Roy Cooper
Governer
Elizabeth Biser
Secretary
Michael A. Abraczinskas
Director

NORTH CAROLINA
Environmental Quality

DAQ-03-004.2 Standard Operating Procedure (SOP)

Thermo Ultimate 3000 Ultra High-Performance Liquid Chromatography by Ultra-Violet Radiation and Mass Spectrometry (UAT and PAMS Carbonyl Analyses)

Operator Responsibilities

North Carolina Department of Environmental Quality | Division of Air Quality
217 West Jones Street | 1641 Mail Service Center | Raleigh, North Carolina 27699-1641
919.707.8400

1.0 Approval Sign-Off Sheet

I certify that I have read and approve DAQ-03-004.2 Standard Operating Procedures for the Thermo Ultimate 3000 Ultra High-Performance Liquid Chromatography by Ultra-Violet Radiation and Mass Spectrometry (UAT and PAMS Carbonyl Analyses) written here with an effective date of June 29, 2021.

Director, Air Quality Division

Michael Abraczinskas

DocuSigned by:
Signature: Mark Cuilla for Michael Abraczinskas Date: 6/30/2021
CBE5596741C4437...

Ambient Monitoring Section Chief

Patrick Butler

DocuSigned by:
Signature: Patrick Butler Date: 6/30/2021
4290A7E2D2A0429...

Laboratory Analysis Branch Supervisor

Jim Bowyer, Environmental Program Supervisor

DocuSigned by:
Signature: Jim Bowyer Date: 6/30/2021
C96894826C9B41D...

Projects and Procedures Branch Supervisor

Joette Steger, Environmental Program Supervisor

DocuSigned by:
Signature: Date: 6/30/2021
B278F8186CEC4DC...

Primary SOP Author

Chaitali Bhaumik, Chemist I

DocuSigned by:
Signature: Chaitali Bhaumik Date: 6/30/2021
BE2629DB877140F...

EPA Region 4

Designated Approving Official

Signature:______________________________ Date: ______________

Disclaimer: This document, and any revision hereto, is intended solely as a reference guide to assist individuals in the operation of the instrument, related to the North Carolina Division of Air Quality's Ambient Monitoring Program

SOP Acronym Glossary

ACS – American Chemical Society

AQS - Air Quality System (EPA's Air database)

CFR – Code of Federal Regulations

COC – Chain of Custody

CV – Coefficient of Variation

DAQ - North Carolina Division of Air Quality

°C - degrees Celsius

DNPH - Dinitrophenylhydrazine

EPA – United States Environmental Protection Agency

FB – Field Blank

g – gram

HPLC – high performance liquid chromatography

IR - infrared

≤ - less than or equal to

L – Liter

L/min – liters per minute

MDL – Method detection limit

mg - milligram

mL- milliliter

mL/min – milliliters per minute

mM - millimolar

mm Hg – millimeters of mercury

MS – mass spectrometer

NIST – National Institute of Standards and Technology

PM – Particulate matter

PAMS – Photochemical Assessment Monitoring Station

PPB – Projects and Procedures Branch

PPE – Personal protective equipment

PTFE – Polytetrafluoroethylene

QA – Quality assurance

QA/QC - Quality assurance/quality control

QAPP - Quality assurance project plan

QC – Quality control

RCL – Reedy Creek Laboratory

RCO – Raleigh central office

RPD – Relative percent difference

SIF – Carbonyl Cartridge Sample Information Form

SN – Serial Number

SOP - Standard operating procedure

SPE – Solid phase extraction

TSA - Technical systems audit

UAT – Urban Air Toxics

UHPLC – Ultra High-Performance Liquid Chromatography

UHPLC-UV-MS – Ultra High-Performance Liquid Chromatograph with Ultra-Violet and Mass Spectrometer Detectors

UPS – United Parcel Service

USB – universal serial bus

UV – ultra-violet

µg/mL – micrograms per milliliter

µL - microliter

µm – micrometer

- number

± - plus or minus

Contents

2.0 SCOPE AND PURPOSE

This standard operating procedure (SOP) describes the process of carbonyl cartridge sample shipping, receiving, extraction, analysis, data processing and review using the Ultra High-Performance Liquid Chromatograph with Ultra-Violet Radiation Detector and Mass Spectrometer (UHPLC-UV-MS) instrumentation.

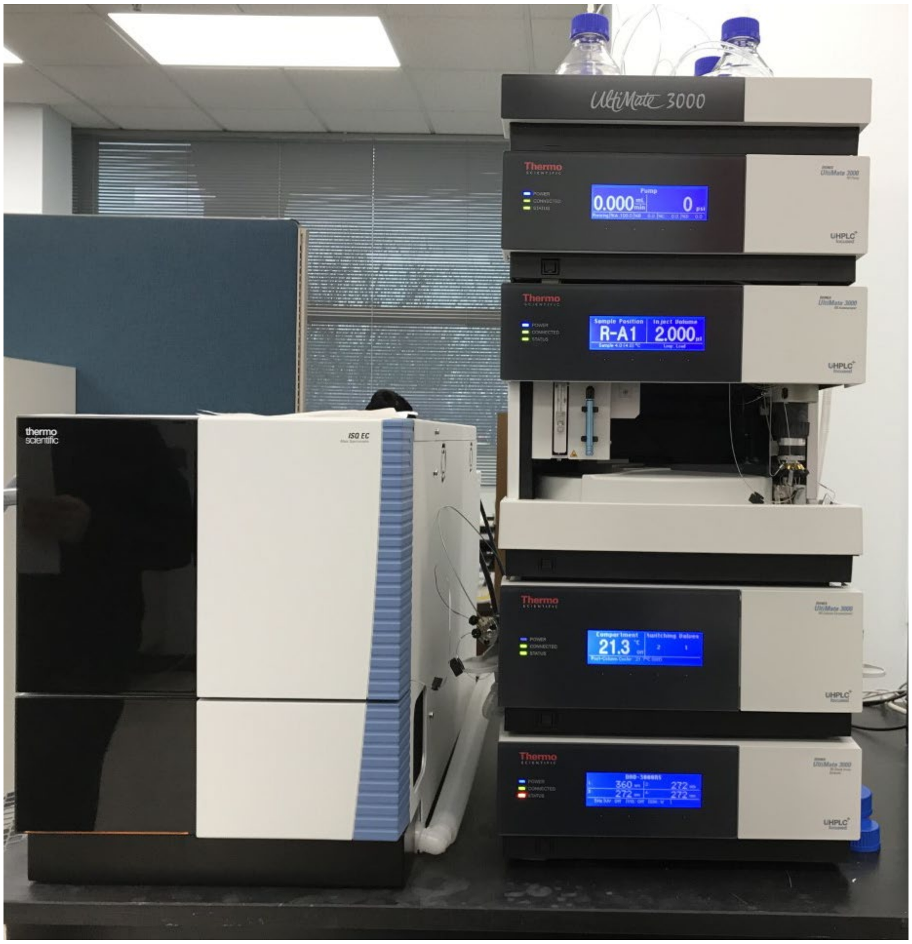


Figure 1: Thermo Ultra High-Performance Liquid Chromatograph with Ultra-Violet Radiation Detector and Mass Spectrometer

3.0 EQUIPMENT CHECKS AND MATERIALS

This section describes the equipment materials that are required to complete the steps described in this document. Additional subsections or SOPs will also describe the equipment and materials as needed.

3.1 Equipment and Material List

- Carbonyl Cartridge Sample Information Forms (SIF)
- Carbonyl Cartridge Chain of Custody Forms (COC)
- Insulated Shippers and Frozen Ice Packs
- Ziploc® Bags
- Infrared (IR) Temperature Monitoring Device
- Waters dinitrophenylhydrazine (DNPH) Coated Sorbent Cartridges (part# WAT037500)
- Carbonyl Cartridge Extraction Manifold Worksheet
- Solid Phase Extraction Manifold
- Vacuum Pump
- Sterilized 5-milliliter (mL) Syringes with Luer-Slip Tips
- Syringe Filters Discs less than or equal to (≤) 0.22-micrometer (µm) Pore Size
- Polyethylene Disposable Transfer Pipettes
- 5-mL Volumetric Flasks
- 1.5-mL Auto-sampler Vials with polytetrafluoroethylene (PTFE) Septa Caps
- 4-mL Storage Vials with PTFE Lined Caps
- Carbonyl-free Grade Acetonitrile
- Optima/High Performance Liquid Chromatography (HPLC)-Grade Water
- Optima-Grade Methanol
- American Chemical Society (ACS)-Grade Ammonium Acetate Salt
- ACS-Grade Glacial Acetic Acid
- 1 or 2-Liter (L) Glass Mobile Phase Reservoirs
- 1-L Volumetric flask
- Restek CARB-1004 Carbonyl standard mix (13 compounds)
- Calibration Standard and Quality Assurance (QA) Check Standard Check List
- Thermo Ultimate 3000 Ultra High-Performance Liquid Chromatography (UHPLC) Gradient Pump (part# LPG-3400RS)
- Thermo Ultimate 3000 UHPLC In-Line Degasser
- Thermo Ultimate 3000 UHPLC Temperature Controlled Autosampler (part# WPS-3000TRS)
- Thermo Ultimate 3000 UHPLC temperature-controlled column compartment (part# TCC-3000RS)
- Thermo Ultimate 3000 UHPLC Diode Array Detector (part# DAD-3000RS)
- Thermo ISQ EC Single Quadrupole Mass Spectrometer Detector (SN: ISQEC1809014)
- Peak Scientific Nitrogen Generator (part # NM32LA 110V)
- Computer Workstation with Chromeleon Software Installed and Connected to UHPLC System
- 10-100-microliter (µL) and 100-1000-µL Pipettes and Tips
- Analytical Balance

3.2 Chemical and Material Checks

This section describes the chemical, material, or equipment checks.

3.2.1 Chemicals and Solvents

Carbonyl-free grade acetonitrile and calibration/quality assurance/quality control (QA/QC) standard mixes must be used within the expiration date printed on the certificate of analysis. Optima grade water and methanol are given a two-year expiration date from the date of receipt, recorded on the bottle label. Acids, bases, and salts are given a 10-year expiration date from the date of receipt, recorded on the bottle label. Lot numbers of chemicals and materials used are recorded where applicable in lab logbooks, extraction records, and dilution records.

3.2.2 DNPH carbonyl cartridges

DNPH carbonyl cartridges must be used within the expiration date printed on the certificate of analysis that accompanies each box of cartridges. Blank Cartridge media must be stored in a dedicated refrigerator at 8 degrees Celsius (°C) or below. Sampled Cartridges must be stored in a different refrigerator at 4°C or below. For each new lot of carbonyl cartridge media, three cartridges per lot or 1 percent of the lot whichever is larger must be extracted and analyzed to assess background contamination of target carbonyl compounds.

The following compounds must meet concentrations and acceptance limits before field use: Formaldehyde ≤ 0.03 micrograms per milliliter (µg/mL); and all other compounds ≤ 0.02 µg/mL to pass. If Formaldehyde and/or Acetaldehyde do not meet acceptance limits, the lot must be disqualified for field use and returned to the manufacturer for replacement.

3.3 UHPLC Instrument Module Checks

1. Visually make sure instrument modules are connected to the instrument computer via Ethernet connections. Instrument modules display a green light next to the word "connected" on the instrument module front panel when successfully connected to the instrument computer.
2. Open Chromeleon using the instrument computer. Make sure the instrument modules (Pump, Sampler, Column oven, ultraviolet (UV), ISQEC, and Nitrogen generator) are connected and display a ready status on the instrument computer control screen.
3. Make sure instrument controller is connected and operating, if not, click on the instrument controller icon in the bottom right corner of the computer screen and click start the instrument controller.
4. Verify the mobile phase reservoirs A, B and C are connected to the appropriate tubing of the pump degasser.
5. Make sure column oven module temperature control is on and left switching valve is appropriate (Valve 1 is for UV analysis only and Valve 2 is for UV and mass spectrometer [MS] analysis).

3.4 Support Equipment Checks

Verify the pipette, balance, and IR-gun are being used before the expiration dates listed on the equipment stickers. Balance and pipette checks are completed quarterly and are explained in more detail in **Sections 3.4.1** and **3.4.2** below. IR-guns are replaced with new devices annually or are verified annually against a National Institute of Standards and Technology (NIST) standard according to SOP# DAQ-13-002.1 (DryWell 3101 Temperature Generator, Electronics and Calibration Branch Responsibilities). Lab refrigerator temperatures are measured with an IR-gun and recorded in lab refrigerator logbooks. Every effort is made to check and record refrigerator temperatures daily, except on holidays and weekends.

3.4.1 Pipette Verification Checks

An accurate pipette is one of the most important tools in performing accurate dilutions of calibration and QA/QC standard mixes and preparing mobile phases. To maintain a valid work product, the pipettes must be verified quarterly. This section describes steps for quarterly pipette verifications.

Equipment and Materials needed for Pipette verifications:

- Mechanical Pipettes
- Pipette Tips
- Analytical Balance
- Weighing Container
- Optima/HPLC-grade water
- Beaker
- NIST-Traceable barometer that reads temperature and relative humidity

Detailed Procedures

1. In advance of testing, make sure the balance verification is current. If the balance verification date has expired perform a balance verification prior to the pipette verification.
2. Fill a beaker partially with Optima/HPLC grade water and turn on the balance to warm it up.
3. Place pipette, pipette tips and a beaker of water in the extraction lab next to balance for about 2 hours before starting the measurements so they can reach equilibrium with room temperature.
4. Place small containers with water inside the balance draft chamber, but not in weigh pan chamber, to add humidity to the chamber to reduce water evaporation during testing.
5. If the balance printer is not being utilized, open a blank pipette verification worksheet (DAQ-03-014) located on internal network drives here: **P:/Toxics/Urban Air toxics/Aldehyde data/Balance and Pipette Checks/DAQ-03-014 Pipette Verification worksheet_TEMPLATE**. Also see **Appendix A** in this document for an example of the pipette verification worksheet. Print the worksheet to record measurements. If a balance printer is being used, a blank copy of the verification form is not needed to record measurements. Instead, the balance printout readings can be transferred by hand to a blank electronic copy of the verification form at the completion of the test.
6. Reference the current room temperature and barometric pressure from the NIST-traceable barometer and follow this link to look up the proper z-factor to use for this pipette verification: https://www.integra-biosciences.com/sites/default/files/Z-factors-calibration_2017_01_26.pdf
7. Load the appropriate pipette tip, adjust the pipette to the highest setting of the operating range of the pipette.
8. Place the weighing container on the analytical balance weigh pan. The weighing container should not be dry, add some Optima/HPLC-grade water to the weigh container.
9. Pre-wet the tip by aspirating and dispensing the nominal volume three times.
10. Aspirate and draw the nominal amount of Optima/HPLC-grade water from the beaker.
11. Tare the balance.
12. Dispense the aliquot in the weigh container. Be sure to dispense along the inner wall of the weigh container and finish by withdrawing the tip end along wall of weigh container to remove residual liquid.

13. Allow balance reading to stabilize, record the mass on the appropriate place in the blank copy of the pipette verification worksheet or use the balance printer to record the measurement.
14. Using the same tip, repeat steps 10 to 13 five consecutive times.
15. Eject the tip and load a new tip. Adjust the pipette to the lowest setting of the operating range. Pre-wet the tip by aspirating and dispensing the nominal volume three times. Perform the steps 10 to 13 five more consecutive times.
16. Transfer the handwritten measurements or the balance printout readings into an electronic copy of the pipette verification worksheet. Make sure all gray fields of the electronic copy of the pipette verification worksheet are filled out. Save the worksheet as "**DAQ-03-014 Pipette Verification_Serial Number_MMDDYY** and store it here: **P:\Toxics\Urban Air Toxics\Aldehyde Data\Balance and Pipette Checks\Pipette Verifications\ Pipette SN_XXXX** on p drive.
17. Print a copy of the electronic pipette verification form and staple the handwritten verification form and/or the balance printout to the printed electronic copy and store them in the designated file cabinet in aldehyde lab.
18. Once measurements are entered, the electronic copy of the verification worksheet will compare the measured accuracy and precision against the limits listed in **Table 1** below and provide a pass/fail status:

Pipette Volume, µL		**Relative Error**	
Nominal	**Setting**	**Accuracy (%) ≤**	**CV or Precision (%) ≤**
10-100	100	2.0	1.0
	10	8.0	3.0
100-1000	1000	2.0	1.0
	100	8.0	3.0

Table 1: Pipette Verification Acceptance Criteria

19. Record the date and status (pass or fail) of the pipette verification in the current extraction logbook (Example: Logbook # EXT-01).
20. If the pipette status passes, then label the pipette with a certification sticker including the certification due date which is 3 months from the current verification date. If it fails repeat the pipette verification one more time following steps 1-19 above for confirmation of the failing status. If failing status is confirmed remove pipette from service. Send pipette for calibration/repair or replace with new pipette. Verify new or repaired pipette prior to use.

3.4.2 Balance Checks

An accurate analytical balance is a crucial and fundamental piece of equipment for a laboratory for precision weighing. To maintain the quality work, the balance must be verified with certified mass(s) quarterly. This section describes steps for balance checks.

Equipment and Materials needed for balance verification.

- Analytical Balance
- Certified mass set
- NIST-Traceable barometer that reads temperature and relative humidity

Detailed Procedures

1. The analytical balance must be verified with certified mass standards. Verify the check masses are not past due for calibration.
2. If a balance printer is not being utilized, open a blank balance verification worksheet (DAQ-03-015) located on internal network drives here: **P:/Toxics/Urban Air toxics/Aldehyde data/Balance and Pipette Checks/DAQ-03-015 Balance Verification Worksheet_TEMPLATE**. Also see **Appendix B** in this document for an example of the balance verification worksheet. Print out the blank worksheet to record measurements. If the balance printer is being used, a blank copy of the worksheet is not needed to record measurements. Instead, the balance printout readings can be transferred by hand to a blank electronic copy of the worksheet at the end of the test.
3. Verify the balance is level, if leveling adjustment is needed, manipulate the leveling feet so that the air bubble is centered within the circle of the balance level indicator.
4. Turn on the balance.
5. Press the tare button if needed to zero the balance readings before placing test mass on balance weigh pan.
6. Open balance draft door and place the first test mass in the center of the balance weigh pan.
7. Close the draft door. Allow several seconds for a stable mass reading to be displayed.
8. Record the balance reading on the blank worksheet or record the balance reading using the balance printer.
9. Repeat steps 5-8 for each nominal test mass (5 milligrams [mg], 200mg, 500mg, and 2 grams [g])
10. Open a blank balance verification worksheet and save the worksheet as: "**DAQ-03-015 Balance Verification_Serial Number_MMDDYY** and store it here: **P:\Toxics\Urban Air Toxics\Aldehyde Data\Balance and Pipette Checks\Balance Verifications.**
11. Enter the handwritten measurements and/or balance printout readings in the recently created worksheet. Make sure all gray colored cells are filled out (serial numbers of equipment, room environmental conditions, etc.). The electronic version of the worksheet will calculate a %Diff and provide a pass/fail status.
12. Save the worksheet then print a hard copy. Staple the handwritten copy and/or the balance printout to the printed hard copy. Store all hard copy records in the designated file cabinet in the aldehyde lab. The electronic file will be stored using the naming convention and storage location listed in step #10 above.
13. Record the balance verification date and pass/fail status in the current balance logbook.
14. If balance passes, label the balance with a certification sticker including the next verification due date (which is 3 months from the current balance verification test date).
15. If the balance fails, repeat the balance verification check procedure again with a new form to confirm the failing status.
16. If failing status is confirmed, do not use the balance until it is calibrated by the balance manufacturer or designated balance calibration service provider.
17. If the balance passes the confirmation test, label the balance with a certification sticker including the next verification due date (which is 3 months from the current balance verification test date).

4.0 SITE CHECKS

This section is reserved. Site checks do not apply to analysis instruments housed at the Reedy Creek Laboratory (RCL).

5.0 DETAILED PROCEDURES

This section describes steps for carbonyl cartridge shipping, receiving, extraction, sample analysis, instrument data analysis, and data package processing.

5.1 Shipping Carbonyl Cartridge Samples

1. Reference the carbonyl sampling schedule located on internal network drive **P:\Toxics\Urban Air Toxics\Sampling Calendars\Current year Sampling Calendar** for Urban Air Toxics (UAT) and Photochemical Assessment Monitoring Station (PAMS) sampling to determine when to ship the carbonyl samples and what type of carbonyl samples to ship.
2. Obtain an un-sampled DNPH cartridge from a lot of cartridges that has passed the lot blank test from RCL Fridge 1 for all samples needed to be shipped and/or assigned.
3. Obtain one blank carbonyl cartridge SIF (DAQ-16-009) for each cartridge being shipped and/or assigned. See **Appendix C** of this document for an example of the carbonyl cartridge SIF.
4. Obtain an insulated shipper or cooler, frozen icepacks, and Ziploc bags.
5. Using a black or blue ink pen, circle the sample program, sample type and/or QC sample type (if applicable); record the sampling site; record the DNPH cartridge lot number (#); sample #; sample channel # and port number [ATEC 8000 only for port number]; and scheduled sample date in the **Laboratory Setup information** section on the carbonyl cartridge SIF. The scheduled sampling date is derived from the sample #. For example, sample # C020321C indicates a scheduled sample date 2/3/2021 at the Candor site.
6. **Sample naming examples for UAT carbonyls include**: C020321M for UAT Millbrook sample date 2/3/2021, C020321C for UAT Candor sample date 2/3/2021, C020321MD for UAT Millbrook Duplicate sample date 2/3/2021, and C020321MFb for UAT Millbrook field blank sample date 2/3/2021.
7. **Sample naming examples for PAMS include**: PC020321M1 for PAMS Millbrook sample date 2/3/2021 collected from 04:00 - 12:00, PC020321M2 for PAMS Millbrook sample date 2/3/2021 collected from 12:00 – 20:00, PC020321M3 for PAMS Millbrook sample date 2/3/2021 collected from 20:00 – 04:00(following day), PC020321MFb for PAMS Millbrook field blank sample date 2/3/2021 on channel # specified on the SIF.
8. Please see **Table 2** for PAMS and UAT carbonyl sample collection times.

PAMS Sample #	Collection Start Time (local standard time)	Collection End Time (local standard time)
PC020321M1	04:00	12:00
PC020321M2	12:00	20:00
PC020321M3	20:00	04:00 (the following day)

UAT Sample #	Collection Start Time (local standard time)	Collection End Time (local standard time)
C020321M	00:00 (midnight)	23:59 or 00:00 (midnight)

Table 2: PAMS and UAT Carbonyl Sampling Times

9. Field blank (FB) samples must be scheduled once per month for UAT and twice per month for PAMS. The UAT field blank samples will rotate between the two UAT carbonyl sites (Millbrook and Candor). For example, if the UAT FB is scheduled for Millbrook in January, the next UAT FB sample should be scheduled for Candor in February.
10. The UAT and PAMS field blank samples will be assigned a specific sampler channel number and port number (typically channel #1 port 1, 2, or 3 for PAMS [ATEC 8000] and channel #1 for UAT [ATEC 2200]).
11. Label the carbonyl cartridge foil pouch with the sample # listed on the carbonyl cartridge SIF and place the labeled cartridge in a Ziploc bag to protect it from moisture generated by melting ice packs.
12. Tear off the gold bottom copy of the carbonyl cartridge SIF, set aside the gold copy and place the remaining carbonyl cartridge SIF in a document sized Ziploc bag to protect it from moisture generated by melting ice packs. Prepare more samples if needed, repeat steps 1-12 above for each cartridge being prepared.
13. Obtain a blank carbonyl cartridge COC form for UAT and PAMS Required sites. See form example in **Appendix D.** Fill out the COC form fields, making sure to include all cartridges assigned a site for the upcoming sample event.
14. Review the carbonyl sample SIF and labeled foil pouches against each other and against the information in the carbonyl cartridge COC form to be sure all recorded information is accurate. If no edits are required, sign and date the first **relinquished by** and **date/time** field on the carbonyl cartridge COC form.
15. Place the labeled foil pouch in the insulated shipper, making every effort to sandwich the foil pouch between the ice packs. Place the filled-out and Ziploc bagged carbonyl sample SIF inside the insulated shipper, on top of the ice packs.
16. Place the filled-out carbonyl cartridge COC form in a document sized Ziploc bag to protect it from moisture generated by melting ice packs. Place the bagged form inside the insulated shipper on top of the ice packs.
17. Gather the gold copies of the carbonyl sample SIF and update the carbonyl sample tracking chart excel file. See **Appendix E** for UAT Carbonyl sample tracking chart and **Appendix F** for PAMS Carbonyl sample tracking chart. The tracking chart file is located on internal network drives here: **P:/Toxics/Urban Air Toxics/VOC Current Year Data/Tracking Chart UAT VOC & Aldehyde**. Enter the shipping date for the samples that were prepared and color the cells using the color code on the tracking chart file.
18. After the tracking chart has been updated, place the gold copies of the carbonyl sample SIF forms in a properly labeled file folder in the designated file cabinet in the carbonyl laboratory. These forms can be used as reference to confirm sample preparation and shipment to the field.

5.2 Receiving Carbonyl Cartridge Samples

Carbonyl cartridges arrive at the laboratory via hand delivery by the site operator or delivery by United Parcel Service (UPS) or designated commercial shipping carrier.

1. If the sampled cartridges are hand delivered by the sampling site operator after business hours, the site operator must store the cooler and samples in a secure, pre-determined storage location at the RCL. This cooler will be received by the UHPLC operator on the following business day.
2. Check the secure storage location for sampled cartridge coolers hand delivered by the site operator. Immediately or as soon as possible open the cooler.
3. Using a certified IR temperature gun, take the temperature of the carbonyl cartridge foil pouch (the operator may have to adjust ice packs to get a clear line of sight to the cartridge foil pouch).
4. Record this temperature on the carbonyl cartridge SIFs for all sampled cartridges present in the insulated shipper. Record the serial number of the IR gun used to measure the temperature.
5. Sampled cartridges may also be delivered to the lab by UPS or a commercial shipping carrier. This typically occurs during normal business hours. Immediately or as soon as possible open the cooler.
6. Using a certified IR temperature gun, take the temperature of the carbonyl cartridge foil pouch (the operator may have to adjust ice packs to get a clear line of sight to the cartridge foil pouch).
7. Record this temperature on the carbonyl cartridge SIFs for all sampled cartridges present in the insulated shipper. Record the serial number of the IR gun used to measure the temperature.
8. Pull out the sampled cartridges, carbonyl cartridge SIF(s), and the carbonyl cartridge COC form. Verify the information on the SIF and COC match the information written on the cartridge foil pouches and match each other.
9. Record the signature and date/time in the second **received by** and **date/time** fields on the carbonyl cartridge COC form.
10. Verify the information recorded on the carbonyl cartridge SIF.
11. Contact the site operator with any questions, or if there are illegible entries and/or blank fields on the carbonyl cartridge SIF or carbonyl cartridge COC.
12. Verify the volume and pre- and post-flows written on the carbonyl cartridge SIF form are within the acceptable ranges. Sample volume should be within 1240L to 1650L for UAT samples and 414L to 550L for PAMS samples. Sample flow should be 0.9 to 1.1 liters per minute (L/min) for both UAT and PAMS samples.
13. If the volume or flow are not within acceptable ranges, place a note in the comments section on the carbonyl cartridge SIF, if a comment was not already recorded by the site operator.
14. Using a stapler, attached the sampled carbonyl cartridge foil pouch to the corresponding carbonyl sample SIF.
15. Fill out the sample received by; date and time sample received; and store in the aldehyde sample refrigerator (RCL Fridge 2) located in the aldehyde laboratory.

***Note**: sampled cartridges must be stored in a different refrigerator from unused cartridges.*

5.3 Extracting Carbonyl Cartridge Samples

***Note**: The extraction process must be completed inside a fume hood located in the extraction lab room at the RCL. Proper personal protective equipment (PPE) including safety glasses must be worn during the extraction procedure. Each extraction batch contains 20 or fewer sample cartridges and must contain at* *least one cartridge blank (method blank), one cartridge spike (laboratory control sample), and with resources permitting one cartridge spike dup (laboratory control sample duplicated) and one cartridge method detection limit (MDL) spike.*

Sometimes a sampler needs to be certified as clean when no sampled cartridges are waiting for extraction. In these cases, if three or less sampler certification cartridges are being extracted, at a minimum extract one cartridge blank (method blank) and one cartridge spike (laboratory control sample) with the sampler certification cartridges.

1. Retrieve 20 or less sampled cartridges or sampler certification cartridges and the attached SIF(s) from the aldehyde sample refrigerator (RCL Fridge 2) located in the aldehyde laboratory. Set aside.
2. Retrieve enough additional un-sampled cartridges from the VOC laboratory refrigerator (RCL Fridge 1) and enough blank SIF(s). These cartridges will be assigned a QC Sample Type such as cartridge blank, MDL spike and cartridge spike. Include a Cartridge SIF for each QC cartridge being prepared.
3. Fill out the aldehyde extractions manifold worksheet shown in **Appendix G** with the samples to be extracted.
4. Record the analyst initials, extraction date, cartridge sample dates, and extraction material lot numbers on the extraction manifold worksheet.
5. Record the extraction date and manifold port number on the carbonyl cartridge SIF(s).
6. Cap the 4mL storage vials and label them using a permanent marker with the sample # for each cartridge being extracted, including QC samples.
7. Cap the 1.5 mL auto-sampler vials and label them with the sample name for each cartridge being extracted, including QC samples.
8. Clean the solid phase extraction (SPE) manifold using the carbonyl-free acetonitrile by opening each port valve and rinsing acetonitrile through the manifolds' flow path, including the outside surfaces of the needle guides. Close the valve of each port. Set aside in the fume hood.
9. Rinse 5mL volumetric flasks using carbonyl-free acetonitrile. Wash each 5mL volumetric flask at least 3 times and place each cleaned flask on the rack. Place the rack containing the washed flasks inside the vacuum chamber of the SPE manifold.
10. Place the cleaned manifold on top of the vacuum chamber. Ensure the needle guides for each SPE site are inserted into the 5mL volumetric flasks. If any needle guides are not lined up, the extract will be lost. Make sure the screw caps of the manifold are closed tightly.
11. Referencing the extraction manifold map show in **Appendix H**, place cartridge spike samples and/or MDL spike samples on the assigned port using a slight twist to lock the Luer tip of the cartridge to the manifold.
12. Open the valve slightly, about a half turn. Turn on the pump to apply a vacuum on the cartridges to be spiked.
13. Spike the amount of standard as per the **DAQ-03-017 Carbonyl Calibration and QC Standard Dilutions Checklist** located on internal network drive **P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Standard Dilution Chart** for making calibration and QA check standards using the appropriate pipette and pipette tip.
14. Release the vacuum and close the manifold valves.
15. Referencing the extraction manifold worksheet **Appendix G** and the manifold map **Appendix H** Install the remaining blank and sample cartridges on the vacuum manifold.

16. Remove the plunger from the 5mL syringe and set aside. Place the 5mL syringe into the cartridge inlet and use a slight twist to lock the syringe onto the cartridge inlet.
17. Dispense approximately 4.5 mL of acetonitrile into each 5mL syringe. Carefully loosen the screw caps of the manifold to allow acetonitrile to flow out of the syringe and through the cartridge. The acetonitrile will continue down through the needle guides and into corresponding 5mL volumetric flask.
18. Apply a slight vacuum to the extraction manifold using the attached vacuum pump. Do not exceed -10 millimeters of mercury (mm Hg). Applying a vacuum to the system will assist in forcing the acetonitrile through the cartridge.
19. Once the acetonitrile has been collected in the 5mL volumetric flasks, turn off the pump and release the vacuum on the system.
20. Carefully remove the rack containing the 5mL volumetric flasks from the vacuum chamber, be sure to not spill any extracts. If a spill occurs the sample must be invalidated as a lab error and the invalidation noted on the SIF form.
21. Using the same syringe during the extraction, attach a syringe filter to the syringe tip using a slight twist motion to lock the filter to the syringe tip. Carefully place the syringe and filter into the correct 4mL storage vial that has been previously labeled with the sample # and extraction date.
22. Using disposable pipettes, bring the volume of the 5mL volumetric flask to the mark with carbonyl free acetonitrile.
23. Mix the extract in the 5mL volumetric flask several times using the same disposable pipette.
24. Transfer the extract into the 5mL syringe with attached filter.
25. Carefully replace the plunger of the syringe and force approximately 2mL of extract into the 4mL storage vial.
26. Stop the plunger, and force approximately 1mL of extract into the 1.5 mL auto-sampler vial.
27. Finally, force the remaining extract into the 4mL storage vial.
28. Cap each vial and set aside.
29. Store extracts in the sample refrigerator of the aldehyde laboratory (RCL Refrigerator 2). Extracts must be analyzed within 30 days of the extraction date.

5.4 Carbonyl Sample Analysis

1. Prepare mobile phases to be used during aldehyde analysis. Mobile phase A is a 2 millimolar (mM) ammonium acetate solution. This mobile phase needs to be made fresh before each run to minimize bacteria growth. If needed, mobile phase A can be filtered in a vacuum filtration assembly. This solution should be removed from the system after completion of the analytical run and the mobile phase lines should be flushed with a solvent/pure water mixture (80:20 water/methanol).
2. Obtain a 1L volumetric flask. Rinse the flask several times, in order, using Optima/HPLC-grade water, then methanol and then finally carbonyl-free acetonitrile.
3. Partially fill the flask with Optima/HPLC-grade water.
4. Weigh out 144 ± 8mg of ammonium acetate salt. Transfer the ammonium acetate salt into the 1L volumetric flask.
5. Pipette 57 ± 3 µL of glacial acetic acid and dispense into the 1L volumetric flask containing the ammonium acetate salt. Dilute to the 1L mark with Optima/HPLC- grade water, mix the solution by inverting the 1L volumetric flask several times.
6. Transfer the 2mM ammonium acetate buffer solution into a clean, empty UHPLC reservoir and sonicate/degas for at least 30 minutes (also mobile phase A can be degassed during the filtration processes).

7. Connect this mobile phase reservoir to channel A of the UHPLC system solvent rack. This solution can also be made directly in a clean, measured 1L UHPLC reservoir.
8. Mobile Phase B is 100% carbonyl-free grade acetonitrile. The solvent is submicron filtered by the manufacturer. Do not filter the acetonitrile as filtering may introduce contamination.
9. Dispense carbonyl free grade acetonitrile into a clean, empty UHPLC reservoir and attached this reservoir to Channel B of the UHPLC system solvent rack.
10. Do not add fresh solvent to reservoirs A and B which contain existing solvent. Completely empty the reservoirs of solvents before dispensing fresh solvents in the reservoirs. Best lab practice is to rinse the reservoir three times with the corresponding solution prior to fill.
11. Mobile Phase C is approximately an 80:20-mixture of Optima/HPLC- grade water and Optima-grade methanol, respectively. This mobile phase is used to flush the system of built-up ammonium acetate salts after analysis is complete. Rinse the C reservoir with Optima/HPLC-grade water.
12. Fill the reservoir with approximately 800 mL of Optima/HPLC-grade water, then add Optima-grade methanol until there is 1000 mL of liquid in the reservoir. This solution should be sonicated/degassed to remove dissolved gas bubbles.
13. Attach this reservoir to Channel C of the UHPLC system solvent rack. For this reservoir, the analyst can add fresh mobile phase to older mobile phase since this solvent is used for flushing purposes only.
14. Power up the UHPLC-ISQEC system modules and the system computer. Open the Chromeleon software and ensure each module is detected by the software and can be controlled using the computer.
15. In Chromeleon, make sure all the system modules are connected. Open the commands dialog box for the pump to purge the mobile phases A, B and C. Set the channel to be purged to 100% of each line.
16. Open the purge valve on the purge unit of the pump (rotate the knob counterclockwise). Start the purge cycle according to the selected settings. To do so, set **Purge** to **on** and follow the prompt. When specified purge time has elapsed, purge is automatically reset to off. **Purge** can be stopped manually, if needed, by setting purge to **off**.
17. Repeat the above steps for all channels until the air bubbles are gone. For the last few minutes, flush with the analytical starting conditions of 52%A/48%B/0%C. After completion close the purge valve.
18. Change the pump flow to 0.4 mL/min. Allow 30-60 minutes of equilibration time. Then record the system backpressure in the logbook.
19. In Chromeleon, open the commands dialog box for the auto-sampler. Under sampler, click on prime syringe and wash buffer loop to help remove any air bubbles or debris trapped in the auto-sampler syringe.
20. In Chromeleon, open the commands dialog box for the column oven. Make sure column temperature control is on. Enter the appropriate switching valve on the left side of the instrument; Valve 1 is for only UV analysis and Valve 2 is for UV and MS analysis.
21. In Chromeleon, open the commands dialog box for the detector. Set VIS lamp on to turn on the tungsten lamp. Then set UV lamp on to turn on the deuterium lamp.
22. In Chromeleon, open the commands dialog box for the ISQ-EC. Start real time scanner using the parameters listed in **Appendix I** to full scan the mobile phase. If scanning indicates the presence of mobile phase, then stop the real time scanner. If contamination is indicated, flush the system for a longer amount of time to purge contaminants or investigate the source (e.g., contaminated acetonitrile solvent).

23. Set the data acquisition to **on** in the detector dialog box and monitor the detector baseline. Allow the system to equilibrate for at least 60 minutes prior to start of analysis. System can also be equilibrated using the smart startup functions of the instrument control software.
24. While the system is equilibrating, prepare the calibration and QC standards as per the "DAQ-03-017 Carbonyl Calibration and QC Standard Dilutions Checklist" located on internal network drive **P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Standard Dilution Chart,** shown in **Appendix J**, using the appropriate pipette and pipette tip. The system can be calibrated before each analytical run at analyst discretion. The calibration standards need to be prepared fresh prior to executing a calibration on any analytical run. When using a previous calibration curve, all check standards and blanks must pass the prescribed criteria in **Table 3**.
25. Prepare the sample sequence by entering each standard and sample to be analyzed. Current run sequence can be made by copying an earlier run or click on **file** and **create sequence**. When copying the earlier run, in Chromeleon, click on **data** select one of the earlier runs, **file**, **save as** the sequence in the current run folder (by year) using this format: MMDDYY_Aldehyde_Run_UV (e.g., ***021721_Aldehyde Run_UV***).
26. Edit all standards and samples to be analyzed. Verify the sample name entered in the sequence match the sample location on the auto-sampler vial tray. An example sequence is shown in **Appendix K** for UV analysis and **Appendix L** for MS analysis. The sample sequence must contain the current instrument method and current data processing method used for analysis. Make sure to **Save** the current sequence.
27. Pull out the sample extracts from the aldehyde sample refrigerator (RCL Fridge 2) to be analyzed. Place the auto-sampler vials in the correct auto-sampler tray locations based on the sequence created.
28. With each sequence, a duplicate injection must be completed, and a reanalysis of a sample from the last run must be analyzed. The reanalysis injection is indicated by this format: C020321CR (for a repeat analysis from a previous run).
29. The duplicate injection is indicated by this format: C020321CS (duplicate injection analysis of a sample from same sequence).
30. After system equilibration is complete and the sequence has been saved, turn off the data acquisition. Then open the **Queue** tab on the UHPLC system control software. Click **Add** and select the sequence to be analyzed. Click **ready check** and the system will diagnose the sequence for any errors. If errors are found, correct, and redo this step. Click on the smart shutdown tab to automatically shut down the system after finishing the sequence.
31. It is very important to verify the auto-sampler vial location against the list in the sample sequence; the software will not find errors associated with vials in the wrong locations. If no errors are found, press **Start** to execute the sequence.
32. If the calibration curve passes criteria and the sequence finished, additional samples may be analyzed under the same calibration curve. The additional samples must be bracketed by solvent blanks and QC checks.
33. If a target compound concentration exceeds the calibration curve range, the sample extract can be diluted and re-analyzed, and diluted results are reported only for compounds exceeding calibration range.

5.5 Carbonyl Data Analysis

Once the run is complete, data analysis can begin. MS raw data must be saved before processing the UV data.

5.5.1 MS Detector Data Processing

NOTE: *MS data results are not reported to AQS. The MS results are being used for informational purposes only.*

1. In Chromeleon Open **data**, select current sequence (e.g., 021721_Aldehyde Run_UV).
2. Click file, save as with raw data for MS (e.g., 021721_Aldehyde Run_MS).
3. Open the MS run sequence, update the current processing method, change the calibration levels from level 0 to level 3 (conc.0.003 ug/ml to 1.00 ug/ml) and save.
4. Double click the chromatogram of Calibration level 2, Click on MS Quantitation channel, go to processing tab, adjust the retention time of all the compounds, if needed, in the component table.
5. In the data processing tab, check all the chromatograms of the sequence and make sure all the compounds are identified correctly. **Data Processing →MS Component → Smart link.** After making those adjustments **save** the processing method.
6. Print out the MS data, calibration, amount summary and sequence. For example, **Report Designer →Integration Tab → File → Print → Current Sequence → Unchecked all but Integration →OK.** Before printing out make sure the current report template has been selected and the current run folder has been created in the instrument computer desktop. Export data files under the current year MS data.
7. Once all chromatograms of the sequence have been checked by the analyst, data needs to be exported into the network drive.
8. First export integration of all injections of the current analysis. **Report Designer → File → Export → Current Sequence → Parent Folder Desktop → Data Export →Year → Select MS Data →Current Run folder → OK →check PDF and /or Excel → Setting (Tool Symbol) → check only Integration → File name Formula (Current Run Date_ Aldehyde Run_MS_ Integration) → Ok.**
9. Similarly, export the Calibration, Sequence, Amount Summary for current injection under the current run folder. **Report Designer → Required Tab (Ex. Calibration) → File → Export → Current Injection → Parent Folder Desktop → Data Export → Year → Select MS Data → Current Run Folder → OK →Uncheck PDF and Check Excel → Setting (Tool Symbol) → check only Calibration → File name Formula (Current Run Date_ Aldehyde Run_MS_ Calibration) → OK**
10. The data need to be copied into the network **P drive** under current year MS Aldehyde data via an external hard drive.
11. After data have been saved in the **P drive**, open Amount Summary of the current MS run folder and convert all n.a. to 0.000 and then save it. Then Complete the current year carbonyl master spreadsheet with all data copied from amount summary report of MS data into the Carbonyls-MS ug_mL tab.

5.5.2 UV Detector Data Processing

1. In the UV run sequence, double click the chromatogram of Calibration level 3 or 4. Click on **processing method** and select acetaldehyde peak. In the Peak Group table, compare the peak group start and end time with the current acetaldehyde peak group start and end time and adjust if needed.

2. Then adjust the retention time of all the compounds in the component table. After making those adjustments **save** the processing method. **Export** the raw data and print out all raw data chromatograms prior to making any manual adjustments to individual chromatograms. Make sure to use current report template and current run folder has been created in the instrument computer desktop. **Export** data file under current year UV data. **Report Designer→ Integration Tab → File → Print → Current Sequence → Unchecked all but Integration → OK**
3. The integration software tends to find many unknown peaks (especially in summer) and tends to draw baselines in the wrong areas. It is common to manually adjust the integration software for each injection in a run. Most samples contain unknown peaks that elute near the known reference standards. Adjusting baselines and splitting peaks is a common data analysis process for each sample analyzed. **Appendix M** shows an unedited chromatogram and **Appendix N** shows the same chromatogram manually adjusted.
4. Make sure the chromatogram is saved after any manual adjustments are made. To remove manual adjustments, click the **processing tab** and click **remove manual adjustments**. This will return the chromatogram to its raw format.
5. Once all chromatograms of the sequence have been checked and/or manually adjusted by the analyst, print out the integration of all the chromatograms. Also print out the calibration curve, Sequence (Overview) and amount summary of the current run folder. **Report Designer → Required Tab (Ex. Calibration) → File → Print → Current Injection→ Unchecked all but Calibration →OK.**
6. Current data needs to be exported into the network drive after each run to complete the QA summary report. First export integration of all injections of the current analysis onto a universal serial bus (USB) storage device. **Report Designer → Integration tab →File → Export → Current Sequence → Parent Folder Desktop → Data Export →Year → Select UV Data → Current Run Folder → OK →check PDF and/or Excel → Setting (Tool Symbol) → check only Integration → File name Formula (Current Run Date_ Aldehyde Run_UV_ Integration) → OK**
7. Similarly, export Calibration, Sequence, and Amount Summary for current injection under the current run folder onto a USB storage device. **Report Designer → Required Tab (Ex. Calibration) → File → Export → Current Injection → Parent Folder Desktop → Data Export → Year → Select UV Data → Current Run Folder → OK →Uncheck PDF and Check Excel → Setting (Tool Symbol) → check only Calibration → File name Formula (Current Run Date_ Aldehyde Run _UV_Calibration) → OK**
8. These data files need to be stored on internal network drives for further review. Transfer the exported instrument files to the proper location on internal network servers under current year's UV Aldehyde data via an external USB hard drive.
9. Open **a blank QA summary report (DAQ-03-016)**. Save this file with new name. See **Section 7.7** for details on naming and storage conventions.
10. Record the current run folder name in the QA Summary report
11. Open the Amount Summary of the current run folder and record the current run folder name.
12. Convert all **n.a**. to **0.000** and save it. Fill up the QA summary report for each criterion by copy and paste from the amount summary.
13. Add sampler information form data and instrument data to the current year's **Carbonyl Master Spreadsheet (DAQ-03-012)** by copy and paste from the Amount Summary Reports or hand entered from the SIFs. See **Section 7.5** for details on naming and storage conventions.
14. Add data to the QA summary report from the **Carbonyl Master Spreadsheet** where applicable.
15. Print out the completed QA summary report and add the printed copy in the aldehyde run folder.
16. Complete the Level 1 data review checklist (see Section 6.1)

17. Once the level 1 data review is completed, notify the level 2 reviewer via email or by verbal contact. The hard copy and electronic copy of the data packet must include paper copies of all the documents mentioned in the **UAT and PAMS Carbonyl Sampling and Analysis Data Level 1/Level 2 Review Checklist**.

5.5.3 UHPLC Data Validation Parameters and Flags

See **Table 3** below for a list of UHPLC data validation parameters, acceptance criteria, and applicable AQS qualifier and null codes. Solvent blanks are analyzed at the beginning of the sequence, after the initial calibration curve, and with each check standard injection.

QA Summary Report Flag	Where the flag comes from?	Acceptance Criteria	AQS Code
1a	Solvent Blank	All compounds ≤ MDL_{spk} to pass.	**QA Qualifier: LB**
	*Extraction Solvent Method Blank	All compounds ≤ MDL_{spk} to pass.	**QA Qualifier: LB**
2a	Cartridge Blank analysis results	All compounds ≤ MDL to pass.	**QA Qualifier: LB**
2b	Field Blank analysis results	Formaldehyde ≤ 0.06 µg/mL Acetone ≤ 0.15 ug/mL; Acetaldehyde ≤ 0.08 µg/mL; Sum of All other ≤ 1.4 µg/mL.	**QA Qualifier: FB**
	**Trip Blank analysis results	Formaldehyde ≤ 0.03 µg/mL; Acetone ≤ 0.06 µg/mL; and all others ≤ 0.02 µg/mL to pass.	
	Lot Blank Analysis	Formaldehyde ≤ 0.03 µg/mL; Acetaldehyde ≤ 0.02 µg/mL; Acetone ≤ 0.06 µg/mL; and all others ≤ 0.02 µg/mL to pass.	**QA Qualifier: LB**
3a	Calibration Curve	Calibration Type: Linear with offset. If the correlation coefficient is less than <0.999	**Null: AT**
3b	Calibration Curve	Relative percent difference (RPD) for each calibration level against the nominal concentration must be ≤20%.	**Null: AT**
3c	Calibration Curve	The absolute value of calibration curve (offset/slope) must be ≤ current MDL_{spk}.	**Null: AT**
4	HL-QA analysis results	The RPD of each compound in the HL-QA against the nominal concentration must be ≤ 15%.	**Null: AS**
5	Replicate analysis (duplicate injection results)	The RPD for each compound detected ≥ 0.1ug/mL must be ≤ 10%. Flag compounds 5 and qualify results QX before reporting to AQS.	**QA Qualifier: 4**
6a	Sampled Cartridge hold time	The (extraction date – sample date) must be ≤ 14 days.	**QA Qualifier: HT**
6b	Sample Extract hold time	The (analysis date – extraction date) must be ≤ 30 days.	**QA Qualifier: HT**

QA Summary Report Flag	Where the flag comes from?	Acceptance Criteria	AQS Code
QX	Cartridge Spike Recovery and Precision	Cartridge spike recovery RPD against nominal ± 20% (formaldehyde); ± 30% (all other compounds). Cartridge spike/cartridge spike duplicate RPD ≤ 20%.	**QA Qualifier: QX**
	Sample receipt temperature	The sampled cartridge receipt temperature must be ≤ 4°C.	**QA Qualifier: TT**
	Collocated/Duplicate sample collection analysis results	The RPD for each compound detected ≥ 0.1ug/mL must be ≤ 20%.	**QA Qualifier: 3**
	DNPH peak area analysis	The DNPH peak area found in samples must be ≥ 50% of the average DNPH peak area from lab blanks and cartridge spikes.	**QA Qualifier: DN**

Table 3: UHPLC Data Validation Parameters and Acceptance Criteria

*Extraction Solvent Method Blanks are not performed regularly. Instead, this QC sample is used as part of corrective action investigations to find sources of contamination in the event of systemic cartridge blank failures of priority compounds (Formaldehyde and Acetaldehyde).

**Trip blanks are not performed regularly. Instead, this QC sample is used as part of the corrective action investigations to find sources of contamination in the event of the systemic field blank failures of priority compounds (Formaldehyde and Acetaldehyde).

6.0 CARBONYL DATA REVIEW

This section of the SOP describes steps required for the primary lab staff to perform a self/Level 1 data review and a trained lab staff not directly involved in the carbonyl collection or analysis method to perform a peer/Level 2 data review on the carbonyl sampling and analysis data.

6.1 Level 1 Carbonyl Sampling and Analysis Data Review

1. After the primary analyst finishes preparing the analytical batch, the primary analyst performs a self/Level 1 review of the analytical batch.
2. Open a blank data review checklist located on internal network drives here: **P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Data Review**. Print out this form to record answers and provide comments where applicable.
3. Save this file in the current run folder on the **P-drive** that is currently under review.
4. Answer the Level 1 reviewer questions in document number DAQ-03-013 (UAT and PAMS Carbonyl Sampling and Analysis Data Level 1/Level 2 Review Checklist).
5. For all questions answered **no** provide a comment with sample identification or failing compounds where appropriate, and/or provide a general comment explaining the situation in concise detail.

6.2 Peer/Level 2 Carbonyl Sampling and Analysis Data Review

The Level 2 reviewer should receive notification from the primary analysist or Level 1 reviewer to start the Level 2 data review process.

1. Open the review checklist already saved by the primary analysist. The review checklist is in the current run folder under review on the p-drive.
2. Answer the questions to verify the entries made by the primary analyst are correct and agree with QA requirements and to verify imbedded calculations are correct and referencing the correct cells.
3. For all questions answered **no** provide a comment with sample identification or failing compounds where appropriate, and/or provide a general comment explaining the situation in concise detail.
4. If error/typos are found during the Level 2 review, coordinate corrections with the primary analyst before approving the results for Level 3 validation.

7.0 CARBONYL FILE MANAGEMENT

7.1 UHPLC Instrument Files

UHPLC instrument files such as chromatograms and results, calibrations, amount summaries, and sequences are stored on the UHPLC instrument computer in appropriately named file folders using a date hierarchy naming system established by the primary analyst.

Additionally, these files are stored electronically on external USB storage devices and on internal network drives. See Sections 5.5.1 and 5.5.2 for more details on UHPLC instrument file naming and storage conventions.

7.2 Sample Information Forms, Chain of Custody Forms, and Extraction Manifold Worksheets

The hard copies of SIFs, COC forms, and Extraction Manifold Worksheets for the current analytical run must be saved in the hard copy of the current run folder under review. Additionally, electronic scans of the SIFs and Extraction Manifold Worksheets must be stored in the current analysis run folder under review on the **P-drive**.

7.3 Standard Dilution Checklist

This document is named “**DAQ-03-017 Carbonyl Calibration and QC Standard Dilutions Checklist**” and can be found here:

P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Standard Dilution Chart

The primary analyst may print several copies to keep in a drawer in the extraction lab room. Filled out copies of these forms are stored with the hard copy run folder and a scanned copy of the filled-out form is stored in the electronic copy of the run folder stored on the **P-drive**.

If changes to this document are needed due to a change in calibration curve concentrations levels or a change to stock standard concentrations, or changes to spiking levels, the document will be saved with the same name as quoted above, but with “**_revision 1**” added to end of file name. The revision number is in the top left corner of the excel file will need to be changed to match the new revision number.

Using the file name example given above, the new file would now be named: "**DAQ-03-017 Carbonyl Calibration and QC Standard Dilutions Checklist_revision 1**" and stored in the same location mapped above.

7.4 Level 1 and Level 2 Data Review Checklists

The data review checklist is named "**DAQ-03-013 UAT and PAMS Carbonyl Sampling and Analysis Data Review Checklist**" and stored here:

P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Data Review

This document is printed and filled out with handwritten entries made by the self and peer reviewer. Once the data have been reviewed and approved by the level 2 reviewer, the handwritten copy of the data review checklist must be saved in the hard copy of the analysis folder under review. Additionally, scan the completed review checklist and store the scan in the electronic copy of the current run folder under review located on the **P-drive**.

If changes are need to the questions and/or documents listed in the form, the form is saved in the same location as mapped above and named **DAQ-03-013 UAT and PAMS Carbonyl Sampling and Analysis Data Review Checklist_Revision 1.**

7.5 Carbonyl Master Spreadsheet

The Carbonyl Master Spreadsheet is named **DAQ-03-012 UAT and PAMS Carbonyl Master Spreadsheet**. The template is located here:

P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Master Spreadsheet

The primary analyst will save this file with the current calendar year in the file name. If creating the master spreadsheet for calendar year 2021, name the file: "**2021_DAQ 03-012 UAT and PAMS Carbonyl Master Spreadsheet** and save this file in the current year's data folder located here:

P:\Toxics\Urban Air Toxics\Aldehyde Data

This spreadsheet is used to track critical sampling and analysis data collected during the 2021 calendar year.

If any part of the form headers or column names change or if columns are added, the above file example is saved as: "**2021_DAQ 03-012 UAT and PAMS Carbonyl Master Spreadsheet_revision 1**" and saved in the same location on the **P-drive**.

7.6 Tracking Chart

The Carbonyl tracking chart is used by the analyst and lab staff to keep track of cartridge shipping and receiving. It provides a reference guide for scheduling cartridge prep and extractions. This file is stored on the **P-drive**: **P:/Toxics/Urban Air Toxics/VOC Current Year Data/Tracking Chart UAT VOC & Aldehyde**

7.7 QA Summary Report

This report is named "**DAQ-03-16 UAT and PAMS QA Summary Report"** and is stored here:

P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Data Review. The primary analyst will open this file and save it with the following name **DAQ-03-16 UAT and PAMS QA Summary Report_MMDDYY** where the MMDDYY represents the current run folder to which the QA Summary Report applies.

For example, if the QA summary report is for the **030521_Aldehyde Run_UV** run folder; the QA Summary report will be named "**DAQ-03-16 UAT and PAMS QA Summary Report_030521"** and stored in the **030521_Aldehyde Run_UV** run folder on the **P-drive**.

See Section 9.5 of this SOP for information on how to document errors found on the QA Summary Report.

If any part of the QA Summary Report headers, worksheets, or column names change or if columns are added, the above file example is saved as: "**DAQ 03-016 UAT and PAMS QA Summary Report_revision 1**" and saved here: **P:\Toxics\Urban Air Toxics\Aldehyde Data\Carbonyl Data Review.**

8.0 QUALITY ASSURANCE FILES AND DATA HANDLING

8.1 Quarterly Pipette and Balance Verification

Quarterly pipette and balance verification worksheets (as discussed in the section 3.4.1 and 3.4.2) records have been entered into the pipette verification worksheet located on internal network drives: **P:/Toxics/Urban Air toxics/Aldehyde data/Balance and Pipette Checks**. Hard copies are stored in the folder in the designated file cabinet.

8.2 Certificate of Analysis Storage

Certificate of analysis of Carbonyl cartridges, solvent (Acetonitrile) and Standards used for the carbonyl analysis are stored in a designated file cabinet in the RCL building.

8.3 Carbonyl Extraction Logbook

This Extraction logbook (Logbook# EXT-XX) will document the analytical work done in the extraction lab such as extraction, preparation of mobile phases, preparation of calibration curve, and quarterly verification of the analytical balance and mechanical pipettes.

8.4 Carbonyl Instrument Logbook

This instrument logbook (Logbook# Inst-XX) will document analysis of carbonyl samples on the UHPLC-UV-MS, calibration and check standard used for the analysis, any maintenance or troubleshooting for the UHPLC-UV-MS.

8.5 Monthly UHPLC Instrument Data Backup

At the conclusion of each month all data must be copied to the external hard drive for data backup and archival.

9.0 TROUBLESHOOTING AND CORRECTIVE ACTIONS

This section of the SOP describes the troubleshooting and corrective actions taken whenever there is an issue with the instrument or its supporting equipment, failing QC checks, and documenting errors found during the data review process, etc.

9.1 Troubleshooting and Corrective Actions for UHPLC

Please see **Table 4** below for a description of troubleshooting and corrective actions of the UHPLC Instrument. The UHPLC system and equipment modules are serviced yearly with basic preventative maintenance performed by certified technicians from Unity Lab Services. The UHPLC and its components are under annual service contracts from Unity Lab Services which also provides for quick repair in the event non-consumable UHPLC parts break or malfunction. Thermo Fisher and Unity Lab Services provide technical support via phone for assistance with UHPLC system troubleshooting that may be repaired by the operator and may not require an onsite visit by a certified technician.

Issue	Possible Cause	Possible Solution
High backpressure	-Plugged inlet frit on column -Plugged frit in inline filter -Column blockage (chemical contamination)	-Backflush column -Inspect frits in filters and replace as necessary -Clean column with solvent or replace column if irreversible
Fluctuating pressure	-Bubble in pump -Leaking check valve or seals	- Check for the bubble in the line; degas the Mobile phase - Replace or clean check valves; replace pump seals;
Peak tailing	Column void	Run in reverse direction; if poor peak shapes or peak doublets are seen for all peaks, a column void may be present; discard column.
Peak fronting	Channeling in column	Replace column
Extra peaks, or 'ghost' peaks	System contamination	Inject the sample solvent (ACN_Blank) injection to ensure that there is nothing in the sample solvent that contributes to the issue
Retention time shifts	-The column is getting old -Change in flow rate	-Replace Column -Check flow rate
Drifting baseline, Baseline Noise	-Look to detector lamp or flow cell. -Occasional sharp spikes - external electrical interference -Periodic - pump pulses -Random - contamination build-up	-Replace if necessary -Use voltage stabilizer/constant voltage power supply for LC system -Service or replace pulse damper -purge air from pump -clean or replace check valves -degas mobile phase. -Flush or backflush column

Issue	Possible Cause	Possible Solution
Poor Recovery	-Damage syringe and syringe valve -Poor alignment in auto sampler injection port -Bad Injector	-Replace syringe if necessary -Realign the auto sampler injection port -Replace injector

Table 4: Troubleshooting and corrective actions of the UHPLC Instrument.

9.2 Troubleshooting and Corrective Actions for Balance and Pipette Checks

1. Analytical balance and pipette equipment must be verified quarterly, and pass acceptance criteria.
2. If a balance or pipette check fails acceptance criteria, confirm the failing condition by performing a second verification check.
3. If two consecutive balance and/or pipette verification checks fail acceptance criteria the equipment must be repaired and or recalibrated where applicable. If the equipment is used while under this failing condition all analysis results must be minimally qualified “QX” back to the most recent expiration of a passing verification check and continue until the equipment is repaired, replaced, or recalibrated. A subsequent verification check is required for recalibrated or new equipment.
4. If there is an instance of a past due quarterly verification, perform a verification as soon as possible according to steps in Section 3.4.1 and 3.4.2.
5. If by any chance the pipette is dropped, perform a pipette verification check. The pipette must pass acceptance criteria before the next use.
6. If the balance is moved or bumped out of place, re-position the balance, re-level the balance, and perform a balance verification check. The balance must past acceptance criteria before next use.

9.3 Troubleshooting and Corrective Actions for Sampling Data

1. As mentioned in section 5.2, sample volume should be within 1240L to 1650L for UAT samples and 414L to 550L for PAMS. Flow should be 0.9 to 1.1 L/min for both PAMS and UAT samples. If the total volume or flow are not within acceptable range, the sample results must be qualified “W” before reporting to AQS. Add a comment noting the flow or volume failure on the carbonyl cartridge SIF if the site operator has not already provided a flow or volume comment on the SIF.
2. If sample collection has been interrupted for more than one hour during the collection period due to a power outage or due to weather, a makeup sample needs to be arranged.
3. If there is any instance of spilling sample extract before bringing to 5mL volume and mixing, the sample needs to be voided and a makeup sample needs to be arranged. A comment should be added to the extraction manifold worksheet and extraction lab logbook summarizing the issue. If the extract is filled to the 5mL mark and mixed and then is spilled, some of the extract may be saved for analysis. In this case note the issue on the extraction manifold worksheet and provide a comment in the master spreadsheet.
4. Makeup samples need to be arranged within 30 days of the original sample date and within the same calendar year for UAT samples whereas for PAMS make up samples should be within two weeks from the originally scheduled collection date and collected within PAMS season (June 1 to August 31).

9.4 Troubleshooting and Corrective Actions for Carbonyl cartridges

When doing the carbonyl cartridges lot blank test, the following compounds must meet concentrations and acceptance limits before field use: Formaldehyde ≤ 0.03 µg/mL; and all other compounds ≤ 0.02 µg/mL to pass. If Formaldehyde and/or Acetaldehyde do not meet acceptance limits, the lot must be disqualified for field use. The failing lot of cartridges should be returned to the manufacturer for replacement if possible.

9.5 Troubleshooting and Corrective Actions for Data Review Process

Once the data processing has been completed by the analyst, Data needs to be verified by a Level 1 and Level 2 data reviewer. Errors found during the review process are documented in the Level 1/Level 2 Data Review Checklist, QA Summary Report, and master spreadsheet. Errors corrected in the master spreadsheet and QA Summary report must be document as follows:

- Go to cell containing the error, remove the error and enter the correct information. Right click on the same cell and select **add comment**. Document the error in the popup comment box, the comment must include the original data and briefly explain the correction.
- The analyst can also document the correction in the **comment** column on the master spreadsheet or the **comment** section in the QA Summary Report.

10.0 REVISION HISTORY

1. Rev 0 – **CB** 6/29/2021 Original Publication

11.0 REFERENCES

1. Environmental Protection Agency (EPA) Compendium Method TO-11A-Determination of Formaldehyde in Ambient Air Using Adsorbent Cartridge Followed by High Performance Liquid Chromatography (HPLC), Center for Environmental Research Information Office of Research and Development U.S. Environmental Protection Agency Cincinnati, OH 45268. January 1999. https://www.epa.gov/sites/production/files/2019-11/documents/to-11ar.pdf

2. Technical Assistance Document for the National Air Toxics Trends Stations Program, Revision 3, Prepared for: U.S. Environmental Protection Agency Office of Air Quality Planning and Standards (C304-06) Research Triangle Park, NC 27711 https://www3.epa.gov/ttnamti1/files/ambient/airtox/NATTS%20TAD%20Revision%203_FINAL%20October%202016.pdf

3. Technical Assistance Document for the Sampling and Analysis of Ozone Precursors for the Photochemical Assessment Monitoring Stations Program, Revision 2, Prepared for: U.S. Environmental Protection Agency Office of Air Quality Planning and Standards (C304-06) Research Triangle Park, NC 27711 https://www.epa.gov/sites/production/files/2019-11/documents/pams_technical_assistance_document_revision_2_april_2019.pdf

4. North Carolina Department of Environmental Quality, Division of Air Quality (2021). Quality Assurance Project Plan for the North Carolina Division of Air Photochemical Assessment Monitoring Stations (PAMS) Required Site Network for Speciated Volatile Organic Compounds, Carbonyls, and Meteorological Parameters Including Mixing Layer Height. Revision 0. Raleigh, NC. DAQ Document ID: DAQ-01-007. Steger, Joette; Walters, Steven and Velleco, Brian D.

5. Pipette Verification Acceptance Criteria: https://www.artel.co/wp-content/uploads/2016/01/Lab-Report-5-Setting-Tolerances-for-pipettes-in-the-laboratory.pdf Pipette Verification Method: https://www.integra-biosciences.com/united-states/en/routine-check-pipettes-best-practices and Z-factor table lookup: https://www.integra-biosciences.com/sites/default/files/Z-factors-calibration_2017_01_26.pdf

12.0 APPENDICES

1. Appendix A – Pipette Verification Worksheet
2. Appendix B – Balance Verification Worksheet
3. Appendix C – Carbonyl Cartridge Sample Information Form
4. Appendix D – Carbonyl Cartridge Chain of Custody Form
5. Appendix E – UAT Carbonyl Samples Tracking Chart
6. Appendix F – PAMS Carbonyl Samples Tracking Chart
7. Appendix G – Aldehyde Extraction Manifold Work Sheet
8. Appendix H – Extraction Manifold Map
9. Appendix I – MS Detector Real-Time Scan Settings
10. Appendix J – Calibration and QC Standard Dilution Chart
11. Appendix K – Example Sequence for UV Detector Analysis
12. Appendix L – Example Sequence for MS Detector Analysis
13. Appendix M – Unedited Chromatogram
14. Appendix N – Edited Chromatogram
15. Appendix O – Example Certificate of Analysis for Carbonyl Standards

Appendix A – Pipette Verification Worksheet

Pipette Verification Worksheet

	Make=			**Room Temp (°C)**			***Document#***	***DAQ-03-014***
	Serial Number=			**Pressure (kPA)**			***Version***	***0.0***
	Date Performed=			**Barometer SN=**			***Effective Date***	***6/1/2021***
	Liquid Used=							
Test #	**Balance Reading (g)**	**Weighing Mass(mg)**	**Z factor**	**Low Settting Volume(uL)**	**Balance Reading (g)**	**Weighing Mass(mg)**	**Z factor**	**High Setting Volume(uL)**
1		0.0000		0.0000		0.0000		0.0000
2		0.0000		0.0000		0.0000		0.0000
3		0.0000		0.0000		0.0000		0.0000
4		0.0000		0.0000		0.0000		0.0000
5		0.0000		0.0000		0.0000		0.0000
Balance S/N								
			MEAN	0.0000			MEAN	0.0000
			STDEV	0.0000			STDEV	0.0000
Expiration Date:			Accuracy	#DIV/0!			Accuracy	#DIV/0!
			CV	#DIV/0!			CV	#DIV/0!
			Accuracy	8.00%			Accuracy	2.00%
			CV	3.00%			CV	1.00%

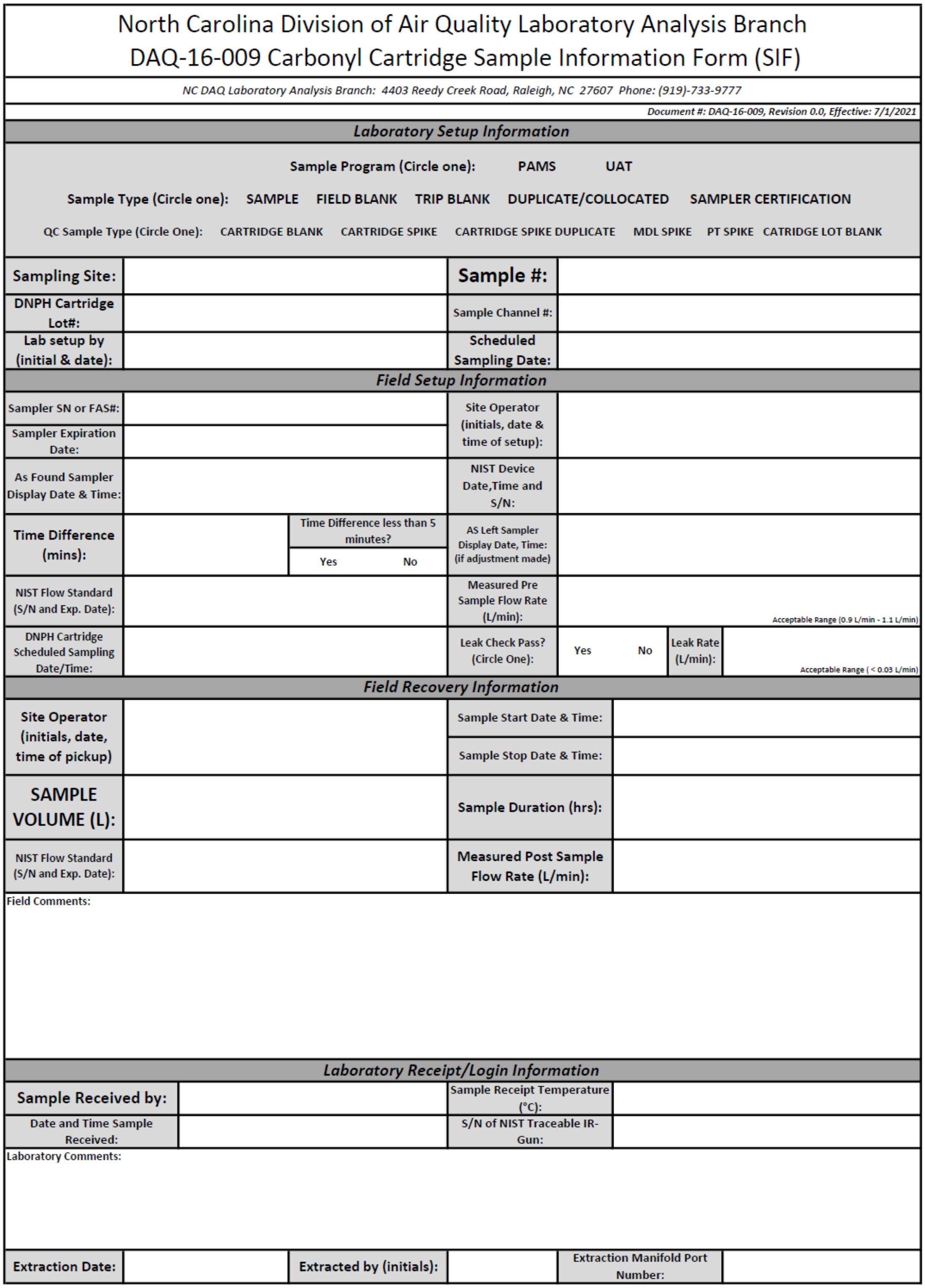
Comments:

Level 2 Review By:

Appendix B – Balance Verification Worksheet

Balance Verification Worksheet			
Balance Make/Model=		***Document#***	***DAQ-03-015***
Balance Serial Number=		***Version***	***0.0***
Weight Make/Model=		***Effective Date***	***6/1/2021***
Weight Serial Number=			
Room Temp (°C)=		**Date of Verification=**	
Pressure (kPA)=		**Analyst Initials=**	
Barometer SN=			
Nominal Weight	**Nominal Weight Unit**	**Measured Weight (g)**	**Accuracy (% Diff)**
5	mg		100.00%
200	mg		100.00%
500	mg		100.00%
2	g		100.00%
		Accuracy	2%
Comment:			
Level 2 Review By:			

Appendix C – Carbonyl Cartridge Sample Information Form



North Carolina Division of Air Quality Laboratory Analysis Branch
DAQ-16-009 Carbonyl Cartridge Sample Information Form (SIF)

NC DAQ Laboratory Analysis Branch: 4403 Reedy Creek Road, Raleigh, NC 27607 Phone: (919)-733-9777

Document #: DAQ-16-009, Revision 0.0, Effective: 7/1/2021

Laboratory Setup Information

Sample Program (Circle one): PAMS UAT

Sample Type (Circle one): SAMPLE FIELD BLANK TRIP BLANK DUPLICATE/COLLOCATED SAMPLER CERTIFICATION

QC Sample Type (Circle One): CARTRIDGE BLANK CARTRIDGE SPIKE CARTRIDGE SPIKE DUPLICATE MDL SPIKE PT SPIKE CATRIDGE LOT BLANK

Sampling Site:		**Sample #:**	
DNPH Cartridge Lot#:		**Sample Channel #:**	
Lab setup by (initial & date):		**Scheduled Sampling Date:**	

Field Setup Information

Sampler SN or FAS#:			**Site Operator (initials, date & time of setup):**		
Sampler Expiration Date:					
As Found Sampler Display Date & Time:			**NIST Device Date,Time and S/N:**		
Time Difference (mins):		**Time Difference less than 5 minutes?** Yes No	**AS Left Sampler Display Date, Time: (if adjustment made)**		
NIST Flow Standard (S/N and Exp. Date):			**Measured Pre Sample Flow Rate (L/min):**	Acceptable Range (0.9 L/min - 1.1 L/min)	
DNPH Cartridge Scheduled Sampling Date/Time:			**Leak Check Pass? (Circle One):**	Yes No	**Leak Rate (L/min):** Acceptable Range (< 0.03 L/min)

Field Recovery Information

Site Operator (initials, date, time of pickup)		**Sample Start Date & Time:**	
		Sample Stop Date & Time:	
SAMPLE VOLUME (L):		**Sample Duration (hrs):**	
NIST Flow Standard (S/N and Exp. Date):		**Measured Post Sample Flow Rate (L/min):**	

Field Comments:

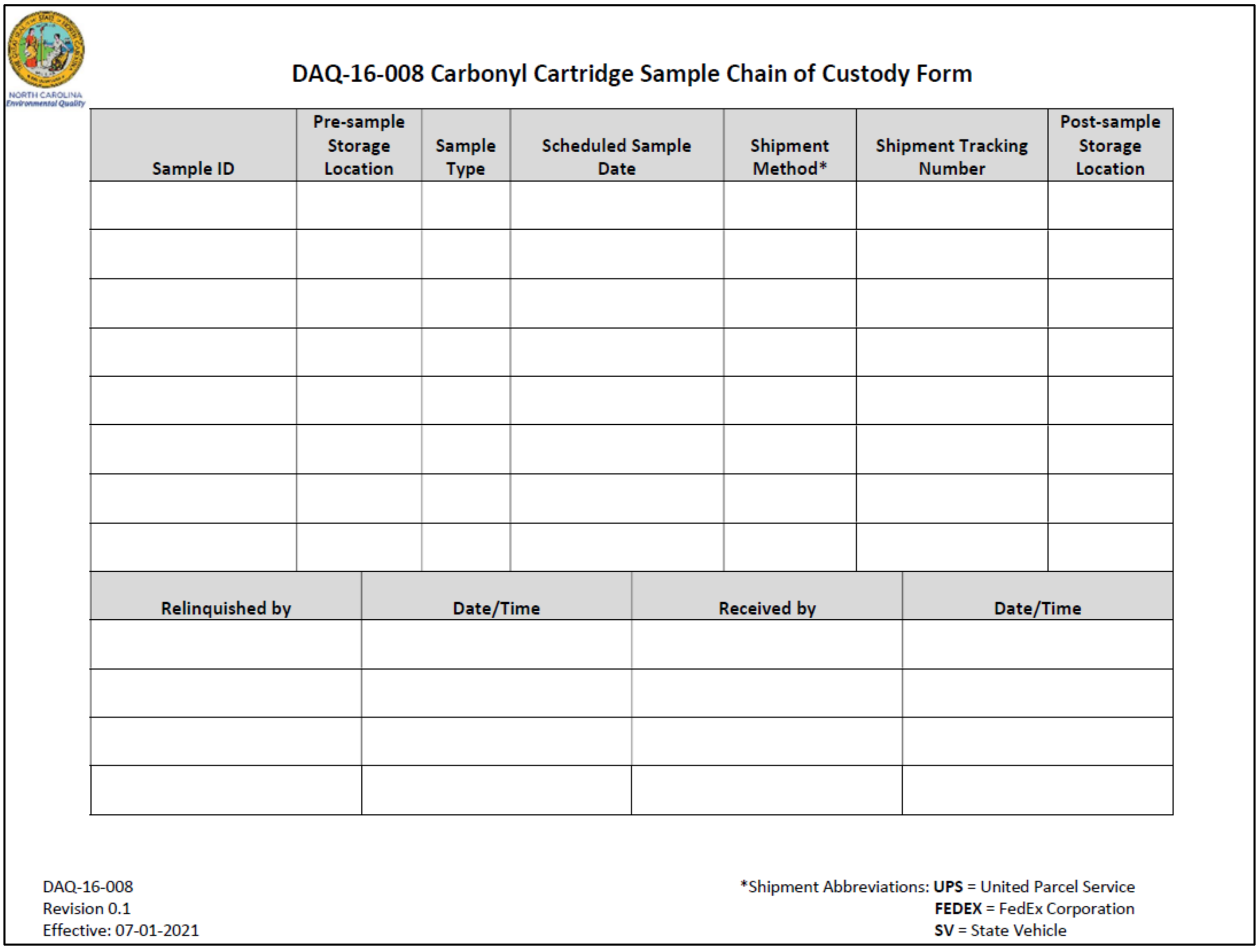
Laboratory Receipt/Login Information

Sample Received by:		**Sample Receipt Temperature (°C):**	
Date and Time Sample Received:		**S/N of NIST Traceable IR-Gun:**	

Laboratory Comments:

Extraction Date:		**Extracted by (initials):**		**Extraction Manifold Port Number:**	

Appendix D – Carbonyl Cartridge Chain of Custody Form



NORTH CAROLINA
Environmental Quality

DAQ-16-008 Carbonyl Cartridge Sample Chain of Custody Form

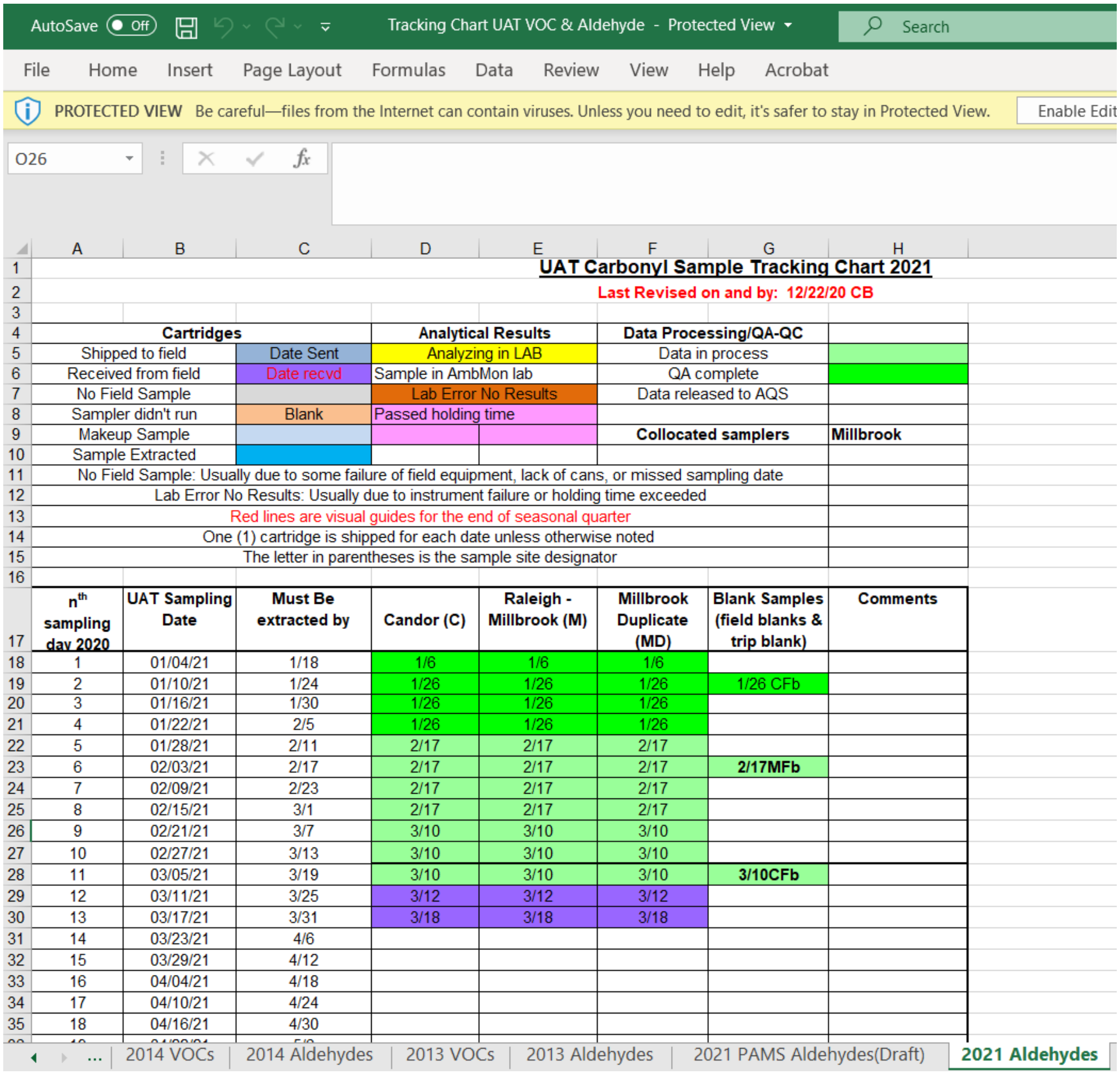
Sample ID	Pre-sample Storage Location	Sample Type	Scheduled Sample Date	Shipment Method*	Shipment Tracking Number	Post-sample Storage Location

Relinquished by	Date/Time	Received by	Date/Time

DAQ-16-008
Revision 0.1
Effective: 07-01-2021

*Shipment Abbreviations: **UPS** = United Parcel Service
FEDEX = FedEx Corporation
SV = State Vehicle

Appendix E – UAT Carbonyl Samples Tracking Chart



UAT Carbonyl Sample Tracking Chart 2021

Last Revised on and by: 12/22/20 CB

Cartridges		Analytical Results	Data Processing/QA-QC	
Shipped to field	Date Sent	Analyzing in LAB	Data in process	🟩
Received from field	Date recvd	Sample in AmbMon lab	QA complete	🟩
No Field Sample	▨	Lab Error No Results	Data released to AQS	
Sampler didn't run	Blank	Passed holding time		
Makeup Sample	🟦		**Collocated samplers**	**Millbrook**
Sample Extracted	🟦			

No Field Sample: Usually due to some failure of field equipment, lack of cans, or missed sampling date

Lab Error No Results: Usually due to instrument failure or holding time exceeded

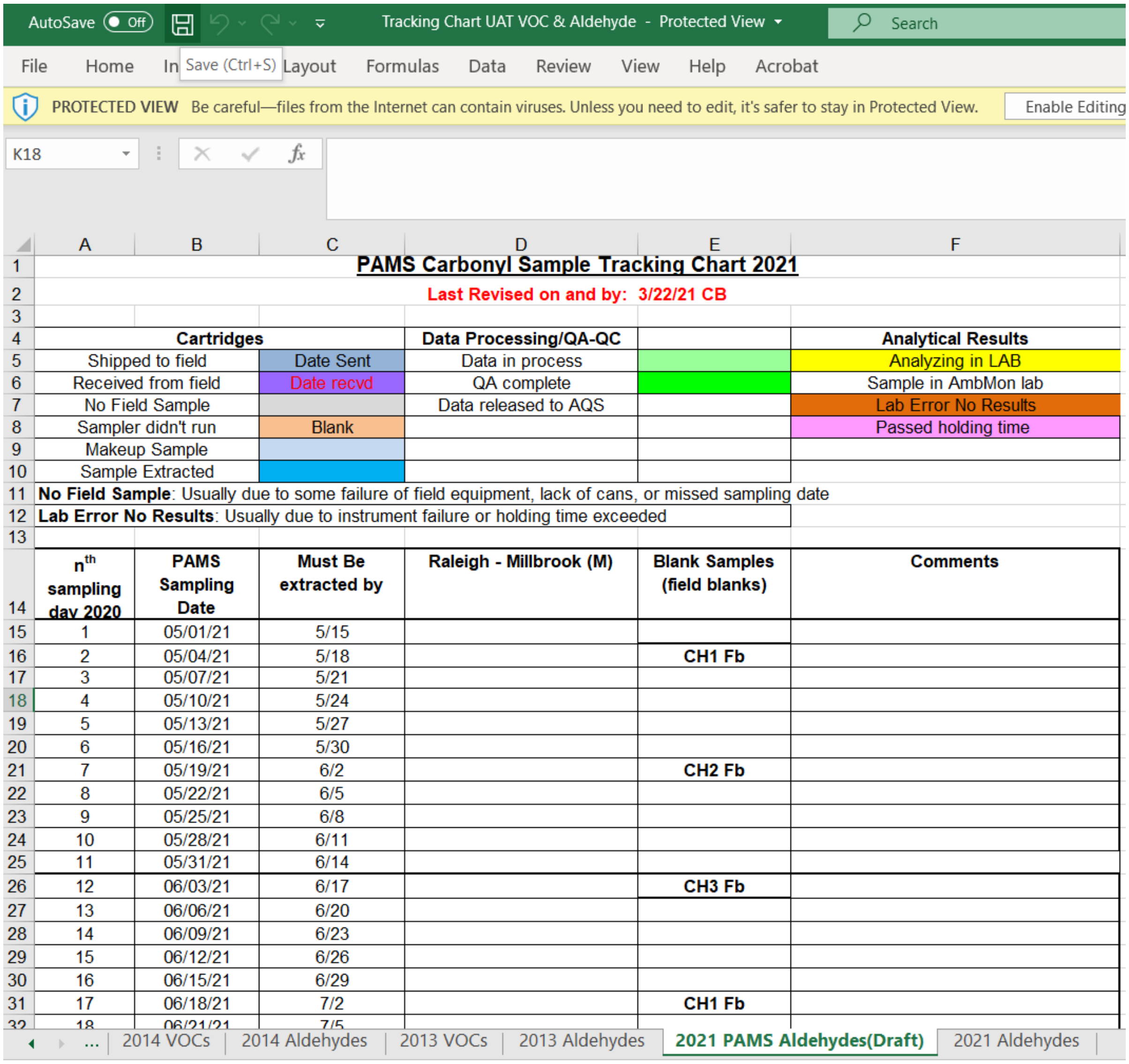
Red lines are visual guides for the end of seasonal quarter

One (1) cartridge is shipped for each date unless otherwise noted

The letter in parentheses is the sample site designator

n^{th} sampling day 2020	UAT Sampling Date	Must Be extracted by	Candor (C)	Raleigh - Millbrook (M)	Millbrook Duplicate (MD)	Blank Samples (field blanks & trip blank)	Comments
1	01/04/21	1/18	1/6	1/6	1/6		
2	01/10/21	1/24	1/26	1/26	1/26	1/26 CFb	
3	01/16/21	1/30	1/26	1/26	1/26		
4	01/22/21	2/5	1/26	1/26	1/26		
5	01/28/21	2/11	2/17	2/17	2/17		
6	02/03/21	2/17	2/17	2/17	2/17	**2/17MFb**	
7	02/09/21	2/23	2/17	2/17	2/17		
8	02/15/21	3/1	2/17	2/17	2/17		
9	02/21/21	3/7	3/10	3/10	3/10		
10	02/27/21	3/13	3/10	3/10	3/10		
11	03/05/21	3/19	3/10	3/10	3/10	**3/10CFb**	
12	03/11/21	3/25	3/12	3/12	3/12		
13	03/17/21	3/31	3/18	3/18	3/18		
14	03/23/21	4/6					
15	03/29/21	4/12					
16	04/04/21	4/18					
17	04/10/21	4/24					
18	04/16/21	4/30					

Appendix F – PAMS Carbonyl Samples Tracking Chart



PAMS Carbonyl Sample Tracking Chart 2021

Last Revised on and by: 3/22/21 CB

Cartridges		Data Processing/QA-QC		Analytical Results
Shipped to field	Date Sent	Data in process	🟩	Analyzing in LAB
Received from field	Date recvd	QA complete	🟩	Sample in AmbMon lab
No Field Sample	⬜	Data released to AQS		Lab Error No Results
Sampler didn't run	Blank			Passed holding time
Makeup Sample	🟦			
Sample Extracted	🟦			

No Field Sample: Usually due to some failure of field equipment, lack of cans, or missed sampling date

Lab Error No Results: Usually due to instrument failure or holding time exceeded

n^{th} sampling day 2020	PAMS Sampling Date	Must Be extracted by	Raleigh - Millbrook (M)	Blank Samples (field blanks)	Comments
1	05/01/21	5/15			
2	05/04/21	5/18		CH1 Fb	
3	05/07/21	5/21			
4	05/10/21	5/24			
5	05/13/21	5/27			
6	05/16/21	5/30			
7	05/19/21	6/2		CH2 Fb	
8	05/22/21	6/5			
9	05/25/21	6/8			
10	05/28/21	6/11			
11	05/31/21	6/14			
12	06/03/21	6/17		CH3 Fb	
13	06/06/21	6/20			
14	06/09/21	6/23			
15	06/12/21	6/26			
16	06/15/21	6/29			
17	06/18/21	7/2		CH1 Fb	
18	06/21/21	7/5			

Appendix G – Aldehyde Extraction Manifold Work Sheet

Aldehyde Extraction Manifold Worksheet

Analyst			
Extraction Date			
Cartridge Sample Dates			

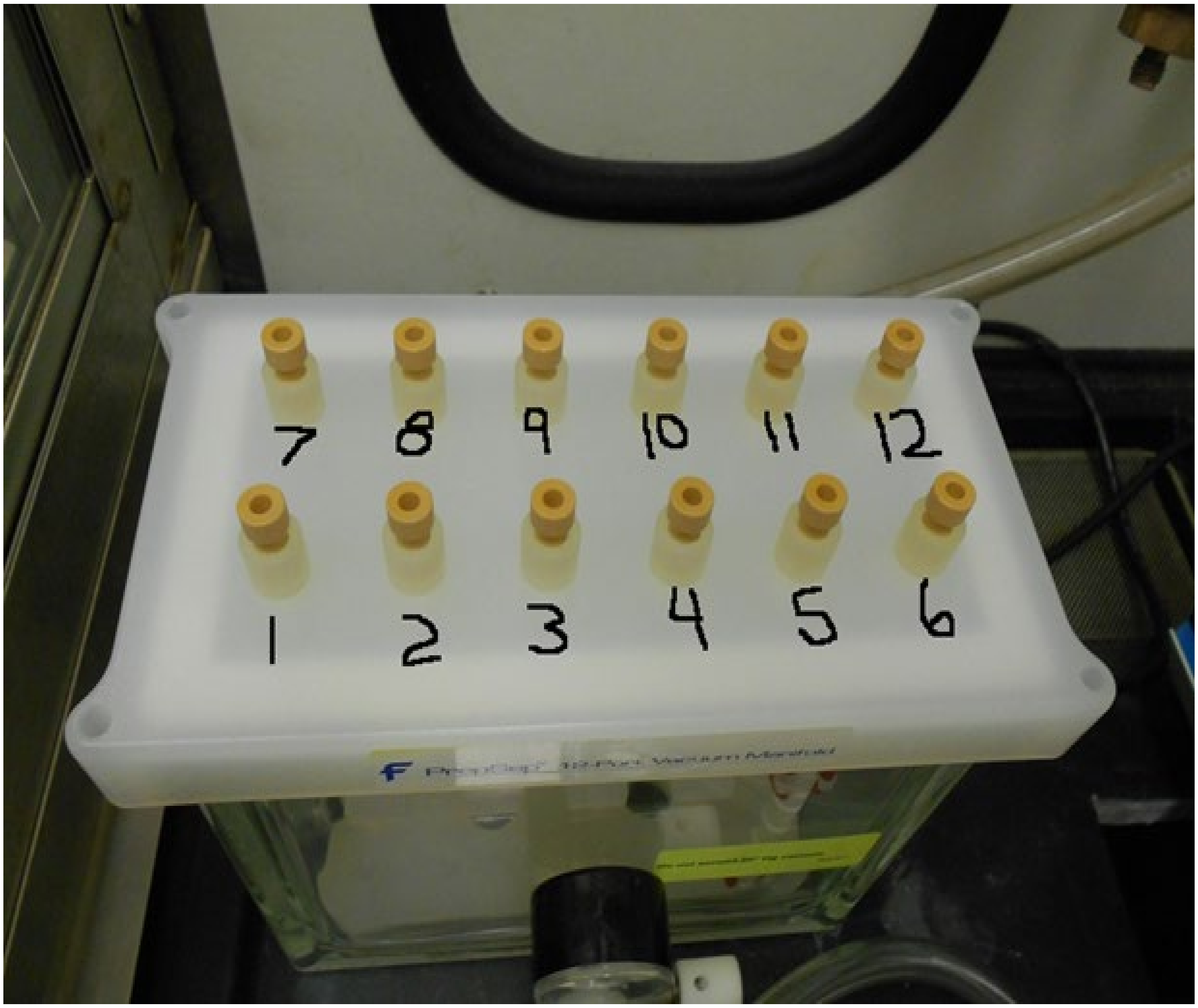
Extraction Material Lot #s

Acetonitrile	
4mL clear storage vials	
1.5 mL autosampler vials	
utosampler caps with septa	
Syringe filters	

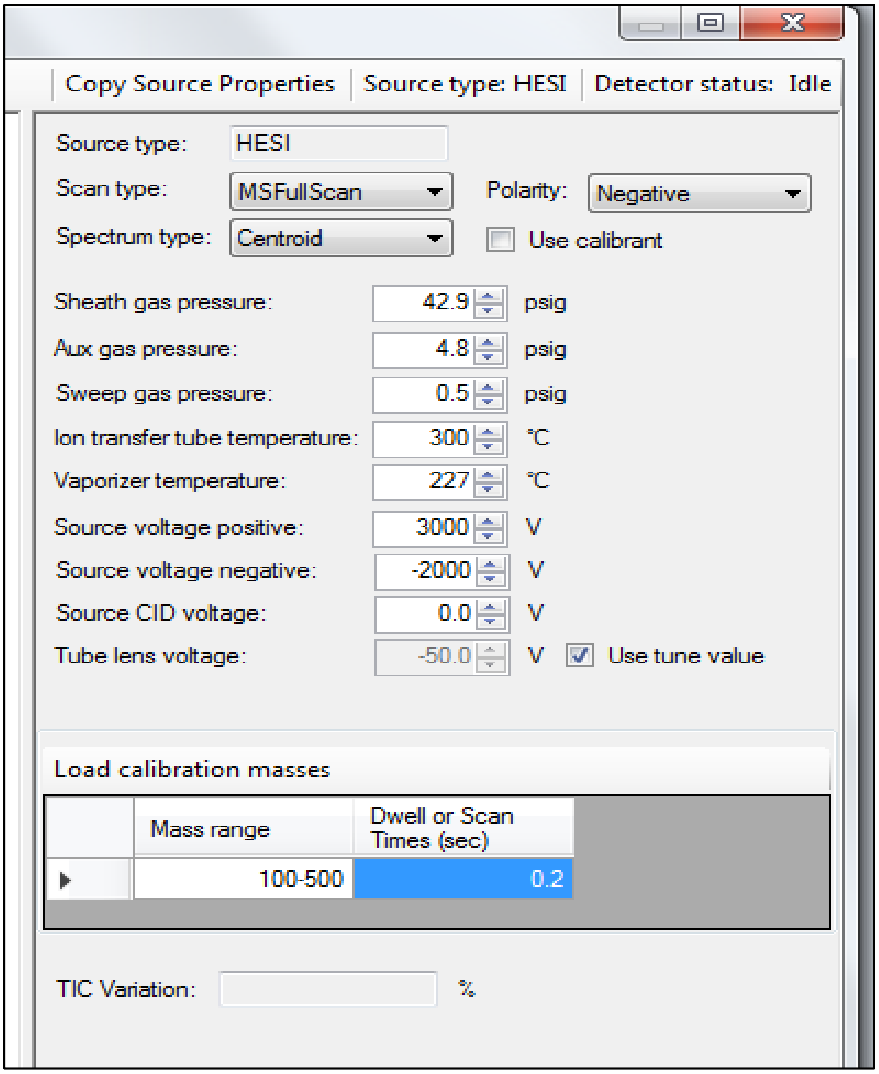
Port #	Sample ID	Port #	Sample ID
1		7	
2		8	
3		9	
4		10	
5		11	
6		12	

COMMENTS:

Appendix H – Extraction Manifold Map



Appendix I – MS Detector Real-Time Scan Settings



Source type: HESI

Scan type: MSFullScan

Polarity: Negative

Spectrum type: Centroid

☐ Use calibrant

Sheath gas pressure: 42.9 psig

Aux gas pressure: 4.8 psig

Sweep gas pressure: 0.5 psig

Ion transfer tube temperature: 300 °C

Vaporizer temperature: 227 °C

Source voltage positive: 3000 V

Source voltage negative: -2000 V

Source CID voltage: 0.0 V

Tube lens voltage: -50.0 V ☑ Use tune value

Load calibration masses

	Mass range	Dwell or Scan Times (sec)
▶	100-500	0.2

TIC Variation: %

Appendix J – Calibration and QC Standard Dilution Chart

Document#	***DAQ-03-017***
Revision	***0***
Effective Date	***4/1/2021***

CHECKLIST FOR MAKING CALIBRATION AND QC STANDARDS FOR CARBONYL ANALYSIS

Extraction Date:				Run Date :	
Standard Name; Identifier	**Contents**	**Manufacturer**	**Concentration**	**Expiration Date**	**Lot #**
CAL1	CARB-1004 mix-DNPH (13 compound mix)	Restek	3.00 ug/mL		
QA	CARB-1004 mix-DNPH (13 compound mix)	Sigma-Aldrich	30.00 ug/mL		
QA1	CARB-1004 mix-DNPH (13 compound mix)	Sigma-Aldrich/Restek (diff lot#)	3.00 ug/mL	Date opened/made:	
TO make QA1 from QA: Pipette 500uL of QA into a 5mL volumetric flask, fill to mark with Carbonyl Free Grade Acetonitrile					

CALIBRATION CURVE DILUTION INSTRUCTIONS

Standard Name; Identifier	**Contents**	**Manufacturer**	**Concentration**	Date Made	Analyst
Level 6 (3.00ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Restek	3.00 ug/mL		
TO make Level 6: None use as purchased from manufacturer (use leftover CAL1 after making Level 5)					
Level 5 (1.00ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Restek	1.00 ug/mL		
TO make Level 5: First pipette 800uL of Carbonyl Free Grade Acetonitrile into a 4mL storage vial then pipette 400uL of CAL1 into the same vial					
Level 4 (0.30ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Restek	0.30 ug/mL		
TO make Level 4: First pipette 840uL of Carbonyl Free Grade Acetonitrile into a 4mL storage vial then pipette 360uL of Level 5 into the same vial					
Level 3 (0.10ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Restek	0.10 ug/mL		
TO make Level 3: First pipette 800uL of Carbonyl Free Grade Acetonitrile into a 4mL storage vial then pipette 400uL of Level 4 into the same vial					
Level 2 (0.03ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Restek	0.03 ug/mL		
TO make Level 2: First pipette 840uL of Carbonyl Free Grade Acetonitrile into a 4mL storage vial then pipette 360uL of Level 3 into the same vial					
Level 1 (0.01ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Restek	0.01 ug/mL		
TO make Level 1: First pipette 800uL of Carbonyl Free Grade Acetonitrile into a 4mL storage vial then pipette 400uL of Level 2 into the same vial					
Level 0 (0.003ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Restek	0.003 ug/mL		
TO make Level 0: First pipette 840uL of Carbonyl Free Grade Acetonitrile into a 4mL storage vial then pipette 360uL of Level 1 into the same vial					

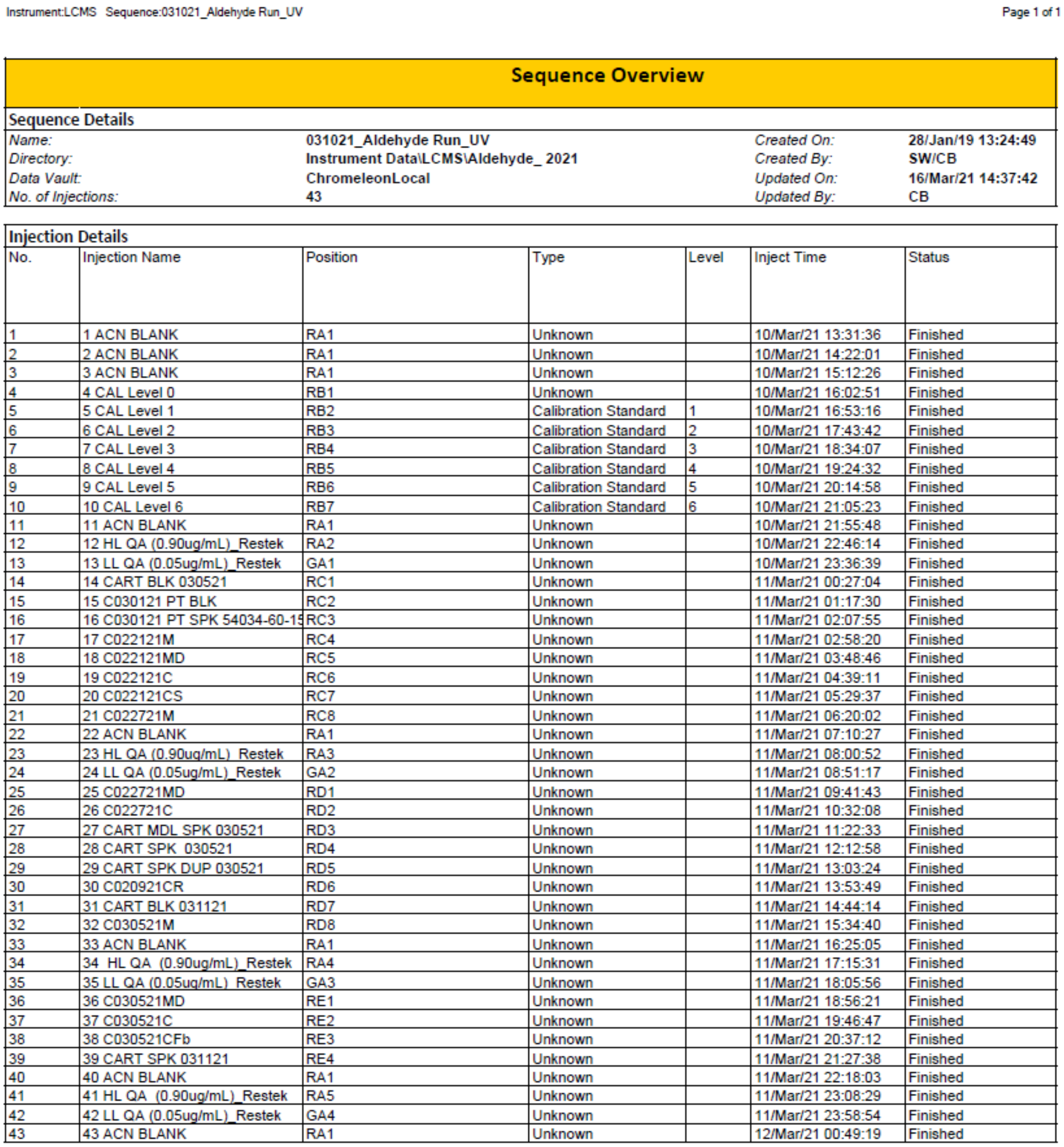
QA SAMPLE DILUTION INSTRUCTIONS

Standard Name; Identifier	**Contents**	**Manufacturer**	**Concentration**	Date Made	Analyst
HL QA (0.9ug/mL)	CARB-1004 mix-DNPH (13 compound mix)	Sigma-Aldrich or Restek (diff lot#)	0.90 ug/mL		
TO make HL QA: First pipette 1400uL of Carbonyl Free Grade Acetonitrile into a 4mL storage vial then pipette 600uL of QA1 into the same vial					
LL QA (0.05ug/mL)	*CARB-1004 mix-DNPH (13 compound mix)*	Sigma-Aldrich or Restek (diff lot#)	*0.05 ug/mL*		
To make LL QA: First Pipette 2950uL of Carbonyl free Acetonitrile into a 4mL storage vial then pipette 50uL of QA1 into the same vial					
CART SPK and CART SPK DUP	CARB-1004 mix-DNPH (13 compound mix)	Sigma-Aldrich or Restek (diff lot#)	0.15ug/mL		
TO make CART SPK or CART SPK DUP: spike a blank unsampled DNPH cartridge with 250uL of or QA1					
MDL SPK	CARB-1004 mix-DNPH (13 compound mix)	Sigma-Aldrich or Restek (diff lot#)	0.021 ug/mL		
TO make MDL SPK: spike a blank unsampled DNPH cartridge with 35uL of QA1					

Pipette SN:

Comments:

Appendix K – Example Sequence for UV Detector Analysis



Instrument:LCMS Sequence:031021_Aldehyde Run_UV

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Sequence Overview

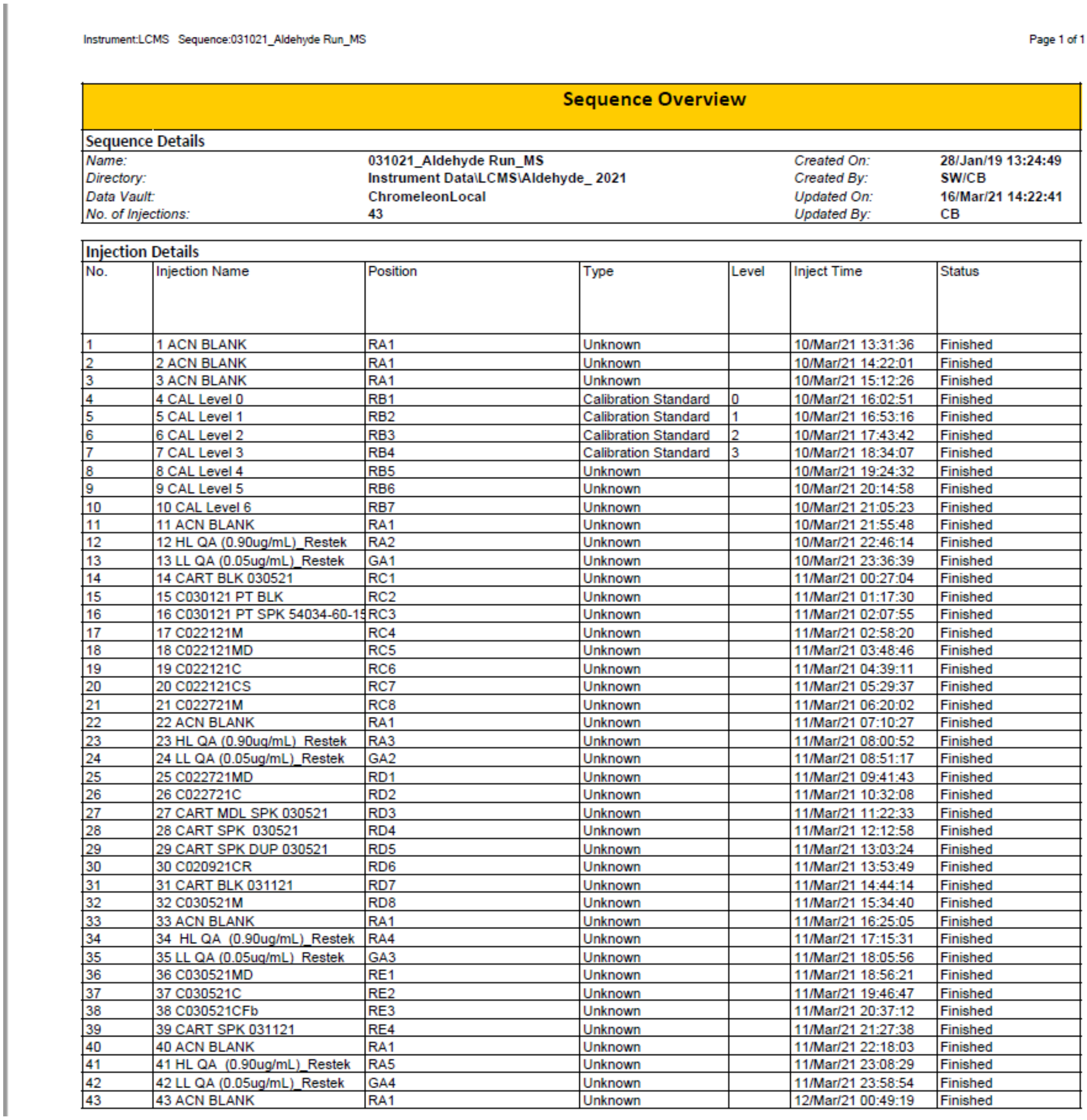
Sequence Details

Name:	**031021_Aldehyde Run_UV**	*Created On:*	**28/Jan/19 13:24:49**
Directory:	**Instrument Data\LCMS\Aldehyde_ 2021**	*Created By:*	**SW/CB**
Data Vault:	**ChromeleonLocal**	*Updated On:*	**16/Mar/21 14:37:42**
No. of Injections:	**43**	*Updated By:*	**CB**

Injection Details

No.	Injection Name	Position	Type	Level	Inject Time	Status
1	1 ACN BLANK	RA1	Unknown		10/Mar/21 13:31:36	Finished
2	2 ACN BLANK	RA1	Unknown		10/Mar/21 14:22:01	Finished
3	3 ACN BLANK	RA1	Unknown		10/Mar/21 15:12:26	Finished
4	4 CAL Level 0	RB1	Unknown		10/Mar/21 16:02:51	Finished
5	5 CAL Level 1	RB2	Calibration Standard	1	10/Mar/21 16:53:16	Finished
6	6 CAL Level 2	RB3	Calibration Standard	2	10/Mar/21 17:43:42	Finished
7	7 CAL Level 3	RB4	Calibration Standard	3	10/Mar/21 18:34:07	Finished
8	8 CAL Level 4	RB5	Calibration Standard	4	10/Mar/21 19:24:32	Finished
9	9 CAL Level 5	RB6	Calibration Standard	5	10/Mar/21 20:14:58	Finished
10	10 CAL Level 6	RB7	Calibration Standard	6	10/Mar/21 21:05:23	Finished
11	11 ACN BLANK	RA1	Unknown		10/Mar/21 21:55:48	Finished
12	12 HL QA (0.90ug/mL)_Restek	RA2	Unknown		10/Mar/21 22:46:14	Finished
13	13 LL QA (0.05ug/mL)_Restek	GA1	Unknown		10/Mar/21 23:36:39	Finished
14	14 CART BLK 030521	RC1	Unknown		11/Mar/21 00:27:04	Finished
15	15 C030121 PT BLK	RC2	Unknown		11/Mar/21 01:17:30	Finished
16	16 C030121 PT SPK 54034-60-1	RC3	Unknown		11/Mar/21 02:07:55	Finished
17	17 C022121M	RC4	Unknown		11/Mar/21 02:58:20	Finished
18	18 C022121MD	RC5	Unknown		11/Mar/21 03:48:46	Finished
19	19 C022121C	RC6	Unknown		11/Mar/21 04:39:11	Finished
20	20 C022121CS	RC7	Unknown		11/Mar/21 05:29:37	Finished
21	21 C022721M	RC8	Unknown		11/Mar/21 06:20:02	Finished
22	22 ACN BLANK	RA1	Unknown		11/Mar/21 07:10:27	Finished
23	23 HL QA (0.90ug/mL)_Restek	RA3	Unknown		11/Mar/21 08:00:52	Finished
24	24 LL QA (0.05ug/mL)_Restek	GA2	Unknown		11/Mar/21 08:51:17	Finished
25	25 C022721MD	RD1	Unknown		11/Mar/21 09:41:43	Finished
26	26 C022721C	RD2	Unknown		11/Mar/21 10:32:08	Finished
27	27 CART MDL SPK 030521	RD3	Unknown		11/Mar/21 11:22:33	Finished
28	28 CART SPK 030521	RD4	Unknown		11/Mar/21 12:12:58	Finished
29	29 CART SPK DUP 030521	RD5	Unknown		11/Mar/21 13:03:24	Finished
30	30 C020921CR	RD6	Unknown		11/Mar/21 13:53:49	Finished
31	31 CART BLK 031121	RD7	Unknown		11/Mar/21 14:44:14	Finished
32	32 C030521M	RD8	Unknown		11/Mar/21 15:34:40	Finished
33	33 ACN BLANK	RA1	Unknown		11/Mar/21 16:25:05	Finished
34	34 HL QA (0.90ug/mL)_Restek	RA4	Unknown		11/Mar/21 17:15:31	Finished
35	35 LL QA (0.05ug/mL)_Restek	GA3	Unknown		11/Mar/21 18:05:56	Finished
36	36 C030521MD	RE1	Unknown		11/Mar/21 18:56:21	Finished
37	37 C030521C	RE2	Unknown		11/Mar/21 19:46:47	Finished
38	38 C030521CFb	RE3	Unknown		11/Mar/21 20:37:12	Finished
39	39 CART SPK 031121	RE4	Unknown		11/Mar/21 21:27:38	Finished
40	40 ACN BLANK	RA1	Unknown		11/Mar/21 22:18:03	Finished
41	41 HL QA (0.90ug/mL)_Restek	RA5	Unknown		11/Mar/21 23:08:29	Finished
42	42 LL QA (0.05ug/mL)_Restek	GA4	Unknown		11/Mar/21 23:58:54	Finished
43	43 ACN BLANK	RA1	Unknown		12/Mar/21 00:49:19	Finished

Appendix L – Example Sequence for MS Detector Analysis



Instrument:LCMS Sequence:031021_Aldehyde Run_MS

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Sequence Overview

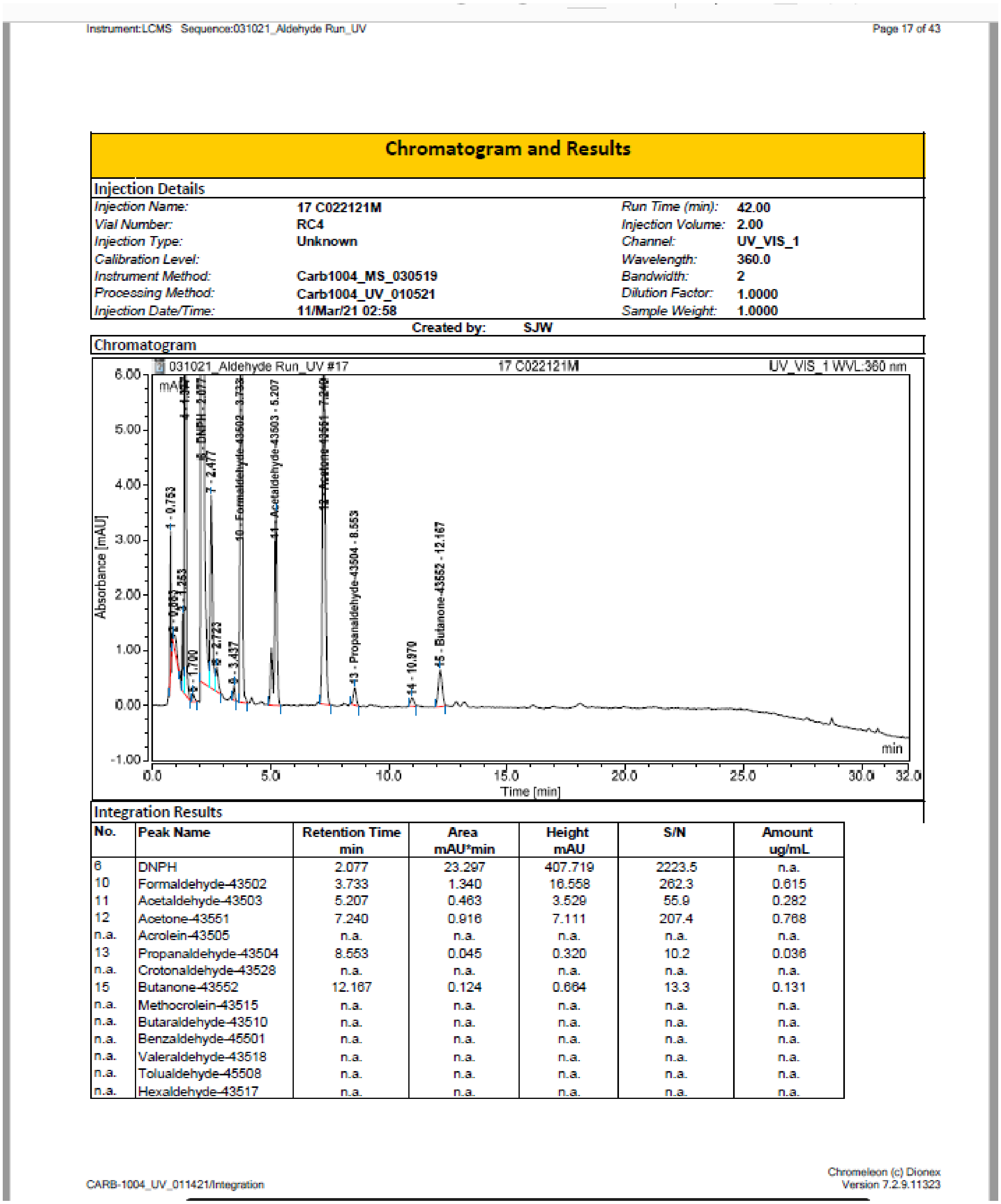
Sequence Details

Name:	**031021_Aldehyde Run_MS**	*Created On:*	**28/Jan/19 13:24:49**
Directory:	**Instrument Data\LCMS\Aldehyde_ 2021**	*Created By:*	**SW/CB**
Data Vault:	**ChromeleonLocal**	*Updated On:*	**16/Mar/21 14:22:41**
No. of Injections:	**43**	*Updated By:*	**CB**

Injection Details

No.	Injection Name	Position	Type	Level	Inject Time	Status
1	1 ACN BLANK	RA1	Unknown		10/Mar/21 13:31:36	Finished
2	2 ACN BLANK	RA1	Unknown		10/Mar/21 14:22:01	Finished
3	3 ACN BLANK	RA1	Unknown		10/Mar/21 15:12:26	Finished
4	4 CAL Level 0	RB1	Calibration Standard	0	10/Mar/21 16:02:51	Finished
5	5 CAL Level 1	RB2	Calibration Standard	1	10/Mar/21 16:53:16	Finished
6	6 CAL Level 2	RB3	Calibration Standard	2	10/Mar/21 17:43:42	Finished
7	7 CAL Level 3	RB4	Calibration Standard	3	10/Mar/21 18:34:07	Finished
8	8 CAL Level 4	RB5	Unknown		10/Mar/21 19:24:32	Finished
9	9 CAL Level 5	RB6	Unknown		10/Mar/21 20:14:58	Finished
10	10 CAL Level 6	RB7	Unknown		10/Mar/21 21:05:23	Finished
11	11 ACN BLANK	RA1	Unknown		10/Mar/21 21:55:48	Finished
12	12 HL QA (0.90ug/mL)_Restek	RA2	Unknown		10/Mar/21 22:46:14	Finished
13	13 LL QA (0.05ug/mL)_Restek	GA1	Unknown		10/Mar/21 23:36:39	Finished
14	14 CART BLK 030521	RC1	Unknown		11/Mar/21 00:27:04	Finished
15	15 C030121 PT BLK	RC2	Unknown		11/Mar/21 01:17:30	Finished
16	16 C030121 PT SPK 54034-60-15	RC3	Unknown		11/Mar/21 02:07:55	Finished
17	17 C022121M	RC4	Unknown		11/Mar/21 02:58:20	Finished
18	18 C022121MD	RC5	Unknown		11/Mar/21 03:48:46	Finished
19	19 C022121C	RC6	Unknown		11/Mar/21 04:39:11	Finished
20	20 C022121CS	RC7	Unknown		11/Mar/21 05:29:37	Finished
21	21 C022721M	RC8	Unknown		11/Mar/21 06:20:02	Finished
22	22 ACN BLANK	RA1	Unknown		11/Mar/21 07:10:27	Finished
23	23 HL QA (0.90ug/mL)_Restek	RA3	Unknown		11/Mar/21 08:00:52	Finished
24	24 LL QA (0.05ug/mL)_Restek	GA2	Unknown		11/Mar/21 08:51:17	Finished
25	25 C022721MD	RD1	Unknown		11/Mar/21 09:41:43	Finished
26	26 C022721C	RD2	Unknown		11/Mar/21 10:32:08	Finished
27	27 CART MDL SPK 030521	RD3	Unknown		11/Mar/21 11:22:33	Finished
28	28 CART SPK 030521	RD4	Unknown		11/Mar/21 12:12:58	Finished
29	29 CART SPK DUP 030521	RD5	Unknown		11/Mar/21 13:03:24	Finished
30	30 C020921CR	RD6	Unknown		11/Mar/21 13:53:49	Finished
31	31 CART BLK 031121	RD7	Unknown		11/Mar/21 14:44:14	Finished
32	32 C030521M	RD8	Unknown		11/Mar/21 15:34:40	Finished
33	33 ACN BLANK	RA1	Unknown		11/Mar/21 16:25:05	Finished
34	34 HL QA (0.90ug/mL)_Restek	RA4	Unknown		11/Mar/21 17:15:31	Finished
35	35 LL QA (0.05ug/mL)_Restek	GA3	Unknown		11/Mar/21 18:05:56	Finished
36	36 C030521MD	RE1	Unknown		11/Mar/21 18:56:21	Finished
37	37 C030521C	RE2	Unknown		11/Mar/21 19:46:47	Finished
38	38 C030521CFb	RE3	Unknown		11/Mar/21 20:37:12	Finished
39	39 CART SPK 031121	RE4	Unknown		11/Mar/21 21:27:38	Finished
40	40 ACN BLANK	RA1	Unknown		11/Mar/21 22:18:03	Finished
41	41 HL QA (0.90ug/mL)_Restek	RA5	Unknown		11/Mar/21 23:08:29	Finished
42	42 LL QA (0.05ug/mL)_Restek	GA4	Unknown		11/Mar/21 23:58:54	Finished
43	43 ACN BLANK	RA1	Unknown		12/Mar/21 00:49:19	Finished

Appendix M – Unedited Chromatogram



Instrument:LCMS   Sequence:031021_Aldehyde Run_UV

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Chromatogram and Results

Injection Details

Injection Name:	**17 C022121M**	*Run Time (min):*	**42.00**
Vial Number:	**RC4**	*Injection Volume:*	**2.00**
Injection Type:	**Unknown**	*Channel:*	**UV_VIS_1**
Calibration Level:		*Wavelength:*	**360.0**
Instrument Method:	**Carb1004_MS_030519**	*Bandwidth:*	**2**
Processing Method:	**Carb1004_UV_010521**	*Dilution Factor:*	**1.0000**
Injection Date/Time:	**11/Mar/21 02:58**	*Sample Weight:*	**1.0000**

Created by: **SJW**

Chromatogram

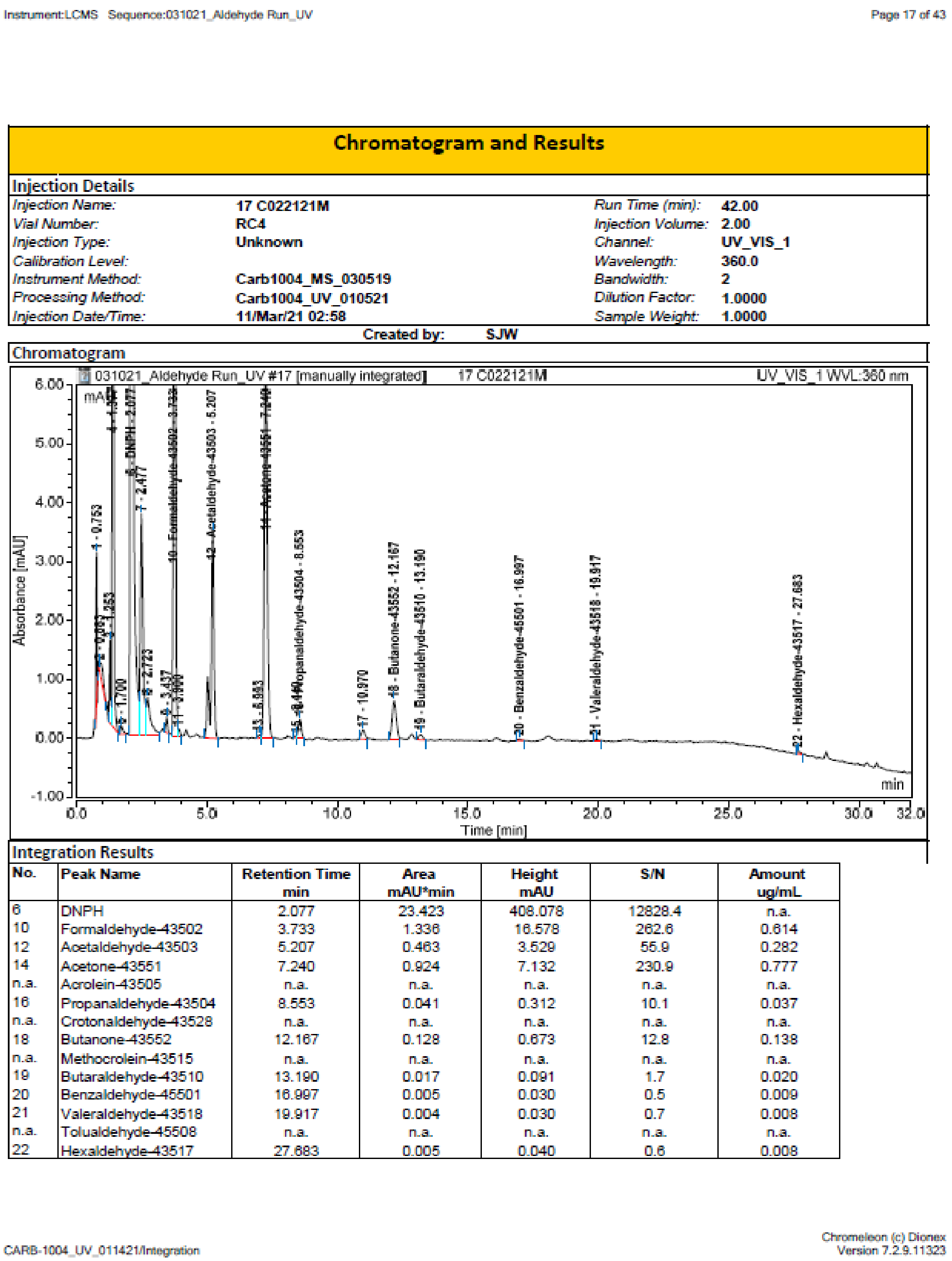
Integration Results

No.	Peak Name	Retention Time min	Area mAU*min	Height mAU	S/N	Amount ug/mL
6	DNPH	2.077	23.297	407.719	2223.5	n.a.
10	Formaldehyde-43502	3.733	1.340	16.558	262.3	0.615
11	Acetaldehyde-43503	5.207	0.463	3.529	55.9	0.282
12	Acetone-43551	7.240	0.916	7.111	207.4	0.768
n.a.	Acrolein-43505	n.a.	n.a.	n.a.	n.a.	n.a.
13	Propanaldehyde-43504	8.553	0.045	0.320	10.2	0.036
n.a.	Crotonaldehyde-43528	n.a.	n.a.	n.a.	n.a.	n.a.
15	Butanone-43552	12.167	0.124	0.664	13.3	0.131
n.a.	Methocrolein-43515	n.a.	n.a.	n.a.	n.a.	n.a.
n.a.	Butaraldehyde-43510	n.a.	n.a.	n.a.	n.a.	n.a.
n.a.	Benzaldehyde-45501	n.a.	n.a.	n.a.	n.a.	n.a.
n.a.	Valeraldehyde-43518	n.a.	n.a.	n.a.	n.a.	n.a.
n.a.	Tolualdehyde-45508	n.a.	n.a.	n.a.	n.a.	n.a.
n.a.	Hexaldehyde-43517	n.a.	n.a.	n.a.	n.a.	n.a.

CARB-1004_UV_011421/Integration

Chromeleon (c) Dionex
Version 7.2.9.11323

Appendix N – Edited Chromatogram



Chromatogram and Results

Injection Details

Injection Name:	**17 C022121M**	*Run Time (min):*	**42.00**
Vial Number:	**RC4**	*Injection Volume:*	**2.00**
Injection Type:	**Unknown**	*Channel:*	**UV_VIS_1**
Calibration Level:		*Wavelength:*	**360.0**
Instrument Method:	**Carb1004_MS_030519**	*Bandwidth:*	**2**
Processing Method:	**Carb1004_UV_010521**	*Dilution Factor:*	**1.0000**
Injection Date/Time:	**11/Mar/21 02:58**	*Sample Weight:*	**1.0000**

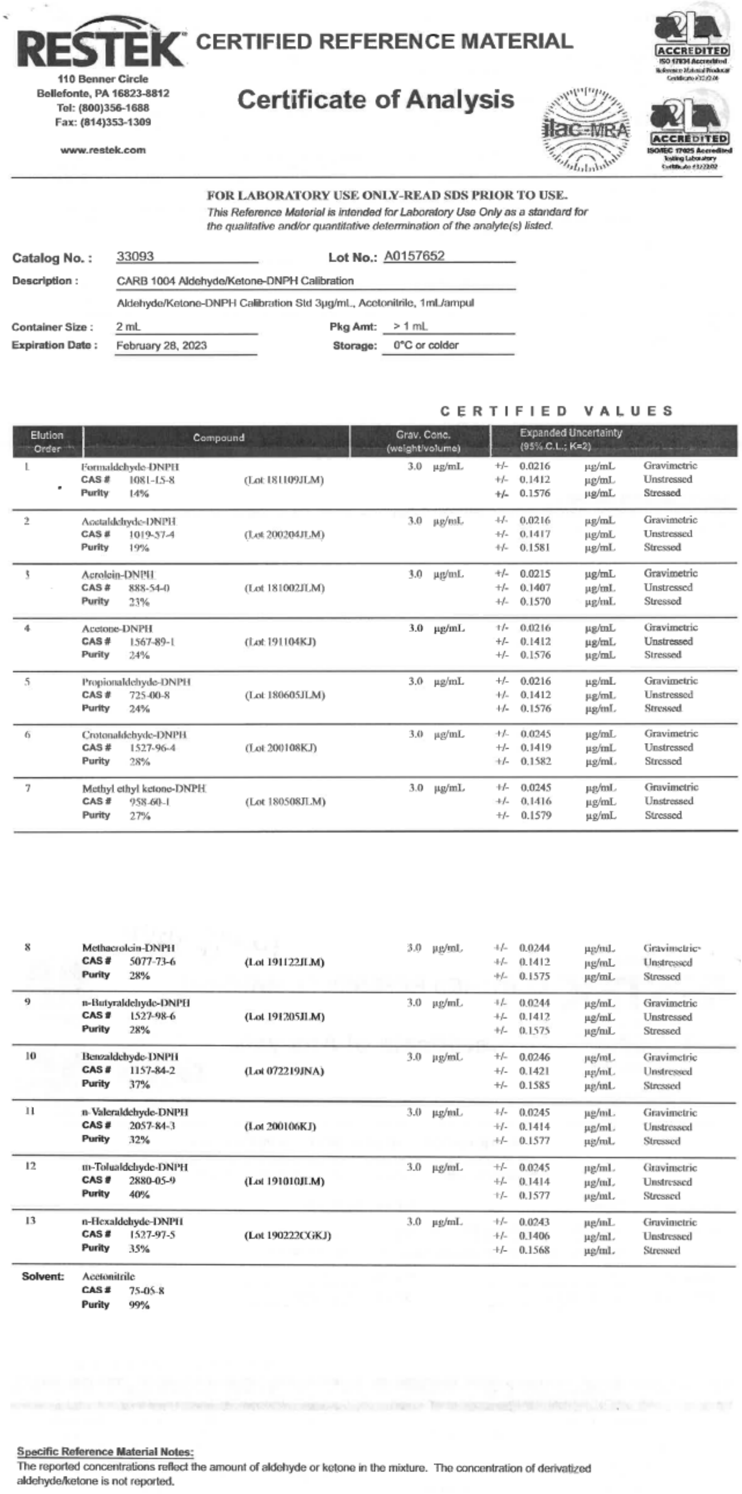
Created by: **SJW**

Chromatogram

Integration Results

No.	Peak Name	Retention Time min	Area mAU*min	Height mAU	S/N	Amount ug/mL
6	DNPH	2.077	23.423	408.078	12828.4	n.a.
10	Formaldehyde-43502	3.733	1.336	16.578	262.6	0.614
12	Acetaldehyde-43503	5.207	0.463	3.529	55.9	0.282
14	Acetone-43551	7.240	0.924	7.132	230.9	0.777
n.a.	Acrolein-43505	n.a.	n.a.	n.a.	n.a.	n.a.
16	Propanaldehyde-43504	8.553	0.041	0.312	10.1	0.037
n.a.	Crotonaldehyde-43528	n.a.	n.a.	n.a.	n.a.	n.a.
18	Butanone-43552	12.167	0.128	0.673	12.8	0.138
n.a.	Methocrolein-43515	n.a.	n.a.	n.a.	n.a.	n.a.
19	Butaraldehyde-43510	13.190	0.017	0.091	1.7	0.020
20	Benzaldehyde-45501	16.997	0.005	0.030	0.5	0.009
21	Valeraldehyde-43518	19.917	0.004	0.030	0.7	0.008
n.a.	Tolualdehyde-45508	n.a.	n.a.	n.a.	n.a.	n.a.
22	Hexaldehyde-43517	27.683	0.005	0.040	0.6	0.008

Appendix O – Example Certificate of Analysis for Carbonyl Standard



RESTEK **CERTIFIED REFERENCE MATERIAL**

110 Benner Circle
Bellefonte, PA 16823-8812
Tel: (800)356-1688
Fax: (814)353-1309

www.restek.com

Certificate of Analysis

ACCREDITED
ISO 17034 Accredited
Reference Material Producer

ilac-MRA

ACCREDITED
ISO/IEC 17025 Accredited
Testing Laboratory

FOR LABORATORY USE ONLY-READ SDS PRIOR TO USE.
This Reference Material is intended for Laboratory Use Only as a standard for the qualitative and/or quantitative determination of the analyte(s) listed.

Catalog No. :	33093	**Lot No.:**	A0157652
Description :	CARB 1004 Aldehyde/Ketone-DNPH Calibration Aldehyde/Ketone-DNPH Calibration Std 3µg/mL, Acetonitrile, 1mL/ampul		
Container Size :	2 mL	**Pkg Amt:**	> 1 mL
Expiration Date :	February 28, 2023	**Storage:**	0°C or colder

CERTIFIED VALUES

Elution Order	Compound		Grav. Conc. (weight/volume)	Expanded Uncertainty (95% C.L.; K=2)
1	Formaldehyde-DNPH **CAS #** 1081-15-8 **Purity** 14%	(Lot 181109JLM)	3.0 µg/mL	+/- 0.0216 µg/mL Gravimetric +/- 0.1412 µg/mL Unstressed +/- 0.1576 µg/mL Stressed
2	Acetaldehyde-DNPH **CAS #** 1019-57-4 **Purity** 19%	(Lot 200204JLM)	3.0 µg/mL	+/- 0.0216 µg/mL Gravimetric +/- 0.1417 µg/mL Unstressed +/- 0.1581 µg/mL Stressed
3	Acrolein-DNPH **CAS #** 888-54-0 **Purity** 23%	(Lot 181002JLM)	3.0 µg/mL	+/- 0.0215 µg/mL Gravimetric +/- 0.1407 µg/mL Unstressed +/- 0.1570 µg/mL Stressed
4	Acetone-DNPH **CAS #** 1567-89-1 **Purity** 24%	(Lot 191104KJ)	3.0 µg/mL	+/- 0.0216 µg/mL Gravimetric +/- 0.1412 µg/mL Unstressed +/- 0.1576 µg/mL Stressed
5	Propionaldehyde-DNPH **CAS #** 725-00-8 **Purity** 24%	(Lot 180605JLM)	3.0 µg/mL	+/- 0.0216 µg/mL Gravimetric +/- 0.1412 µg/mL Unstressed +/- 0.1576 µg/mL Stressed
6	Crotonaldehyde-DNPH **CAS #** 1527-96-4 **Purity** 28%	(Lot 200108KJ)	3.0 µg/mL	+/- 0.0245 µg/mL Gravimetric +/- 0.1419 µg/mL Unstressed +/- 0.1582 µg/mL Stressed
7	Methyl ethyl ketone-DNPH **CAS #** 958-60-1 **Purity** 27%	(Lot 180508JLM)	3.0 µg/mL	+/- 0.0245 µg/mL Gravimetric +/- 0.1416 µg/mL Unstressed +/- 0.1579 µg/mL Stressed
8	Methacrolein-DNPH **CAS #** 5077-73-6 **Purity** 28%	(Lot 191122JLM)	3.0 µg/mL	+/- 0.0244 µg/mL Gravimetric +/- 0.1412 µg/mL Unstressed +/- 0.1575 µg/mL Stressed
9	n-Butyraldehyde-DNPH **CAS #** 1527-98-6 **Purity** 28%	(Lot 191205JLM)	3.0 µg/mL	+/- 0.0244 µg/mL Gravimetric +/- 0.1412 µg/mL Unstressed +/- 0.1575 µg/mL Stressed
10	Benzaldehyde-DNPH **CAS #** 1157-84-2 **Purity** 37%	(Lot 072219JNA)	3.0 µg/mL	+/- 0.0246 µg/mL Gravimetric +/- 0.1421 µg/mL Unstressed +/- 0.1585 µg/mL Stressed
11	n-Valeraldehyde-DNPH **CAS #** 2057-84-3 **Purity** 32%	(Lot 200106KJ)	3.0 µg/mL	+/- 0.0245 µg/mL Gravimetric +/- 0.1414 µg/mL Unstressed +/- 0.1577 µg/mL Stressed
12	m-Tolualdehyde-DNPH **CAS #** 2880-05-9 **Purity** 40%	(Lot 191010JLM)	3.0 µg/mL	+/- 0.0245 µg/mL Gravimetric +/- 0.1414 µg/mL Unstressed +/- 0.1577 µg/mL Stressed
13	n-Hexaldehyde-DNPH **CAS #** 1527-97-5 **Purity** 35%	(Lot 190222CGKJ)	3.0 µg/mL	+/- 0.0243 µg/mL Gravimetric +/- 0.1406 µg/mL Unstressed +/- 0.1568 µg/mL Stressed
Solvent:	Acetonitrile **CAS #** 75-05-8 **Purity** 99%			

Specific Reference Material Notes:
The reported concentrations reflect the amount of aldehyde or ketone in the mixture. The concentration of derivatized aldehyde/ketone is not reported.