
8	1.07	1.86	2.31	2.37	3.36	4.28
9	1.06	1.83	2.26	2.32	3.25	4.09
10	1.05	1.81	2.23	2.28	3.17	3.96
11	1.05	1.80	2.20	2.25	3.11	3.85
12	1.04	1.78	2.18	2.23	3.05	3.76
13	1.04	1.77	2.16	2.21	3.01	3.69
14	1.04	1.76	2.14	2.20	2.98	3.64
15	1.03	1.75	2.13	2.18	2.95	3.59
16	1.03	1.75	2.12	2.17	2.92	3.54
17	1.03	1.74	2.11	2.16	2.90	3.51
18	1.03	1.73	2.10	2.15	2.88	3.48
19	1.03	1.73	2.09	2.14	2.86	3.45
20	1.03	1.72	2.09	2.13	2.85	3.42
25	1.02	1.71	2.06	2.11	2.79	3.33
30	1.02	1.70	2.04	2.09	2.75	3.27
35	1.01	1.70	2.03	2.07	2.72	3.23
40	1.01	1.68	2.02	2.06	2.70	3.20
45	1.01	1.68	2.01	2.06	2.69	3.18
50	1.01	1.68	2.01	2.05	2.68	3.16
100	1.005	1.660	1.984	2.025	2.626	3.077
∞	1.000	1.645	1.960	2.000	2.576	3.000

(a)For a quantity z described by a normal distribution with expectation μz and standard deviation σ, the interval $\mu z \pm k\,\sigma$ encompasses p = 68.27, 95.45, and 99.73 percent of the distribution for k = 1, 2, and 3, respectively.

DR-04 Attachment 2 Controlled Substances Scale Scenarios

The following are scenarios when a scale will have to be calibrated and/or undergo a Measurement Assurance Study or a Function Test.

A. Purchase a new scale

1. Calibrate, by outside vendor
2. A Measurement Assurance Study with all analysts in the lab or the analyst assigned to the scale, using different scenarios (paper, weigh boat, tubs) using calibrated weights needs to be performed.
3. Ensure the uncertainty on new scale is less than uncertainty being reported by lab system, if it's not, then the uncertainty for lab system will need to be changed.

B. Move a scale between labs

1. Calibrate, by outside vendor
2. IF the scale has undergone a Measurement Assurance Study with different scenarios, then perform:
   a. Perform a function test on the scale using the calibrated steel weights.
   b. Ensure the uncertainty on scale is still less than the uncertainty being reported by lab system.
3. IF the scale has NOT undergone a Measurement Assurance Study with different scenarios, then perform:
   a. A Measurement Assurance Study with different people in lab or the analyst assigned to the scale, using different scenarios (paper, weigh boat, tubs) using calibrated weights.
      - Ensure the uncertainty on the scale is less than uncertainty being reported by lab system, if it's not, then the uncertainty for lab system will need to be changed.

C. Move a scale within a lab

1. IF the scale *has been used for casework within 30 days* of the moving of the scale, then perform:
   a. A function test on the scale using the calibrated steel weights
      - IF the function test results **are the same or within 1%**, of the last monthly check, the scale can be used for casework.
      - IF the function test results **are NOT the same or within 1%**, of the last monthly check, the scale can NOT be used for casework.
        i. The scale will need to be recalibrated by an outside vendor
        ii. A Measurement Assurance Study using with different people in lab or the analyst assigned to the scale, using different scenarios (paper, weigh boat, tubs) and using calibrated weights will need to be performed.
        iii. Ensure the uncertainty on the scale is less than uncertainty being reported by lab system, if it's not, then the uncertainty for lab system will need to be changed.

2. IF the scale *has NOT been used for casework within 30* days of the moving of the scale and:
   a. IF the scale has NOT undergone a Measurement Assurance Study with different scenarios, then perform:
      i. A Measurement Assurance Study with different people in lab, using different scenarios (paper, weigh boat, tubs) and using calibrated weights.
      ii. Ensure the uncertainty on the scale is less than uncertainty being reported by lab system, if it's not, then the uncertainty for lab system will need to be changed.
   b. IF the scale has undergone a Measurement Assurance Study with different scenarios, then perform:
      i. A function test on the scale using the calibrated steel weights
         - IF the function test results are the same or within 1%, of the last monthly check, the scale can be used for casework.
         - IF the function test results are NOT the same or within 1%, of the last monthly check, the scale can NOT be used for casework.
           1. The scale will need to be recalibrated by an outside vendor.
           2. A Measurement Assurance Study with person assigned to scale, using calibrated weights needs to be performed.
           3. Ensure the uncertainty on scale is still less than the uncertainty being reported by lab system.

D. Hire a new Criminalist

- Receive uncertainty of measurement training
- IF the scale they are assigned has undergone a Measurement Assurance Study with different scenarios, then:
  1. The person needs to perform a Measurement Assurance Study with different scenarios (paper, weigh boat, tubs) using calibrated weights, on the scale they are assigned to and any common use scales (i.e. large capacity scale).
  2. Ensure the uncertainty on the scales is less than uncertainty being reported by lab system, if it's not, then the uncertainty for lab system will need to be changed.
- IF the scale they are assigned has NOT undergone a Measurement Assurance Study with different scenarios, then:
  1. The new person needs to perform a Measurement Assurance Study with different scenarios (paper, weigh boat, tubs) using calibrated weights, on the scale they are assigned AND
  2. Ensure the uncertainty on the scale is less than uncertainty being reported by lab system, if it's not, then the uncertainty for lab system will need to be changed.
- The new person needs to perform a Measurement Assurance Study with different scenarios (paper, weigh boat, tubs) using calibrated weights, on any common use scales (i.e. large capacity scale).
- Ensure the uncertainty on each scale is less than uncertainty being reported by lab system, if it's not, then the uncertainty for lab system will need to be changed.

DR-04 Attachment 3 Uncertainty of Measurement for OSBI Labs

Introduction:

Purpose

OSBI Laboratories performing drug testing must report uncertainty of measurement for cases involving trafficking levels of controlled substances. This Attachment is a record of the uncertainties for the OSBI Controlled Substance Laboratories.

Summary of Method:

Method

Individual scales within the OSBI Controlled Substance Laboratory system have a Measurement Uncertainty Budget and an expanded uncertainty. The uncertainty for scales with similar ranges (i.e. large capacity or bench scales) within an individual Controlled Substances Laboratory will be compared. The largest expanded uncertainty from those scales will be selected as the uncertainty reported for that laboratory.

The uncertainties for the OSBI Controlled Substance Laboratory system will be documented in the spreadsheets for each scale and will include historical data, including effective dates. The spreadsheets can be found at \\VM-FSC-FILES\ContSub\Balance. The values may change when a significant change is made in a laboratory, i.e. addition of new personnel.

The following reported uncertainties do not include the scales that are not currently being used for casework in the Controlled Substances Unit or scale 14547 (located on the FSC 3rd floor). If these scales are used for casework the individual reported uncertainties will be used.

Reported Uncertainties:

FORENSIC SCIENCE CENTER (Edmond)

Scale	Reported Uncertainty	Effective Date
Bench	0.09 gram	12-31-2023
Large Capacity	0.05 pound	12-31-2023
Analytical	0.00088 grams	12-31-2023
Bench	0.09 gram	12-31-2024
Large Capacity	0.05 pound	12-31-2024
Analytical	0.00088 grams	12-31-2024

NORTHEASTERN REGIONAL LAB (Tahlequah)

Scale	Reported Uncertainty	Effective Date
Bench	0.08 gram	12-31-2023
Large Capacity	0.04 pound	12-31-2023
Bench	0.08 gram	12-31-2024
Large Capacity	0.04 pound	12-31-2024
Analytical	TBD	TBD

DR-05 Pharmaceutical Identification by Literary Reference

Purpose:

Tablets and capsules can be tentatively identified by visual examination and comparison to literary references. This method of examination is intended to expedite analysis of drug cases by simplifying the identification process for tablets and capsules that only need tentative identification. It is also useful as a presumptive examination prior to gas chromatograph/mass spectrometer or FTIR examination

Associated Protocols:

DR-30 Gas Chromatography (Flame Ionization Detector)
DR-60 Drug Analysis by FTIR
DR-70 Gas Chromatograph Mass Spectrometer Methods for Drug Analysis

Specimen Required (Sample Handling and Preservation):

1. Pharmaceutical preparations can be identified by literary reference once the following requirements have been met:
    a. A controlled substance of the same schedule or higher, excluding marijuana, has been conclusively identified.
    b. A pharmaceutical preparation of the same schedule or higher has been conclusively identified.

2. Any tablet or capsule that appears to be a non-scheduled preparation.

3. "Look alike" and clandestinely manufactured tablets and capsules are not suitable for this examination.

Individual Steps of Protocol:

1. Examine tablet or capsule visually.

2. Look up description, including lettering and numbers on the tablet or capsule, in one of the following references.
    a. Physician's Desk Reference
    b. Drug Identification Bible
    c. www.drugs.com

3. Record the literary reference, literary reference result, and the amount/concentration of the literary reference result in the designated fields in the matrix panel in the BEAST. The literary reference result should be the active ingredient; listings such as No CDS are not acceptable.
    a. When listing the concentrations for a mixture (i.e. Acetaminophen/Hydrocodone) list the concentrations in the same order (i.e. 325 mg/10 mg).

4. Refer to Controlled Substances Quality Assurance Manual 7.3.B through 7.3.F when inventorying and describing tablets.

Protocol Notes:

1. Analysts must be aware of the possibility of clandestinely manufactured tablets that may or may not contain the indicated ingredient.

2. Pharmaceuticals that do not have any type of label should be analyzed for controlled substances whenever a schedule I or schedule II substance has not been conclusively identified.

3. Pharmaceutical identification by literary reference is considered a type of examination. Therefore, if an analyst searches for a literary reference and one cannot be located, it is not acceptable to report "No analysis" or equivalent report wording. Instrumental analysis is to be performed.

4. If a clandestinely manufactured tablet with a commercial logo is identified using instrumental analysis, other pharmaceuticals with the same logo in the case cannot be reported using a literary reference.

5. Clorazepate and nordiazepam cannot be differentiated on the mass spectrometer and must be reported using literary reference.

6. Pregabalin does not chromatograph and cannot be identified using instrumentation. Tablets/capsules of pregabalin must be reported using a literary reference. For instances in which instrumental analysis was attempted see DR-70 Recommended Report Wording.

7. Dexmethylphenidate and Methylphenidate
    a. Dexmethylphenidate is one of 4 isomers of methylphenidate. When methylphenidate is sold, it is sold as a mixture of isomers. When separated from the mixture and sold as dexmethylphenidate, dexmethylphenidate is reported to be approximately twice as strong as methylphenidate. Dexmethylphenidate is sold as Focalin by Novartis, Attenade by Celgene and as a generic by a couple of different companies.

    b. Methylphenidate is listed in OK Statutes and in the Code of Federal Regulations (CFR) as a Schedule II substance. Oklahoma Statutes has a statement regarding isomers of methylphenidate. CFR does not a have statement regarding isomers of methylphenidate. The CFR specifies isomer for amphetamine and meth and has an overall statement for isomers in the next section regarding Depressants; dexmethylphenidate is a stimulant. The Oklahoma Pharmacy Board interprets dexmethylphenidate as controlled only at the federal level.

    c. Per an email from DEA Headquarters in 2014: *The CFR names methylphenidate and does not distinguish any one of its four isomers. Thus, all isomers are controlled. … all the isomers are controlled and they have the same drug code of 1724. The "dex" isomer is likely the most commonly encountered because it is the most active and produced commercially. As a suggestion, it may be best to just report it as methylphenidate as that is the term used in the CFR.*

d. If a tablet or capsule of dexmethylphenidate is encountered in casework, use a literary reference and the following report wording:

> Visual and literary reference indicates Dexmethylphenidate Federal Schedule II. Instrumental analysis was not performed.

If analysis is required, Methylphenidate, Schedule II will be reported.

e. Testimony should be that dexmethylphenidate is an isomer of methylphenidate.

Recommended Report Wording/Interpretation of Test Data:

1. For literary references without instrumental analysis:

   Visual and literary examinations indicate Clonazepam, Schedule IV. Instrumental analysis was not performed.

2. For non-controlled preparations, report using one of the following examples:
   a. No controlled dangerous substances indicated by literary reference.
   b. After visual and literary examination, no controlled dangerous substance was indicated. No further analysis was performed.
   c. Visual and literary examinations indicate Cyclobenzaprine, a non-controlled substance. Instrumental analysis was not performed.

3. For crudely manufactured or suspected clandestine tablets do not report a tentative conclusion by literary reference. Further analysis is required.

4. For controlled drugs in an exempt preparation (see Oklahoma Pharmacy Law book, 535:1-14-4. Exclusion of Rx Only products not federally scheduled from Oklahoma Controlled dangerous substances scheduling):

   Visual and literary examinations indicate the presence of Methyltestosterone, a schedule III substance in the form of an exempt preparation. Instrumental analysis was not performed.

5. Verbal reports and testimony in court will follow the above guidelines for wording. The fact that this is not a conclusive examination shall be emphasized.

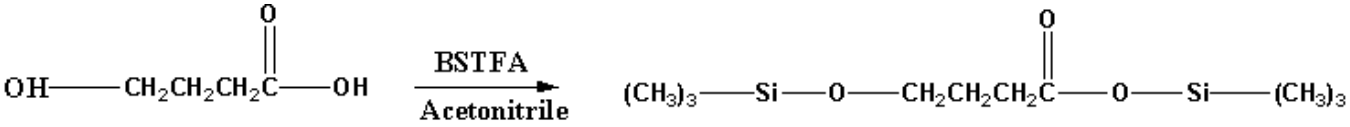
References:

1. Physician's Desk Reference
2. Oklahoma Statute Title 63
3. Drug Identification Bible
4. www.drugs.com

DR-07 Identification of Gamma-Hydroxybutyric Acid through Derivatization

Purpose:

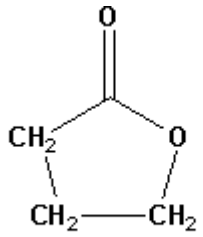
Gas Chromatography (GC) and Mass Spectroscopy (GC/MS) are unable to differentiate between Gamma-Hydroxybutyric Acid (GHB) and Gamma-Butyrolactone (GBL). However, when GHB is derivatized with bis-(trimethylsilyl) trifluoroacetamide (BSTFA) this results in a more stable compound that can be readily chromatographed and identified by GC and GC/MS. In the derivatization, the BSTFA reacts with the carboxylic and hydroxyl functional groups of the GHB molecule by replacing the hydrogen with a trimethylsilyl group; the trimethylchlorosilane is a catalyst for the reaction.



Gamma Hydroxybutyric Acid

Derivatized Gamma Hydroxybutyric Acid

Since GBL does not consist of functional groups that will react with BSTFA, no derivatization will occur.



Gamma Butyrolactone (GBL)

According to literature, it should be noted that GHB can readily convert to GBL in an acidic solution and/or with the application of heat, and GBL can readily convert to GHB in a basic solution.

Associated Protocols:

DR-30 Gas Chromatography Analysis (FID)
DR-70 Gas Chromatograph Mass Spectrometer
DR-60 Drug Analysis by FTIR

Specimen(s) Required (Sample handling and preservation):

Approximately 2 to 10 milligrams for a powder sample, or approximately ½ to 1 milliliter for a liquid sample.

Reagents:

1. Bis-(trimethylsilyl) trifluoroacetamide, choose one of the following:
   a. without trimethylchlorosilane

   b. with 1% trimethylchlorosilane
   c. with 10% trimethylchlorosilane
2. Acetonitrile
3. Chloroform

Standards and Controls:

1. Gamma Hydroxybutyric Acid Sodium Salt
2. Gamma Butyrolactone

Apparatus and Materials:

1. Test Tubes
2. Pipettes
3. Glass auto sampling vials and caps

Individual Steps of the Protocol:

For a dry sample:

1. In a disposable culture tube, place approximately 2 to 10 milligrams of the powder sample.
2. Add approximately ½ milliliter acetonitrile to the sample.
3. Add approximately 1 milliliter BSTFA to the sample and shake.
4. Analyze sample by GC and GC/MS using the appropriate drug method.
5. Compare retention times, retention indices and mass spectra of the sample to a known derivatized standard.

For a liquid sample:

1. Place ½ to 1 milliliter of a liquid sample into a disposable culture tube.
2. Wash the liquid sample with multiple 1 mL aliquots of chloroform.
   a. The first and last wash should be kept separate from the other washes and both should be shot on the GC.
   b. If the GC shows signs of the presence of GBL in the first wash, the GBL needs to be identified by GC/MS utilizing the Slow method.
   c. If signs that GBL is still present in the last wash, the sample should be washed until no signs of GBL are present.
   d. In the event a two-layer separation is not created with the chloroform wash, the analyst must document the attempted wash and then may analyze the sample for the identification of GBL using an analysis of the "neat" liquid sample.
3. Evaporate the washed sample to as close to dryness as possible.
4. Add approximately ½ milliliter acetonitrile to the sample.
5. Add approximately 1 milliliter BSTFA to the sample and shake.
6. Analyze sample by GC and GC/MS using the appropriate drug methods.
7. Compare retention times, retention indices and mass spectra of the sample to a known derivatized standard.

Due to the alternate temperature parameters associated with the identification of GBL, the retention index is not calculated. A GBL standard must be shot on the GC and GC/MS each time an

identification is made for comparison of retention times. The sample and standard must be analyzed using the same methods on both instruments.

Frequency and Tolerance of Controls and Corrective Action to Be Taken If Tolerances Are Exceeded:

Acetonitrile/BSTFA blanks are to be utilized for the negative control. If the negative controls (blanks) contain contamination, do not report any results. Refer to DR-70 for specific instructions/corrective actions.

Protocol Notes:

1. Sample must be reconstituted in acetonitrile. Due to the hydroxyl group in methanol, the derivatization of GHB will not take place in methanol. GHB is not soluble in chloroform; however, GBL is very soluble in chloroform.

2. Research has shown that 5 chloroform washes performed on a liquid sample are generally sufficient to remove all GBL. The number of washes may vary to ensure the removal of all GBL from the sample.

3. GHB became a Schedule I controlled substance in Oklahoma on April 13th, 1998.

4. GBL when packaged marketed, manufactured, or promoted for human consumption became a Schedule I controlled substance in Oklahoma on April 3, 2000. Legitimate uses for GBL which have been excluded in Schedule I include pesticides, photochemical etching, electrolytes of small batteries or capacitors, viscosity modifiers in polyurethane, surface etching of metal coated plastics, organic paint disbursements for water soluble inks, pH regulators in the dyeing of wool and polyamide fibers, foundry chemistry as a catalyst during curing, and curing agents in many coating systems based on urethanes and amides. Products containing GBL may also be exempted by the Director of OBN when labeled, marketed, manufactured, and distributed for legitimate industrial uses.

5. If the case is a question regarding a product that is being commercially sold which shows signs of GHB being present as a result of the addition of GBL, then the case should be referred to the Food and Drug Administration. To contact the FDA in Oklahoma, call (405) 231-4544.

Recommended Report Wording:

For a Dry Sample:

1. When the sample has been derivatized, and GC and GC/MS confirms the GHB derivative to be present, report as:

   Item #: Gamma-hydroxybutyrate, Schedule I.

2. When a GC examination has been performed and is positive for GHB derivative but the GC/MS is not conclusive due to the insufficient sample, report as:

   Item #: Examinations indicate the presence of a controlled substance; however,

the laboratory's reporting criteria have not been met,

3. When the sample has been derivatized, and GC and GC/MS results are negative or indicate another controlled substance, report according to DR-30 and DR-70 protocol, as appropriate.

For a Liquid Sample:

1. When the sample has been derivatized and GC and GC/MS confirms the GHB derivative to be present and no GBL is detected in the first chloroform wash, report as:

   Item # Gamma-hydroxybutyrate, Schedule I.

2. When the sample has been derivatized and GC and GC/MS confirms the GHB derivative to be present and the presence of GBL is identified in the first chloroform wash, report as:

   Item #: Gamma-hydroxybutyrate, Schedule I and Gamma-butyrolactone, Schedule I in accordance with 63 O.S. Section 2-204 (D)(E).

3. When the sample has been derivatized, and GC and GC/MS do not indicate the presence of GHB, but GBL has been identified, report as:

   Item #: Gamma-butyrolactone, Schedule I in accordance with 63 O.S., Section 2-204 (D)(E).

4. When a GC examination has been performed and is positive for GHB derivative but the GC/MS is not conclusive due to the insufficient sample, report as:

   Item #: Examinations indicate the presence of a controlled substance; however, the laboratory's reporting criteria have not been met,

5. When a GC examination has been performed and is positive for GBL, but the GC/MS is not conclusive due to the insufficient sample, report as:

   Item #: Quantity not sufficient for conclusive analysis.

6. When the sample has been derivatized, and GC and GC/MS results are negative or indicate another controlled substance, report according to DR-30 and DR-70 protocol, as appropriate.

References:

1. Blackledge, RD and Miller, MD. Identification of GHB. Microgram. XXIV:7.172.
2. Bommarito, C. Analytical profile of gamma hydroxybutyric acid. Journal of the Clandestine Laboratory Investigation Chemists Association. 3:3.10 July 1993
3. Reynolds, CS. Miami Dade Police Department Crime Lab, Miami, Florida. GHB Derivatization.

DR-10 Color Tests: Marquis

Purpose:

The Marquis test is a preliminary screening test that responds to particular functional groups causing a characteristic color change. Depending upon the compound and its functional groups, addition of sulphuric acid can cause oxidation, sulphonation, dehydration, esterification, polymerization, hydrolytic splitting and other reactions. A positive result to a color examination is not a conclusive identification and should be substantiated by other methods of analysis. However, these examinations can give the analyst a basis as to which extractions and/or examinations are necessary for further conclusive instrumental analysis.

Associated Protocol(s):

DR-13 Bates Test

Specimen Required (Sample Handling and Preservation):

Approximately 1 milligram.

Reagents:

Marquis Reagent

10 ml concentrated H_2SO_4 per 1 ml 37% formaldehyde

Preparation of Reagent:

1. H_2SO_4 added slowly to formaldehyde while stirring.
2. The marquis reagent is to be prepared and stored in a fume hood.
3. Spot test with a known sample of methamphetamine and observe an orange color change. Add a drop to a clean dish and observe no color change.
4. Transfer to dropper bottles

All chemicals are reagent grade or better.

Standards and Controls:

Once a month, the reagent will be tested with a positive control. Methamphetamine will be used as a positive control; for a positive result, the reagent will turn orange when it is dropped onto the methamphetamine. This test and results are to be recorded in the Excel spreadsheet "Monthly_Spot_Test_Verifications" which is located in \\VM-FSC-FILES\ContSub\Spot_Test_Verification_Drugs. If a reagent fails to pass QA/QC as expected it is to be discarded.

Each day the reagent is used for casework, a drop of the reagent will be placed in a clean weigh dish. If no color reactions are observed, the reagent is suitable for use that day. If color reactions are observed, the reagent will be discarded immediately. The negative control is to be documented in each case utilizing this reagent.

Apparatus and Materials:

1. Spot plates or weigh dishes
2. Dropper bottle

Individual Steps of Protocol:

1. Transfer a small amount of the sample to a spot plate or weigh dish.
2. Place 1 - 2 drops of Marquis reagent on the sample.
3. Observe and record any changes in the color of the reagent. The change should occur within 5-15 seconds for amphetamine, methamphetamine, and opiates depending on the concentration of the sample being tested and the age of the reagent.

Expected Values; Values Requiring Special Notification:

Orange	Purple	Black
Amphetamine (→brown)	Opiates	Methylenedioxyamphetamine
Methamphetamine (→brown)	Guaifenesin	3,4-Methylenedioxymethamphetamine
Phentermine (→brown)	Morphine	
Mescaline	Codeine	
Pethidine	Propoxyphene	
Psilocybin		

Yellow	Red
Chlordiazepoxide	Aspirin
Hydrocodone (→brown→violet)	Pentazocine
Hydromorphone (→red→violet)	Thebaine
Lorazepam	
Oxycodone (→brown→violet)	

For further information on color reactions refer to literary references.

Protocol Notes:

1. This reagent is volatile due to the formaldehyde. Marquis reagent is also very sensitive to water. Store in a dry place. If reagent turns brown, discard by flushing down the drain with large amounts of water.

2. This spot test is to be performed in a fume hood.

References:

1. Clarke's isolation and identification of drugs. The Pharmaceutical Press. London, 1986.
2. Colorado Bureau of Investigation. Standard Operating Procedures. 1987, revised May 1, 1992.
3. Illinois Dept. of State Police. Chemistry Procedures Manual. September, 1986.
4. Berman E. Analysis of drugs of abuse. 1977

DR-11 Color Tests: Cobalt Thiocyanate

Purpose:

Color tests are preliminary screening tests that react to particular functional groups causing a characteristic color change. The results of these color examinations are not a positive identification; however, these examinations can give the analyst a basis as for which extractions and/or examinations are necessary for further conclusive analysis. The cobalt thiocyanate spot test is used primarily as a screening tool to detect cocaine hydrochloride.

Associated Protocol(s):

DR-13 Bates Test
DR-21 Cocaine Free Base Determination by Hexane Solubility

Specimen Required (Sample Handling and Preservation):

Approximately 1 milligram.

Reagents:

Cobalt Thiocyanate Reagent

2 gm Cobaltous Thiocyanate
100 ml deionized H2O

Preparation of Reagent:

1. Weigh 2 grams of cobalt thiocyanate.
2. Dissolve in approximately 100 ml of deionized water.
3. Filter (optional)
4. Transfer to dropper bottle.
5. Spot test with a known sample of cocaine HCl and observe blue color change. Add a drop to a clean dish and observe no color change.

Cobalt thiocyanate reagent has an indefinite storage life.
All chemicals are reagent grade or better.

Standards and Controls:

Once a month, the reagent will be tested with a positive control. Cocaine HCl will be used as a positive control; for a positive result, the reagent will create a blue precipitate when it is dropped onto the cocaine HCl. This test and results are to be recorded in the Excel spreadsheet "Monthly_Spot_Test_Verifications" which is located in \\VM-FSC-FILES\ContSub\Spot_Test_Verification_Drugs. If a reagent fails to pass QA/QC as expected it is to be discarded.

Each day the reagent is used for casework, a drop of the reagent will be placed in a clean weigh dish. If no color reactions are observed, the reagent is suitable for use that day. If color reactions are observed, the reagent will be discarded immediately. The negative control is to be documented in each case utilizing this reagent.

Apparatus and Materials:

1. Spot plates or weigh dishes
2. Dropper bottle

Individual Steps of Protocol:

1. Transfer a small amount (1-2 milligram) of the sample to a spot plate or weigh dish.
2. Place 1 - 2 drops of cobalt thiocyanate reagent on the sample.
3. Observe and record any color changes. Changes should occur within 1-2 seconds depending on the concentration of the sample being tested.

Expected Values; Values Requiring Special Notification:

Blue Precipitate:

Cocaine HCl	Procaine HCl	Lidocaine HCl

Blue Precipitate and/or Blue Solution:

Antihistamines	Codeine	Meperidine	Nicotine
Atropine	Heroin	Methqualone	Pentazocine
Methadone	Opium	Propoxyphene	Quinine
Methylphenidate	PCP	Pyrilamine	

For further information on color reactions refer to literature references.

Protocol Notes:

1. Spot tests are to be performed in fume hood.

2. Intended use for cobalt thiocyanate reagent is for a preliminary exam for cocaine HCl.

3. Cocaine free base will show no change.

References:

1. Clarke's isolation and identification of drugs. The Pharmaceutical Press, London, 1986.
2. Colorado Bureau of Investigation. Standard Operating Procedures, 1987, revised May 1, 1992.
3. Illinois Dept. of State Police. Chemistry Procedures Manual, September, 1986.

DR-13 Color Tests: Bates Examination

Purpose:

Color tests are preliminary screening tests that respond to particular functional groups causing a characteristic color change. The results of these color examinations are not a positive identification; however, these examinations can give the analyst a basis as to which extractions and/or examinations are necessary for further conclusive analysis. The Bates examination is used primarily as a screening tool for cocaine base.

Associated Protocol(s):

DR-10 Marquis
DR-11 Cobalt Thiocyanate
DR-21 Cocaine Free Base Determination by Hexane Solubility

Specimen Required (Sample Handling and Preservation):

Approximately 1 milligram.

Reagents:

Reagent #1: Cobalt Thiocyanate

2 gm Cobaltous Thiocyanate per 100 ml deionized H2O
See DR-11 for Preparation of Reagent

Reagent #2: Marquis

10 ml Concentrated H2SO4 per l ml of 37% Formaldehyde
See DR-10 for Preparation of Reagent

All chemicals are reagent grade or better.

Standards and Controls:

Once a month, the reagents will be tested with a positive control. Cocaine base will be used as a positive control; for a positive result, the reagents will turn blue when the marquis is dropped onto the cobalt thiocyanate and cocaine base. This test and results are to be recorded in the Excel spreadsheet "Monthly_Spot_Test_Verifications" which is located in \\VM-FSC-FILES\ContSub\Spot_Test_Verification_Drugs. If a reagent fails to pass QA/QC as expected it is to be discarded.

Each day the reagent is used for casework, a drop of the reagent will be placed in a clean weigh dish. If no color reactions are observed, the reagent is suitable for use that day. If color reactions are observed, the reagent will be discarded immediately. The negative control is to be documented in each case utilizing this reagent.

Apparatus and Materials:

1. Spot plates or weigh dishes
2. Dropper bottle

Individual Steps of Protocol:

1. Transfer a small amount (1-2 milligram) of the sample to a spot plate or weigh dish.
2. Place 1 drop of cobalt thiocyanate reagent on the sample, followed by one drop of Marquis reagent.
3. Observe and record any changes in the color of the reagent. Changes should occur rapidly, within 1-3 seconds.

Expected Values; Values Requiring Special Notification:

Cocaine HCl turns blue after cobalt thiocyanate is added.

Cocaine base - no reaction occurs with the addition of Cobalt thiocyanate, however, turns blue after Marquis reagent is added.

Protocol Notes:

1. Cobalt Thiocyanate reagent is a very stable reagent with an indefinite shelf life.

2. The spot test is to be performed in a fume hood.

References:

1. Clarke's isolation and identification of drugs. The Pharmaceutical Press, London, 1986.
2. Colorado Bureau of Investigation. Standard Operating Procedures, 1987, revised May 1, 1992.
3. Illinois Dept. of State Police. Chemistry Procedures Manual, September, 1986.

DR-21 Cocaine Free Base Determination by Hexane Solubility

Purpose:

Cocaine HCl and cocaine free base give identical gas chromatograph retention times and mass spectra because the HCl is disassociated from the cocaine molecule in the heated injection port of the gas chromatograph. Cocaine HCl and cocaine free base can be differentiated based on their solubility in hexane. Cocaine HCl is not soluble whereas cocaine free base is readily soluble in hexane.

Associated Protocol(s):

DR-11 Color Tests: Cobalt Thiocyanate
DR-13 Color Tests: Bates Examination
DR-30 Gas Chromatography Analysis (Flame Ionization Detector)
DR-60 Drug Analysis by FTIR
DR-70 Gas Chromatography Mass Spectrometer Methods for Drug Analysis

Specimen(s) Required (Sample Handling and Preservation):

Sample believed to be cocaine free base.

Reagents:

Hexane or hexanes, reagent grade or better.

Standards:

Follow DR-30, DR-60, and DR-70 regarding standards.

Controls:

Follow DR-30, DR-60, and DR-70 regarding controls.

Apparatus and Materials:

1. Agilent Gas Chromatograph or equivalent.
2. Agilent Gas Chromatograph/Mass Spectrometer or equivalent.
3. Fourier Transform Infrared Spectrometer
4. Test tubes or sample vials

Individual Steps of Protocol:

Gas Chromatograph and Gas Chromatograph Mass Spectrometer

1. Place an appropriate amount of sample in a test tube.
2. Add Hexane.
3. Vortex or shake the test tube.
4. Centrifuge, filter, or allow the undissolved sample to settle.
5. Analyze Hexane solution with GC and GC/MS.
6. If the Hexane is negative, utilize other extraction methods and re-analyze for other compounds.

FTIR

1. Perform a Cobalt Thiocyanate and Bates examination color test.
2. Analyze sample on the FTIR using an ATR accessory.

Calculations:

Refer to DR-30, DR-60, and DR-70.

Frequency and Tolerance of Controls and Corrective Action to Be Taken If Tolerances Are Exceeded:

Refer to DR-30, DR-60, and DR-70.

Protocol Notes:

1. Cobalt Thiocyanate, DR-11 and the Bates test DR-13 are presumptive tests the analyst may find useful in providing a preliminary indication of whether the base or HCl form of cocaine is present.

2. The analyst must be aware of the possibility of a mixed sample containing both the free base and the HCl form of cocaine.

3. For an identification, the requirements in DR-30, DR-60, and DR-70 must be met.

Recommended Report Wording/Interpretation of Test Data:

When cocaine is dissolved in hexane and conclusively identified by instrumental analysis using the protocol DR-21, report as follows:

Item #: Cocaine base, Schedule II.

References:

Clarke's Isolation and Identification of Drugs

DR-30 Gas Chromatography Analysis (Flame Ionization Detector)

Purpose:

Gas chromatography is an instrumental analysis that separates the volatile organic components in a mixture. The gas chromatograph consists of three basic parts: an injection port, a chromatographic column, and a detector. A prepared sample is injected into the heated injection port, where it is vaporized. As the vaporized sample enters and travels through the chromatographic column, the different components are separated based on their boiling point, molecular size, and affinity for the liquid phase in the column. When the separated components of the sample reach the end of the column, they enter the flame ionization detector where they encounter a hydrogen flame. The flame ionizes the emerging chemical substance, thus generating an electrical signal. The electrical signal is converted into a digital signal that is processed, stored, and printed out by a data system. The time from injection of the sample until the detector produces a signal is called the retention time. The retention time of the unknown sample can be compared to that of a known standard to indicate the presence of a particular substance. The gas chromatograph is a presumptive identification for drug samples.

Associated Protocol(s):

DR-70 Gas Chromatography Mass Spectrometer Methods for Drug Analysis
DR-75 Gas Chromatograph Infrared Detector Methods for Drug Analysis

Reagents:

1. Methanol
2. Chloroform
4. Hexanes
5. Or other suitable solvent

All must be Reagent Grade or better.

Standards:

Standards are to be purchased from reputable manufacturers. Samples of unknown origin or from cases are not suitable for making a comparison. All standards will be verified by the Technical Manager or designee prior to use in casework. All records will be maintained with the standard log at FSC for future reference or in the Standard Verification folder which can be found at \\VM-FSC-FILES\ContSub\Standard_Verifications.

Controls:

Each day that casework is performed, the first sequence is to contain a solvent blank as a negative control and an approximate one to two milligram/milliliter concentration cocaine standard for a positive control. The solvent blank data file will be named “blank” and the cocaine standard data file will be named “cocaine”. When sequences continuously run for an extended period of time, for example 2 or more days, it is not necessary to re-run the positive control. A solvent blank will be shot at least every fifth sample (4 samples followed by a blank) thereafter on each column. Controls will be run and documented in each case file. Sequence logs will be digitally archived; therefore, sequences should be allowed to finish for archiving reasons.

Apparatus and Materials:

1. Agilent Gas Chromatograph
2. J&W/Agilent/Phenomenex or Restek DB/HP/ZB-1/Rtx-1 (15m x 530um x 1.5 um), DB/HP/ZB-50 (15m x 530um x 1 um). The Technical Manager of Controlled Substances or designee may approve other columns for use.
3. Five (5) or ten (10) microliter volume syringes.
   NOTE: *The method must have the correct syringe size listed.*
4. Glass inserts for vials.
5. Caps for auto sampling vials.
6. Gas purifier, if necessary.
7. Glass wool.
8. Low bleed septa.
9. Four mm large bore split/splitless injection liners.
10. Inlet gold seals and washers.

Methods Approved for Analysis:

Methods and macros for drug analysis using the Agilent GC are available from the OSBI Technical Manager. These methods are to be utilized by all drug analysis sections of the OSBI using the Agilent GC. Any requests for new methods/macros or changes to existing methods/macros should be submitted in writing to the technical manager of drug analysis for review and approval prior to any changes being made.

Unless indicated in the description, methods utilize the analysis of 1 microliter injections. The current approved methods are as follows:

Drug1:	General drug analysis method.
Drug2:	General drug analysis method utilizing a 2 microliter injection.
Drug1F:	General drug analysis method used on the 8890 GC/5977B MS.
Drug1FE:	General drug analysis method with extended final time for compounds, such as steroids used on the 8890 GC/5977B MS.
Drug1FSV:	General drug analysis method for solvents used on the 8890 GC/5977B MS.
Extend1:	General drug analysis method with extended final time for compounds such as steroids.
Extend2:	General drug analysis method with extended final time for compounds such as steroids with a 2-microliter injection to place a larger amount of sample on the column.
Extend45:	General drug analysis method with extended final time for late eluting compounds such as synthetic cannabinoids on ZB-50 column.
Extend55:	General drug analysis method with extended final time for late eluting compounds.
Extend1F:	General drug analysis method with extended final time for compounds, such as steroids used on the 8890 GC/5977B MS.
Lowflow:	Same method as Drug1, although it turns off the air and hydrogen, while

reducing the helium flow through the column from 10ml/min to 1 milliliter/min. When a GC sequence stops, it will revert to the method loaded in the instrument panel. Therefore, this method should always be loaded in the instrument control panel, as it conserves gases and lowers the demand on the hydrogen generators and air compressors.

Method1:	Nine-minute run designed to separate Ephedrine and Pseudoephedrine. (See protocol note 7 for requirements to use this method.) (DB/HP/ZB-50 columns only.)
Method2:	Twenty-minute run designed to separate Ephedrine and Pseudoephedrine. (This method to be used if protocol requirements in protocol note 7 is not met for abbreviated runs.) (DB/HP/ZB-50 columns only.)
OpiateSep:	Method to separate oxycodone and oxymorphone on a DB/HP/ZB -1 column. It can also separate hydrocodone and morphine on the DB/HP/ZB-50 column.
PCPTCP:	Run for the separation of PCP and TCP
Steroid:	General steroid analysis method with extended final time. This method utilizes a steroid ladder in place of the standard drug ladder.
TFMPP1:	Utilizes the Drug1 method, but replaces the standard drug ladder with the TFMPP ladder

Temperature programming must be capable of eluting C34 except as specifically provided for in the protocol notes.

The methods listed above utilize Helium as a carrier gas.

Individual Steps of Protocol:

Suggested procedure:

1. Dilute the sample in an amount of suitable solvent, which would give a concentration of approximately 1.0 mg/milliliter.
2. Enter case and sample information into Chemstation sample log table, including case number, sample item number, extraction solvent, analyst's name, method to be used for analysis and vial barcode.
3. Inject the sample onto the gas chromatograph. Sample size shall be an amount that when vaporized will not exceed 990 microliters. Do not inject more than one microliter of methanol or an unknown solvent (i.e., clan labs). See "Calculations" for solvent expansion volumes.
4. Compare unknown retention time to known standard's retention time. Unknown retention time values must be within 2% plus or minus that of the standard. It is not necessary to document the retention time difference.
5. If needed, perform sample cleanup extraction, i.e. NaOH/CHCl3 or a back extraction using HCl/water/NaOH/CHCl3.
6. If unknown sample exhibits sign of overloading the GC column, dilution and re-injection is needed.

Calculations:

During injection, the solvent vaporizes. If the vapor exceeds the volume of the glass inlet liner, it may contaminate metal lines and parts within the injector, which can subsequently result in carry over or contamination of future injections.

Expansion volumes for different pressures or inlet liner sizes can be calculated using the following formulas:

$$\text{Sample and solvent expansion volume} = \frac{nRT}{P}$$

$$n = \#\text{ of moles of solvent and sample} = \frac{\text{volume (milliliter) x density (gram/milliliter)}}{\text{MW (grams/mole)}}$$

T = absolute temperature of injector (K)
P = column head pressure (Atm) + 1 Atm
R = gas law constant = 82.06 cc atm/mole x deg. K

$$\text{Volume of inlet liner} = \pi r^2 L$$

r = radius (cm)
L = length (cm)

Solvent Expansion Volumes in µl at Various Head Pressures

Solvent	Density (gram/milliliter)	MW (gram/mole)	5 psig	10 psig	15 psig
Hexane	0.66	86	264	210	175
Chloroform	1.49	119	430	343	285
Methanol	0.79	32	852	680	565
Toluene	0.87	92	325	260	216
BSTFA	0.96	257	128	103	85
Acetonitrile	0.78	41	656	523	435

The expansion volumes were determined using a 1.0 µl injection volume, a 290°C injection port temperature, and a head pressure of 5, 10, and 15 psig. Double the volumes for 2 µl injections.

The 4mm inlet liner has a 990 µl volume. Solvent expansion volumes are not to exceed the capacity of the liner.

Frequency and Tolerance of Controls and Corrective Action for exceeded Tolerances:

Retention time values for unknown samples are to be within 2% of the known standard. All blanks shot during the course of an analysis sequence must be free from sample contamination, and the only integrated peak will be the solvent peak. If contamination or baseline integration is present, determine cause of deviation immediately and correct.

The following requires corrective action before casework can be resumed:

1. The contamination of blanks during the course of an analysis sequence. Contamination is defined as carry over from a previous sample into a later sample or control in a sequence or manual run:
    - Do not report any results following a contaminated blank.

2. Methanol blanks run for QA purposes, during the course of an analysis sequence, with baseline integration. Baseline integration occurs when the baseline has risen above the set threshold and Chemstation gives the baseline a value. Analysts will not change the baseline threshold without consulting the Technical Manager. The only peak integrated in a methanol blank should be the solvent peak.
    - Do not report any results following the integrated baseline.

3. The failure to produce a satisfactory chromatogram for the cocaine standard (positive control) or standard ladder:
    - Do not report any results from the analysis sequence.

    Note: A satisfactory cocaine standard is one in which the solvent and cocaine are the only peaks integrated in the QA/QC for case samples. A satisfactory standard ladder is one that all standard peaks are integrated by the instrument, including on all scans thereafter used in casework. If a cocaine or standard ladder peak is not integrated by the instrument and manually entered by an analyst, this will need to be noted on all chromatograms used in casework.

4. The value for the retention time of a single unknown and the standard exceed a plus or minus 2% difference:
    - The operator is to diagnose the problem. Typically, column overloading due to large sample size is the situation. Dilution of the sample will give suitable chromatography.
    - The sample in which this occurs will not be reported until it is within the 2% window.
    - If sample is outside the 2% window it will need to be rejected and entered into the rejection panel.

If the problem cannot be corrected, the instrument should be taken out of service and vendor contacted for repairs.

Any instrument removed from service will be documented in the GC maintenance log (DR30-1) and reported immediately to the Supervisor and Technical Manager in charge of drug analysis.

Protocol Notes:

1. Injection liners and septa are to be changed at or before 250 injections. The analysis of un-extracted or dirty samples may necessitate more frequent injection liner changes. The gold seal in the injector is to be changed at or before 1250 injections. When the gold seal is changed, the metal sides of the injector are to be cleaned with cotton tipped swabs and methanol until no discoloration of the swabs is present and metal septum nuts are to be cleaned and rinsed in

methanol. The split vent line is to be changed at or before 5000 injections. The split vent filter is to be changed at or before 5000 injections, on the Agilent GC.

2. Routine maintenance (i.e. septum change, column changes, liner changes, gold seal changes, injector/septum nut cleaning, standard ladders) and repairs are to be documented on the instrument maintenance log.

3. Inject a sufficient number of solvent blanks after liner and septa changes on each column to eliminate extraneous background noise. A new QA/QC blank must be shot before casework resumes.

4. Ensure that the solvent bottles contain sufficient solvent to rinse the syringe. The wash bottle is to be labeled with the minimum liquid level.

5. Split ratios are to be recorded in header information in macros.

6. Partial or abbreviated GC runs are not normally to be utilized except as specifically provided for in the protocol. Abbreviated runs increase the risk of retaining compounds on the GC column which may elute in a later run. All runs will be of sufficient duration and programming to elute C34 with the exception of the following circumstance. Based upon the interpretation of previously run gas chromatographic data, a short GC run of 9 minutes or longer may be utilized in the determination of ephedrine or pseudoephedrine. The gas chromatographic data must show no sign of any further analyte in a sample beyond 1.0 minutes past the retention time of the pseudoephedrine peak on the standard ladder. All blank runs between samples and cases must still meet the C34 requirement.

7. All gas chromatograph comparisons will be made by comparing the question sample to a chromatogram of a known documented standard that has been analyzed on the same column. The question sample retention time values must be within 2% plus or minus that of the standard. The data for comparisons will be kept in the case file.

8. A standard ladder will be prepared at approximately a one to two milligram/milliliter concentration in methanol or other suitable solvent and analyzed monthly. If the standard ladder lot number is recorded on casework printouts, then it is not necessary to record the individual lot numbers of each drug. Standards that are not in the drug standard ladder or specialty standard ladders will be analyzed individually. These standards should include the lot number on instrument printouts and be within 60 days of the sample. Specialty standard ladders, such as Ephedrine/Pseudoephedrine or Steroid ladder, are not required to be analyzed monthly. These ladders can be run on an as needed basis.

9. Refer to www.agilent.com for instrumentation user's manual.

Recommended Report Wording/Interpretation of Test Data:

1. The gas chromatograph examination is a presumptive identification that should be followed by a confirmatory examination by FTIR, GC/MS, or GC/IRD. All samples that are screened on the GC should be further analyzed by FTIR, GC/MS, or GC/IRD.

2. The OSBI Quality Manual requires all items in a container be listed on each report. In the case of evidence containing items needing additional analysis and items that have been tested and reported in a previous report, report the untested items using the following example:

   Item #: No Additional Analysis

Associated Form(s):

DR30-1 Maintenance Log for OSBI GC

References:

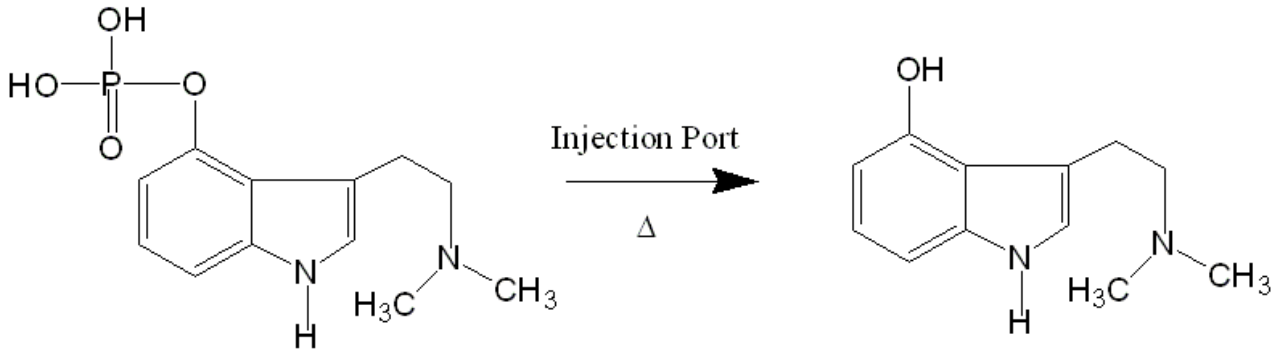
1. Bonelli EJ, McNair HM. Basic gas chromatography. Berkeley: Varian, 1969.
Hewlett Packard. Practical capillary chromatography: A short course. Atlanta: HP, 1988

DR-45 Analysis of Mushrooms to Determine Presence of Psilocyn or Psilocybin

Purpose:

Suspected mushroom samples or any other submittal which may contain psilocyn or psilocybin will be subjected to the examinations set forth in this protocol. The materials shall be analyzed using GC and GC/MS according to DR-30 and DR-70.

Under GC injection port conditions psilocybin rapidly dephosphorylates into psilocyn[1], therefore an additional test must be conducted in order to determine if the sample contains psilocyn, psilocybin, or a mixture of the two substances.



Psilocybin
$C_{12}H_{17}N_2O_4P$
MW: 284.25

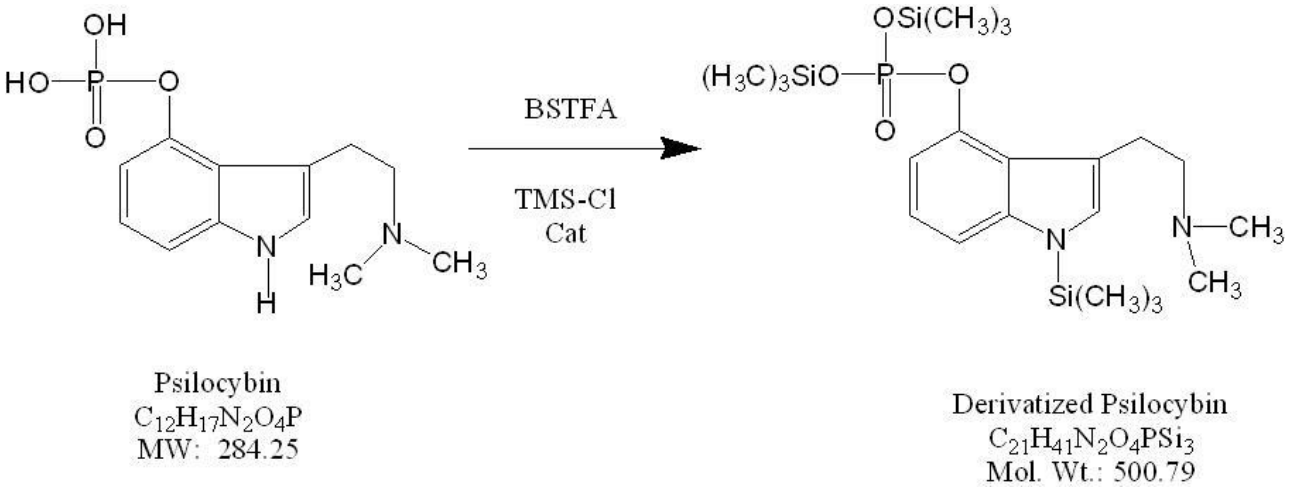
Psilocyn
$C_{12}H_{16}N_2O$
MW: 204.27

Figure 1: Conversion of Psilocybin in Injection Port

To differentiate between psilocyn and psilocybin, the analyst has a choice of either using thin layer chromatography to determine the compound present or derivatizing the sample using GC and GC/MS for identification.

The thin layer chromatography method works without subjecting the sample to high temperature conditions that promote dephosphorylation of psilocybin into psilocyn.

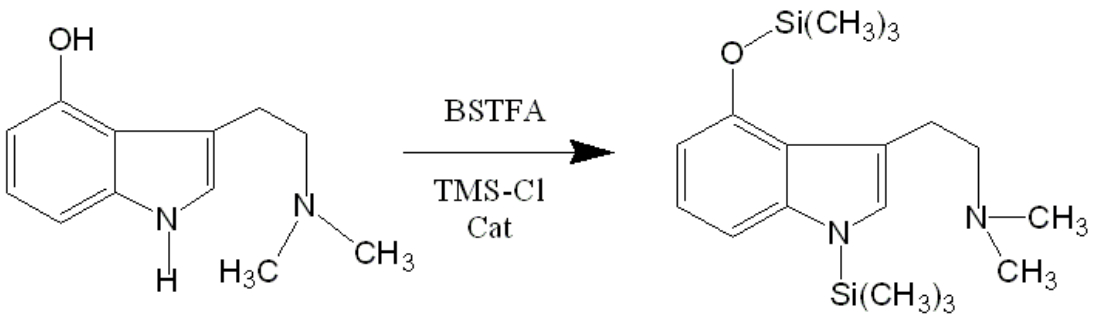
The derivatization method works by adding multiple trimethylsilyl groups to functional groups on both psilocyn and psilocybin; the trimethylchlorosilane is a catalyst for the reaction(s). The derivatized psilocybin is stable and will not dephosphorylate under GC inlet conditions.



Psilocybin
$C_{12}H_{17}N_2O_4P$
MW: 284.25

Derivatized Psilocybin
$C_{21}H_{41}N_2O_4PSi_3$
Mol. Wt.: 500.79

Figure 2: Derivatization of Psilocybin



Psilocyn
$C_{12}H_{16}N_2O$
MW: 204.27

Derivatized Psilocyn
$C_{18}H_{32}N_2OSi_2$
MW: 348.63

Figure 3: Derivatization of Psilocyn

Associated Protocol(s):

DR-30 Gas Chromatography Analysis (Flame Ionization Detector)
DR-70 Gas Chromatograph / Mass Spectrometer Methods for Drug Analysis

Specimen Required (Sample Handling and Preservation):

The sample size used for analysis may vary from residue to approximately 50 milligrams depending on the size of the submitted exhibit. Fresh or damp suspected mushroom material is to be dried before chemical examination. The portion removed for analysis should be representative of the entire exhibit. Per Oklahoma State Statute §63-2-204.C any material, compound, mixture, or preparation which contains any quantity of psilocybin or psilocyn, the whole sample shall be

considered as controlled. As a result of this allowance by state law, this protocol should not be considered a sampling plan.

Analysis will not be performed on suspected mushroom spores.

Reagents:

1. Thin Layer Chromatography method
    a. Methanol
    b. n-Butanol
    c. Deionized Water
    d. Acetic Acid
    e. Fast blue BB salt
    f. Marquis
    g. Ethanol
    h. P-Dimethylaminobenzaldeyde

2. Derivatization method
    a. Methanol
    b. Bis(trimethylsilyl)trifluoroacetamide (BSTFA) with 1% or 10% trimethylchlorosilane

Standards:

1. Psilocyn
2. Psilocybin

Controls:

1. Thin Layer Chromatography Method:
   Psilocyn and psilocybin standards are used as positive controls.
   Methanol is used as a negative control.

2. Derivatization Method:
   Derivatized psilocyn, psilocybin, or both are used as positive controls.

   The blanks/negative controls required must receive the same treatment as the sample.
    a. When extracting the sample in methanol only, a methanol blank is required.
    b. If the sample is extracted in methanol, dried and then derivatized with BSTFA, a blank in which methanol has been dried down and BSTFA has been added then heated to 140°C for approximately 15 minutes is required.
    c. If the glacial acetic acid extraction is used for the sample extraction, then a blank in which isopropyl (or reagent) alcohol & glacial acetic acid must be dried down, then methanol or BSTFA (whichever is used) will be added and appropriate steps followed.

Apparatus and Materials:

1. Thin Layer Chromatography Method:
    a. Thin Layer Chromatography Plates (250 Micron Silica Gel G or Silica Gel GHLF)
    b. Developing Tank
    c. Disposable culture tubes
    d. Capillary tubes

2. Derivatization Method:
    a. GC oven
    b. Disposable culture tubes

Individual Steps of Protocol:

1. **Preparation of Reagents:**

TLC Method

1. TLC Developing Reagent
    a. 50 mL of n-Butanol, 25 mL of deionized water, and 25 mL of acetic acid.
    b. Combine the ingredients and shake or stir until they are thoroughly mixed.
    Recommended to store in the refrigerator.

2. TLC Visualization Reagent
    a. Combine 1 gram Fast Blue BB Salt with 100 mL of distilled water
    b. Combine the ingredients and shake or stir until they are thoroughly mixed.
    Caution: Fast Blue BB Salt is a suspected carcinogen.

3. Ehrlich's Reagent (TLC Visualization Reagent)
    0.5 gram p-Dimethylaminobenzaldehyde
    Caution: p-dimethylaminobenzaldehyde is a suspected carcinogen.
    12.5 mL Ethanol
    12.5 mL concentrated Hydrochloric Acid

    Preparation of Ehrlich's Reagent:
        a. Add p-dimethylaminobenzaldehyde to ethanol
        b. Add hydrochloric acid to solution.
    Recommended to store in the refrigerator.

Derivatization Method

1. BSTFA – no reagent preparation is needed.

2. **Preparation of Sample**

Methanol Extraction

1. Place dried and pulverized mushroom sample into a minimal volume of methanol.
2. Let soak approximately 24 hours minimizing light exposure.
3. Separate extract from plant material.
4. Conduct GC and GC/MS analysis per DR-30 and DR-70.

5. If psilocyn or psilocybin is indicated in the extract, continue with TLC or derivatization. If psilocyn or psilocybin is not indicated, derivatize the sample.

3. **Analysis of Sample**

Thin Layer Chromatography Method (TLC)

TLC compares an unknown sample with the known psilocyn, psilocybin, or both standards. This examination is to be performed in a fume hood.

1. Spot a small portion of the extract at the origin of a TLC plate with a capillary tube.
2. Spot a psilocyn and psilocybin standard at the origin on the same TLC plate next to or near the question sample for purposes of comparison.
3. Spot a solvent blank on the TLC plate.
4. Develop the plate using the TLC developing reagent in a developing chamber.
5. Allow the plate to dry. The spot may be visible to the naked eye.
6. Spray the TLC plate using a visualizing agent.
7. A positive test for a particular compound is indicated by the sample developing a spot traveling equal distance and turning the same color as the comparison standard.
8. If needed, acidic vapors can be used to help visualize the spots.
9. Record results of the test in the Matrix. The results of the sample, controls and the number of TLC plates used must be recorded. *A record of a negative sample is not a rejection of a test result.*
   a. If TLC results are rejected due to QA/QC issues, the reason and date will be recorded in the Rejected Data panel. The assigned analyst is the only individual that can add information in the matrix. The assigned analyst will be the only individual rejecting the data; therefore, documenting the analyst's name in the "Notes" or Rejected Data panel is not required.

10. When testing using TLC:

a. If testing a single sample, record the date in the "Date of Analysis" on the Matrix Panel of the suspected sample.

b. If testing multiple samples and all testing occurs on the same date, record the date in the "Date of Analysis" and the number of TLC plates used in the "Notes" section of the first suspected sample. *Individual test and QA/QC results must still be recorded in the individual item panels.*

Example

Notes:
Items 1A-1H, TLC (2 plates)

c. If testing multiple samples and testing occurs on multiple dates: record the dates, item numbers and the number of TLC plates used in the "Notes" section of the first suspected sample. *The multiple date information only needs to be recorded one time in a case, i.e. in the Notes of the first item tested using TLC. Individual test and QA/QC results must still be recorded in the individual item panels.*

Example

Notes:
07-24-17, Items 1A-1H, TLC (1 plate)
07-25-17, Items 1I-1T, TLC (2 plates)
07-26-17, Items 2A-2D, TLC (1 plate)

Derivatization Method

1. Evaporate the methanol extract to dryness. Do not apply heat.
2. Add 0.5 mL or less of BSTFA to the dried extract.
3. Heat the mixture at 140°C for approximately 15 minutes.
4. Analyze the mixture with GC and GC/MS.
5. Run the derivatized psilocyn, psilocybin, or both as needed on the GC.
6. The requirements in DR-30 must be met.

Protocol Notes:

1. The Safety Data Sheets for the chemicals listed in this protocol shall be reviewed before beginning the procedure. Special care should be taken while using the Fast Blue BB Salt. (It is a suspected carcinogen.) BSTFA can cause eye and skin burns.

2. When utilizing the derivatization method, use a metal crimp capsule when heating the sample. Be careful when heating the BSTFA/sample mixture and ensure that the vial cannot tip over while in the oven.

3. Psilocyn and psilocybin may be light sensitive. Minimize light exposure during the extraction process and during storage.

4. In situations where there is any reason to believe additional controlled substances may be present, the analyst should continue their analysis using GC and GCMS.

5. The psilocybin, psilocyn and BSTFA lot numbers must be recorded in the casefile; the lot numbers may be recorded in the BEAST Matrix or on the GC or GC/MS printouts. If an analyst records the lot numbers in both places (Matrix and printouts), it is up to the analyst to ensure the lot numbers match.

6. For an identification, the requirements in DR-70 must be met.

Recommended Report Wording/Interpretation of Test Data:

1. When the GC/MS examination indicates psilocyn, and either TLC or derivatization test is positive for psilocyn, report as:

   Item #: Psilocyn, Schedule I.

2. When the GC/MS examination indicates psilocybin, and either TLC or derivatization test is positive for psilocybin, report as:

   Item #: Psilocybin, Schedule I.

3. When the GC/MS examination indicates psilocyn or psilocybin, and either TLC or derivatization test is positive for both psilocyn and psilocybin, report as:

   Item #: Psilocyn, Schedule I and Psilocybin, Schedule I.

4. When results are negative or indicate another controlled substance, report according to DR-30 and DR-70 protocol, as appropriate.

References:

1. Cole, M. The Analysis of Controlled Substances, John Wiley and Sons Ltd, 2003, Chapter 8
2. Anastos, N.; et al. The Determination of Psilocyn and Psilocybin in Hallucinogenic Mushrooms by HPLC Utilizing a Duel Reagent Acidic Reagent Acidic Potassium Permanganate and Tris(2,2'-bipyridyl)ruthenium(II) Chemiluminescence Detection system, J. Forensic Sci, Jan 2006, Volume 51, No 1, p 45-51
3. Gross, S. Detecting Psychoactive Drugs in the Developmental Stages of Mushrooms, J. Forensic Sci, May 2000, Volume 45, No 3, p 527-537
4. Tryptamines, Vol II, CLIC 11th annual training seminar, page 20, 2003.

DR-50 PDF Examination Documentation Procedure

Purpose:

This protocol will outline how the PDF document is generated, stored, transferred, and finally attached into the BEAST image vault and it will discuss the implemented security and tracking features that ensure the integrity of the PDF documents.

There are four major stages to the PDF process:

1. Utilize OSBI Chemstation macros to:
   a. Generate instrumental data reports with case information embedded within the Other.ini file, which is located in the Data file, and to appear on the printed report.
   b. Create an .txt file that will assign a unique number, a Validation Code, to each sample. The Validation Code .txt file contains the validation number, case number, item information, and analyst name, the Validation Code appears on the printed report and each file is archived on the instrument computer and in a secure folder on an OSBI server.
   c. Archive the sequence logs to a secure folder on an OSBI server.
2. The OSBI Chemstation generated instrumental data reports will be sent to a PDF printer which will put the analyst's name in the PDF document. The case number for the report will be used in the naming of the PDF document. Each analyst assigned to the instrument has a specific folder for their documents. The PDF document will then be placed into the analyst specific folder.
3. The FTIR generated data will include the name of the analyst performing the analysis.
4. The instrumental data reports will be transferred to the analysts through the OSBI network. The analyst will place their data reports into a folder where the PDF documents will be stored until they are uploaded into the BEAST Image Vault.

Associated Protocol(s):

DR-30 Gas Chromatography Analysis (Flame Ionization Detector)
DR-70 Gas Chromatograph / Mass Spectrometer Methods for Drug Analysis
DR-60 Drug Analysis by FTIR
DR-75 Gas Chromatograph Infrared Detector Methods for Drug Analysis

PDF Procedure:

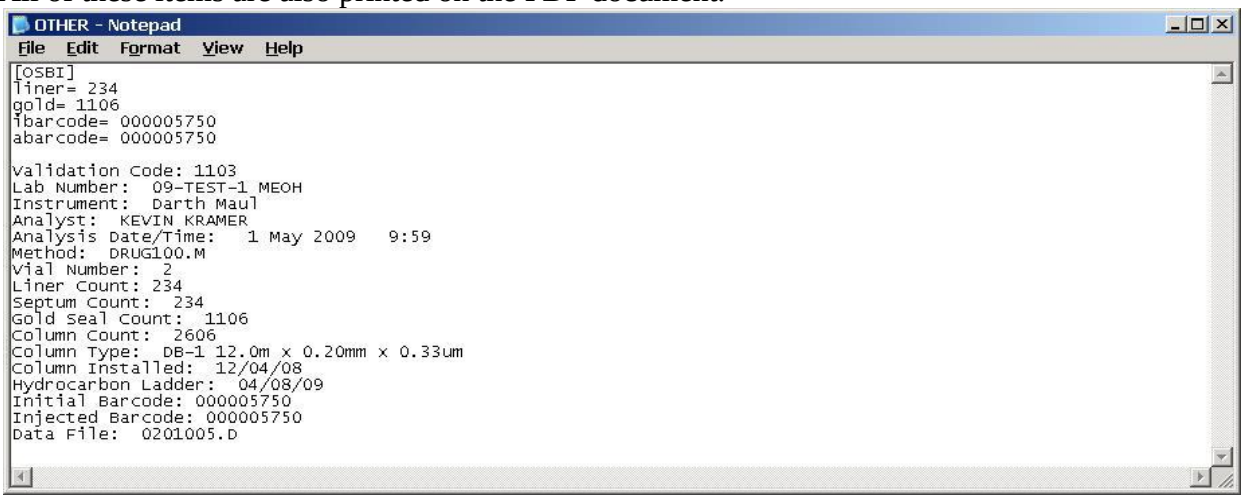
All OSBI Drug Lab instruments and computers are kept in secure limited access areas. Each instrument set, one Agilent Gas Chromatograph and one Agilent Gas Chromatograph / Mass Spectrometer or a Thermo Fourier Transform Infrared Spectrometer (FTIR) is controlled by a computer and each analyst has a specific folder for instrumental examination documentation on that computer.

The OSBI has created a file within the Data file/folder for each sample called Other.ini. Within this file several key pieces of information are kept for each sample.

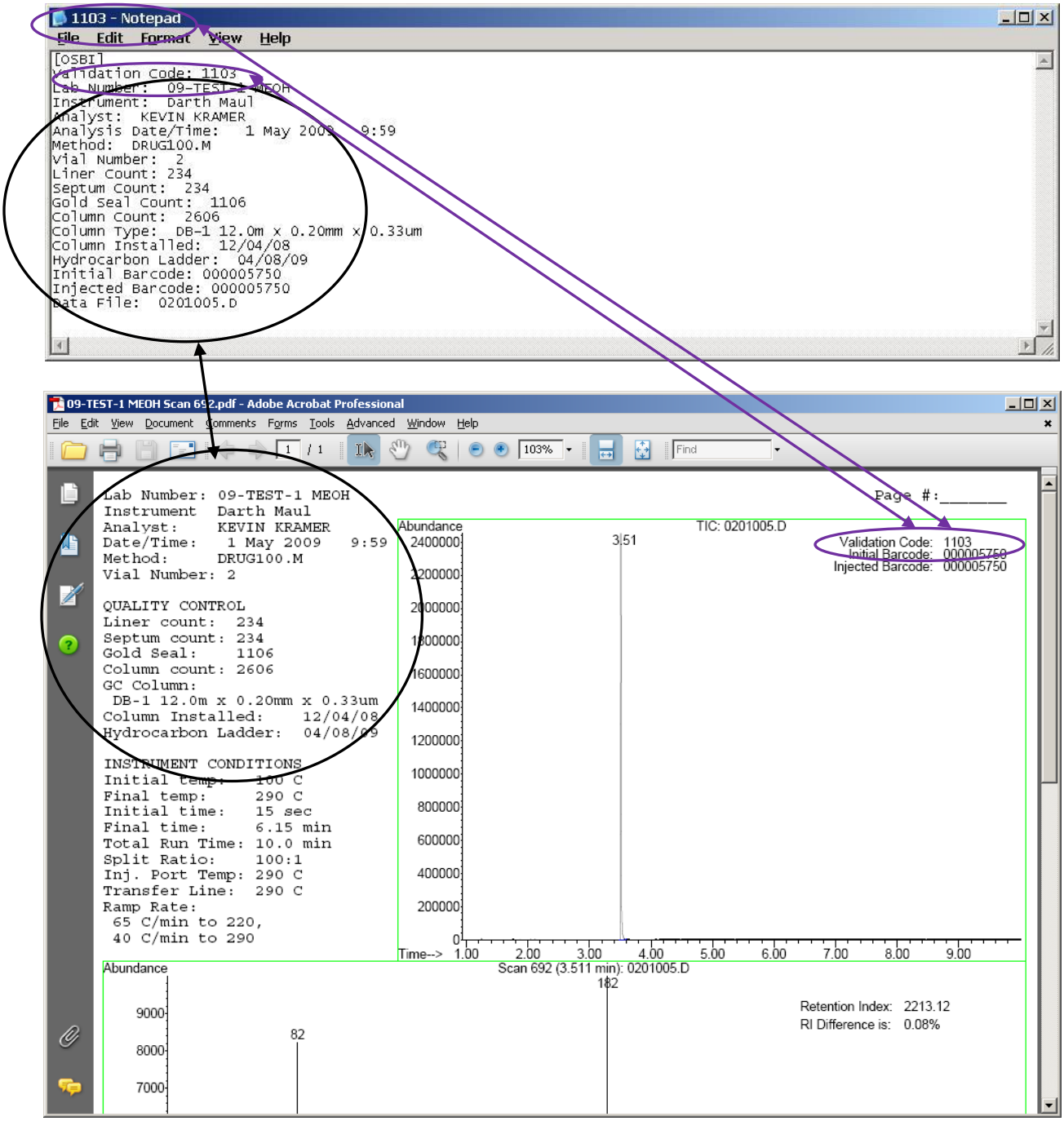
Included are:

- liner and gold seal counts
- initial and actual/injected barcodes found on the sample vial
- validation code
- lab number, sample number and solvent system used for extraction
- instrument used for analysis
- analyst name
- date and time of analysis
- method used to analyze sample
- vial number in tray
- column information
- date hydrocarbon analyzed
- data file name for that sample

All of these items are also printed on the PDF document.



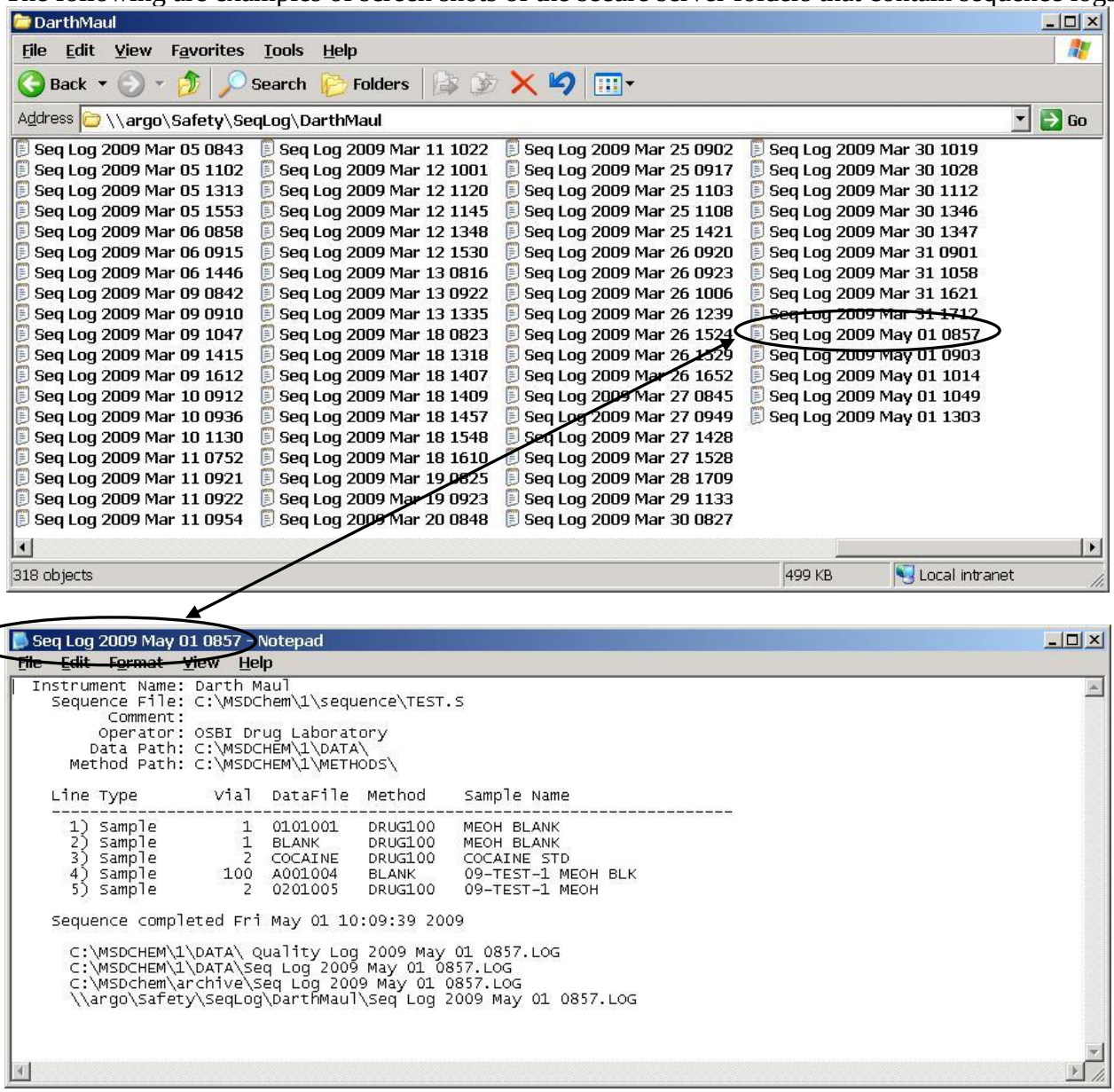
A Validation Code system has been implemented to generate and assign a unique number to each sample analyzed on an instrument. Each sample will have a Validation Code printed on the PDF documents and the number will be used to name the .txt file. The same information found in the Other.ini file can be found in the Validation Code.txt file.



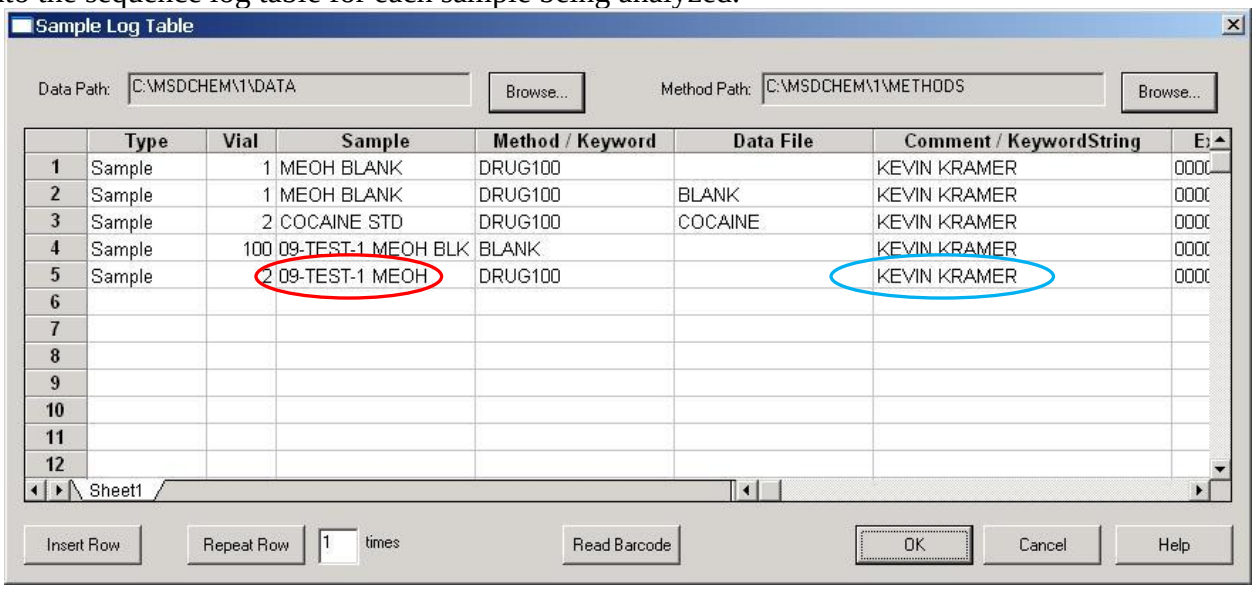
The Agilent macros have been modified to archive the Validation Code files and the Sequence Logs to the instrument computer. The Validation Code file, the Other.ini file and the PDF files are all created by the computer for each sample simultaneously and these three files are all stored in different locations. If there was reason to believe a person was tampering with documents, these files

could be compared to the document in question. This comparison will verify whether the document in question contains the original information generated at the time of analysis.

The following are examples of screen shots of the secure server folders that contain sequence logs.



The process begins with the preparation and loading of samples onto the instruments for analysis. When an analyst creates a sequence log, they are required to enter the case number and their name into the sequence log table for each sample being analyzed.

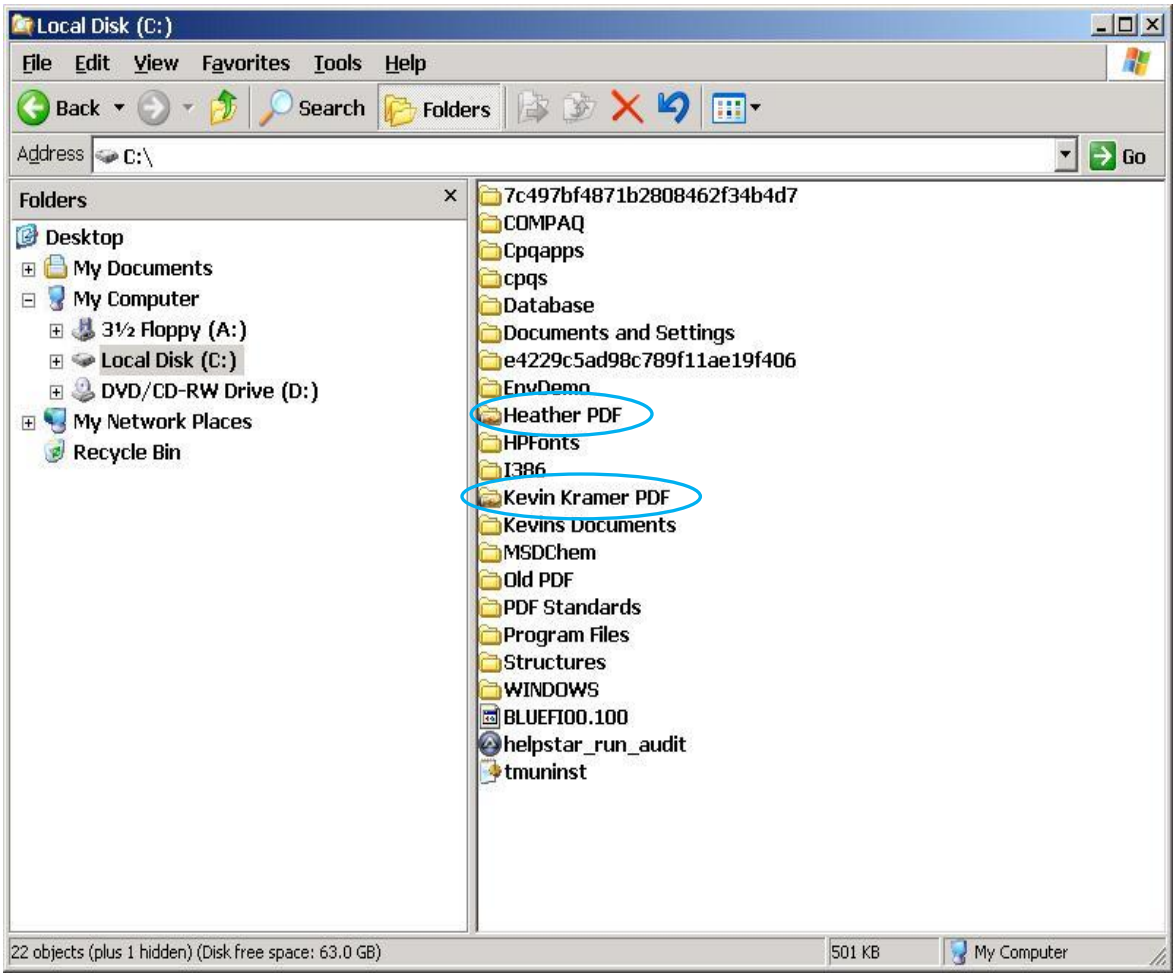


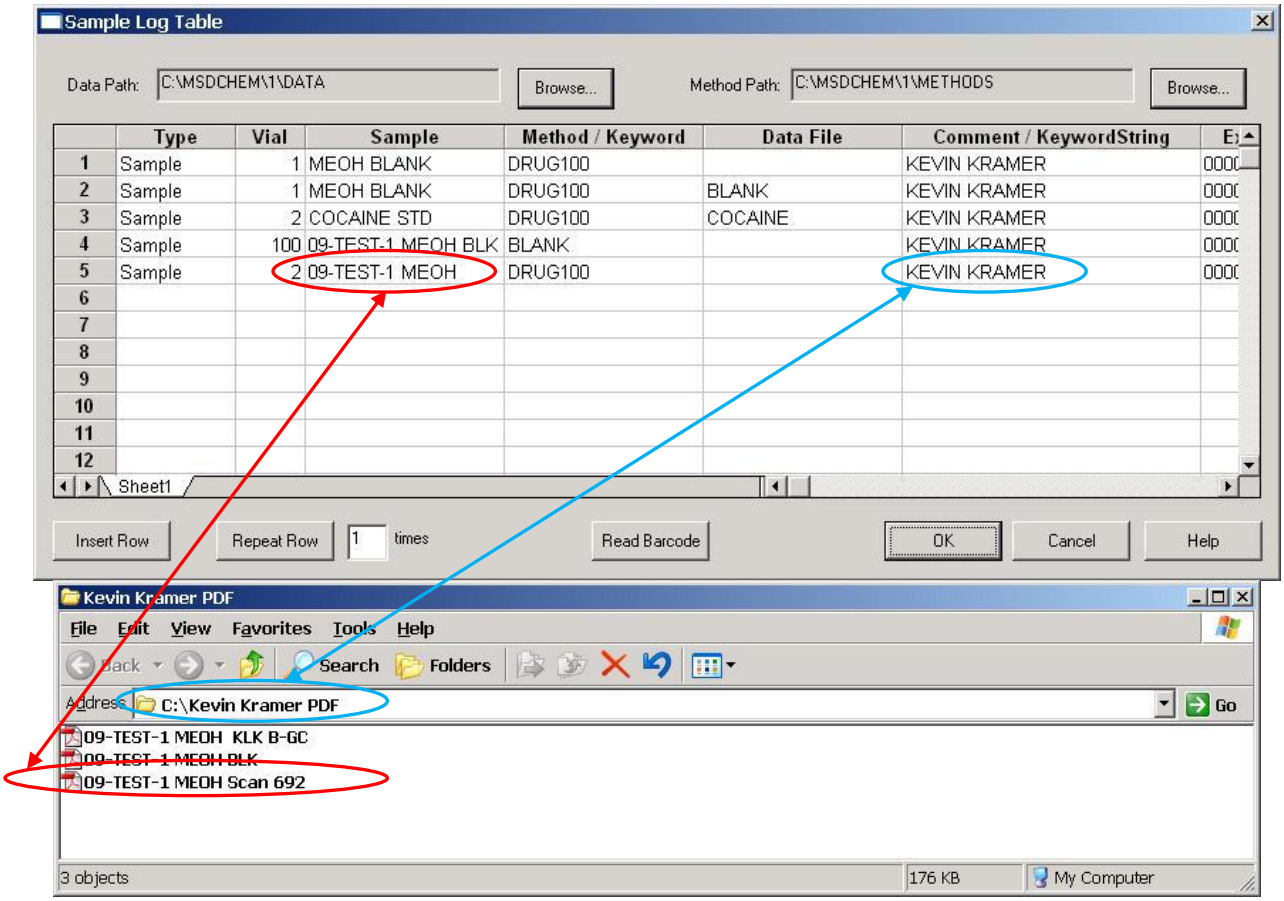
The case number, item number, and extraction for each sample is placed into the Sample field. The information in the Sample field is used to generate the PDF report name for each sample analyzed. The information in this field is also placed into the Other.ini file located in the Data file and the Validation Code file generated for each sample.

Entering a name into the Comment/KeywordString field sets a variable in the OSBI Chemstation data editing macros. This variable has three functions: 1. place the analyst's name in the Other.ini file located in the Data file and in the Validation Code file for the sample; 2. generate a PDF document with the analyst's name printed on it; 3. direct the PDF document to be placed into the analyst specific folder located on the computer.

The folders for each analyst are set up on the hard drive of the instrument computer. For this illustration the folders are Kevin Kramer PDF and Heather PDF. The name of this folder can contain the analyst's first name or first and last name. The analyst's name entered into the Comment/KeywordString in the sequence log will direct the PDF document to the analyst's folder. If the name in the sequence log is Kevin Kramer, then the PDF will go to the Kevin Kramer PDF folder. If the name is Heather, then the PDF will go to the Heather PDF folder.

The following are screen shots to illustrate where the analyst specific folders are located, the sequence log table for a specific sample and the analyst folder with the sample's PDF document.





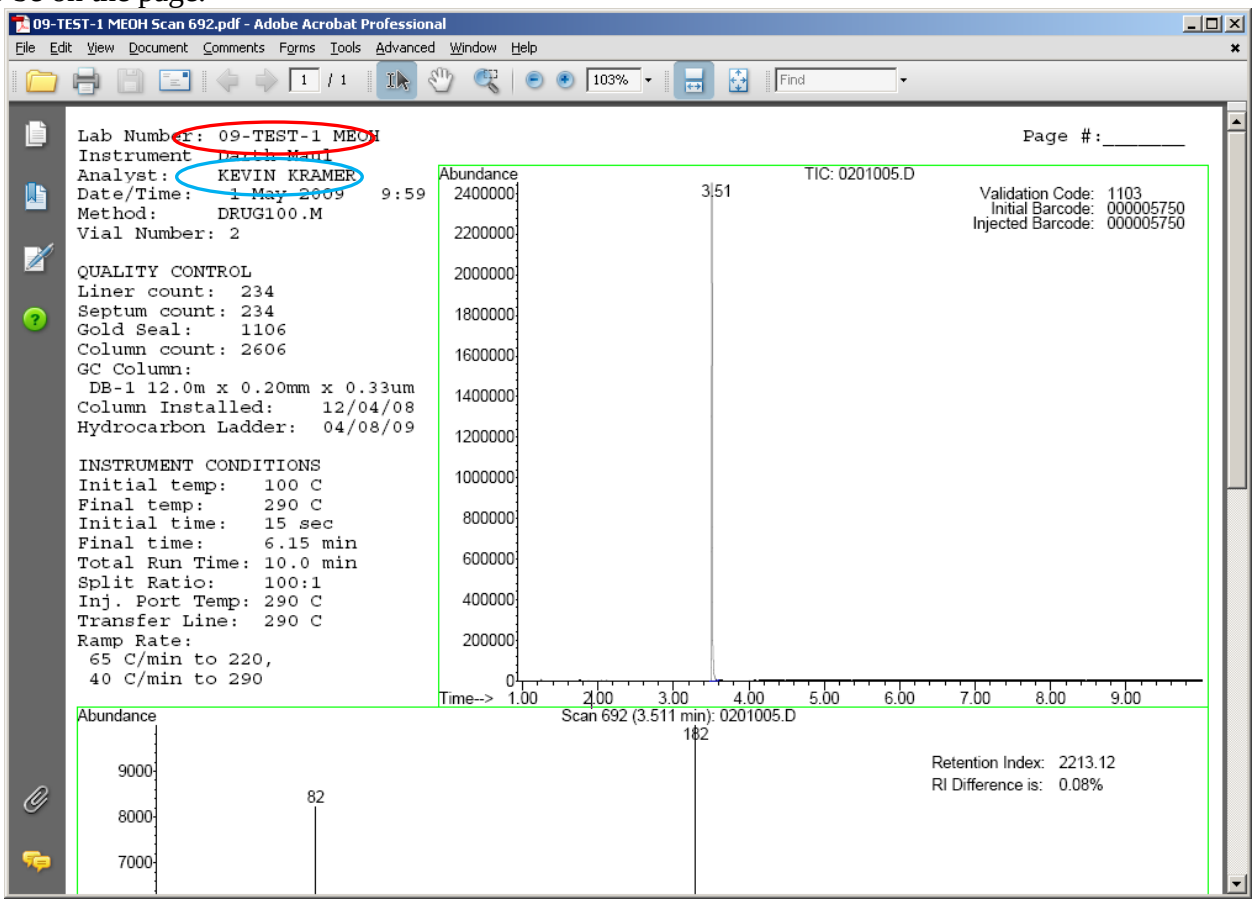
Note the sequence log table with the sample name and analyst name, Kevin Kramer, and the same information appearing in the title of the PDF document in Kevin Kramer's PDF folder. The title of the PDF document is the sample name from the sequence.

The folders Kevin Kramer PDF and Heather PDF are shared through the OSBI network. The permissions to access these folders are limited to the specific analyst. Only Kevin Kramer has permission to access the Kevin Kramer PDF folder from any computer other than the instrument computer. Specifically, the computers that will be used are either the analyst laboratory computer or the office computer; however, it does not have to be limited to those computers. Authentication at the Windows log in by giving the proper user's name and OSBI network password is required.

Once the sample has run on the (GC, GC/MS or FTIR) instrument and the PDF created, the analyst transfers the data from the instrument computer to their computer for review, approval, and uploading to the BEAST. The data is transferred through the OSBI network. This requires the analyst to use an OSBI computer, on which they have been authenticated as the user by entering their user's name and network password.

The analyst does not do a final review of the PDF documents until they are securely stored on their individual computer. At this time the analyst will confirm their name or initials are on the PDF document. If an analyst is using data (i.e. FTIR Val-Pro, GC drug standard, or GC/IRD QA/QC)

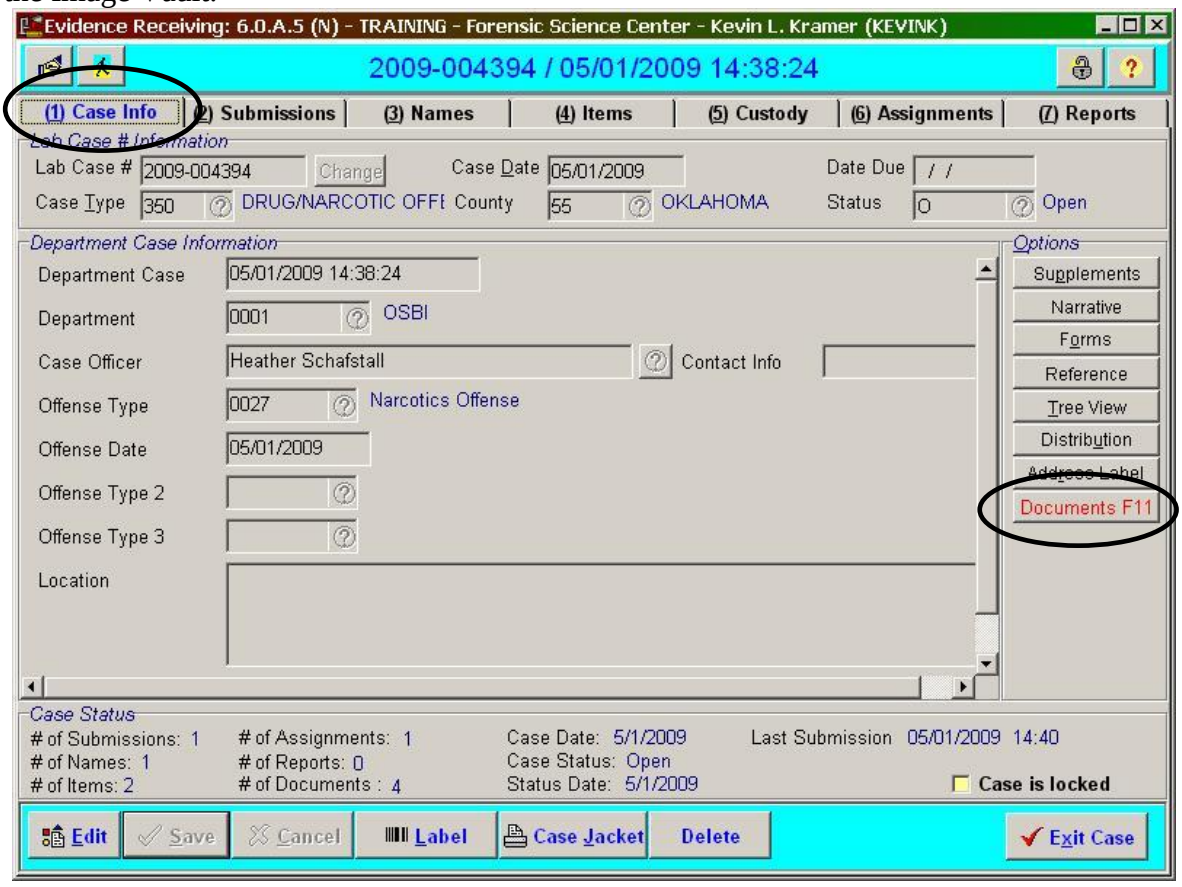
created by another analyst, only the name of the analyst conducting that step of analysis is required to be on the page.



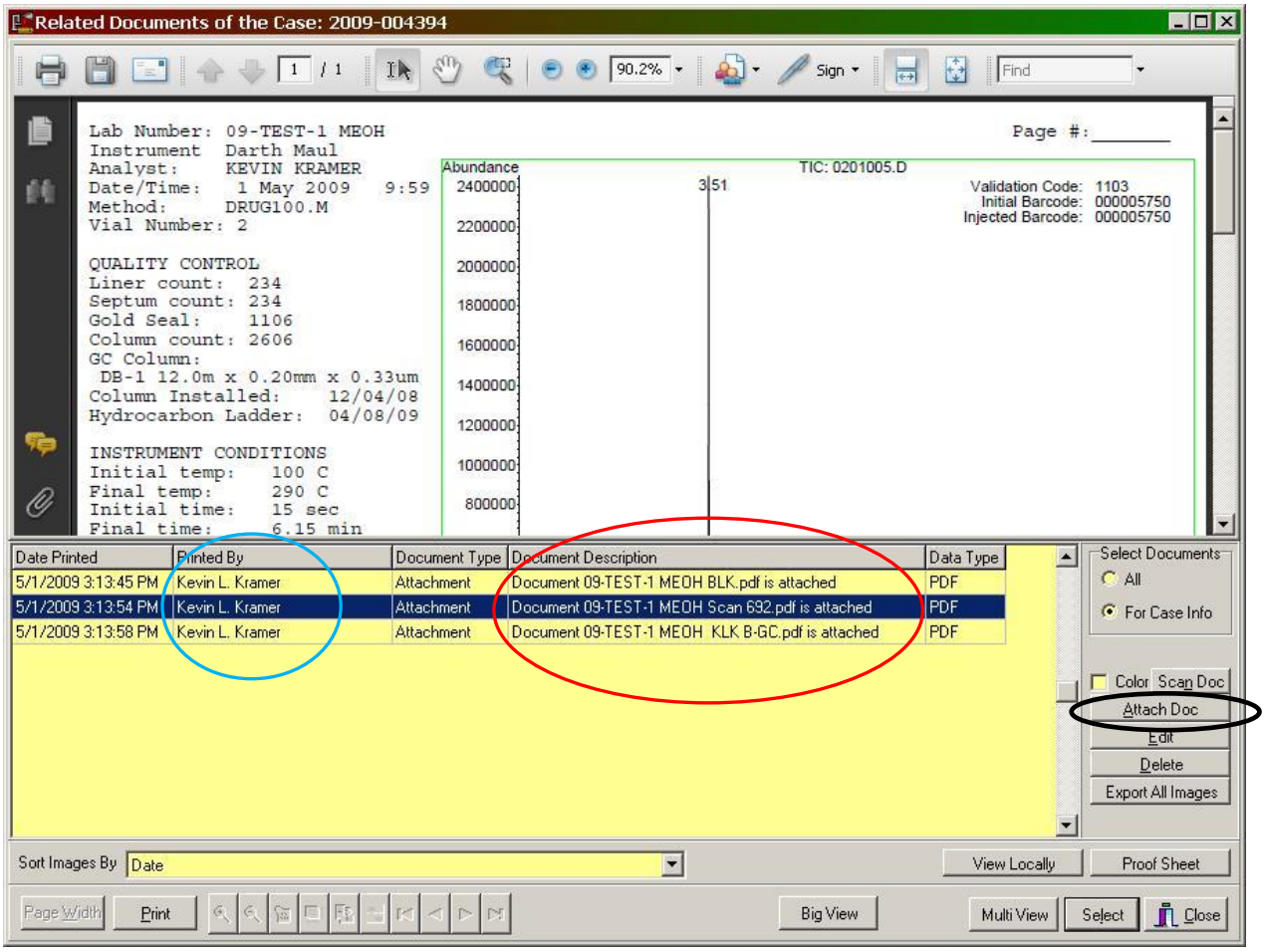
When the analyst has completed reviewing all the PDF documents for an item, they will have the option of directly uploading each document into the Beast or merging the documents into one document using a PDF software.

To upload documents into the Beast Image Vault, the analyst must first log into the Beast using their username and password.

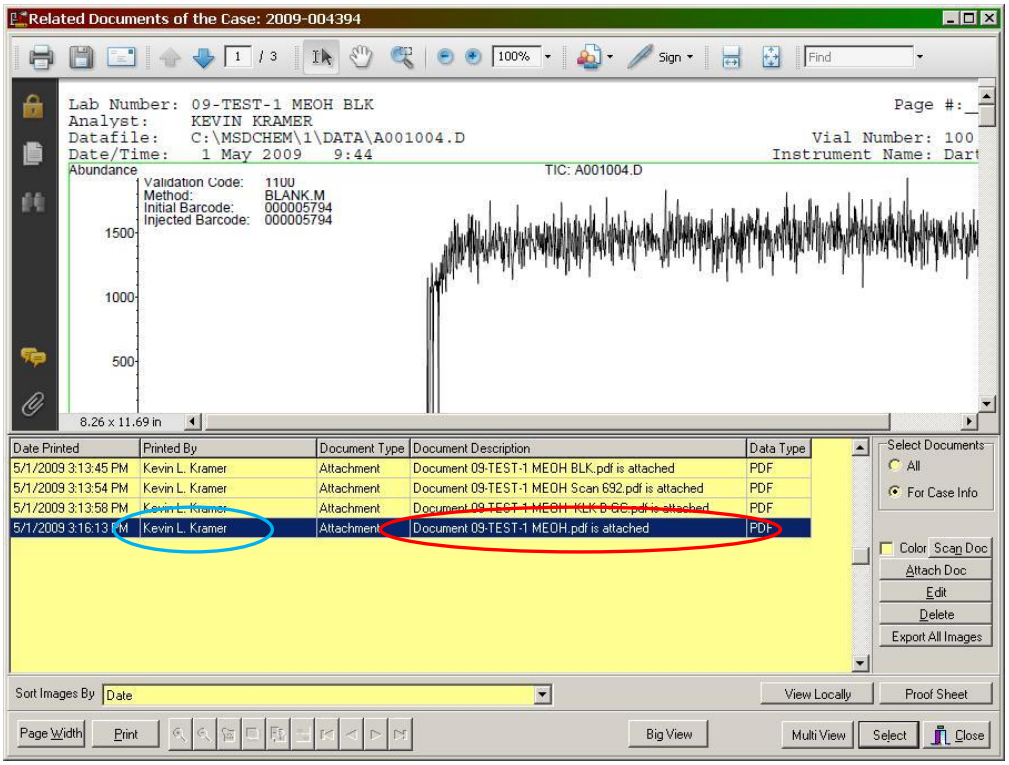
After logging into the Beast the proper case file must be retrieved and the Case Info tab or Matrix accessed. The Documents F11 button will take the analyst to the next screen to upload documents into the Image Vault.



The Attach Doc button will open a window that allows the analyst to choose the document to be uploaded. The documents will be uploaded from the folder found on the analyst's computer. The image viewer shows which documents have been previously uploaded and by whom for the individual case.

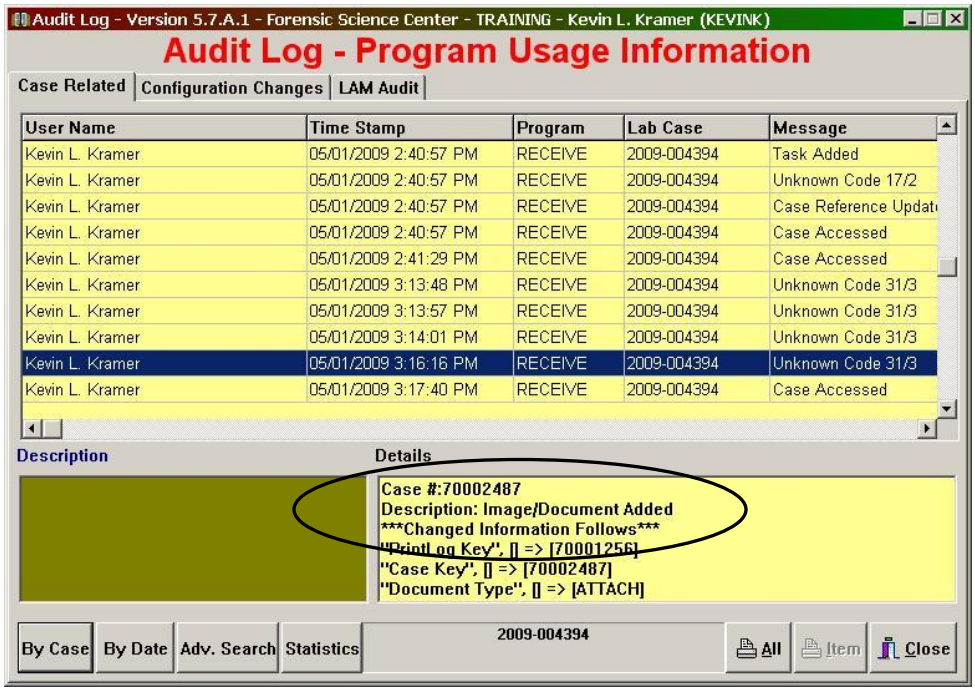


Here the three individual documents have been uploaded for this case by the analyst.

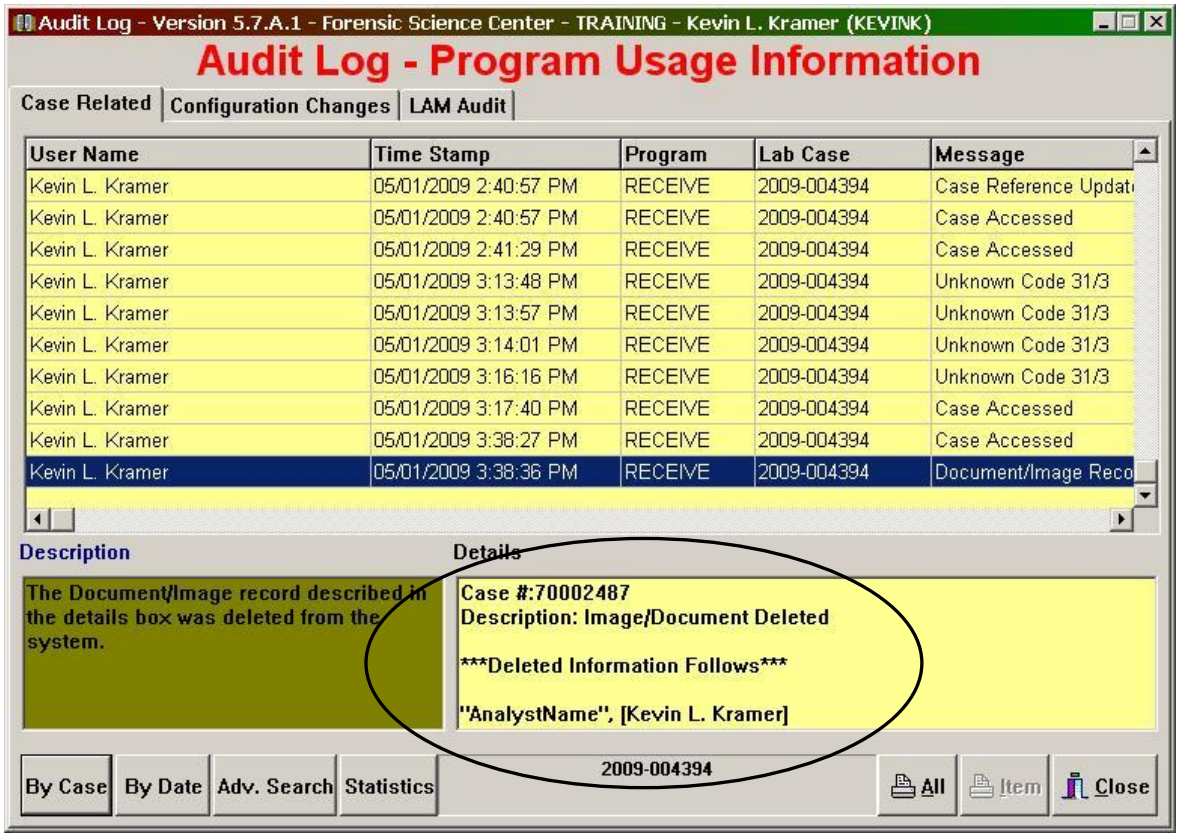


Here is an example of the merged document uploaded into the Image Vault.

When the documents are uploaded into the Beast the Audit Log tracks everything that occurs in the case file.



Here is an example of the Audit Log tracking a document being uploaded into the Beast.



If a document is deleted from a case file, this is also documented in the Audit Log.

Whenever a change is made, a document uploaded or the case is accessed, the Audit Log tracks:

- User Name – full name associated with the analyst's Beast username
- Time Stamp – time of action
- Program – which Beast program used
- Lab Case – case number
- Message – action was taken
  - Case Reference Update
  - Case Accessed
  - Unknown Code 31/3 (document uploaded)
  - Document/Image Record Deleted
- OS Computer – OSBI computer used to perform the action
- OS User Name – analyst logged into computer
- Details – detail of the action taken

Following all of the steps outlined in this protocol will allow the OSBI Drug Chemists to create a paperless case file using data reports created in PDF format and uploaded into the Beast Image Vault following all OSBI policy and procedures.

The document security will be ensured by:

- Case number and analyst name/initials in the sequence log table.
- Date the document was originally generated, case number, analyst name/initials and sample information in the Other.ini file and Validation Code file for each item.
- The Other.ini, the Validation Code file and PDF document are all generated for a sample by the computer simultaneously, but are sent and stored to three different locations. This will allow comparison of data if a document's authenticity is ever questioned.
- Validation Code files and Sequence Log files are archived to the instrument computer and uploaded to the technical manager's computer for archiving.
- Analyst name printed on the instrumental data report generated by PDF printer. Case number used to generate the PDF document name.
- PDF document placed into folder on instrument computer only accessible by the specific analyst from any computer other than the instrument computer.
- Document transfer via password protected OSBI network. This requires the analyst to use an OSBI computer, on which they have been authenticated as the user by entering their user name and network password.
- Placement of documents on to analyst's individual computer.
- To upload documents into the Beast Image Vault, the analyst must log into the Beast using a username and password.
- Once uploaded into the Beast, all activity to a case file is tracked using the Audit Log.

Corrections to PDF Documents:

Corrections made to PDF documents are to be made using a red font. The "mistake" must be lined out with the corrected value, the date and initials near the lined-out data.

If the error is on multiple pages, the error will need to be corrected on each page. Making the correction on the first page only is not acceptable. Verification of a barcode does not have to be documented on each page; documentation on the first page only is acceptable.

DR-60 Drug Analysis by FTIR

Purpose:

Infrared spectroscopy can be used to identify most organic compounds based on absorption of radiation over the mid-infrared range from 4000 to 400 wave numbers. In most instances, the mid-infrared spectrum of an organic compound provides a unique fingerprint, which is readily distinguished from the absorption patterns of all other compounds; only optical isomers absorb in exactly the same way[1]. In order for a molecule to absorb infrared radiation, a molecule must undergo a net change in dipole moment as a consequence of its vibrational or rotational motion[1]. The frequency that causes a change in dipole is specific for the vibrational motions in each bond and function group within all organic compounds. Only those frequencies of infrared radiation which match the natural vibrational frequencies of the molecule in question will be absorbed, and the energy absorbed will serve to increase the amplitude of the vibrational motions of the bonds in the molecule[2]. The IR radiation, which is produced from a mid-IR source, is directed into an interferometer. In the interferometer the beam is split and recombined producing a single beam containing all frequencies of mid-IR light. The light then interacts with the sample where selective absorption takes place, and then strikes the detector producing an electrical signal. The resulting interferogram is then converted into an infrared spectrum through a mathematical formula called Fourier Transform[1]. This spectrum displays absorbance of radiation vs. the wavelength of radiation, which is absorbed. This infrared spectrum can be compared with that of a known standard for a conclusive identification.

Associated Protocols:

DR-10 Color Test: Marquis
DR-11 Color Test: Cobalt Thiocyanate
DR-13 Color Test: Bates Examination
DR-30 Gas Chromatography Analysis

Specimen(s) Required (Sample handling and preservation):

Sample size can vary from a residue to approximately twenty milligrams of sample. In order to make a conclusive identification, an extraction may be necessary to remove diluents.

Reagents:

1. Solvents to perform suitable extraction
2. Methanol

Note: All reagents are reagent grade or better.

Standards:

All identifications will be made by comparing the question sample to a known documented standard spectrum acquired using the same technique (i.e. transmittance or reflectance). The standard spectra must be available in the case file for future peer review and possible legal review. Such standards are to be purchased from Alltech, Fluka, Sigma, Supelco, U.S. Pharmacopeia, or other reputable manufacturers. Spectra from literary references, computer references (i.e. Georgia Bureau or Thermo), and samples of unknown origin or from cases are not suitable for making a conclusive comparison for reporting. Spectra retrieved from a user-generated computer library are

recommended, but high-resolution spectra must be used for comparison and print out. Libraries generated for this purpose are to be backed up.

The following information is to be readily and permanently available for each standard:

- Name of standard
- Manufacturer or source
- Date spectra was produced
- Lot number (if used by manufacturer or source)
- Analyst who produced the spectra and date produced
- Literary or other reference to which the spectra was compared, analyst making comparison and date of comparison

Controls:

Each month, the bench will be checked with the ValPro System Qualification using the validation wheel equipped with polystyrene standards. This report will be archived.

Each day before casework is performed, if an ATR accessory is to be used (or any other accessory), a polystyrene standard will be run using the macro entitled, Macro OSBI. This report will be archived and a copy placed in each case file.

Each day casework is to be performed, if an accessory is not to be used, the spectra of a polystyrene film will be obtained via transmission and compared to a literature spectrum as a quality control. This scan will be archived and a copy placed in each case file.

Apparatus and Materials:

1. Thermo 6700 Infrared Spectrometer equipped with OMNIC software and ValPro System Qualification software.
2. Thermo SmartOrbit Attenuated Total Reflectance (ATR) accessory with diamond crystal.
3. Nicolet iS5 Infrared Spectrometer equipped with OMNIC software and ValPro System Qualification software.
4. Thermo iD7 Attenuated Total Reflectance (ATR) accessory with diamond crystal.
5. Mortar and Pestle
6. Spatula
7. Kim Wipes

Individual Steps of Protocol:

1. FTIR Instrument Parameters:
   Experiment setup
   Collect:
   a. Number of Scans: 32 or greater
   b. Resolution (cm-1): 4
   c. Final format: Absorbance or % Transmission
   d. Correction: None
   e. Aperture: 100
   f. Gain: Autogain
   (Instrument will determine and print the Gain used on the sample printout)

2. Sample preparation:
   Decide if the spectra will be collected using transmission or reflectance. The instrument will automatically detect if an accessory is in the sample compartment and load the experiment setup for that accessory.

   Reflectance – Using ATR Accessory
   a. Obtain sample of unknown substance.
   b. Perform a color test, literary reference, or run on the GC.
   c. Perform suitable extraction, if needed, to separate unknown from diluents.
   d. Clean the ATR accessory and arm with a suitable solvent (i.e. Methanol, Acetone, DI Water).
   e. Collect a background spectrum on the spectrometer with nothing making contact with the crystal. A background must be taken before each sample.
   f. Place the sample on the ATR accessory, lower the arm, and collect the sample spectrum.
   g. Sample can be recovered for additional testing, if needed.

3. Bench Alignment using the ATR Accessory:
   a. Ensure the tower is up.
   b. Open the OMNIC software.
   c. Select “Collect.”
   d. Select “Experiment Setup.”
   e. Click on “Diagnostic,” then “Align.”
   f. Once the alignment is complete, click “Save” and then “OK.”

4. Comparison and Acceptable Criteria for Identification with Infrared Spectroscopy
   Directly compare the question sample spectrum to a standard spectrum. Similar peak shape and position (in wavenumbers) along with no missing peaks or added significant peaks indicating a mixture will be considered acceptable identification.

Frequency & Tolerance of Controls & Corrective Action To Be Taken If Tolerances Are Exceeded:

If a ValPro System Qualification of the bench fails, the instrument is to be removed from service and the following actions taken:

1. Notify the Supervisor and Technical Manager regardless of diagnosis.
2. Determine performance problem, repair, verify instrument performance, and document in the maintenance log.
3. Return the instrument to service once a satisfactory ValPro System Qualification of the bench is obtained.

If an unsatisfactory polystyrene spectrum from an ATR accessory is obtained, the following actions will be taken.

1. Obtain a 2nd polystyrene spectrum from the ATR accessory, if this remains unsatisfactory then see the next step.
2. Perform a ValPro System Qualification of the bench. If this fails, remove the instrument from service, notify the Supervisor and Technical Manager.
3. Determine performance problem, repair, verify instrument performance, and document in the maintenance log.
4. Return the instrument to service once a satisfactory ValPro System Qualification of the bench is obtained and a satisfactory polystyrene spectrum is obtained from the ATR accessory.

Expected Values: Values Requiring Special Notification:

Any item listed in the preceding section requiring that an instrument be removed from service will be documented in the maintenance log and reported immediately to the Supervisor and Technical Manager.

Protocol Notes:

1. Routine maintenance (i.e., monthly ValPro System Qualification of the bench), repairs and bench alignments are to be documented on the instrument maintenance log (DR60-1).

2. If the signal is greater than 100% transmittance, a bench alignment should be performed to return the signal to normal. This will be recorded in the maintenance log. To prevent this issue, be sure to run the sample without delay after the background has been collected. See Individual Steps of Protocol for steps to perform a bench alignment.

3. When using the "Reprocess" macro, be sure to have all data windows closed. Not doing so will cause the incorrect sample information (the title, collection time, and background time) to be printed on the data being reprocessed.

4. When an instrument is removed from service, label the instrument "Not in Service" and document in the maintenance log. When the instrument is returned to service, document the event, relevant repairs and maintenance. Note in the maintenance log that the instrument is returned to service.

Recommended Report Wording/Interpretation of Test Data:

1. The FTIR is normally considered a conclusive examination by itself. However, the Controlled Substance Unit requires that a second test be conducted. When used in conjunction with a positive color test, GC/MS, GC/IRD, literature reference, GC, or thin layer chromatograph, report as follows:

   Item #: Cocaine, Schedule II

2. If no controlled dangerous substance is conclusively identified using FTIR, an analyst cannot report these findings unless they are verified by other means of analysis (i.e. GC and GC/MS).

Associated Form(s):

DR60-1 Maintenance Log

References:

1. Skoog DA, Leary JJ. Principles of Instrumental Analysis, 4th Ed. Saunders College Publishing, 1992: 113, 252-4.
2. Pavia DL, Lampman GRAM, Krix GRAMS. Introduction to Spectroscopy: A guide for students of organic chemistry. Saunders College Publishing, 1979: 14.

DR-70 Gas Chromatograph Mass Spectrometer Methods for Drug Analysis

Purpose:

The dissolved sample is introduced into the inlet of the gas chromatograph portion of the instrument. The inlet temperature is high enough to volatilize most organic substances with a molecular weight: below approximately 600. The helium carrier gas transporting the sample flows into the capillary column from the injector. The capillary column separates the components of the sample based on their solubility in the liquid phase and volatility. Each component of the sample exits the column directly into the mass spectrometer where it is bombarded with electrons (Electron Impact or EI) and fragmented into molecular and fragment ions. Fragmentation patterns are characteristic for each organic compound. The fragments then pass into the mass filter.

A quadrupole mass filter is comprised of four parallel cylindrical electrodes or rods assembled in a precise square array. Diagonal rods are electrically connected and a radio frequency voltage and positive direct current voltage is applied to one pair and a radio frequency voltage with a 180° phase shift and a negative direct current voltage applied to the other pair. A field is set up in the space between the rods that will only allow the passage from one end of the assembly to the other of ions with a particular m/z value. All other ions will undergo oscillation with increasing amplitude until they collide with one of the rods, where they are neutralized and pumped away by the vacuum system. A mass scan can be achieved by increasing both the direct current and radio frequency voltages, which focuses ions on an electron multiplier for detection.

An analysis by gas chromatography mass spectrometer is normally considered a conclusive analysis capable of distinguishing all known organic compounds that can be chromatographed, including diastereomers. One notable exception is ephedrine and pseudoephedrine when analyzed on a nonpolar column such as an HP 1.

Associated Protocols:

DR-30 Gas Chromatography Analysis

Sample required (sample handling and preservation):

A dissolved sample of sufficient strength to give a detailed mass spectrum of the suspected compound, but also of an amount that does not overload the column is utilized. Phenethylamines such as methamphetamine, amphetamine, ephedrine, and pseudoephedrine are to be injected as the free base, whenever possible. Residual samples may be injected as the salt form if there is insufficient sample to extract for further analysis. Unless they are extracted, phenethylamines may not chromatograph well. Other compounds may be injected in the salt form.

Reagents:

1. Reagents as needed for analysis of samples.

Refer to Approved Suppliers list for reagent grades and suppliers.

Standards:

All identifications will be made by comparing the question sample to a known, documented standard spectrum that is to be available in the case file for future peer review and possible legal review. Such standards are to be purchased from Alltech, Sigma, or other reputable manufacturers. Spectra from literary references, computer references, (i.e., NBS, Georgia, TOX) and samples of unknown origin or from cases are not suitable for making a conclusive comparison for reporting. Spectra retrieved from an OSBI-generated computer library are required to make an identification. The following information is to be readily and permanently available for each standard:

- Name of standard
- Manufacturer or source
- Date spectra was produced
- Lot number (if used by manufacturer or source)
- Analyst who produced the spectra
- Literary reference which the spectra was compared to, analyst making the comparison, and date.

Controls:

1. Each day that casework is performed, the first sequence is to contain a solvent blank as a negative control and an approximate one to two milligram/milliliter concentration cocaine standard for a positive control. The solvent blank data file will be named "blank" and the cocaine standard data file will be named "cocaine". When sequences continuously run for an extended period of time, for example 2 or more days, it is not necessary to re-run the positive and negative control. The controls will be documented in each case file. Sequence logs will be digitally archived; therefore, sequences should be allowed to finish for archiving reasons.

2. Solvent and extraction blanks will be used to routinely verify that instrumentation, reagents, and solvents being used for casework are free from contamination. A blank that consists of the same solvent or extraction used in a case will be shot during the case samples and at least every sixth injection (five samples then a blank) within a case. For example, if a case has three samples that are in hexane, then a hexane blank made from the hexane used that day will be shot during the case samples. If a case involves samples that have been extracted with sodium hydroxide and chloroform and samples that are in methanol, then a sodium hydroxide/chloroform extraction blank and methanol blank should be prepared from solvents used that day, and both blanks will be shot during these casework samples. If unknown solvents are analyzed from clandestine laboratories, the solvent used for diluting the sample should be used as the blank extraction. If no dilution occurs, any extraction blank will be acceptable. All extraction blanks should be clearly labeled on the GC/MS scan. Abbreviations such as "extraction blank" or "n/c" are not acceptable. The blank TIC will be kept in the case file.

3. Solvent and extraction blanks will be run at split ratios and run times consistent with analysis runs. The standard blank run has a split ratio of 50:1 and a run time of 10 minutes. This method can be utilized for all Drug50 runs or higher. Lower split ratio runs should utilize the blank run with an equal or lower split ratio. Extended runs should utilize the BlankE methods available. LowM, LSD, Method1, Method2, Meth50, and Meth100 methods should use Blank methods with the equivalent split ratios. All blanks and solvents should be tested to the lowest level that sample analysis occurs for a specific case.

4. Solvent and extraction blanks that have a 58 ion need to be re-analyzed and casework samples following such blanks cannot be used for identification. The analyst will need to troubleshoot to determine the source of the 58 ion.

Apparatus and Supplies:

1. Agilent Gas Chromatograph/Mass Spectrometer.
2. J&W/Agilent DB/HP-1 (12m x 0.20mm x 0.33 um) columns.
   Phenomenex ZB-1 (12m x 0.20mm x 0.33µm) columns.
   Phenomenex ZB-50 (12m x 0.25mm x 0.25µm) columns.
   Restek Rxi-1ms (12m x 0.20mm x 0.33µm) columns.
   The Technical Manager of Drug Analysis may approve other columns for use.
3. Autosampler syringes; five (5) or ten (10) microliters.
   NOTE: *The method must have the correct syringe size listed.*
4. Glass auto sampler vials
5. Glass inserts for vials
6. Caps for auto sampler vials
   NOTE: Do not use polypropylene vial capsule with chloroform or hexane solvent
7. Glass wool
8. Low bleed septa
9. Four millimeter large bore split/splitless injection liners
10. Inlet gold seals and washers

Methods Approved for Analysis:

Methods for drug analysis using the Agilent GCMS are available from the technical manager. These methods are to be utilized by all OSBI drug analysis sections utilizing the Agilent GCMS. Any requests for new methods/macros or changes to existing methods/macros should be submitted in writing to the technical manager of drug analysis for review and approval prior to any changes being made.

The current approved methods are as follows:

Blank:	Blank run utilized for general drug methods with a 50:1 split ratio or higher when required.
Blank0:	Blank run utilized for general drug methods with a 0:1 split ratio or higher when required.
Blank10:	Blank run utilized for general drug methods with a 10:1 split ratio or higher when required.
Blank25:	Blank run utilized for general drug methods with a 25:1 split ratio or higher when required.
BlankE:	Blank run utilized for general extended drug methods with a 50:1split ratio or higher when required.
BlankE10:	Blank run utilized for general extended drug methods with a 10:1 split ratio or higher when required.
BlankE25:	Blank run utilized for general extended drug methods with a 25:1 split ratio or higher when required.

Blank40E25: Blank run utilized for 40-minute extended drug method with a 25:1 split ratio or higher when required.

BlankSL: Blank run utilized for specialized methods Slow50 and Slow100.

Drug0: General drug analysis method, 0 split ratio, 1µl injection volume.

Drug10: General drug analysis method, 10:1 split ratio, 1µl injection volume.

Drug25: General drug analysis method, 25:1 split ratio, 1µl injection volume.

Drug50: General drug analysis method, 50:1 split ratio, 1µl injection volume.

Drug100: General drug analysis method, 100:1 split ratio, 1µl injection volume.

Drug200: General drug analysis method, 200:1 split ratio, 1µl injection volume.

Drug300: General drug analysis method, 150:1 split ratio, 0.5 µl injection volume.

Drug750: General drug analysis method, 75:1 split ratio, 0.1µl injection volume.

Drug1000: General drug analysis method, 100:1 split ratio, 0.1µl injection volume.

Drug2000: General drug analysis method, 200:1 split ratio, 0.1µl injection volume.

Drug3000: General drug analysis method, 300:1 split ratio, 0.1µl injection volume.

Extnd10: General drug analysis method with extended final time for compounds such as steroids, 10:1 split ratio, 1µl injection volume.

Extnd25: General drug analysis method with extended final time for compounds such as steroids, 25:1 split ratio, 1µl injection volume.

40Extnd25: General drug analysis method with extended 40-minute final run time for compounds such as sildenafil, 25:1 split ratio, 1µl injection volume.

Extnd50: General drug analysis method with extended final time for compounds such as steroids, 50:1 split ratio, 1µl injection volume.

Extnd100: General drug analysis method with extended final time for compounds such as steroids, 100:1 split ratio, 1µl injection volume.

Extnd200: General drug analysis method with extended final time for compounds such as steroids, 200:1 split ratio, 1µl injection volume.

Extnd300: General drug analysis method with extended final time for compounds such as steroids, 150:1 split ratio, 0.5µl injection volume.

HCLad: Extended method for hydrocarbon calibration. This method does not search the hydrocarbon peaks, eliminating the unnecessary printouts of mass spectra. Method utilizes a 40:1 split ratio, 1µl injection volume.

LowM50: Specialized Drug50 method that scans to lower masses, 50:1 split ratio, 1µl injection volume.

LowM100: Specialized Drug100 method that scans to lower masses, 100:1 split ratio, 1µl injection volume.

LSD: Method specifically for LSD analysis. Method utilizes lower mass scanning range for the entire analysis time, 10:1 split ratio, 1µl injection volume.

LSD0: Method specifically for LSD analysis. Method utilizes lower mass scanning range for the entire analysis time, 0:1 split ratio, 1µl injection volume.

Meth50: Five minute run which can be utilized when specific requirements are met, 50:1 split ratio, 1µl injection volume. Requirements listed in “Protocol Notes”.

Meth100: Five minute run which can be utilized when specific requirements are met, 100:1 split ratio, 1µl injection volume. Requirements listed in “Protocol Notes”.

PCPTCP25: Specialized method for the differentiation and identification of PCP and TCP, 25:1 split ratio, 1µl injection volume. **See Individual Steps of Protocol**

PCPTCP50: Specialized method for the differentiation and identification of PCP and TCP, 50:1 split ratio, 1µl injection volume. **See Individual Steps of Protocol**

PCPTCP200: Specialized method for the differentiation and identification of PCP and TCP, 200:1 split ratio, 1µl injection volume. **See Individual Steps of Protocol**

Slow50: Specialized method for the analysis of compounds that elute quickly, 50:1 split ratio, 1µl injection volume.

Slow100: Specialized method for the analysis of compounds that elute quickly, 100:1 split ratio, 1µl injection volume.

Solvent: Specialized method for obtaining mass spectra of solvents, 500:1 split ratio, 0.1µl injection volume.

Individual Steps of Protocol:

1. Generally, samples are dissolved in methanol or extracted.
2. Prior to injection on the GCMS, all samples are analyzed by gas chromatograph (GC) to make a tentative identification of the sample and to determine if the sample needs to be diluted or concentrated.
3. Enter case and sample information into Chemstation sample log table, including case number, sample item number, extraction solvent, analyst's name or initials, method to be used for analysis and vial barcode.
4. Inject a portion of the sample onto the GCMS utilizing one of the approved methods for drug analysis. NOTE: A sample analyzed by GC in methanol and determined an extraction is necessary; the extraction must be re-analyzed by GC to determine proper concentration for GCMS analysis.
5. The analyst will directly compare the question sample to a standard spectra and its retention index or retention time for interpretation.
   a. The analyst will evaluate the quality of the chromatography and spectrum to decide if it is of sufficient quality to compare with a standard spectra or if a different extraction, dilution, or concentration of the sample is needed. The analyst will also evaluate the spectrum to determine if it is a single compound or if there is co-elution.
   b. The retention index of the question sample must not exceed the retention index of the standard by more than 2.0%. It is not necessary to document the retention index difference, as long as both retention indexes are documented.

Calculations:

Retention Index:

Retention index values are calculated by the data system using the macro "msdchem\osbimac\ri_100.mac" or similar macro as follows:

```
if Ret_time >= RT_C9 then
if Ret_time < RT_C10 then
RI_UNK = C9 + (C10-C9) * (RET_time - RT_C9)/(RT_C10 - RT_C9)
endif
endif
if Ret_time >= RT_C10 then
if Ret_time < RT_C11 then
RI_UNK = C10 + (C11-C10) * (RET_time - RT_C10)/(RT_C11 - RT_C10)
endif
endif
.....etc. until C36 is reached
```

Expansion Volumes:

During injection, the solvent vaporizes. If the vapor exceeds the volume of the glass inlet liner, it may contaminate metal lines and parts within the injector which can subsequently result in carry over or contamination of future injections.

Expansion volumes for different pressures or inlet liner sizes can be calculated using the following formulas:

Sample and solvent expansion volume = $\frac{nRT}{P}$

n = # of moles of solvent and sample = $\frac{\text{volume (milliliter) x density (gram/milliliter)}}{\text{MW (grams/mole)}}$

T = absolute temperature of injector (K)

P = column head pressure (Atm) + 1 Atm

R = gas law constant = 82.06 cc atm/mole x deg. K

Volume of inlet liner = $\pi r^2 L$

r = radius (cm)

L = length (cm)

Solvent Expansion Volumes in µl at Various Head Pressures

Solvent	Density (g/ml)	MW (g/mole)	5 psig	10 psig	15 psig
Hexane	0.66	86	264	210	175
Chloroform	1.49	119	430	343	285
Methanol	0.79	32	852	680	565
Toluene	0.87	92	325	260	216
BSTFA	0.96	257	128	103	85

The expansion volumes were determined using a 1.0 µl injection volume, a 290°C injection port temperature, and a head pressure of 5, 10, and 15 psig.

The 4mm inlet liner has a 990 µl volume. Solvent expansion volumes are not to exceed the capacity of the liner.

Frequency and Tolerance of Controls and Corrective Action for exceeded Tolerances:

A gas chromatograph mass spectrometer will be removed from service immediately and listed actions taken, if any of the following occur:

1. The value for retention index of the cocaine standard exceeds plus or minus 2% of the expected value:
    - Run hydrocarbon ladder, verify instrument performance, and document in maintenance log if the problem continues,
    - Do not report any results from the analysis sequence.

2. The failure to produce a satisfactory cocaine spectrum for the cocaine standard (positive control).
    - Determine the performance problem, repair, verify instrument performance, and document in maintenance log if the problem continues,
    - Do not report any results from the analysis sequence.

Any instrument removed from service will be documented in the GC/MS maintenance log (DR70-1) and reported immediately to the Supervisor and Technical Manager.

The following require corrective action, but not the removal of the instrument from service:

1. When high levels (above approximately 10% on the scale of an autotune) of O2, N2, or H2O are present in the autotune:
    - Find and repair the leak immediately and document in the maintenance log if the problem continues.

2. Failure of instrument tune evaluation:
    - The operator is to diagnose and correct the problem, and document in the maintenance log if the problem continues.
    - If the operator is unable to determine the cause of the failure, contact the technical manager to determine the best course of action.

3. The value for the retention index or retention time of a single unknown and the standard exceed a plus or minus 2% difference:
    - The operator is to diagnose and correct the problem, and document in the maintenance log if the problem continues.
    - The sample in which this occurs will not be reported until it is within the 2% window.

4. Any contamination peaks in the blank chromatograms, including controlled substances or related compounds or interfering substances present in any concentration, during the course of an analysis sequence:
    - The operator must diagnose the source of the peaks and correct the problem
    - In order to report results from a sample or a series of samples, the blank preceding the sample(s) must show no signs of contamination.

Protocol Notes:

1. Casework will not be reported from instruments where the injection liners and septa counts exceed 250 injections. The analysis of unextracted or dirty samples may necessitate more frequent injection liner changes. Inject a sufficient number of solvent blanks after liner and septa changes to eliminate extraneous background noise. Casework will not be reported from instruments where the gold seal count exceeds 1500 injections. When the gold seal is changed, the metal sides of the injector are to be cleaned with cotton tipped swabs and a solvent until no discoloration of swabs is present, and the metal septum nuts are to be cleaned and rinsed in methanol. The split vent filter is to be changed at or before 2500 injections. The split vent line is to be changed at or before 5000 injections.

2. Partial or abbreviated GC or GC/MS runs are not normally to be utilized except as specifically provided for in the protocol. Abbreviated runs increase the risk of retaining compounds on the GC column that may elute in a later run. All runs will be of sufficient duration and programming to elute C34 with the exception of the following circumstance. Based upon the interpretation of previously run gas chromatographic data, a short GCMS method (Meth50, Meth100) of 5 minutes or longer may be utilized for the identification of methamphetamine. The gas chromatographic data must show no sign of any further analyte in a sample beyond 2.5 minutes past the retention time of the methamphetamine peak on the standard ladder.

   Based upon the interpretation of previously run gas chromatographic data, a short GCMS method (Method1) may be utilized for the identification and separation of Ephedrine and Pseudoephedrine. The gas chromatographic data must show no sign of any further analyte in a sample beyond 1.0 minutes past the retention time of the pseudoephedrine peak on the standard ladder. All blank runs between samples and cases must still meet the C34 requirement.

3. Routine maintenance (i.e., hydrocarbon ladder, oil changes, column changes, liner changes, cleaning of metal septum nuts, cleaning injection port, updating macros, etc.) and repairs are to be documented on the instrument maintenance log (DR70-1). When an instrument is removed from service, label the instrument “Not in Service” and document in the maintenance log. When the instrument is returned to service, document the repairs and maintenance. Note that the instrument is returned to service.

4. A hydrocarbon ladder consisting of the hydrocarbons C10 through C30, C32, C34, C36, and C38 will be analyzed within the first 5 working days of each month or before casework is analyzed on the instrument for that month.

5. Retention index programs on the GC/MS are to be updated at least monthly when the instrument is in service. Hydrocarbon ladders will be archived in PDF format on each instrument. The file will be stored in a primary folder titled "HCLadders" which contains secondary folders by year (2011, 2012, etc…). The analyst approving the TIC of the hydrocarbon ladder will name the PDF file "mm-dd-yy HCLad xxxxxx AAA", xxxxxx being the six-digit validation code and AAA being the initials of the verifying person. See Appendix B for instructions on reprocessing the hydrocarbon ladder.

6. Ensure that the solvent wash bottles contain sufficient solvent to rinse the syringe. The wash bottle is to be labeled with the minimum liquid level.

7. Autotunes are to be performed prior to the first sequence of the day when the instrument is in use. All autotunes will be evaluated utilizing the Agilent tune evaluation. If the tune evaluation indicates the autotune passes, the instrument is ready to use.

   When sequences continuously run for an extended period of time, for example 2 or more days, it is not necessary to stop the instrument to perform an autotune. Autotunes are to be archived in PDF format; the filename should be renamed to contain the initials of the person verifying the autotune. The format should follow "mm-dd-yy AAA", AAA being the initials of the verifying analyst. Tune evaluations are to be archived in PDF format; the filename should be renamed to contain the initials of the person verifying the tune evaluation. The format should follow "mm-dd-yy TE AAA", TE indicating tune evaluation and AAA being the initials of the verifying analyst. It is not necessary to document the autotune or tune evaluation in the maintenance log.

8. Maximum injection volume on the GC/MS is to be determined by utilizing solvent expansion volumes in the "Calculation" portion of the protocol. Expanded vapor volume is not to exceed the volume of the inlet liner (990 µl). One microliter is the maximum injection volume when using methanol as a solvent.

9. Refer to www.agilent.com for instrumentation user's manual.

10. To report a substance as "No controlled dangerous substance identified" the sample should be analyzed using either a Drug25, Drug10, Extend25 or Extend10 method, unless using one of them would overload the detector with sample. If Drug25, Drug10, Extend25 or Extend10 are not used, the analyst must put a narrative in the item notes explaining why they were not used.

11. When performing GC/MS analysis and no peaks in the TIC were matched to any reference spectra in an OSBI library, a manual scan should be performed on all peaks that appear significant.

    a. If manual scans indicate no controlled substances, then an indication on the TIC should be made showing a manual scan was performed and no controlled substances were identified.
    b. If a manual scan indicates a non-controlled substance that the analyst chooses to identify, then further analysis must be performed. To make an identification, the sample spectra must be directly compared to a standard spectra and meet the requirements found in this protocol.

c. If a manual scan indicates a controlled substance, then either further analysis should be performed or the case file should include information as to why the substance could not be identified.
   i. The reason that the controlled substance cannot be identified can be included in the notes section for that item within the BEAST or a printout of the mass spectra showing the ions of interest can be included in the case file.
   ii. If an analyst is utilizing PDF exam documentation, indication should be made in the notes section for that item within the BEAST or on the PDF document, indicating a manual scan was performed on the PDF document.
   iii. Appropriate report wording should also be used to reflect the results of analysis.

Recommended Report Wording:

1. When preliminary instrumental and GCMS examination does not indicate a controlled substance, report using the following:

   Item #: No controlled dangerous substance identified.

2. When preliminary instrumental and GCMS examination has been performed and a controlled substance is indicated but is not conclusive, report using one of the following examples:

   Item #: Examinations indicate the presence of a controlled substance; however, the laboratory's reporting criteria have not been met.

3. The GCMS is normally considered a conclusive examination by itself, however the OSBI requires that a second test be conducted. When used in conjunction with a positive gas chromatography, FTIR, literary references for pharmaceuticals (GC screening still required for concentration determinations) or thin layer chromatography examination, report using the following examples:

   Item #: Methamphetamine, Schedule II

   Item #: Camazepam, Federal Schedule III

   Item #: Lidocaine, a non-controlled substance

4. When visual and literary examinations indicate a substance that cannot be identified using GC and GCMS, but instrumental analysis is started, report using the following example:

   Item #: Visual, literary and instrumental analysis was performed. Visual and literary results indicate Pregabalin, Schedule V; however, this could not be confirmed by instrumental analysis.

5. The OSBI CSD Quality Manual requires all items in a container be listed on each report. In the case of evidence containing items needing additional analysis and items that have been tested and reported in a previous report, report the untested items using the following example:

   Item #: No additional analysis was performed.

6. When GCMS cannot identify a substance and further instrumental analysis is needed, but will be performed by a different analyst, report using the following example:

   Item #: Instrumental analysis indicates the presence of a controlled substance, however this was not confirmed. This item will be transferred for additional analysis.

Associated Form(s):

DR70-1 Maintenance Log for OSBI GC/MS

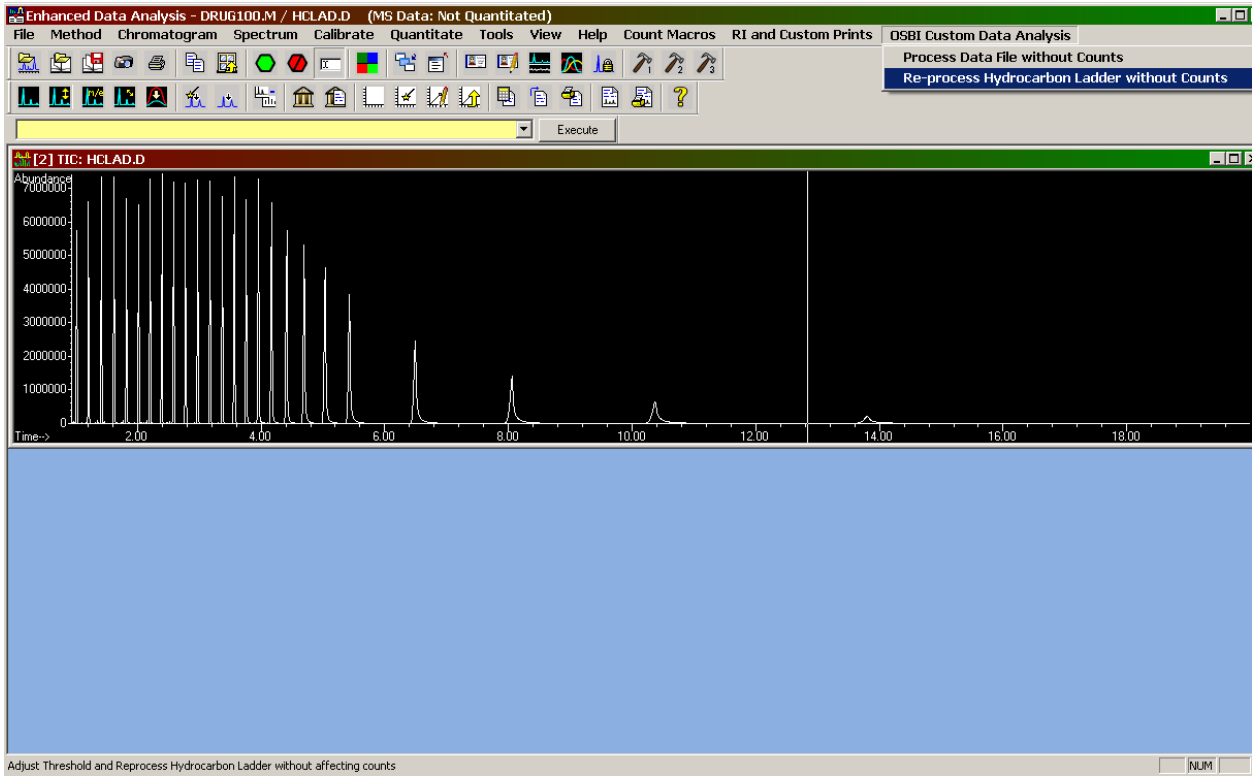
References:

1. Saferstein, Criminalistics, an Introduction to Forensic Science. 5th Edition: Prentice Hall, 126-32, 145-51.
2. Saferstein, editor. Forensic Science Handbook. Prentice Hall, 92-138

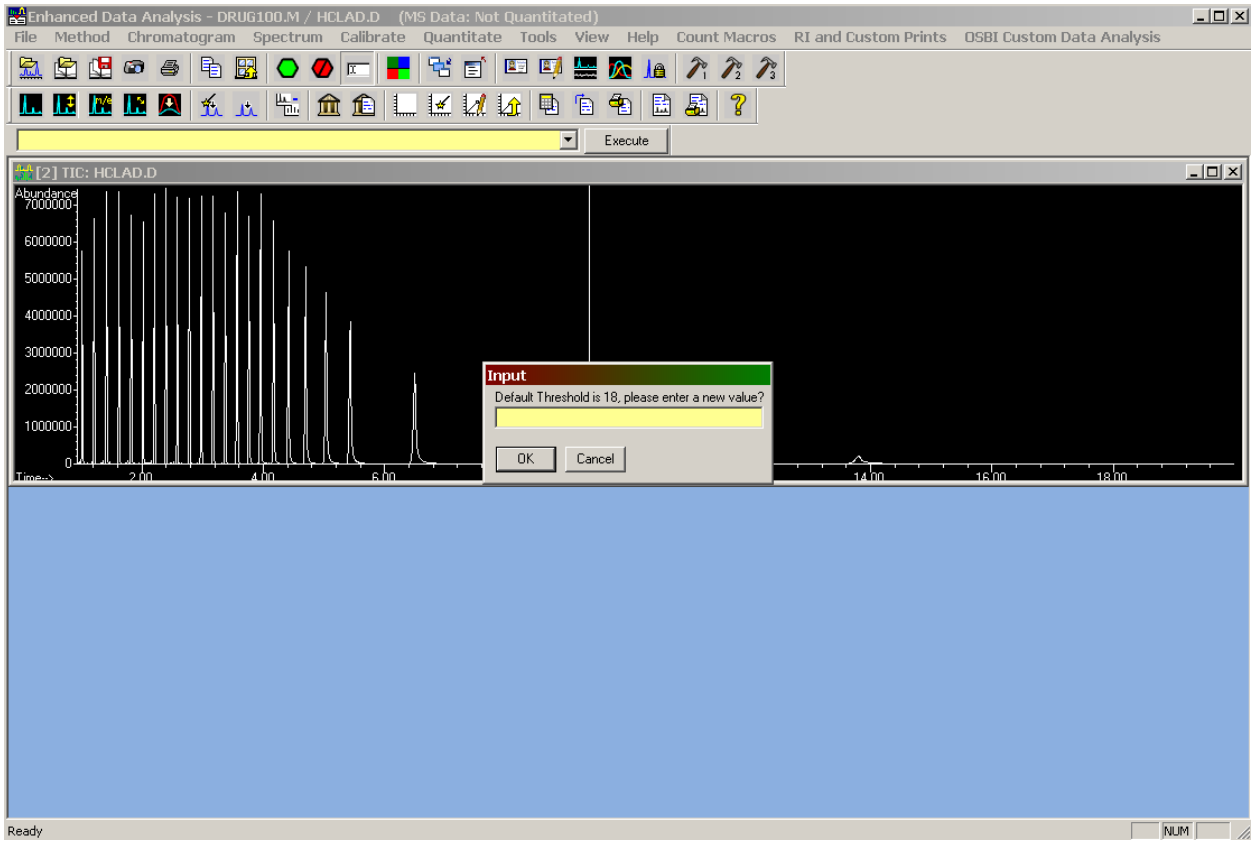
Appendix B: Instructions for Reprocessing Hydrocarbon Ladders

The hydrocarbon ladder can only be reprocessed on the day it was run! When it is reprocessed, it uses the date of the day it was reprocessed and not the day it was run.

The automated processing of the hydrocarbon ladder and updating of the retention times can occasionally integrate either too many or too few peaks. To alleviate the need for re-shooting the ladder, under the GCMS data analysis toolbar heading "OSBI Custom Data Analysis" is a selection "Re-process Hydrocarbon Ladder without Counts". This selection will prompt analysts to change the threshold setting and then re-process the data file without changing the current counts. This macro is intended for use when the hydrocarbon ladder is run and either there is too many or too few peaks integrated with the new automated hydrocarbon ladder processing.



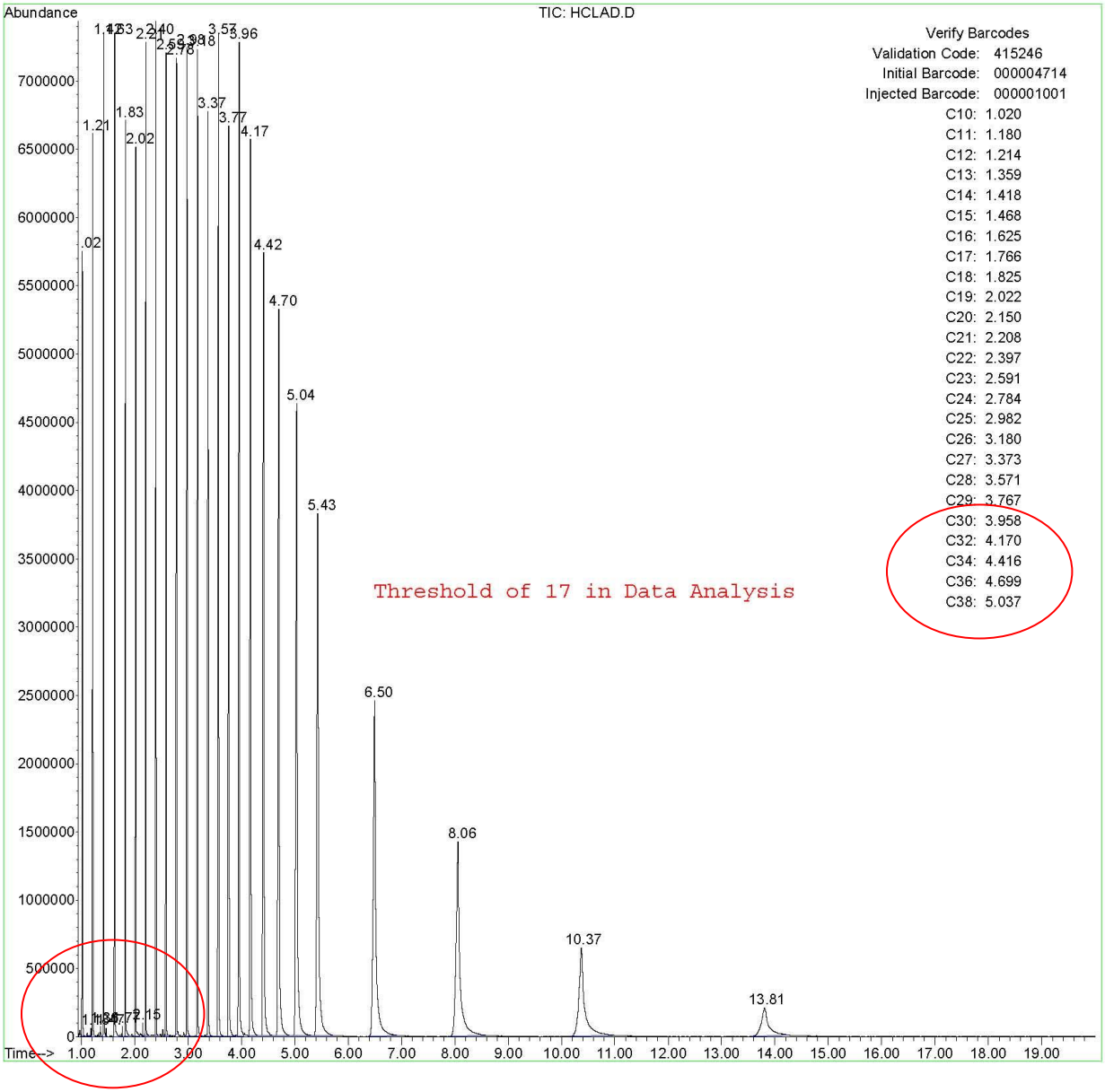
Load the current HCLAD.D data in data analysis. IT does not matter what method is loaded. You will be prompted for the threshold value you want to process the data file with. The default threshold setting for data analysis is 18. If you have changed this value at some time in the past, then you're setting may be set at a different number. The allowed values are -12 through 25, where -12 specifies the most sensitive (lowest) threshold value and 25 the least sensitive (highest) threshold. Each change of 1 in the threshold changes the sensitivity of the integrator by a factor of 2 (ex: a value of 8 is four times less sensitive than a value of 6). In most cases, you will only need to change the value by one or two at the most, working within a range of 16-20.



Once you enter your threshold value, click OK and the macros will process the data file at the new value. The processing will not change any of your current counts. The processing will update the values stored for your hydrocarbon ladder. Once the file has been processed correctly, you can archive that printout and continue with case work. Below are three examples at different threshold values.

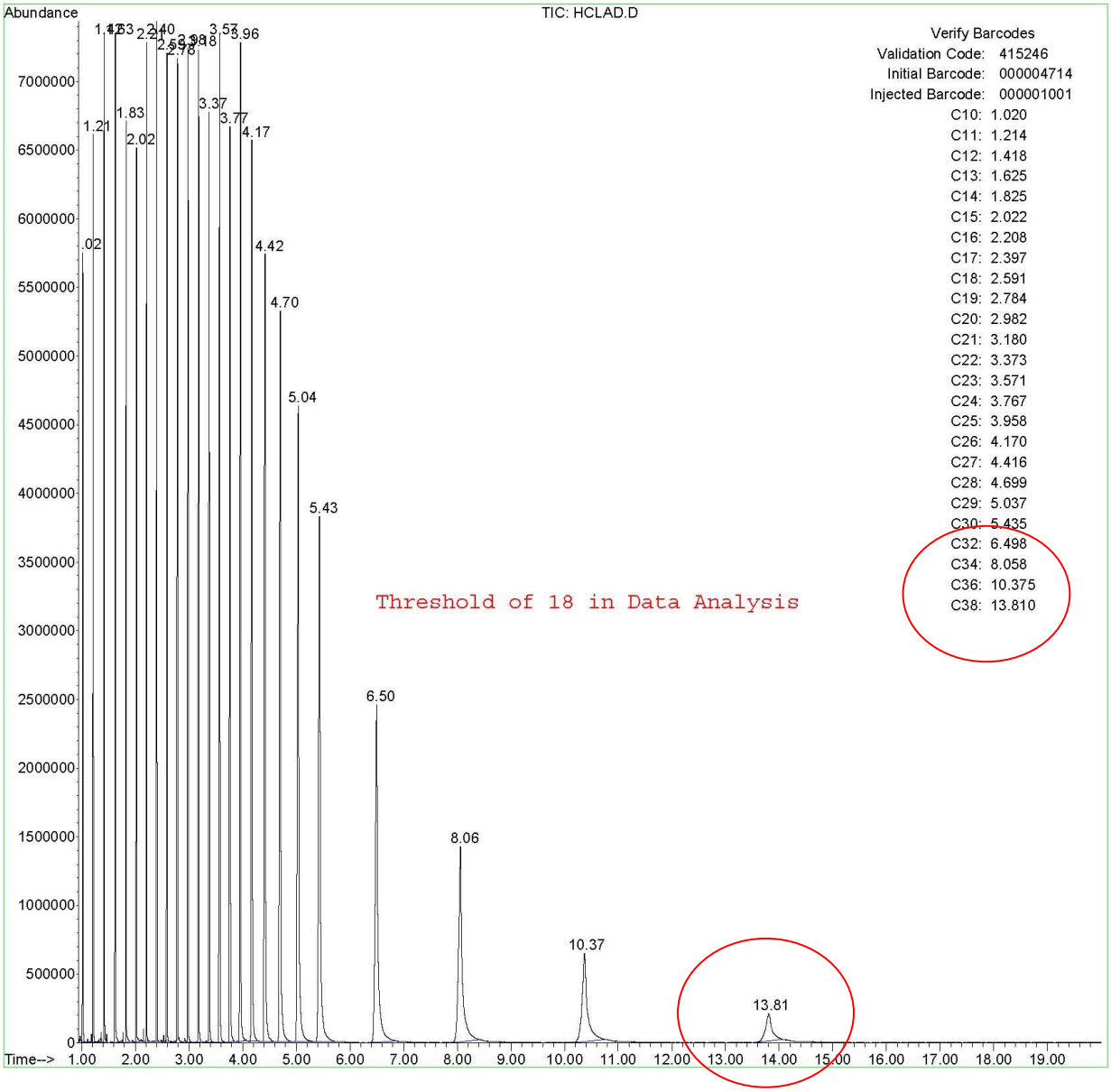
Page #:_____

```
Lab Number:  HYDROCARBON LADDER
Instrument   YODA
Analyst:     K. HEARD
Date/Time:    2 Nov 2010   7:04
Method:      HCLAD.D Cust DA
Vial Number: 90

QUALITY CONTROL                  INSTRUMENT CONDITIONS
Liner count:  214                Initial temp:    100 C
Septum count: 214                Final temp:      290 C
Gold Seal:    1449               Initial time:    15 sec
Column count: 5531               Final time:      16.15 min
GC Column:                       Total Run Time:  20.0 min
 ZB-1MS 12.0m x 0.20mm x 0.33um  Split Ratio:     25:1
Column Installed:    06/10/10    Inj. Port Temp:  290 C
Hydrocarbon Ladder:  11/10/10    Transfer Line:   290 C
                                 Ramp Rate:        65 C/min to 220,
                                                   40 C/min to 290
```



Page #:_____

```
Lab Number:  HYDROCARBON LADDER
Instrument   YODA
Analyst:     K. HEARD
Date/Time:    2 Nov 2010   7:04
Method:      HCLAD.D Cust DA
Vial Number: 90

QUALITY CONTROL                  INSTRUMENT CONDITIONS
Liner count:  214                Initial temp:     100 C
Septum count: 214                Final temp:       290 C
Gold Seal:    1449               Initial time:     15 sec
Column count: 5531               Final time:       16.15 min
GC Column:                       Total Run Time:   20.0 min
 ZB-1MS 12.0m x 0.20mm x 0.33um  Split Ratio:      25:1
Column Installed:     06/10/10   Inj. Port Temp:   290 C
Hydrocarbon Ladder:   11/10/10   Transfer Line:    290 C
                                 Ramp Rate:         65 C/min to 220,
                                                    40 C/min to 290
```

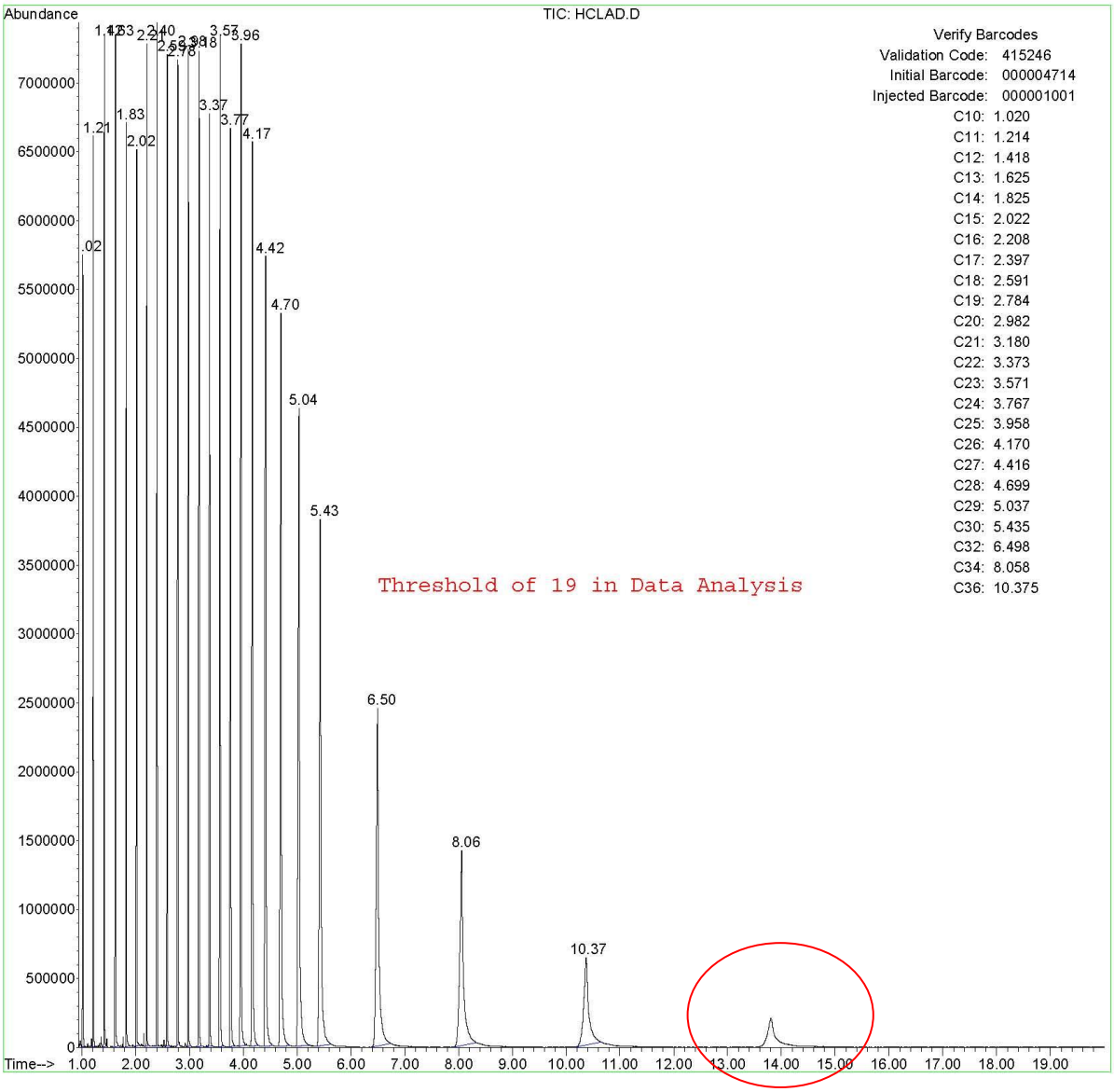


Lab Number: HYDROCARBON LADDER
Instrument YODA
Analyst: K. HEARD
Date/Time: 2 Nov 2010 7:04
Method: HCLAD.D Cust DA
Vial Number: 90

Page #:_____

QUALITY CONTROL
Liner count: 214
Septum count: 214
Gold Seal: 1449
Column count: 5531
GC Column:
ZB-1MS 12.0m x 0.20mm x 0.33um
Column Installed: 06/10/10
Hydrocarbon Ladder: 11/10/10

INSTRUMENT CONDITIONS
Initial temp: 100 C
Final temp: 290 C
Initial time: 15 sec
Final time: 16.15 min
Total Run Time: 20.0 min
Split Ratio: 25:1
Inj. Port Temp: 290 C
Transfer Line: 290 C
Ramp Rate: 65 C/min to 220,
40 C/min to 290



The data file can be re-processed as many times as necessary. All counts that were current at the time of analysis will be present on the re-processed print out. All hydrocarbon numbers will be updated so all retention index calculations will be correct. The pdf file name will contain the word "Reprocessed" in the file name. On the print out itself you will notice "Cust DA" after the method. This indicates that a custom data analysis was performed to generate this print out. Therefore, it is easy to tell if it is the original hydrocarbon ladder or a re-processed hydrocarbon ladder.

Note: The threshold you select for re-processing is not stored permanently in the method/data analysis.

DR-75 Gas Chromatograph Infrared Detector Methods for Drug Analysis

Principle of the Protocol:

Infrared spectrophotometry deals with the interaction between molecules and infrared radiation. The molecules of almost every chemical compound are capable of absorbing some infrared radiation. The atoms in a molecule vibrate in complicated patterns, with frequencies that depend on the masses of the atoms and the strengths of the bonds connecting them together. The molecule can only absorb those frequencies of infrared radiation that exactly match its own vibrational frequencies. Monatomic gases and diatomic gases with atoms of the same element such as helium, hydrogen, oxygen, and nitrogen cannot absorb infrared radiation.

The dissolved sample is introduced into the inlet of the gas chromatograph portion of the instrument. The inlet temperature is high enough to volatilize most organic substances with a molecular weight below approximately 600. The helium carrier gas transporting the sample flows into the capillary column from the injector. The capillary column separates the components of the sample based on their solubility in the liquid phase and volatility. The end of the GC column extends from the GC oven to the flow cell through a heated interface to maintain thermal uniformity. As each component elutes from the column, it passes through the flow cell. The sample is then returned to the GC oven through a second heated interface.

The IRD instrument uses an IR source to create infrared light. The light from the source passes through the interferometer and then through a flow cell. Any light that reaches the end of the flow cell is collected and focused on a detector. The detector converts the intensity of the beam into an electrical signal. This signal is then converted to digital numbers that can be processed by the computer. The spectrometer measures, repeatedly and as fast as possible, the infrared spectrum of anything in the flow cell. When only carrier gas is present, there is no detectable absorption. When a component of an unknown mixture elutes, a spectrum is obtained. The wavelengths absorbed depend on the identity of the compound and the amount of radiation absorbed depends on the amount of the compound present.

The IRD detector provides some molecular identification that a mass spectrometer cannot. The IRD utilizes a flow cell to analyze the intact molecule. The molecules are freely rotating and isolated from each other in a low energy environment in which the molecular geometry is kept intact. The IRD's vapor phase spectra are molecularly unique and highly reproducible.

Associated Protocol(s):

DR-30 Gas Chromatography Analysis

Specimen Required (Sample Handling and Preservation):

Dissolved samples of sufficient strength to give a detailed total response chromatogram (TRC) of the suspected compound. Depending on the compound, concentrations can range from 0.25 to 5 or more milligrams per milliliter. Phenethylamines such as methamphetamine, amphetamine, ephedrine, pseudoephedrine are to be injected as the free base, whenever possible. However, residues may be injected either as the salt or free base. Unless they are extracted, phenethylamines normally do not chromatograph well. Other compounds may be injected in the salt form.

Reagents:

1. Reagents as needed for extractions.
2. High purity helium carrier gas

All reagents are Reagent Grade or better.

Standards:

All identifications will be made by comparing the question sample to a known documented standard TRC that is to be available in the case file for future peer review and possible legal review. Such standards are to be purchased from Alltech, Fluka, Sigma, Supelco, U.S. Pharmacopeia, or other reputable manufacturers. Spectra from literary references, computer references, and samples of unknown origin or from cases are not suitable for making a conclusive comparison for reporting. TRC's retrieved from a user-generated computer library are recommended, but detailed TRC's must be used for comparison and print out. The following information is to be readily and permanently available for each standard:

- Name of standard
- Manufacturer or source
- Date spectra was produced
- Lot number (if used by manufacturer or source)
- Analyst who produced the spectra
- Literary reference that the spectra were compared to, analyst making the comparison, and date.

Controls:

A blank that consists of each of the appropriate solvents or extractions used in a case will be shot during the case samples and at least every sixth injection (five samples then a blank) within a case. Place the blank TRC printout in the case file of the item that follows it during analysis. If the required types of solvent blanks have been analyzed by GCMS, it is not necessary to repeat these for the IRD. Methanol is acceptable for quality control. Each day that casework is performed, the first sequence is to contain a solvent blank as a negative control and an approximate two to three milligram/milliliter concentration cocaine standard for a positive control. The solvent blank data file will contain the name "blank" and the cocaine standard data file will contain the name "cocaine". Controls will be run and documented whether or not the autosampler is utilized (manual injection). Archive cocaine standard and run tables.

Apparatus and Materials:

1. Agilent Gas Chromatograph
2. J&W/Agilent/Restek DB/HP-1/Rxi-1 (12m x 0.20mm x 0.33 um) columns. The Technical Manager of Controlled Substances may approve other columns for use.
3. Syringes, Five (5) microliter
4. Glass auto sampling vials
5. Glass inserts for vials
   NOTE: Do not use polypropylene or any polymer vial with chloroform or hexane solvent
6. Caps for auto sampling vials
7. Glass wool.

Individual Steps of Protocol:

A. Methods for drug analysis using the Agilent GC / IRD detector are available from the technical manager. These methods are to be utilized by all drug analysis sections of the OSBI utilizing this instrument. Any requests for new methods/macros or changes to existing methods/macros should be submitted in writing to the Technical Manager of Controlled Substances for review and approval prior to any changes being made.

The current approved methods are as follows:

Ird2ml: General drug analysis method used for blanks and samples. 2ml/min column flow.
Ird2ml05: General drug analysis method used for blanks and samples. 2ml/min column flow with a split ratio of 5:1.
Ird2ml10: General drug analysis method used for blanks and samples. 2ml/min column flow with a split ratio of 10:1.
Ird2ml20: General drug analysis method used for blanks and samples. 2ml/min column flow with a split ratio of 20:1.
Ird2ml30: General drug analysis method used for blanks and samples. 2ml/min column flow with a split ratio of 30:1.
Ird10ml: General drug analysis method used for blanks and samples of higher molecular weight compounds. 10ml/min column flow.
IrdXml: General drug analysis method where X is the number of milliliters/min of column flow. This is to allow for flexibility in the method for analysis of specific drugs if necessary.
IrdSolv: Method to specifically identify solvents.
IRDFKIT01
IRDFKIT02
IRDFKIT05
IRDFKIT10
IRDFKIT20

B. Procedure:
1. Generally, samples are dissolved in methanol or ethanol. Extraction of some samples may be required.
2. Prior to injection on the GC/IRD, all samples are analyzed by a gas chromatograph to make a tentative identification of the sample and to determine if the sample needs to be diluted or concentrated.
3. Inject a portion of the sample onto the GC/IRD. Sample size shall be an amount that when vaporized will not exceed 990 microliters. Do not inject more than one and one-half (1.5) microliters of methanol or an unknown solvent (i.e., clan labs) at 25-PSI inlet pressure. See "Calculations" for solvent expansion volumes.
4. A sample analyzed by GC in methanol and determined an extraction is necessary; the extraction must be re-analyzed by GC to determine proper concentration for GC/IRD analysis.

5. The analyst will directly compare the question sample to a standard spectrum for interpretation.

Calculations:

During injection, the solvent vaporizes. If the vapor exceeds the volume of the glass inlet liner, it may contaminate metal lines and parts within the injector which can subsequently result in carry over or contamination of future injections.

Expansion volumes for different pressures or inlet liner sizes can be calculated using the following formulas:

Sample and solvent expansion volume = $\frac{nRT}{P}$

n = # of moles of solvent and sample = $\frac{\text{volume (ml) x density (gm/ml)}}{\text{MW (grams/mole)}}$

T = absolute temperature of injector (K)

P = column head pressure (Atm) + 1 Atm

R = gas law constant = 82.06 cc atm/mole x deg. K

Volume of inlet liner = $\pi r^2 L$

r = radius (cm)

L = length (cm)

Solvent Expansion Volumes in µl at 25 psig Head Pressure

Solvent	Density (g/ml)	MW (g/mole)	25 psig
Hexane	0.66	86	196
Chloroform	1.49	119	320
Methanol	0.79	32	634

The expansion volumes were determined using a 1.5 µl injection volume, a 290°C injection port temperature, and a head pressure of 25 psig.

The inlet liner for the GC/IRD has a 990 µl volume. Solvent expansion volumes are not to exceed the capacity of the liner.

Frequency and Tolerance of Controls and Corrective Action to Be Taken If Tolerances Are Exceeded:

A GC/IRD will be removed from service immediately and listed actions taken if any of the following occur:

1. The failure to produce a satisfactory TRC for the cocaine standard (positive control).
   - Determine performance problem, repair, verify instrument performance, and document in maintenance log,
   - Do not report any results from the analysis sequence.

Any instrument removed from service will be documented in the GC/IRD maintenance log (DR75-1) and reported immediately to the Technical Manager.

The following require corrective action, but not the removal of the instrument from service:

1. Contamination of blanks during the course of an analysis sequence. Contamination is defined as carry over from a previous sample into a later sample or control in a sequence or manual run:
   - Do not utilize the instrument until problem is identified, fixed, and proper performance demonstrated, and documented in the maintenance log.
   - In order to report results from a sample or series of samples, the blank preceding and following the samples must show no signs of contamination.

Protocol Notes:

1. Casework will not be reported from instruments were the injection liners and septa counts exceed 250 injections. The analysis of un-extracted or dirty samples may necessitate more frequent injection liner changes. Every 1500 injections the gold seal is to be changed, the metal sides of the injector are to be cleaned with cotton tipped swabs and a solvent until no discoloration of swabs is present, and the metal septum nuts are to be cleaned and rinsed in methanol. Inject a sufficient number of solvent blanks after liner and septa changes to eliminate extraneous background noise.

2. Routine maintenance (liner/septa changes & inlet cleaning) and repairs are to be documented on the instrument maintenance log. The format of the instrument log must have a place for documenting what was done, the date, and who did the maintenance or repair. When an instrument is removed from service, label the instrument “Not in Service” and document in the maintenance log. When the instrument is returned to service, document the event, relevant repairs and maintenance in the maintenance log. Note that the instrument is returned to service.

3. Ensure that the solvent wash bottles contain sufficient solvent to rinse the syringe. The wash bottle is to be labeled with the minimum liquid level.

4. Split ratios are to be recorded in header or footer information in macros.

Recommended Report Wording/Interpretation of Test Data:

1. The GC/IRD is normally considered a conclusive examination by itself, however the OSBI requires that a second test be conducted. When used in conjunction with a positive gas chromatography, literary references for pharmaceuticals (GC screening still required for concentration determinations), thin layer chromatography examination, or GCMS, report using the following example:

   Item #: Methamphetamine, Schedule II

2. If no controlled dangerous substance is conclusively identified using GC/IRD, an analyst cannot report these findings unless they are verified by GC/MS.

Associated Form(s):

DR75-1 Maintenance Log for OSBI GC/IRD

References:

1. IRD3 Operations Manual, Rev 1-3, ASIC
2. Essential FTIR Operations Manual for GC IRD Users, Rev 0-3

DR-101 Identification of Lysergic Acid Diethylamide

Purpose:

Identification of lysergic acid diethylamide (LSD) from case samples is made using Gas Chromatography (GC) and Gas Chromatography/Mass Spectrometry (GC/MS).

LSD has two isomers that may be encountered, lysergic acid methyl propylamide (LAMPA) and lysergic acid sec-butylamide (sec-LSB). Differentiation of these compounds is made by use of retention time differences, retention index (RI) differences and ion ratios in the mass spectra. OSBI uses GC/MS macros that compare retention index differences, with a requirement that the RI difference is less than 1%. The macros also compare ion ratios specific for LAMPA and LSD, the ion ratios must be within defined limits for the compound to be selected by the macro. The analyst also does comparisons of the standard to the unknown and makes the final determination.

Associated Protocol(s):

DR-30 Gas Chromatography Analysis (Flame Ionization Detector)
DR-70 Gas Chromatograph / Mass Spectrometer Methods for Drug Analysis

Specimen Required (Sample Handling and Preservation):

Samples from many connected dosage units (2 or more), depending upon the quantity submitted, may be used for analysis; refer to the Controlled Substances Quality Manual, Case Management and Analysis for specifics regarding analyzing blotter squares/gelatin squares/candy/individual or multiple dosage units. For liquids, one half to one milliliter depending upon the volume submitted. Samples should be kept in a suitable cool and dark area while they are not being analyzed.

LSD has been observed in many forms. Primarily, LSD dosage units occur as segmented paper with or without ink designs. LSD has also been encountered in liquids, tablets, sugar cubes and in gelatinized samples. The concentration for individual dosage units is typically low and can range from 20 to 100 micrograms per dosage unit.

Reagents:

1. Methanol
2. Chloroform
3. Hexane
4. Concentrated Sulfuric acid
5. Sodium bicarbonate
6. Deionized water

All chemicals are reagent grade or better.

Standards:

1. Lysergic acid diethylamide (LSD)
2. Lysergic acid-n-methylpropylamide (LAMPA)

Apparatus and Materials:

1. Refer to DR-30 and DR-70 for instrumentation and columns
2. Test tubes
3. N-Evap or similar apparatus.
4. Autosampler vials

Reagents:

1. 0.4N H_2SO_4
   1 ml concentrated H_2SO_4
   89 ml de-ionized water
   Preparation of Reagent:
   a. Dilute H_2SO_4 with deionized water to 90 ml.

2. Mixed Solvent
   5 ml de-ionized water
   ~6 ml $CHCl_3$
   ~10-12 ml Hexanes
   Preparation of Reagent:
   a. In 25 ml graduated cylinder with ground glass stopper, add water and $CHCl_3$.
   b. Add hexane until the organic mixture floats on the water layer.
   c. Allow to stand for fifteen (15) minutes.

Individual Steps of Protocol:

Caution: ***Samples suspected of being LSD are to be handled with gloves or a mechanical device; never with bare hands***

1. Extraction recommendations
   a. Blotter paper
      1. Sample the blotter paper following the requirements in the Controlled Substances Quality Manual and place into a conical vial.
      2. Add methanol just to exceed the saturation point of the paper (1 to 4 drops).
      3. It is suggested that the vial be allowed to stand for approximately one (1) hour before analysis.
   b. Mixed Solvent extraction for liquid samples, tablets or gelatinized samples –
      1. Add ½ to 1 ml of a liquid sample or one to four dosage units of tablets or gelatinized samples, following the requirements in the Controlled Substances Quality Manual, to 1 ml of 0.4N H_2SO_4 in a test tube.
      2. Vortex.
      3. Centrifuge and transfer the supernate to another test tube. (This step is unnecessary for liquid samples).
      4. Carefully add solid sodium bicarbonate to the supernate until the H_2SO_4 is neutralized and the solution is saturated.
         This should be performed slowly to avoid evolving too much gas.
      5. Extract the solution with 0.5 ml of Mixed Solvent two times
      6. Evaporate the combined organic solvent extracts to dryness.
      7. One or two drops of methanol are then added to reconstitute the residue.

2. Perform analysis using GC
   a. Analysis must be performed on the sample and the LSD standard; analysis of the LAMPA standard is optional.
   b. Compare unknown retention time to known standard's retention time.
   Unknown retention time values must be within 2% plus or minus that of the standard.
   c. Prior to injection on the GCMS, all samples are analyzed by GC to make a tentative identification of the sample and to determine if the sample needs to be diluted or concentrated.

3. Perform analysis using GC/MS
   Analysis must be performed on the sample utilizing one of the approved methods for LSD analysis. The current approved methods for LSD analysis are:
   LSD: Method specifically for LSD analysis. Method utilizes lower mass scanning range for the entire analysis time.
   LSD0: Method specifically for LSD analysis. Method utilizes lower mass scanning range for the entire analysis time and a 0:1 split ratio.

Protocol Notes:

1. See DR-30 and DR-70 for Calculations.

2. See DR-30 and DR-70 for Frequency and Tolerance of Controls for GC and GC/MS.

3. See DR-30 and DR-70; all QA/QC elements in the protocols are applicable.

4. Most samples (paper, tablets) can be placed directly into methanol and then analyzed using GC and GC/MS. This method should be tried first, but, if purification is needed or if the sample is not suitable for methanol extraction, (liquids, gelatinized samples) then the Mixed Solvent extraction can be performed.

5. In the case of a sheet(s) of paper that are not perforated or segmented in some manner, the dimension of 1/4 inch by 1/4 inch square will be used in determining a dosage unit.

Recommended Report Wording/Interpretation of Test Data:

The report must specify the number analyzed and confirmed/not confirmed. Refer to the Controlled Substances Quality Manual, Case Management and Analysis for specific information.

1. To conclusively identify LSD, positive GC and GC/MS examinations are required. Report using the following example:

   Item #: Lysergic acid diethylamide, Schedule I.

2. When a gas chromatograph examination has been performed and is positive for LSD, but the quantity is not sufficient for a GC/MS, or the GC/MS is not conclusive, refer to DR-70 for report wording.

3. When GC and GC/MS examinations are negative, refer to DR-70 for report wording.

References:

1. Jacobs, JL. A simplified method for the clean-up and identification of LSD. Microgram, Vol. XVII, No. 6 June 1984.
2. Smith, F., Handbook of Forensic Drug Analysis, 2005, page 186.
3. Laing, R., Hallucinogens: A Forensic Drug Handbook, 2003 pages 38-39

DR-103 Classification of Synthetic Cannabinoids – Date of Offense August 21, 2015 or later

Purpose:

As of November 1, 2014, Oklahoma Statute was amended to allow for certain synthetic cannabinoids to be controlled based upon a structural class determination. 63 O.S. Section 2-204 Section G initially defined ten specific chemical groups that will be controlled as Schedule I. This statute was amended as of August 21, 2015 to include fourteen specific chemical groups. This protocol is designed to allow the analyst to determine if a synthetic cannabinoid belongs to one of these groups. This protocol is only for suspected synthetic cannabinoids with a date of offense of August 21, 2015 or later where the synthetic cannabinoid has been conclusively identified and are not specifically listed elsewhere in Oklahoma or Federal Statutes. For suspected synthetic cannabinoid cases with a date of offense between November 1, 2014 and August 20, 2015, see TM for archived DR-102.

Associated Protocol(s):

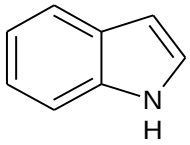
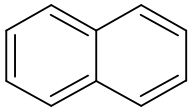
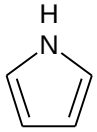
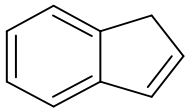
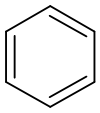
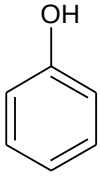
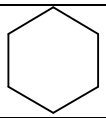
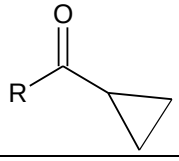
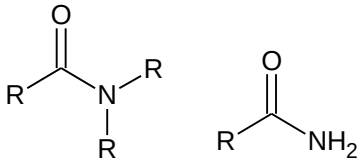
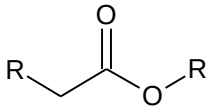
DR-30	Gas Chromatography Analysis
DR-60	Drug Analysis by FTIR
DR-70	Gas Chromatograph Mass Spectrometer Methods for Drug Analysis
DR-75	Gas Chromatograph Infrared Detector Methods for Drug Analysis

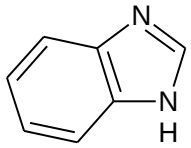
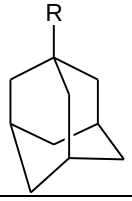
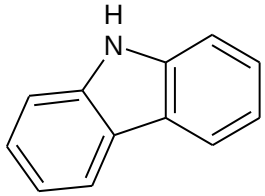
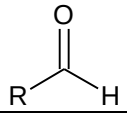
Definitions:

Alkanes	any series of hydrocarbons that contain only single bonds
Alkenes	any series of hydrocarbons that contain a double bond.
Alkenyl	an alkene where one hydrogen is removed allowing it to be a functional group alkene
Alkyl	an alkane where one hydrogen is removed allowing it to be a functional group (examples: methyl, ethyl, propyl)
Benzyl	a benzene ring attached to a CH2 group
Cycloalkanes	alkanes in which the carbon atoms are arranged in a ring
Cylcoalkyl	a cylcoalkane where one hydrogen is removed allowing it to be a functional group (examples: cyclopropyl, cycohexyl)
Cycloalkylmethyl	a cylcoalkane with a methyl group attached
Cycloalkylethyl	a cycloalkane with an ethyl group attached
Halobenzyl	a benzyl with one or more halogens attached
Haloalkyl	an alkyl with one or more halogens attached
Halogens	a specific group in the periodic chart consisting of five elements: Fluorine (F), Chlorine (Cl), Bromine(Br), Iodine (I), and Astatine (At)
Phenyl	a benzene ring where one hydrogen is removed allowing it to be a functional group

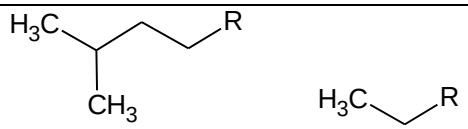
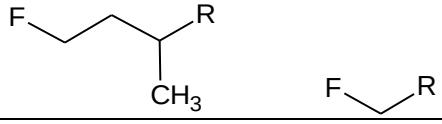
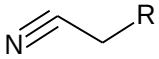
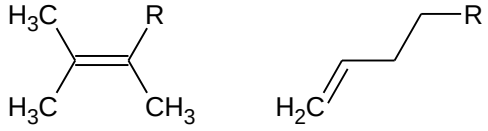
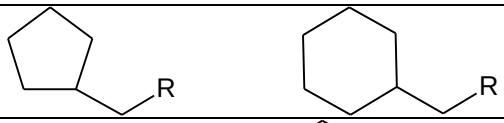
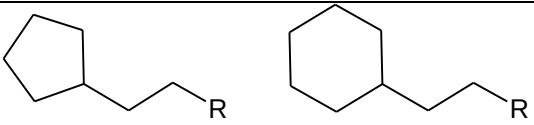
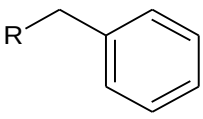
Structures:

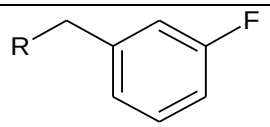
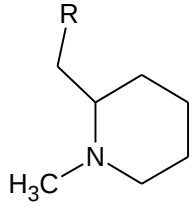
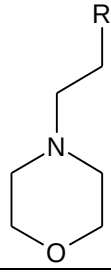
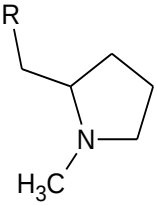
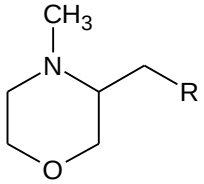
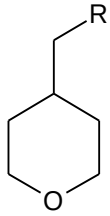
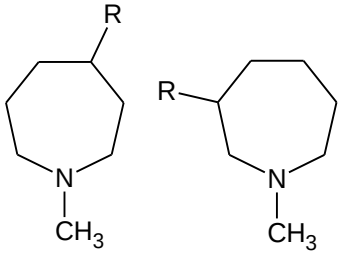
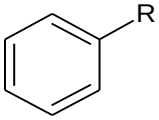
The following table provides structures for groups that will be found in the structure of the chemical groups defined in statute. This information is provided for reference purposes.

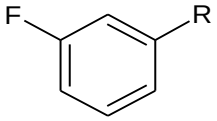
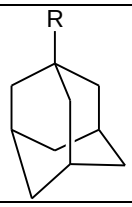
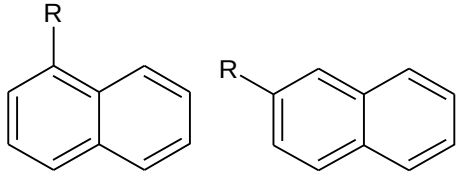
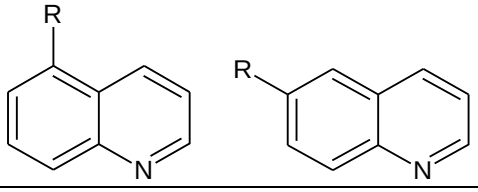
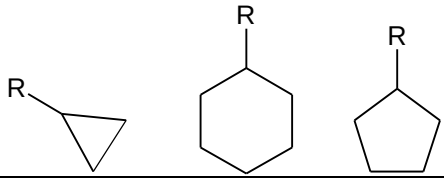
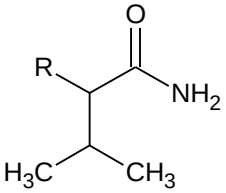
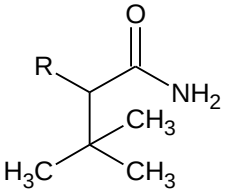
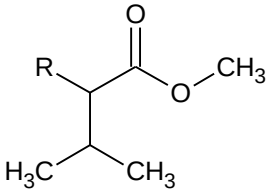
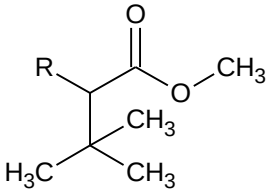
Group in Base Structure	Structure
Indole	
Naphthyl	
Pyrrole	
Indene	
Phenyl	
Phenol	
Cyclohexyl	
Cyclopropoyl	
Carboxamide	
Carboxylate	

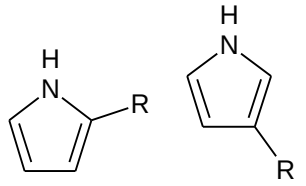
Benzimidazole	
Adamantyl	
Carbazole	
Methanone	

The following table provides structures for the substitute groups listed in statute. Where the functional group attaches to the compound is designated by "R".

Functional Groups	**Structure**
alkyl	
haloalkyl	
cyanoalkyl	
alkenyl	
cycloalkylmethyl	
cycloalkylethyl	
benzyl	

halobenzyl	
1-(N-methyl-2-piperidinyl)methyl	
2-(4-morpholinyl)ethyl	
1-(N-methyl-2-pyrrolidinyl)methyl	
1-(N-methyl-3-morpholinyl)methyl	
(tetrahydropyran-4-yl)methyl	
1-methylazepanyl	
phenyl	

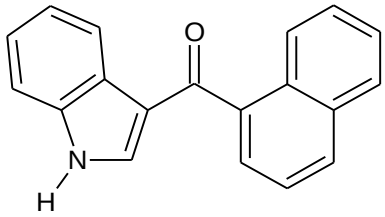
halophenyl	
adamantyl	
naphthyl	
quinolinyl	
cycloalkyl	
1-amino-3-methyl-1-oxobutan-2-yl	
1-amino-3,3-dimethyl-1-oxobutan-2-yl	
1-methoxy-3-methyl-1-oxobutan-2-yl	
1-methoxy-3,3-dimethyl-1-oxobutan-2-yl	

pyrrole	

Individual Steps of Protocol:

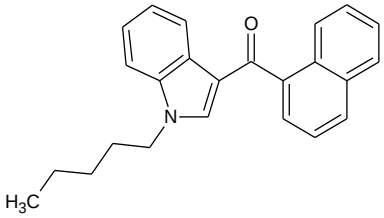
After the compound has been conclusively identified, determine if the compound is specifically listed in either state or federal schedules. If it is specifically listed, the compound will be reported without determining if it falls into one of the groups listed. If the compound is not specifically listed in state or federal schedules, locate the structure of the compound from a reputable source (ie manufacturer, DEA, SWGDRUG, Forendex) and determine, based on its structure, if it is a member of one of the following chemical groups as defined by 63 O.S. Section 2-204 Section G.

1. Naphthoylindoles (63 O.S. Section 2-204 Section G.1): any compound containing a 3-(1-naphthoyl)indole structure with or without substitution at the nitrogen atom of the indole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not further substituted on the indole ring to any extent, and whether or not substituted on the naphthyl ring to any extent.

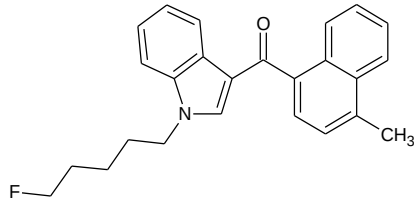


3-(1-naphthoyl)indole

Examples of Naphthoylindoles:

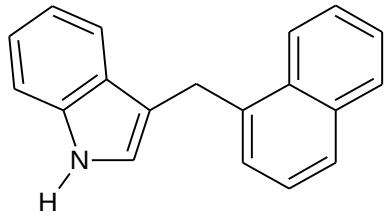


1-pentyl-3-(1-naphthoyl)indole
JWH-018



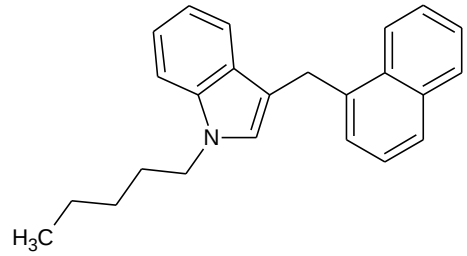
1-(5-fluoropentyl)-3-(4-methyl-1-naphthoyl)
MAM-2201

2. Naphthylmethylindoles (63 O.S. Section 2-204 Section G.2): any compound containing a 1H-indol-3-yl-(1-naphthyl)methane structure with or without substitution at the nitrogen atom of the indole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3-morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not further substituted on the indole ring to any extent, and whether or not substituted on the naphthyl ring to any extent.



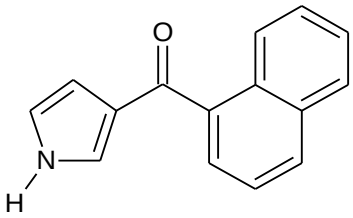
1H-indol-3-yl-(1-naphthyl)methane

Example of a Naphthylmethylindole:



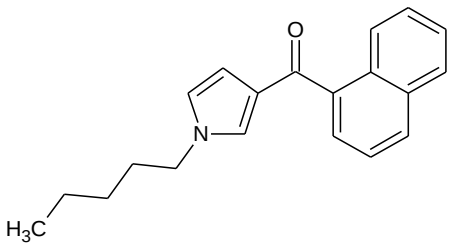
(1-pentylindol-3-yl)(1-naphthyl)methane
JWH-175

3. Naphthoylpyrroles (63 O.S. Section 2-204 Section G.3): any compound containing a 3-(1-naphthoyl)pyrrole structure with or without substitution at the nitrogen atom of the pyrrole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not further substituted on the pyrrole ring to any extent, and whether or not substituted on the naphthyl group to any extent.

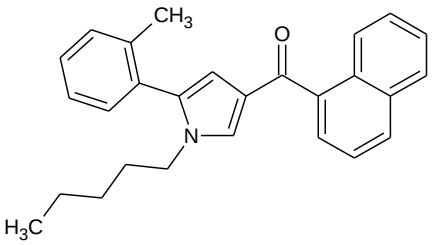


3-(1-napthoyl)pyrrole

Examples of Naphthoypyrroles:

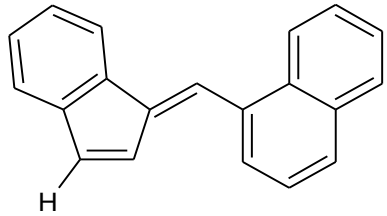


1-hexyl-2-phenyl-4-(1-naphthoyl)pyrrole
JWH-147



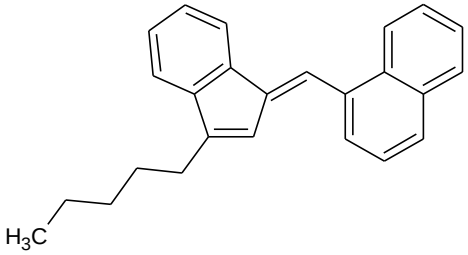
1-pentyl-5-(2-methylphenyl)-3-(1-naphthoyl)pyrrole
JWH-370

4. Naphthylideneindenes (63 O.S. Section 2-204 Section G.4): any compound containing a 1-(1-naphthylmethylene)indene structure with or without substitution at the 3-position of the indene ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not further substituted on the indene group to any extent, and whether or not substituted on the naphthyl group to any extent.



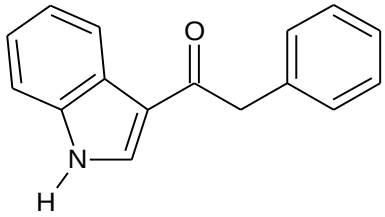
1-(1-naphthylmethylene) indene

Example of a Naphthylideneindene:



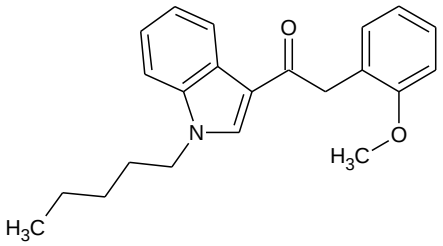
(1-[(3-pentyl)-1H-inden-1-ylidene)methyl]naphthalene
JWH-176

5. Phenylacetylindoles (63 O.S. Section 2-204 Section G.5): any compound containing a 3-phenylacetylindole structure with or without substitution at the nitrogen atom of the indole ring by alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group,, whether or not further substituted on the indole ring to any extent, and whether or not substituted on the phenyl ring to any extent.

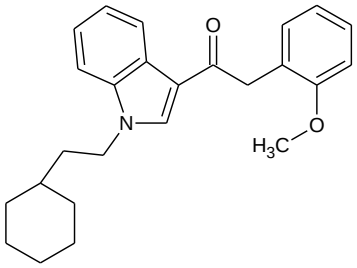


3-phenylacetylindole

Examples of Phenylacetyindoles:

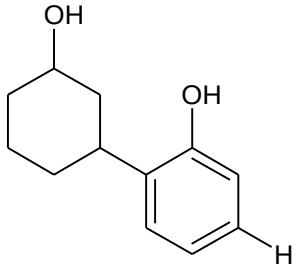


1-pentyl-3-(2-methoxyphenylacetyl)indole
JWH-250



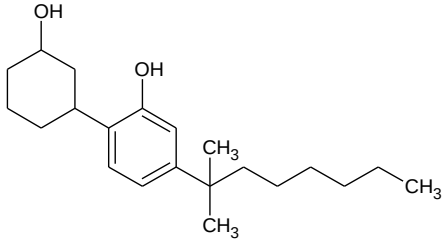
1-(2-cyclohexylethyl)-3-(2-methoxyphenylacetyl)indole
RCS-8

6. Cyclohexylphenols (63 O.S. Section 2-204 Section G.6): any compound containing a 2-(3-hydroxycyclohexyl)phenol structure with or without substitution at the 5-position of the phenolic ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl 1-methylazepanyl, phenyl, or halophenyl group, and whether or not further substituted on the cyclohexyl ring to any extent.



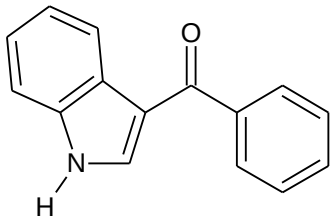
2-(3-hyroxycyclohexyl)phenol

Example of a Cyclohexylphenols:



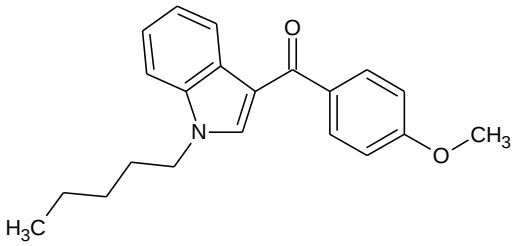
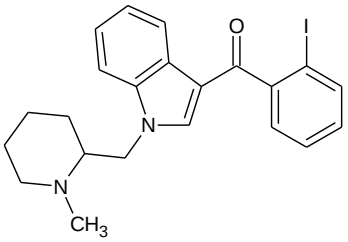
5-(1,1-dimethylheptyl)-2-[(1R,3S)-3-hydroxycyclohexyl]-phenol
CP-47, 497

7. Benzoylindoles (63 O.S. Section 2-204 Section G.7): any compound containing a 3-(benzoyl)indole structure with or without substitution at the nitrogen atom of the indole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not further substituted on the indole ring to any extent, and whether or not substituted on the phenyl group to any extent.

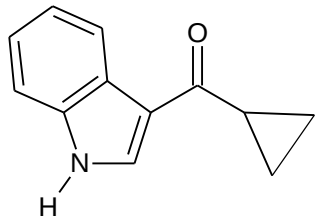


3-(1-benzoyl)indole

Examples of Benzoylindoles:

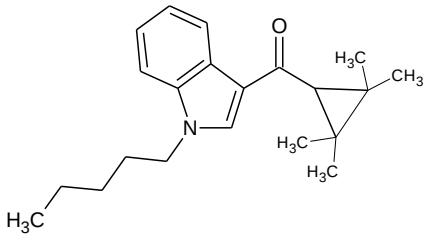
 1-pentyl-3-(4-methoxybenzoyl)indole RCS-4	 1-[1-(N-methyl-2-piperidinyl)methyl]-3-(2-iodobenzoyl) AM2233

8. Cyclopropoylindoles (63 O.S. Section 2-204 Section G.8): Any compound containing a 3-(cyclopropoyl)indole structure with substitution at the nitrogen atom of the indole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not further substituted in the indole ring to any extent and whether or not substituted in the cyclopropyl ring to any extent.

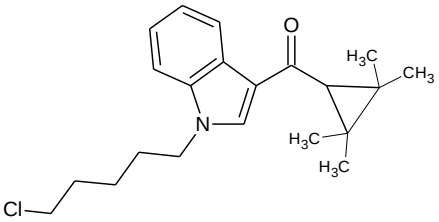


3-(cyclopropoyl)indole

Examples of Cyclopropoylindoles:

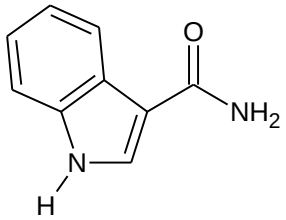


1-pentyl-3-(2,2,3,3-tetramethylcyclopropoyl)indole
UR-144



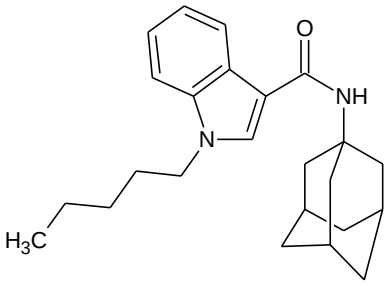
1-(5-chloropentyl)-3-(2,2,3,3-tetramethylcyclopropoyl)indole
5Cl-UR-144

9. Indole Amides (63 O.S. Section 2-204 Section G.9): Any compound containing a 1H-Indole-3-carboxamide structure with or without substitution at the nitrogen atom of the indole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not substituted at the carboxamide group by an adamantyl, naphthyl, phenyl, benzyl, quinolinyl, cycloalkyl, 1-amino-3-methyl-1-oxobutan-2-yl, 1-amino-3,3-dimethyl-1-oxobutan-2-yl, 1-methoxy-3-methyl-1-oxobutan-2-yl, 1-methoxy-3,3-dimethyl-1-oxobutan-2-yl or pyrrole group, and whether or not further substituted in the indole, adamantyl, naphthyl, phenyl, pyrrole, quinolinyl, or cycloalkyl rings to any extent.

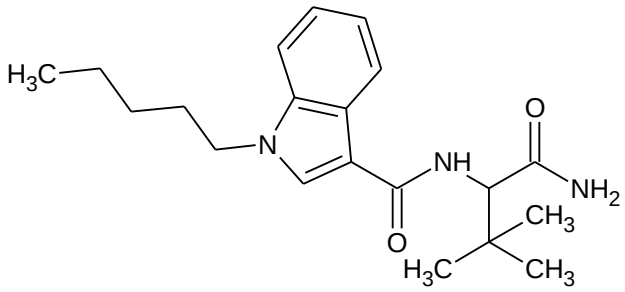


1H-Indole-3-carboxamide

Examples of Indole Amides:

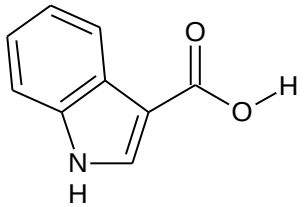


N-(1-adamantyl)-1-pentyl-1H-indole-3-carboxamide
2NE1



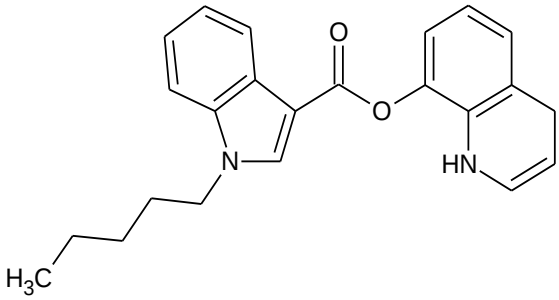
N-(1-amino-3,3-dimethyl-1-oxobutan-2-yl)-1-pentyl-1H-indole-3-carboxamide
AD-BICA

10. Indole Esters (63 O.S. Section 2-204 Section G.10): Any compound containing a 1H-Indole-3-carboxylate structure with or without substitution at the nitrogen atom of the indole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not substituted at the carboxylate group by an adamantyl, naphthyl, phenyl, benzyl, quinolinyl, cycloalkyl,1-amino-3-methyl-1-oxobutan-2-yl, 1-amino-3,3-dimethyl-1-oxobutan-2-yl, 1-methoxy-3-methyl-1-oxobutan-2-yl, 1-methoxy-3,3-dimethyl-1-oxobutan-2-yl, or pyrrole group, and whether or not further substituted in the indole, adamantyl, naphthyl, phenyl, pyrrole, quinolinyl, or cycloalkyl rings to any extent.

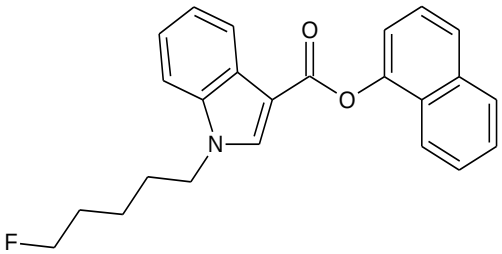


1H-Indole-3-carboxylate

Examples of Indole Esters:

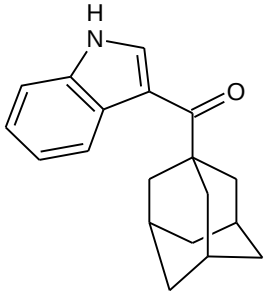


Quinolin-8-yl 1-pentyl-1H-indole-3-carboxylate
PB-22



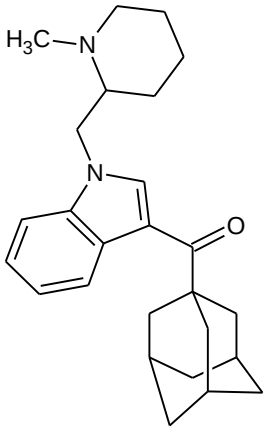
Naphthalen-1-yl 1-(5-fluoropentyl)-1H-indole-3-carboxylate
NM2201

11. Adamantanoylindoles (63 O.S. Section 2-204 Section G.11): Any compound containing a adamantanyl-(1H-indol-3-yl)methanone structure with or without substitution at the nitrogen atom of the indole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, whether or not further substituted in the indole ring to any extent and whether or not substituted in the adamantyl ring to any extent.

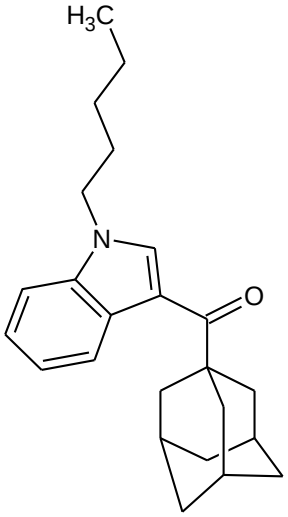


Adamantanyl-(1H-indol-3-yl)methanone

Examples of Adamantanoylindoles:

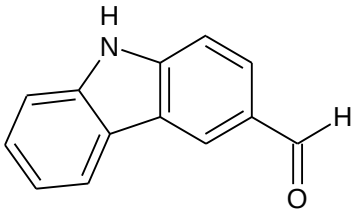


Adamantan-1-yl[1-[(1-methyl-2-piperidinyl)methyl]-1H-indol-3-yl]methanone AM1248



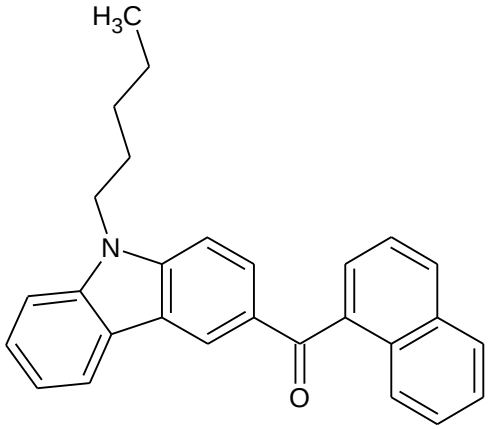
Adamantan-1-yl-(1-pentyl-1H-indol-3-yl)methanone
AB-001

12. Carbazole Ketones (63 O.S. Section 2-204 Section G.12): Any compound containing a (9H-carbazole-3-yl) methanone structure with or without substitution at the nitrogen atom of the carbazole ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3-morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, with substitution at the carbon of the methanone group by an adamantyl, naphthyl, phenyl, benzyl, quinolinyl, cycloalkyl,1-amino-3-methyl-1-oxobutan-2-yl, 1-amino-3,3-dimethyl-1-oxobutan-2-yl, 1-methoxy-3-methyl-1-oxobutan-2-yl,1-methoxy-3,3-dimethyl-1-oxobutan-2-yl, or pyrrole group, and whether or not further substituted at the carbazole, adamantyl, naphthyl, phenyl, pyrrole, quinolinyl, or cycloalkyl rings to any extent.



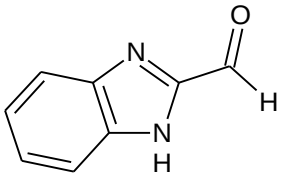
(9H-carbazole-3-yl) methanone

Example of Carbazole Ketones:



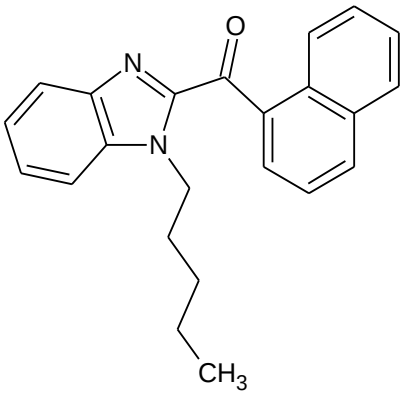
Naphthalen-1-yl(9-pentyl-9H-carbazol-3-yl)methanone
EG-018

13. Benzimidazole Ketones (63 O.S. Section 2-204 Section G.13): Any compound containing a (benzimidazole-2-yl)methanone structure with or without substitution at either nitrogen atom of the benzimidzaol ring by an alkyl, haloalkyl, cyanoalkyl, alkenyl, cycloalkylmethyl, cycloalkylethyl, benzyl, halobenzyl, 1-(N-methyl-2-piperidinyl)methyl, 2-(4-morpholinyl)ethyl, 1-(N-methyl-2-pyrrolidinyl)methyl, 1-(N-methyl-3- morpholinyl)methyl, (tetrahydropyran-4-yl)methyl, 1-methylazepanyl, phenyl, or halophenyl group, with substitution at the carbon of the methanone by an adamantyl, naphthyl, phenyl, benzyl, quinolinyl, cycloalkyl,1-amino-3-methyl-1-oxobutan-2-yl, 1-amino-3,3-dimethyl-1-oxobutan-2-yl, 1-methoxy-3-methyl-1-oxobutan-2-yl,1-methoxy-3,3-dimethyl-1-oxobutan-2-yl, or pyrrole group, and whether or not further substituted in the benzimidazole, adamantyl, naphthyl, phenyl, pyrrole, quinolinyl, or cycloalkyl rings to any extent.

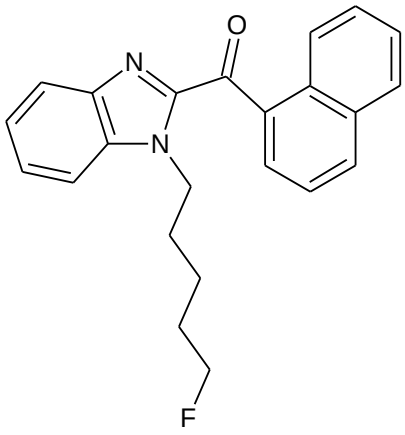


(benzimidazole-2-yl)methanone

Examples of Benzimidazole Ketones:



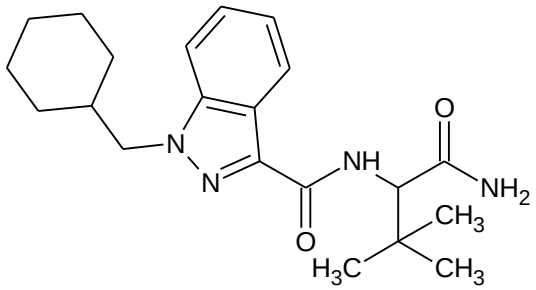
Naphthalen-1-yl(1-pentyl-1H-benzo[d]imidazol-2-yl)methanone
(JWH-018 benzimidazole analog)



(1-(5-fluoropentyl)-1H-benzo[d]imidazol-2-yl)(naphthalen-1-yl)methanone
FUBIMINA

14. Modified by Replacement (63 O.S. Section 2-204 Section G.14): any compound in one of the above groups (1-13) that is modified by replacement of a carbon with nitrogen in the indole, naphthyl, indene, benzimidazole, or carbazole ring.

    Example of a modified by replacement:



N-(1-amino-3,3-dimethyl-1-oxbutan-2-yl)-1-(cyclohexylmethyl)-1H-indazole-3-carboxamide
MAB-CHMINACA

Protocol Notes:

1. The case record must include documentation of the classification assigned to each compound. For example, case notes may state: 5-Fluoro NNEI is an indole amide substituted at the nitrogen in the indole ring with a haloalkyl and substituted at the carboxamide group with a napthyl group. An attached document from the Technical Manager, or their designee, with documentation regarding classification of compound, maybe be substituted for case notes. The classification documents can be found at \\VM-FSC-FILES\ContSub\Synthetic Cannabinoids Classification

2. Specifically listed in Oklahoma statute, includes compounds listed in Schedules I-V, including the examples listed in 63 O.S. Section 2-204 Section G. For example, if AM-2232 is conclusively identified, it would be reported as 1-(4-cyanobutyl)-3-(1-naphthoyl)indole (AM-2232), Schedule I.

3. Report writing should mirror statute. If the chemical name is listed in statute, it should be included in the results section of the report. For example, AM-2201 is listed in 63 O.S. Section 2-204 Section F as AM-2201, but also in 63 O.S. Section 2-204 Section G as 1-(5-fluoropentyl)-3-(1-naphthoyl)indole (AM2201). This would be reported as 1-(5-fluoropentyl)-3-(1-naphthoyl)indole (AM2201), Schedule I.

4. If the same compound is listed in Federal statute and Oklahoma statute, it will be reported as it appears in Oklahoma statute.

Recommended Report Wording:

1. When the compound has been conclusively identified and is specifically listed in state or federal statute, report as:

   Item #: URB597, Schedule I

   Item #: N-(1-Amino-3,3-Dimethyl-1-Oxobtan-2-yl)-1-Pentyl1H-Indazole-3-Carboxamide (ADB-PINACA), Schedule I

2. When the compound has been conclusively identified, is not specifically listed in state or federal statute, and does not belong in any of the fourteen chemical groups listed in 63 O.S. Section 2-204 Section G, report as:

   Item #: 5-Fluoro-CUMYL-PINACA.

3. When the compound has been conclusively identified, is not specifically listed in state or federal statute, and does belong in one of the ten chemical groups listed in 63 O.S. Section 2-204 Section G, report the name of the compound and provide the statute reference to which chemical group the compound belongs. For example:

   Item #: MAB-CHMINACA, Schedule I per 63 O.S. Section 2-204 G.14

DR-110 Extractions

Principle of the Protocol:

This protocol will serve as a reference collection of information for samples that are not the "norm" and require a specific extraction method or instrumental analysis for identification. Information regarding reporting or scheduling of samples with multiple schedules may be found in this protocol as well.

Associated Protocols:

DR-05 Pharmaceutical Identification by Literary Reference
DR-30 Gas Chromatography Analysis (Flame Ionization Detector)
DR-60 Drug Analysis by FTIR
DR-70 Gas Chromatograph Mass Spectrometer Methods for Drug Analysis
DR-75 Gas Chromatograph Infrared Detector Methods for Drug Analysis

Apparatus and Materials:

See appropriate Associated Protocol

Protocol / Recommended Report Wording / Interpretation of Test Data:

1. 2-CI and 2,5I-NBOH

   2,5I-NBOH converts to 2C-I in the inlet of the GC at 290 degrees. Therefore, 2CI and 25I-NBOH cannot be differentiated using the GC and GC/MS. The two compounds can be differentiated using an Infrared Detector. If 2C-I is tentatively identified via GC and GC/MS, the sample/case will need to be analyzed on the FTIR to confirm identification. **See Appendix C-1**.

   Note: 2,5I-NBOH and 2C-I-NBOH are synonyms.

2. 3-Fluoromethcathinone (3-FMC) and 4-Fluoromethcathinone (4-FMC)

   3-FMC and 4-FMC have the same retention times on a GC DB1 column. To separate and confirm the identification of the isomer(s) present, the sample and both the 3-FMC and 4-FMC standards will need to be analyzed on the GC using a 50 column and the method "Method1". **See Appendix C-2.**

3. 2, 3 and 4-Fluoroamphetamines

   As of November 1, 2013, 4-Fluoroamphetamine is a schedule I substance, therefore an isomer identification is required. 2, 3 and 4-fluoroamphetamines cannot be differentiated on the GC-MS. A sample with one of these compounds will need to be transferred to FSC to be analyzed on the GC-IRD. **See Appendix C-3.**

4. JWH-203 and JWH-250

   JWH-203 and JWH-250 co-elute on the GC DB-1 column. The compounds separate on the GC DB-50 column using the Drug1 method. The compounds will co-elute on the GC/MS, however it should be obvious that both compounds are present and should not be missed. JWH-203 will elute first, with the JWH-250 as a shoulder peak. **See Appendix C-4.**

5. JWH-201, JWH-250 and JWH-302

   The mass spectra for JWH-201, JWH-250 and JWH-302 are very similar. There are minor spectral differences and the GC-MS macro should print the spectra with all three on the page. The three compounds do separate on the DB-1 columns, but can achieve baseline separation on the DB-50 columns. **See Appendix C-5**.

6. Acid and Base Samples

   Evidence samples, those typically associated with clandestine laboratories, that when tested for pH are found to be a strong acid (pH~0) or a strong base (pH~14) should not be analyzed on the instruments. The following report wording should be used:

   Results:

   1A Consistent with an acid.
   1B Consistent with a base.

7. Codeine and Morphine

   Morphine is amphoteric and can react with NaOH to create codeine.

   Amphoteric drugs, such as morphine, require careful adjustment of pH for efficient extraction, using liquid-liquid procedures. Amphoteric compounds contain both acidic and basic functional groups. If the aqueous phase is too acidic or too basic, one of the functional groups will be ionized, and the extraction will be inefficient. The pH must be close to the isoelectric point for high recovery by liquid-liquid extraction. (Source: Drug Abuse Handbook, edited by Steven Karch, 2nd edition, 2007)

   Liquid or residue samples that are extracted and codeine is identified, must be re-analyzed using methanol or sodium bicarb/chloroform as a second extraction to determine if morphine or codeine is present. If the sample is a tablet that literary references codeine or a labeled cough syrup with codeine, the second extraction is not required. **See Appendix C-6**.

8. Codeine

   Per the Oklahoma State Board of Pharmacy, the following are the correct schedules for the reporting of Codeine.

   a. Codeine encountered in a liquid or cough syrup found in a container other than retail packaging or a prescription bottle, report: Codeine, Schedule V.
   b. Codeine-containing solid dosage forms when combined with other products, report: Codeine, Schedule III.
   c. Codeine only products, report: Codeine, Schedule II.

9. 6-Mononacetylmorphine

   Black tar is a product of postprocessing hydrolysis of heroin. Hydrolysis can be mediated by water or excess acid in heroin samples. 6-MAM content above 10% can be an indicator of postprocessing hydrolysis of heroin.

   The production of black tar heroin results in significant amounts of 6-MAM in the final product. 6-MAM is approximately 30 percent more active than diacetylmorphine itself. This

is why despite lower heroin content; black tar heroin may be more potent than some other forms of heroin.

Pharmacology: Hydrolysis converts heroin to its active metabolite, 6-monoacetylmorphine (6-MAM), and its inactive one, 3-MAM, followed by transformation to morphine.

Unless 6-MAM is the only controlled substance in a sample, 6-MAM is typically not identified in casework.

10. Oxymetholone

Oxymetholone is a schedule III substance in the state of Oklahoma. Oxymetholone, when put into solution, is prone to breaking down quickly. One of the breakdown products is Mestanolone, also a schedule III substance in the state of Oklahoma. It has been theorized that breakdown can occur in the injection port; however, current data indicates putting Oxymetholone in solution is the primary cause.

When analysis of a suspected Oxymetholone sample indicates the presence of other compounds or if Mestanolone is indicated, a second sample must be taken for analysis. The second sample should be placed in solution and analyzed on the GC followed immediately by analysis on the GC/MS. The sample should be placed at the front of the sequence for both instruments and analyzed as quickly as possible.

It is imperative that the false identification of Mestanolone, or any other compound, does not occur. Therefore, if both Oxymetholone and Mestanolone are present in a sample, do NOT report Mestanolone, as it is a known breakdown product of Oxymetholone.

11. Butalbital

Per the Oklahoma State Board of Pharmacy, Oklahoma does not have any exempt preparations of butalbital. All samples with butalbital are Schedule III.

a. Butalbital/acetaminophen/caffeine is specifically listed as Schedule III. If all three compounds are reported, all three will need to be identified.
    i. Report using:
        Butalbital/Acetaminophen/Caffeine, Schedule III
b. All other samples will fall under Title 63, §63-2-208. Schedule III: "3. Any substance which contains any quantity of a derivative of barbituric acid, or any salt of a derivative of barbituric acid."
    i. Report using:
        Butalbital, Schedule III *(only identify butalbital)*

12. 3.4-Methylenedioxymethamphetamine (3,4-MDMA) and Trifluoromethylphenylpiperazine (TFMPP)

When analyzed on the mass spectrometer, 3, 4-MDMA and TFMPP co-elute. The only indication both compounds are present is the presence of 230 and a few other ions present in the mass spectra for MDMA. For instructions on how to background subtract **see Appendix C-7.**

13. Trifluoromethylphenylpiperazine (TFMPP) Isomers

When analyzed on the DB-50 or DB-1columns, the three TFMPP isomers, 1,2-TFMPP, 1,3-TFMPP and 1,4-TFMPP elute in numerical order. A TFMPP ladder can be used on the GC for isomer determination; use GC method "TFMPP1". **See Appendix C-8**.

14. MAPB Compounds

5-MAPB has a 2, 3, 4, 5, 6, and 7 isomer. The GC and GCMS cannot differentiate them. The retention time, retention index, and mass specs are all almost identical. The GC-IRD can easily differentiate these isomers. Take the appropriate steps to have it identified by IRD.

15. Aspirin with Methamphetamine or MDMA

Clandestine tablets or residues in which **aspirin is identified** when using methanol as a solvent, must be re-extracted with NaOH/CHCl3. Aspirin will acetylate MDMA and methamphetamine in the injection port of the GC. If the concentration of MDMA and/or methamphetamine is too low, the aspirin can acetylate the entire amount of the drug, giving a false negative. Extracting the sample with NaOH/CHCl3 will remove the aspirin.

16. Methylone

Methylone is also known as 3,4-methylenedioxymethcathinone and 3,4-methylenedioxy-N-methyl-B-ketoamphetamine (name used in Title 63, Schedule I of Oklahoma Statutes). Street names include M1, MDMC and bk-MDMA.

17. Eutylone

Eutylone and caffeine will co-elute when using the DB-1 or any "1" column; separation can be achieved using a DB-50 column. If a DB-1 column is used for analysis on the GC, the analyst must include the caffeine scans from the mass spec. This will allow the technical reviewer to review the mass spec data for extra ions in caffeine, which is an indication of co-elution. A mass spec scan for eutylone/pentylone with extra ions is another indicator of co-elution.

If a sample(s) has an indication of caffeine/eutylone co-elution, extracting with NaOH into Hexanes will eliminate the caffeine. The extraction needs to be done fairly quickly to ensure keeping the caffeine to a minimum. The normal Drug methods on the mass spec can then be used to make the identification of eutylone (using the RI from the library).

18. Tenocycline (TCP) and Phencyclidine (PCP)

TCP and PCP will co-elute on the GC and the GC/MS, but can be separated using the following methods: GC use "PCPTCP" and on the GC/MS use "PCPTCP50". If a different split ratio is needed for the mass spec, see the technical manager for a new method. Due to the modification of the PCPTCP method, the retention indexes for PCP and TCP will not be within the requirements of the GC/MS macros and a Simple Deviation will be required to use the GC/MS retention times for a comparison for identification and not the Retention Index.

19. Eszopiclone and zopiclone

Eszopiclone is the active dextrorotatory stereoisomer of zopiclone. Eszopiclone is not marketed in Europe, but is commercially available in the United States as a Schedule IV controlled substance. Zopiclone is not commercially available in the United States and is a Schedule IV controlled substance; although it is readily available in other countries. Eszopiclone and zopiclone cannot be differentiated using the GC and GC/MS.

20. Tianeptine

Tianeptine as a relatively pure substance can be identified using the GC and FTIR. The standard must be run on the GC and the break down peaks compared to the sample. Tianeptine cannot be identified on the GC/MS, as the compound breaks down during analysis. Example of report wording for Tianeptine using the GC and FTIR:

Item#: Tianeptine, Schedule II

Tianeptine in a mixture, commonly seen in tablet form, cannot currently be separated for identification on the FTIR. Example of report wording for Tianeptine in a mixture:

Item#: Examination indicates the presence of Tianeptine, a non-controlled substance; however, this could not be confirmed by instrumental analysis.

21. Reporting of Ephedrine/Pseudoephedrine

Guideline 1:

a. When the charges being filed **are** either "Manufacturing of a Controlled Substance" or "Attempting to Manufacture a Controlled Substance"
b. There is indication of Ephedrine or Pseudoephedrine in the sample
c. The evidence/chemicals present indicate that methamphetamine was the drug to be produced for example: Solvents, Iodine, Red Phosphorous, Ammonia, Sodium or Lithium metal, Red Devil Lye, Muriatic Acid, Sulfuric Acid, two layered liquids, etc. Most or all of these items should be present.
d. Methamphetamine ***has been identified*** within the evidence/chemicals in the particular case
e. It is the analyst's opinion that methamphetamine was the drug to be made and the analyst's testimony will reflect this opinion

Report using one of the following examples:

Item#: Ephedrine, Schedule IV
or
Item#: Pseudoephedrine, Schedule V
or
Item#: Ephedrine/Pseudoephedrine, a precursor substance.

Note: Analysts need to make every effort to identify a precursor in each manufacturing case. Once a precursor has been identified the analyst may use reasonable judgment on additional samples whether the precursor should be identified.

Guideline 2:

a. When the charges being filed **are** either "Manufacturing of a Controlled Substance" or "Attempting to Manufacture a Controlled Substance"
b. There is indication of Ephedrine or Pseudoephedrine in the sample
c. Methamphetamine ***has not been identified*** within the evidence/chemicals in the particular case

Report using one of the following examples:

Item#: Ephedrine, Schedule IV
or
Item#: Pseudoephedrine, Schedule V

Guideline 3:

a. When the charges being filed are ***something other than*** "Manufacturing of a Controlled Substance", "Attempting to Manufacture a Controlled Substance", or "Possession of a Precursor Substance"
b. Other drugs are present in the ***sample*** with controlled substances of a higher schedule.

Report using one of the following examples:

Item#: Higher scheduled drug and Ephedrine, Schedule IV
or
Item#: Higher scheduled drug and Pseudoephedrine, Schedule V
or
Item#: Higher scheduled drug

Note: If analysis is begun on an individual sample that ***only*** contains Ephedrine or Pseudoephedrine, the analysis should be carried through and the determination made, regardless of what is in other samples of the submittal.

Guideline 4:

a. When the charges being filed are ***something other than*** "Manufacturing of a Controlled Substance" or "Attempting to Manufacture a Controlled Substance"
b. Other items are present that ***do not*** contain any controlled substances, or the item submitted is the only item in the case.

Report using one of the following examples:

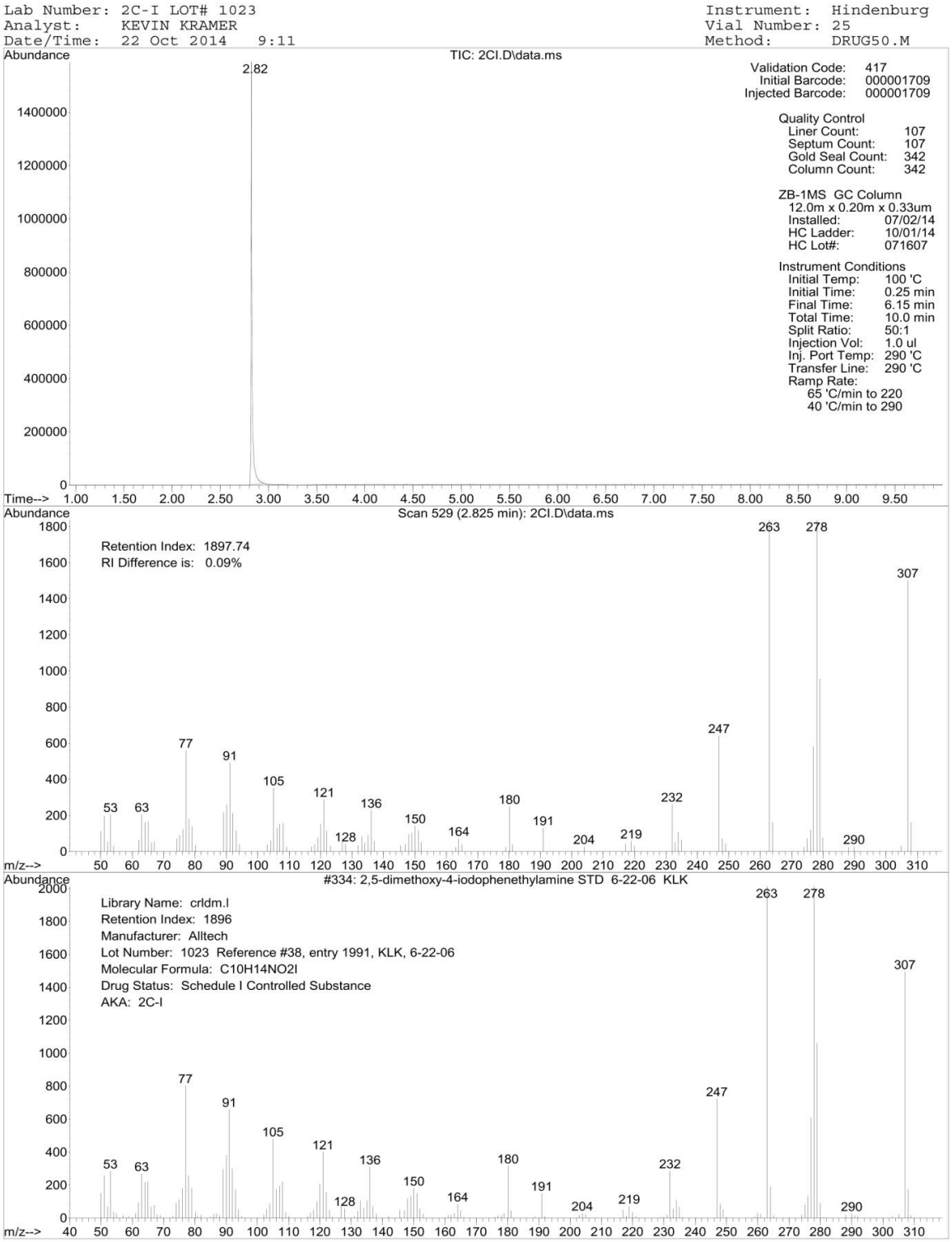
Item#: Ephedrine, Schedule IV
or
Item#: Pseudoephedrine, Schedule V

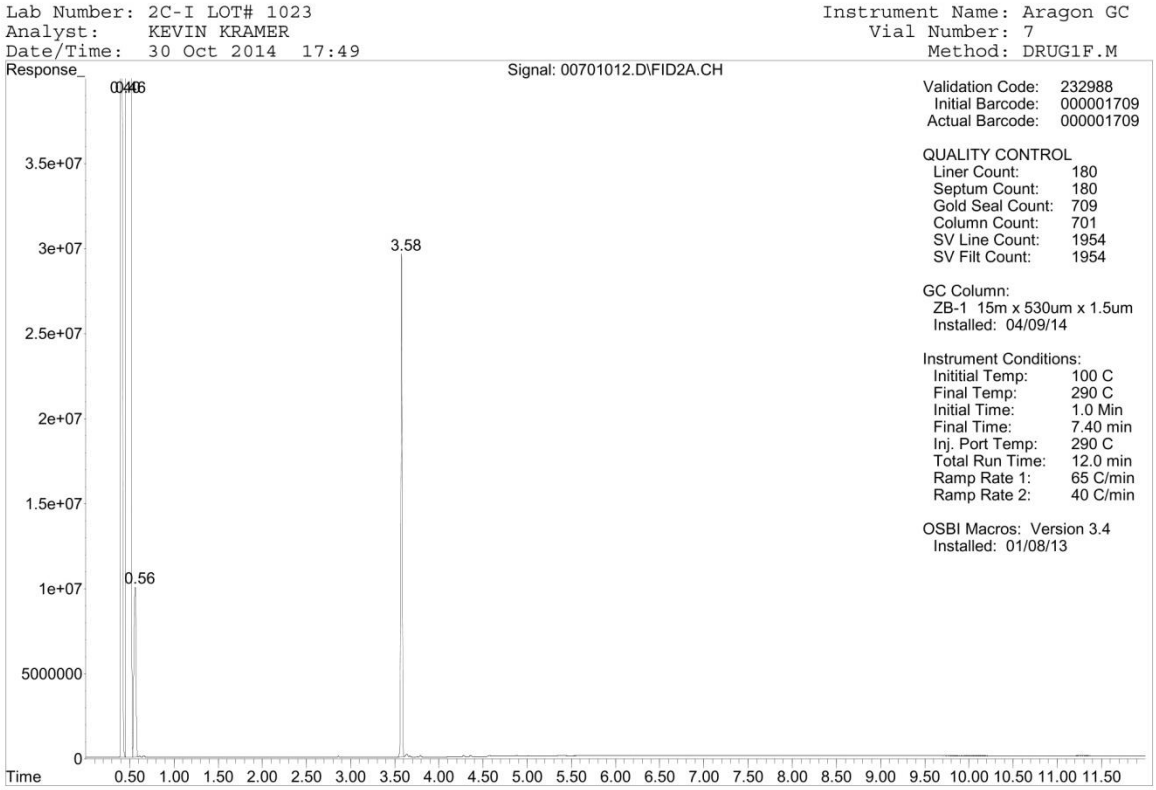
Note: If the item is the ***only item in the case*** and is positive for Ephedrine or Pseudoephedrine, the determination of which one must be made. A literary reference along with GCMS is not a confirmatory test procedure for the determination of Ephedrine

or Pseudoephedrine. A GC run utilizing method1 or method2 must be run to determine which is present, or the use of FTIR or GC/IRD.

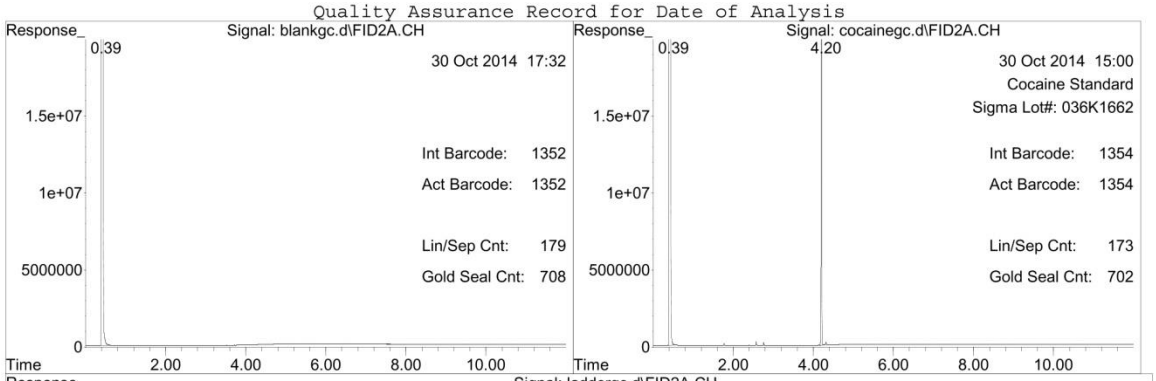
Appendix C: Extraction Data

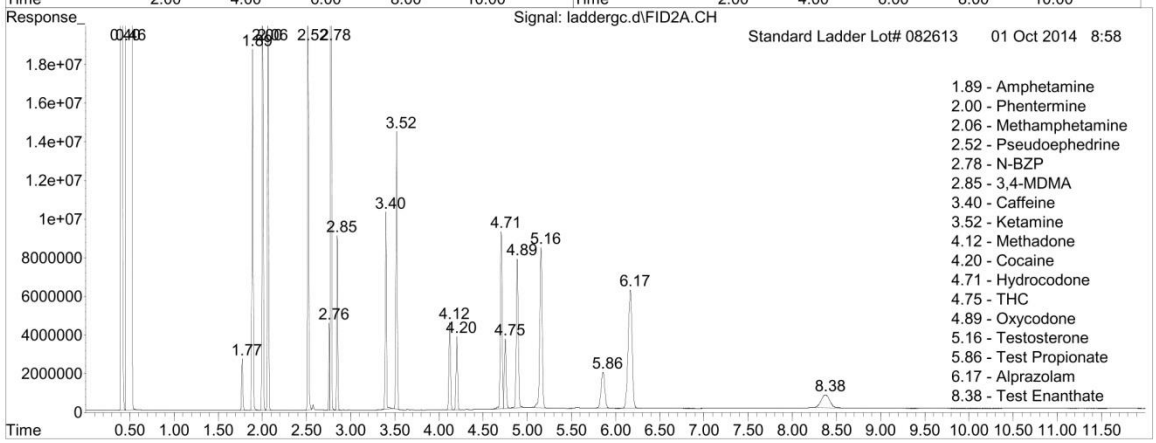
Appendix C-1: 2-CI and 2,5I-NBOH Data

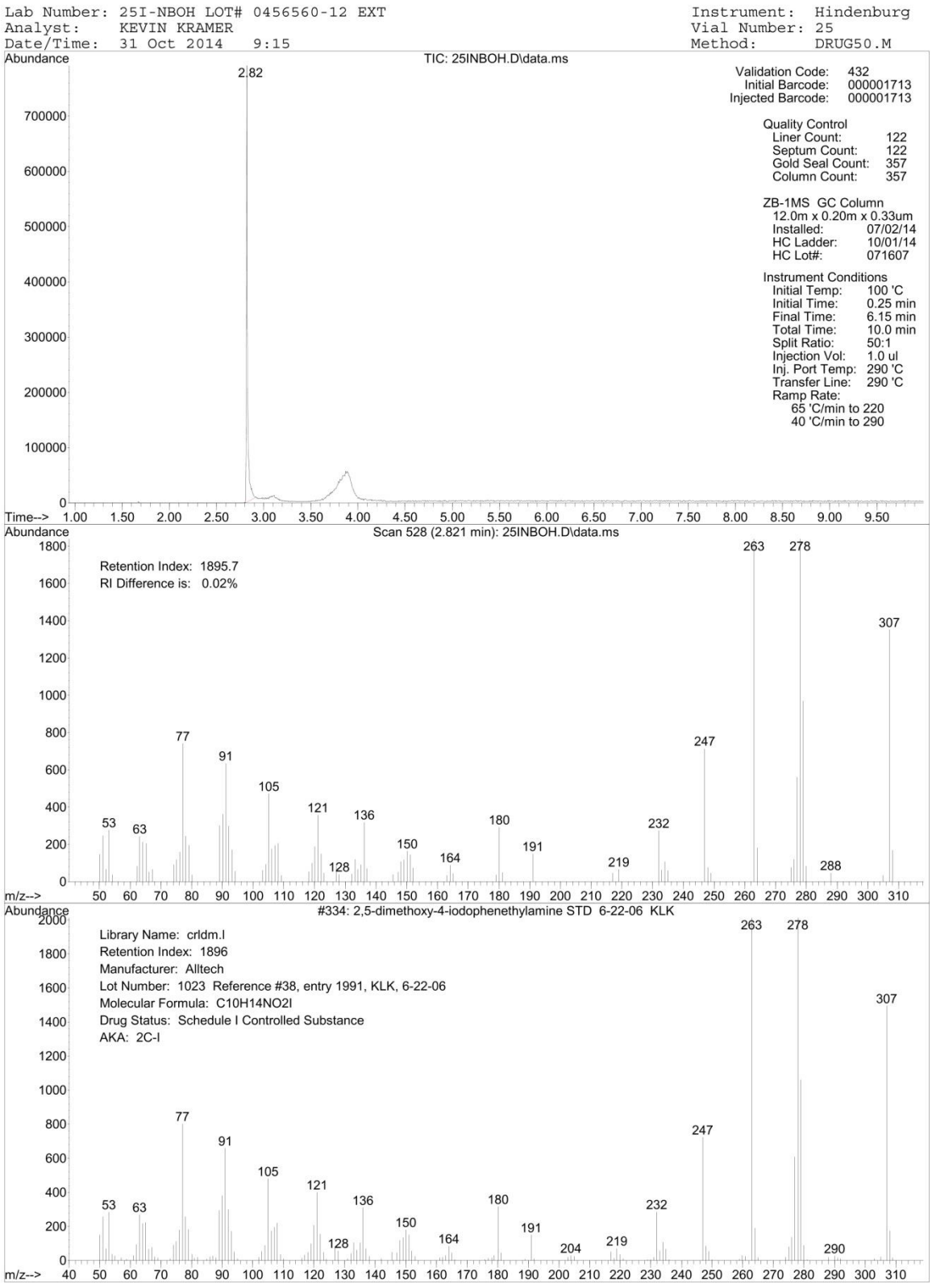


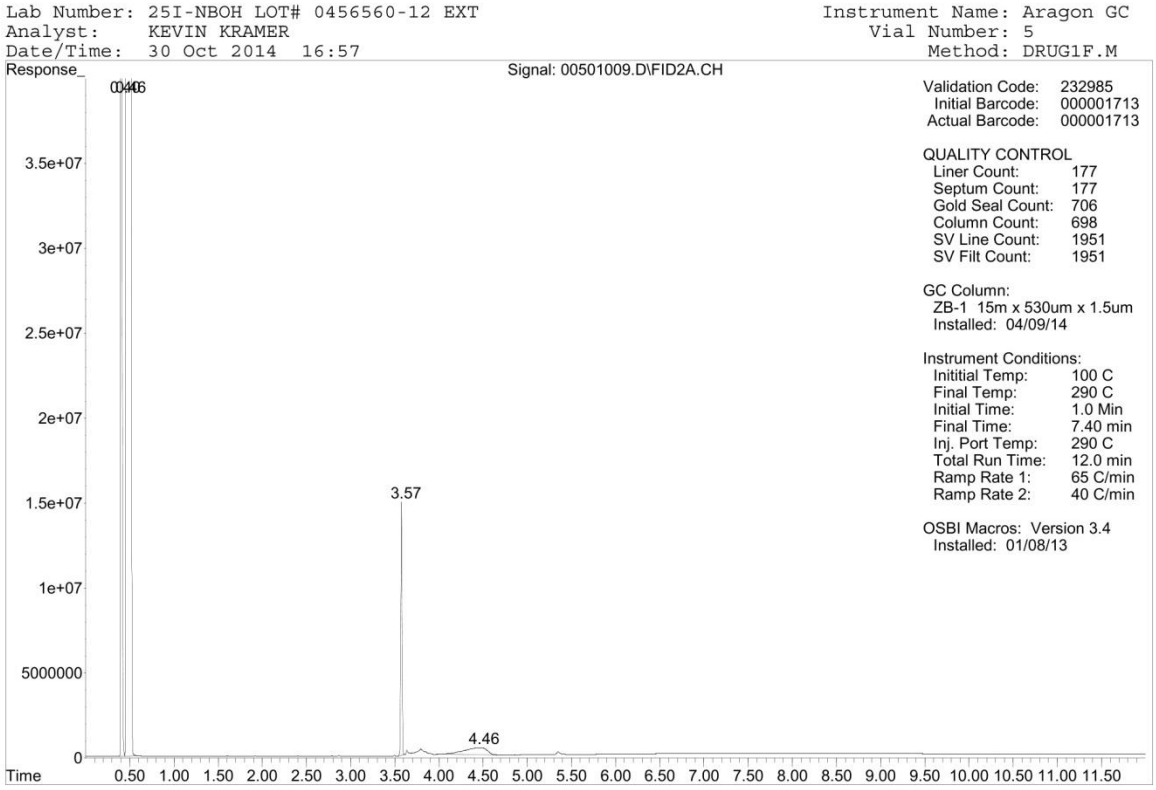


Quality Assurance Record for Date of Analysis

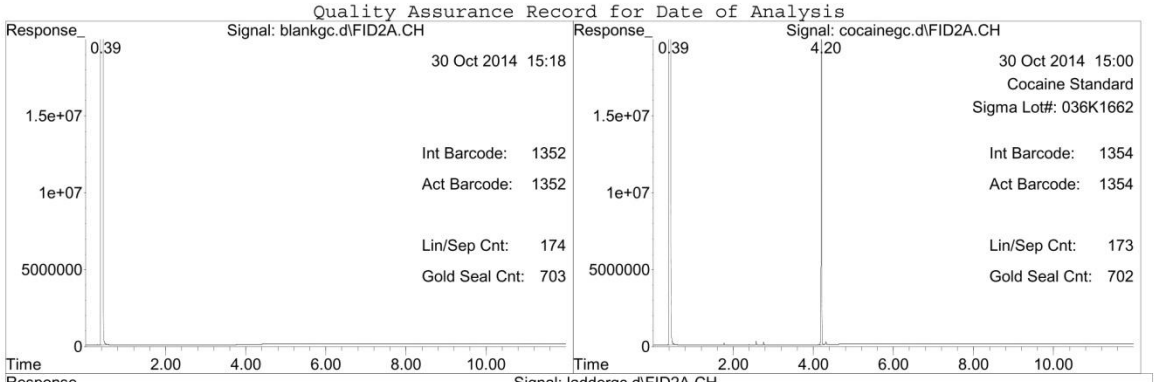


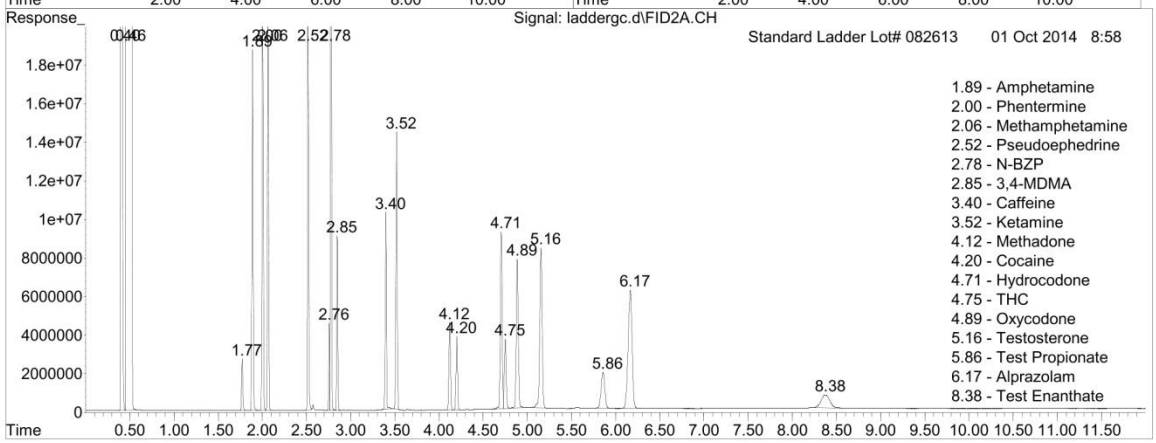


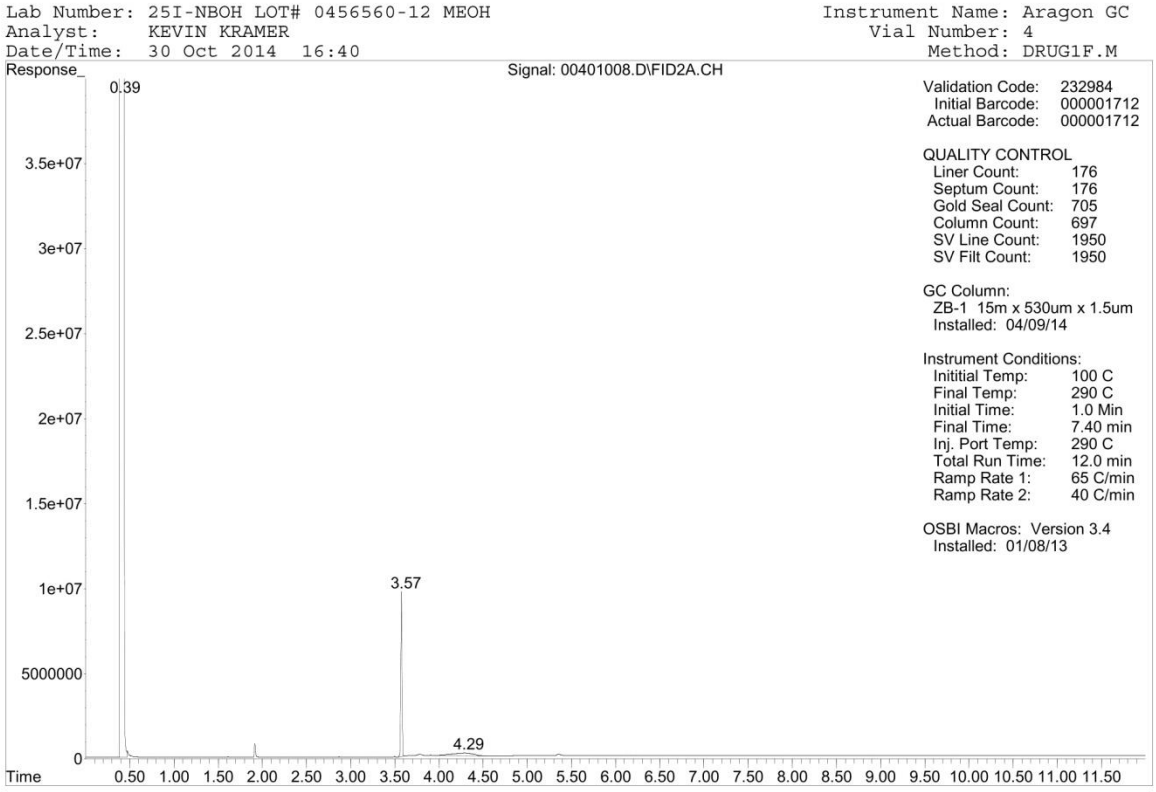

Lab Number: 25I-NBOH LOT# 0456560-12 EXT
Analyst: KEVIN KRAMER
Date/Time: 31 Oct 2014 9:15
Instrument: Hindenburg
Vial Number: 25
Method: DRUG50.M
TIC: 25INBOH.D\data.ms
Validation Code: 432
Initial Barcode: 000001713
Injected Barcode: 000001713
Quality Control
Liner Count: 122
Septum Count: 122
Gold Seal Count: 357
Column Count: 357
ZB-1MS GC Column
12.0m x 0.20m x 0.33um
Installed: 07/02/14
HC Ladder: 10/01/14
HC Lot#: 071607
Instrument Conditions
Initial Temp: 100 'C
Initial Time: 0.25 min
Final Time: 6.15 min
Total Time: 10.0 min
Split Ratio: 50:1
Injection Vol: 1.0 ul
Inj. Port Temp: 290 'C
Transfer Line: 290 'C
Ramp Rate:
65 'C/min to 220
40 'C/min to 290
Abundance
Time-->
Scan 528 (2.821 min): 25INBOH.D\data.ms
Retention Index: 1895.7
RI Difference is: 0.02%
m/z-->
#334: 2,5-dimethoxy-4-iodophenethylamine STD 6-22-06 KLK
Library Name: crldm.l
Retention Index: 1896
Manufacturer: Alltech
Lot Number: 1023 Reference #38, entry 1991, KLK, 6-22-06
Molecular Formula: C10H14NO2I
Drug Status: Schedule I Controlled Substance
AKA: 2C-I



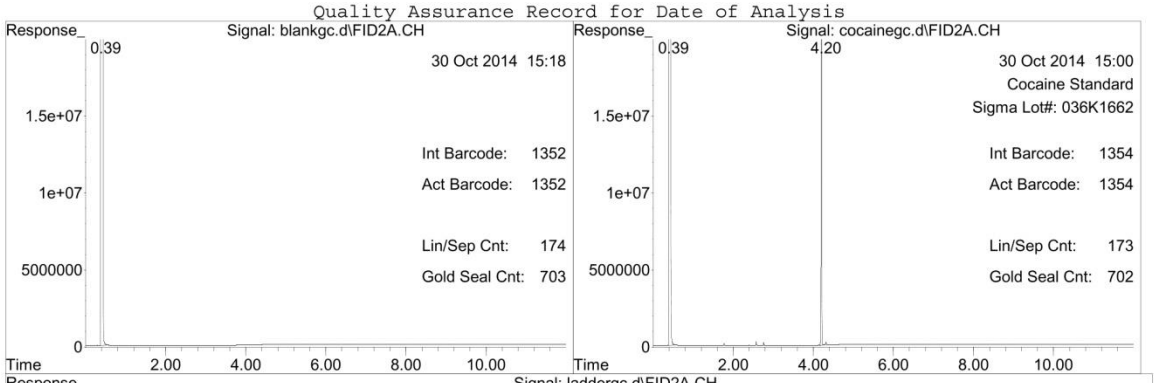
Quality Assurance Record for Date of Analysis

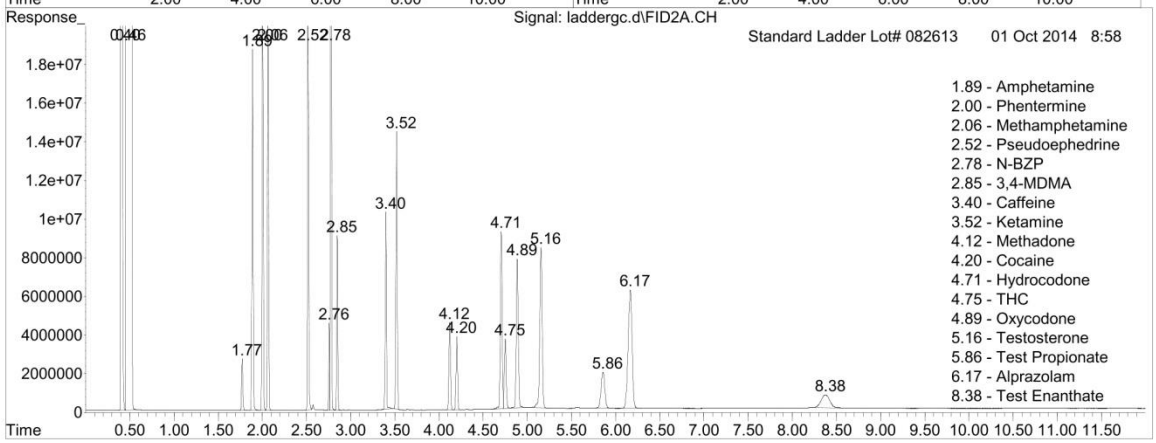


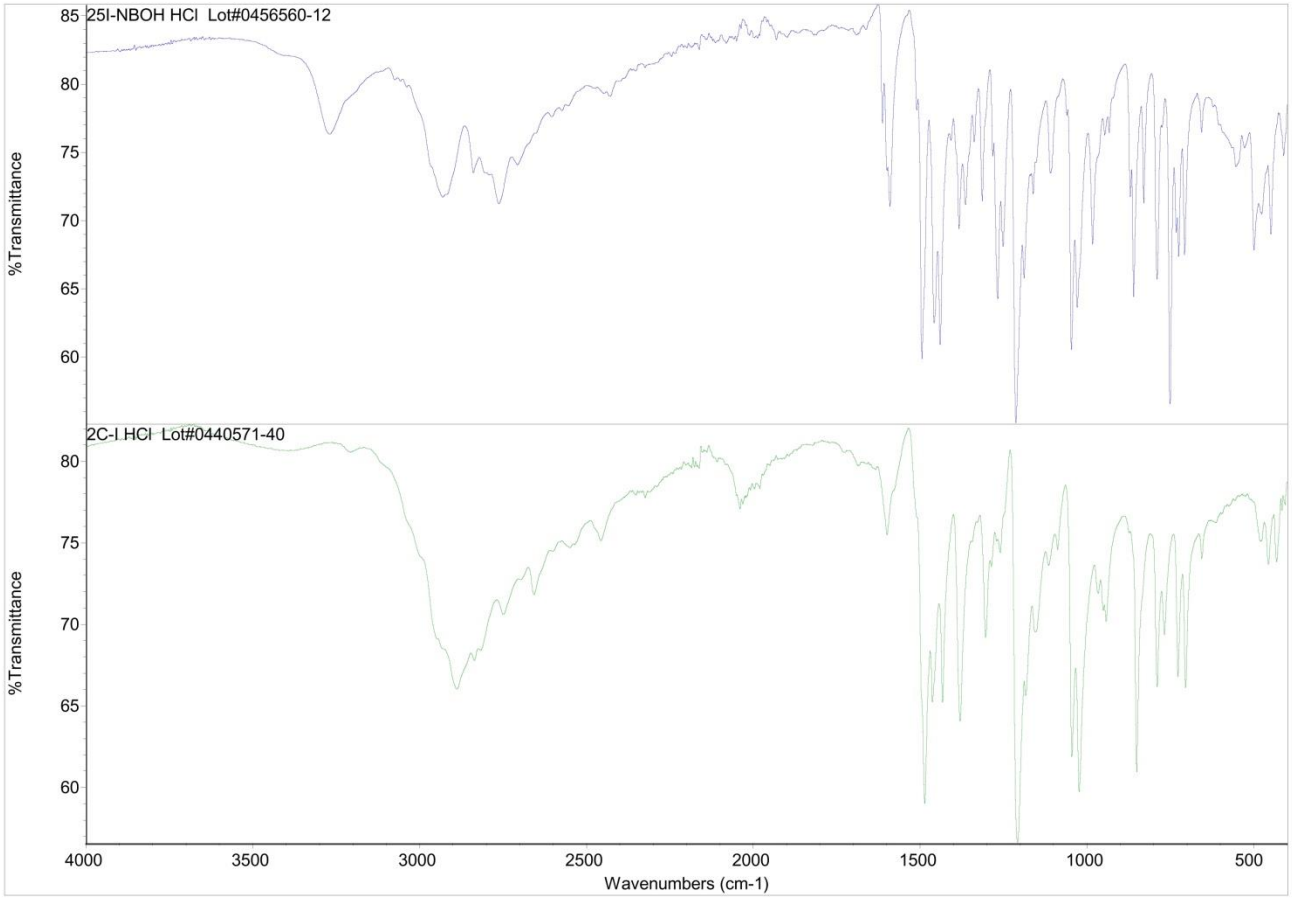




Quality Assurance Record for Date of Analysis

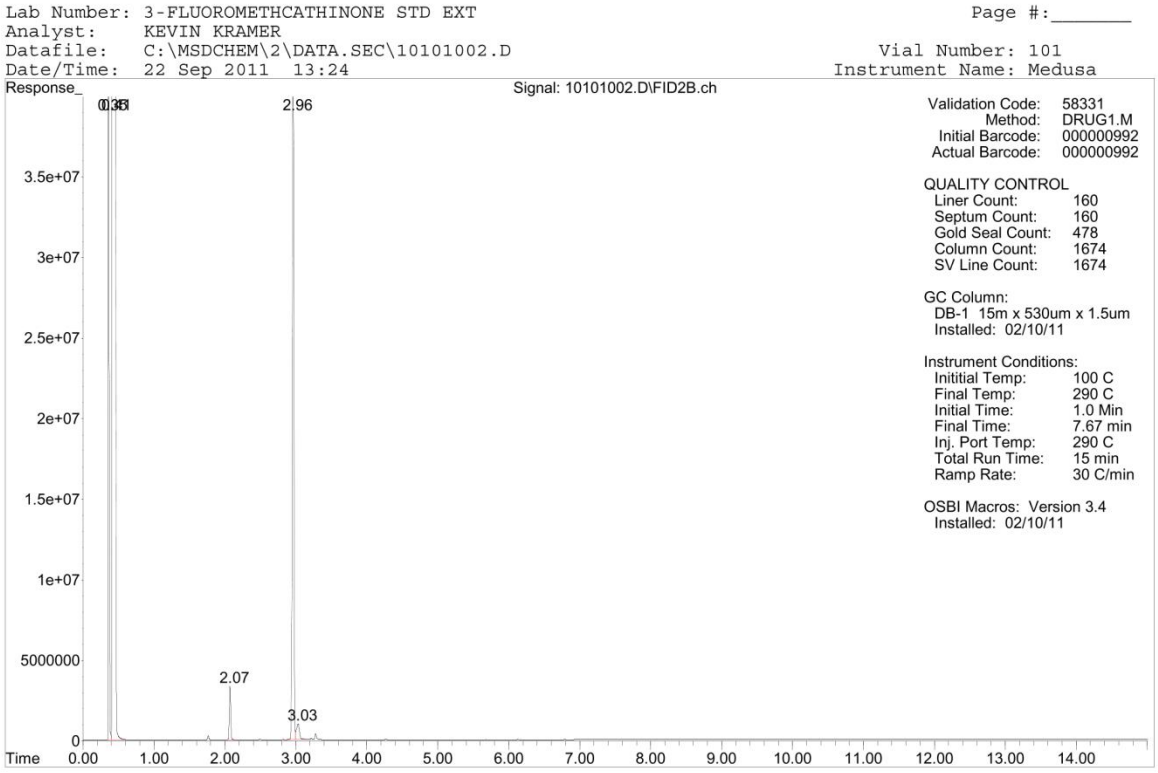




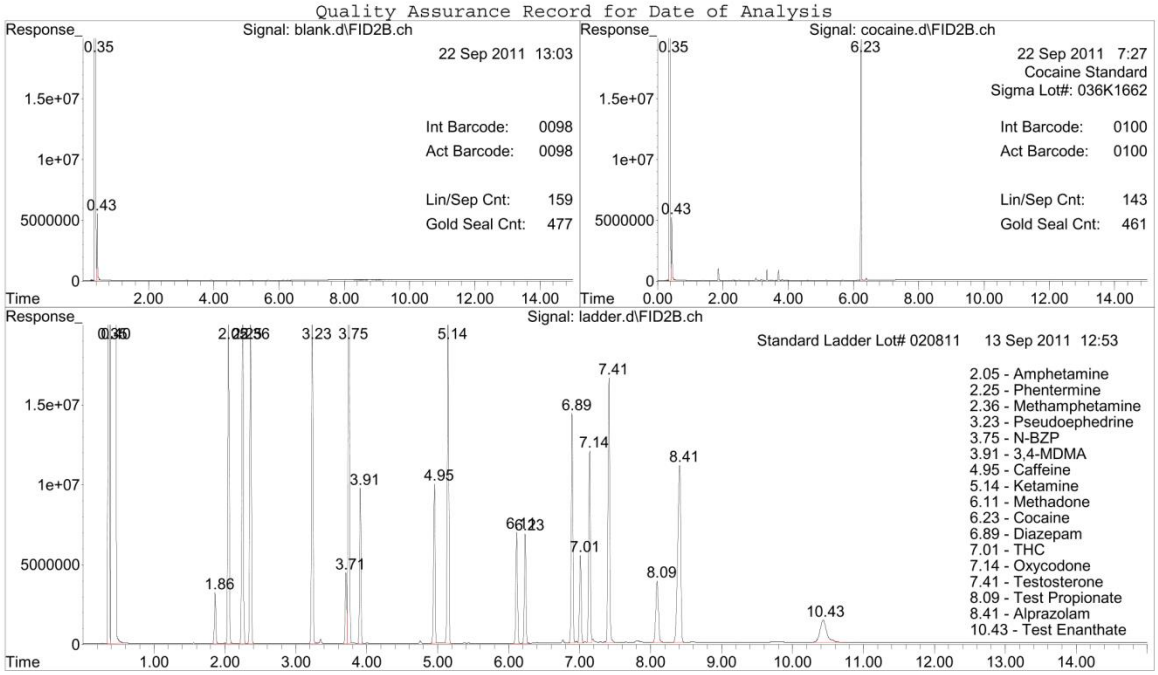

25I-NBOH HCl Lot#0456560-12
%Transmittance
85
80
75
70
65
60
2C-I HCl Lot#0440571-40
%Transmittance
80
75
70
65
60
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)

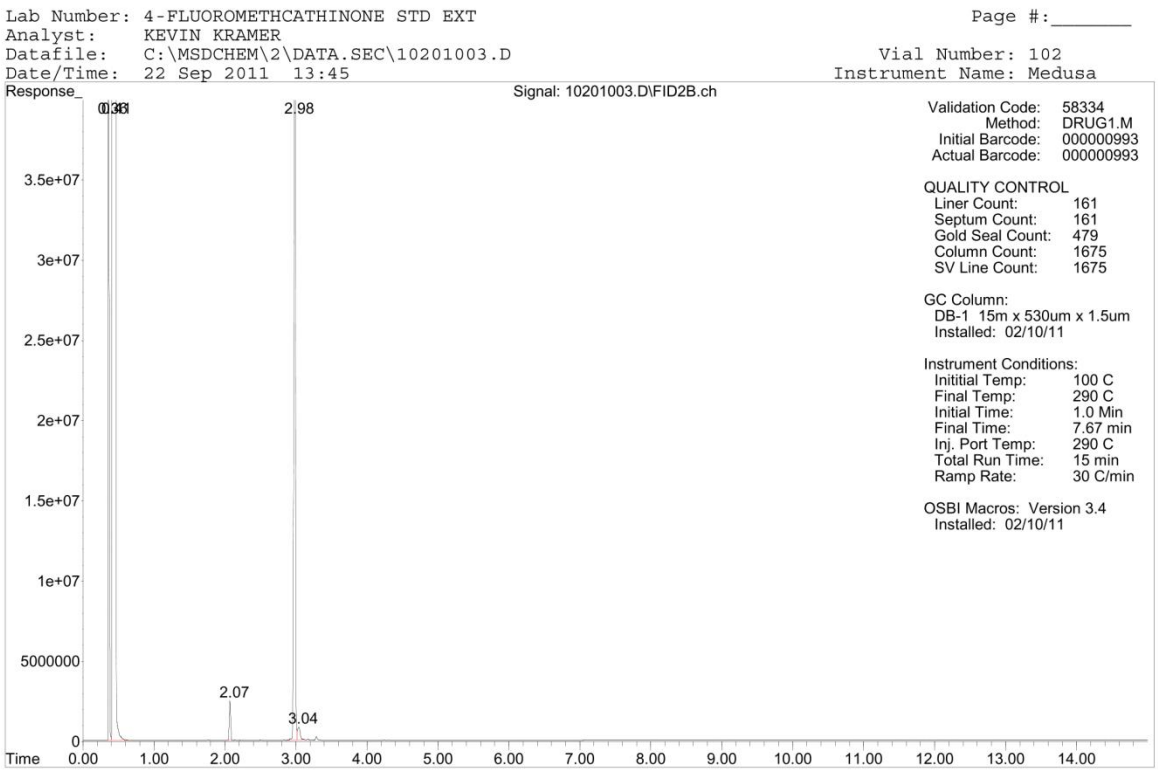
Appendix C: Extraction Data

Appendix C-2: 3-FMC and 4-FMC Data

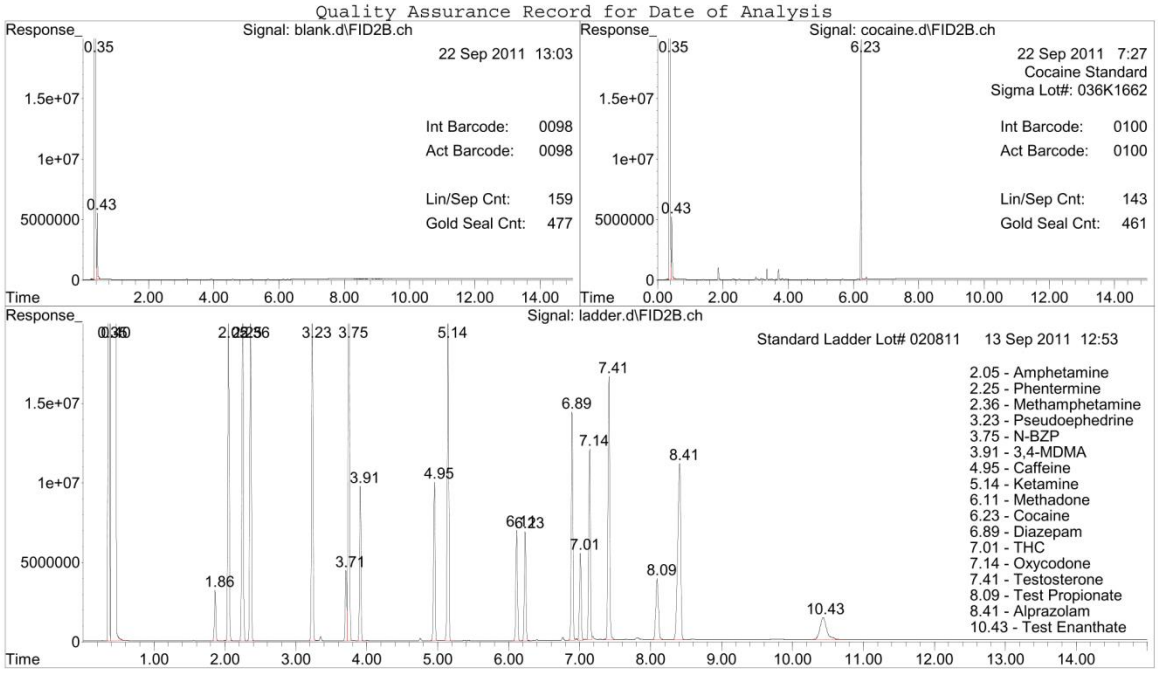


Quality Assurance Record for Date of Analysis

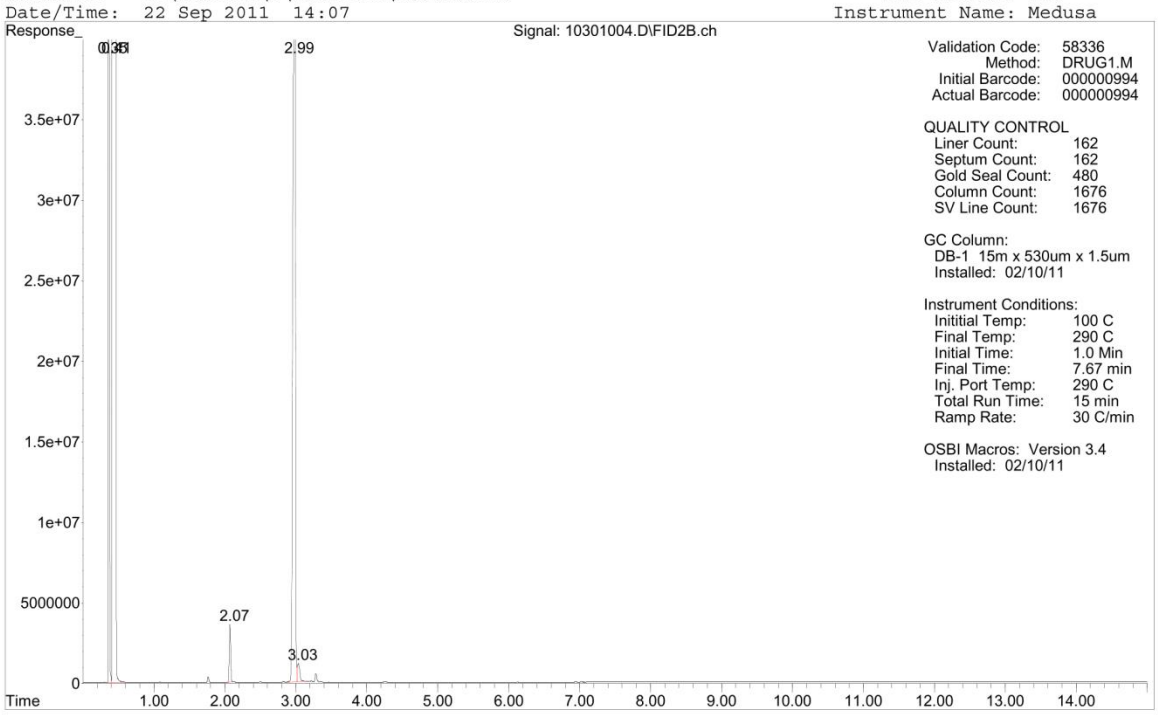




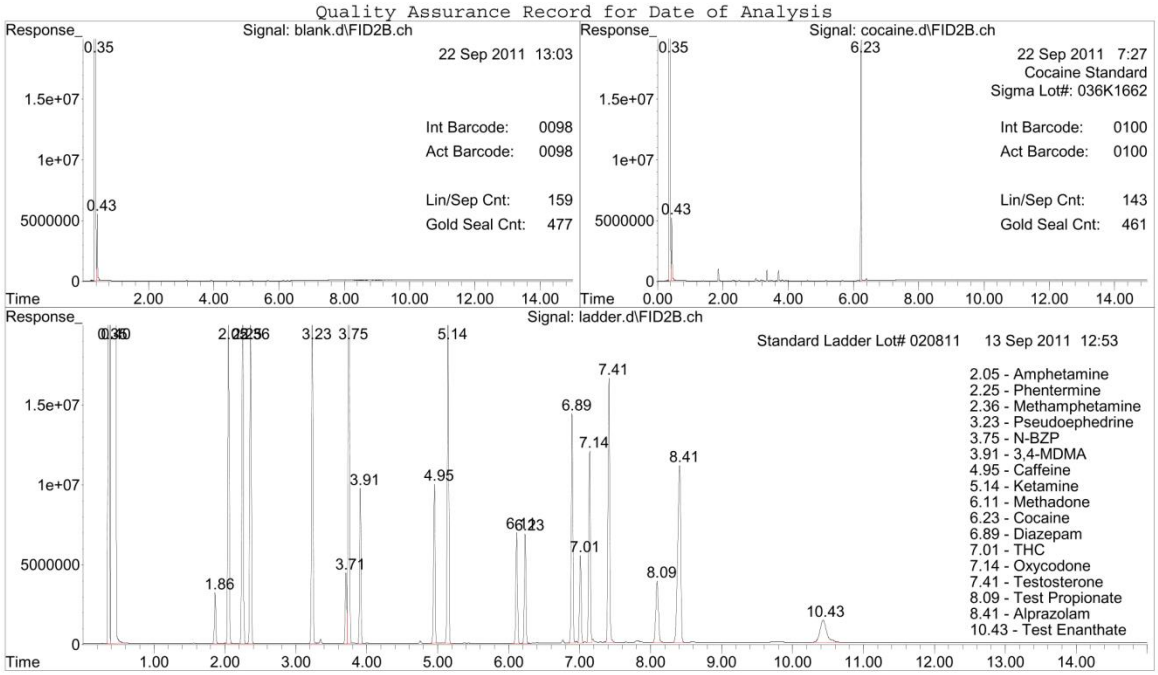
Quality Assurance Record for Date of Analysis



Lab Number: 3 and 4-FLUOROMETHCATHINONE STD MIX EXT Page #:_____
Analyst: KEVIN KRAMER
Datafile: C:\MSDCHEM\2\DATA.SEC\10301004.D Vial Number: 103
Date/Time: 22 Sep 2011 14:07 Instrument Name: Medusa

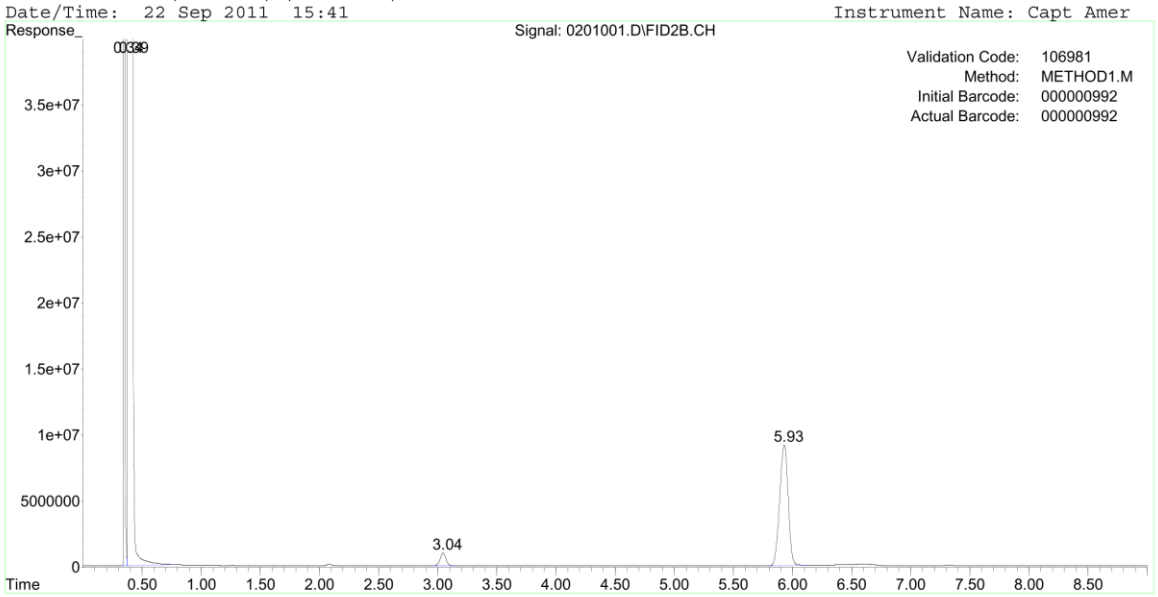


Quality Assurance Record for Date of Analysis



Lab Number: 3-FMC STD EXT KLK
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\2\DATA.SEC\0201001.D
Date/Time: 22 Sep 2011 15:41

Page #:_____
Vial Number: 76
Instrument Name: Capt Amer



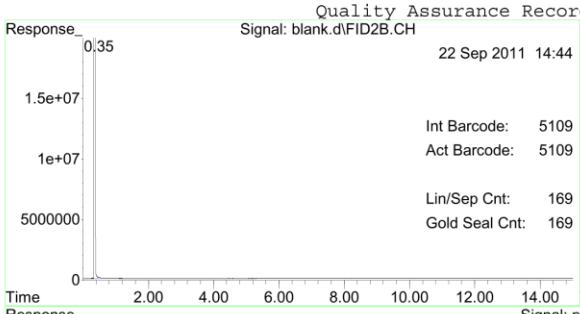
INSTRUMENT CONDITIONS

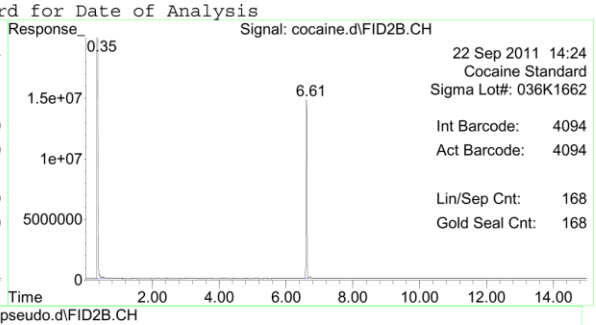
Initial temp:	100 C	Final temp:	140 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	0 min	Total Run Time:	9 min
		Ramp Rate:	5 C/min		

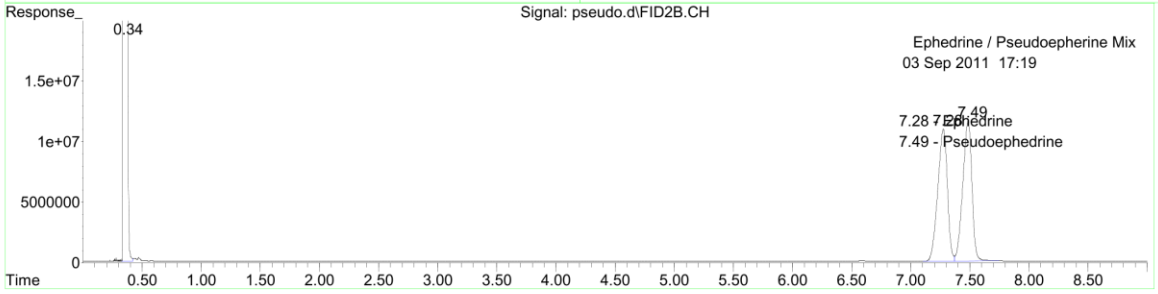
QUALITY CONTROL

Liner count:	170	GC Column:		OSBI MACROS	
Septum count:	170	ZB-50 15m x 530um x 1.0um		Version 3.4	
Gold seal count:	170	Date Column Installed:	06/03/11	Install Date:	
Column count:	882	Split Vent Line Count:	2925	02/03/11	

Quality Assurance Record for Date of Analysis

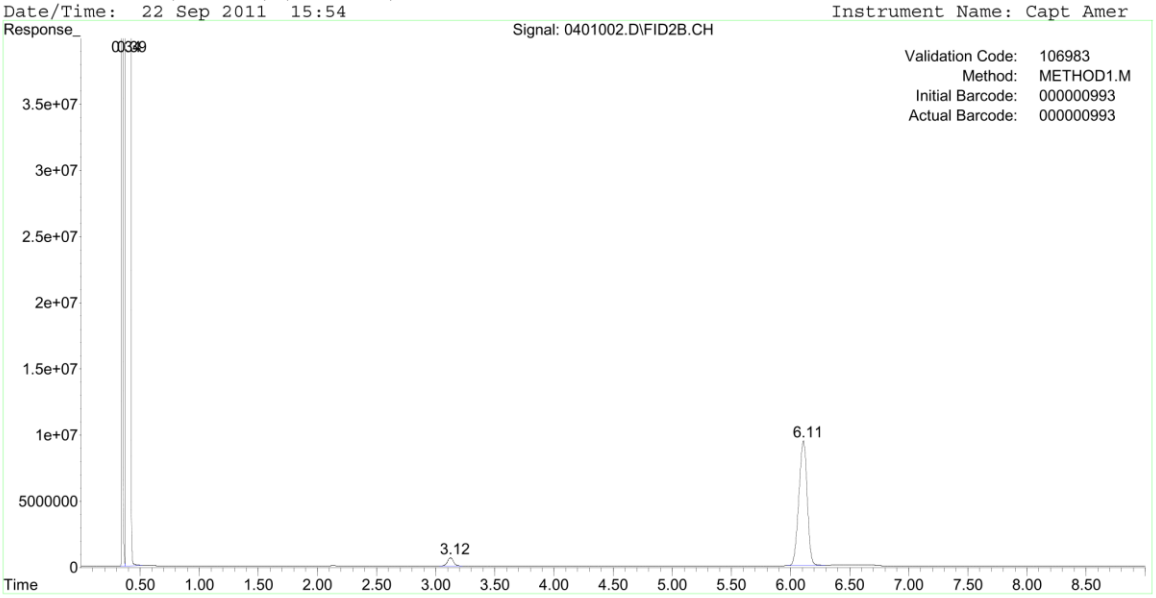






Lab Number: 4-FMC STD EXT KLK
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\2\DATA.SEC\0401002.D
Date/Time: 22 Sep 2011 15:54

Page #:_____
Vial Number: 77
Instrument Name: Capt Amer



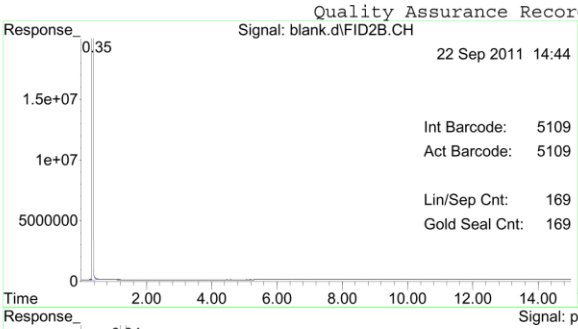
INSTRUMENT CONDITIONS

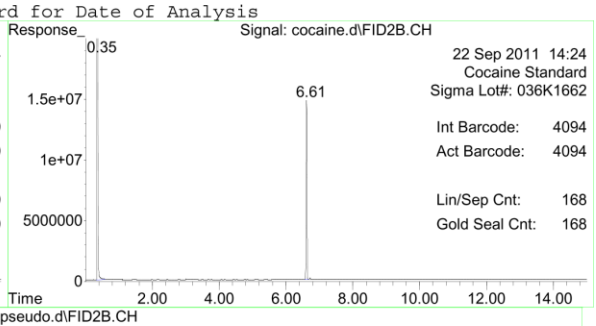
Initial temp:	100 C	Final temp:	140 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	0 min	Total Run Time:	9 min
		Ramp Rate:	5 C/min		

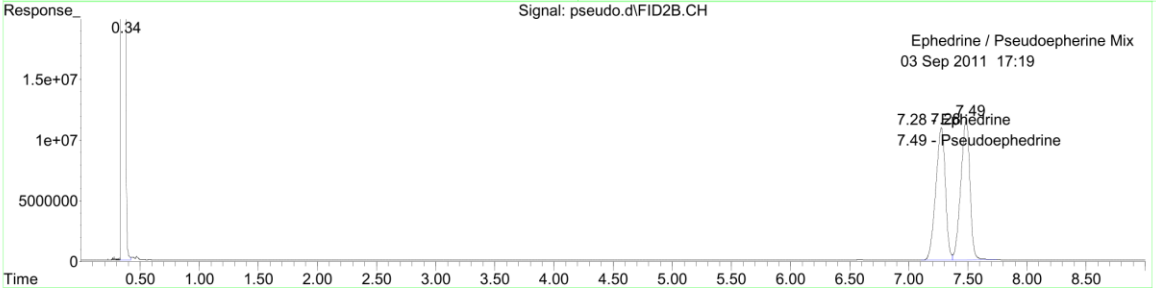
QUALITY CONTROL

Liner count:	171	GC Column:		OSBI MACROS	
Septum count:	171	ZB-50 15m x 530um x 1.0um		Version 3.4	
Gold seal count:	171	Date Column Installed:	06/03/11	Install Date:	
Column count:	883	Split Vent Line Count:	2926	02/03/11	

Quality Assurance Record for Date of Analysis

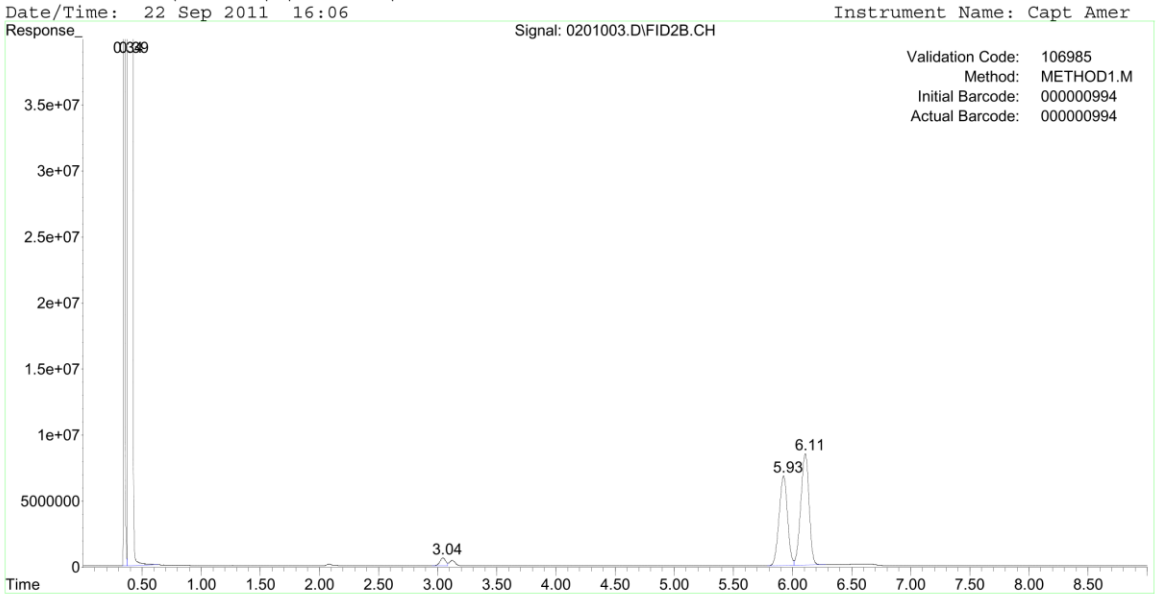






Lab Number: 3 and 4-FMC STD MIX EXT
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\2\DATA.SEC\0201003.D
Date/Time: 22 Sep 2011 16:06

Page #:_____
Vial Number: 78
Instrument Name: Capt Amer



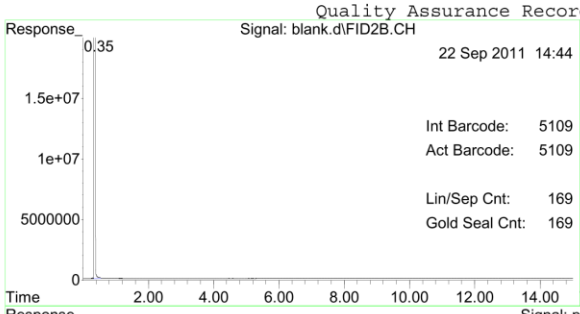
INSTRUMENT CONDITIONS

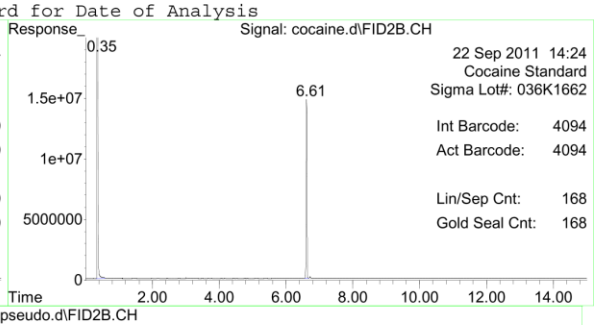
Initial temp:	100 C	Final temp:	140 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	0 min	Total Run Time:	9 min
		Ramp Rate:	5 C/min		

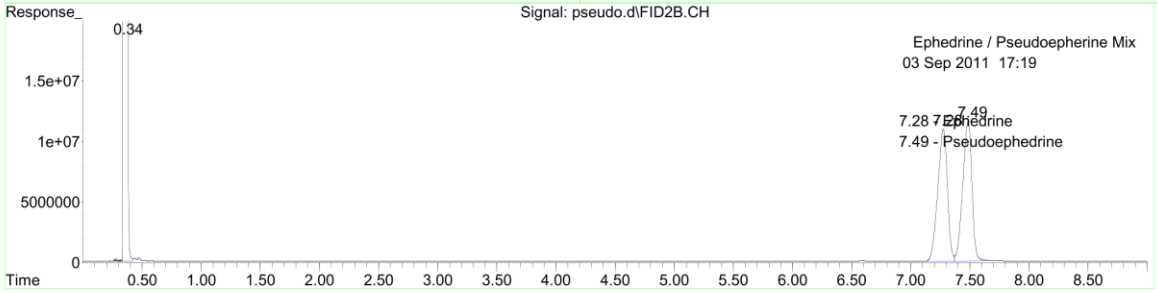
QUALITY CONTROL

Liner count:	172	GC Column:		OSBI MACROS	
Septum count:	172	ZB-50 15m x 530um x 1.0um		Version 3.4	
Gold seal count:	172	Date Column Installed:	06/03/11	Install Date:	
Column count:	884	Split Vent Line Count:	2927	02/03/11	

Quality Assurance Record for Date of Analysis

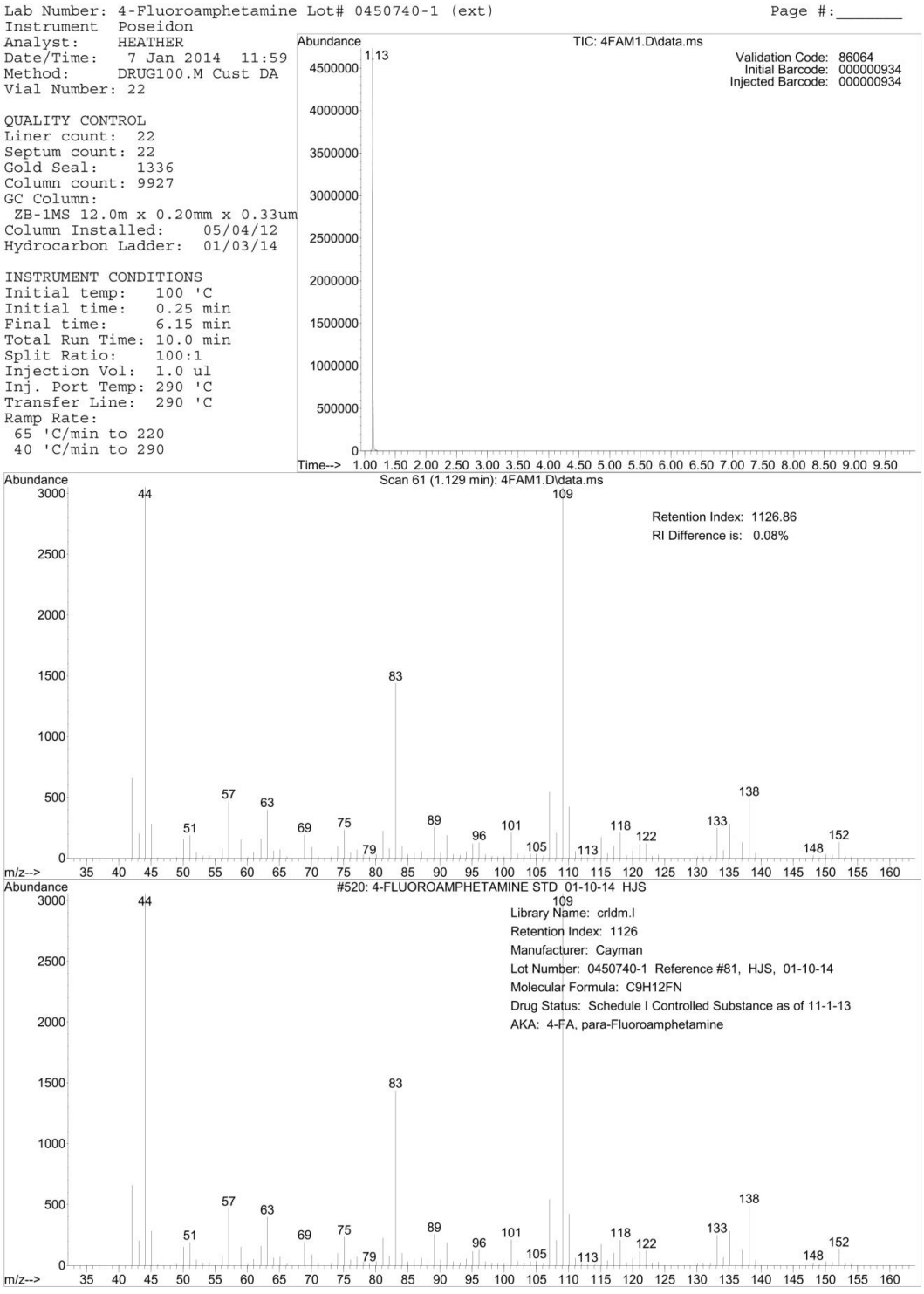




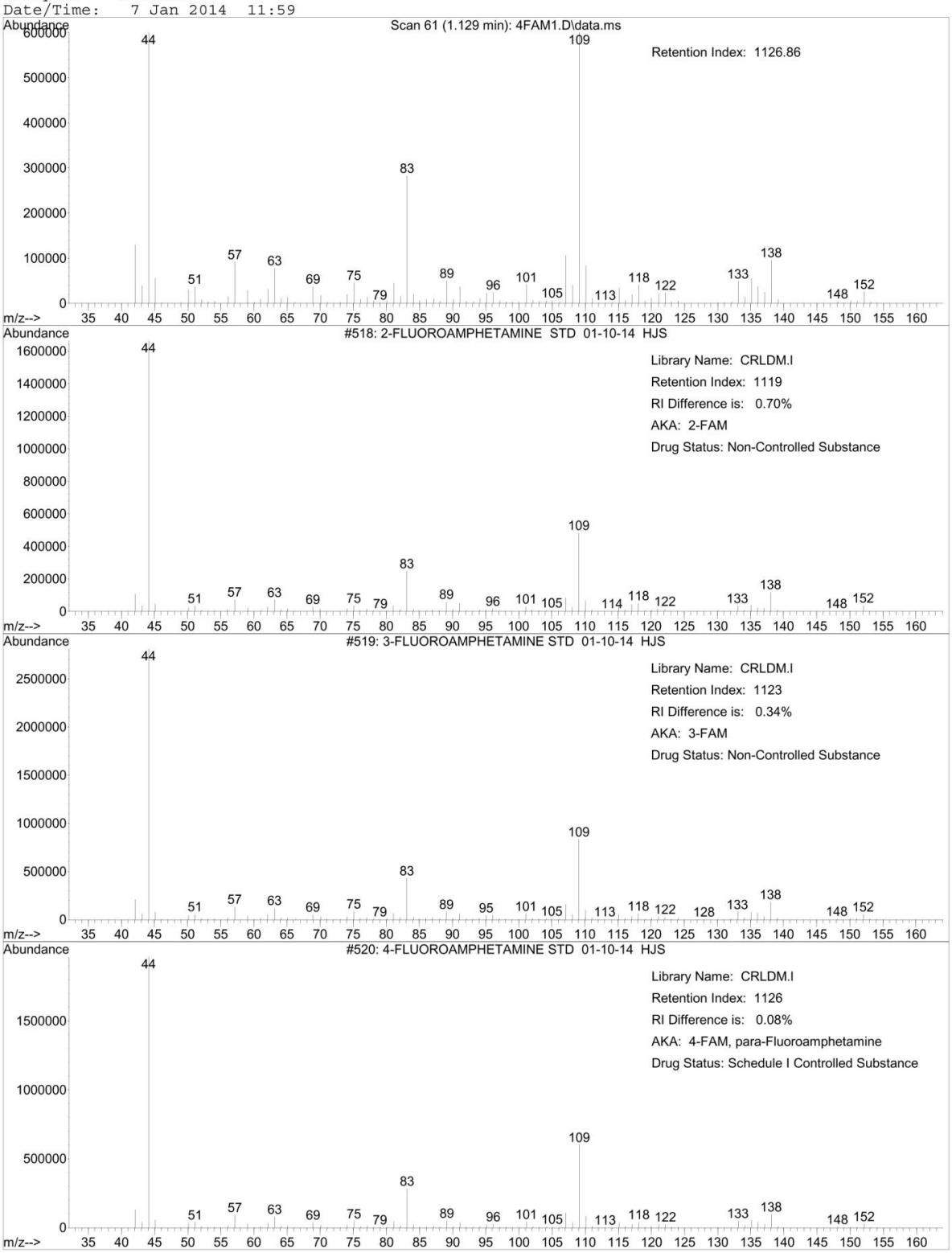


Appendix C: Extraction Data

Appendix C-3: 2, 3 and 4-Fluoroamphetamines Data

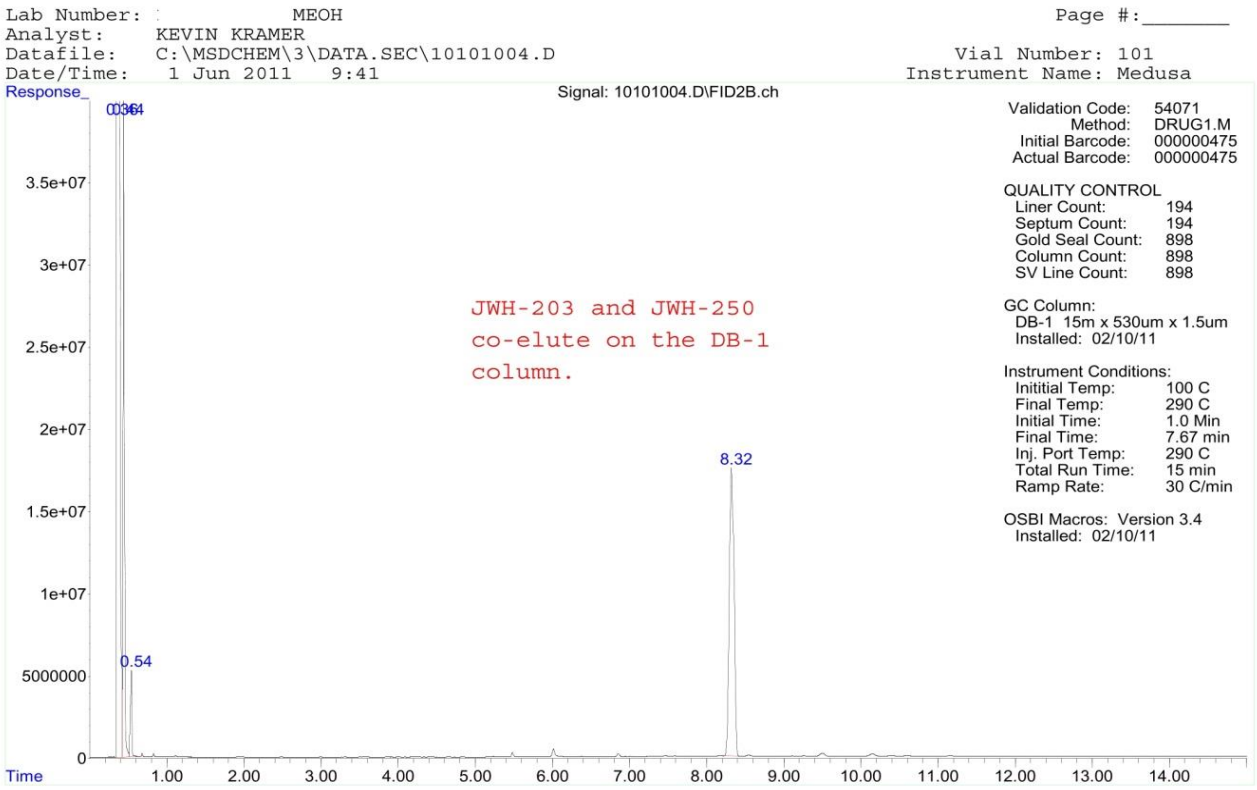


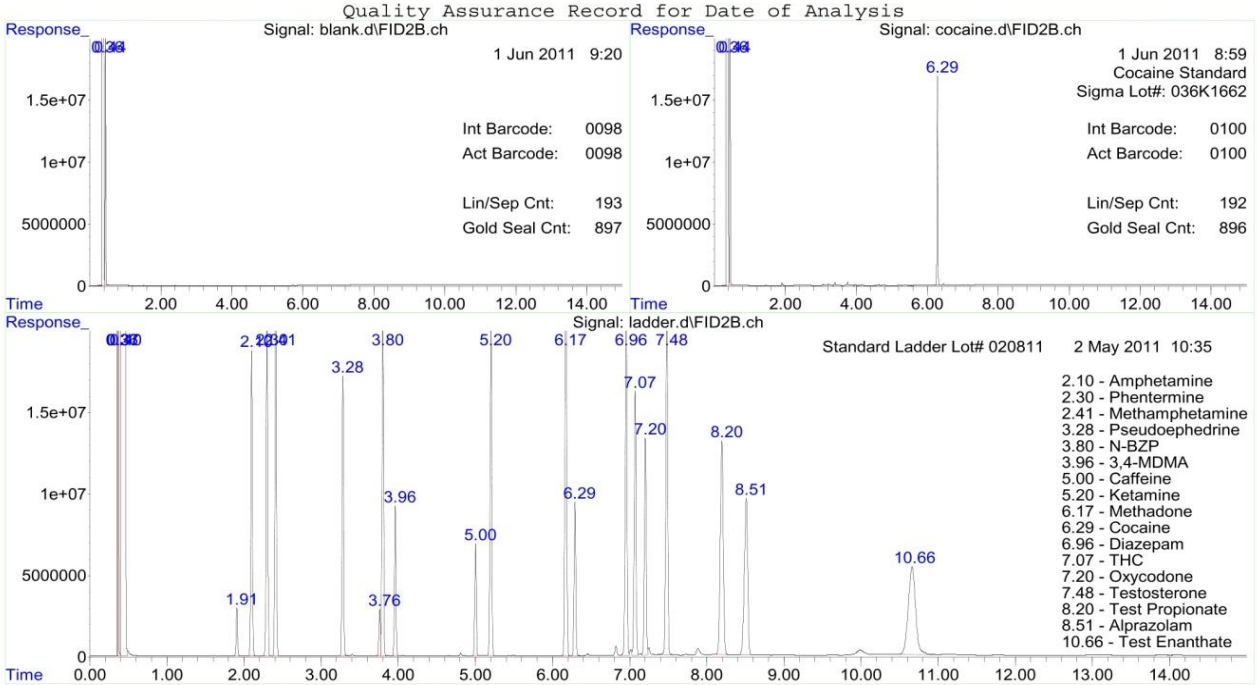
Lab Number: 4-Fluoroamphetamine Lot# 0450740-1 (ext) Page #:_____
Instrument Poseidon
Analyst: HEATHER
Date/Time: 7 Jan 2014 11:59



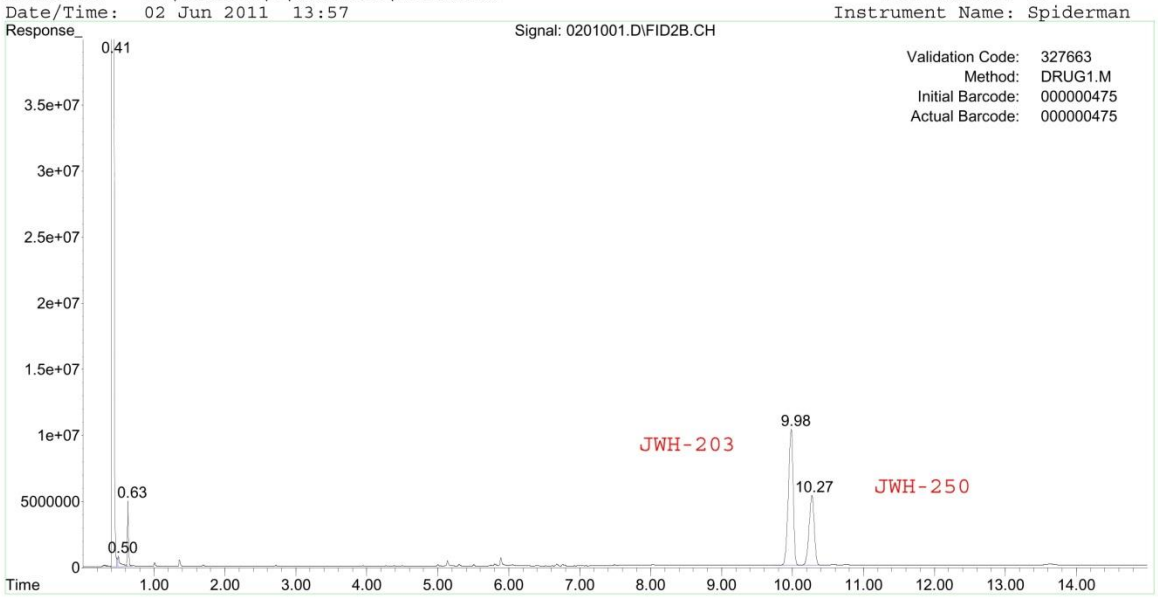
Appendix C: Extraction Data

Appendix C-4: JWH-203 and JWH-250 Data





Lab Number: [redacted] MEOH KLK Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\1\DATA.SEC\0201001.D Vial Number: 25
Date/Time: 02 Jun 2011 13:57 Instrument Name: Spiderman



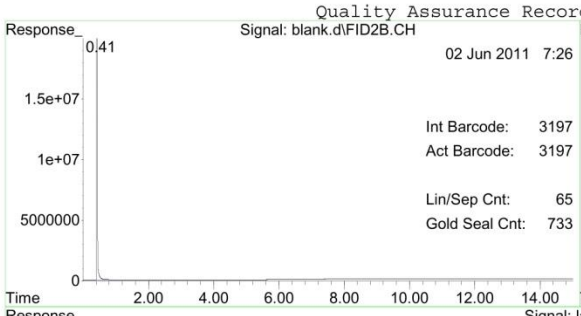
INSTRUMENT CONDITIONS
Initial temp: 100 C
Initial time: 1.0 Min
Final temp: 290 C
Final time: 7.67 min
Ramp Rate: 30 C/min
Inj. Port Temp: 290 C
Total Run Time: 15 min

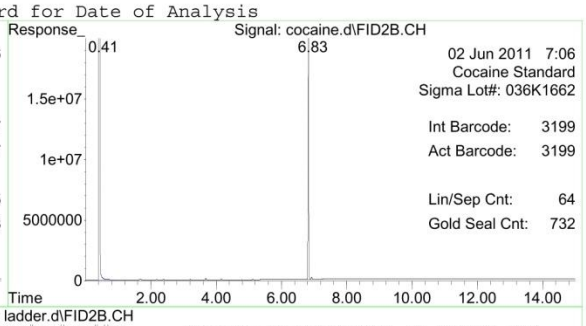
QUALITY CONTROL
Liner count: 68
Septum count: 68
Gold seal count: 736
Column count: 736

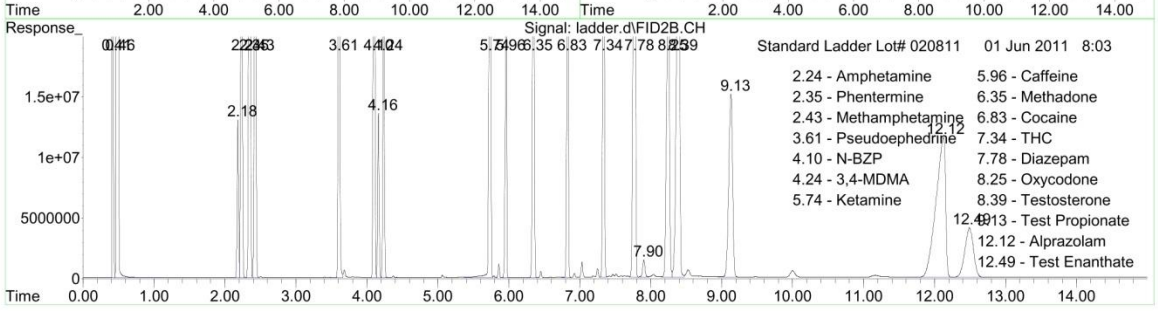
GC Column:
ZB-50 15.0m x 0.53mm x 1.0um
Date Column Installed: 03/23/11
Split Vent Line Count: 4727

OSBI MACROS
Version 3.4
Install Date:
02/04/11

Quality Assurance Record for Date of Analysis





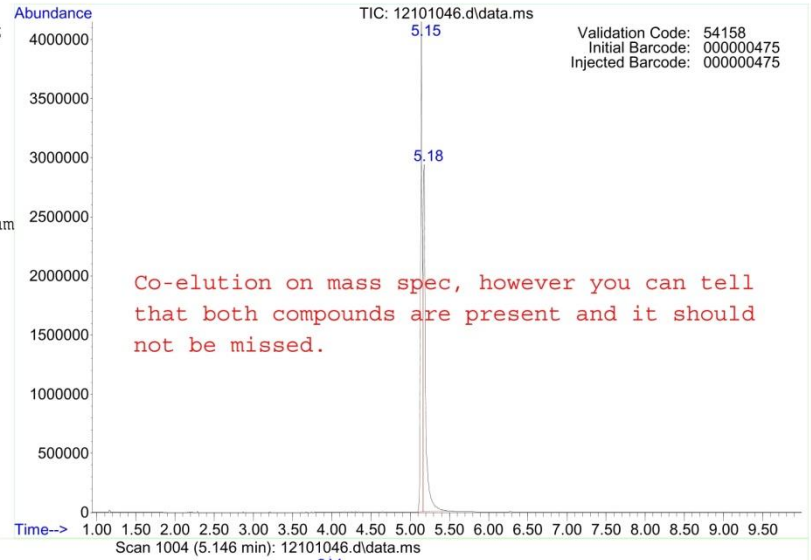


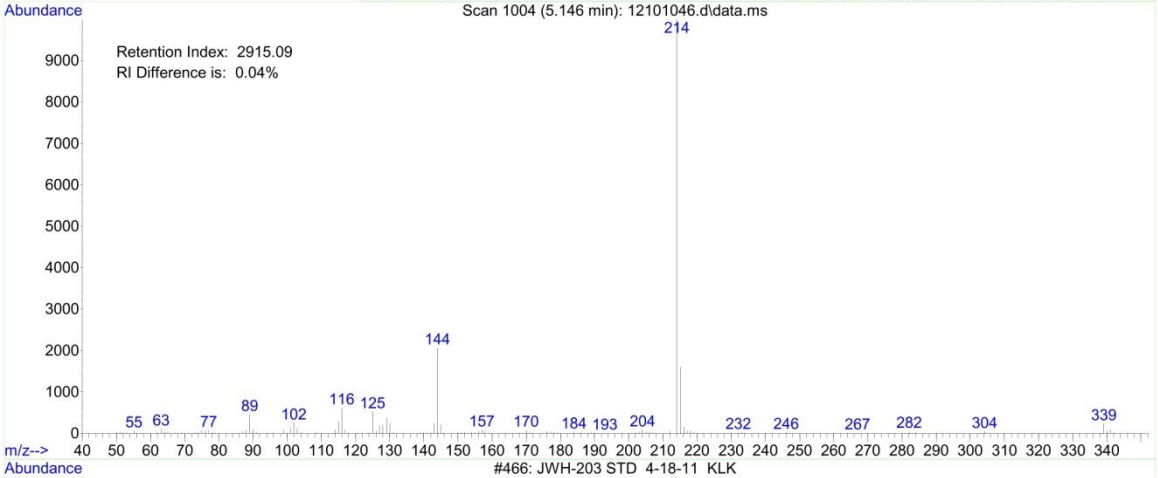
Lab Number: : MEOH Page #:_____

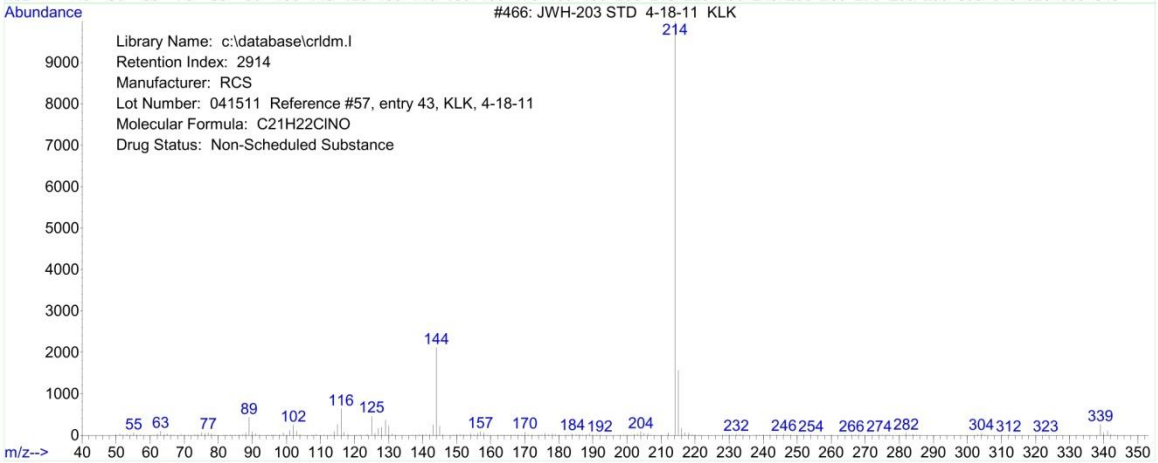
Instrument Poseidon
Analyst: KEVIN KRAMER
Date/Time: 1 Jun 2011 18:46
Method: DRUG100.M
Vial Number: 121

QUALITY CONTROL
Liner count: 172
Septum count: 172
Gold Seal: 841
Column count: 2310
GC Column:
ZB-1MS 12.0m x 0.20mm x 0.33um
Column Installed: 02/18/11
Hydrocarbon Ladder: 05/02/11

INSTRUMENT CONDITIONS
Initial temp: 100 'C
Initial time: 0.25 min
Final time: 6.15 min
Total Run Time: 10.0 min
Split Ratio: 100:1
Injection Vol: 1.0 ul
Inj. Port Temp: 290 'C
Transfer Line: 290 'C
Ramp Rate:
65 'C/min to 220
40 'C/min to 290

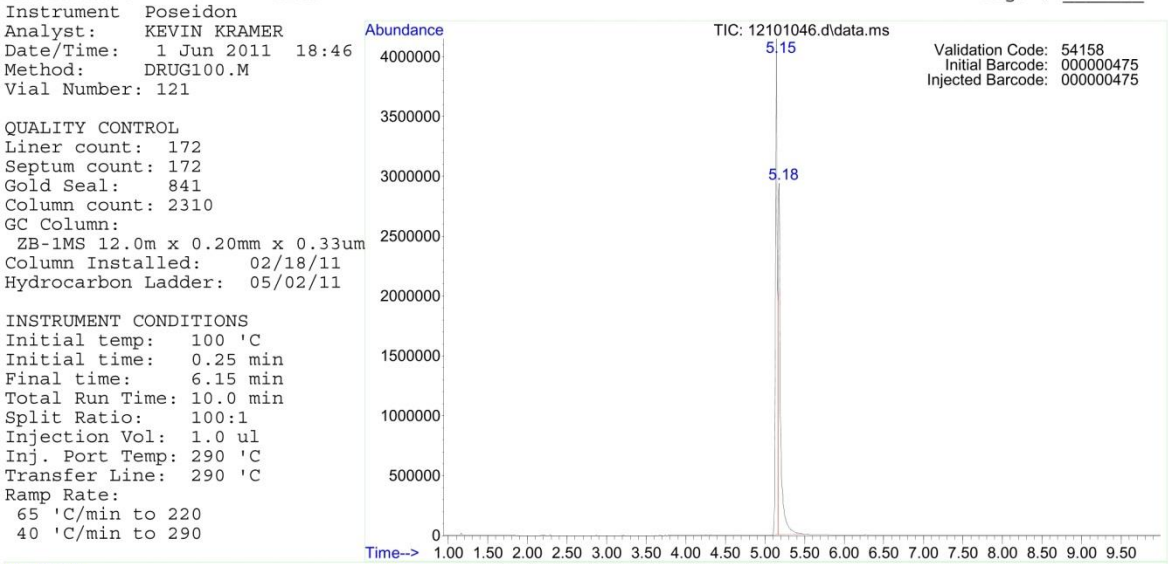


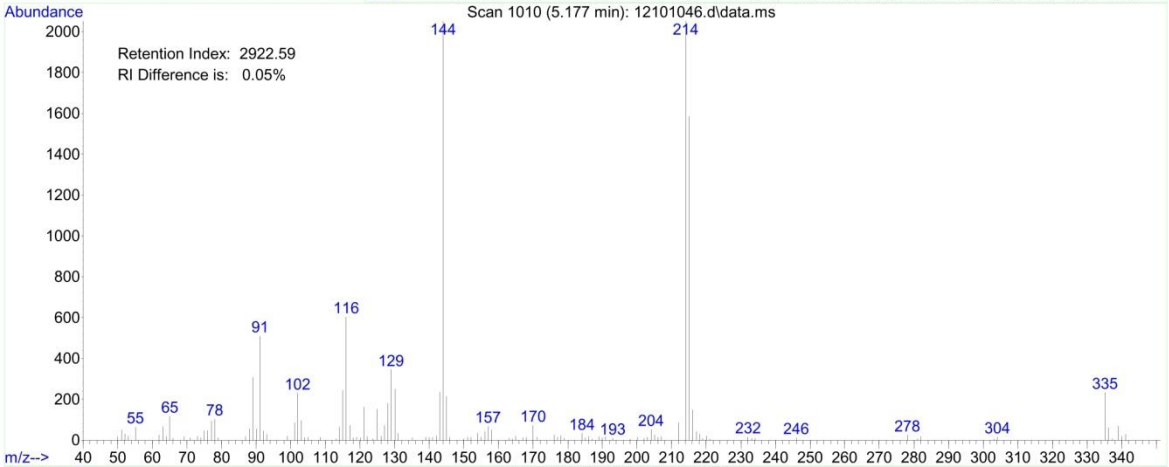


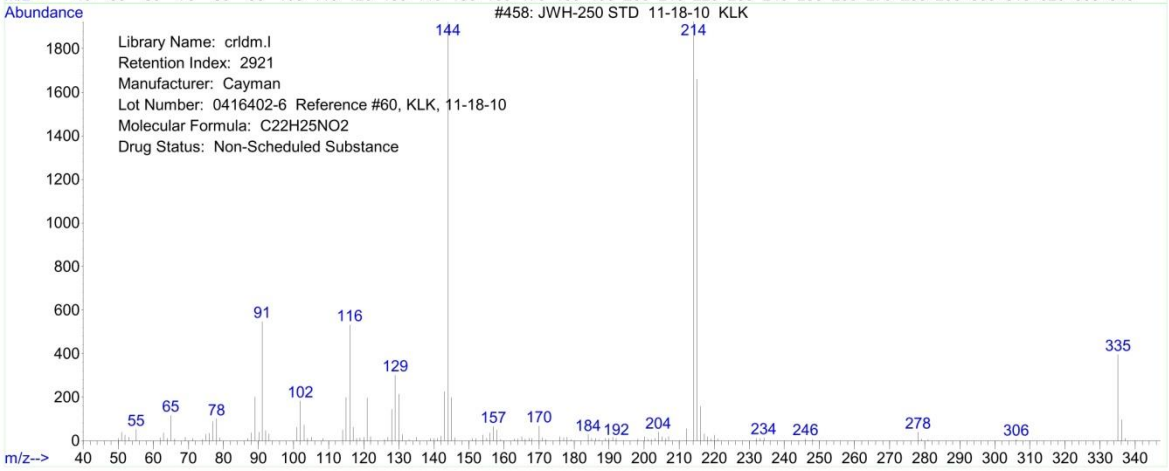


Page #:_____

```
Lab Number:            MEOH
Instrument  Poseidon
Analyst:    KEVIN KRAMER
Date/Time:   1 Jun 2011  18:46
Method:     DRUG100.M
Vial Number: 121

QUALITY CONTROL
Liner count:  172
Septum count: 172
Gold Seal:    841
Column count: 2310
GC Column:
 ZB-1MS 12.0m x 0.20mm x 0.33um
Column Installed:    02/18/11
Hydrocarbon Ladder:  05/02/11

INSTRUMENT CONDITIONS
Initial temp:   100 'C
Initial time:   0.25 min
Final time:     6.15 min
Total Run Time: 10.0 min
Split Ratio:    100:1
Injection Vol:  1.0 ul
Inj. Port Temp: 290 'C
Transfer Line:  290 'C
Ramp Rate:
 65 'C/min to 220
 40 'C/min to 290
```







Appendix C: Extraction Data

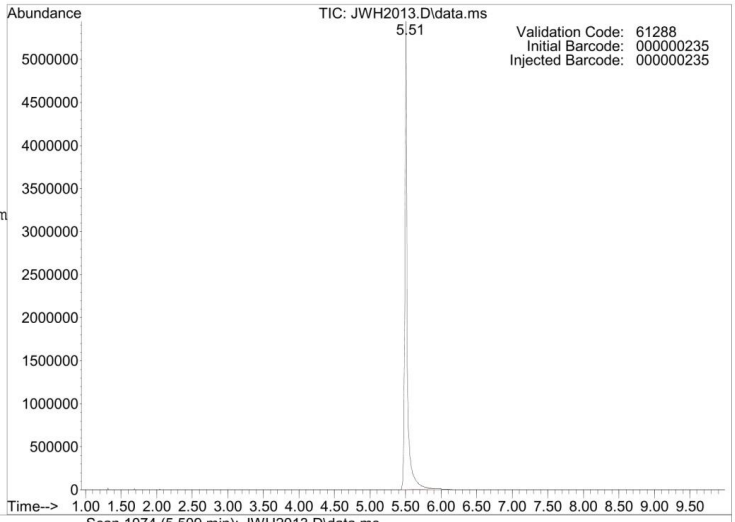
Appendix C-5: JWH-201, JWH-250 and JWH-302 Data

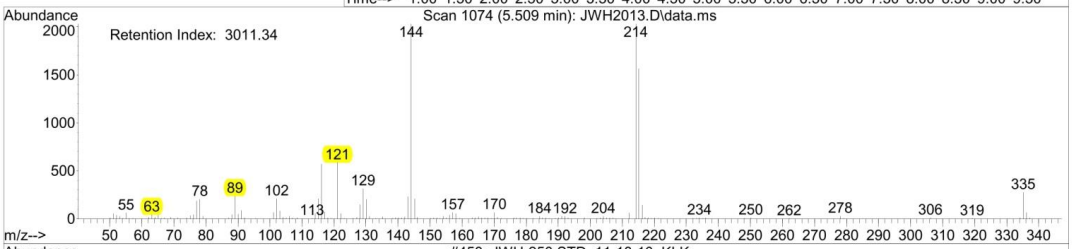
Lab Number: JWH-201 LOT# 0431965-6 Page #:_____

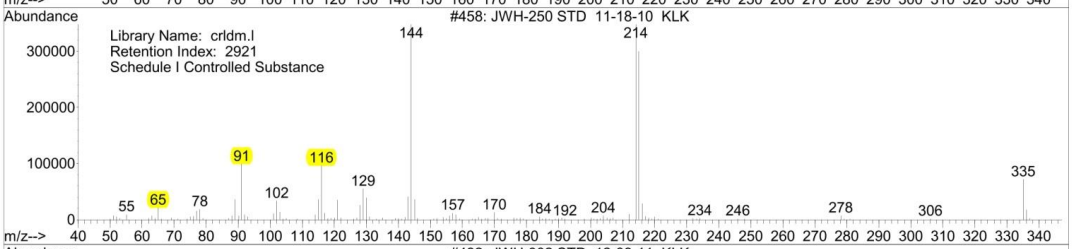
Instrument Poseidon
Analyst: KEVIN KRAMER
Date/Time: 8 Dec 2011 10:52
Method: DRUG50.M Cust DA
Vial Number: 135

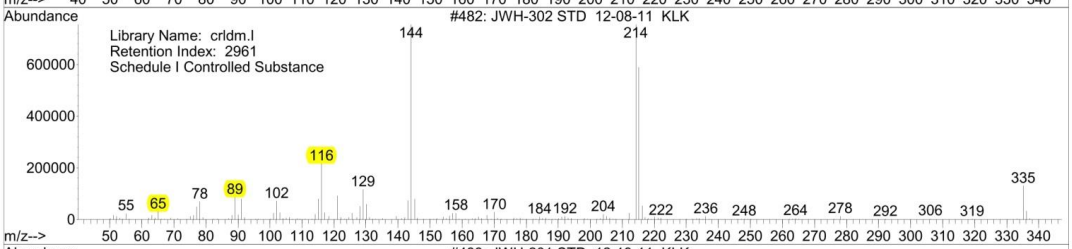
QUALITY CONTROL
Liner count: 74
Septum count: 74
Gold Seal: 534
Column count: 6614
GC Column:
ZB-1MS 12.0m x 0.20mm x 0.33um
Column Installed: 02/18/11
Hydrocarbon Ladder: 12/01/11

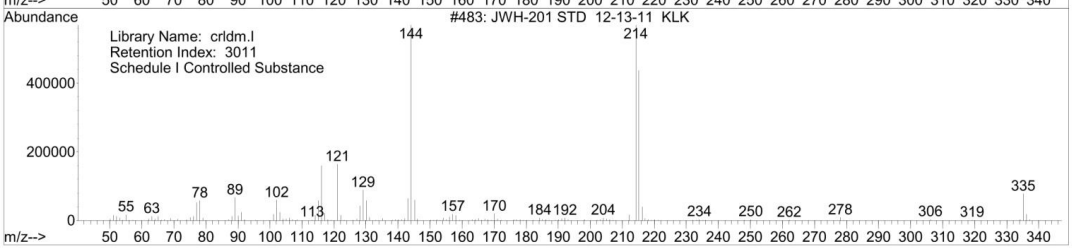
INSTRUMENT CONDITIONS
Initial temp: 100 'C
Initial time: 0.25 min
Final time: 6.15 min
Total Run Time: 10.0 min
Split Ratio: 50:1
Injection Vol: 1.0 ul
Inj. Port Temp: 290 'C
Transfer Line: 290 'C
Ramp Rate:
65 'C/min to 220
40 'C/min to 290

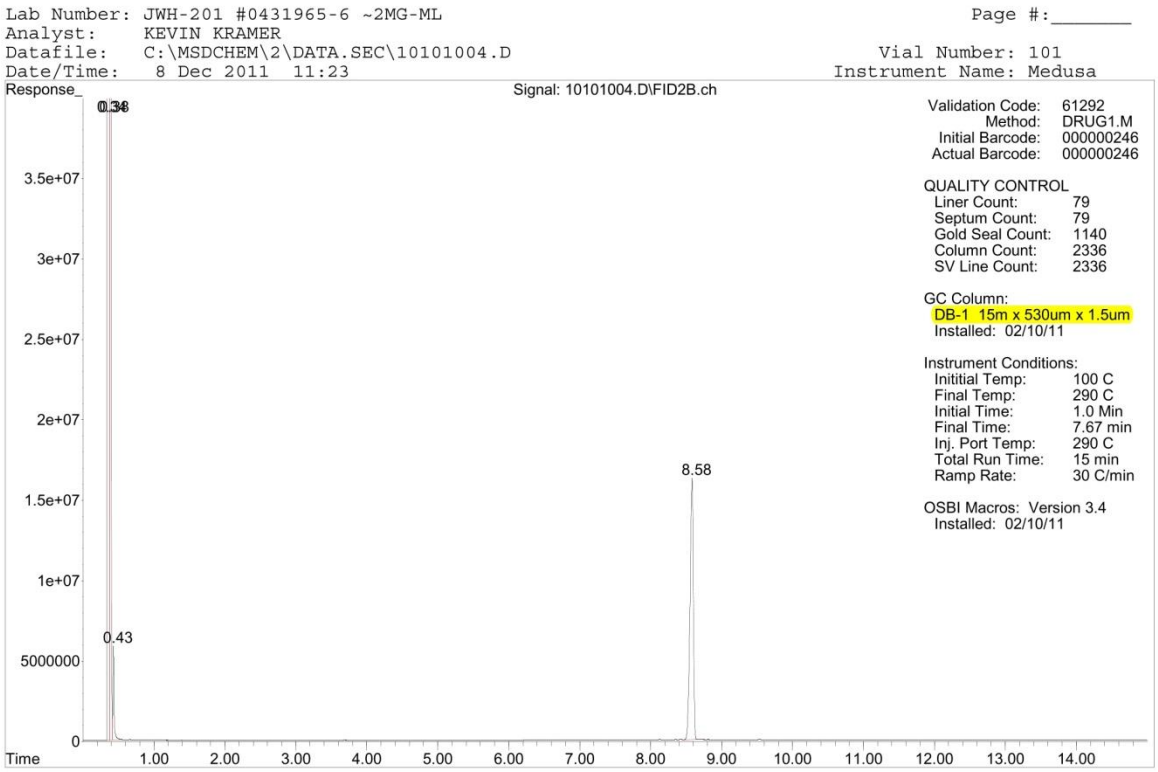


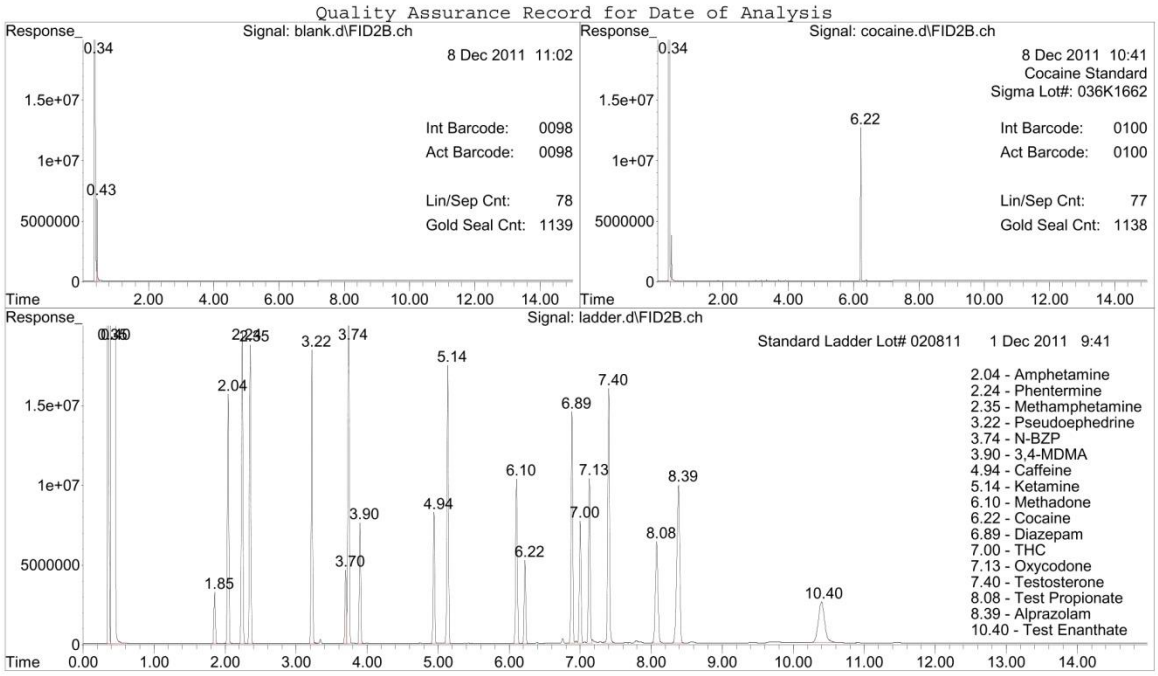


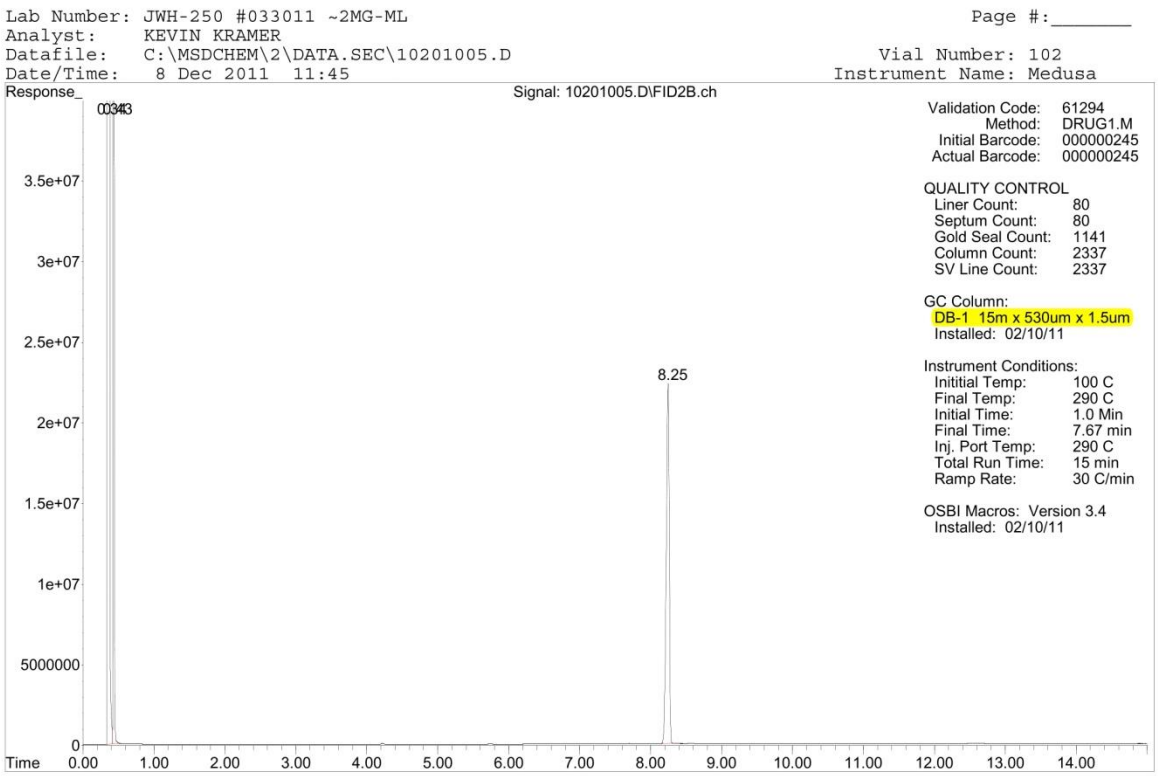




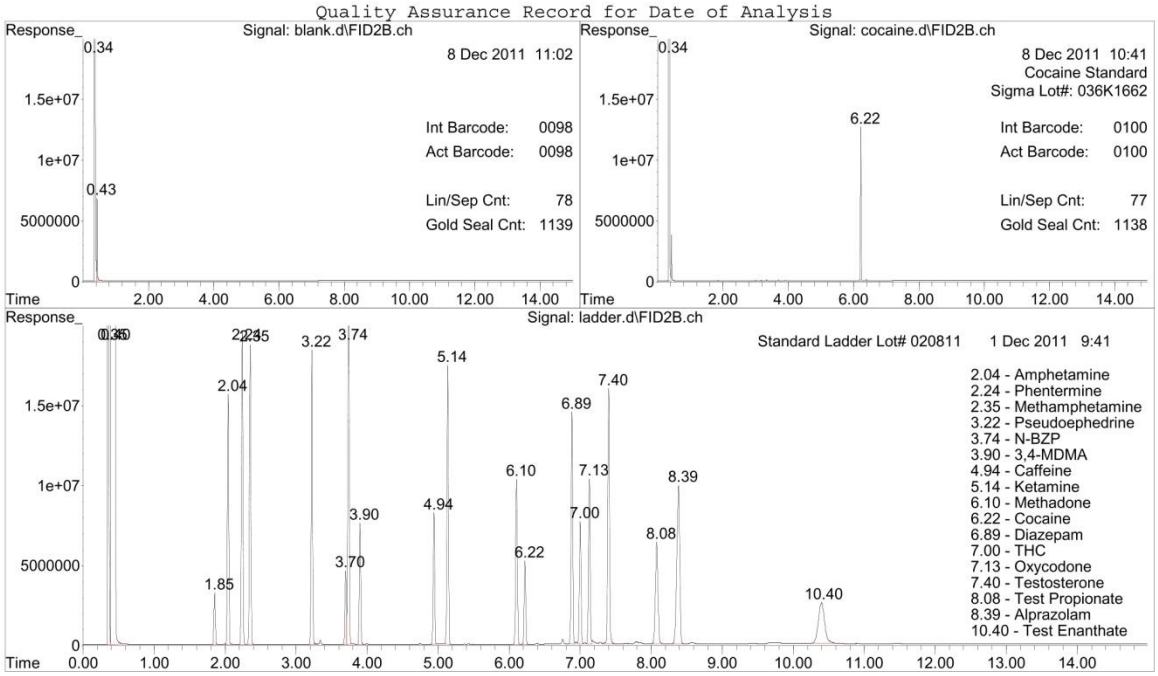


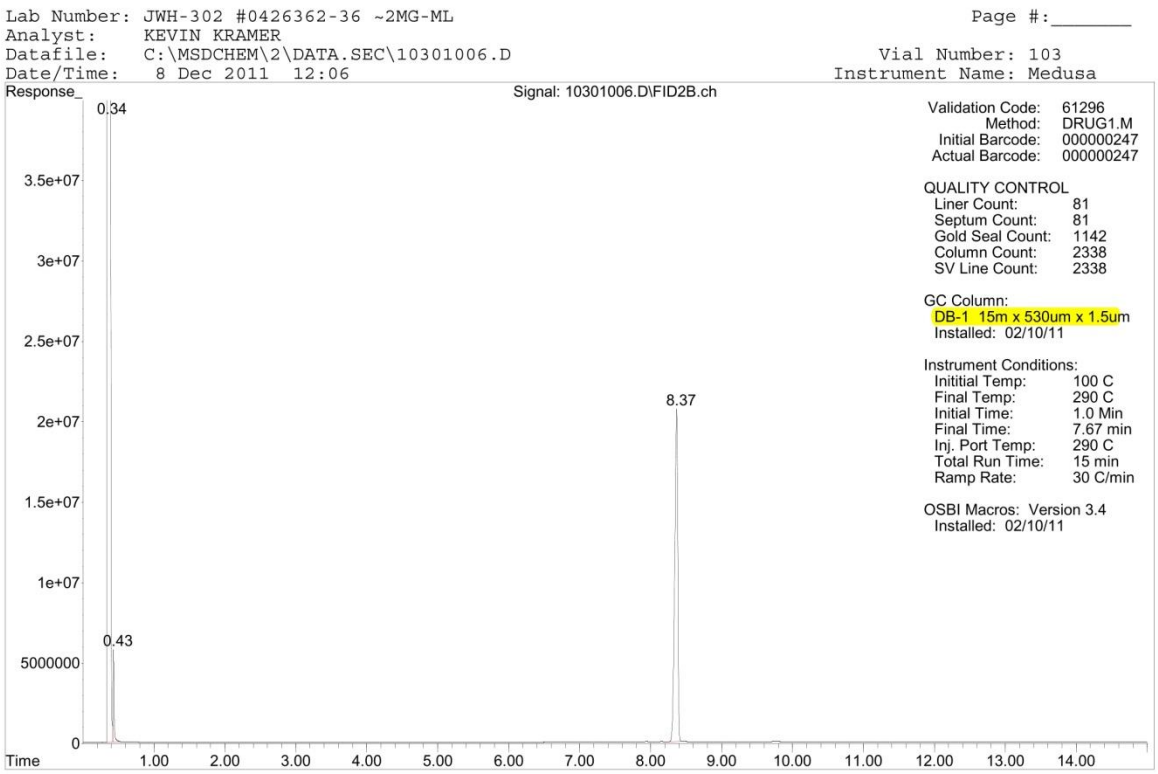

Lab Number: JWH-201 #0431965-6 ~2MG-ML
Page #:_____
Analyst: KEVIN KRAMER
Datafile: C:\MSDCHEM\2\DATA.SEC\10101004.D
Vial Number: 101
Date/Time: 8 Dec 2011 11:23
Instrument Name: Medusa
Response_
Signal: 10101004.D\FID2B.ch
Validation Code: 61292
Method: DRUG1.M
Initial Barcode: 000000246
Actual Barcode: 000000246
QUALITY CONTROL
Liner Count: 79
Septum Count: 79
Gold Seal Count: 1140
Column Count: 2336
SV Line Count: 2336
GC Column:
DB-1 15m x 530um x 1.5um
Installed: 02/10/11
Instrument Conditions:
Inititial Temp: 100 C
Final Temp: 290 C
Initial Time: 1.0 Min
Final Time: 7.67 min
Inj. Port Temp: 290 C
Total Run Time: 15 min
Ramp Rate: 30 C/min
OSBI Macros: Version 3.4
Installed: 02/10/11
0.34
0.43
8.58
Time


Quality Assurance Record for Date of Analysis
Response_
Signal: blank.d\FID2B.ch
8 Dec 2011 11:02
Int Barcode: 0098
Act Barcode: 0098
Lin/Sep Cnt: 78
Gold Seal Cnt: 1139
0.34
0.43
Signal: cocaine.d\FID2B.ch
8 Dec 2011 10:41
Cocaine Standard
Sigma Lot#: 036K1662
Int Barcode: 0100
Act Barcode: 0100
Lin/Sep Cnt: 77
Gold Seal Cnt: 1138
6.22
Signal: ladder.d\FID2B.ch
Standard Ladder Lot# 020811 1 Dec 2011 9:41
2.04 - Amphetamine
2.24 - Phentermine
2.35 - Methamphetamine
3.22 - Pseudoephedrine
3.74 - N-BZP
3.90 - 3,4-MDMA
4.94 - Caffeine
5.14 - Ketamine
6.10 - Methadone
6.22 - Cocaine
6.89 - Diazepam
7.00 - THC
7.13 - Oxycodone
7.40 - Testosterone
8.08 - Test Propionate
8.39 - Alprazolam
10.40 - Test Enanthate
Time

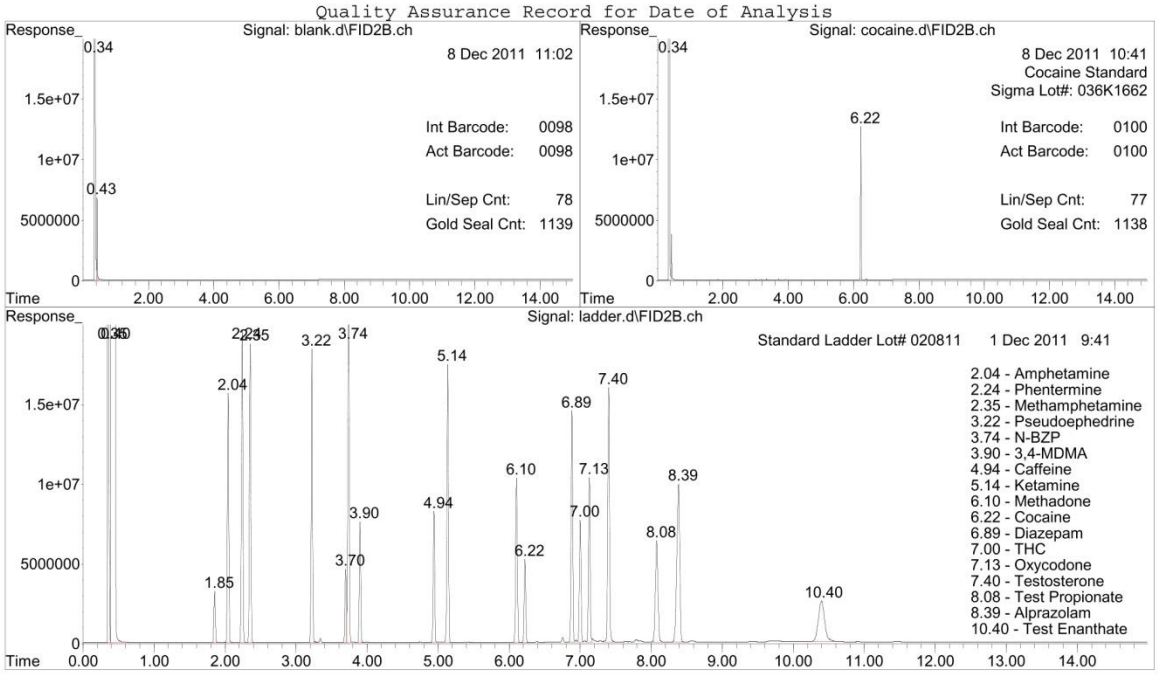


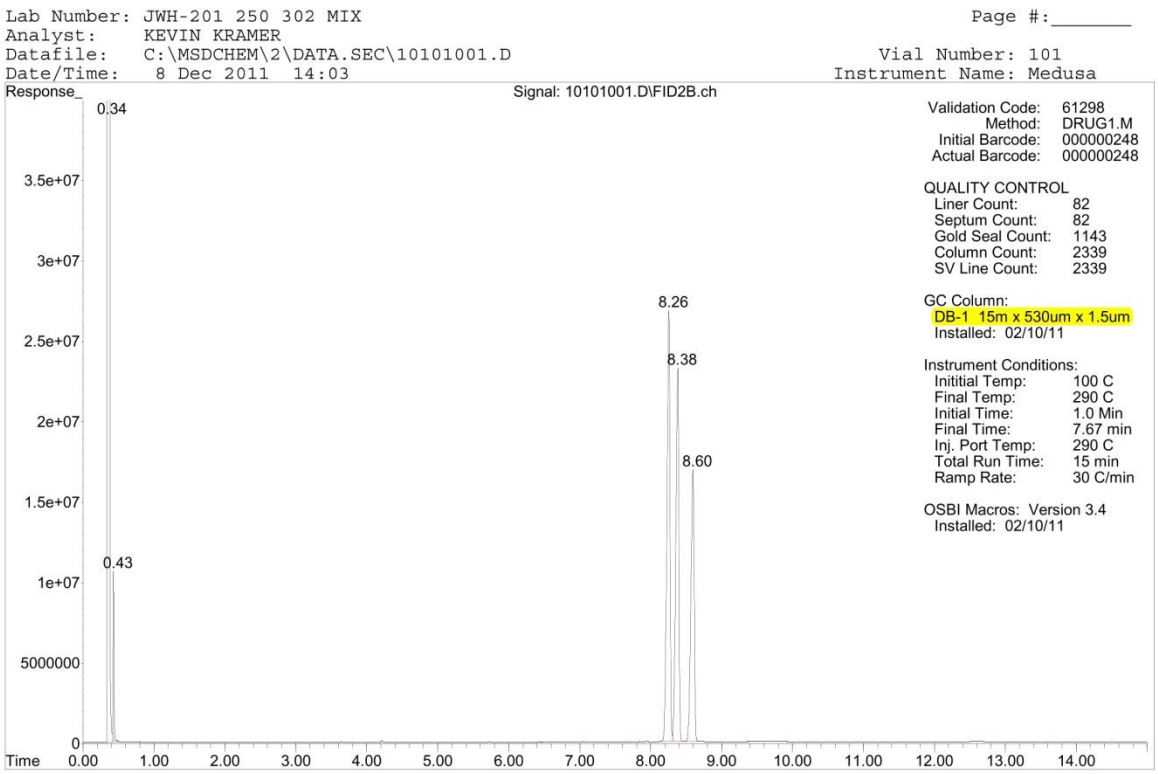
Quality Assurance Record for Date of Analysis

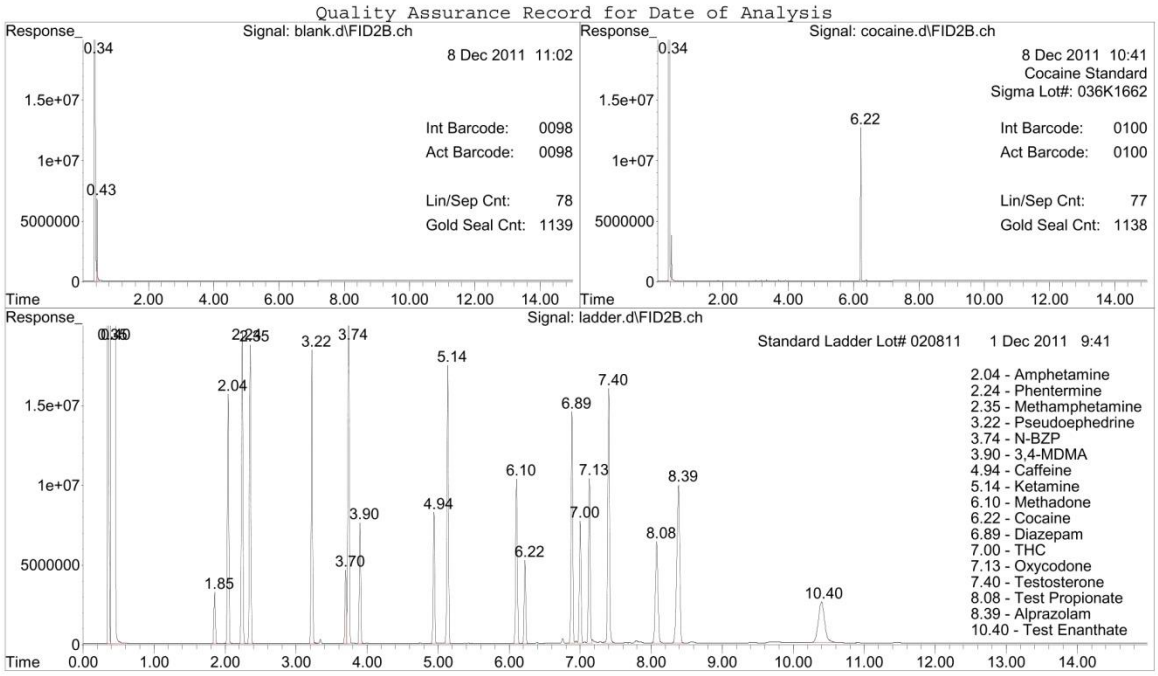




Quality Assurance Record for Date of Analysis

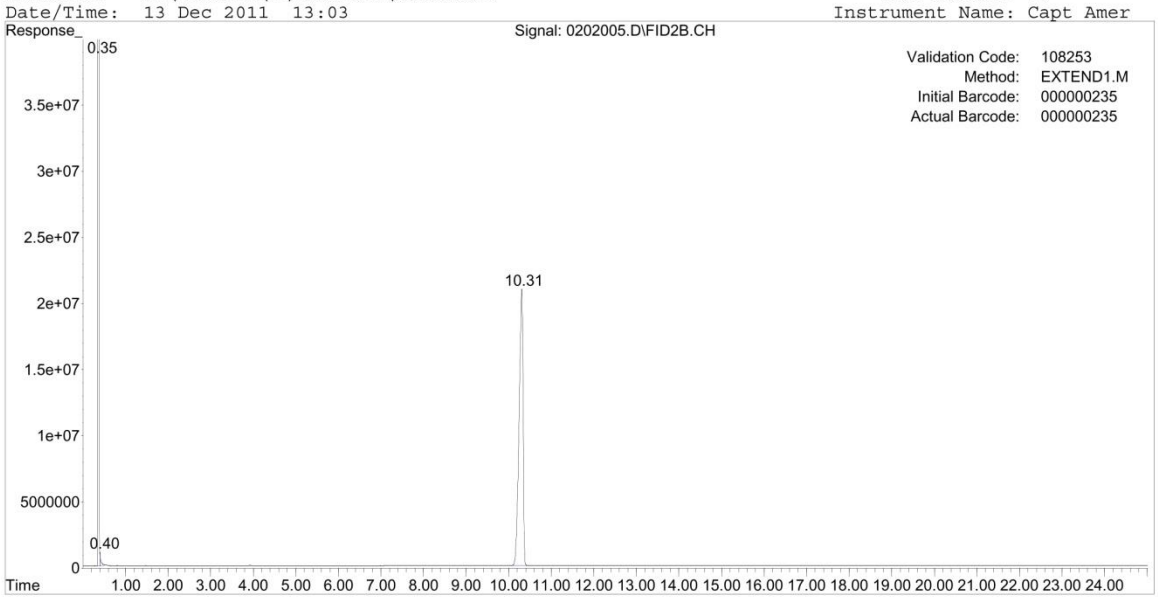



Lab Number: JWH-201 250 302 MIX
Page #:_____
Analyst: KEVIN KRAMER
Datafile: C:\MSDCHEM\2\DATA.SEC\10101001.D
Vial Number: 101
Date/Time: 8 Dec 2011 14:03
Instrument Name: Medusa
Response_
Signal: 10101001.D\FID2B.ch
Validation Code: 61298
Method: DRUG1.M
Initial Barcode: 000000248
Actual Barcode: 000000248
QUALITY CONTROL
Liner Count: 82
Septum Count: 82
Gold Seal Count: 1143
Column Count: 2339
SV Line Count: 2339
GC Column:
DB-1 15m x 530um x 1.5um
Installed: 02/10/11
Instrument Conditions:
Inititial Temp: 100 C
Final Temp: 290 C
Initial Time: 1.0 Min
Final Time: 7.67 min
Inj. Port Temp: 290 C
Total Run Time: 15 min
Ramp Rate: 30 C/min
OSBI Macros: Version 3.4
Installed: 02/10/11
0.34
0.43
8.26
8.38
8.60
Time


Quality Assurance Record for Date of Analysis
Response_
Signal: blank.d\FID2B.ch
8 Dec 2011 11:02
Int Barcode: 0098
Act Barcode: 0098
Lin/Sep Cnt: 78
Gold Seal Cnt: 1139
Signal: cocaine.d\FID2B.ch
8 Dec 2011 10:41
Cocaine Standard
Sigma Lot#: 036K1662
Int Barcode: 0100
Act Barcode: 0100
Lin/Sep Cnt: 77
Gold Seal Cnt: 1138
Signal: ladder.d\FID2B.ch
Standard Ladder Lot# 020811 1 Dec 2011 9:41
2.04 - Amphetamine
2.24 - Phentermine
2.35 - Methamphetamine
3.22 - Pseudoephedrine
3.74 - N-BZP
3.90 - 3,4-MDMA
4.94 - Caffeine
5.14 - Ketamine
6.10 - Methadone
6.22 - Cocaine
6.89 - Diazepam
7.00 - THC
7.13 - Oxycodone
7.40 - Testosterone
8.08 - Test Propionate
8.39 - Alprazolam
10.40 - Test Enanthate
Time

Lab Number: JWH-201 #0431965-6 ~2MG-ML
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\2\DATA.SEC\0202005.D
Date/Time: 13 Dec 2011 13:03

Page #:_____
Vial Number: 76
Instrument Name: Capt Amer



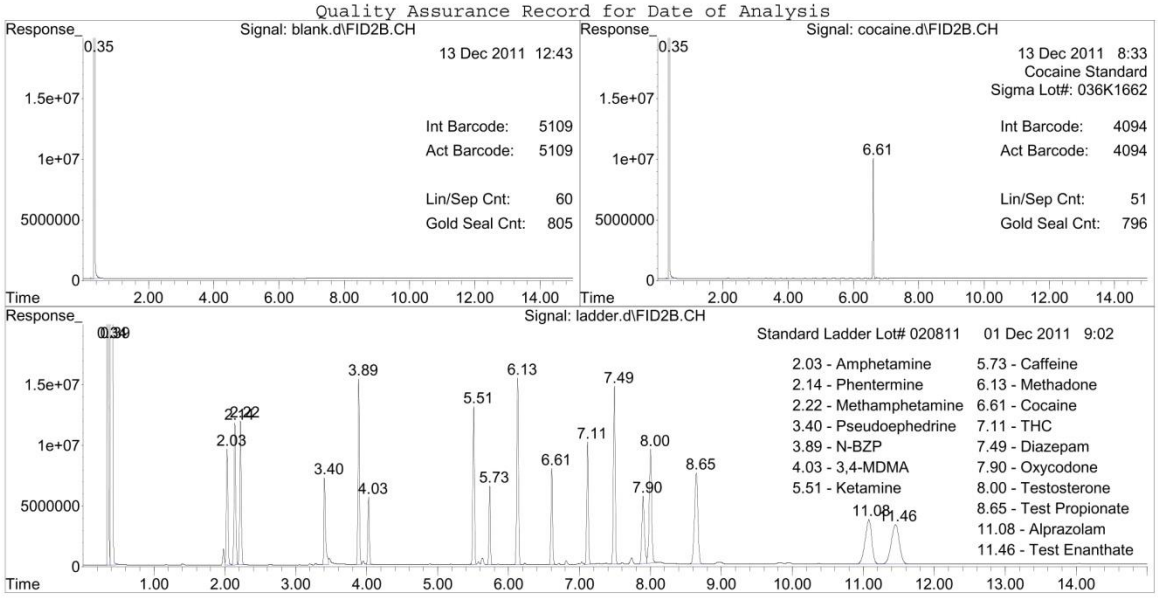
INSTRUMENT CONDITIONS
Initial temp: 100 C
Initial time: 1.0 Min
Final temp: 290 C
Final time: 17.67 min
Ramp Rate: 30 C/min
Inj. Port Temp: 290 C
Total Run Time: 25 min

QUALITY CONTROL
Liner count: 61
Septum count: 61
Gold seal count: 806
Column count: 1518

GC Column:
ZB-50 15m x 530um x 1.0um
Date Column Installed: 06/03/11
Split Vent Line Count: 3561

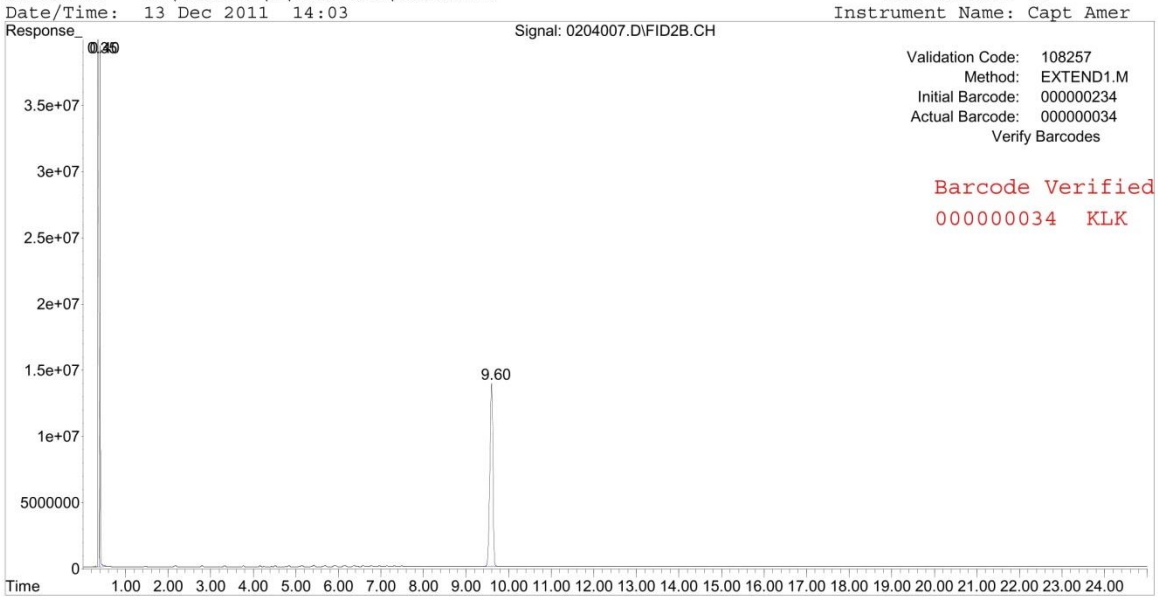
OSBI MACROS
Version 3.4
Install Date:
02/03/11

Quality Assurance Record for Date of Analysis



Lab Number: JWH-250 #033011 ~2MG-ML
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\2\DATA.SEC\0204007.D
Date/Time: 13 Dec 2011 14:03

Page #:_____
Vial Number: 78
Instrument Name: Capt Amer



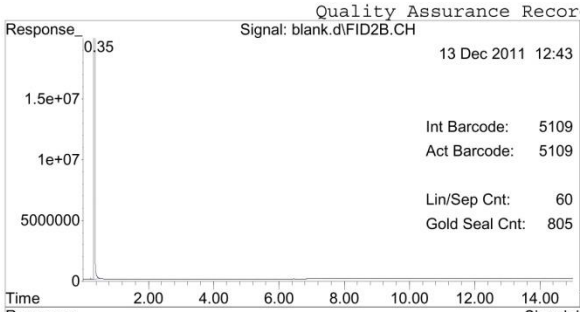
INSTRUMENT CONDITIONS
Initial temp: 100 C
Initial time: 1.0 Min
Final temp: 290 C
Final time: 17.67 min
Ramp Rate: 30 C/min
Inj. Port Temp: 290 C
Total Run Time: 25 min

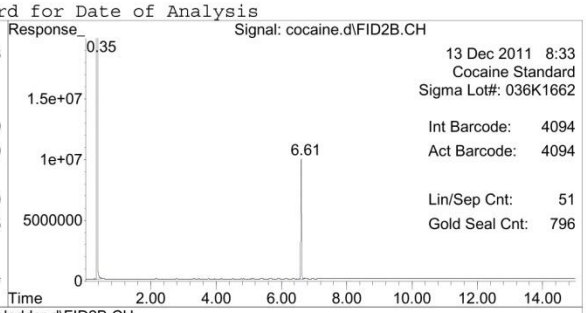
QUALITY CONTROL
Liner count: 63
Septum count: 63
Gold seal count: 808
Column count: 1520

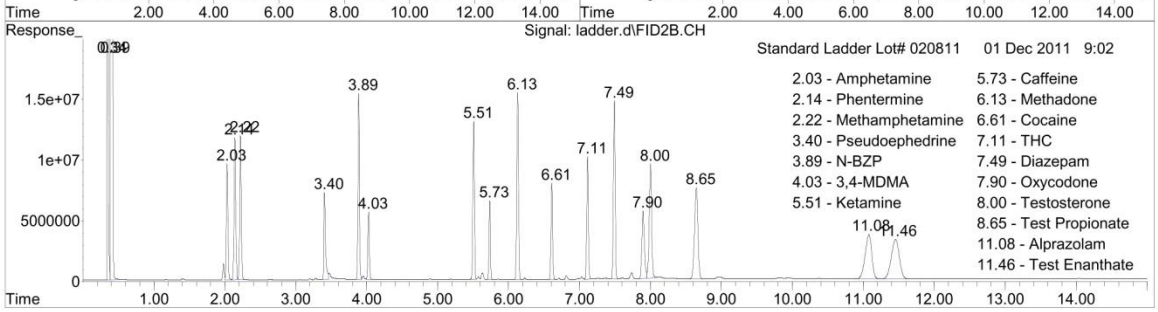
GC Column:
ZB-50 15m x 530um x 1.0um
Date Column Installed: 06/03/11
Split Vent Line Count: 3563

OSBI MACROS
Version 3.4
Install Date:
02/03/11

Quality Assurance Record for Date of Analysis

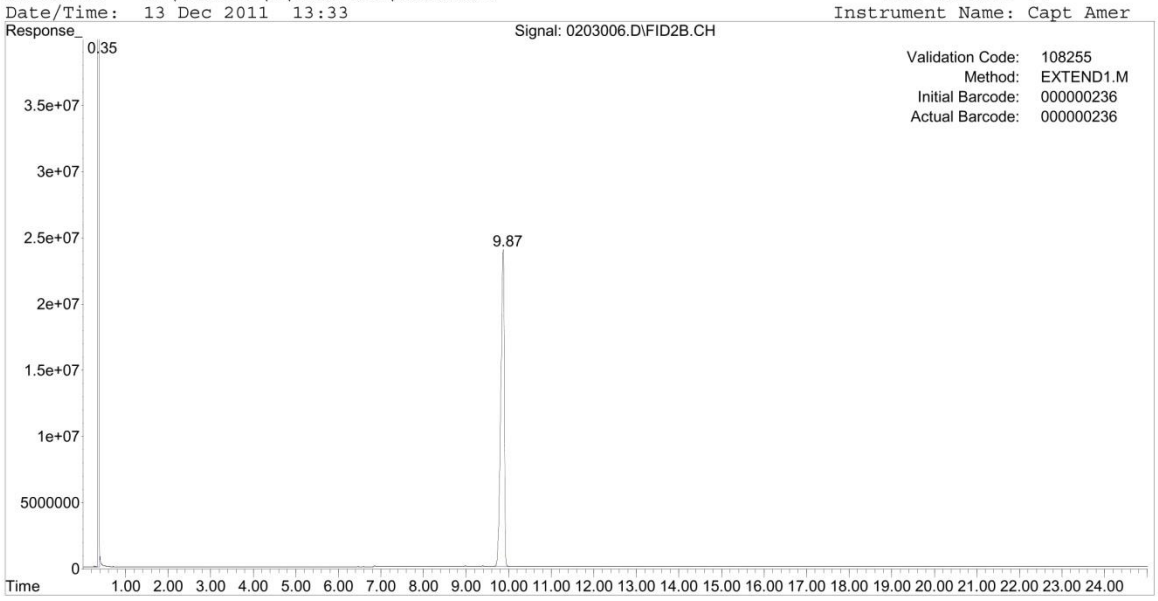






Lab Number: JWH-302 #0426362-36 ~2MG-ML
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\2\DATA.SEC\0203006.D
Date/Time: 13 Dec 2011 13:33

Page #:_____
Vial Number: 77
Instrument Name: Capt Amer



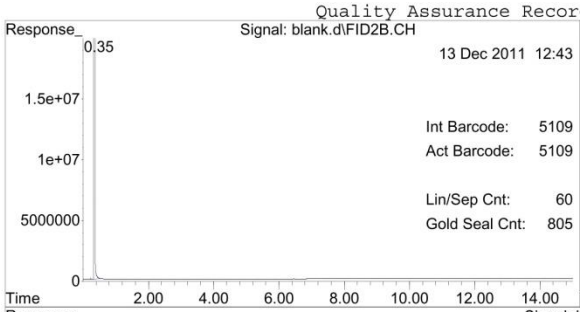
INSTRUMENT CONDITIONS

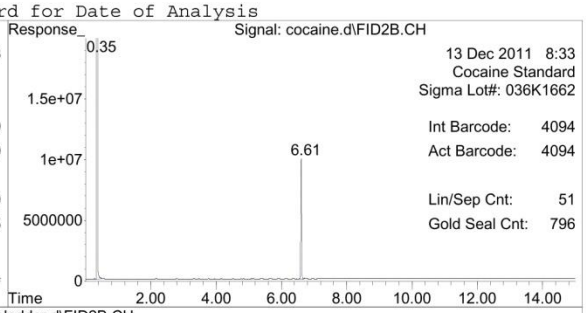
Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	17.67 min	Total Run Time:	25 min
		Ramp Rate:	30 C/min		

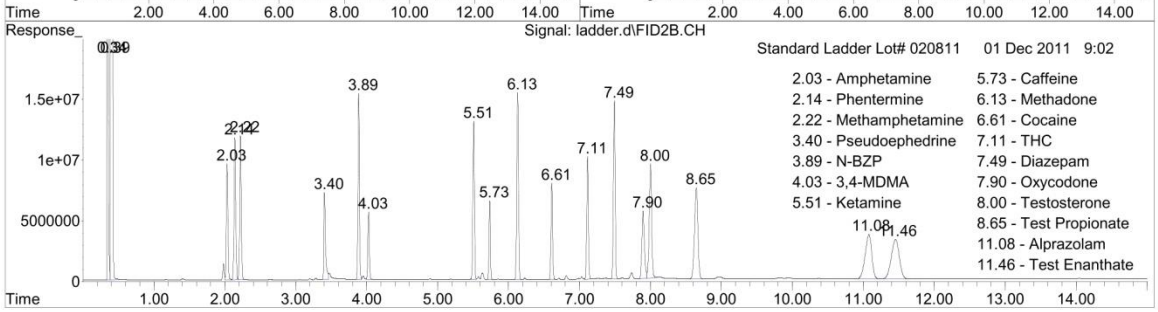
QUALITY CONTROL

Liner count:	62	GC Column:		OSBI MACROS	
Septum count:	62	ZB-50	15m x 530um x 1.0um	Version 3.4	
Gold seal count:	807	Date Column Installed:	06/03/11	Install Date:	
Column count:	1519	Split Vent Line Count:	3562		02/03/11

Quality Assurance Record for Date of Analysis

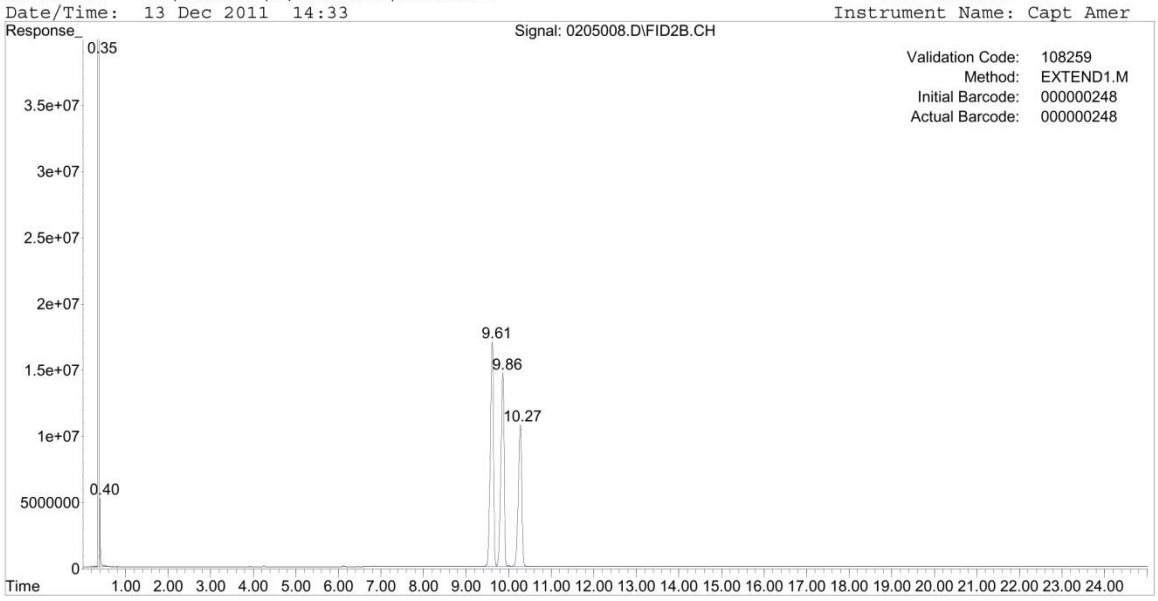






Lab Number: JWH-201 250 302 MIX
Operator: OSBI Drug Laboratory
Datafile: C:\MSDCHEM\2\DATA.SEC\0205008.D
Date/Time: 13 Dec 2011 14:33

Page #:_____
Vial Number: 79
Instrument Name: Capt Amer



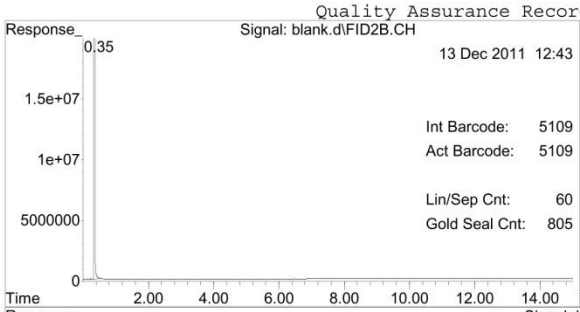
INSTRUMENT CONDITIONS
Initial temp: 100 C
Initial time: 1.0 Min
Final temp: 290 C
Final time: 17.67 min
Ramp Rate: 30 C/min
Inj. Port Temp: 290 C
Total Run Time: 25 min

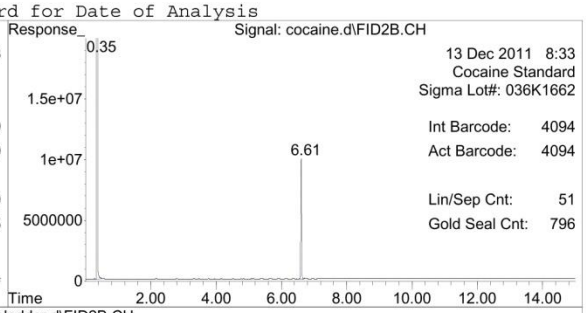
QUALITY CONTROL
Liner count: 64
Septum count: 64
Gold seal count: 809
Column count: 1521

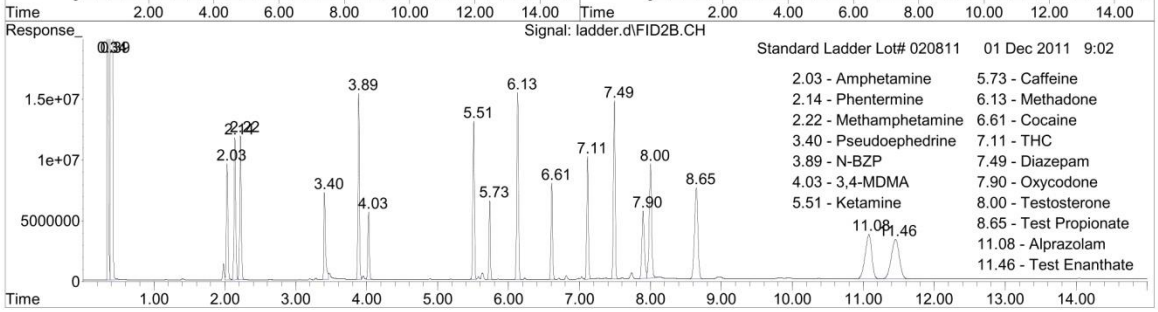
GC Column:
ZB-50 15m x 530um x 1.0um
Date Column Installed: 06/03/11
Split Vent Line Count: 3564

OSBI MACROS
Version 3.4
Install Date:
02/03/11

Quality Assurance Record for Date of Analysis

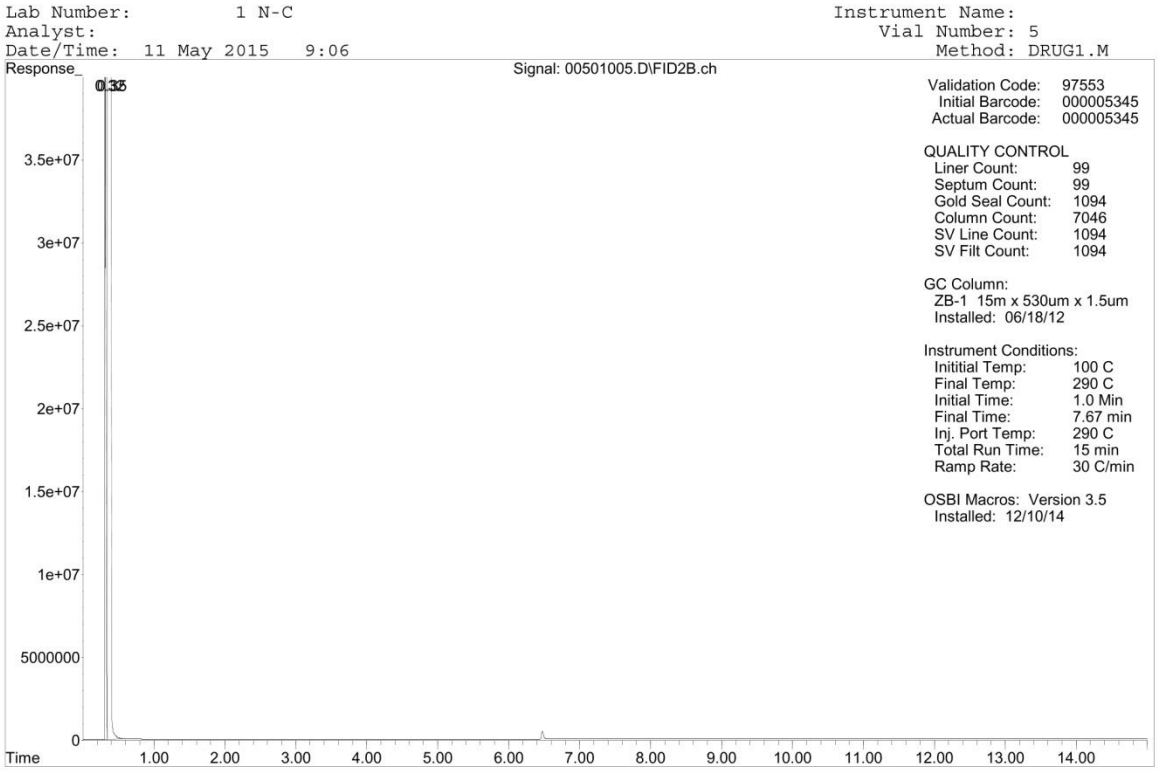


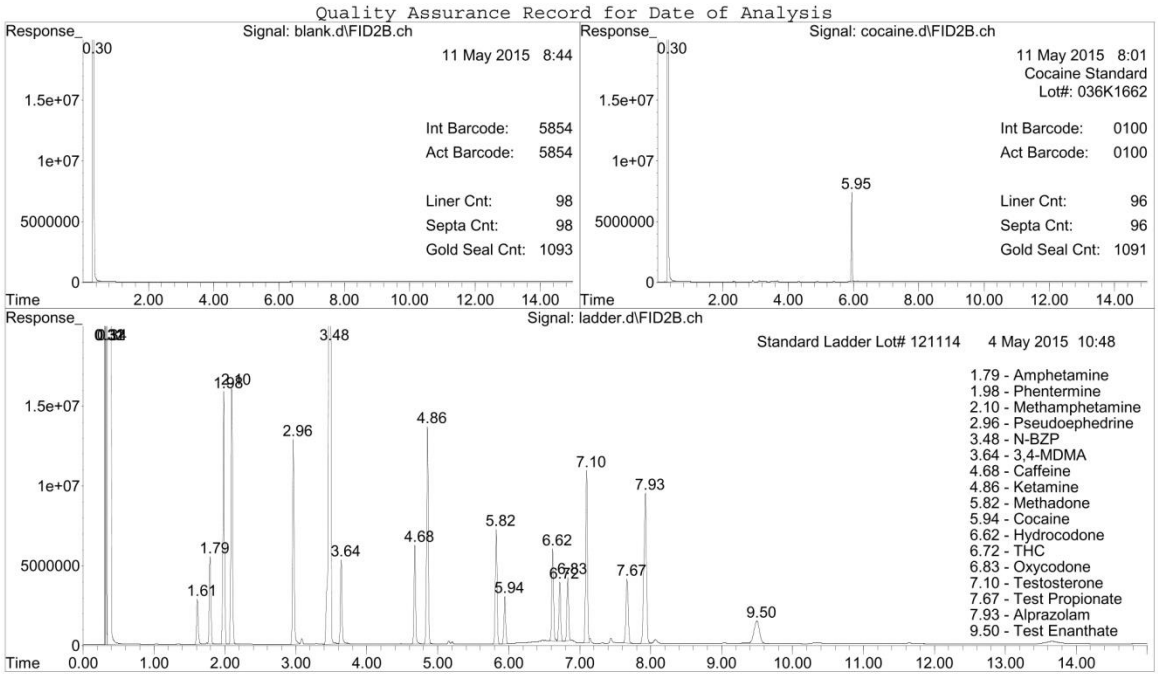




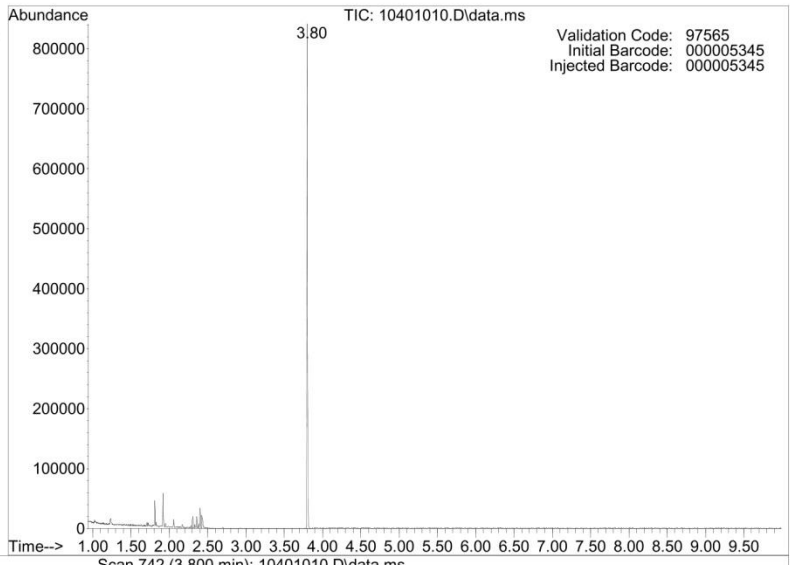
Appendix C: Extraction Data

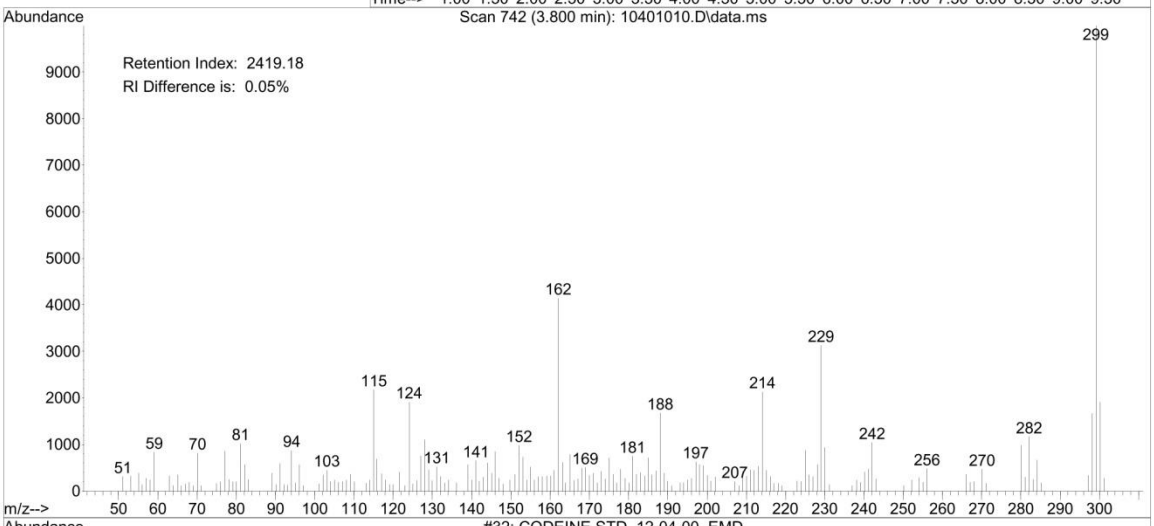
Appendix C-6: Morphine to Codeine Data

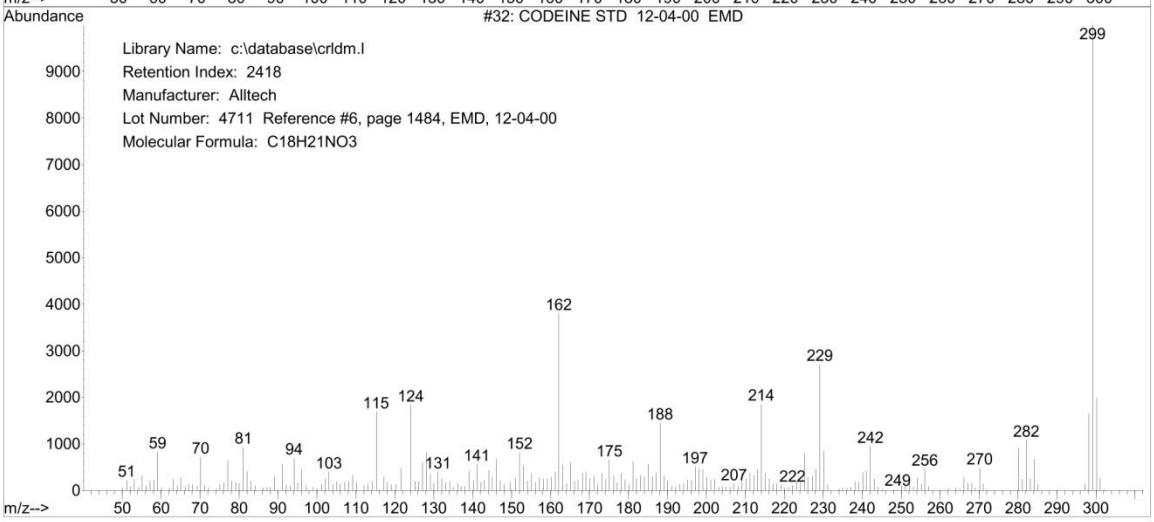


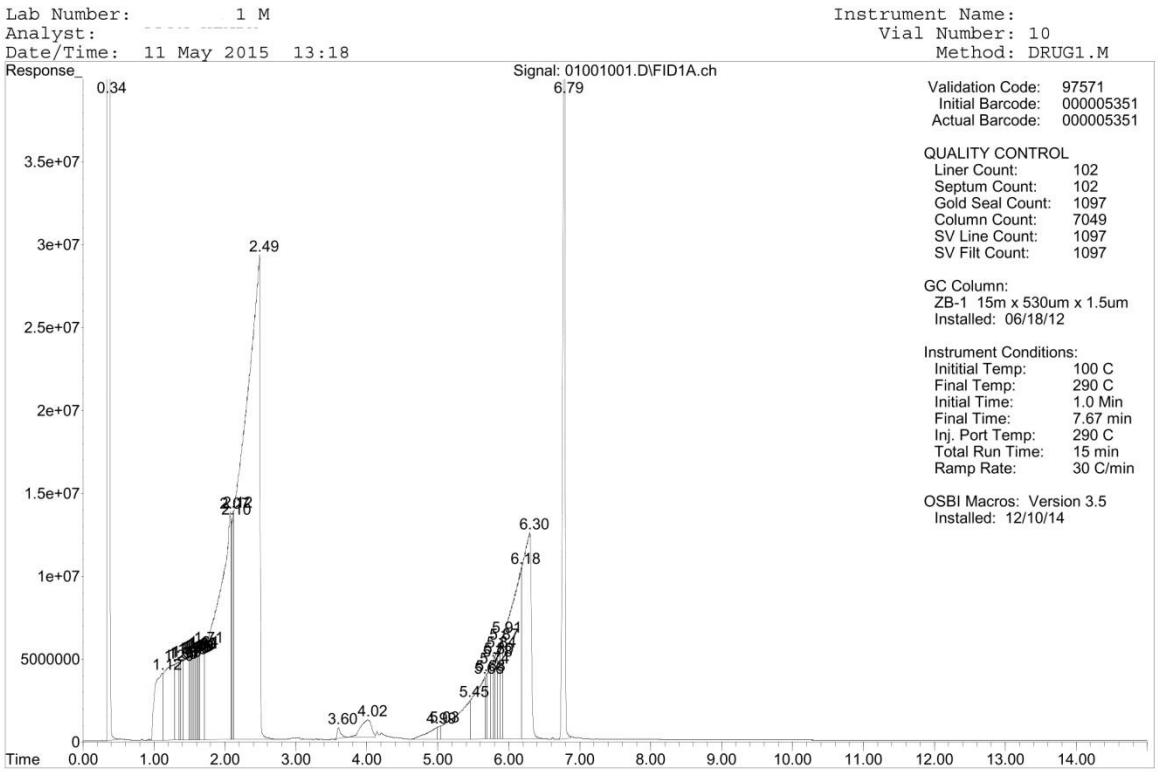


```
Lab Number:          1 N-C                                  Vial Number: 104
Instrument
Analyst:
Date/Time:   11 May 2015   12:03
Method:      DRUG10.M

QUALITY CONTROL
Liner count:  237
Septum count: 237
Gold Seal:    136
Column count: 1635
GC Column:
 ZB-1 12.0m x 0.20m x 0.33um
Column Installed:     02/23/15
 Hydrocarbon Ladder: 05/04/15
 Hydrocarbon Lot#:   071607

INSTRUMENT CONDITIONS
Initial temp:   100 'C
Initial time:   0.25 min
Final time:     6.15 min
Total Run Time: 10.0 min
Split Ratio:    10:1
Injection Vol:  1.0 ul
Inj. Port Temp: 290 'C
Transfer Line:  290 'C
Ramp Rate:
 65 'C/min to 220
 40 'C/min to 290
```

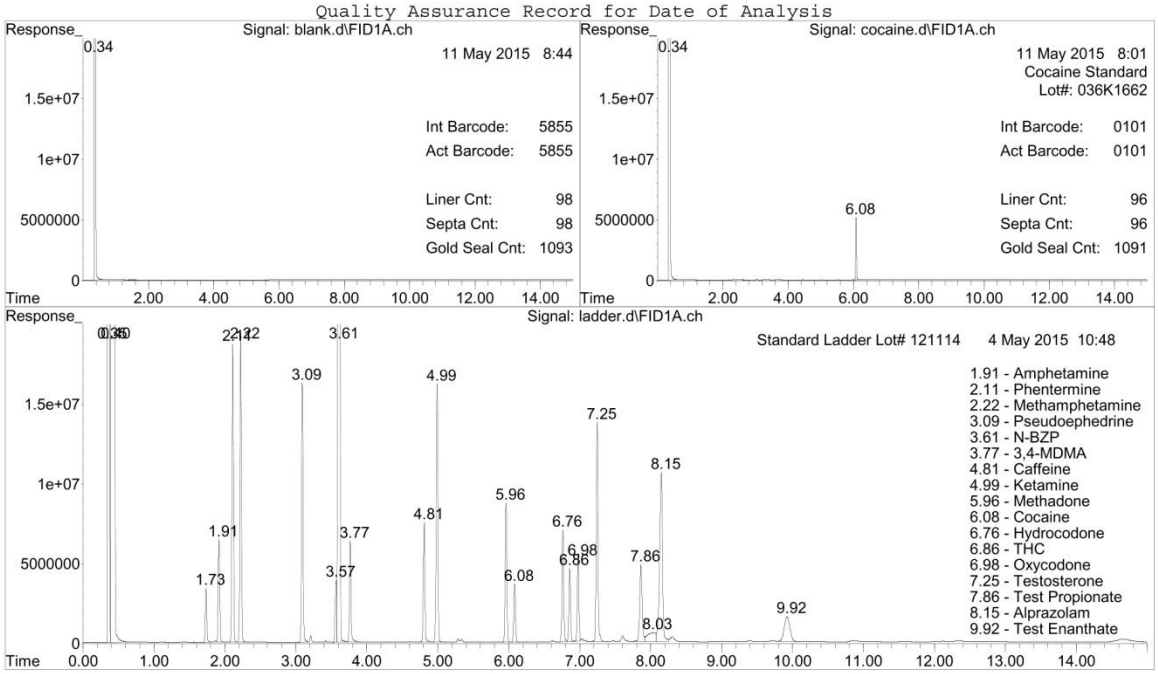




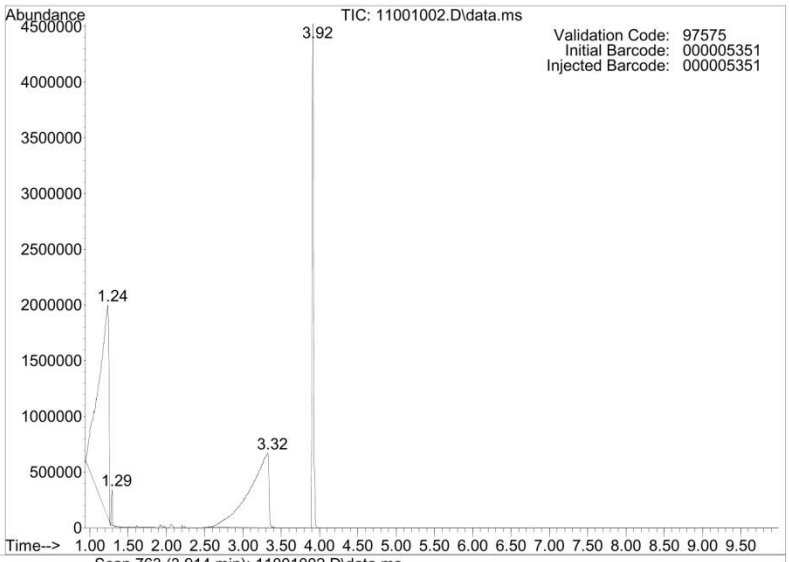


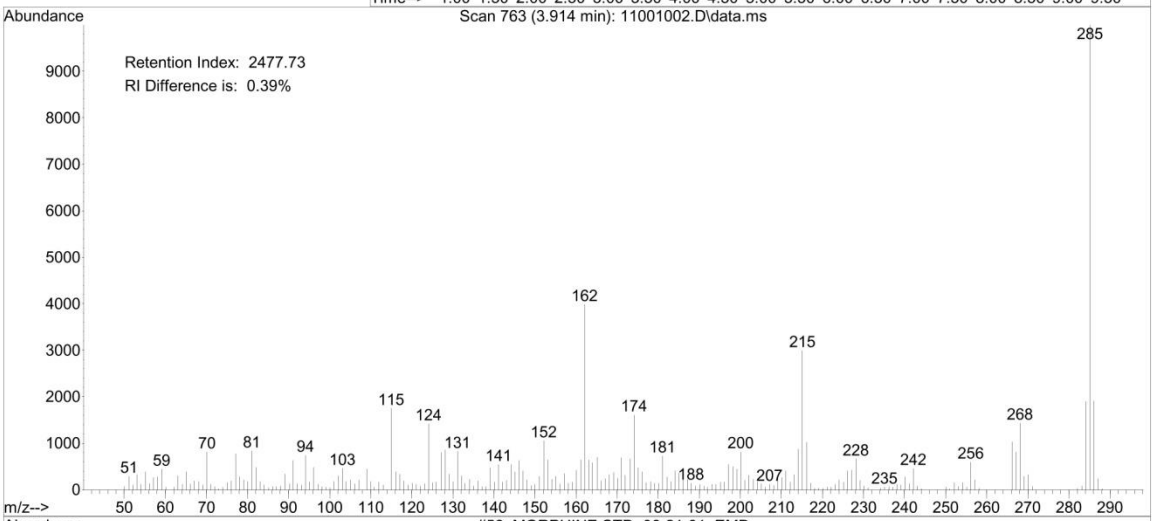


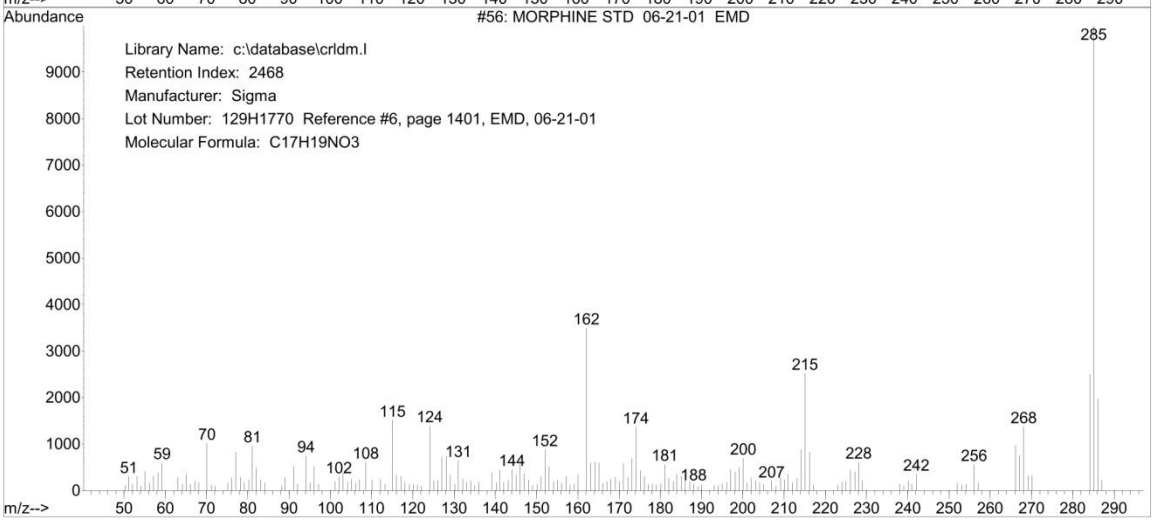
Quality Assurance Record for Date of Analysis



```
Lab Number:          1 M                                  Vial Number: 110
Instrument
Analyst:
Date/Time:  11 May 2015  13:58
Method:     DRUG100.M

QUALITY CONTROL
Liner count:  245
Septum count: 245
Gold Seal:    144
Column count: 1643
GC Column:
 ZB-1 12.0m x 0.20m x 0.33um
Column Installed:    02/23/15
 Hydrocarbon Ladder: 05/04/15
 Hydrocarbon Lot#:   071607

INSTRUMENT CONDITIONS
Initial temp:   100 'C
Initial time:   0.25 min
Final time:     6.15 min
Total Run Time: 10.0 min
Split Ratio:    100:1
Injection Vol:  1.0 ul
Inj. Port Temp: 290 'C
Transfer Line:  290 'C
Ramp Rate:
 65 'C/min to 220
 40 'C/min to 290
```



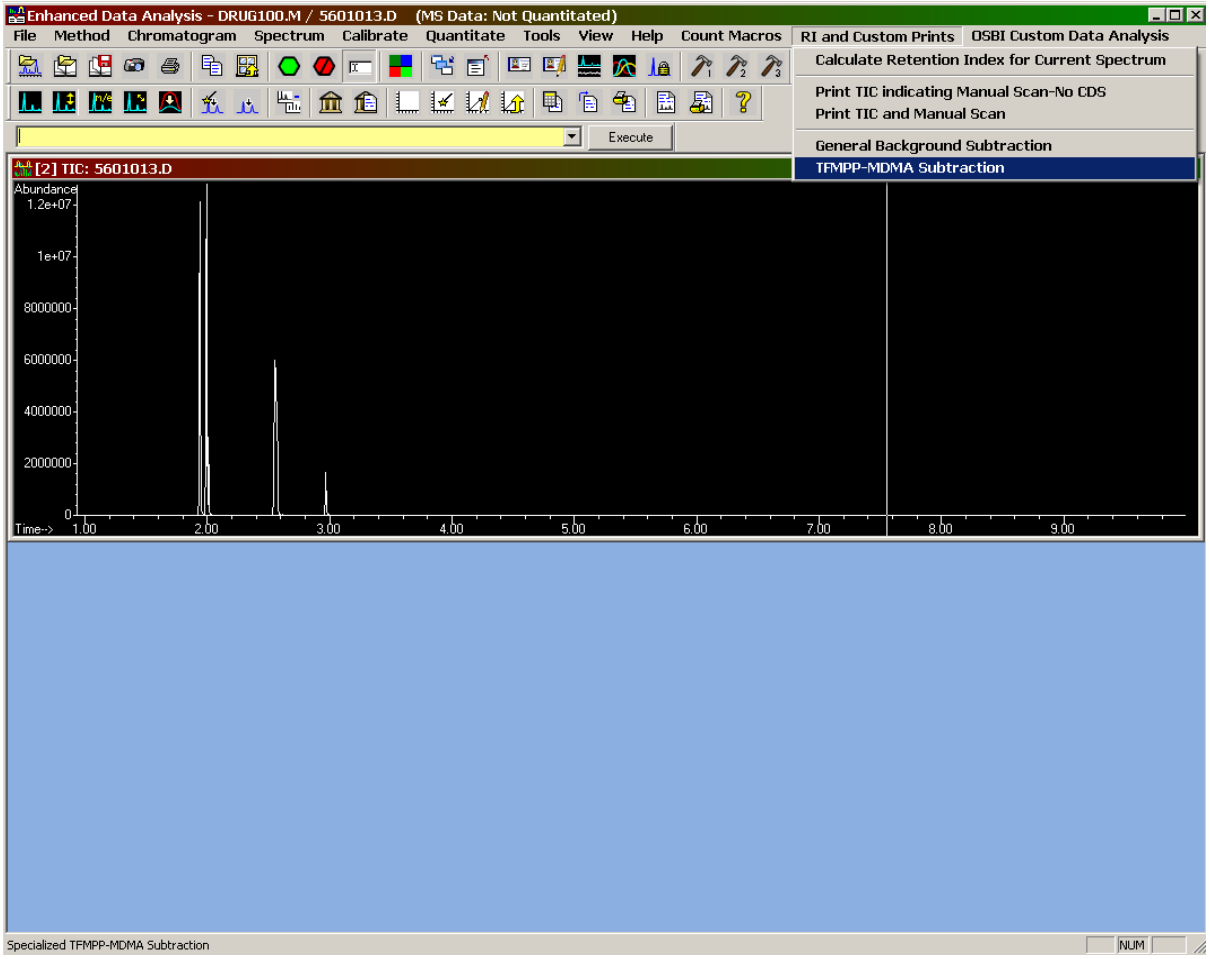




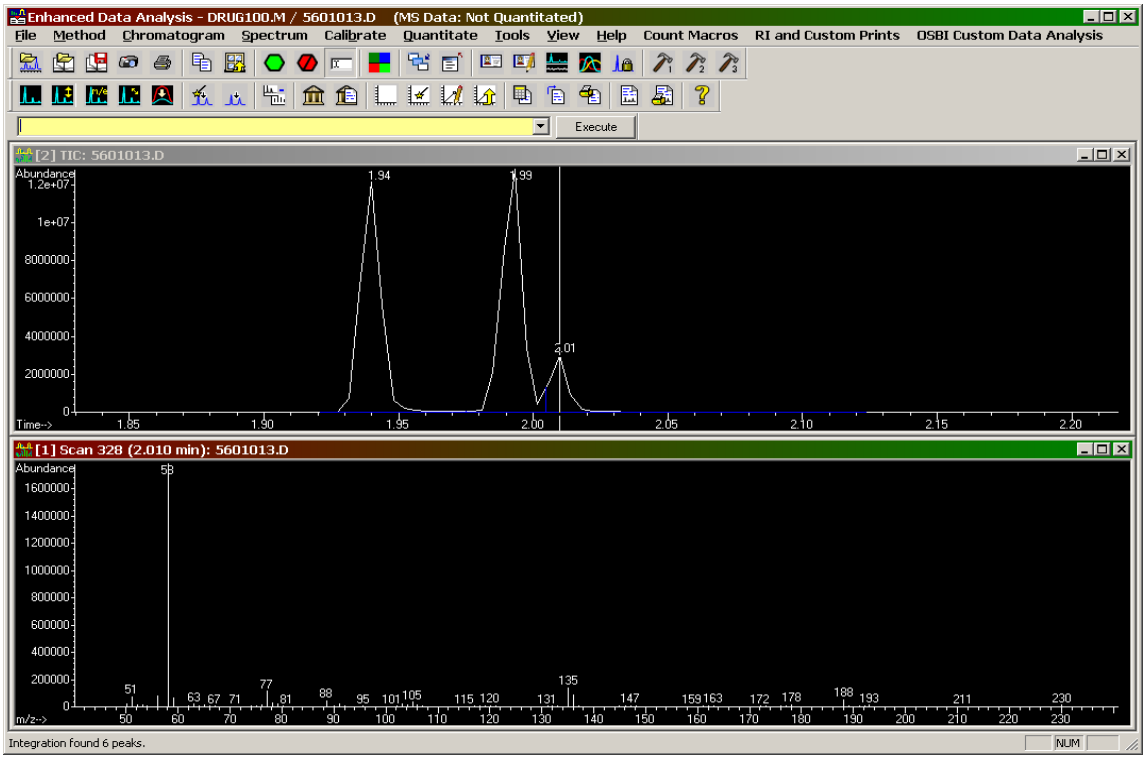
Appendix C: Extraction Data

Appendix C-7: Instructions for Specialized TFMPP-MDMA Subtraction

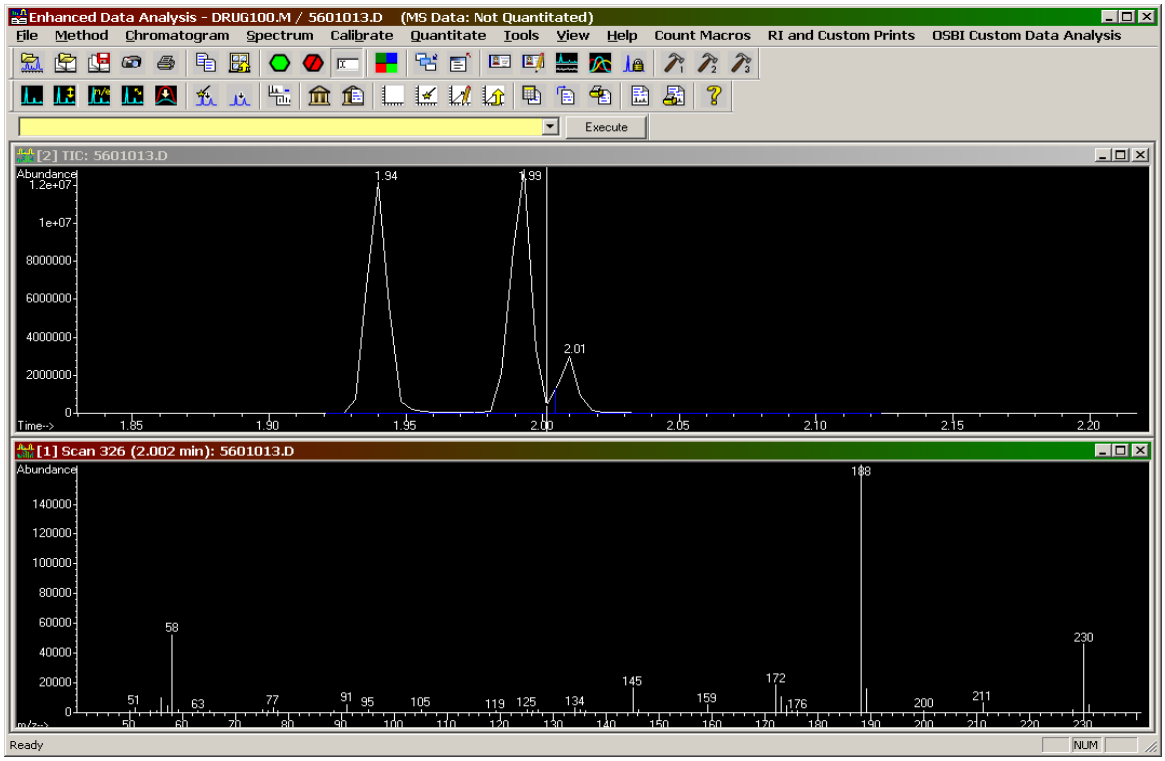
Under the GCMS data analysis toolbar heading “RI and Custom Prints” is a selection “TFMPP-MDMA Subtraction”. This is a specialized printout specifically for subtracting TFMPP from 3, 4-MDMA.



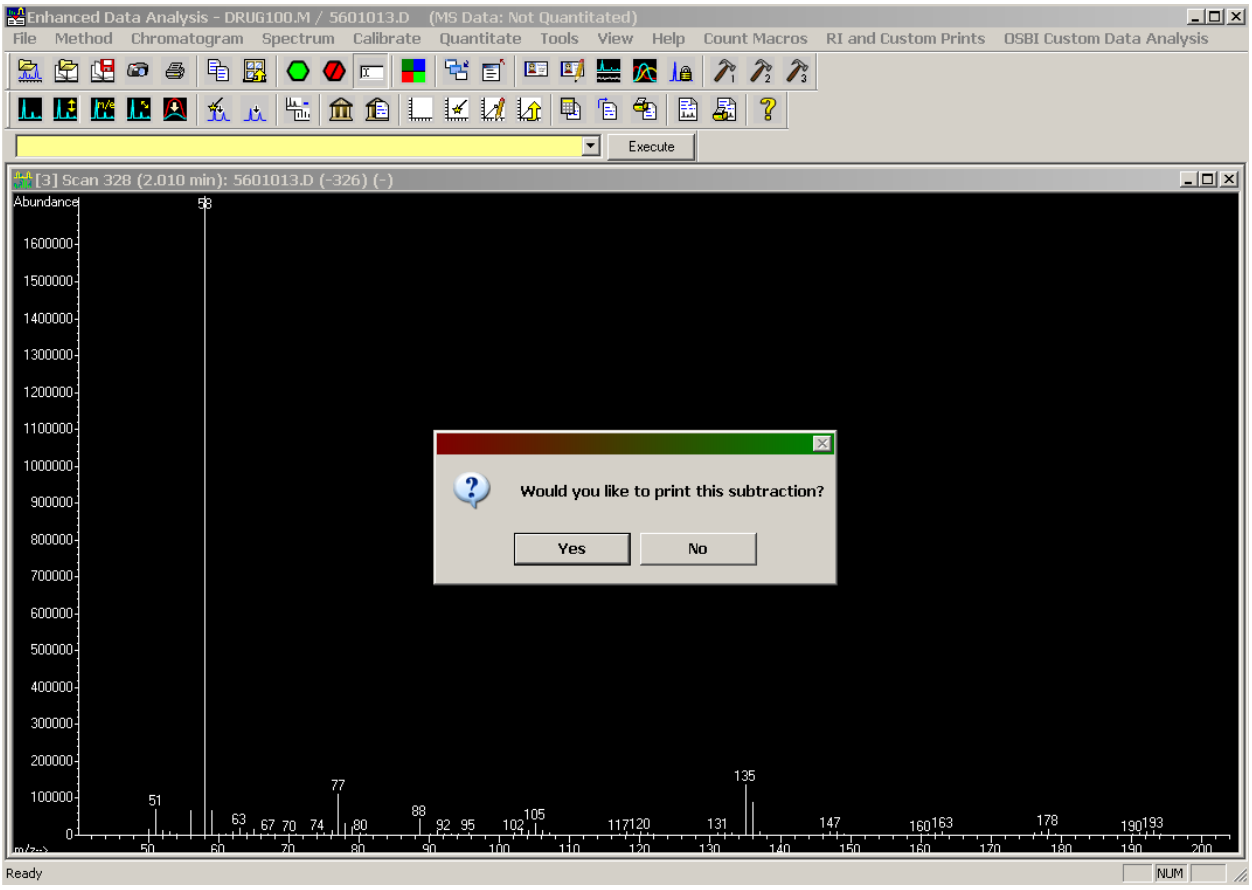
The first step is to select the scan of MDMA containing the co-eluting TFMPP. Select the same scan that was selected on the original print out from the data editing macros. This will keep the retention index calculations valid.



Then select the scan to background subtract:



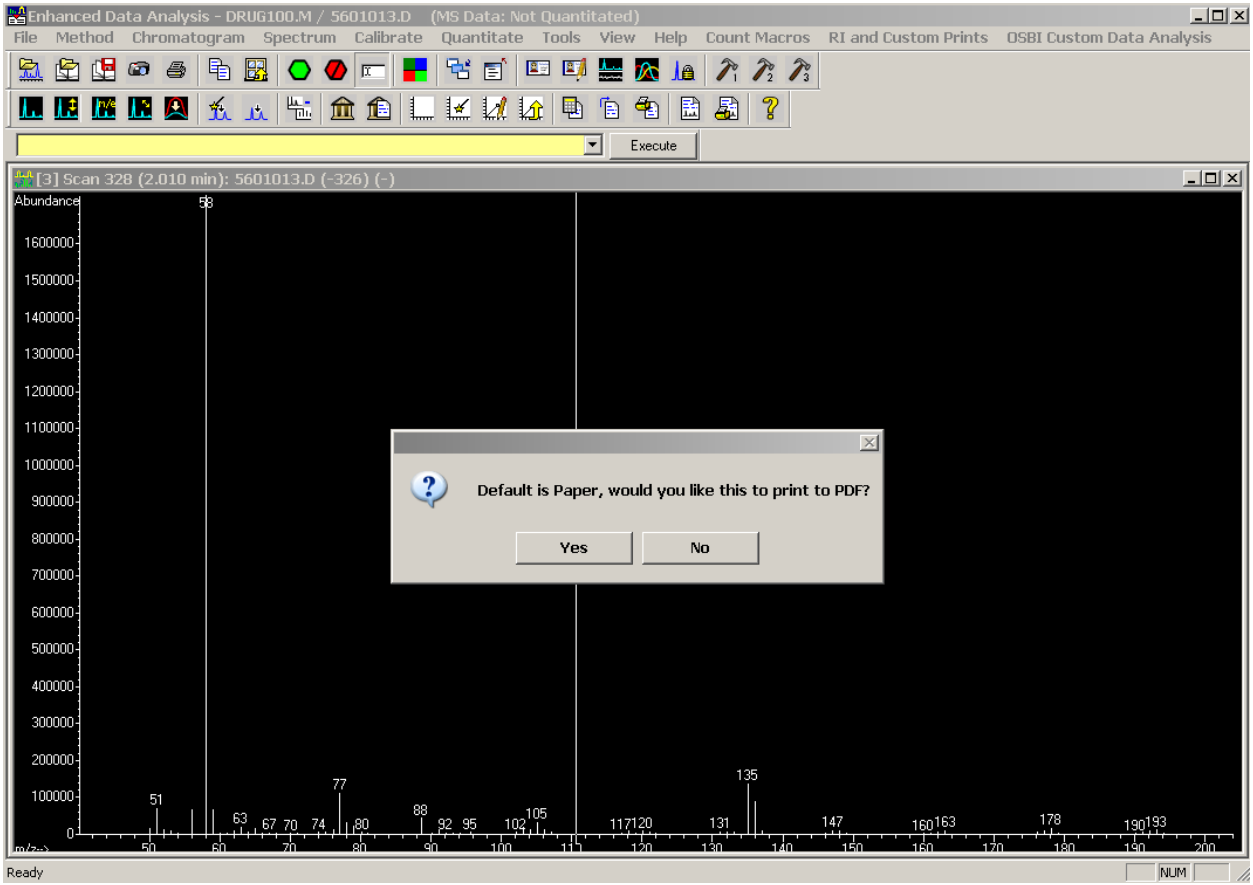
This is where the specialized printout for this background subtraction is used. Go to the "RI and Custom Prints" menu and select "TFMPP-MDMA Subtraction". The macro will perform the subtraction, display the subtraction, and ask if you would like to print this extraction:



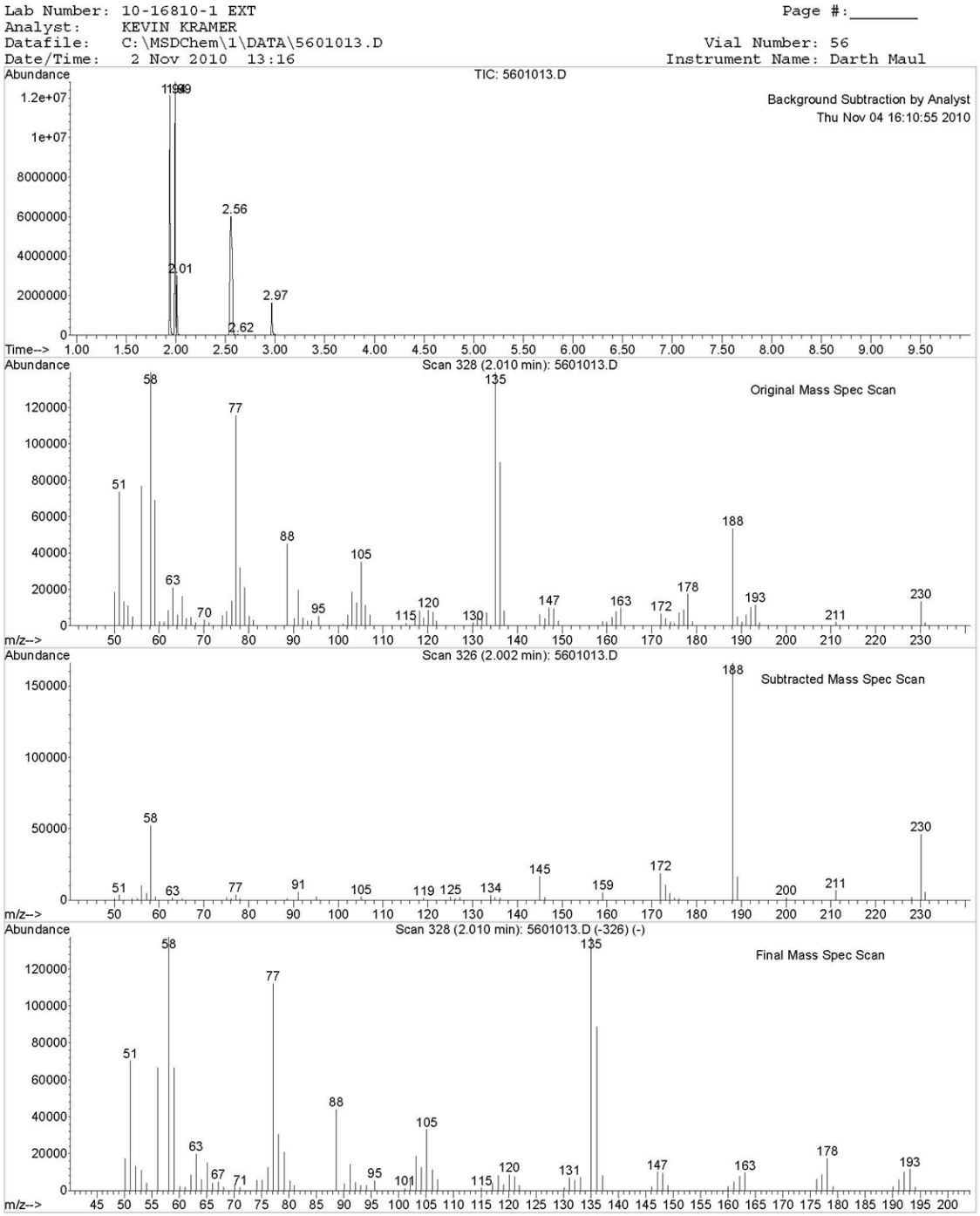
If "No" is selected, the macro will ask if you want to subtract the same spectrum again. Prompting to subtract the same spectrum again is really meant for subtracting a column background peak such as 341. If "No" is selected again, you will be returned to your screen without any print outs. At this point, the process can be either started over by selecting the scan with co-elution and then selecting a different scan to background subtract or data analysis can be exited.

This process is exactly how it is done using the "Subtract" selection under "Spectrum" on the data analysis toolbar. First select the sample scan and the spectrum to be subtracted second, and then subtract.

If "Yes" is selected, the prompt will ask "Default is Paper, would you like this to print to PDF?" Select "No" for paper, "Yes" for PDF.



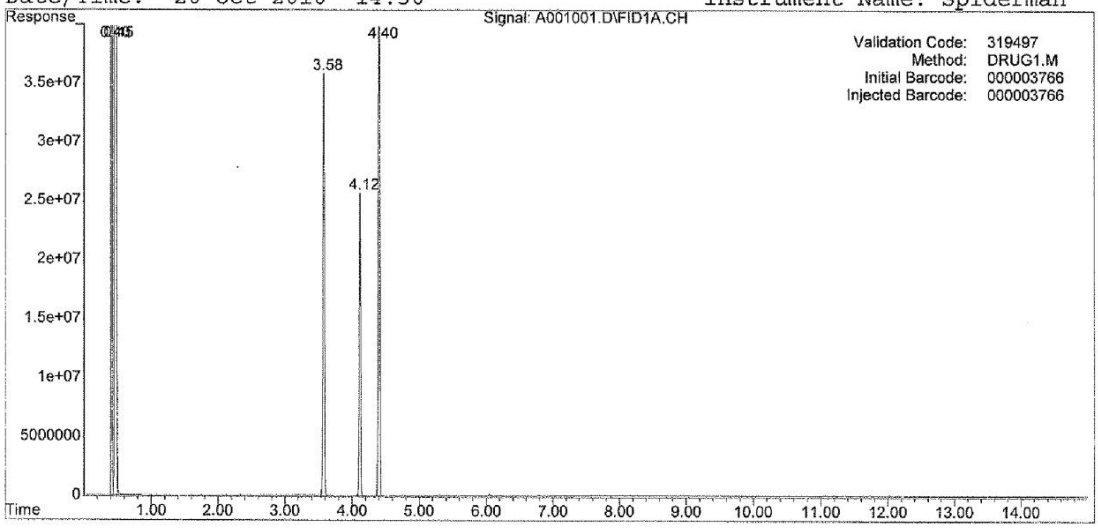
You will receive the following printout. This printout contains information required for the case file and can be easily uploaded into the BEAST or stored in a file. The printout contains the original TIC, original scan prior to being subtracted, the scan that is being subtracted, and the final scan after the subtraction has been done.



Appendix C: Extraction Data

Appendix C-8: TFMPP Isomer Order

Lab Number: TFMPP MIX N/C MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\1\data\A001001.D Vial Number: 100
Date/Time: 20 Oct 2010 14:50 Instrument Name: Spiderman



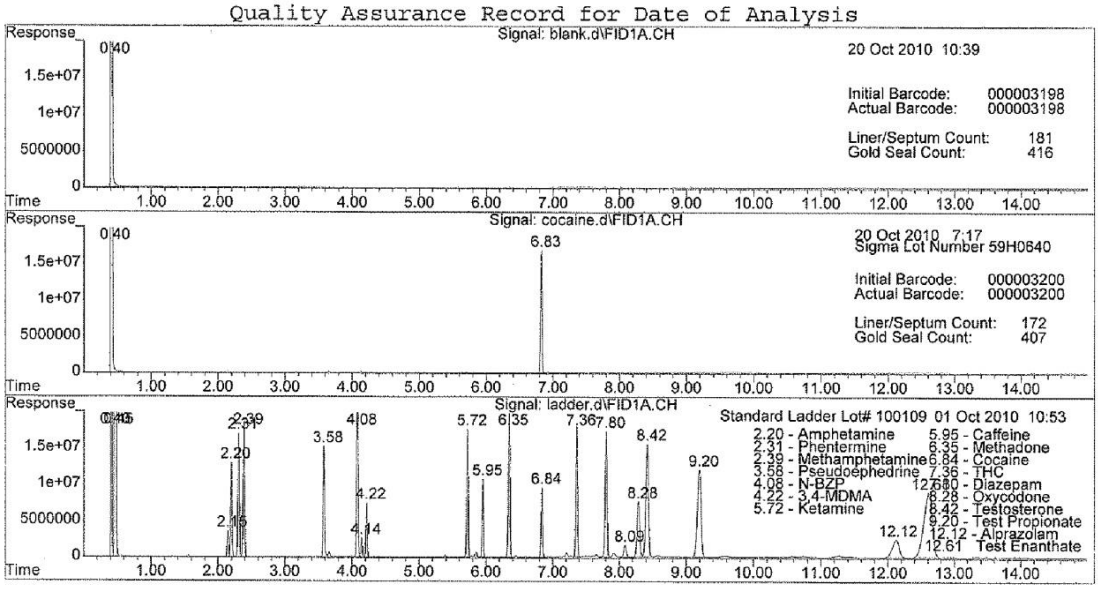
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

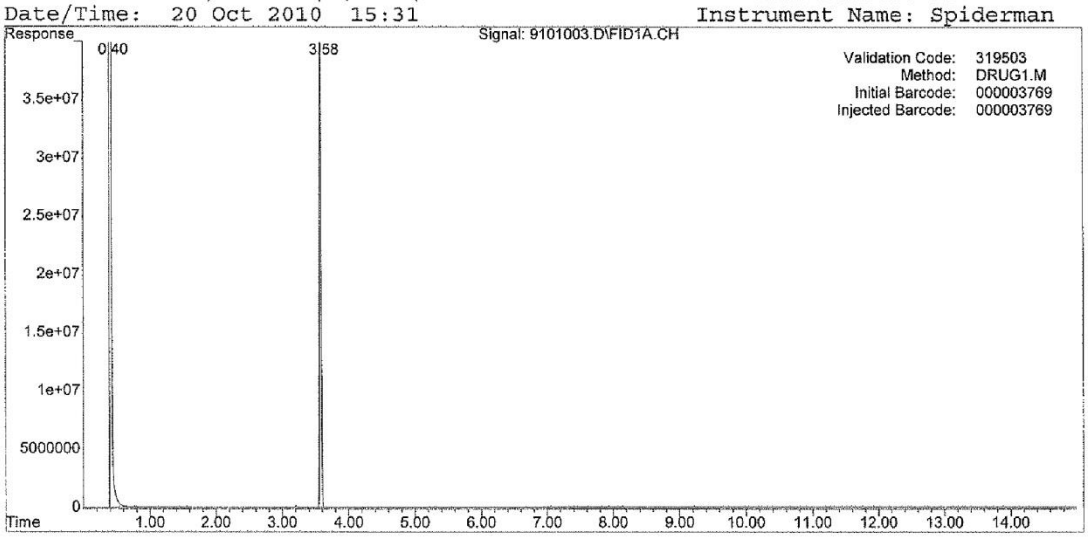
QUALITY CONTROL

Liner count:	185	GC Column:		OSBI MACROS	
Septum count:	185	ZB-50 15.0m x 0.53mm x 1.0um		Version 3.2	
Gold seal count:	420	Date Column Installed:	05/19/10	Install Date:	
Column count:	1448	Split Vent Line Count:	2711	05/19/10	

Quality Assurance Record for Date of Analysis



Lab Number: 1,2 TFMPP MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\1\data\9101003.D Vial Number: 91
Date/Time: 20 Oct 2010 15:31 Instrument Name: Spiderman



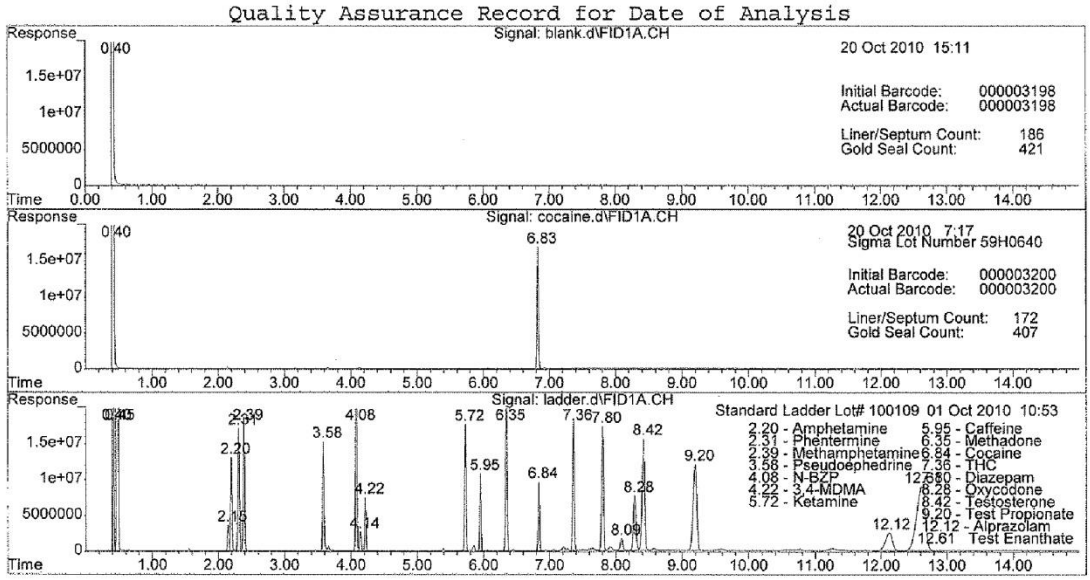
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

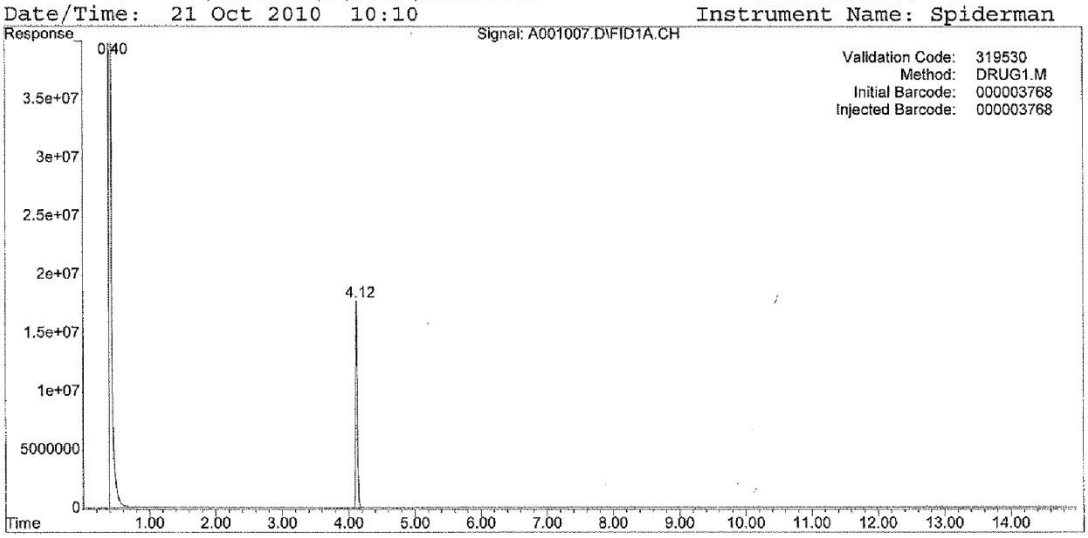
QUALITY CONTROL

Liner count:	187	GC Column:		OSBI MACROS	
Septum count:	187	ZB-50 15.0m x 0.53mm x 1.0um		Version 3.2	
Gold seal count:	422	Date Column Installed:	05/19/10	Install Date:	
Column count:	1450	Split Vent Line Count:	2713	05/19/10	

Quality Assurance Record for Date of Analysis



Lab Number: 1,3 TFMPP MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\1\data\A001007.D Vial Number: 100
Date/Time: 21 Oct 2010 10:10 Instrument Name: Spiderman



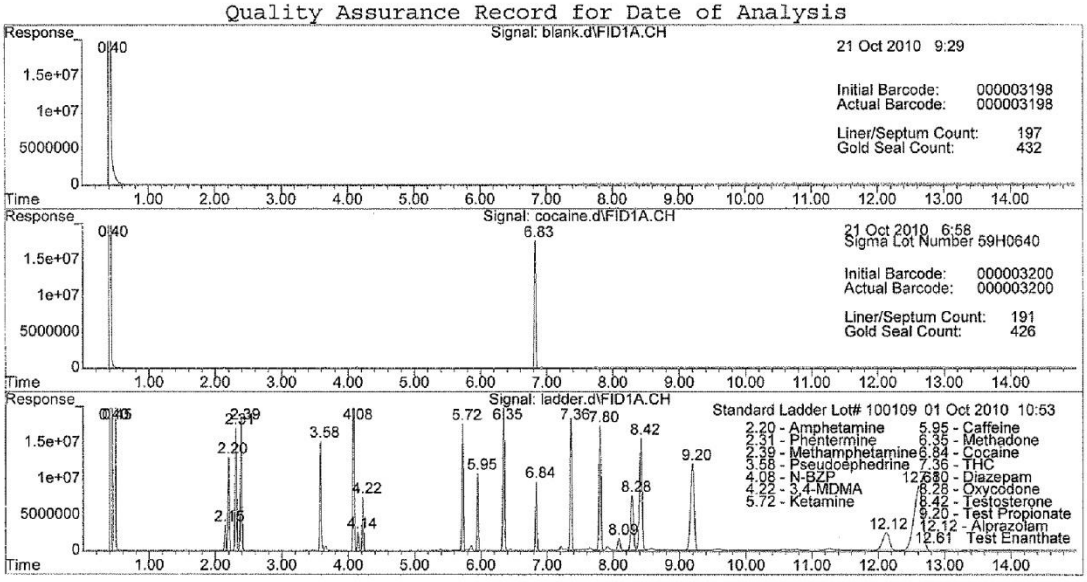
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

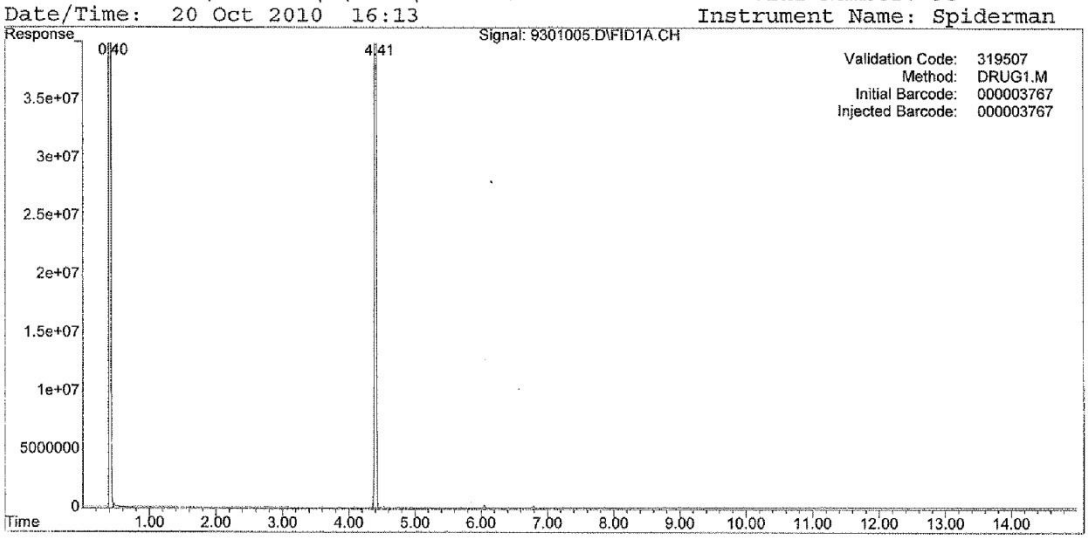
QUALITY CONTROL

Liner count:	199	GC Column:		OSBI MACROS	
Septum count:	199	ZB-50 15.0m x 0.53mm x 1.0um		Version 3.2	
Gold seal count:	434	Date Column Installed:	05/19/10	Install Date:	
Column count:	1462	Split Vent Line Count:	2725	05/19/10	

Quality Assurance Record for Date of Analysis



Lab Number: 1,4 TFMPP MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\1\data\9301005.D Vial Number: 93
Date/Time: 20 Oct 2010 16:13 Instrument Name: Spiderman



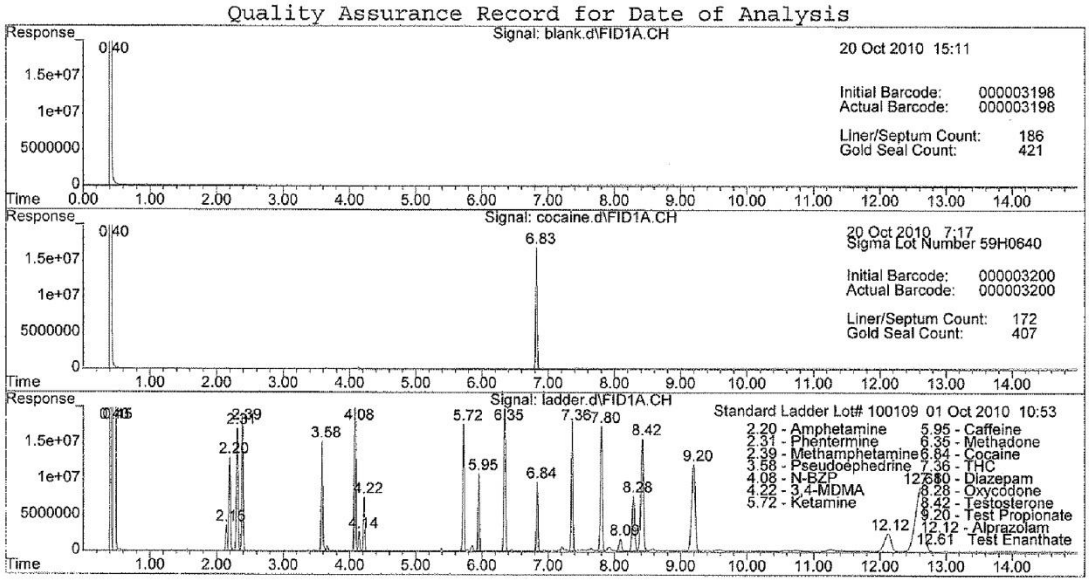
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

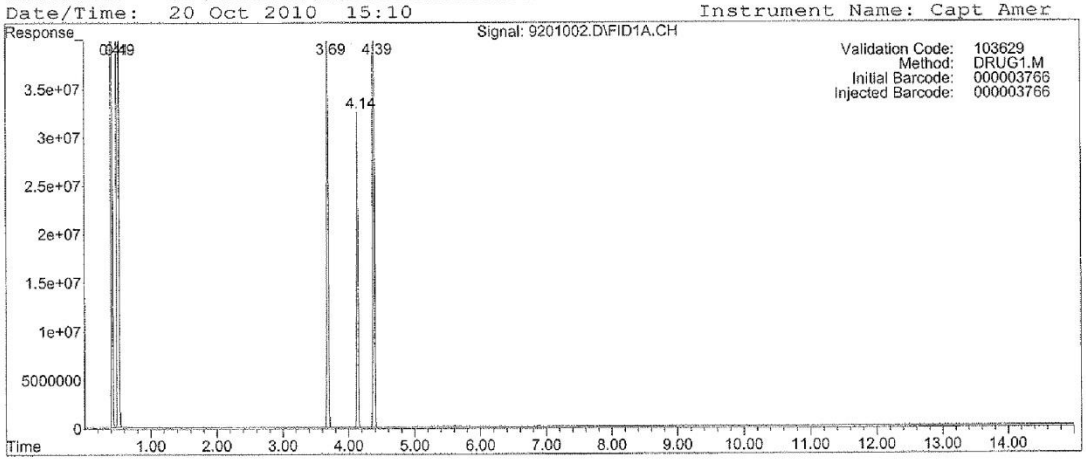
QUALITY CONTROL

Liner count:	189	GC Column:		OSBI MACROS	
Septum count:	189	ZB-50 15.0m x 0.53mm x 1.0um		Version 3.2	
Gold seal count:	424	Date Column Installed:	05/19/10	Install Date:	
Column count:	1452	Split Vent Line Count:	2715	05/19/10	

Quality Assurance Record for Date of Analysis



Lab Number: TFMPP MIX N/C MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\2\DATA\9201002.D Vial Number: 92
Date/Time: 20 Oct 2010 15:10 Instrument Name: Capt Amer



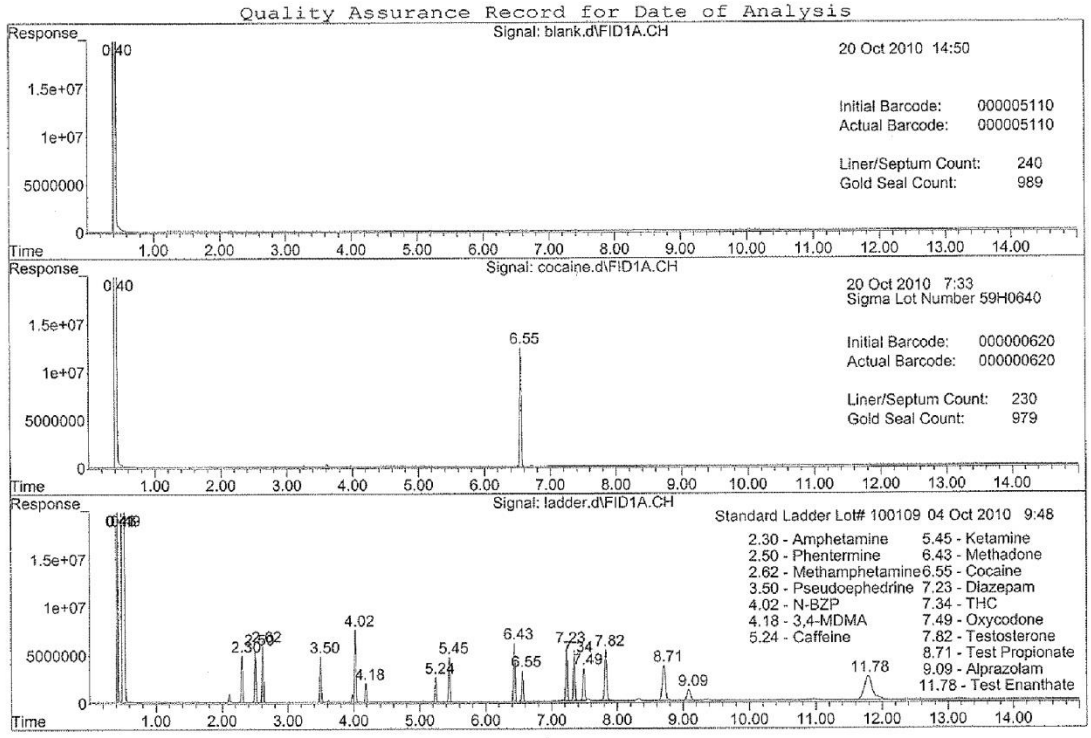
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

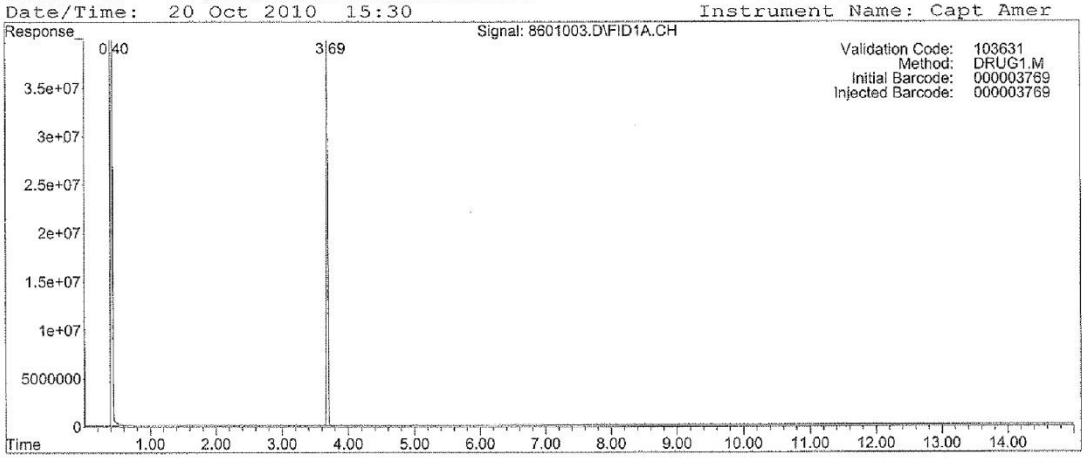
QUALITY CONTROL

Liner count:	241	GC Column:		OSBI MACROS	
Septum count:	241	DB-1 15m x 530um x 1.5um		Version 3.2	
Gold seal count:	990	Date Column Installed:	01/15/09	Install Date:	
Column count:	2380	Split Vent Line Count:	1248	04/05/10	

Quality Assurance Record for Date of Analysis



Lab Number: 1,2-TFMPP MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\2\DATA\8601003.D Vial Number: 86
Date/Time: 20 Oct 2010 15:30 Instrument Name: Capt Amer



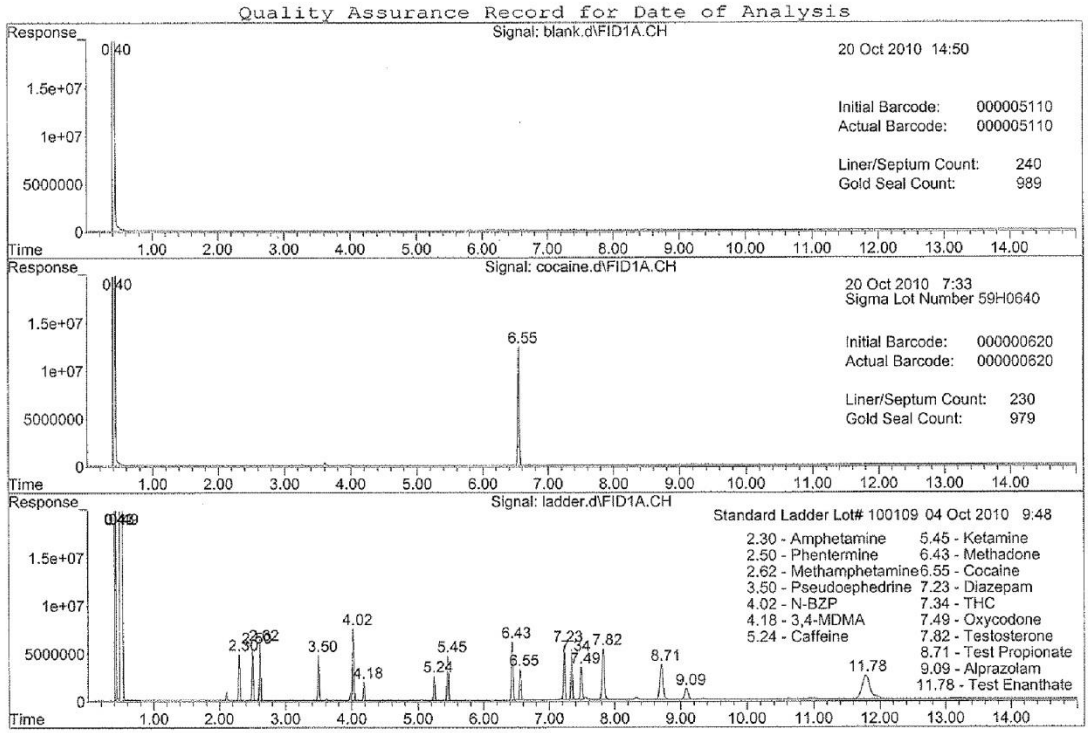
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

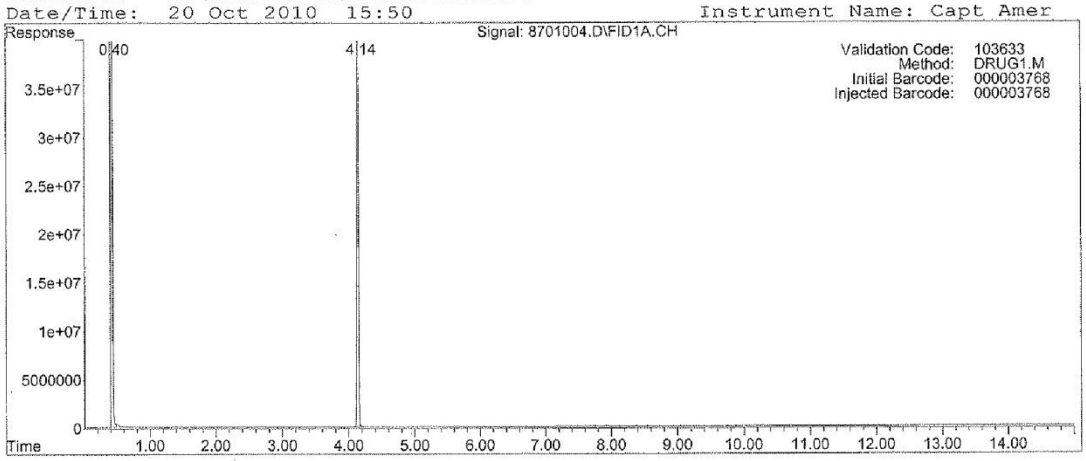
QUALITY CONTROL

Liner count:	242	GC Column:		OSBI MACROS	
Septum count:	242	DB-1 15m x 530um x 1.5um		Version 3.2	
Gold seal count:	991	Date Column Installed:	01/15/09	Install Date:	
Column count:	2381	Split Vent Line Count:	1249	04/05/10	

Quality Assurance Record for Date of Analysis



Lab Number: 1,3-TFMPP MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\2\DATA\8701004.D Vial Number: 87
Date/Time: 20 Oct 2010 15:50 Instrument Name: Capt Amer



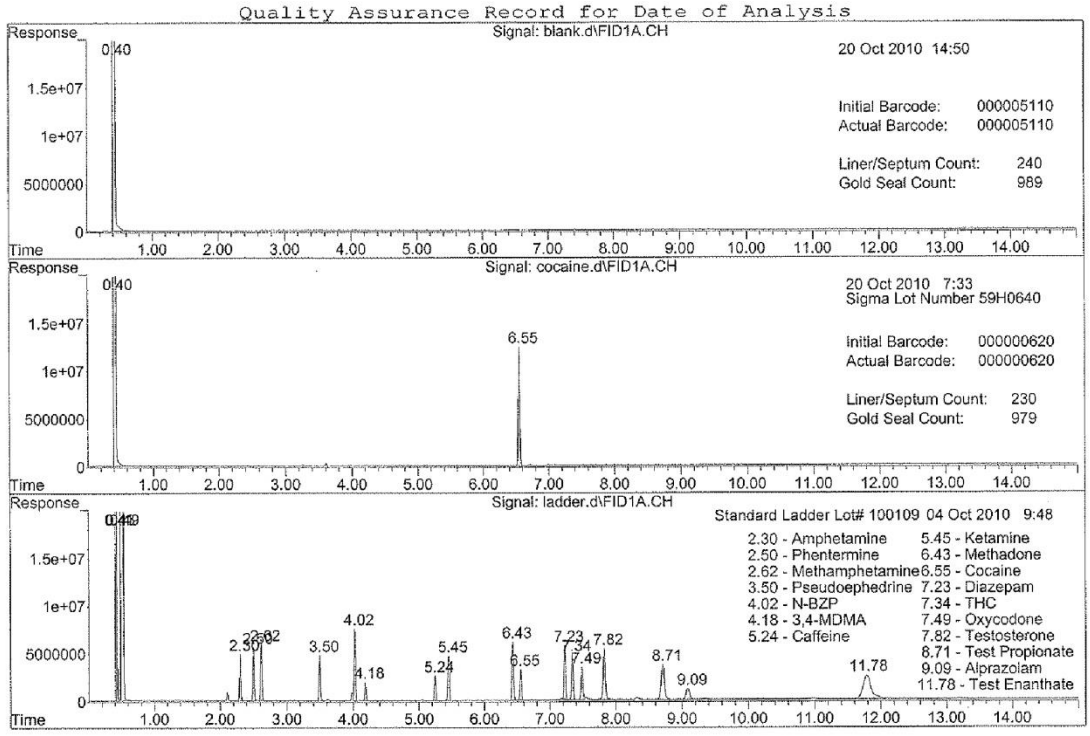
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

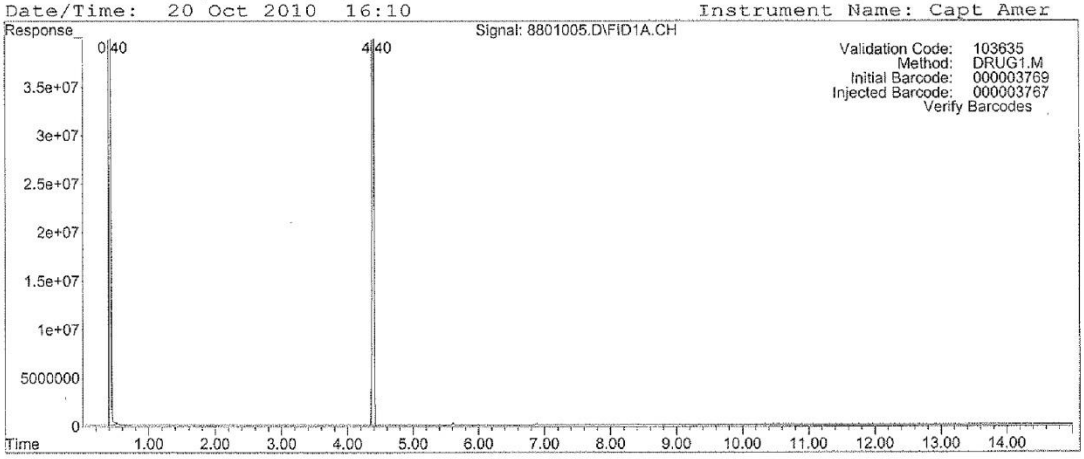
QUALITY CONTROL

Liner count:	243	GC Column:		OSBI MACROS	
Septum count:	243	DB-1 15m x 530um x 1.5um		Version 3.2	
Gold seal count:	992	Date Column Installed:	01/15/09	Install Date:	
Column count:	2382	Split Vent Line Count:	1250	04/05/10	

Quality Assurance Record for Date of Analysis



Lab Number: 1,4-TFMPP MDB Page #:_____
Operator: OSBI Drug Laboratory
Datafile: C:\MSDChem\2\DATA\8801005.D Vial Number: 88
Date/Time: 20 Oct 2010 16:10 Instrument Name: Capt Amer



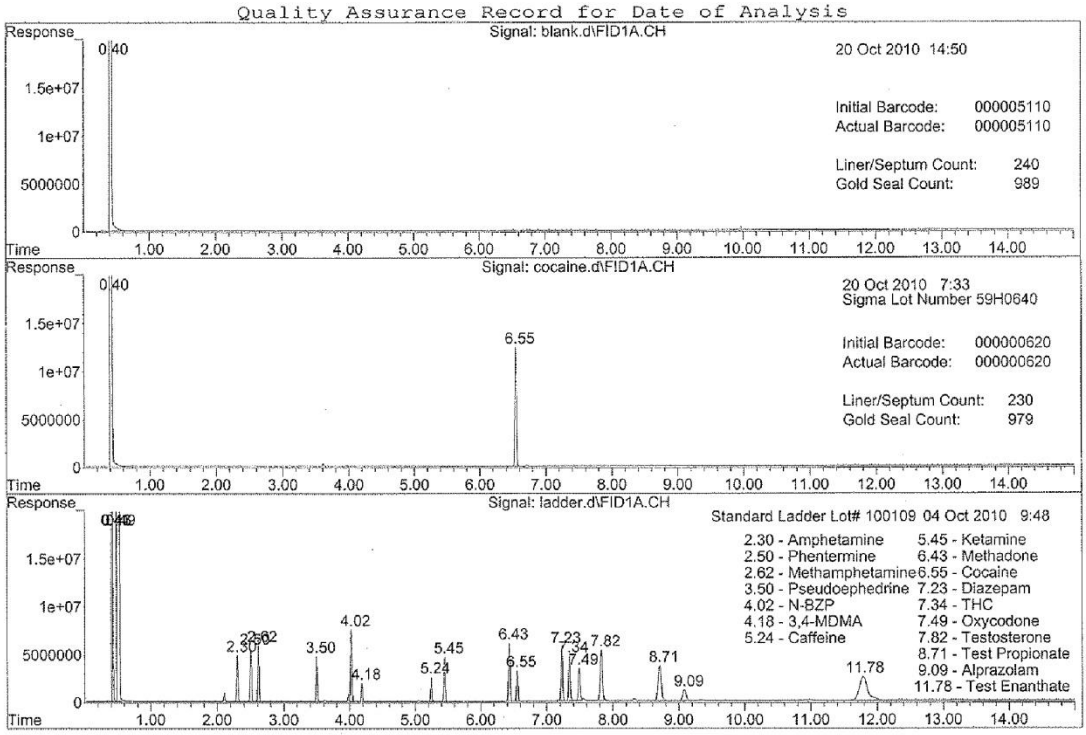
INSTRUMENT CONDITIONS

Initial temp:	100 C	Final temp:	290 C	Inj. Port Temp:	290 C
Initial time:	1.0 Min	Final time:	7.67 min	Total Run Time:	15 min
		Ramp Rate:	30 C/min		

QUALITY CONTROL

Liner count:	244	GC Column:		OSBI MACROS	
Septum count:	244	DB-1 15m x 530um x 1.5um		Version 3.2	
Gold seal count:	993	Date Column Installed:	01/15/09	Install Date:	
Column count:	2383	Split Vent Line Count:	1251	04/05/10	

Quality Assurance Record for Date of Analysis



Author(s) and Approvals

Author(s):

Revision 0 Heather Schafstall, Technical Manager of Forensic Chemistry
Revision 1 Heather Schafstall, Technical Manager of Forensic Chemistry
Revision 2 Heather Schafstall, Technical Manager of Forensic Chemistry
Revision 3 Heather Schafstall, Technical Manager of Forensic Chemistry
Revision 4 Michella Carter, Technical Manager of Controlled Substances
Revision 5 Michella Carter, Technical Manager of Controlled Substances
Revision 6 Michella Carter, Technical Manager of Controlled Substances
Revision 7 Michella Carter, Technical Manager of Controlled Substances

Approval:

Technical Manager: Michella Carter Date: 12/20/24

OSBI CSD Director: Date: 12/26/2024

History

Rev #	Issue Date	History
7	12-31-24	Moved the References for each protocol to the end of each protocol.
		DR-01 Incorporated the deviation from 10-01-24. Individual Steps of Protocol: Added "allow the samples to derivatize overnight" to the Other Suspected Cannabis Products. Removed TLC positive for delta 9-THC from the Recommended Wording for DR-01 Attachment 1.
		DR-02: Updated the Associated Protocols. Updated the name of the Specimen(s) Required section name to be consistent and changed the sample amount that needs to be collected. Reordered 2.a and b in Individual Steps of Protocol and added "made on paper or in Excel" to b. so it is clear that this is in the notes and not in the Matrix. Moved Protocol Note 7 up to 2. Clarified in Recommended Wording 3 that only the sampled bundles would be included in the UOM.
		DR-03: Removed kilograms from Recommended Wording 1. Updated the name of the reports in Recommended Wording 6. Added "approximate volume: 5 milliliters" to Recommended Wording 7.
		DR-04: Removed "5-pound weight" from the Steps of Protocol Large Capacity and replace it with individual weight to be consistent with the Analytical Balance section. In Frequency and Tolerance of Controls "(FSC Employees)" was removed since FSC and NERL will both have an analytical balance. Added that the analytical balance needs to have bracketing weights when used for trafficking cases. Removed the use of balance to count tablets in the Uncertainty of Measurement section since it can't be used with UOM. Updated the UOM on the examples in the Uncertainty of Measurement section. Added hyperlinks to the Associated Forms.
		DR-04 Attachment 3: The UOM was updated. The UOM for the Enid lab was archived since the lab closed in 2023.
		DR-07: Changed "general method" to "appropriate method" throughout. Added chloroform to the Reagents list. Removed the instruments from the Apparatus and Materials list to be consistent with the other protocols. Updated the Recommended Report Wording to include "examinations indicate the presence of a controlled substance; however, the laboratory's reporting criteria have not been met."
		DR-21: Specified the steps listed are for GC and GC/MS. Added the steps for FTIR.
		DR-30: Updated the Apparatus and Materials list to include Agilent GCs. Removed Protocol Note 6 because it was a repeat of Protocol Steps 3 and it is discussed in the calculations.
		DR-45: Updated the Reagents list to include the components of Ehrlichs. Added a caution statement for Fast Blue BB Salt in the preparation of Reagents. Added recommendation to store TLC reagent

		and Ehrlichs visualizing agent in the refrigerator. Removed Protocol Note 3 since it is covered in the Specimen Required section.
		DR-60: Added Associated Protocols. Updated the name of Specimen(s) Required section to be consistent.
		DR-70: Changed name of protocol to DR-70 Gas Chromatography Mass Spectrometer Methods for Drug Analysis to match DR-75. Changed "Instrument and Supplies" to "Apparatus and Supplies" for consistency. Updated Recommended Report Wording to include report wording for when an examination indicates a controlled substance.
		DR-75: Removed that the libraries are stored on a floppy disk, etc in the Standards section. Added a link to the Associated Form(s).
		DR-101: Updated the Associated Protocols so they are consistent with other protocols.
		DR-103: Updated the last sentence of the Purpose to say "see TM for archived DR-102." Removed DR-102 from the Associated Protocols.