THE RESTEK ADVANTAGE

Analyzing Free Fatty Acids Without Derivatization[1]

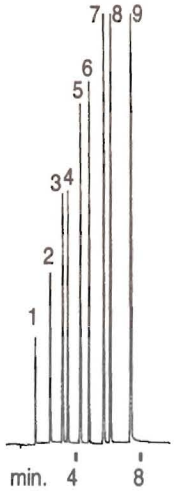
Dietary intake of fats has become an important concern for the health conscious public. Therefore, laboratories involved in analyzing food products are often faced with the difficult task of analyzing fatty acids. The lack of volatility coupled with their adsorptive nature has complicated gas chromatographic analysis of free fatty acids. As a result, many analysts derivatize fatty acids to their methyl ester state. Derivatization increases the volatility of free acids and reduces their activity making them more amenable to GC analysis. However, derivatization adds additional sample prep costs and some uncertainty to the analysis due to the possibility for sample loss or incomplete methylation. Therefore, many analysts would prefer to analyze fatty acids in their free state.

Volatile Fatty Acids

Several capillary column techniques have been developed for the analysis of volatile fatty acids. One approach is to use very thick film non-polar stationary phases for analyzing volatile free acids. Thick film columns are necessary to increase the sample capacity of non-polar phases since they have little affinity for highly polar acids. Figure 1 shows the analysis of C_2 to C_7 organic acids on a 30 meter, 0.53mm ID, 5.0µm Rtx®-1 column. Excellent separation of these volatile acids can be achieved in less than 8 minutes with minimal peak tailing. Another approach for analyzing volatile fatty acids is to use

Figure 1 - A thick film Rtx®-1 column resolves lower molecular weight free fatty acids.

1	acetic acid
2	propionic acid
3	isobutyric acid
4	n-butyric acid
5	isovaleric acid
6	n-valeric acid
7	isocaproic acid
8	caproic acid
9	heptanoic acid



30m, 0.53mm ID, 5.0µm Rtx®-1 (cat.# 10179)
0.2µl injection of a 10-20ng/µl free fatty acid standard in water
Direct injection using a Uniliner®.
Oven temp.: 60°C to 180°C @ 15°C/min.
Inj. & det. temp.: 250°C **Carrier Gas:** hydrogen
Linear velocity: 50cm/sec. (flow rate: 6cc/min.)
FID sensitivity: 4 x 10^{-11} AFS

(article is continued on page 2)

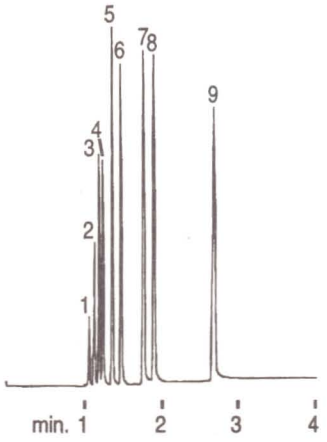
in this issue...

Another approach for analyzing volatile fatty acids is to use highly polar bonded Carbowax® capillary columns that have been deactivated for acidic compounds. The strong affinity of the acids on this polar column results in excellent separation of peaks, even with a 15 meter column. The acidic deactivation of the Carbowax® column produces excellent peak shape for these free fatty acids.

Moderately polar stationary phases can also be used for the analysis of volatile free fatty acids. Figure 2 shows the analysis of C_2 to C_7 organic acids on a 30 meter, 0.25mm ID, 0.25µm Rtx®-200 column. This trifluoropropylmethyl stationary phase shows good separation and peak shape of these volatile acids and the analysis time is less than 3 minutes, making the Rtx®-200 an excellent screening column.

Figure 2 - Fast analysis of low molecular weight free fatty acids can be achieved with an Rtx®-200 column.

(same peak list as Figure 1)



30m, 0.25mm ID, 0.25µm Rtx®-200 (cat.# 15023)
0.8µl split injection of a free fatty acid standard
Concentration approximately 10 to 20ng/µl
Oven temp.: 90°C isothermal
Inj. & det. temp.: 250°C **Carrier Gas:** hydrogen
Linear velocity: 40cm/sec. (flow rate: 1.4cc/min.)
FID sensitivity: 4 x 10^{-11} AFS **Split vent:** 40cc/min.

Saturated & Unsaturated Fatty Acids

Polar stationary phases are also capable of resolving saturated and unsaturated fatty acids. Figure 3 shows the analysis of saturated and unsaturated fatty acids on a 30 meter, 0.53mm ID, 0.25µm Stabilwax®-DA column. Palmitic acid (C16:0) can easily be resolved from Palmitoleic acid (C16:1), and Stearic acid (C18:0) can be resolved from Oleic (C18:1), Linoleic (C18:2), and Linolenic acid (C18:3) on this column.

Methylation of fatty acids for GC analysis may not always be necessary. Several options are available for analyzing fatty acids in their free form using capillary columns. Column selection will depend on the molecular weight range of fatty acids and the resolution required to separate saturated from unsaturated forms.

Product Listing

Rtx®-1
30m, 0.53mmID, 5.0µm
cat.#10179

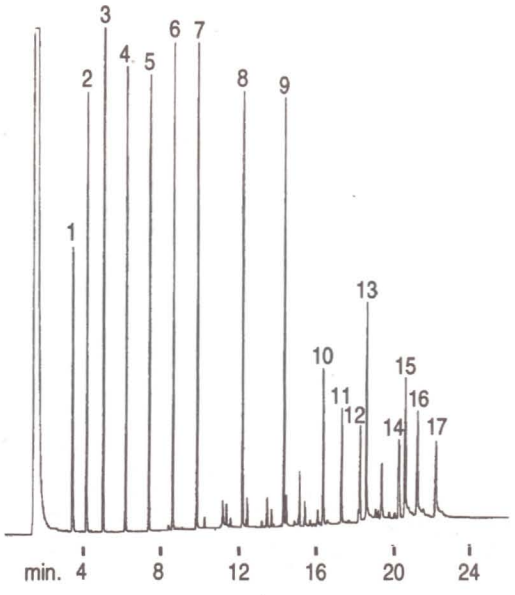
Rtx®-200
30m, 0.25mmID, 0.25µm
cat.#15023

Stabilwax®-DA
15m, 0.53mmID, 1.0µm
cat.#11052

30m, 0.53mmID, 0.25µm
cat.#11025

[1]This article is a partial reprint from *The Restek Advantage*, Vol. 3 No. 5, September 1992, p. 8-9.

Figure 3 - Saturated and unsaturated free fatty acids can be separated on a 30-meter Stabilwax®-DA column.



1	C2:0 - acetic acid	10	C14:0 - myristic acid
2	C3:0 - propionic acid	11	C15:0 - pentadecanoic acid
3	C4:0 - butyric acid	12	C16:0 - palmitic acid
4	C5:0 - valeric acid	13	C16:1 - palmitoleic acid
5	C6:0 - caproic acid	14	C18:0 - stearic acid
6	C7:0 - enanthic acid	15	C18:1 - oleic acid
7	C8:0 - caprylic acid	16	C18:2 - linoleic acid
8	C10:0 - capric acid	17	C18:3 - linolenic acid
9	C12:0 - lauric acid		

30m, 0.53mm ID, 0.25µm Stabilwax-DA (cat.# 11025)
0.5µl direct injection of a 5mg/ml standard.
Oven temp.: 100°C (hold 2 min.) to 250°C @ 8°C/min.
Inj. & det. temp.: 280°C **Carrier Gas:** helium
Linear velocity: 40cm/sec. (flow rate: 5.2cc/min.)
FID sensitivity: 8 x 10^{-11} AFS

Cover Your Bases with Rtx®-5 Amine Columns

The Rtx®-5 Amine column has been developed with a unique deactivation technology that reduces adsorption and improves the response of basic compounds. Analyses that previously required derivatization or another analytical technique such as HPLC can now be performed on the Rtx®-5 Amine column. Clinical chemists can now save time and money when analyzing pharmaceutical compounds with the Rtx®-5 Amine. (Related article on page 5.) Because basic samples can be analyzed in their free base or salt form with the Rtx®-5 Amine, derivatization is not necessarily required. This column can also be used to analyze neutral and mildly acidic compounds with the same efficiency as our standard Rtx®-5 columns.

The Rtx®-5 Amine can be used to analyze a wide variety of basic compounds such as alkylamines, diamines, triamines, and nitro-containing heterocycles. Additionally, the Rtx®-5 Amine permits the analysis of neutral and weakly acidic analytes. Figure 1 shows the simultaneous analysis of several basic compounds and phenols in water. Excellent peak symmetry of both the basic and acidic compounds is obtained with the Rtx®-5 Amine.

Chemists using guard columns for the analysis of basic compounds frequently experience peak tailing and low recovery. This problem occurs because the deactivated surface of the guard column can be adsorptive to basic compounds. Restek now offers base deactivated guard columns for a completely inert, basic pathway for basic compound analysis. These base deactivated guard columns are made with the same technology as the Rtx®-5 Amine columns to guarantee excellent response of basic compounds.

If your lab is analyzing amines or other strongly basic compounds, the Rtx®-5 Amine is guaranteed to give you consistent, reproducible results from run-to-run. The Rtx®-5 Amine can also be used for samples where both basic and weakly acidic compounds must be analyzed. Choose Rtx®-5 Amine columns and base deactivated guard columns for your next basic compound analysis. These products were designed to meet your chromatographic challenges!

Product Listing

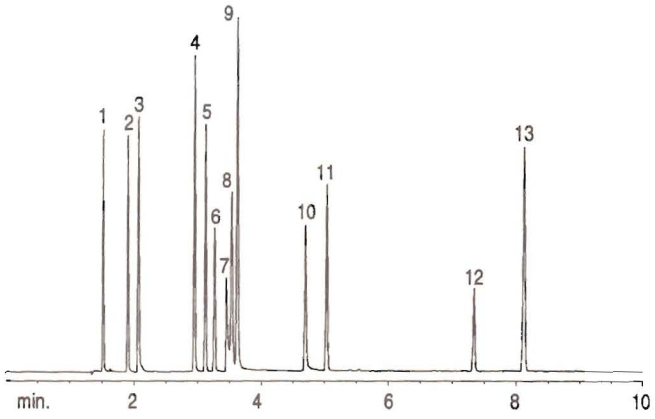
Rtx®-5 Amine Columns

	df(µm)	15-meter	30-meter
0.25mm ID	0.50	12335	12338
	1.00	12350	12353
0.32mm ID	1.00	12351	12354
	1.50	12366	12369
0.53mm ID	1.00	12352	12355
	3.00	12382	12385

New! Base Deactivated Guard Columns

5m, 0.25mm ID	cat.# 10000, each
5m, 0.32mm ID	cat.# 10001, each
5m, 0.53mm ID	cat.# 10002, each

Figure 1 - The versatility of the Rtx®-5 Amine column permits the analysis of both basic and weakly acidic compounds - simultaneously!



1. diethylamine
2. pyridine
3. morpholine
4. phenol
5. aniline
6. 2-chlorophenol
7. diethylenetriamine
8. octylamine
9. 1-methyl-2-pyrrolidinone
10. 2-nitrophenol
11. 2,6-dimethylaniline
12. nicotine
13. 2-nitroaniline

30m, 0.32mm ID, 1.0µm Rtx®-5 Amine column (cat.# 12354)
1.0µl split injection of miscellaneous amines and phenols in water, on-column concentration: 22ng

Oven temp.:	120°C to 220°C @ 10°C/min.		
Inj. & det. temp.:	305°C	**Detector:**	FID
Carrier gas:	hydrogen	**Linear velocity:**	38cm/sec. set @ 120°C
Detector sensitivity:	6.4 x 10^{-11} AFS	**Split ratio:**	25:1

Clinical Corner

Analysis of Antiepileptic Drugs by Capillary GC

Epilepsy is a central nervous system disorder characterized by seizures resulting from excessive electrical discharges in the brain. Epileptic syndromes can be categorized into two main divisions. Partial seizures are limited to one part of the brain and are manifested by convulsions of single muscles or muscle groups. Generalized seizures affect the entire brain and can result in varying degrees of impaired consciousness and major convulsions that involve all body muscle groups. Status epilepticus is a prolonged period of seizure activity, either partial or generalized, during which normal consciousness is not regained, usually greater than 30 minutes[1]. Untreated status epilepticus is associated with higher morbidity rates and can result in permanent brain damage.

Measuring concentrations of antiepileptic drugs in serum or plasma is especially important in establishing control of seizure activity, monitoring patient compliance and minimizing side effects.

Drugs used to control epileptic disorders focus on inhibiting the initial discharge of electrical stimuli from seizure foci or preventing the spread of uncontrolled electrical activity to other neurons[2]. Choice of drug therapy to control convulsions is dependent upon the type of seizure experienced by the patient as well as other associated clinical factors. Hydantoins, succinimides, barbiturates and a few specific, non-related compounds make up the bulk of drugs used in the long term treatment of epileptic disorders.

All of the antiepileptic drugs have polar characteristics through the inclusion of keto, azo and carboxylic acid functional groups. Absence seizures are treated most frequently with ethosuximide or valproic acid. However, concerns over hepatotoxicity limit the use of valproic acid in children. Carbamazepine and diphenylhydantoin are the agents of choice in the treatment of generalized tonic-clonic seizures.

Measuring concentrations of antiepileptic drugs in serum or plasma is especially important in establishing control of seizure activity, monitoring patient compliance and minimizing side effects. Therapeutic concentrations of most antiepileptic drugs are in the µg/ml range and extraction of antiepileptic drugs from biological matrices is straightforward. Low limits of detection are easily achieved with commonly used detectors such as FID.

Despite being characterized as polar compounds, antiepileptic drugs can be analyzed without being derivatized. Intermediate polarity columns are recommeded over non-polar columns to obtain symmetrical peak shapes and maximum resolution. Figure 1 (on page 5) shows the analysis of a set of antiepileptic drugs on an Rtx®-20 column. All compounds exhibit good peak shape and resolution except for carbamazepine and diphenylhydantoin, which are only partially resolved. By using another intermediate polarity column with a different selective mode of retention, this coelution can be resolved without compromising the resolution of any of the other compounds in this group. Figure 2 (on page 5) shows the analysis of the same group of antiepileptic drugs using the same chromatographic conditions on an Rtx®-1701 column. Changes in retention time and relative elution order allow both of these columns to be used for screening and confirming the presence of antiepileptic drugs in a dual column configuration.

Monitoring the concentrations of antiepileptic drugs is critical in providing optimal drug therapy for epileptic patients. Capillary gas chromatography provides a quick and versatile method to screen and quantitate a wide variety of antiepileptic drugs. Intermediate polarity columns like the Rtx®-20 and the Rtx®-1701 provide the best overall peak shape and resolution for these compounds.

References

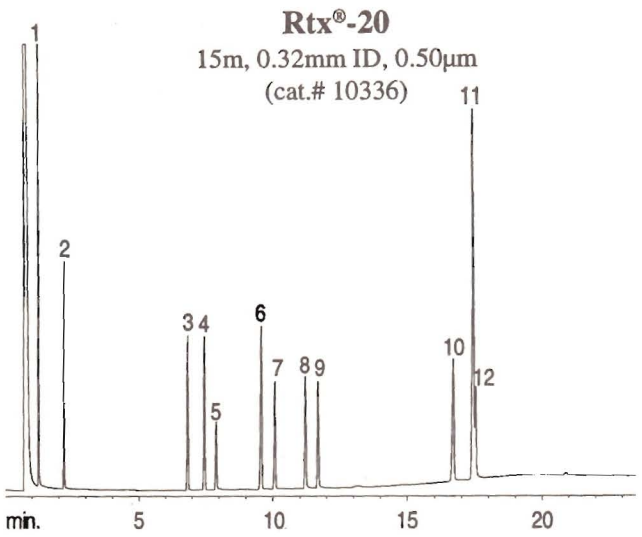
1. Martindale, *The Extra Pharmacopeia*, 30th Edition, 1993, p. 292.
2. Goodman and Gillman's, *The Pharmacological Basis of Therapeutics*, 8th Edition, p. 438-439.

Product Listing

Rtx®-20
15m, 0.32mm ID, 0.50µm
cat.# 10336, each

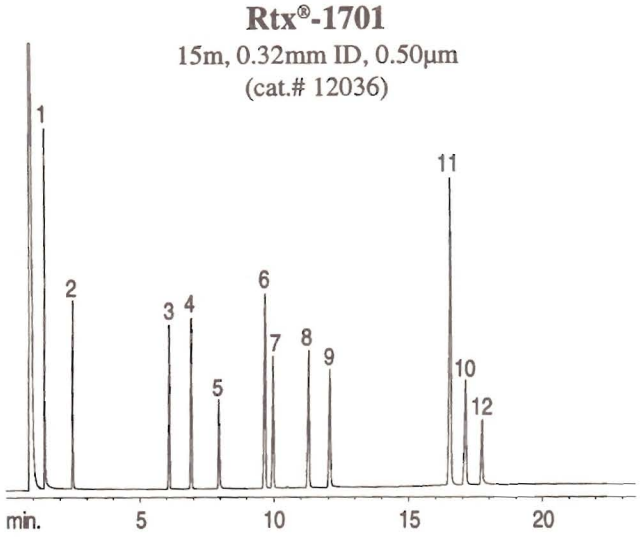
Rtx®-1701
15m, 0.32mm ID, 0.25µm
cat.# 12036, each

Figure 1 - The Rtx®-20 shows good peak shape and resolution for all compounds in the antiepileptic drug sample except for a coelution of carbamazepine and diphenylhydantoin.



COMPOUNDS	
1. valproic acid	7. ethotoin
2. ethosuximide	8. PEMA
3. methsuximide	9. phenobarbital
4. phensuximide	10. primidone
5. desmethyl methsuximide	11. carbamazepine
6. mephenytoin	12. diphenylhydantoin

Figure 2 - Analysis of the same compounds on an Rtx®-1701 resolves the coelution of carbamazepine and diphenylhydantoin without compromising resolution of the other compounds.



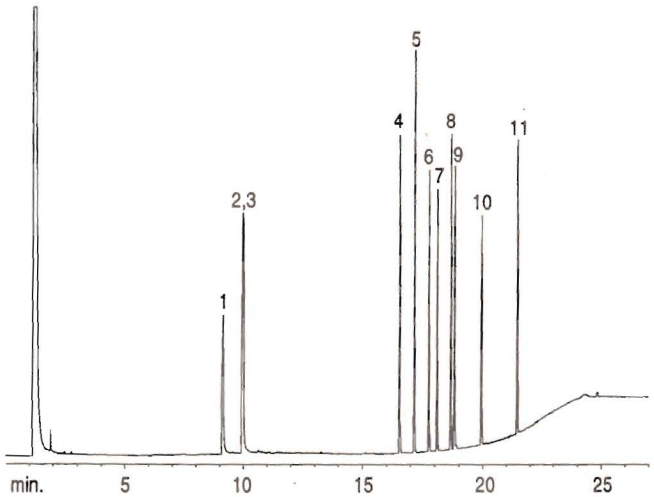
1.0µl split injection of Antiepileptic Drugs
on-column concentration: 30ng

Oven temp.:	150°C to 280°C @ 7°C/min. (hold 5 min.)
Inj./det. temp.:	250°C/270°C
Detector:	FID **Carrier gas:** helium
Linear velocity:	30cm/sec. set @ 150°C
Detector sensitivity:	1.28 x 10^{-10} AFS
Split ratio:	30:1

Analysis of Antihistamines on an Rtx®-5 Amine Column

Antihistamines are common ingredients in over-the-counter cold medicines. Antihistamines typically have amine functional groups in their structure and are classified as basic compounds. Because of their basic nature, antihistamines often exhibit adsorption or tailing peaks on poorly deactivated columns. With the Rtx®-5 Amine, excellent peak symmetry and enhanced response is obtained for eleven antihistamines in their salt form, as illustrated in Figure 1.

Figure 1 - Analyze antihistamines in their salt form on the Rtx®-5 Amine.



COMPOUNDS	
1. phenylpropanolamine	7. phenyltoloxamine
2. ephedrine	8. methapyrilene
3. pseudoephedrine	9. chlorpheniramine
4. Pheniramine	10. brompheniramine
5. diphenhydramine	11. triprolidine
6. doxylamine	

30m, 0.32mm ID, 1.0µm Rtx®-5 Amine column (cat.# 12354)
1.0µl split injection of antihistamines in methanol,
on-column concentration: 22ppm

Oven temp.:	130°C (hold 5 min.) to 305°C @ 10°C/min. (hold 5 min.)
Inj. & det. temp.:	305°C
Detector:	FID
Carrier gas:	hydrogen
Linear velocity:	43.3cm/sec. set @ 130°C
Detector sensitivity:	6.4 x 10^{-11} AFS
Split ratio:	27:1

See page 3 for more information on the Rtx®-5 Amine column.

Rtx®-1 - A New Bonded Packed Column for Simulated Distillation

- Rtx®-1 bonded packed column requires minimal conditioning.
- Meets or exceeds all specifications of ASTM D2887-93.
- Stable baseline to 350°C and repeatable RT's "right from the box".
- Deactivated Silcosteel® tubing and Silcoport™ packing for high inertness.
- Column lifetime superior to existing Sim Dist columns.

Simulated Distillation (Sim Dist), according to The American Society for Testing Materials (ASTM) test method D2887-93, can be performed using either packed or capillary columns. Some of the advantages of capillary columns are the columns are preconditioned so hey can be used after only minimal conditioning, and the bonded stationary phases exhibit stable baselines and retention times. There are many laboratories, currently using packed columns, which would like to take advantage of bonded phases but do not have GC equipment which can be easily converted for use with capillary columns. Restek's Rtx®-1 Sim Dist column is the first in a new generation of bonded packed columns having superior inertness and stability compared to conventional packed columns. Improvements are obtained by preparing the columns with Silcosteel® tubing and bonding the Rtx®-1 stationary phase to a highly deactivated Silcoport™ support. The column dimensions and packing (1/8" Silcosteel® with 10% Rtx®-1 on Silcoport™) are designed to exceed all requirements specified in ASTM Test Methods D2887-93 and D3710-93.

Table I - Retention Time Repeatability for Calibration after only 30 minutes conditioning.

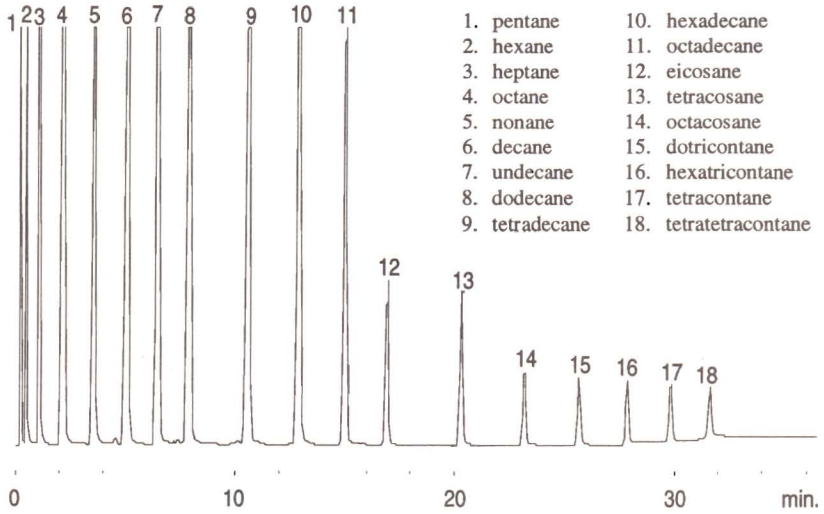
Hydrocarbon	Min Rt	Max Rt	Avg. RT	Stand. Dev.
C_5	0.241	0.243	0.242	0.001
C_6	0.493	0.497	0.495	0.002
C_{10}	5.746	5.765	5.752	0.005
C_{20}	18.482	18.491	18.486	0.004
C_{28}	25.093	25.103	25.098	0.004
C_{40}	32.160	32.171	32.166	0.004
C_{44}	34.316	34.328	34.326	0.007

n=9

Bonded stationary phases require minimal conditioning and give stable baselines and retention times "right from the box"

Simulated distillation is a gas chromatographic procedure which differs from typical GC analyses requiring peak resolution and integration. The sample is analyzed using a linear temperature program so that the retention time of the hydrocarbons are proportional to their boiling points. The sample boiling range distribution is calculated by comparing the sample area and its retention time with that of an alkane calibration standard. In order for the calibration to be valid for sample analysis, it is crucial retention times be repeatable until the next calibration is performed. Figure 1 is an example of the analysis of the Restek D2887 Calibration Mix (cat.# 31222) illustrating the typical pattern obtained for the alkanes under temperature programmed conditions. To demonstrate the stability of the Rtx®-1 column, a series of calibration standards were analyzed after only 30 minutes of conditioning at 350°C. Table I shows the excellent retention time repeatability obtained with the column, indicating the column is suitable for sample analysis after minimal conditioning.

Figure 1 - C5 to C44 calibration analysis after only 30 minutes conditioning.

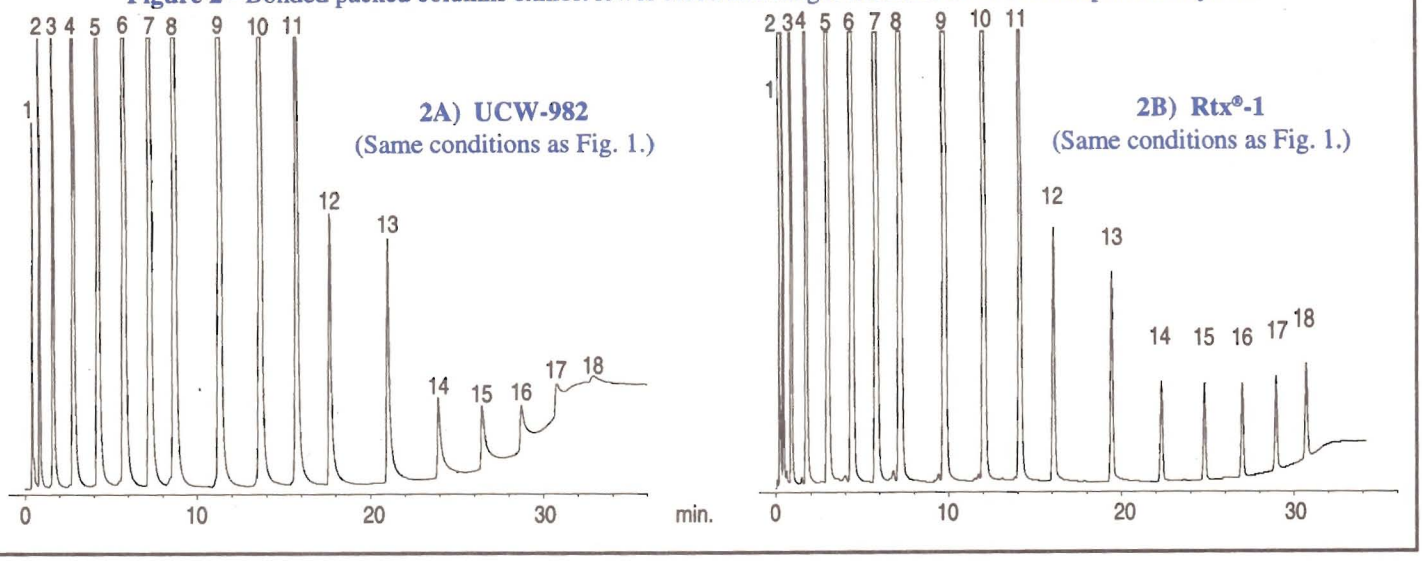


25" x 1/8" Rtx®-1 Sim Dist 2887 Silcosteel® column
1.0µl direct injection of D2887 Calibration Mix (cat.# 31222)

Oven temp.:	35°C to 350°C @ 10°C/min. (hold 5 min.)
Inj. & det. temp.:	350°C
Carrier gas:	helium @ 25ml/min.
FID sensitivity:	256 x 10^{-11} AFS

Column bleed is another important consideration for selecting a Sim Dist column. The baseline must be stable and free of any artifacts during the temperature program up to 350°C. Although baseline subtraction is permitted in the method, this compensation will produce errors if the baseline rise is not consistent. Conventional packed columns require up to 14 hours of conditioning and frequent updating of the baseline compensation run because the stationary phase is not bonded. Rtx®-1 columns, however, exhibit stable and reproducible baselines with just 30

Figure 2 - Bonded packed columns exhibit lower bleed and longer lifetimes after 170 temperature cycles.



minutes of conditioning. This results in fewer baseline blanks and less frequent calibration increasing laboratory productivity.

Bonded stationary phases extend column lifetime

The Rtx®-1 stationary phase is bonded to the diatomite particles resulting in an immobilized coating which is resistant to solvents and lower in bleed than conventional packings. Since the packing is preconditioned, there is no need for extended conditioning in the GC oven. This conditioning must be carried out in an oxygen-free system, otherwise high bleed will result. Since GC systems often have leaks or carrier gas which contain oxygen, it is more likely that conventional columns will be damaged during the conditioning process. Figure 2A shows a conventional UCW-982 column after only 170 temperature cycles, demonstrating higher bleed and more tailing than the Rtx®-1 Sim Dist column (Figure 2B). Although actual column lifetimes depend upon the system and types of samples analyzed, the bonded stationary phase should result in longer lifetime than its non-bonded equivalent.

Rtx®-1 Sim Dist columns have equivalent polarity to OV-101 and UCW-982

In order for a stationary phase to be acceptable for ASTM methods, the column must not exhibit selective retention for aromatic hydrocarbons compared to aliphatic hydrocarbons. This is an important test because if the polarity of a column is different, the boiling point results will demonstrate a bias, especially for highly aromatic samples. The "polarity" of the bonded Rtx®-1 column was compared with OV-101 and UCW-982, two of the most common stationary phases currently used for simulated distillation. The results of the calculated boiling points for aromatics compared to the published boiling points appear in Table II. All three silicone columns tested are essentially identical in they elute aromatics at a slightly lower temperature than the alkanes. This confirms the polarity of the Rtx®-1 column is equivalent, and the boiling range values obtained will agree with OV-101 and UCW-982 columns.

Table II - Comparison of bonded and conventional packed columns indicates no polarity differences.

Aromatic Hydrocarbon	Published BP[1] (°C)	Calculated BP		
		Rtx®-1	UCW-982	OV-101
benzene	80	81.3	82	80.3
p-xylene	139	138.6	140.2	137.7
naphthalene	218	204.6	206.9	204.3
acenaphthylene	280	252.7	255.6	252.2
anthracene	342	304.1	307.2	303.4
chrysene	447	385.6	389.2	384.9
dibenzo(a,h)anthracene	524	452.3	455.7	450.4

Rtx®-1 is an excellent choice for Sim Dist using packed columns

Simulated Distillation is one of the most common GC analyses performed in the petroleum laboratory. ASTM test methods D2887 and D3710 can be performed with either packed or capillary columns, but until now the benefits of bonded phases were available only to capillary users. The Rtx®-1 packed column uses a bonded stationary phase which is immobilized on Silcoport™, a specially deactivated support. The columns are prepared using Silcosteel® tubing for inertness unavailable with conventional metal tubing. Rtx®-1 bonded packed columns require minimal conditioning and give stable baselines and retention even after only 30 minutes of operation at 350°C. If your laboratory has been looking for a better solution to Sim Dist analysis, Restek's Rtx®-1 packed columns are the answer.

Product Listing

Rtx®-1 Sim Dist 25" x 1/8"
Silcosteel® Packed Column
cat.# 80000, each

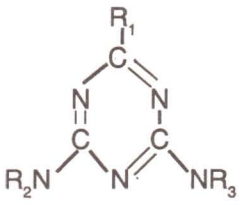
D2887 Calibration Mix
cat.# 31222, each cat.# 31322, 10-pk.

D3710 Calibration Mix
cat.# 31223, each cat.# 31323, 10-pk.

Analysis of Triazine Herbicides

Triazine herbicides are a class of herbicides that have risen in popularity and usage over the last decade. Because of their low toxicity towards mammals, they are not considered to be a threat to the environment. However, triazine herbicides are relatively stable and can be detected in the environment for long periods of time following their application. Due to their long residence time, monitoring for the presence of triazine herbicides has become a concern. The Environmental Protection Agency (EPA) has addressed these issues by drafting a method for the determination of triazine herbicides in industrial and municipal wastewater.

Figure 1 - Structure of diamino-s-triazine.



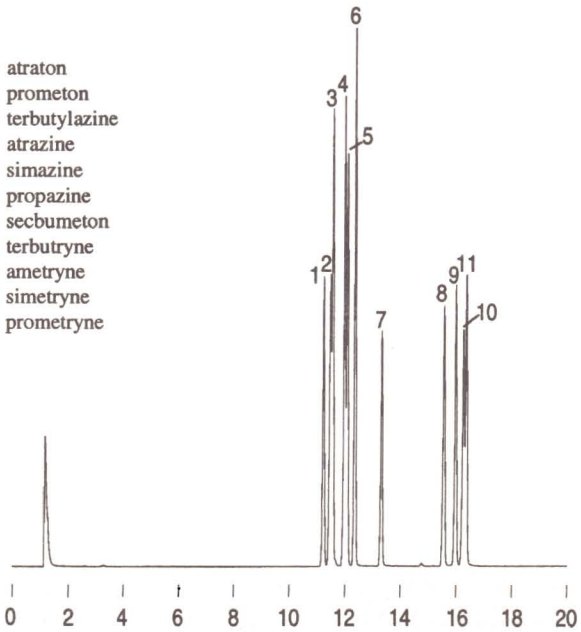
EPA Method 619 is a gas chromatographic method designed for the analysis of triazine herbicides using packed columns. This method can be modified for use with capillary columns. Resolving the eleven analytes for this method, even with highly efficient capillary columns, can be very difficult. The structures for all of the triazine herbicides are very similar and most common stationary phases do not offer the polarity or selectivity needed to obtain efficient resolution of these compounds. By matching the chemical characteristics of these compounds to stationary phases with selective retention mechanisms, better peak shape and resolution can be achieved.

Triazine herbicides are referred to as s-triazines, meaning they are symmetrical in structure. Triazines that have greater bioactivity generally contain halogenated (R1) and diamino (R2 and R3) functionalities (Figure 1). These herbicides are based on a six membered ring containing nitrogen constituents at the 1,3 and 5 positions. The electron density resulting from the inclusion of the nitrogen in the ring and the diamino substituted groups imparts significant polarity to these compounds. The degree of polarity will change depending on the functional groups present at either the R1, R2 or R3 substitution sites. Columns containing stationary phases with intermediate polarity are better suited for these compounds.

Figure 2 shows the analysis on a Rtx®-50 column (50% methyl-50% phenyl polysiloxane). The Rtx®-50 is a common intermediate polarity stationary phase used for the analysis of pesticides and herbicides. It completely resolves five compounds and partially resolves the remaining six; thus, the Rtx®-50 can effectively be used for the primary analytical column. Figure 3 shows the same compounds analyzed on a Rtx®-200 column. The Rtx®-200 is a trifluoropropylmethyl polysiloxane that has unique selectivity for compounds containing lone pair electrons. The Rtx®-200 completely resolves four compounds, partially resolves five, with two remaining unresolved. The elution pattern for the Rtx®-200 differs greatly from the Rtx®-50 making it an excellent choice as a confirmational column.

Figure 2 - The Rtx®-50 is an excellent choice for the primary column in triazine analysis.

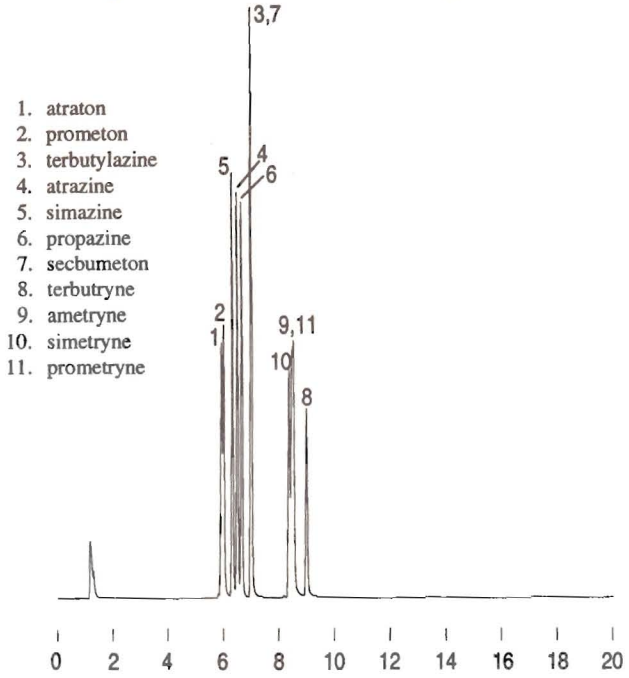
1. atraton
2. prometon
3. terbutylazine
4. atrazine
5. simazine
6. propazine
7. secbumeton
8. terbutryne
9. ametryne
10. simetryne
11. prometryne



Rtx®-50
30m, 0.53mm ID, 0.50µm
cat.# 10540, each

0.5µl direct injection of EPA 619 - triazine herbicides
on-column concentration: 50ng
Oven temp.: 150°C to 250°C @ 4°C/min. (hold 5 min.)
Inj./det. temp.: 250°C/275°C
Detector: TSD
Carrier gas: helium
Linear velocity: 40cm/sec. set @ 150°C

Figure 3 - The elution order change of the Rtx®-200 makes it a good secondary column choice.



Rtx®-200
30m, 0.53mm ID, 0.50µm
cat.# 15040, each

0.5µl direct injection of EPA 619 - triazine herbicides
on-column concentration: 50ng
Oven temp.: 150°C to 250°C @ 4°C/min. (hold 5 min.)
Inj./det. temp.: 250°C/275°C
Detector: TSD
Carrier gas: helium
Linear velocity: 40cm/sec. set @ 150°C

Used together, these columns combine to help the analyst positively identify and quantitate the compounds of interest. Since both columns are operated with the same temperature program conditions, simultaneous confirmation can be used. Several techniques for simultaneous dual column confirmation are available including a "Y" Press-Tight® Connector, a dual column direct injection tee, or a 2-hole ferrule.

Triazine herbicides are commonly applied to agricultural fields containing corn, apples, grapes, etc. Their stability increases their residence time in the environment, creating a general concern for their potential hazards. The Rtx®-50 and Rtx®-200 columns enhance the performance of triazine herbicide analyses. These columns offer a basis of quality that leads to accurate, reproducible results as required in EPA Method 619.

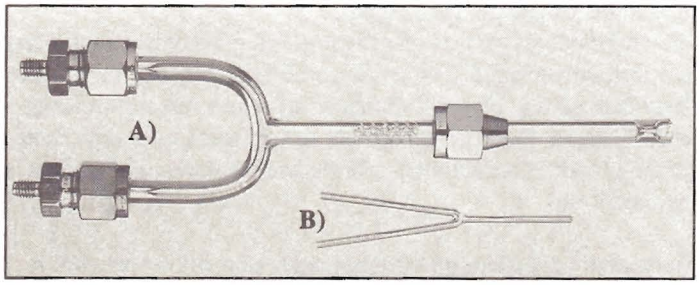
Reference
Kearney, P.C. and Kaufman, D.D. *Herbicides-Chemistry, Degradation, and Mode of Action.* 2ed., Marcel Dekker, Inc., 1969.

Product Listing

Rtx®-50
30m, 0.53mm, 0.50µm
cat.# 10540, each

Rtx®-200
30m, 0.53mm, 0.50µm
cat.# 15040, each

Products for Dual Column Analysis



A) Dual Column Direct Injection Tee Kit (includes all fittings/ferrules): cat.# 20412, kit
Replacement Tee: cat.# 20411, each
Replacement ferrules:

0.5mm ID graphite:	0.8mm ID graphite:
cat.# 20201, 10-pk.	cat.# 20202, 10-pk.
cat.# 20228, 50-pk.	cat.# 20224, 50-pk.

1/4" graphite cat.# 20210, 10-pk.

B) Universal "Y" Press-Tight® Connectors
- Split sample flow onto two different columns.
- Split a single column flow into two different detectors.
- Perform confirmational analysis with a single injection.

cat.# 20405, each
cat.# 20406, (3-pack)

Also Available:

Restek's new Dual Column *mini*-Lam™
Direct Injection Tee Kit
cat.# 20436, kit
Call your local distributor to request the October 1994 International Version of *The Restek Advantage* for more information.

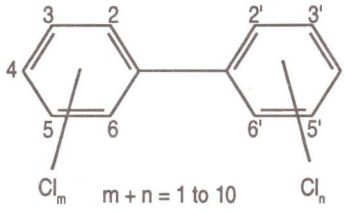
Polychlorinated Biphenyls in Used Engine Oil

Production of polychlorinated biphenyls began in the early part of this century and proved very useful in a variety of applications. They were used in transformers, paints, printing inks, and pesticides. PCBs are also by-products of many processes such as the manufacturing of chlorinated solvents and chlorinated benzenes. They were primarily synthesized by chlorination of biphenyl with chlorine gas.[1] The world's primary producer was Monsanto. They produced complex mixtures of PCBs under the tradename Aroclor®. A four digit numbering system was assigned to the mixtures. The first two numbers indicate the number of carbon atoms and the second numbers provide the weight percent of chlorine. PCBs were manufactured by other companies under trade names like Clophen (Bayer, GFR), Phenoclor (Caffaro, Italy), Fenclor (Prodelec, France), etc.[2] PCBs or Aroclors® are very stable species and do not readily degrade in the environment. Consequently, their wide use resulted in their regular presence in soil, water, and air. Due to this stability, they tend to be present in much of the food chain. The toxicity level of PCBs is still not clearly known--some being more lethal than others. In 1976, PCBs became regulated under the Toxic Substances Control Act[3] and disposal became a major consideration. Monsanto stopped producing Aroclors® in 1977.

Identification of PCBs

The general structure of PCBs is shown in Figure 1. There are 209 congeners, 10 homologs, and 1 to 46 isomers. The commercial production of PCBs resulted in mixtures or blends. Consequently, identification is based more on the type of mixture, eg., Aroclor® 1232, Aroclor® 1254 than on individual PCBs. This can be difficult and identification relies heavily on pattern recognition.

Figure 1 - Structure of PCBs.



Even though PCBs are relatively stable, some microbial and environmental degradation can occur making it more difficult to ascertain the parent Aroclor®. PCBs are hydrophobic and with their low vapor pressure tend to accumulate in soil sediments. In the soil, anaerobic microbes can attack the PCBs, dechlorinating them primarily at the meta and para positions. The extent of PCB dechlorination by microbial activity is dictated by environmental surroundings. Variables such as the presence of oils, grease, sulfur, and metals govern how and if anaerobic microbial dechlorination will occur.[4]

Environmental degradation involves photolysis. The monochlorobiphenyls can have half lives as little as a few days. On the other hand, the half life of pentachloro-biphenyl can be as long as a few months.[5] Any degradation can change the "fingerprint" of the Aroclor®, making identification a very difficult process.

Clean-up is Crucial

Since PCBs were widely used in a variety of applications and remain prominent in the environment, their presence is visible in almost every sample matrix--pesticide residue, milk, waste oils, fish, etc. Complicating a PCB blend with a waste oil matrix can make it virtually impossible to correctly identify and/or quantify. A series of clean-up steps are vital in order to properly identify the PCB mixture. A typical waste oil sample can include not only used motor oil, but sediment and other non-volatile components. The typical mode of PCB analysis is capillary GC using an Electron Capture Detector (ECD). In order to produce acceptable chromatography, the above interferents need to be removed and concentration levels should typically be in the ppb range. Even though the ECD is predominantly responsive to halogenated compounds, severe interferences, such as large amounts of hydrocarbons (ng concentrations), will disrupt the integrity of the chromatography. Figure 2 shows a 2µl injection of PCBs in waste oil with little sample preparation. Peak broadening is a dominant factor producing an almost packed column type chromatogram. There is no indication of an Aroclor® fingerprint. Figure 3 shows the same sample after several clean-up steps. Now the characteristic Aroclor® pattern is readily apparent. Both samples were spiked with decachlorobiphenyl as the internal standard. The 5-point calibration curve indicated the retention time of DCB to be 19.038 minutes (an additional amount of DCB was added to confirm the retention time of DCB). The chromatogram in Figure 2 shows a distinctive shift in the retention time of DCB indicating that significant amounts of interferents such as motor oils can cause peak shifting. This phenomenon can result in incorrect component identification and unreliable quantitation. After proper sample clean-up, the resulting chromatogram in Figure 3 can be properly identified and quantified. The retention time for DCB (19.038 minutes) is well within its retention window.

Common Clean-up Techniques

Several methods exist for the clean-up of PCB type samples and are well documented. The clean-up of waste oils can be particularly demanding. Figure 2 indicated a major source of interference from oil. Diluting the sample in hexane or isooctane, then mixing it with concentrated sulfuric acid removes much of the oil by oxidation.

Even after acid clean-up, contamination can still be present. It may be necessary to treat the sample with a magnesium silicate slurry (Florisil). Shake the sample mixture with Florisil, allow the magnesium silicate to settle, remove the final solution, and then analyze the sample. If the sample still exhibits interferences, the sample should undergo a silica gel slurry clean-up, following the same procedure as the Florisil method.

Figure 2 - Inadequate sample clean-up results in poor chromatography and loss of resolution.

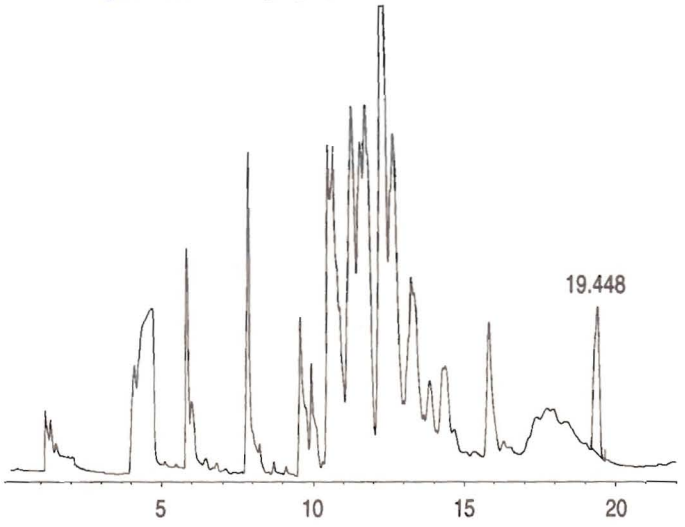
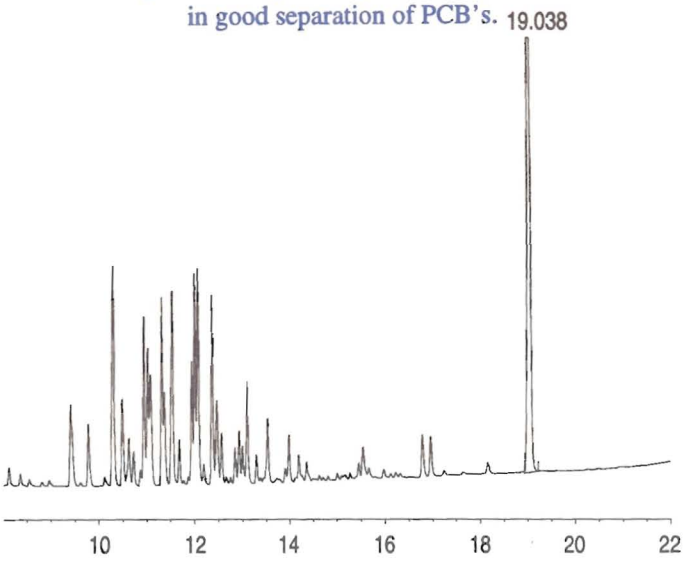


Figure 3 - Proper sample clean-up results in good separation of PCB's.



Another step that can be taken for dirty waste oils is a magnesium silicate column clean-up. A 10ml disposable pipet is plugged with glass wool, followed with anhydrous sodium sulfate, magnesium silicate, sodium sulfate, and moistened with some hexane. Filter the sample solution through the column. Collect the eluant in a vial. The sample should now be completely free of oil contamination.

If the sample also contains elemental sulfur, the impurity will be seen as a large peak on the chromatogram in the early portion of the analysis. Mixing the sample with copper powder or mercury and following it with an acid-wash will remove the sulfur. Depending on the degree of contamination some or all of these steps may be necessary to prepare the sample for analysis. More detail on these clean-up procedures is discussed in EPA Methods 3620, 3630, and 3660.

Column Choice

After clean-up, PCB samples can be effectively analyzed on a 30m, 0.32mm ID, 0.25µm XTI®-5 (5% phenyl / 95% dimethyl polysiloxane) capillary column. Since Aroclors® are very complex samples, a high resolution capillary column such as the XTI®-5 ensures that the Aroclor® will be identified correctly. The high degree of the XTI®-5 columns' reliability guarantees maximum resolution and efficiency. The thermal stability of the XTI®-5 (360°C maximum temperature) allows faster elution of higher boiling contamination.

The analysis of PCB mixtures can be additionally complicated by contamination, photodegradation of the PCBs, dechlorination by anaerobic microbes, and more than one Aroclor® present in the sample. Knowledge of the sample's origin, an effective clean-up procedure, and a high resolution capillary column will significantly decrease the problems associated with PCB analyses.

References:

1. Abramowicz, Daniel A., Brennan, Michael J., Van Dort, Heidi M., and Gallagher, Edith L., *Environ. Sci. Technol.*, Vol. 27, No. 6, 1993, p. 1125.
2. *Analytical Chemistry of PCBs*, Erickson, Michell D., Lewis Publishers,Inc.,1992, p.18.
3. *Analytical Chemistry of PCBs*, Erickson, Michell D., Lewis Publishers,Inc.,1992, p.2.
4. Alder, Alfredo C., Häggblom, Max M., Oppenheimer, Stephanie R., Young, L.Y., Environ. *Sci. Technol.*, Vol. 27, No. 3, 1993, pp. 530-538.
5. *Analytical Chemistry of PCBs*, Erickson, Michell D., Lewis Publishers,Inc.,1992, p.37.

PRODUCTS

XTI®-5 30m, 0.32mm ID, 0.25µm cat .# 12224, each

Aroclor® Standards

1000µg/ml each in 1ml hexane	Individual	Individual w/data pack	5-pk.	5-pk. w/data pack	10pk. w/data pack
Aroclor® 1221	32007	32007-500	32007-510	32007-520	32107
Aroclor® 1232	32008	32008-500	32008-510	32008-520	32108
Aroclor® 1242	32009	32009-500	32009-510	32009-520	32109
Aroclor® 1248	32010	32010-500	32010-510	32010-520	32110
Aroclor® 1254	32011	32011-500	32011-510	32011-520	32111
Aroclor® 1016/1260	32039	32039-500	32039-510	32039-520	32139

Hints for the Capillary Chromatographer

Selecting the Proper Ferrule for Capillary Columns[1]

Proper ferrule selection is critical for capillary column installation. Characteristics such as thermal stability, ruggedness, and compressibility are determined by the different materials used to make ferrules. It is important to choose the right ferrule type and size to ensure proper column installation. The wrong ferrule type could cause damage to sensitive detectors such as ECDs, ELCDs, and MSDs. The wrong ferrule size or type can cause system leaks that result in decreased sensitivity and deterioration.

Ferrule Materials

Since metal ferrules would damage fused silica tubing, softer materials are used for capillary column ferrules. The two most common materials for capillary column ferrules are graphite and Vespel®. These materials can also be combined to form hybrid ferrules with the benefits of each material. Other ferrule materials, such as Teflon® and silicone, are commonly used with packed columns, but because of their limited thermal stability they are not typically used with capillary columns. Table I lists the maximum operating temperatures and the characteristics of common capillary ferrule materials.

Table I - Common Characteristics of Capillary Ferrules

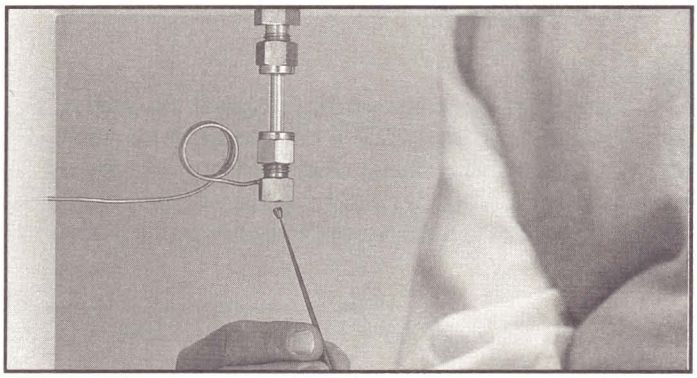
Material	*Max Temp.*	*Characteristics*
Graphite	450°C	Soft, easily conforms to all column sizes. Excellent for high temperature applications. Can flake or deposit particles in inlet & detector fittings. Easily deforms, resulting in limited reusability. Not recommended for vacuum interfaces.
Vespel®/Graphite	400°C	Hard, must be sized to exact column OD. Contracts when cooled causing leakage if not retightened after several thermal cycles. Excellent reusability.

Properties of Graphite Ferrules

Many chromatographers prefer graphite ferrules because they are soft and easily conform to any fitting dimension. Most graphite ferrules are made by tightly winding graphite ribbon around a pin and compressing it into a mold. The graphite ribbon increases ferrule pliability and allows it to deform easily. Increased pliability makes it possible to seal a 0.4mm OD (0.25mm ID) fused silica column with a 0.8mm ID ferrule. In addition, the ferrule can accomodate larger columns if the graphite bore is cored out. These features allow chromatographers to always have the right size ferrule on hand.

Graphite ferrules should be tightened using minimal force. Usually ¼-turn past finger-tight is sufficient to form a leak-tight seal. If a graphite ferrule is over-tightened, it will extrude out of the bottom of the nut, deform into the fitting cavity, and create ferrule fragments. These particles can be driven further into the inlet or make-up gas fitting, causing adsorption or peak tailing when a column is reinstalled. Graphite ferrules can also flake or abrade and emit particles that can clog small orifices. Because graphite is porous, graphite ferrules leak under vacuum. Therefore, graphite ferrules are not recommended for detectors operated under vacuum, such as MSDs.

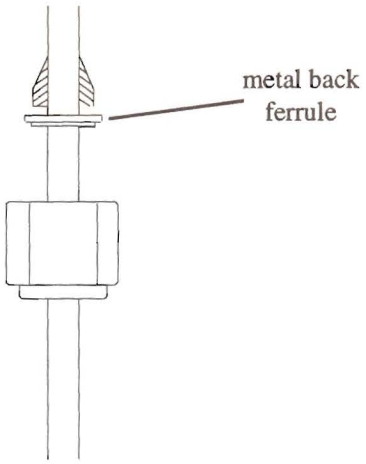
Graphite ferrules must be carefully removed, otherwise fragments and flakes remaining in the fitting can contaminate the GC system. Ferrules are easily dislodged by inserting a tapered needle file into the bore and moving it side-to-side. If the graphite ferrule does not come out in one piece, the inlet or detector fitting should be completely disassembled to ensure that no ferrule fragments remain.



Needle files easily remove graphite ferrules from injector and detector fittings or nuts. Gently insert the file into the ferrule bore and move it from side-to-side to dislodge the ferrule.

The life of a graphite ferrule is limited because they compress so easily. Some chromatographers obtain new life from a crushed ferrule by installing a reversed Swagelok®-type back ferrule between the fitting and the ferrule (Figure 1). The back ferrule raises the graphite ferrule higher in the fitting, allowing it to seal again.

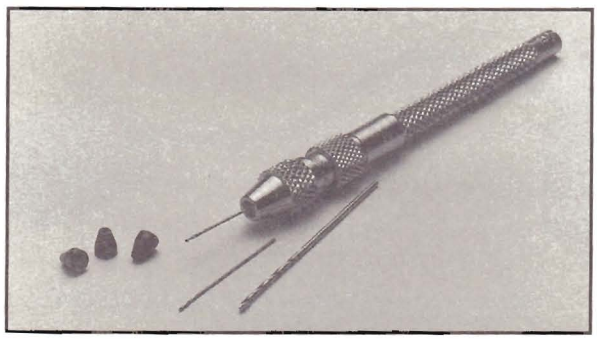
Figure 1 - Give a used graphite ferrule new life by installing a reversed metal back ferrule in the fitting.



Properties of Vespel® or Vespel®/graphite

Both 100% Vespel® and Vespel®/graphite ferrules are available. Vespel®-type ferrules are often preferred because they do not flake, deposit particles, or fall apart in a fitting. Most chromatographers choose the Vespel®/graphite ferrule combination. These ferrules are made by compressing a graphite/polyimide powder under high pressure in a heated mold. They retain their shape and can easily be removed intact. Vespel®/graphite has a higher thermal stability than Vespel® (400°C vs. 350°C) and the graphite impregnation makes the ferrule feel softer and seal with less torque. Vespel®/graphite ferrules are currently available in combinations ranging from 85% Vespel®/15% graphite to 60% Vespel®/40% graphite. The 60/40 Vespel®/graphite combinations are preferred by most chromatographers because they seal with the least amount of torque.

Unlike graphite, the inside diameter of Vespel®-type ferrules must be very close to the column OD in order to seal properly. If the ID of a Vespel®-type ferrule is too large for the column OD, it will not compress properly and allow a leak. Usually, the ferrule forms an oval shape, gripping the tubing but not sealing at the ends of the oval. If the ID of a Vespel®-type ferrule is too small to fit over the column, the bore must be enlarged with a small drill.



If the Vespel®/graphite ferrule's ID is too small to fit over the column, a pin vise drill can be used to enlarge the bore.

Vespel®/graphite ferrules will deform to the exact fitting dimension when heated. Usually this deformation process causes the ferrule to become loose and leak during the cool down cycle of a GC oven. Therefore, they must be subsequently retightened after several thermal cycles or carrier gas leakage will occur. No additional shrinkage or loosening occurs once the ferrule has conformed to the internal dimensions of the fitting cavity.

Vespel® ferrules can be removed from a fitting using a tapered needle file in the same manner as a graphite ferrule. Vespel® ferrules sometimes stick to the fitting and column after they have been in use for a prolonged period. Stuck ferrules can be removed by tapping the fitting with a solid object such as a wrench and gently pulling outward on the column. This problem is greatly minimized by using Vespel®/graphite combination ferrules.

What are common ferrule sizes?

Most column connections in the GC inlet and detector are made using 1/16" Swagelok®-type fittings. The ID or opening of the ferrule depends on the outside diameter of the column. Table II lists common fused silica capillary column IDs, ODs, and recommended ferrule sizes.

Table II - Common Ferrule Sizes for Fused Silica Capillary Columns

Column ID	*Column OD*	*Ferrule Opening*
0.18 to 0.25mm	0.35 to 0.40mm	0.4mm
0.32mm	0.45 to 0.48mm	0.5mm
0.53mm	0.69 to 0.72mm	0.8mm

The choice of ferrule material is often personal preference. If you are installing a capillary column for the first time, we suggest using a graphite ferrule. Graphite easily forms a leak-tight seal and conforms to any column OD. If you frequently install new columns, Vespel®/graphite is recommended to eliminate particle evolution and minimize maintenance downtime. However, when connecting columns to MSDs or Mass Spectrometer transfer lines, Vespel®/graphite is the only ferrule you should use to ensure a leak-free seal under vacuum. We recommend trying both ferrule types to choose a ferrule that best fits your needs. ■

[1]This article is a reprint from the *The Restek Advantage*, Vol. 4 No. 2, March 1993, p. 12-13.

Peak Performers

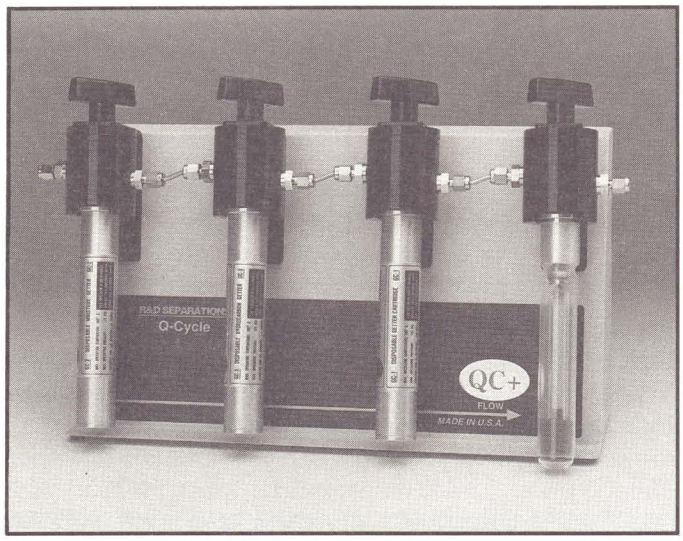
Simplify Gas Purification with the QC+ Purifier Panel

- Replace GC gas purifiers without flow interruption.
- Self aligning, trouble-free installation.
- Three and four panels units available.
- Impurities reduced to low part per billion levels.

GC carrier, make-up, and fuel gases require purification in order to provide stable baselines and longer column life. Traditional gas purifiers are installed at the gas source or in-line near the instrument, but require instrument downtime to be replaced. The new QC+ Purifier panels can be bench or wall mounted and allow a complete set of gas purifiers to be quickly changed without significant interruption in gas flow, eliminating instrument downtime.

Each QC+ filter mounting head is equipped with an internal valve that provides two flow paths. In the up position, it provides a closed gas path through the mounting head. As the valve is moved to the down position, it punctures the seal on the cartridge top and shifts the gas flow through the purifier cartridge. QC+ cartridges are self-aligning for trouble-free installation and are quickly replaced via a simple knurled retaining nut. As many as four cartridges can be replaced in less than two minutes without contamination of your GC gas system.

Restek offers two QC+ panels. The QC+ 3 cartridge panel features moisture, oxygen and indicating oxygen cartridges. The QC+ 4 cartridge panel is the ultimate in GC purification. It features the moisture, hydrocarbon, oxygen and indicating oxygen cartridges. The indicating oxygen cartridge can be regenerated to further increase the value of these units. Both panels will reduce gas impurities to low part per billion levels, regardless of source gas condition.



Product Listing

QC+ 3 head purifier panel
(with Moisture, O_2, & Indicating O_2 traps):
cat.# 21676

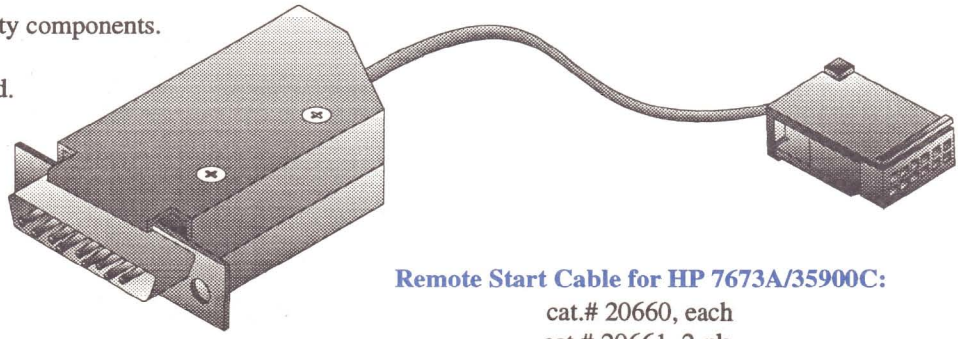
QC+ 4 head purifier panel
(with Hydrocarbon, Moisture, O_2, & Indicating O_2 traps):
cat.# 21677

QC+ Replacement Cartridges
Oxygen Cartridge: cat.# 21678
Moisture Cartridge: cat.# 21679
Hydrocarbon Cartridge: cat.# 21680
Indicating O_2 Cartridge: cat.# 21681

Remote Start Cable for HP 7673A/35900C

- Manufactured with only the highest quality components.
- Tested for 100% signal integrity.
- Instructions and wiring diagrams included.
- Priced less than original HP equipment.
- Instructions and wiring diagrams included.

Connect an HP 5890 GC to an HP 35900C Interface or an HP 7673 Autosampler to enable remote starts. Similar to HP part # 35900-60700.



Remote Start Cable for HP 7673A/35900C:
cat.# 20660, each
cat.# 20661, 2-pk.

MXT® Capillary Columns

The unbreakable alternative for process control and portable GCs

- Stationary phases available for most applications.
- High degree of inertness.
- Priced equivalent to fused silica.
- Extreme flexibility eliminates spontaneous breakage.
- Standard coil diameter approximately 5" OD, smaller custom coil sizes available.

What are MXT® columns?

MXT® columns are made by depositing a uniform, micron layer of flexible fused silica on the inner surface of thin walled stainless steel tubing. The surface is then deactivated by the same process used to treat our fused silica capillary columns.

MXT® columns are easy to use!

Available in both 0.28 and 0.53mm IDs (other IDs available upon request), MXT® columns can be installed in most instruments without modification. MXT columns are easy to cut using a small file or ceramic scoring wafer.

If the idea of an unbreakable capillary column sounds like the solution to your analytical problems, try an MXT® column. They are backed by Restek's 100% satisfaction guarantee. For a complete list of MXT® products, call your local distributor for a free copy of our *MXT® Columns & Accessories* catalog.

			0.28mm ID columns			0.53mm ID columns			
MXT®-1 Applications: Sim Dist, waxes, fuel oils, pharmaceutical, solvents	df (µm)	max. temp.	15-meter cat.#	30-meter cat.#	60-meter cat.#	15-meter cat.#	30-meter cat.#	60-meter cat.#	
	0.25	360°C	70121	70124	70127	70122	70125	70128	
	0.50	330°C	70136	70139	70142	70137	70140	70143	
	1.00	325°C	70151	70154	70157	70152	70155	70158	
	1.50	320°C	-------	-------	-------	70167	70170	70173	
	3.00	300°C	70181	70184	70187	70182	70185	70188	
	5.00	275°C	-------	-------	-------	70177	70179	70183	
	7.00	250°C	-------	-------	-------	70191	70192	70193	
MXT®-5 Applications: semi-volatiles, pesticides, PCBs, environmental samples, essential oils	df (µm)	max. temp.	15-meter cat.#	30-meter cat.#	60-meter cat.#	15-meter cat.#	30-meter cat.#	60-meter cat.#	
	0.25	360°C	70221	70224	70227	70222	70225	70228	
	0.50	330°C	70236	70239	70242	70237	70240	70243	
	1.0	325°C	70251	70254	70257	70252	70255	70258	
	1.5	320°C	-------	-------	-------	70267	70270	70273	
	3.0	300°C	70281	70284	70287	70282	70285	70288	
	5.0	275°C	-------	-------	-------	70277	70279	70283	
MXT®-Wax Applications: FAMEs, flavors, solvents, BTEX, essential oils, EPA Method 603	df (µm)	max. temp.	15-meter cat.#	30-meter cat.#	60-meter cat.#	15-meter cat.#	30-meter cat.#	60-meter cat.#	
	0.25	250°C	70621	70624	70627	70622	70625	70628	
	0.50	240°C	70636	70639	70642	70637	70640	70643	
	1.0	230°C	70651	70654	70657	70652	70655	70658	
	1.5	220°C	-------	-------	-------	70666	70669	70672	
	2.0	230°C	-------	-------	-------	70667	70670	-------	
MXT®-502.2 Applications: volatile organics	df (µm)	max. temp.	30-meter cat.#	60-meter cat.#	105-meter cat.#	30-meter cat.#	60-meter cat.#	105-meter cat.#	
	1.6	270°C	70919	70920	70921	-------	-------	-------	
	3.0	270°C	-------	-------	-------	70908	70909	70910	
MXT®-624 Applications: EPA Methods 502.2, 524, 624, 8240, and 8260	df (µm)	max. temp.	30-meter cat.#	60-meter cat.#	105-meter cat.#	30-meter cat.#	60-meter cat.#	75-meter cat.#	105-meter cat.#
	3.0	240°C	-------	-------	-------	70971	70973	70974	70975
Sim Dist MXT®-1	6m, 0.53mm ID, 0.15µm, cat.# 70101				cat.# 70101's are tested at 400°C, but may be run at higher temperatures and at a lower sensitivity with additional conditioning.				
Sim Dist MXT®-2887	10m, 0.53mm ID, 2.65µm, cat.# 70199				cat.# 70199's maximum temperature is 360°C.				

Note: All maximum operating temperatures are for 30m columns. Maximum temperatures may be slightly lower for longer lengths.

CHROMalytic TECHnology >>>

" some" **promos / Products / Offers** in the **ADVNews**
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