GC Best Practices & Troubleshooting
Troubleshooting Tips

1. Isolate the problem.
   (Blank Runs, Inject Un-retained Compound, Know what it is not)

2. Change only one variable at a time.

3. Compare before/after chromatograms.
   (Peak shape, response, retention, baseline rise, background, look for trends, etc.)
4. Make sure it makes sense to do what you’re doing…
5. Be careful of distractions
6. Sometimes just a fresh set of eyes is all that is needed.

“Well, here’s your problem, Mr. Schueler.”
What Can Possibly Go Wrong?

INJECTOR – contamination, flow settings, flow path issues, overload, valve settings, faulty consumables

COLUMN – contamination, flow settings, flow path issues, damage (activity & bleed), breakage

DETECTOR – contamination, flow settings, flow path issues, electronics
Logical Troubleshooting

- Troubleshooting Starts with Isolating the problem
  - There are 5 basic areas from where the problem arises
    - FLOW
    - INJECTOR
    - COLUMN
    - DETECTOR
    - ELECTRONICS (Temperature)
  - But of course it can always be some COMBINATION

- Knowing what can & can’t cause the symptom is the key
Typical Gas Chromatographic System

Selection of type and velocity influences efficiency and retention time

Cylinders or Generators
van Deemter Curves

\[ \bar{u} \text{ (cm/sec)} \]

\[ h \]

\[ \text{He} \]

\[ \text{N}_2 \]

\[ \text{H}_2 \]
# CARRIER GAS

<table>
<thead>
<tr>
<th>Type</th>
<th>Velocity Range ($u_{opt} - \text{OPGV}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrogen</td>
<td>8-16</td>
</tr>
<tr>
<td>Helium</td>
<td>20-40</td>
</tr>
<tr>
<td>Hydrogen</td>
<td>30-55</td>
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Gas Clean Filters
Typical Gas Chromatographic System

Have to be able to get things to their gas state

Key to starting the Chromatographic process right.

Cylinders or Generators

Air

Hydrogen

Carrier Gas

Mol-Sieve Traps

Fixed Restrictors

Regulators

Flow Controller

Injection Port

Detector

Column

Electrometer

Recorder/Integrator
Influence of Injection Efficiency

Short Concentrated

Long Diffuse

Solute Bands

Same column, same chromatographic conditions
Split Injector
Flow Path

Carrier gas source

Septum purge

Split vent
Splitless Injector
Purge Off At Injection

Carrier gas source

Septum purge

Split vent

Flow through injector = Column flow only
**Splitless Injector**

**Purge On After Injection**

Flow through injector = Column flow + Split Vent Flow
Split Injector
Major Variables

Split ratio - determines amount of sample onto column and efficiency of injection (sensitivity vs peak shape)

Liner - influences efficiency of vaporization/discrimination

Temperature - hot enough to vaporize sample without degradation or causing backflash

Injection volume - typically 1-3uL, increasing it does not have as much of an effect as one might think
Split Liners – What’s What?

- Straight tube
- Straight tube with glass wool
- Fixed glass wool
- Inverted cup
- Baffle
Split Liner

Packed with Glass Wool

Without Glass Wool Packing

\[ \frac{C_{10}^{40}}{C_{10}^{10}} = 0.64 \]

\[ \frac{C_{10}^{40}}{C_{10}^{10}} = 0.37 \]
Split Liner

Packed with Glass Wool

Peak Area Ratio
\[ \frac{n-C_{40}}{n-C_{10}} = 0.64 \]

Without Glass Wool Packing

Peak Area Ratio
\[ \frac{n-C_{40}}{n-C_{10}} = 0.37 \]
Splitless Injector
Overview

Most of the sample is introduced into the column

Used for low concentration samples

Wider peaks are obtained than for split injections
Splitless Injector
Major Variables

Purge activation time - determines amount of sample onto column and efficiency of injection (sensitivity vs peak shape)

Liner - preventing backflash more critical than vaporization properties (double tapered type recommended)

Injection volume - typically 1uL or less (backflash)

Temperature – long residence times allow for lower temps
Splitless Injector
Liners

- Straight tube
- Bottom restriction
- Dual restriction
Solvent Vapor Volume Calculator

![Vapor Volume Calculator interface with two examples: one for Acetone and one for Water. Each example shows the solvent properties, injection volume, estimated volume, and percentage capacity. The interface includes options for adding, removing, and setting defaults for solvents and injection liners.]
Backflash (carry-over) can give false positives!
Cleaning the Split/Splitless Injector

1. Carrier gas flow off
2. Disconnect split vent line
3. Replace split vent trap
4. Remove column, reducing nut, gold seal, washer and liner

- MeOH
- MeCl₂
- Ace tone

GC Off
Finding the Split Vent Trap

Follow the split vent line back to the EPC
Finding the Split Vent Trap

Remove cover at Split Vent
Replacing the Split Vent Trap

Finger Tight Knurled Nut

G1544-80530
Split Vent Trap Changed (Column Bleed?!?)

Before

After
Split/Splitless Pulsed Injection

Pressure Pulse contains sample expansion and transfers analytes to the column faster.

**Pulsed Split**
- the most volatile components and solvent effected most
- faster sample transfer not as critical since it’s already fast

**Pulsed Splitless**
- sample containment more critical than in split injection
- much sharper peaks than in traditional splitless injection
Select Pulsed Splitless Mode in Inlets
Check the Splitless Pressure
Double or Triple the Pressure for ~1 sec less than the Purge Activation Time
The BIGGEST Problem in GC is…

There are more things that DON’T go through a GC than DO!

….therefore, don’t inject anything and you’ll never have problems.

OK, inject, but realize that everything just got dirty…deal with it!
Where Does it Get Dirty?
What Are You Doing!?
Bonus Peaks or Ghost Peaks

Before

Contamination in INJECTOR or FLOW (carrier gas)
- Contaminated consumables
- Carryover from a backflash or previous sample
- Bad tank of gas or traps have expired
- Septum bleed

*TIP = Run a blank run… it should be blank!

After
Bonus Peaks - Ferrule Contamination

![Graph showing Ferrule Contamination with Agilent and Vespel/Graphite Ferrule from a bag highlighted.]

- **Agilent**
- **Vespel/Graphite Ferrule from a bag**
More “Off-Brand” O-Ring Issues
Controlled Substances Analysis, H2 Carrier

Residue on top of inlet weldment

Problem Resolution:
Agilent Non-Stick Liner O-Ring
p/n 5188-5365, 10PK
Septa Bleed vs Column Bleed (MSD)

Source of peaks from outside of the column

Columns: HP 5MS
30mx0.25mmx0.25um

Oven: 80 to 160C at 25 C/min,
160 to 320 C at 3 C/min(4),
320 to 325 C at 20C/min(4)

Injection: split 100:1; 1ul of 100ng/ul

Detector: MSD (HP-5973)
run at max sensitivity- full scan

Peak height approx. equiv. to 5 ppb of PAH
(actually impurities)
Septa Bleed vs Column Bleed (MSD)

Source of peaks from outside of the column *ELIMINATED*

Columns: HP 5 MS
30mx0.25mmx0.25um

Oven: 
80 to 160 C at 25 C/min,
160 to 320 C at 3 C/min (4)
320 to 325 C at 20C/min (4)

Injection: split 100:1; 1ul of 100ng/ul

Detector: MSD (HP-5973)
run at max sensitivity- full scan

**New Septa Installed**
HP Advanced Green Septa
P/N 5183-4594 (11 mm)
Peak Tailing

**Before**

INJECTOR or COLUMN is Active
- Reversible adsorption of active compounds
  (-OH, -NH, -SH)

FLOW problem
- dead volume, obstruction, poor installation, or severe column contamination

Miscellaneous – temperature issues for late eluters, overloading of PLOT columns, co-elution, polarity mismatch between phase, solute or solvent, and some compounds always tail

*Tip = Inject a light hydrocarbon, should not tail unless flow path problem.

**After**
A flat, 90° square cut will be optimal for all connections
Symptom – Tailing of Active Compounds

Sample: 0.5 ng on column loading with ISTD
Column: 20m 0.18mm 0.18µm
Carrier: Helium 37cm/sec, Ramped flow; 0.7ml/min (0.1min) to 1.3ml/min (15ml/min²)
Oven: 35°C (2.5 min) to 80°C (40°C/min), 15°C/min to 200°C, 8°C/min to 275°C (2 min)
Injection: 0.5µl, Splitless, 280°C, purge flow 30ml/min at 0.75 min
MSD: Transfer Line 290°C, Source 300°C, Quad 180°C

1. n-Nitrosodimethylamine
2. Aniline
3. 1,4-Dichlorobenzene-d4
4. Benzoic Acid
5. Naphthalene-d8
6. Acenaphthene-d10
7. 2,4-Dinitrophenol
8. 4-Nitrophenol
9. 2-Me-4,6-dinitrophenol
10. 4-Aminobiphenyl
11. Pentachlorophenol
12. Phenanthrene-d10
13. Benzidine
14. Chrysene-d12
15. 3,3’-Dichlorobenzidine
16. Benzo[b]fluoranthene
17. Benzo[k]fluoranthene
18. Perylene-d12
Solution – Ultra Inert GC Column

- n-Nitrosodimethylamine
- Aniline
- 1,4-Dichlorobenzene-d₄
- Benzoic Acid
- Naphthalene-d₈
- Acenaphthene-d₁₀
- 2,4-Dinitrophenol
- 4-Nitrophenol
- 2-Me-4,6-dinitrophenol
- 4-Aminobiphenyl
- Pentachlorophenol
- Phenanthrene-d₁₀
- Benzidine
- Chrysene-d₁₂
- 3,3’-Dichlorobenzidine
- Benzo[b]fluoranthene
- Benzo[k]fluoranthene
- Perylene-d₁₂

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Improved Performance
Improving the Entire Flowpath

UltiMetal Inlet Weldment, Shell and Transfer Lines

Ultra Inert Inlet Liner

Ultra Inert Gold Seal

UltiMetal Capillary Flow Technology Devices, Ultimate Union

New UltiMetal FlexiMetal Ferrules

Ultra Inert GC Column

UltiMetal – TCD, FPD, NPD/FID Jets

...now from a single supplier
Split Peaks

- Injector (poor sample introduction)
  - Injecting the sample twice (some how?)
  - Mixed sample solvent (polarity difference)
  - Sample in syringe needle (manual inject)

- Injector (activity)
  - Breakdown (not really a split peak, 2 peaks)
  - Sample degradation in injector

VOLATILITY
- High boilers dropping out on Cold Spots
  - Transfer line temps
  - Unions or fittings not tracking column temp
Broad Peaks

INJECTOR
- Poor Installation
- Change in settings (temps/flows)
- Poor sample focusing
- Large change in sample concentration

FLOW
- Change in gas velocity
- Constant Flow vs Constant Pressure

COLUMN
- Contamination
- Damaged/old stationary phase
- Reverse Solvent Effect
No Peaks

DETECTOR (not on or not operational)

INJECTOR (not working)

- Plugged syringe/plunger not moving

- Wrong injector (or detector)

- Huge leak/no carrier gas flow (older systems)

NOT the COLUMN Unless…

- Broken column

- No column
Symptom – No Peaks
Solution - Unplugged Syringe
Peak Response
All Change in Size

**INJECTOR**
- Leaky syringe
- Split ratio set incorrectly
- Wrong purge activation time
- Septum purge flow too high
- Injector temperature too low*

**DETECTOR (response problem)**
- Settings or flows changed
- Electronics failing

*Tip = Ask is it all of them or some of them, if all then injector or detector
Peak Response
Some Change in Size

INJECTOR or COLUMN is active/contaminated
- Irreversible adsorption of active compounds (-OH, -NH, -SH)
- Decomposition of sample
- Temperature Change – Discrimination
- Evaporation from sample

*Tip = If only some change, then ask which ones? If active compounds then activity. If tracks volatility then cold spots or inlet discrimination.
Typical Gas Chromatographic System

Picking the appropriate stationary phase and optimum dimensions for the column will give the greatest resolution in the shortest analysis time.
Peak Fronting
Shark Fin Shaped or Just Slight

COLUMN (contaminated)
- Overload (More pronounced with large solute and phase polarity differences)

INJECTOR
- Poor efficiency (flow/temp)
- Column installation
- Compound very soluble in injection solvent (need retention gap)
- Mixed sample solvent

OTHER
- Co-elution
- Breakdown
Retention Time Shift

INJECTOR
- Change in injection solvent
- Large change in sample concentration

FLOW
- Leak in the septum
- Change in gas velocity

COLUMN
- Contamination
- Damaged stationary phase
- Loss of stationary phase
- Change in temperature
Under constant pressure conditions, flow decreases as temperature increases. (viscosity of a gas increases as temperature increases)
Gas Viscosity vs Temperature

J.V. Hinshaw, Column Connections, LCGC Asia Pacific, 12(2), 1100 (2009).
Loss of Resolution

Resolution is a function of separation and peak width.
Loss of Resolution - Separation Decrease

COLUMN
- Different column temperature
- Contamination (more phase?)
- Matrix components co-eluting
- Different column phase?
Loss of Resolution - Peak Broadening

FLOW
- Change in carrier gas velocity
- Make-up gas

COLUMN
- Contamination
- Phase degradation

INJECTOR (efficiency)
- Settings, Liner, Installation, etc.

- Peak Tailing – Flow Path or Activity
- Bonus Peaks – In Sample or Back Flash (Carry Over)
- Split Peaks – Injector Problems, Mixed Solvent
- No Peaks – Wasn’t Introduced, Wasn’t Detected
- Response Changes – Activity, Injector Discrimination, Detector Problem
- Peak Fronting – Overload or Solubility Mismatch, Injector Problems
- Shifting Retention – Leaks, Column Aging, Contamination or Damage
- Loss of Resolution – Separation Decreasing, Peak Broadening
- Baseline Disturbances – Column Bleed, Contamination, Electronics
- Noisy or Spiking Baseline – Electronics or Contaminated Detector
- Quantitation Problems – Activity, Injector or Detector Problems
Quantitation Problems

DETECTOR
- Poor stability (electronics) or Baseline disturbances (contamination)
- Outside detector's linear range or wrong settings

Activity (adsorption) in INJECTOR or COLUMN

OTHER

INJECTOR
- Technique, settings, conditions
- Syringe worn

- Co-elution
- Matrix effects
- Sample evaporation – leaky vials
- Sample decomposition
Baseline Disturbances
Sudden Changes, Wandering, or Drifting

WANDER

COLUMN or DETECTOR
- Not fully conditioned or stabilized (electronics)
- Contamination

FLOW
- Changes in carrier and/or detector gas flows
- Valves switching, leaks

DRIFT
Noisy Baseline

**FLOW**
- Contaminated gas
- Incorrect detector settings

**COLUMN**
- Bleed if at high temperature
- In detector flame (poor installation)

**DETECTOR**
- Air leak - ECD, TCD
- Electronics malfunction
Spiking Baseline

DETECTOR

- Particles entering the detector
- Random: poor connection
- Regular: nearby "cycling" equipment (electronics)
Remember

Complete system = Carrier Gas + Injector + Column + Detector + Data System

Multiple cause and effect

Do not change too many variables at once
Self-Tightening Column Nuts
Innovative spring-driven piston continuously presses against ferrule

• **Less wasted time:** No retightening needed after repeated thermal cycles

• **Ease of use:** Finger-tight, consistent connections *without tools*

• **Leak Free = Lower column bleed:** Longer column life

Video at [agilent.com/chem/STnutvideo](http://agilent.com/chem/STnutvideo)
Self-Tightening Column Nuts

Standard column nuts new fitting

Slight baseline rise due to air leak

Standard column nuts after 25 injections

Dramatic baseline rise due to air leak

Agilent Self Tightening Column Nuts after 400 injections

- No baseline rise
- No air leak
- No retightening
Changes in Column Dimensions, Gas Type or Velocity Require Changes in Temp Program Rates

Method Translation Software to the Rescue!
Troubleshooting Resources

Online Troubleshooting and Maintenance Videos


GC Troubleshooting Guide


Method Translation Software

Agilent Better GC Connections

www.agilent.com/chem/betterGCconnections

Order the poster…

View the video…
1 (214) 883-2260 (Eric)

E-mail:

gc-column-support@agilent.com

Eric.pavlich@agilent.com