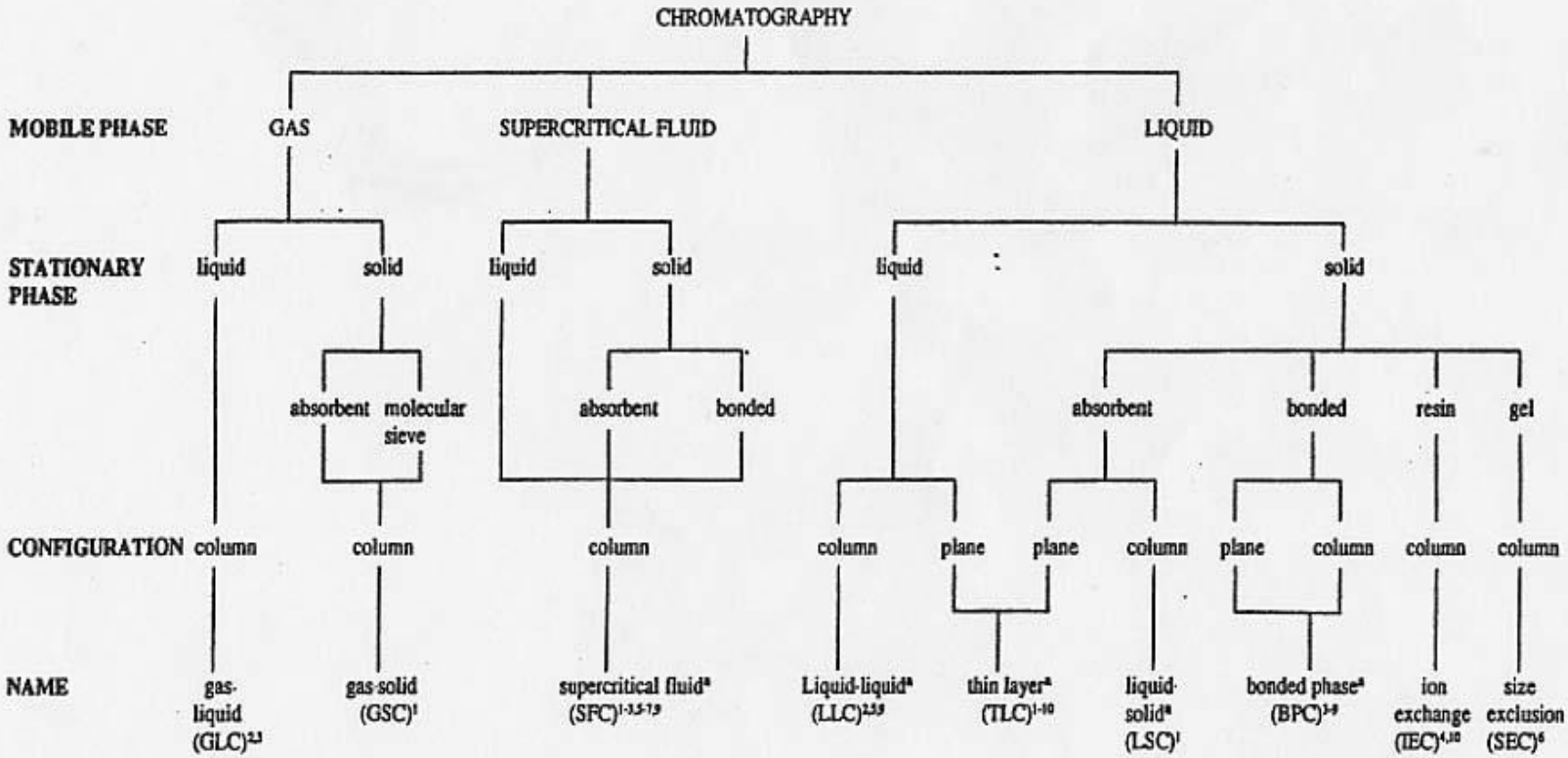
CHEMISTRY 5570

Advanced Analytical Chemistry

Lecture 14

Chromatography

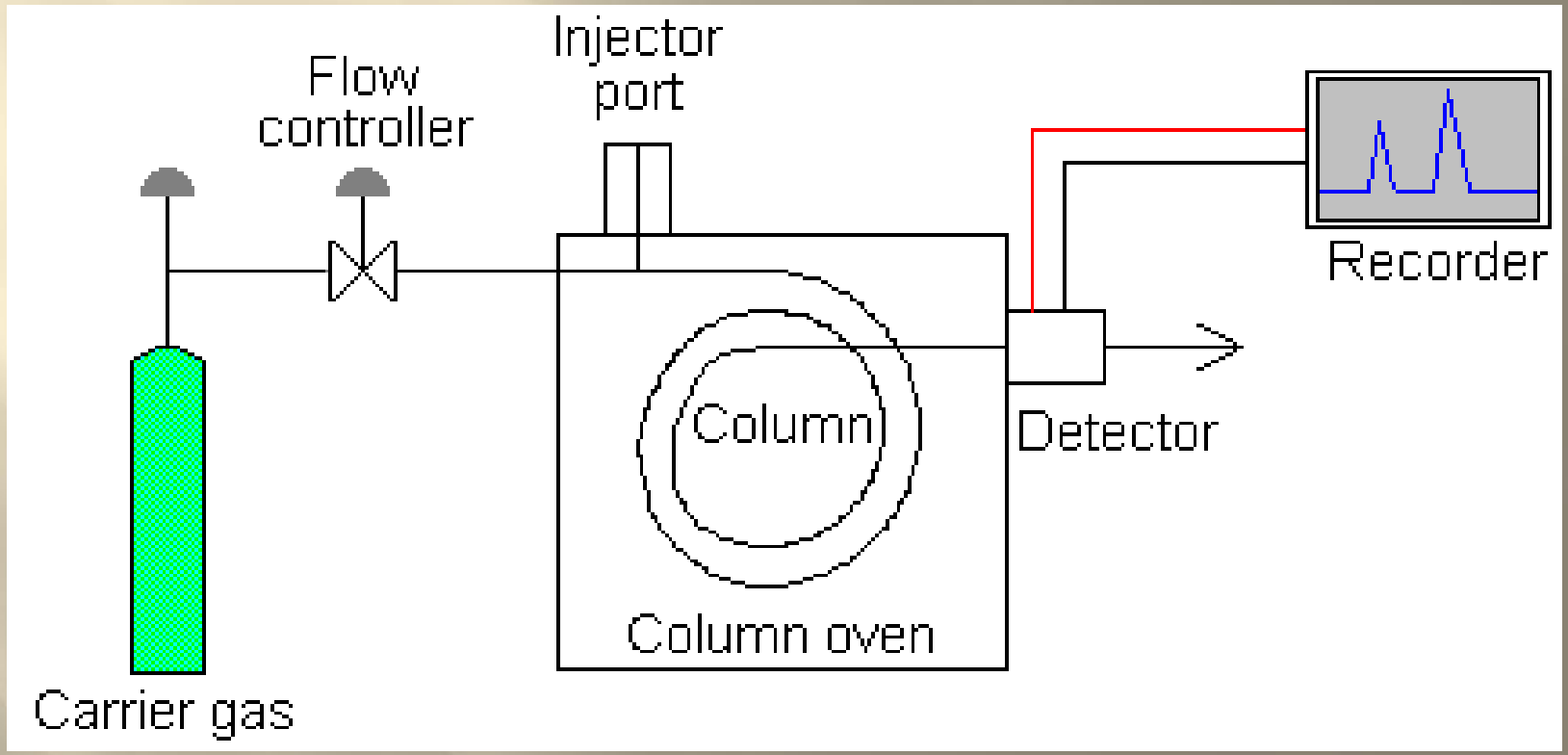


[a]For these techniques the combination of mobile and stationary phase can be varied to generate either a normal phase or reversed phase system. Mechanisms which have been exploited in the various techniques are identified as: [1]adsorption, [2]partition, [3]bonded phase, [4]ion exchange, [5]ion interaction, [6]size exclusion, [7]affinity, [8]micellar, [9]chelation, [10]ion exclusion.

Fig. 1.3. Classification of chromatographic systems.

Chromatography

Gas Chromatography – Columns



Chromatography

Gas Chromatography

Types of Columns

Packed Columns

Older type of column

Both solid and liquid stationary phase

Best column for preparatory GC and for use with thermal conductivity detectors

Sometimes used for very specific applications (low production volume less of an issue)

Chromatography

Gas Chromatography

Types of Columns

Open Tubular Columns

More modern columns

Much better analytical performance (large N values)

Most common in wall coated format (WCOT)

Variety of diameters (0.25 to 0.53 mm most common) allow high resolution vs. easier injection

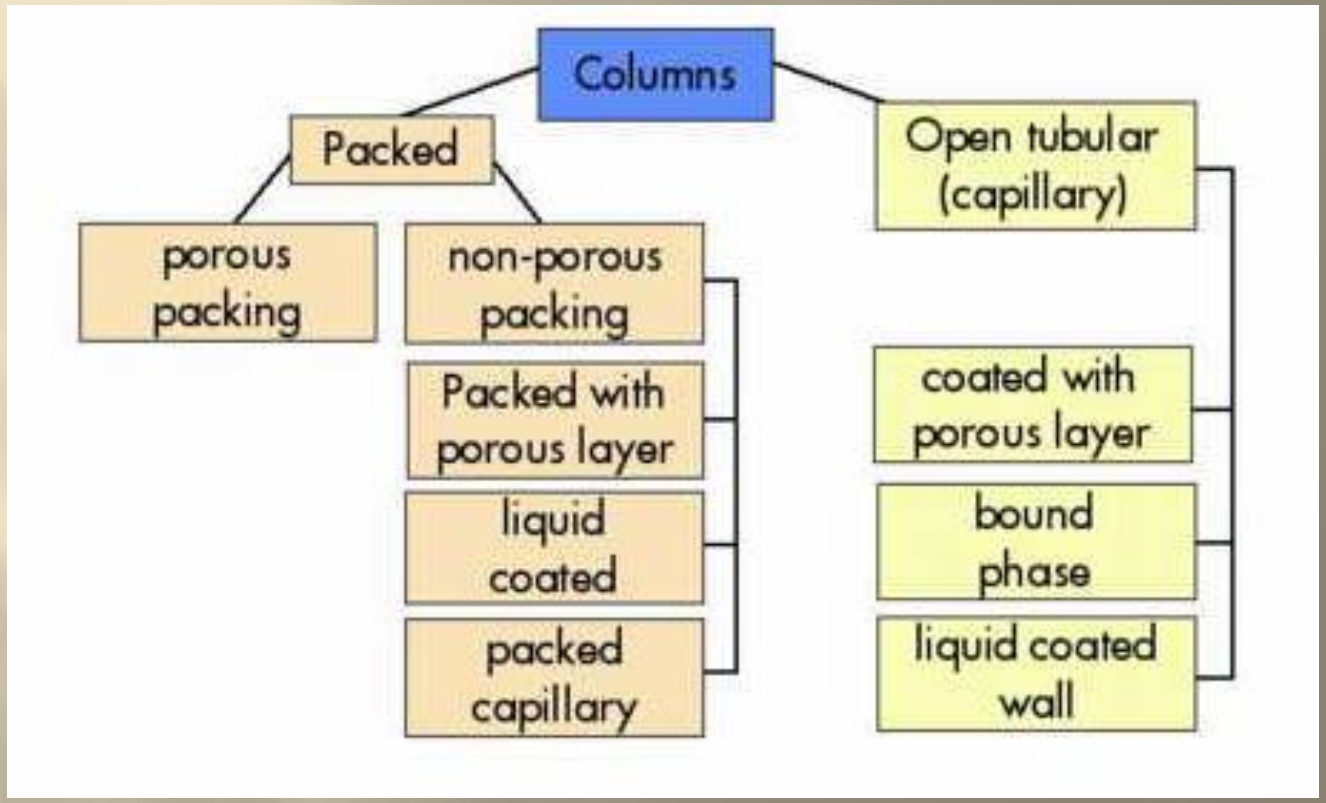
Stationary phases are mainly bonded of varying amounts of polarity

Good reliability

Disadvantages: harder to make and less capacity

Chromatography

Gas Chromatography



Chromatography

Gas Chromatography

Columns

Packed columns contain a finely divided, inert, solid support material (commonly based on diatomaceous earth) coated with liquid stationary phase. Most packed columns are 1.5 – 10 m in length and have an internal diameter of 2 – 4 mm.

Capillary columns have an internal diameter of a few tenths of a millimeter.
They can be one of two types;
wall-coated open tubular (WCOT) or
support-coated open tubular (SCOT).

Chromatography

Gas Chromatography

Columns

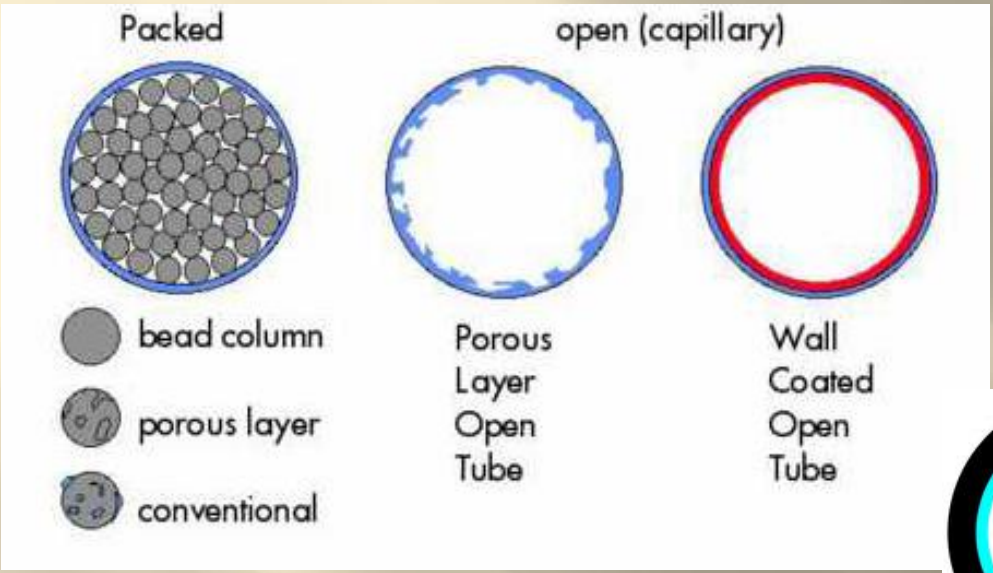
Wall-coated columns consist of a capillary tube whose walls are coated with liquid stationary phase.

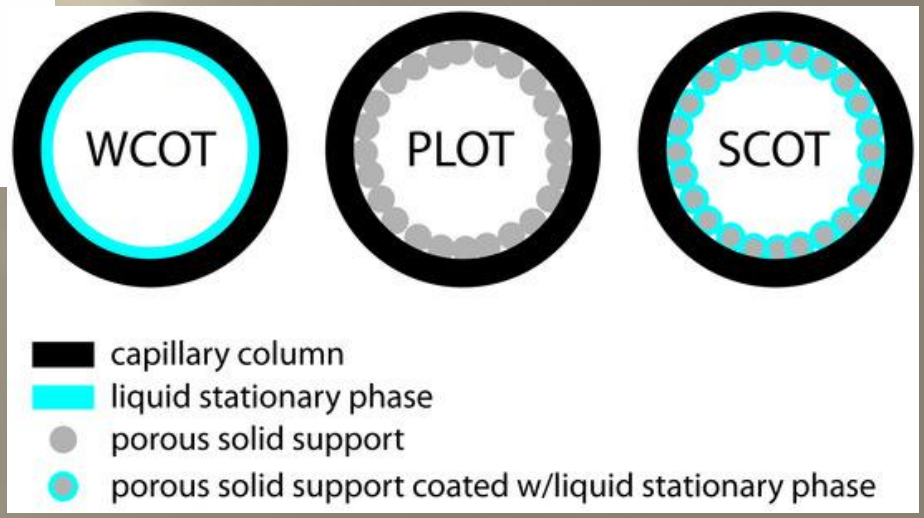
In support-coated columns, the inner wall of the capillary is lined with a thin layer of support material such as diatomaceous earth, onto which the stationary phase has been adsorbed.
SCOT columns are generally less efficient than WCOT columns.

Both types of capillary column are more efficient than packed columns.

Chromatography

Columns





Chromatography

Gas Chromatography

Column Materials

- Column tubing supports the stationary phase.

- Needs to be:

-chemically inert (to prevent sample decomposition)
-thermally stable
-robust
-flexible – form a coil to fit into oven

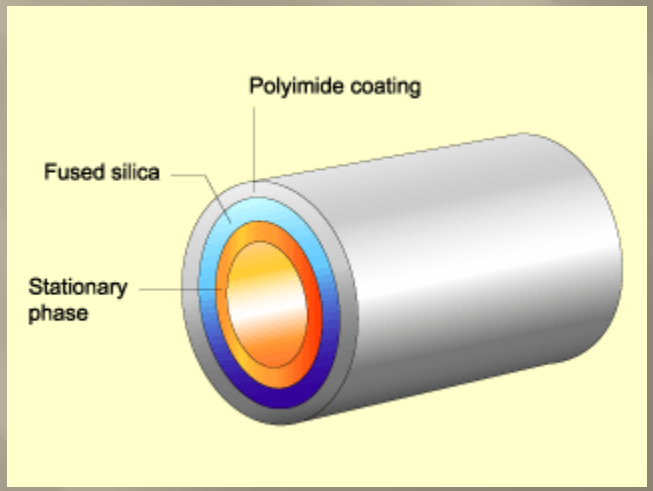
Chromatography

Column Materials

Early columns were metal (stainless steel, Cu, Al, Ni) but these were too reactive.

Glass is popular – but fragile

Fused silica columns – weak and subject to atmospheric corrosion. Use a protective outer Al or polymer sheath. These columns are used for most applications.



Chromatography

Parameter	Capillary	Packed
Efficiency (plates/m)	2 000 – 4 000	500 - 1 000
Sample size (ng)	10 - 75	10 – 1 000 000
Relative pressure	Low	High
Relative speed	Fast	Slow
Chemical inertness	Best	Poorest
Flexible column	Yes	No

Chromatography

Gas Chromatography

Columns

Column Activity leads to:

- tailing or skewed peaks
- partial or total disappearance of a solute
  - reversible peak adsorption
  - non-reversible peak adsorption
  - breakdown

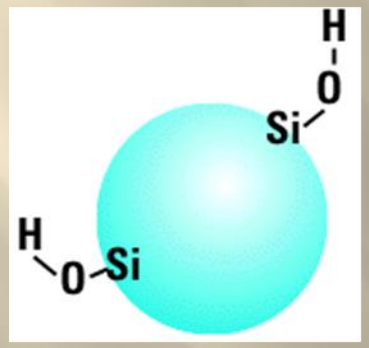
The surface properties of the column – catalytic or sorptive are important to consider to eliminate column activity.

Chromatography

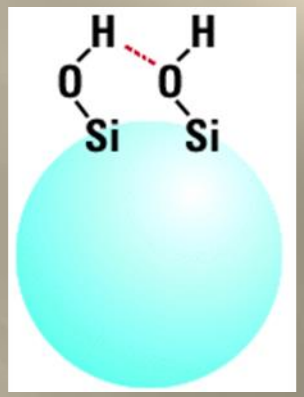
Gas Chromatography

Columns

Various surface groups attributed to the silica surface have been identified:

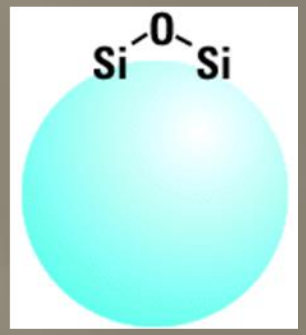


Isolated Silanol



Vicinal Silanol

Geminal Silanol



Siloxane

Chromatography

Gas Chromatography

Columns

Surface hydroxyl groups can act as proton donors in hydrogen bonding interactions

- very strong sorptive sites for molecules with localized high electron density.

Surface siloxane bridge can act as

- proton acceptors functioning as sorptive sites for molecules such as alcohols

Chromatography

Columns

Metallic contamination

Metal ions present on the surface of soda lime or borosilicate glasses can function as Lewis acid sites absorbing molecules that have regions of localized high electron density such olefins, aromatic compounds, alcohols, ketones and amines. (Lewis acids – a species that accepts an electron pair)

Chromatography

Gas Chromatography

Columns

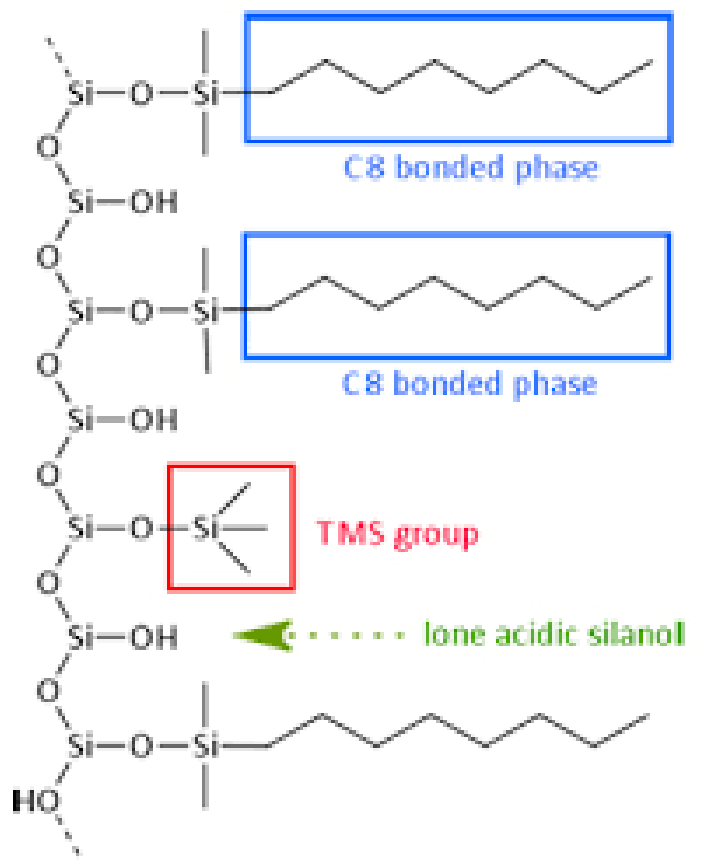
Columns are deactivated before use. One method deactivates reactive hydroxyl groups by silanizing or silylating the inner surface of the column.

Typical derivatization reactions are:

- Silylation
- Acylation
- Alkylation
- Esterification

Chromatography

Gas Chromatography



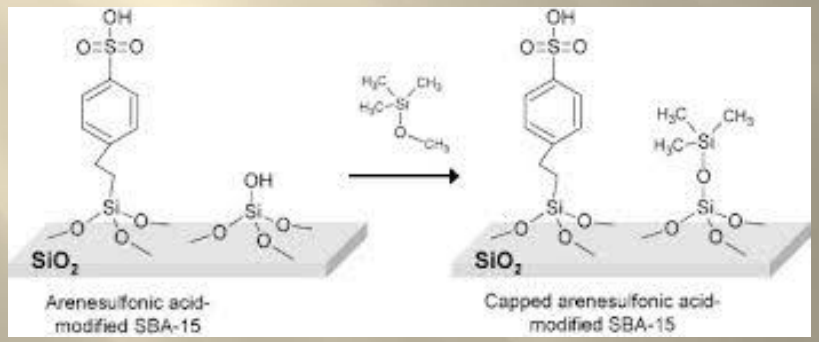
Columns are deactivated before use. Remember that these reactions are used to take care of a "problem" – but can just as well be used to "create" new stationary phases… we will cover this more later.

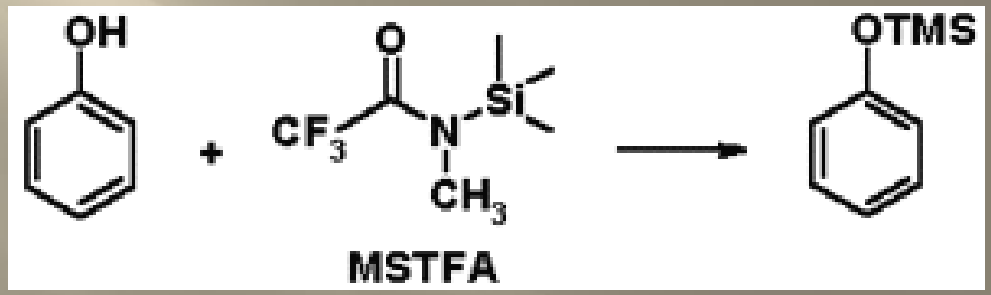
Chromatography

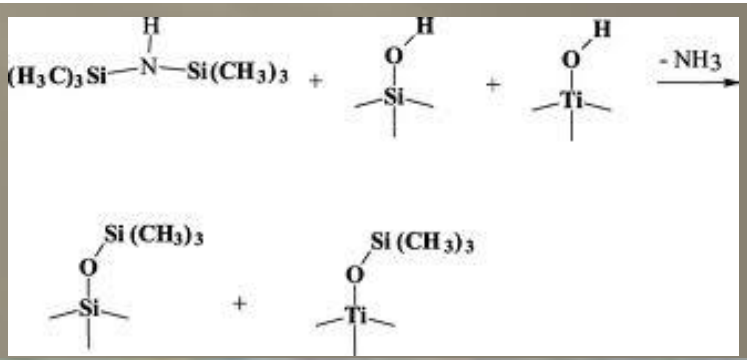
Gas Chromatography

Columns

Silylation - It involves the replacement of an acidic hydrogen on the compound with an alkylsilyl group, for example, -$SiMe_3$. The derivatives are generally less polar, more volatile and more thermally stable.



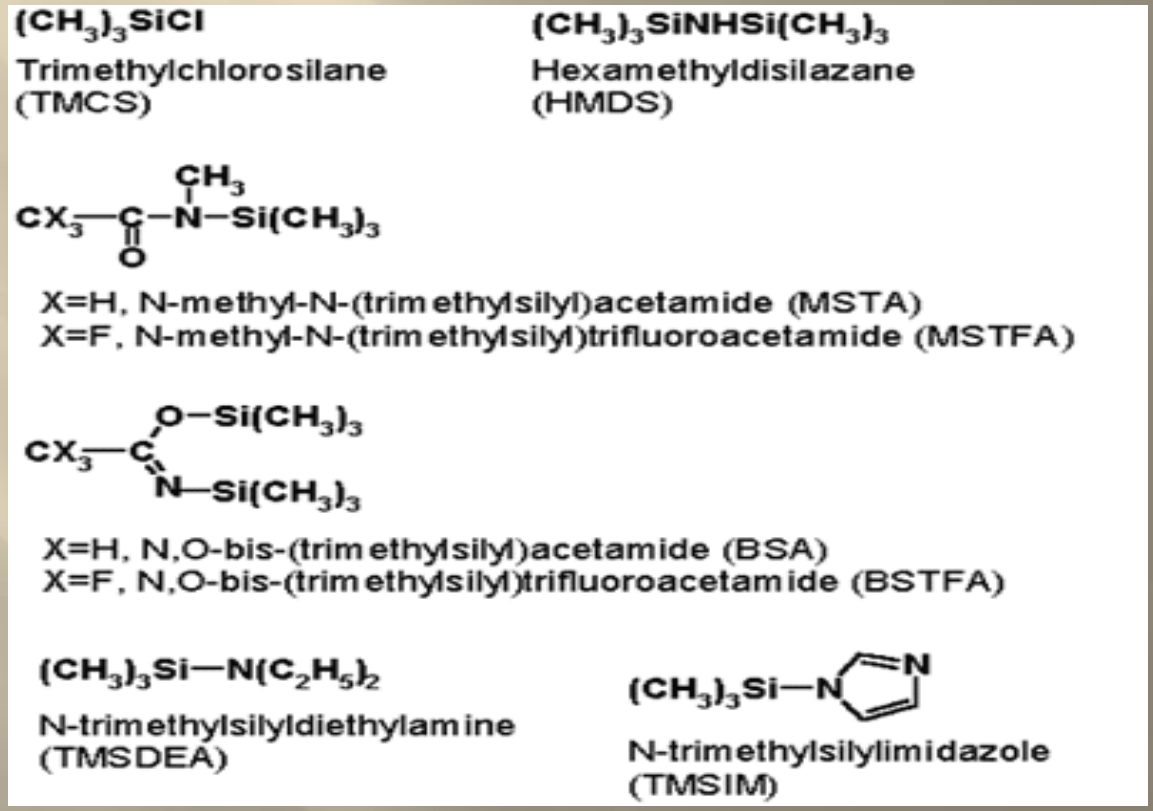




Chromatography

Columns

Various silanizing reagents can be used. The most common reagents for silylation are the trimethylsilyl (TMS) reagents.



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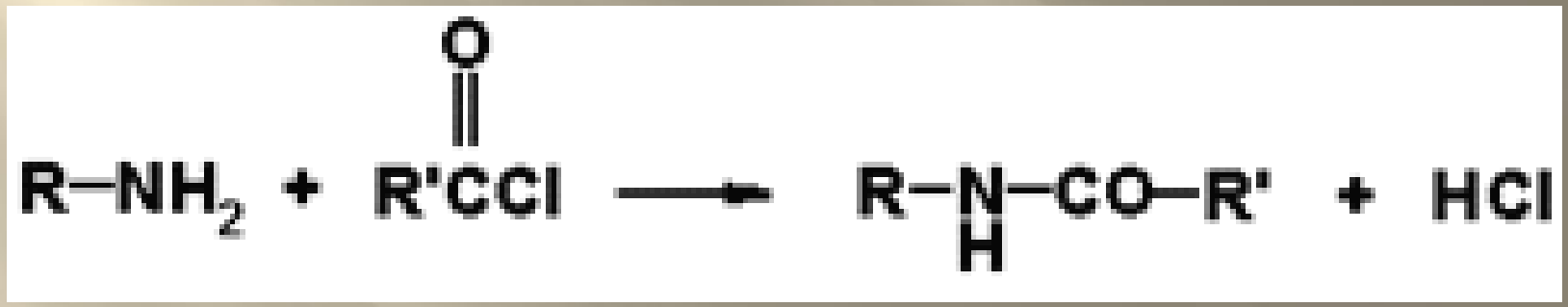
Gas Chromatography

Columns

Acylation - Acylation is one of the most widely used derivatization procedures for gas chromatography. Acylation reduces the polarity of compounds that contain active hydrogens (-NH, -OH, -SH amino, hydroxyl and thiol groups) into amides, esters, or thioesters by using carboxylic derivatives.

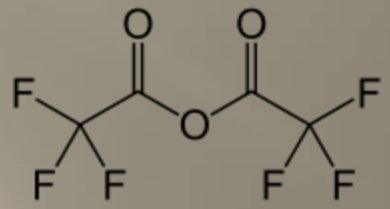


Chromatography

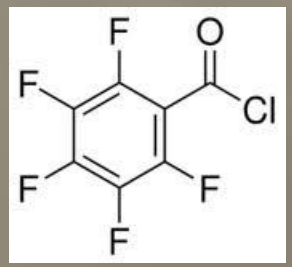
Columns

The most widely used acylating reagents is acetic anhydride, $(CH_3CO)_2O$.

Acylating reagents



Trifluoroacetic anhydride (TFAA)
Pentafluoropropionic anhydride (PFPA)
Heptafluorobutyryl anhydride (HFBA)
Pentafluorobenzoyl chloride



Chromatography

Columns

Alkylation- **Alkylation is the replacement of a active hydrogen in R-COOH, R-OH, R-SH, and R-NH_2 with an alkyl group or, sometimes aryl group.**

Chromatography

Columns

Alkylating reagents

- Alkyl halide (eg. pentafluorobenzyl bromide, PFBBr, 3,5-ditrifluoromethylbenzyl bromide)
- Diazoalkane (eg. diazomethane)
- N,N-Dimethylformamide dialkyl acetal
- 3-alkyl-1-p-tolyltriazene
- Fluorosulfonate
- Trimethylanilinium hydroxide (TMAH)
- Trialkyloxonium fluoroborate
- Alkyl trichloroacetimidate

Chromatography

Columns

Functional group	Derivatization
-OH (hydroxyl group) in primary, secondary and tertiary alcohols; phenols; carbohydrates)	Silylation Acylation Benzoylation Alkylation Dansylation Reaction with Dis-Cl Reaction with FDNB Reaction with NBD-Cl Ion-pair formation
-COOH (carboxylic acids)	Esterification S ilylation Ion-pair formation
-C=O (carbonyl group) in aldehydes and ketones	Oxime formation Oxime formation and silylation Ketal/acetal formation Hydrazone formation Schiff's base formation Silylation

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Functional group	Derivatization
-NH2 (amino group) in primary amines, amino acids, amino sugars	Acylation Benzoylation Silylation Treatment with CS2 Thiourea formation Schiff's base formation 2,4-Dinitrophenylation Sulphonamide formation Carbamate formation Treatment with pyridoxal Treatment with NBD-Cl Alkylation Ion-pair formation
-NH-R (amino group) in secondary amines, imino acids, substituted amino sugars	Acylation Benzoylation Silylation 2,4-Dinitrophenylation Sulphonamide formation Treatment with NBD-Cl Ion-pair formation
-NH2 and -COOH in amino acids	Silylation Esterification + Acylation
-NO2 (nitro compounds)	Chromatograph without derivatization

Chromatography

Columns

Stationary Phases

Selection of stationary phase affects k and α values.

Main concerns of stationary phase are:
- polarity,
- functional groups,
- maximum operating temperature, and
- column bleed (loss of stationary phase due to decomposition)

More polar columns suffer from lower maximum temperatures and greater column bleed

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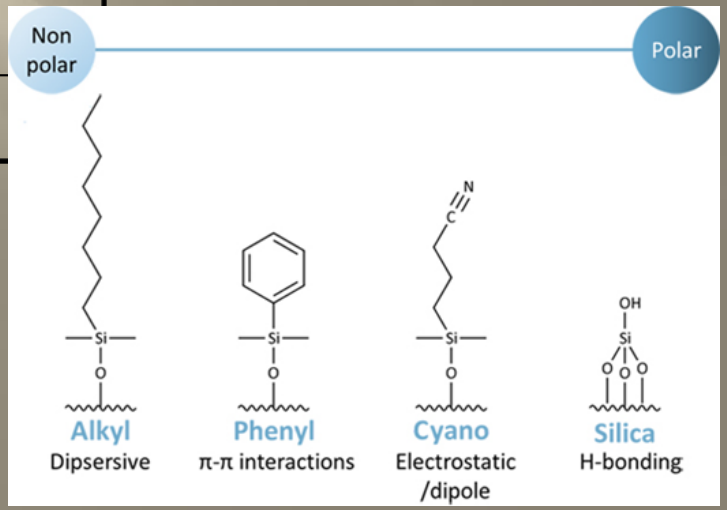
Columns

Stationary Phases (examples)

Type	Functional Groups	Polarity
OV-1	methyl	Non-polar
OV-17	50% methyl/50% phenyl	Somewhat polar
OV-225	Cyanopropyl, methyl, and phenyl	More polar
carbowax	Ether groups	polar



Chromatography

Columns

Stationary Phases (GSC)

GSC of inorganic gases is the one area where packed columns are still used almost exclusively. Main adsorbents for packed columns are based on:

- **silica**
- **charcoal**
- **alumina**
- **molecular sieves**

Chromatography

Columns

Stationary Phases (GSC)

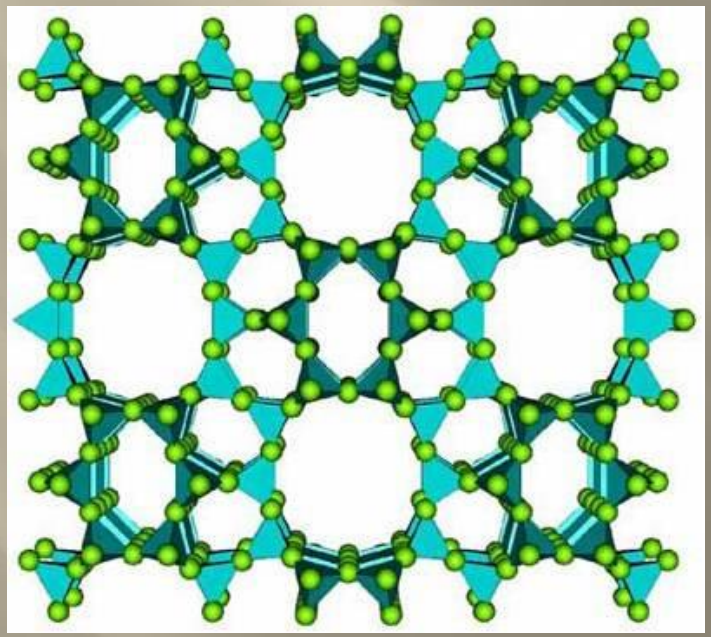
- Porous silica is used with a variety of surface areas and pore diameters (separate low molecular mass hydrocarbons).

- Alumina has a variety of surface areas – but different selectivities due to Lewis acid sites associated with aluminum ions on the surface. To modify selectivity and improve efficiency, both silica and alumina can be coated with a non-volatile liquid.

Chromatography

Stationary Phases (GSC)

- **Molecular sieves are artificially prepared zeolites (i.e. Na, K, Ca – aluminosilicates) – small pore diameter used to separate low molecular weight molecules and hydrocarbons.**



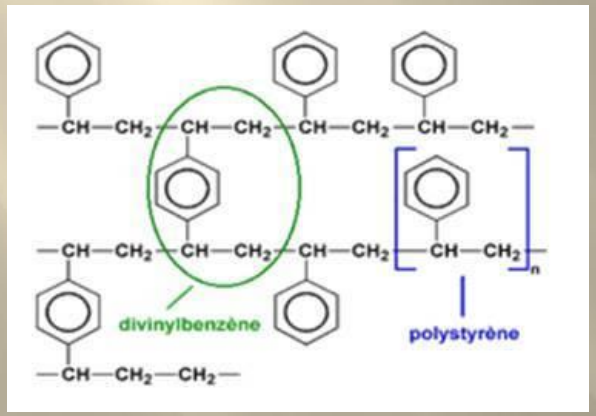
Chromatography

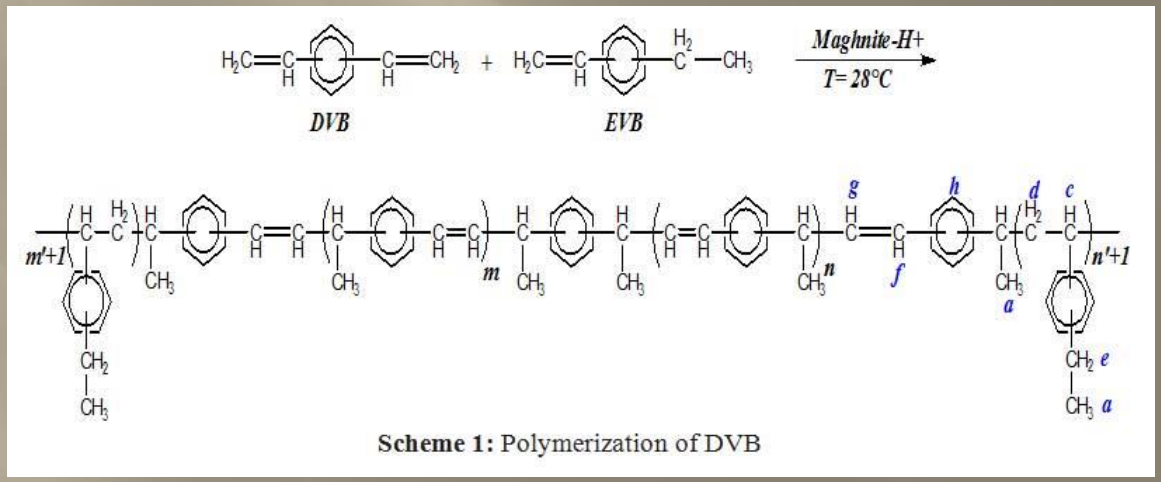
Stationary Phases (GSC)

Porous polymers – good for separating polar analytes. Usually consist of copolymers of divinylbenzene (DVB), CH_2=CH-(benzene ring)-CH=CH_2, with another aromatic olefin (ethylene CH_2=CH_2).

Easy to use and has superior performance and unique separating abilities.

The nature of the separation is adsorption, but the mechanism is unclear.





Scheme 1: Polymerization of DVB

Chromatography

Columns

Stationary Phases (GSC)

Adsorbents for GSC

Chemical Type	Specific Surface Area (m^2g^{-1})	Pore Diameter (nm)
Silica	~ 100 – 185	15 – 30
Alumina	-	-
Carbon Black	10 – 1000	1 – 2
Molecular Sieves	700 – 1000	0.5 – 2.0
Porous Polymers	~ 100 – 800	5 – 500

Chromatography

Columns - GLC

Wall-coated columns consist of a capillary tube whose walls are coated with liquid stationary phase.

In support-coated columns, the inner wall of the capillary is lined with a thin layer of support material such as diatomaceous earth, onto which the stationary phase has been adsorbed.

Liquid Phase – required properties

- **low vapor pressure**
- **thermal and chemical stability**
- **high viscosity**
- **nonreactive toward sample components**
- **wide operating temperatures (-80 to 450°C)**
- **reasonable solvent properties**

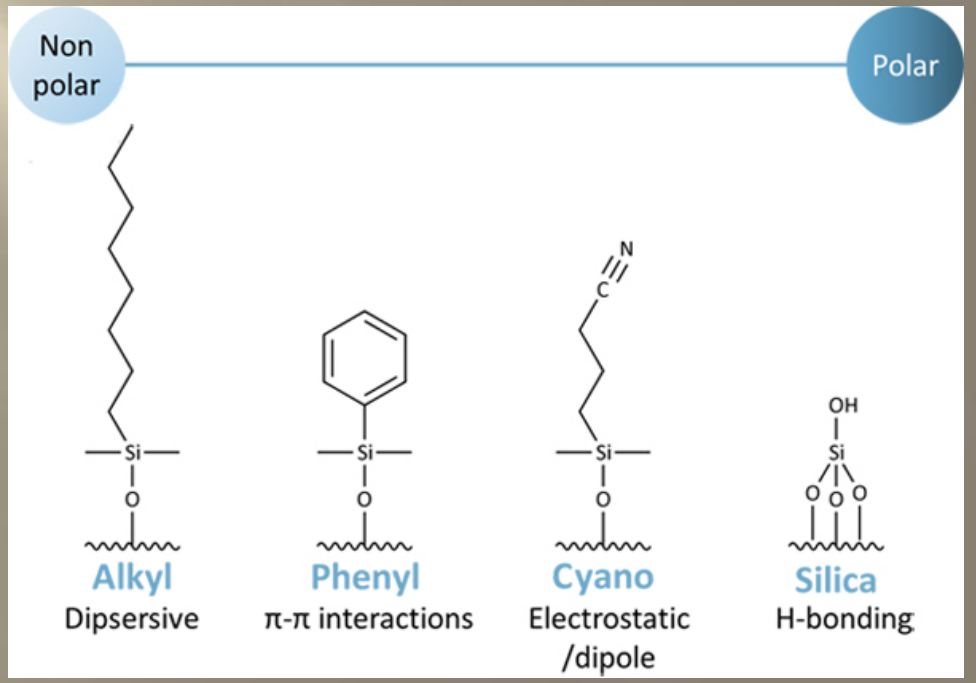
Chromatography

Columns

Stationary Phases (GLC)

Categories of stationary phases

- non-polar
- polar
- specialty phases



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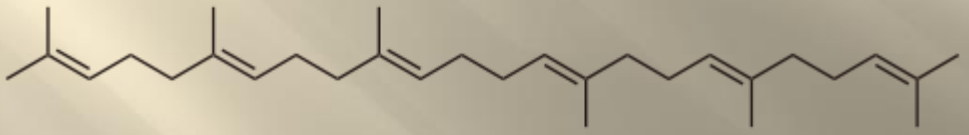
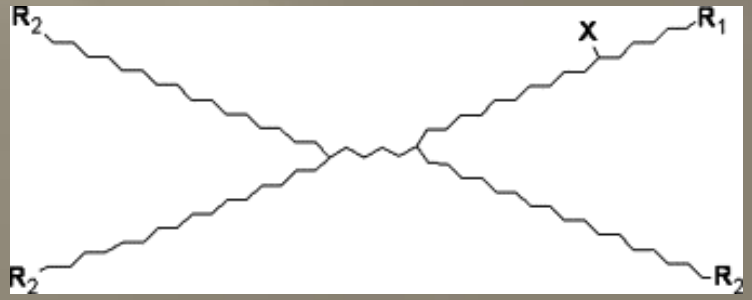
Columns

Stationary Phases (GLC)

Hydrocarbons – non-polar – have been used as non-polar stationary phase.
Most have a high molecular weight for low voltality.
Examples: Squalane ($C_{30}H_{62}$), Apolane C87, long-chain n-alkanes, Apiezon L-not used much.





Chromatography

Columns

Stationary Phases (GLC)

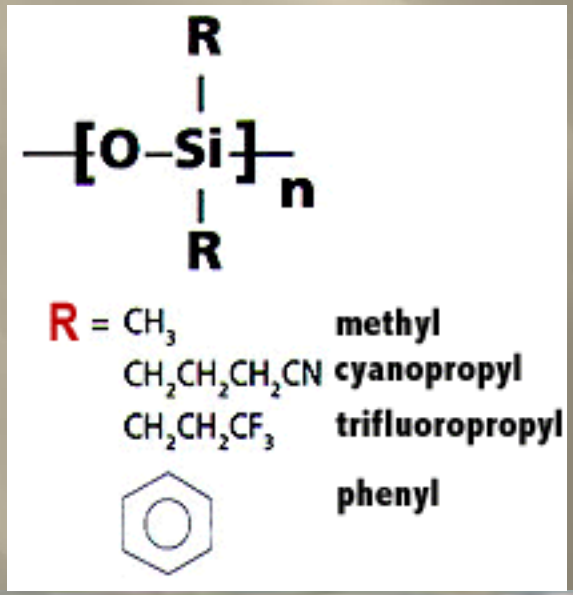
Alkylsilicone Phases – can be non-polar or polar. Polymers based on silicon-oxygen-silicone backbone – most widely used group of stationary phases. Differences in these stationary phases differ mainly in the degree of substitution on the silicon backbone.

Chromatography

Columns

Stationary Phases (GLC)

Standard polysiloxanes are characterized by the repeating siloxane backbone. Each silicon atom contains two functional groups. The type and amount of the groups distinguish each stationary phase and its properties.



Chromatography

Columns

Stationary Phases (GLC)

The most basic polysiloxane is the 100% methyl substituted.
When other groups are present, the amount is indicated as the percent of the total number of groups.
For example, a 5% diphenyl-95% dimethyl polysiloxane contains 5% phenyl groups and 95% methyl groups.
The "di-" prefix indicates that each silicon atom contains two of that particular group.
Sometimes this prefix is omitted even though two identical groups are present.

If the methyl percentage is not stated, it is understood to be present in the amount necessary to make 100% (e.g., 50% phenyl-methyl polysiloxane contains 50% methyl substitution).

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Columns

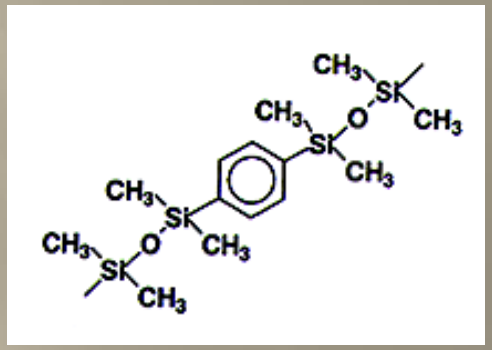
Stationary Phases (GLC)

Cyanopropylphenyl percent values can be misleading.

A 14% cyanopropylphenyl-dimethyl polysiloxane contains 7% cyanopropyl and 7% phenyl (along with 86% methyl).

The cyanopropyl and phenyl groups are on the same silicon atom, thus their amounts are summed.

Chromatography



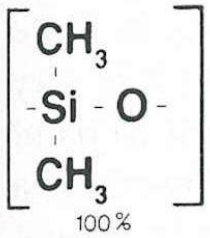
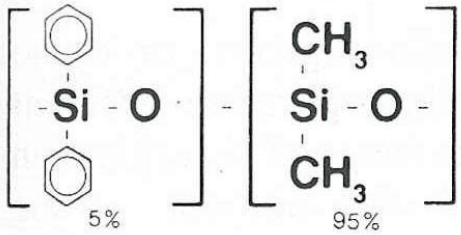
Columns

Stationary Phases (GLC)

- For select stationary phases, a low bleed or "ms" version is available.
- These stationary phases incorporate phenyl or phenyl type groups into the backbone of the siloxane polymer.
- These types of stationary phases are commonly called arylenes.
- The phenyl group strengthens and stiffens the polymer backbone which inhibits stationary phase degradation at higher temperatures.
- This results in lower column bleed and, in most cases, higher temperature limits.

Chromatography

Stationary Phases (GLC)

Chemical structure	Classification	Uses
100% dimethyl silicone 	Non-polar	Boiling point separations (solvents, petroleum products, pharmaceuticals)
95% dimethyl, 5% phenyl silicone 	Non-polar	Boiling point separations (aromatics, flavours, aromatic hydrocarbons)
86% dimethyl, 7% phenyl, 7% cyanopropyl silicone	Intermediate polarity	Pesticides, alcohols

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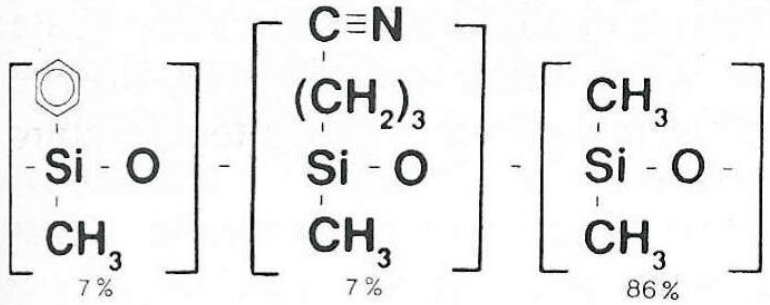
Stationary Phases (GLC)

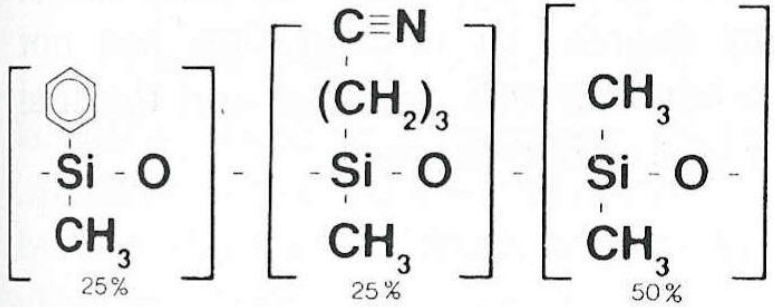
86 % dimethyl
7 % phenyl
7 % cyanopropyl
} silicone



Intermediate polarity

Pesticides, alcohols

50 % dimethyl
25 % phenyl
25 % cyanopropyl
} silicone



Polar

Triglycerides, phthalate esters

Chromatography

Columns

Stationary Phases (GLC)

Polyether Phases – polar phases – based on polyethylene glycol,
$HO\text{-}CH_2\text{-}CH_2\text{-}[O\text{-}CH_2\text{-}CH_2]_n\text{-}OH$.
Marketed under the trade name, carbowax or superox.

Polyethylene glycols stationary phases are not substituted, thus the polymer is 100% of the stated material. They are less stable, less robust and have lower temperature limits than most polysiloxanes.
With typical use, they exhibit shorter lifetimes and are more susceptible to damage upon over heating or exposure to oxygen.

Chromatography

Columns

Stationary Phases (GLC)

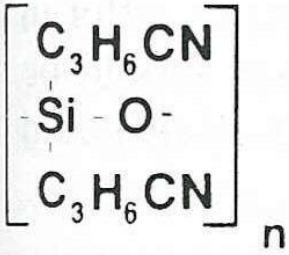
Polyether Phases – polar phases – based on polyethylene glycol,
$HO\text{-}CH_2\text{-}CH_2\text{-}[O\text{-}CH_2\text{-}CH_2]_n\text{-}OH$.

Advantage - Average molecular weight of 20,000, so can be used at high temperatures.
Disadvantage is that it reacts with even trace amounts of oxygen at high temperatures, as well as decomposing to acetaldehyde and acetic acid.

Chromatography

Stationary Phases (GLC)

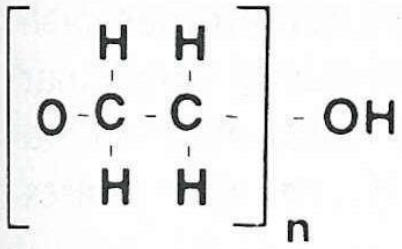
100 % cyano propyl silicone



Polar

Fatty acid methyl esters, carbohydrates

polyethylene glycol 20 M



Polar

Flavours, fatty acid methyl esters, acids, amines

Fig. 3.17. Structure of polysiloxane and polyethylene glycol stationary phases.

Chromatography

Columns

Stationary Phases (GLC)

- **High-temperature phases – substituted silicone-carborane copolymers.**
- **Stable up to 450°C - 500°C.**
- **Their advantage is in their low stationary bleed – for use with highly sensitive detectors.**

Chromatography

Stationary Phases (GLC)

High-temperature phases

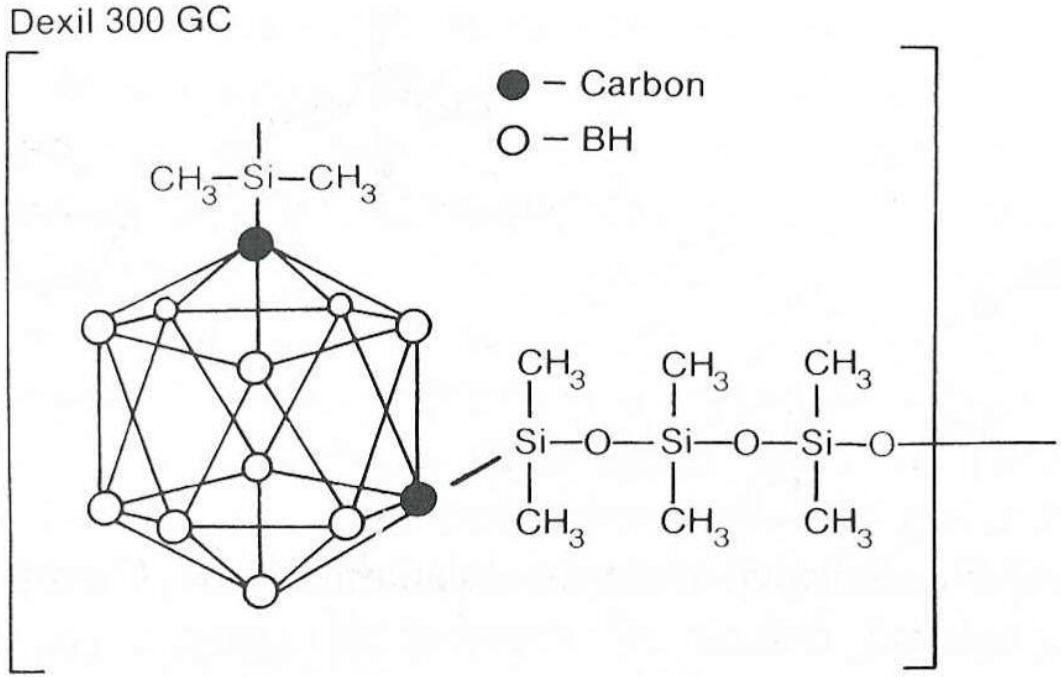


Fig. 3.18. A silicone–carborane copolymer used as a high temperature stationary phase.

Chromatography

Columns

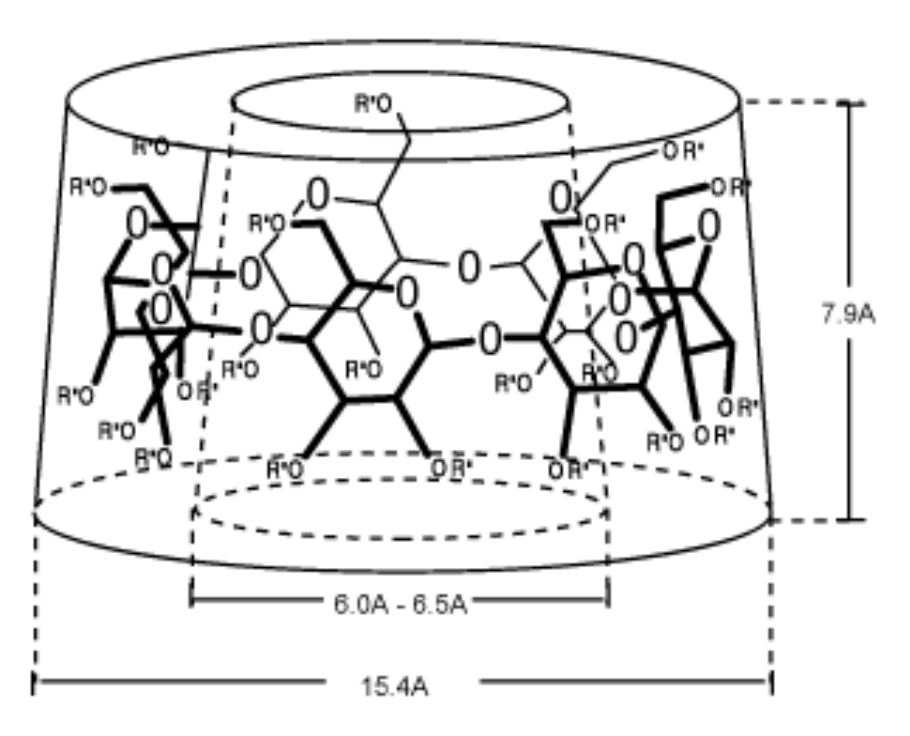
Stationary Phases (GLC)

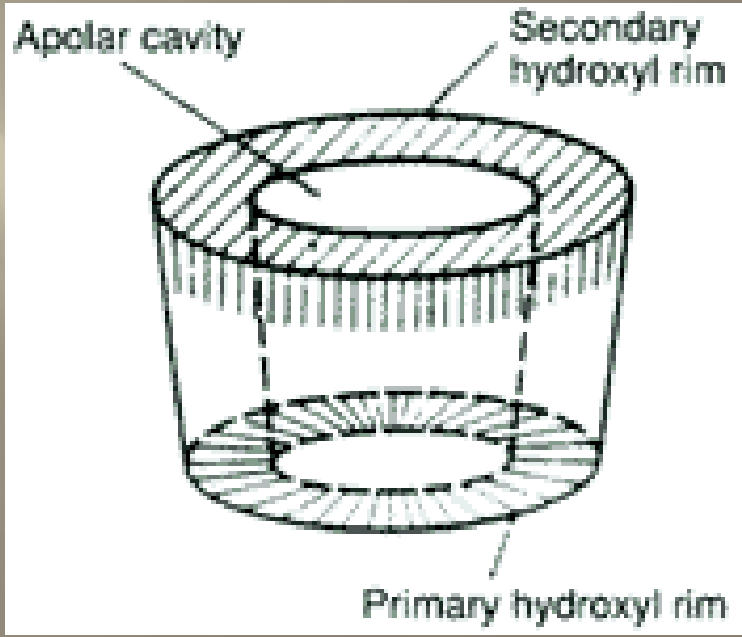
Chiral Phases - used to distinguish between enantiomers of optically active compounds. α,β,γ – cyclodextrins – composed of six or more D(+)-glucose units bonded through a α-(1,4) – glycosidic linkage.

Chromatography

Columns

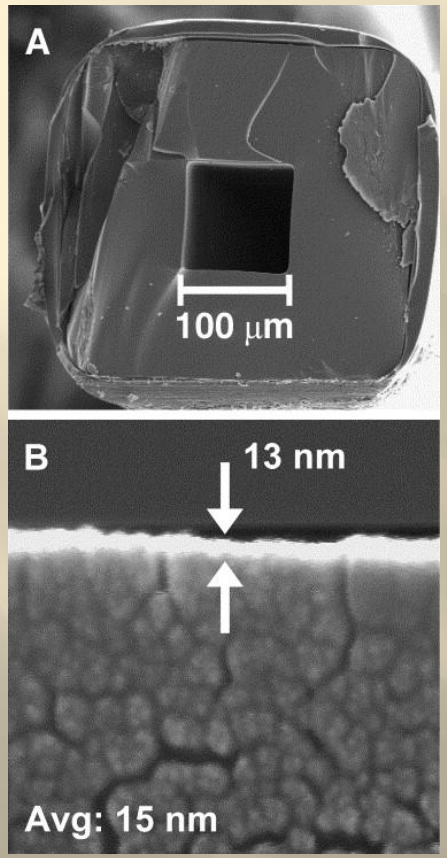
Stationary Phases (GLC)



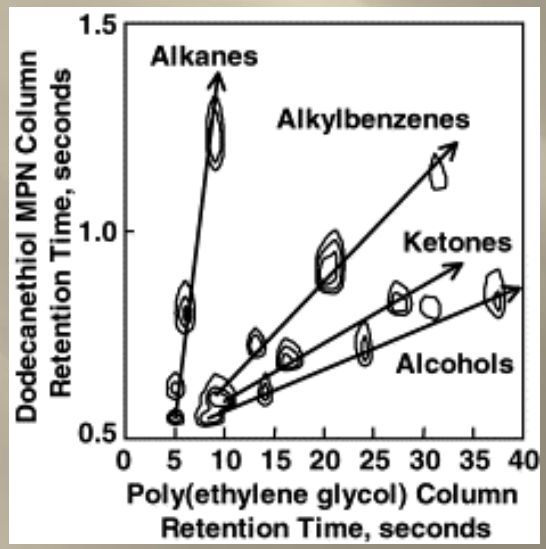


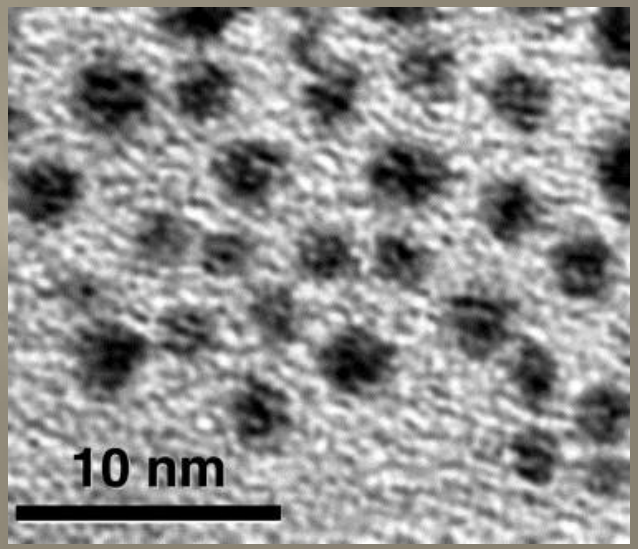
Chromatography

Stationary Phases



Dodecanethiol MPNs, in which the monolayer is dodecanethiol linked to the gold nanoparticle





- Journal of Chromatography A Volume 1060, Issues 1–2, 10 December 2004, Pages 225–236

Chromatography

Columns

Temperature Limits

Columns have lower and upper temperature limits.

If a column is used below its lower temperature limit, rounded and wide peaks are obtained (i.e., loss of efficiency).

No column damage has occurred; however, the column does not function properly.

Using the column at or above its lower limit maintains good peak shapes.

Chromatography

Columns

Temperature Limits

Columns have lower and upper temperature limits. Upper temperature limits are often stated as two numbers.

The lower one is the isothermal temperature limit. The column can be used indefinitely at this temperature and reasonable column bleed and lifetime are realized.

The upper number is the temperature program limit. A column can be maintained at this temperature for 10-15 minutes without severely shortening column lifetime or experiencing excessively high column bleed.

Exceeding the upper temperature limits may damage the stationary phase.

Chromatography

Columns

Temperature Programming

The column sits in an oven. If the temperature is held constant during the entire analysis it is isothermal.
If you vary the temperature during the analysis, you typically use a temperature program.

With homologues, the retention time increases exponentially with the number of carbon.
As t_R increases, width increases and the height decreases, making detection impossible after a few peaks have eluted.

Chromatography

Columns

Temperature Programming

General steps to create a program assuming that the separation is possible.

- Determine initial temperature and time based on best possible separation of first few peaks.

- Repeat 1 for the last few peaks to find the best final temperature and time.

- Experiment with various ramps to account for the rest of the components.

Chromatography

Columns

Capacity

Column capacity is the maximum amount of a solute that can be introduced into a column before significant peak distortion occurs. No damage occurs if a column is overloaded.

Column capacity is related to stationary phase polarity, film thickness, column diameter and solute retention. Higher column capacities are obtained for solutes that are similar in polarity to the stationary phase.

For example, a polar column has a higher capacity for a polar solute than a non-polar solute.

Thicker film and wider diameter columns have higher capacities.

Chromatography

Columns

Stationary Phase Film Thickness

Stationary phase loadings were 20-30% w/w prior to 1970's.

Now loadings are in the order of 1-3%.

Film thickness, d_f, has a direct effect on the retention, sample capacity and elution temperature.

For best column efficiency, the film thickness is kept as thin as possible in order to reduce resistance to mass transfer in the stationary phase, C_s.

Chromatography

Columns

Column Capacity in ng

Film Thickness (µm)	Column Diameter (mm)			
	0.18-0.20	**0.25**	**0.32**	**0.53**
0.10	20-35	25-50	35-75	50-100
0.25	35-75	50-100	75-125	100-250
0.50	75-150	100-200	125-250	250-500
1.00	150-250	200-300	250-500	500-1000
3.00		400-600	500-800	1000-2000
5.00		1000-1500	1200-2000	2000-3000

Chromatography

Columns

Film Thickness Selection

1. For 0.18-0.32 mm I.D. columns, a film thickness of 0.18-0.25 µm is average or standard (i.e., not thin or thick) and used for most analyses.

2. For 0.45-0.53 mm I.D. columns, a film thickness of 0.8-1.5 µm is average or standard (i.e., not thin or thick) and used for most analyses.

3. Thick film columns are used to retain and resolve volatile solutes (e.g., light solvents, gases). Thick columns are more inert and have higher capacities. Thick film columns exhibit higher column bleed and decreased upper temperature limits.

4. Thin film columns are used to minimize the retention of high boiling, high molecular weight solutes (e.g., steroids, triglycerides). Thin columns are less inert and have lower capacities. Thin film columns exhibit lower column bleed.

Chromatography

Columns

Column Length

Column length influences three parameters:

- efficiency,

- retention (analysis time) and

- pressure.

Chromatography

Columns

Column Length

Column efficiency (N) is proportional to column length.

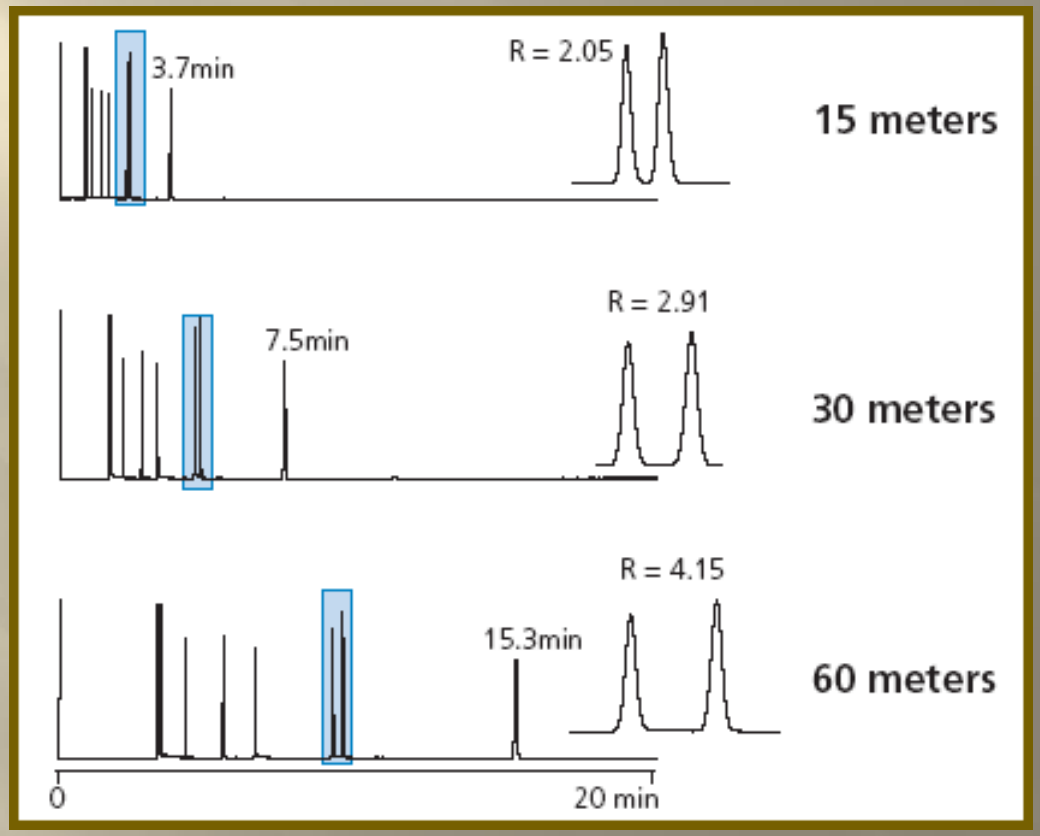
Resolution is a square root function of the theoretical plate number. For example, doubling column length (thus efficiency) theoretically increases resolution by 1.41 times (closer to 1.2-1.3 times in real practice).

Longer columns are used when peak separation is small and high column efficiency (i.e., narrow peaks) is needed.

Chromatography

Columns

Column Length



Longer columns are used when peak separation is small and high column efficiency (i.e., narrow peaks) is needed.

Chromatography

Columns

Column Length Selection

1. Start with 25-30 meter columns when the best length is unknown.

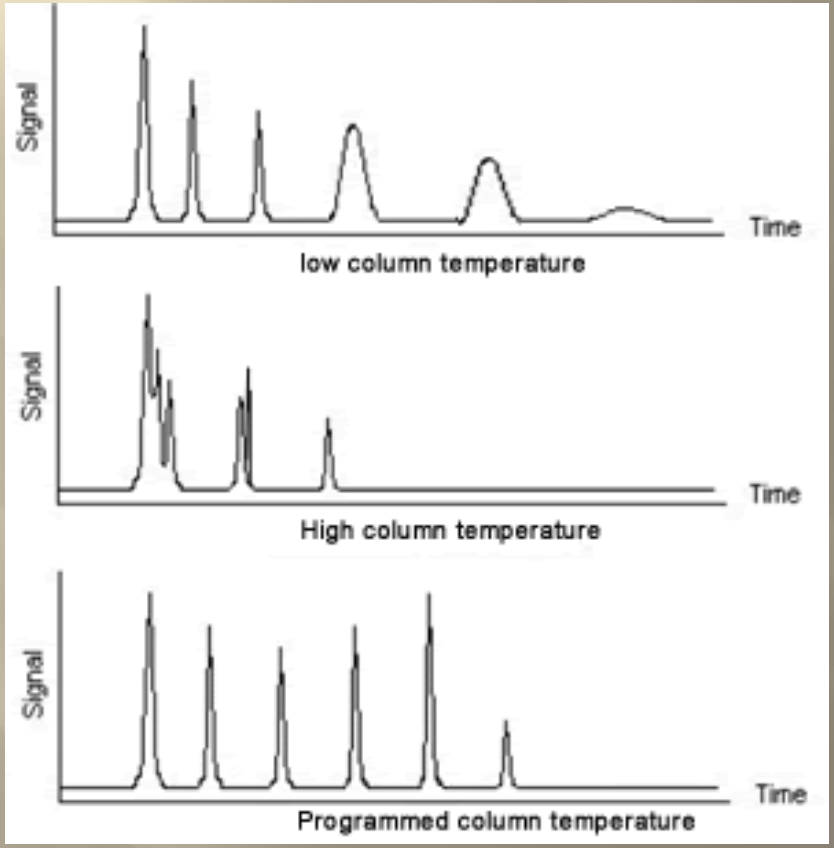
2. 10-15 meter columns are well suited for samples containing very well separated solutes or very few solutes. Shorter lengths are used for very small diameter columns to reduce head pressures.

3. 50-60 meters should be used when resolution is not possible by other means (smaller diameter, different stationary phase, change in column temperature). Best suited for complex samples containing a large number of solutes. Long columns have long analysis times and higher cost.

Chromatography

Columns

Column Temperature Programming



Chromatography

Columns

Stationary Phase Selection

Usually historically based – do what someone else did.

Selecting the Stationary Phase

Consider:

- **polarity**
- **temperature limits**
- **column efficiency**
- **detector compatibility.**

Chromatography

Columns

Column Selection

Non-polar phases usually exhibit superior lifetimes – so best to select least polar phase needed for a particular separation.

Most difficult factor to access is the ability of a phase to produce a good separation. In theory selection is based on knowing the types of interactions.

Chromatography

Columns

Column Selection

Types of interactions:

- London or dispersive forces (weak, non-specific). Temporarily induced dipole. As molecular weight increases dispersive forces increase (electrons further from nucleus)
- Dipole-dipole interactions (polar molecules)
- Acid – Base interactions (proton sharing or electron transfer) (hydrogen bonding)

-Dispersion is the dominant interaction for all polysiloxane and polyethylene glycol stationary phases.

-Dispersion can be simplified into the concept of volatility. The more volatile a solute, the faster it elutes from the column (i.e., shorter retention time).

Chromatography

Columns

Types of interactions

- Empirical results have shown that dipole interaction stationary phases are well suited for samples containing compounds that have base or central structures to which different groups are attached in various positions.

- Examples include substituted aromatics, halocarbons, pesticides and drugs.

Chromatography

Columns

Types of interactions

- **The hydrogen bonding interaction occurs if there is hydrogen bonding between the solute molecules and the stationary phase.**
- **It is the difference in the strength of the hydrogen bonding that is critical.**

Relative Hydrogen Bonding Strengths

Strength	Compounds
Strong	Alcohols, carboxylic acids, amines
Moderate	Aldehydes, esters, ketones
Weak to None	Hydrocarbons, halocarbons, ethers

Chromatography

Columns

Stationary phase interaction

Functional Group	Dispersion	Dipole	Hydrogen Bonding
Methyl	Strong	None	None
Phenyl	Very Strong	None	Weak
Cyanopropyl	Strong	Very Strong	Moderate
Trifluoropropyl	Strong	Moderate	Weak
PEG	Strong	Strong	Moderate

Chromatography

Columns

Each chromatographic setup will vary to some degree.

Retention times for a known set of species can be hard to reproduce from one lab to another or even one instrument to another.

Retention indexing helps to standardize your results.

Chromatography

Columns

Column Selection

Methods to characterize stationary phases (Chromatographic systems):

Kovats Index/Rohrschneider Constants/McReynolds Constants

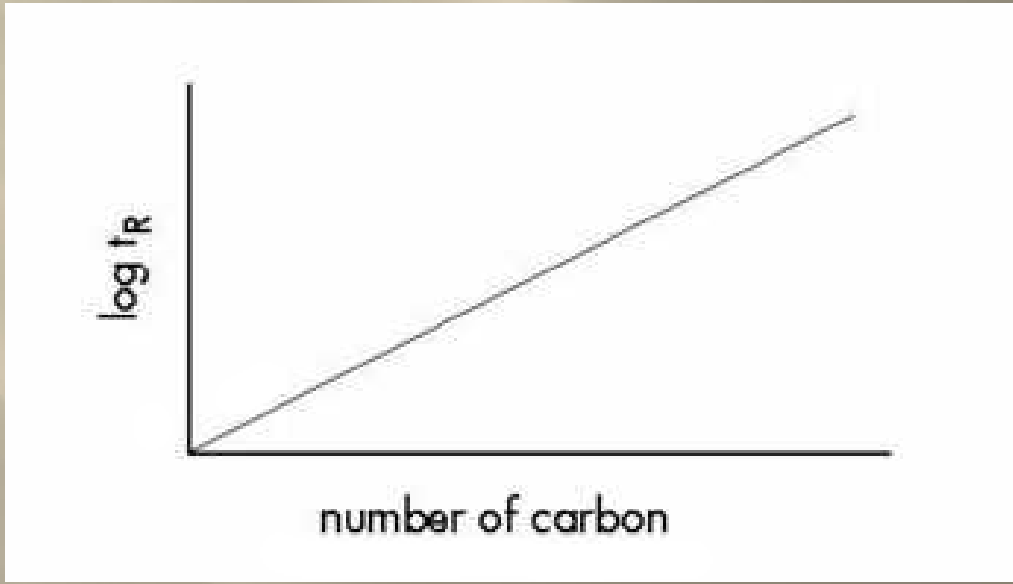
Use the relative retention of a solute – plot of the logarithm of the adjusted retention time versus Kovats retention index, I (linear).

Chromatography

Columns

Method is based on results of a homologous series.

Plot of the logarithm of the adjusted retention time versus Kovats retention index, I (linear).



Chromatography

Columns

The Kovat index value for any material can be found by assigning the retention index (I) of an n-alkane to equal a value 100 times its carbon number.

n-octane = 800
n-decane = 1000
n-dodecane = 12000, etc.

An I value of something different than n-alkane can be determined by "spiking" the mixture with n-alkanes. Plot log t'_r vs. I (or use equation) and I of solute is determined.

Chromatography

Columns

Equation:

$$I_x = 100Z + \frac{100(\log t'_{r,x} - \log t'_{r,z})}{\log t'_{r,z+1} - \log t'_{r,z}}$$

where,

Z - carbon # of n-alkane.

$t'_{r,x}$ - adjusted retention time of component under consideration.

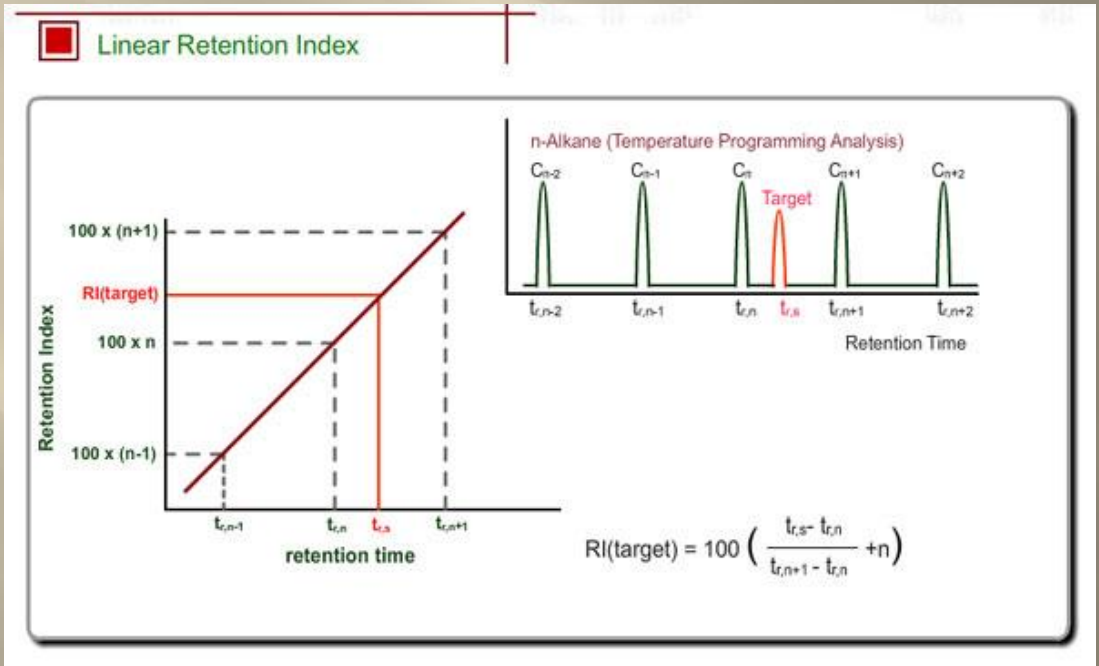
$t'_{r,z}$ - adjusted retention time of alkane eluting before x

$t'_{r,z+1}$ - adjusted retention time of alkane eluting after x.

Chromatography

Columns

All that is really being done is to normalize each component compared to n-alkanes. It assumes that you are dealing with either identical or at least very similar columns or packings.



Chromatography

Columns

Methods to evaluate a wide range of phases are based on measuring performance for a set of representative substances.

Can then tell if two phases should give comparable performance or if a phase is better for specific functional groups.

McReynolds Constants or Rohrschneider Constants

Chromatography

Columns

Use a series of reference compounds.

McReynolds used 10 probe solutes – each with a different functionality to measure a specific interaction with a liquid phase. He measured I for the 10 compounds on over 200 phases, including squalene (serves as a reference).

Calculated ΔI value for each probe = $I_{liquid\ phase} - I_{squalene}$.

Chromatography

Columns

As ΔI increases for a probe on a given liquid phase, the degree of a specific interaction associated with the probe increases.

The probes are assigned symbols of X', Y', Z', U', S' (Rohrschneider Constants) and McReynolds expanding it adding H', J', K', L', M'.

For squalene all = 0

Chromatography

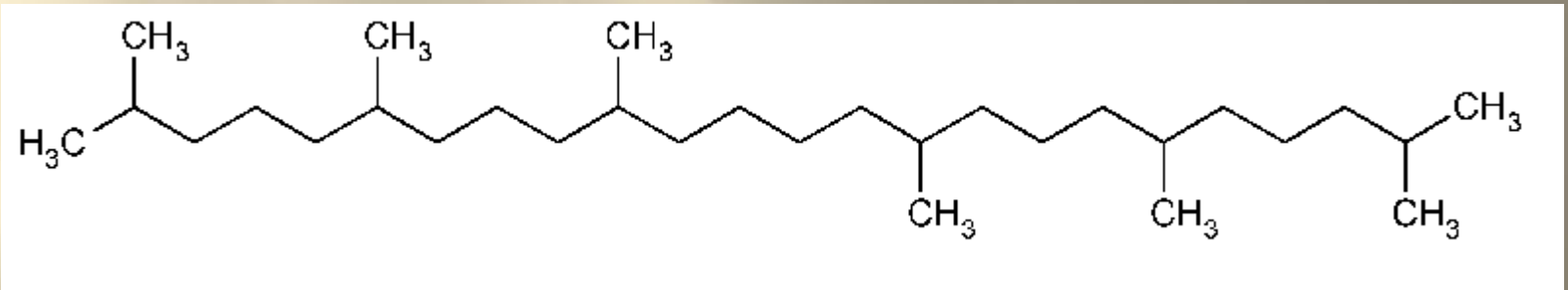
Columns

Group	Substance	Symbol
aromatic, olefinic	benzene	X'
alcohols, phenols, acids	1-butanol	Y'
ketones, ethers, esters, aldehydes	methyl-n-propyl ketone	Z'
nitro, nitriles	nitropropane	U'
bases, aromatic hetrocyclics	pyridine	S'

Chromatography

Columns

Squalane is used as the reference material and all other packings are normalized to it.



Packing	TMAX	X'	Y'	Z'	U'	S'
squalane	150	0	0	0	0	0
SE-30	350	15	53	44	64	41
OV-7	350	69	113	111	171	128
Carbowax 20M	250	322	536	368	572	59

Chromatography

Columns

TABLE 3.4 Probes Used in the McReynolds and Rohrschneider Classifications of Liquid Phases

Symbol	McReynolds Probe	Rohrschneider Probe	Measured Interaction
X′	Benzene	Benzene	Electron density for aromatic and olefinic hydrocarbons
Y′	*n*-Butanol	Ethanol	Proton donor and proton acceptor capabilities (alcohols, nitriles)
Z′	2-Pentanone	2-Butanone	Proton acceptor interaction (ketones, ethers, aldehydes, esters)
U′	Nitropropane	Nitromethane	Dipole interactions
S′	Pyridine	Pyridine	Strong proton acceptor interaction
H′	2-Methyl-2-pentanol	—	Substituted alcohol interaction similar to *n*-butanol
J′	Iodobutane	—	Polar alkane interactions
K′	2-Octyne	—	Unsaturated hydrocarbon interaction similar to benzene
L′	1,4-Dioxane	—	Proton acceptor interaction
M′	*cis*-Hydrindane	—	Dispersion–interaction

Assignment

- Test II

Chem 5570