Water Injections in Capillary GC

“How Wet Can You Get?”

Why Inject Water?

- Convenient
  - Aqueous samples (waste water, drinking water, etc.)
  - Biological samples
- Necessary
  - Purge & Trap/Static Headspace

Potential Problems with Water

- Injector Issues
  - Large expansion volume
  - Backflash

Potential Problems with Water

- Detector Issues
  - Extinguish FID flame
  - Decrease sensitivity of ECD
  - “Wet” MSD

Potential Problems with Water

- Solvent - stationary phase mismatch
  - poor wettability of many stationary phases by water
  - “puddles”

Potential Problems with Water

- Damage to stationary phase
  - Change in Retention Times
  - Change in Selectivity
  - Increase in Bleed

What is Normal Column Bleed?

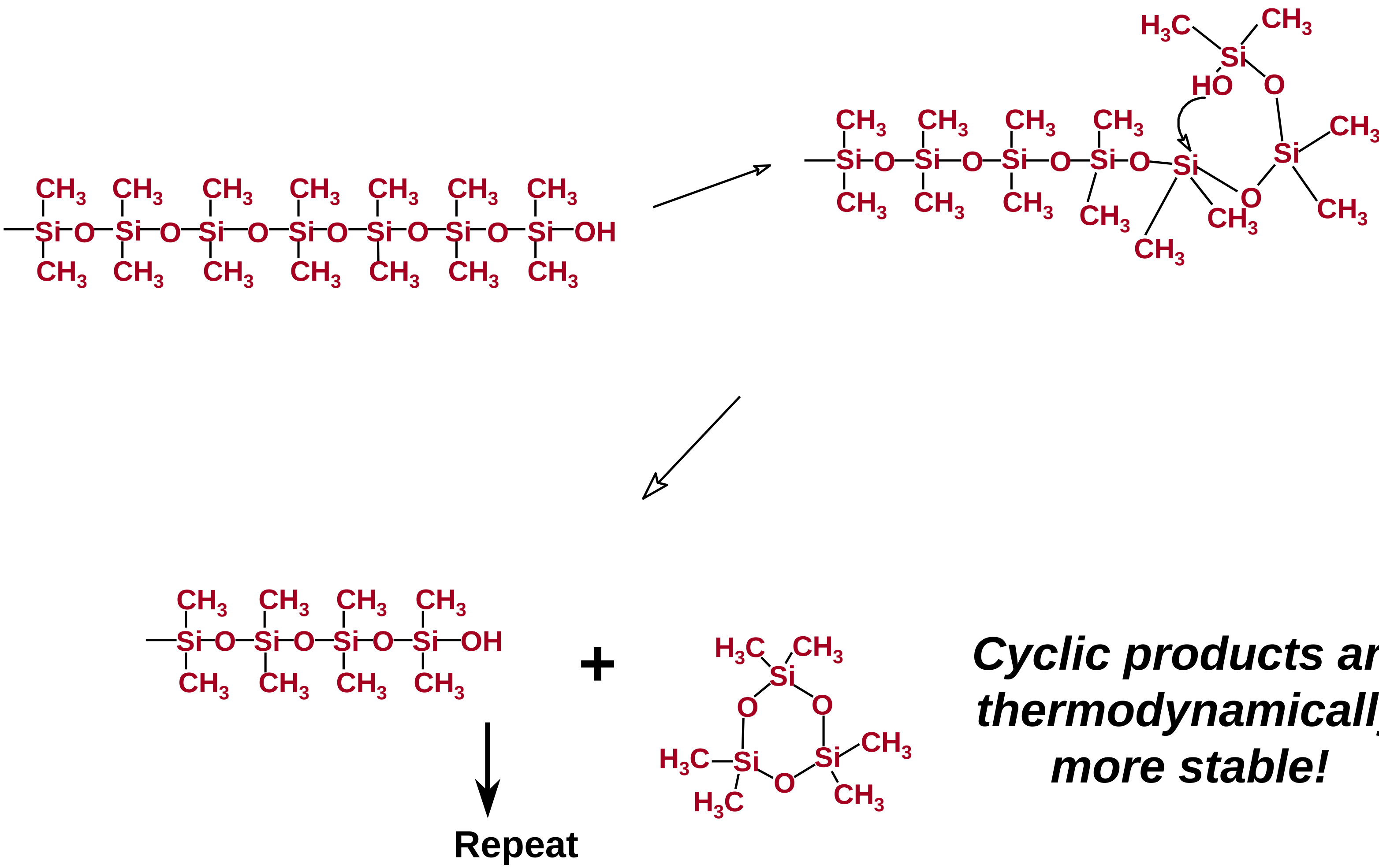
Normal background signal generated by the elution of normal degradation products of the column stationary phase

What is Bleed?

- Thermodynamic equilibrium process occurs to some degree in all columns
- Polysiloxane backbone releases low molecular weight, cyclic fragments
- Occurs at low level in low temperature, O_2-free, clean system
- Increased by increased temperature, oxygen exposure or chemical contamination

Bleed: Why Does It Happen?

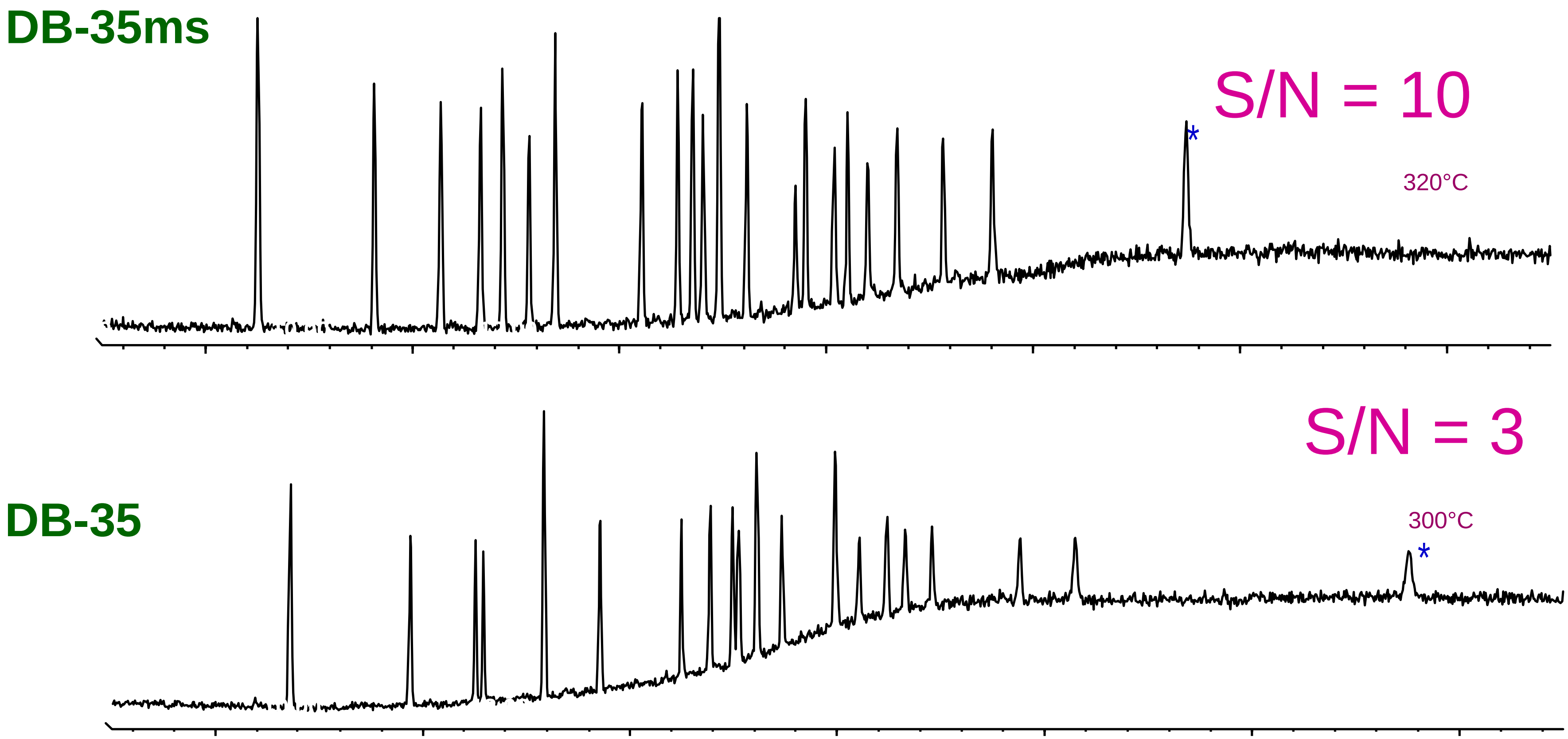
"Back Biting" Mechanism of Product Formation



Cyclic products are thermodynamically more stable!

Low Bleed Stationary Phases

DB-35ms vs. DB-35



* 5ng Decachlorobiphenyl

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Dial 1-816-650-0621 for e-Seminar Audio

Low Bleed Stationary Phases

Spectral Purity

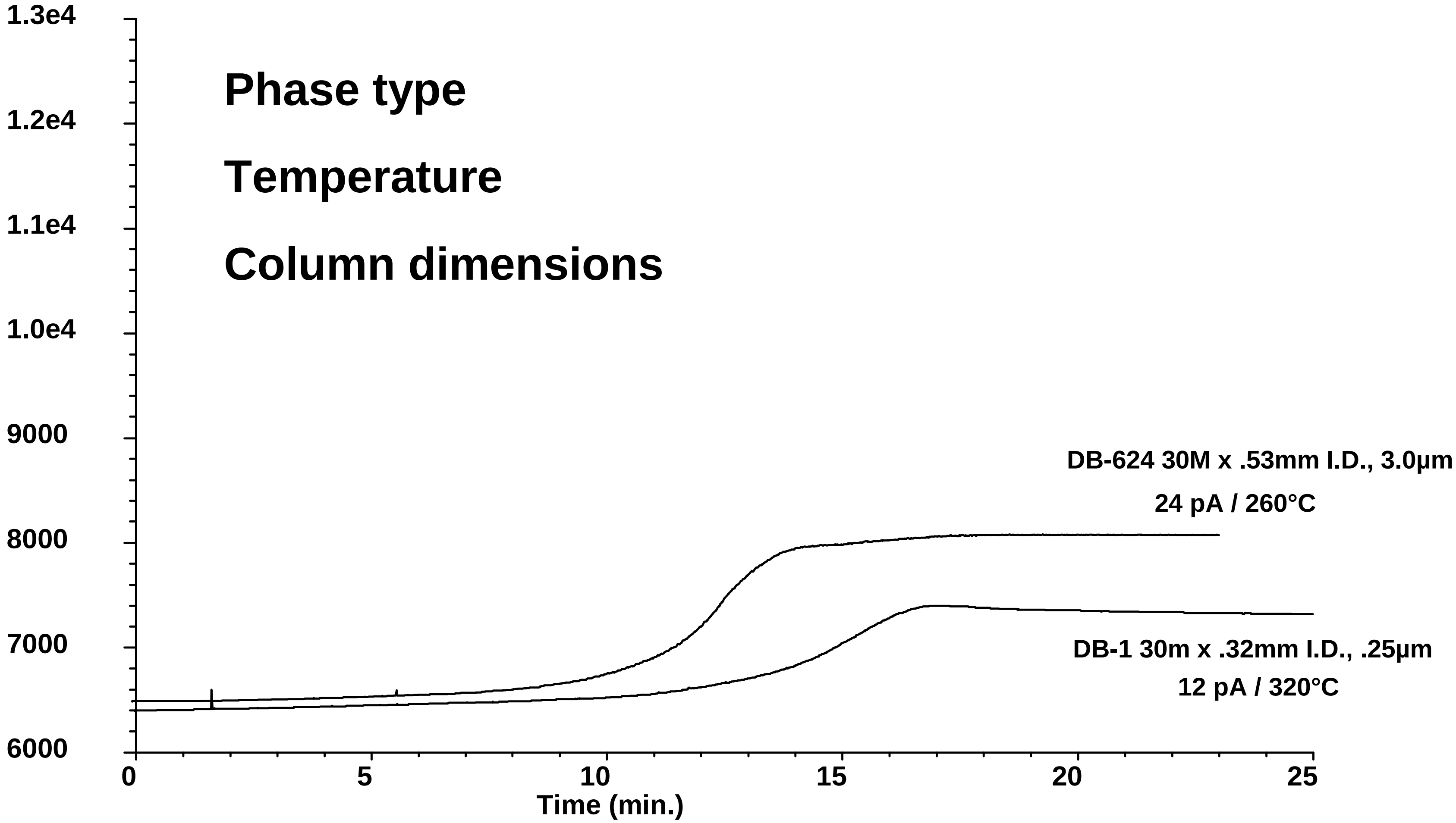
DB-35ms

Primary Ion

DB-35

Primary Ion

Column Bleed is Influenced by:



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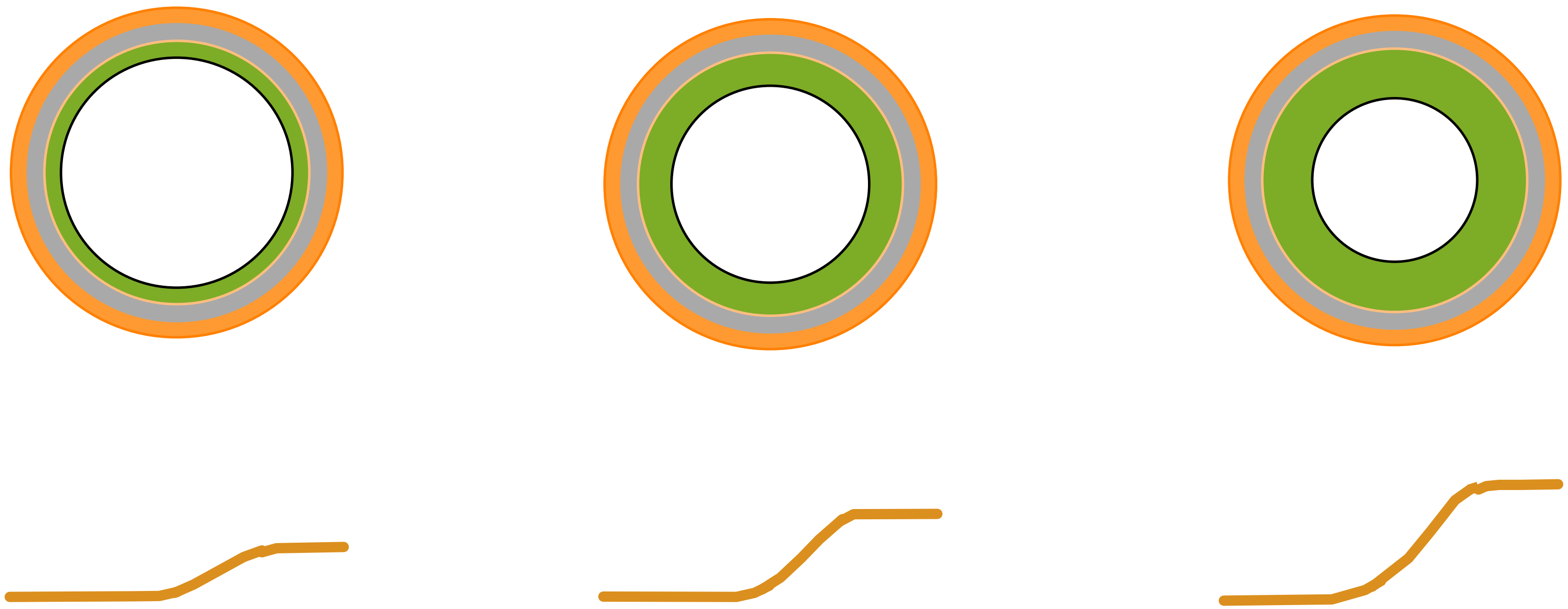
FILM THICKNESS
Bleed

More stationary phase = More degradation products



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What is a Bleed Problem?

An abnormal elevated baseline at high temperature

IT IS NOT

A high baseline at low temperature

Wandering or drifting baseline at any temperature

Discrete peaks

Break

For Questions and Answers

Press 1 on Your Phone to

Ask a Question

Goals of Water Injection Study

Who cares?

- Frequent Inquiries
- Water does cause problems (but what?)
- Quantitative data
- Establish “guidelines”

STATIONARY PHASES

- Dimethylpolysiloxane (DB-1)
- Polyethylene glycol (DB-Wax)
- Cyanopropylphenyl (DB-225)
- Cyclodextrin (CycloSil B)
- Divinylbenzene/ethylene glycol dimethacrylate (HP-PLOT U)

Experimental Conditions

- Instrument: Agilent 6890 with Autoinjector
- Injector: 250^{o} C, *1:5 split*
- Injection volume: *1 μL*
- Detector: FID, 300^{o} C

Experimental Conditions

- Oven: *130º C, 60º* C, or *200º* C* isothermal
- Column Dimensions: *30 m x 0.53 mm I.D. x 1.0 μm*#
- *1,000* water injections at *each* temperature on each column
- Bleed profile after 250, 500, and 1,000 water injections
- Test mix after 250, 500, and 1,000 water injections

*for Cyclosil B and DB-Wax only

#CycloSil B: 30 m x 0.32 mm x 0.25 *μm*

Test Mixes

Compound List for Chromatograms DB-1

Peak	Compound
1	2-Chlorophenol (skew)
2	Undecane
3	2,4-Dimethylaniline (skew)
4	<u>1-Undecanol (RI 1, skew)</u>
5	Tetradecane
6	<u>Acenaphthalene (RI 2)</u>
7	*Pentadecane (k, N)*

Test Mixes

Compound List for Chromatograms DB-Wax

Peak	Compound
1	2-Octanone
2	Tetradecane
3	1-Octanol
4	Methyl deconate
5	Methyl undecanoate
6	<u>Naphthalene (RI 1)</u>
7	<u>1-Decanol (RI 2, skew)</u>
8	*Methyl dodecanoate (k, N)*
9	2,6-Dimethylaniline (skew)
10	2,6-Dimethylphenol (skew)

Test Mixes

Compound List for Chromatograms DB-225

Peak	Compound
1	Tetradecane
2	2-Chlorophenol (skew)
3	Hexadecane
4	<u>Naphthalene (RI 1)</u>
5	2,4-Dimethylaniline (skew)
6	<u>1-Undecanol (RI 2, skew)</u>
7	Octadecane
8	*Ethyl dodecanoate (k, N)*

Test Mixes

Compound List for Chromatograms CycloSil B

Peak	Compound
1	Dodecane
2	(R)-Linalool (RI 1)
3	(S)-Linalool (RI 2)
4	naphthalene
5	1-nonanol
6	gamma-heptalactone
7	gamma-heptalactone
8	*methyl decanoate*
9	Tetradecane (k, N)

Test Mixes

Compound List for Chromatograms HP- PLOT U

Peak	Compound
1	Methanol
2	Ethanol
3	Pentane
4	Ethyl ether
5	<u>Acetone (RI 1)</u>
6	Hexane (k, N)
7	*Ethyl acetate*

Results DB-1

Parameter	Before Inj.	After Inj.
Ret. Factor	14.6	14.5
Ret. Index 1	1349.88	1350.02
Ret. Index 2	1427.77	1428.16
Theor. plates	1448	1474
Bleed (pA)	12.8	11.2

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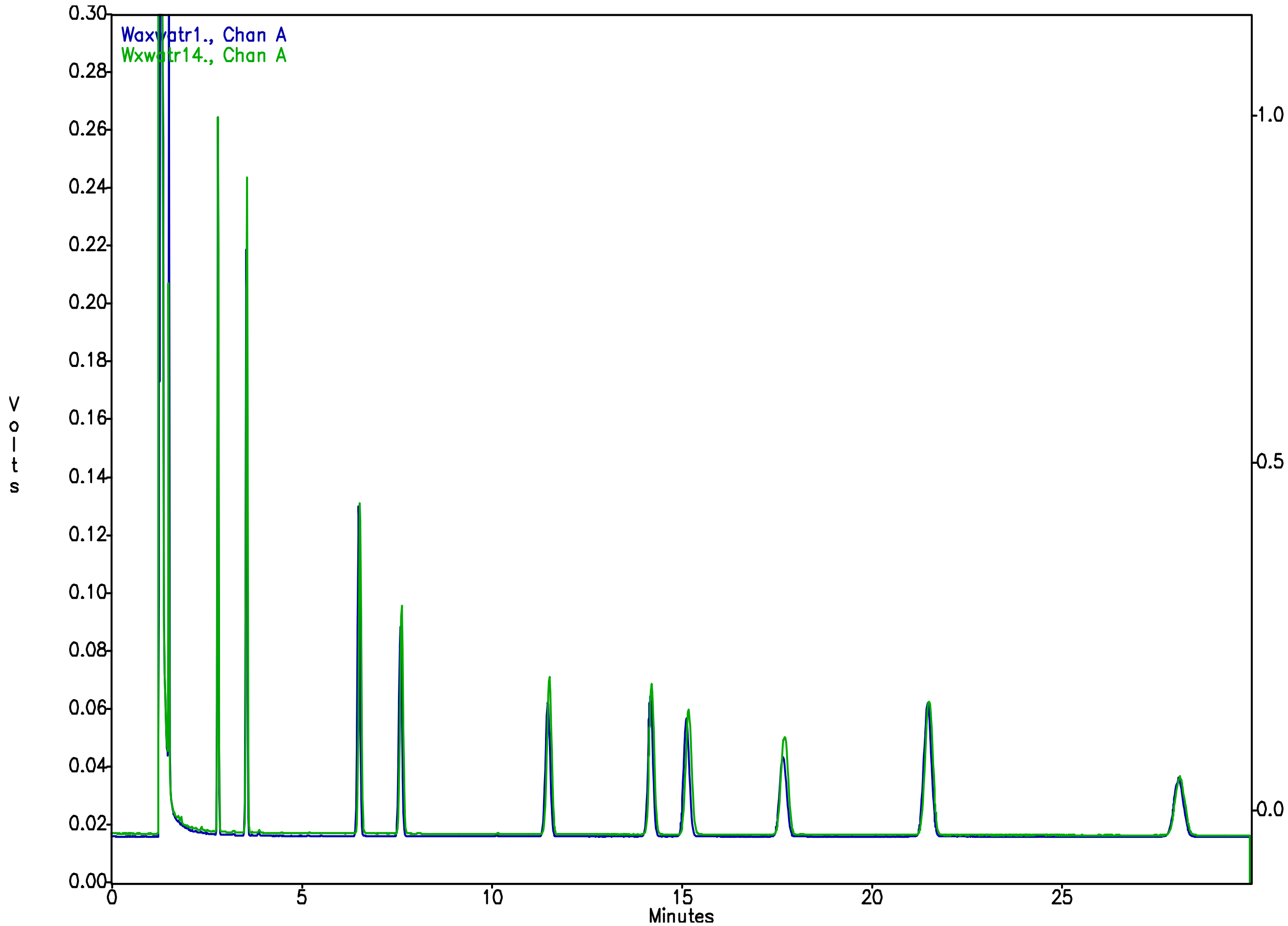
Results DB-Wax

Parameter	Before Inj.	After Inj.
Ret. Factor	12.6	12.6
Ret. Index 1	1149.54	1149.73
Ret. Index 2	1163.44	1163.71
Theor. plates	1277	1261
Bleed (pA)	44.8	32.1

DB-Wax before and after 2,000 water injections



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Results DB-225

Parameter	Before Inj.	After Inj.
Ret. Factor	11.5	11.4
Ret. Index 1	1622.30	1621.26
Ret. Index 2	1711.51	1711.03
Theor. plates	1101	1110
Bleed (pA)	34.5	39.3

Results CycloSil B

Parameter	Before Inj.	After Inj.
Ret. Factor	7.8	7.6
Ret. Index	1306.3	1306.0
Resolution	1.9	1.3
Theor. plates	2631	2025
Bleed (pA)	28.4	15.1

Results HP-PLOT U

Parameter	Before Inj.	After Inj.
Ret. Factor	5.2	5.3
Ret. Index	538.0	540.0
Theor. plates	950	982
Bleed (pA)	74.2	35.6

Asymmetry (Skew)

The numbers represent the deviation from an ideal Gaussian, i.e., perfectly symmetrical, peak.

The smaller the number, the better.

Noticeable tailing starts at ~0.8.
Obvious tailing start at ~1.2

Asymmetry (Skew)

Skew numbers are based on a modified Gaussian-peak model*. It relies more on the zeroth and first moment calculations, which are less influenced by baseline noise.

*W.W.Yau, Anal. Chem., vol. 49, No. 3 (1977), pp 395-398

Results DB-1

Asymmetry

Compound	Before Inj.	After Inj.
Chlorophenol	0.48	0.47
Timethylaniline	0.47	0.46
Undecanol	0.51	0.46

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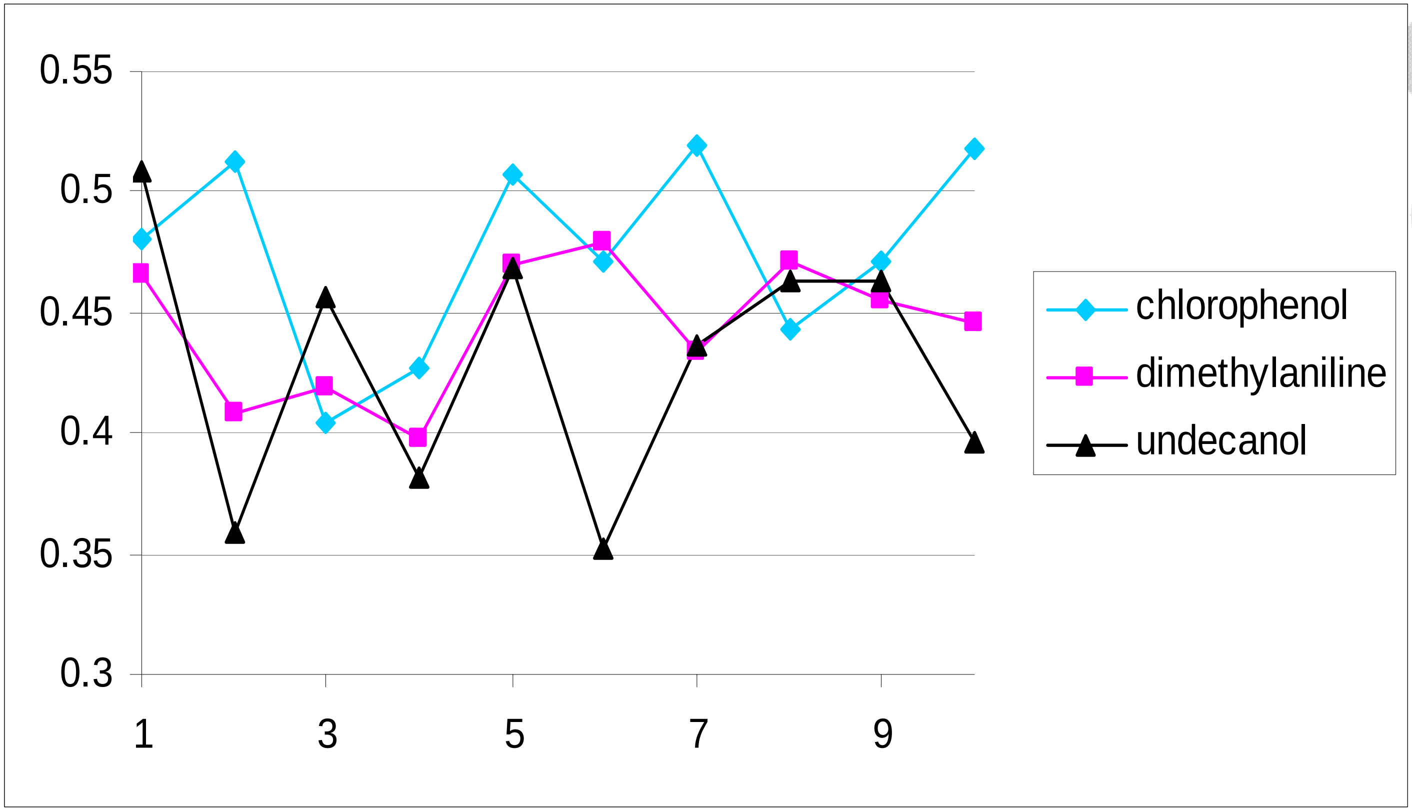
Skew Factors DB-1



Results DB-Wax

Asymmetry

Compound	Before Inj.	After Inj.
Decanol	0.38	0.47
Dimethylaniline	0.30	0.32
Dimethylphenol	0.25	0.27

Results DB-225

Asymmetry

Compound	Before Inj.	After Inj.
Chlorophenol	0.46	0.47
Dimethylaniline	0.42	0.43
Undecanol	0.63	0.58

Break

For Questions and Answers

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Summary

For water injections on all columns with bonded stationary phases, no change in:

polarity
selectivity
retention
efficiency
activity
bleed

is observed.

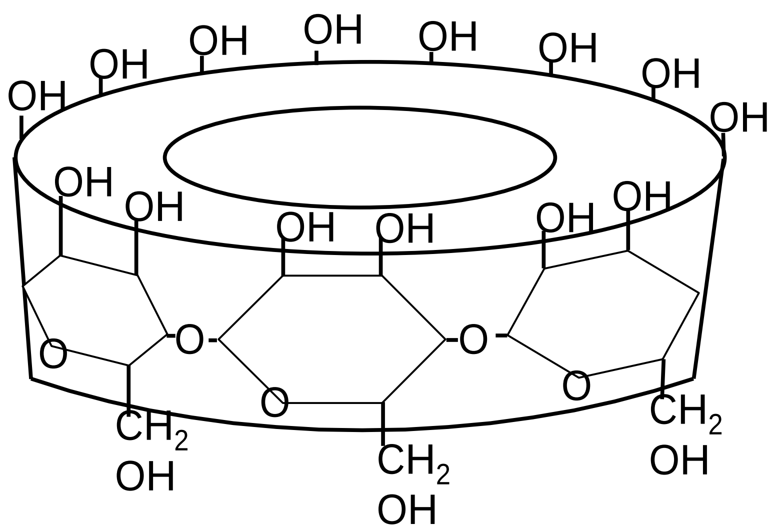
Summary

For <u>non-bonded phases</u>, like CycloSil B, water injections can wash out part of the non-bonded stationary phase - loss of resolution, retention, and, possibly, efficiency.

Summary

•Amount of wash-out is dependent on temperature: at 130º C and 200º C it is minimal, at 60º C it is noticeable.

• Solubility of phase material is greater in liquid water.



-7 Glucose molecules joined through 1,4-glycosidic linkages
-Primary hydroxyl groups partially block the base
-Cavity size is about 6-6.5 Å in diameter
-Hydroxy groups are peralkylated

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Summary

Amount of wash-out is gradual!

Most likely dependent on solubility of phase in water (cyclodextrin = high).

No change in selectivity.

No increase in bleed!

Summary

For bonded PLOT columns, no negative effects of injecting water were observed.

Non-bonded PLOT columns (e.g., Alumina and Molesieve), are not suitable for water injections. Observe manufacturers recommendations.

Summary

For all phases, the time to bleed down, or re-condition, a column after injecting water is dependent on the run temperature:

Low-temperature injections will take a lot longer than high-temperature injections.

Periodic bake-out is recommend if injecting water at <80º C.

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Conclusions

Non-polar columns (bonded and cross-linked) : *safe to inject and rinse with water*

Polar columns (bonded and cross-linked) :
water injections are safe
water-rinsing not recommended

Non-bonded columns: *water injections can wash out stationary phase - use with caution*

Conclusions

Problems associated with water injections are most often caused by injector related phenomena, e.g., backflash.

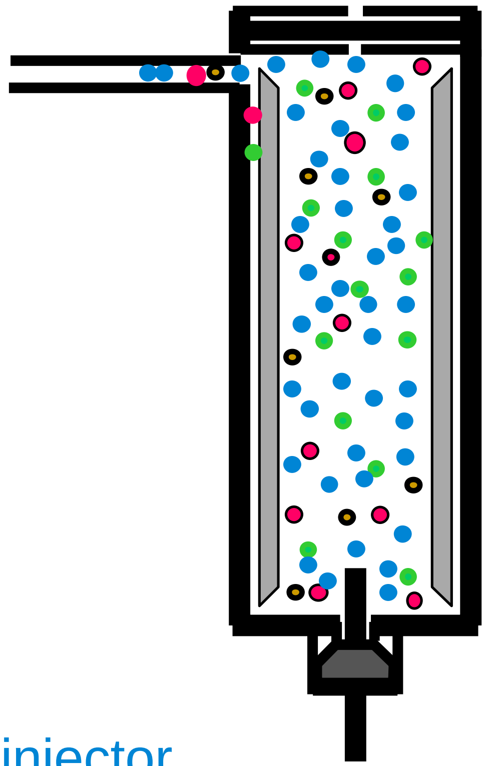
Solvent Expansion Volumes

Solvent	Vapor volume (µL) of 1µL liquid
Water	*1010*
Methanol	450
Carbon disulfide	300
Methylene chloride	285
Acetone	245
n-Hexane	140

BACKFLASH
Cause

- Vaporized sample expands 100 -1000 X
- Portions may leave the liner
- Occurs when vapor volume > liner volume

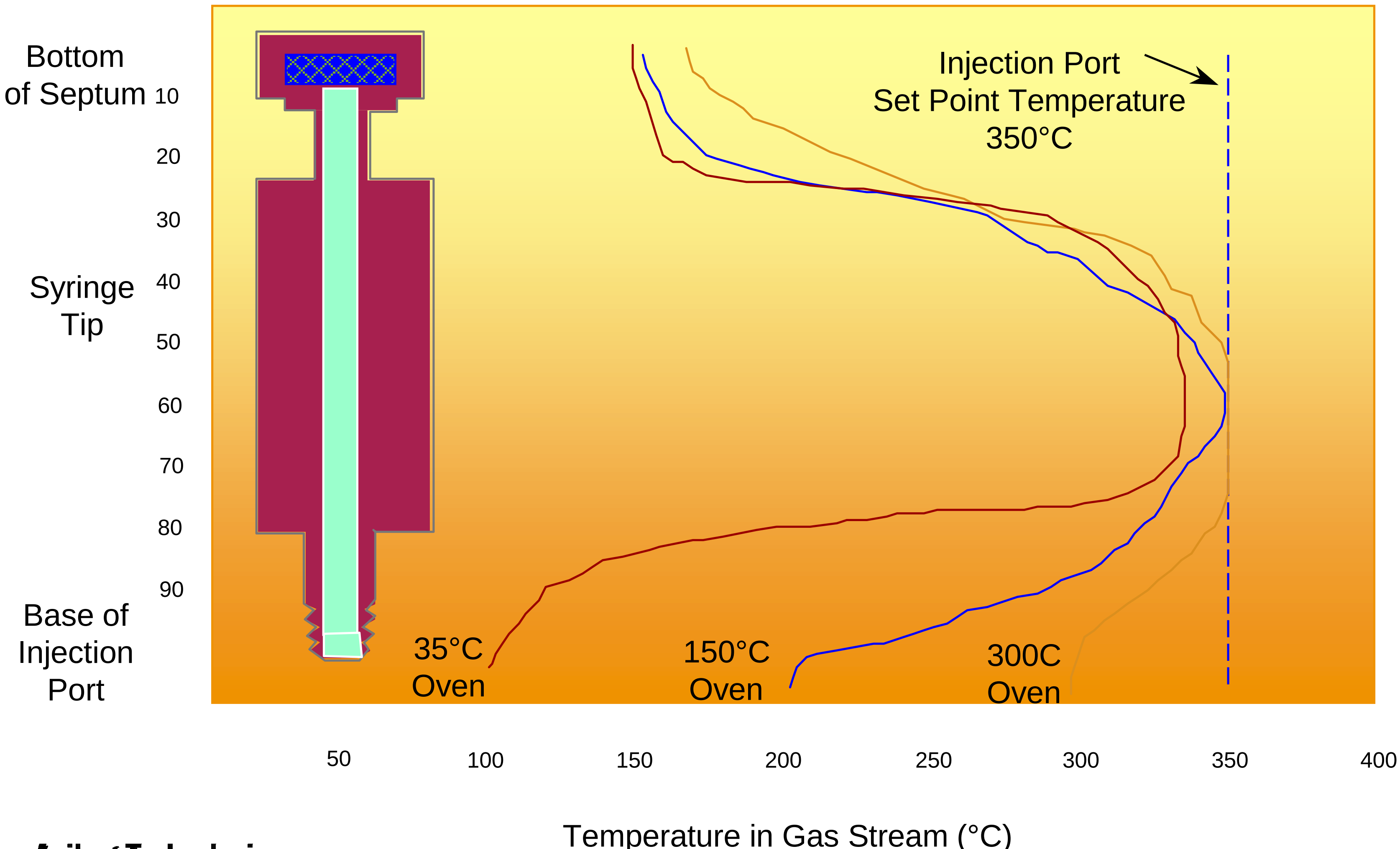
BACKFLASH



Sample expands to overfill injector
Some sample condenses on cooler areas (bottom of septum, metal body, etc.)

Next injections or carrier gas dislodges condensed sample
Sample then enters the column

Temperature Profile of a Typical Vaporization Injector vs Oven Temperature



BACKFLASH
Problems

- Loss of sample
- Baseline interferences
- "Ghost" peaks
- Tailing solvent front

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BACKFLASH
Minimizing

- Large volume liner
- Small injection volume
- Low expansion solvent
- Low injector temperature
- High carrier gas flow rates
- High head pressures

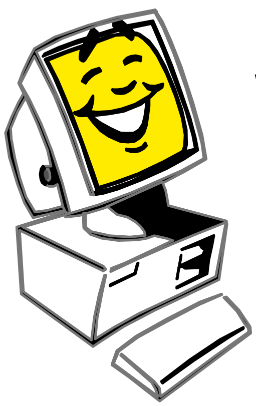
J&W Scientific GC Columns Technical Support

800-227-9770 (phone: US & Canada)*

302-993-5304 (phone)*

**** Select option 4, then option 1.***

916-608-1964 (fax)



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