Improved Phases for the GC Analysis of Organophosphorus Pesticides

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Abstract

Organophosphorus pesticide (OPP) analysis is common in agricultural analytical laboratories. With the decline of organochlorine pesticide use, OPPs have become increasingly popular as the active ingredients in insecticides. In addition, they are used in termite treatments, lawn and garden sprays, indoor insect sprays and baits, and in pet flea collars and sprays. Testing of agricultural products is required because of potential health risks associated with these compounds.

The gas chromatographic (GC) analysis of OPPs is demanding because some of the compounds are light and temperature sensitive. In addition, specialty detectors (e.g. the nitrogen phosphorus detector or flame photometric detector) and dual-column analyses are required in OPP methods in order to obtain low-level detection and identification. This presentation will discuss a primary and confirmational column set developed for the analysis of OPPs. The phases were developed using a proprietary phase modeling software program, which aids in the optimization of the phase chemistry, film thickness, and column dimensions.

Developing an Organophosphorus Column

In developing a GC column for organophosphorus pesticide analysis, the column should be capable of separating USEPA 8141 compounds, serve as a companion column to columns such as the Rtx-OPPesticides, and have a total run time of 20 minutes or less. In addition, it should be compatible with GC/MS analysis (i.e. low bleed, and able to chromatographically resolve compounds with similar spectra).

Traditional column selection has involved asking a "senior" person in the laboratory, consulting a column manufacturer's applications section, matching the polarity of the column to the compounds of interest or trying to force the separation onto an existing phase. Conversely, modern column selection involves columns and stationary phases that are designed around applications. Specific phases and columns can be developed for a target separation. This approach requires an understanding of and the ability to model analyte-phase interactions.

Stationary Phase Optimization

Stationary phase optimization can be achieved through window diagramming or by computer simulation of the retention time, R_t, and peak width at half height, $W_{1/2}$. Simulation programs such as ezGC have been applied in the development of phases such as the Rtx®-CLPesticides and Rtx-CLPesticides2.

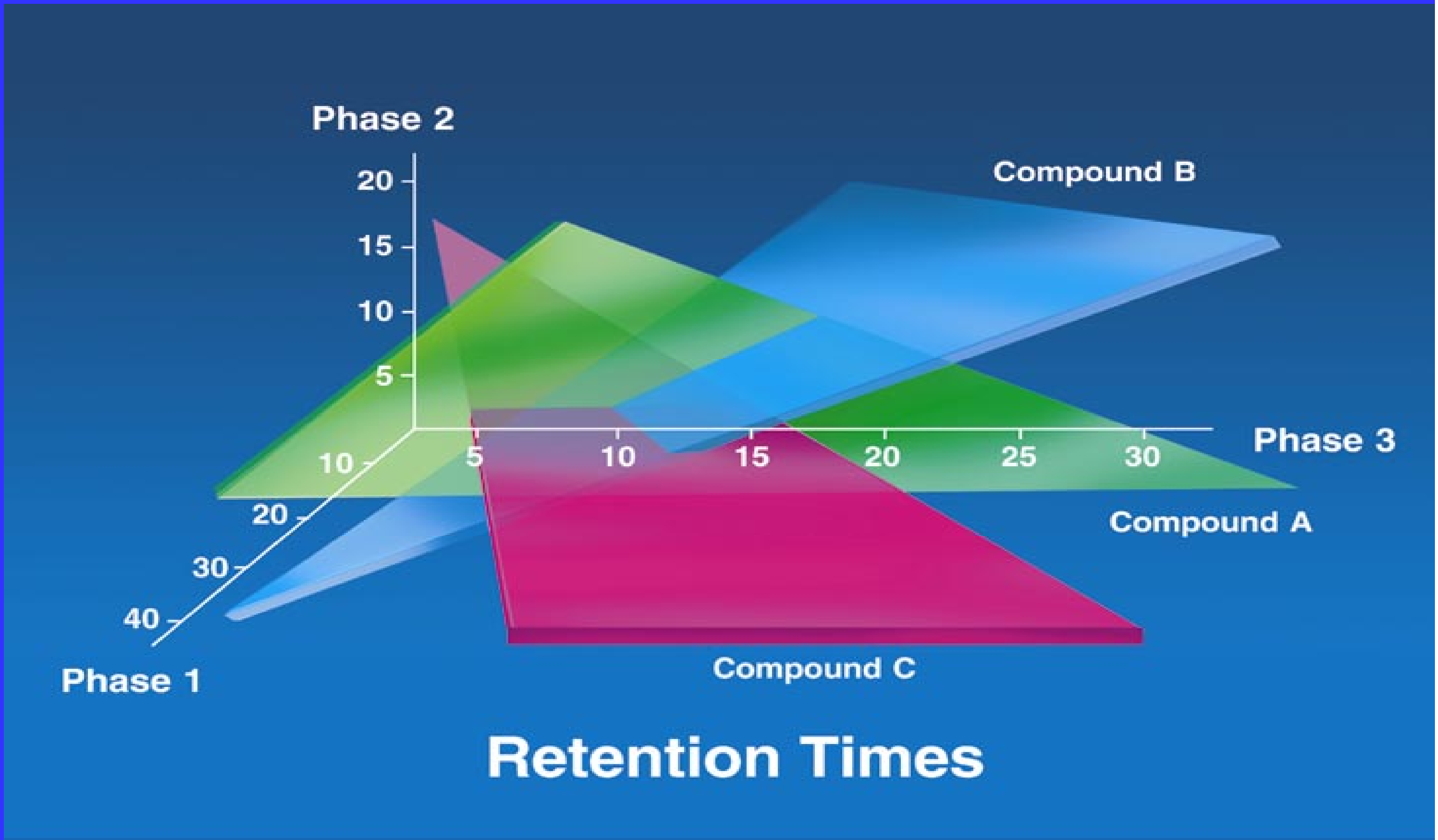
The next step in polymer development involves the computer prediction of optimized stationary phase composition and column dimensions. This approach uses computer prediction of solute/stationary phase interactions to aid in the design of new polymers. Columns such as the Rtx-TNT, Rtx-TNT2, Rtx-VMS, Rtx-VGC, Rtx-5SilMS, Rtx-VRX, and Rtx-OPPesticides2 were developed using this approach.

Development of the Rtx-OPPesticides2 Column

In the development of the Rtx-OPPesticides2 column, data was first acquired for target compounds under two temperature programs. Columns with functionalities that displayed selectivity for these compounds were used. Computer Assisted Stationary Phase Design (CASPD) was used to calculate ΔH and ΔS for each compound. By working in Retention Index, the phase and dimensions could be optimized for the target separation. The stationary phase can then be synthesized and the column coated at the appropriate phase thickness.

Below is a simple 3-D model depicting a polymer composition for 3 compounds. This is a simplified model describing the 53 compounds which were modeled for the Rtx-OPPesticides2 column.

3-Space Selectivity Model for 3 Compounds



CASPD Modeling Results

The modeling software output includes not only the predicted phase composition but also predicted retention times and run conditions. The following table shows the comparison of the modeled results to the experimental results for the column made from the predicted polymer.

As indicated, the compounds eluted within 0.2 minutes of the CASPD prediction. The accuracy of the software benefits both R&D and Innovations chemists involved in new product development. Instead of making various polymers with different percentages of functional groups, computer modeling can be performed to generate polymer formulations that result in optimal resolution in short analytical run times.

Comparison of Predicted vs Actual Retention Times of OP Pesticides

Target Compound	Predicted Rt	Actual Rt	Difference (min)
dichlorvos	4.08	4.05	-0.03
HMPA	4.70	4.70	0.00
mevinphos	6.43	6.34	-0.09
trichlorfon	6.44	6.43	-0.01
TEPP	8.20	8.40	0.20
demeton-o	8.46	8.52	0.06
thionazin	8.58	8.52	-0.06
TBP	8.60	8.52	-0.08
ethoprop	8.84	8.74	-0.10
naled	9.34	9.32	-0.02
sulfotepp	9.42	9.56	0.14
phorate	9.53	9.56	0.03
dicrotophos	9.61	9.59	-0.02
monocrotophos	9.70	9.62	-0.08
demeton-s	9.80	9.62	-0.18
terbufos	10.44	10.32	-0.12
dimethoate	10.67	10.62	-0.05
dioxathion	10.78	10.77	-0.01
fonophos	10.91	10.79	-0.11
diazinon	10.93	10.90	-0.04
disulfoton	11.13	11.09	-0.03
phosph isomer	11.19	11.16	-0.04
dichlorofenthion	11.38	11.37	-0.01
chlorpyrifos methyl	11.94	12.03	0.09
phosphamidon	12.14	12.03	-0.11

Analysis OP Pesticides by FPD

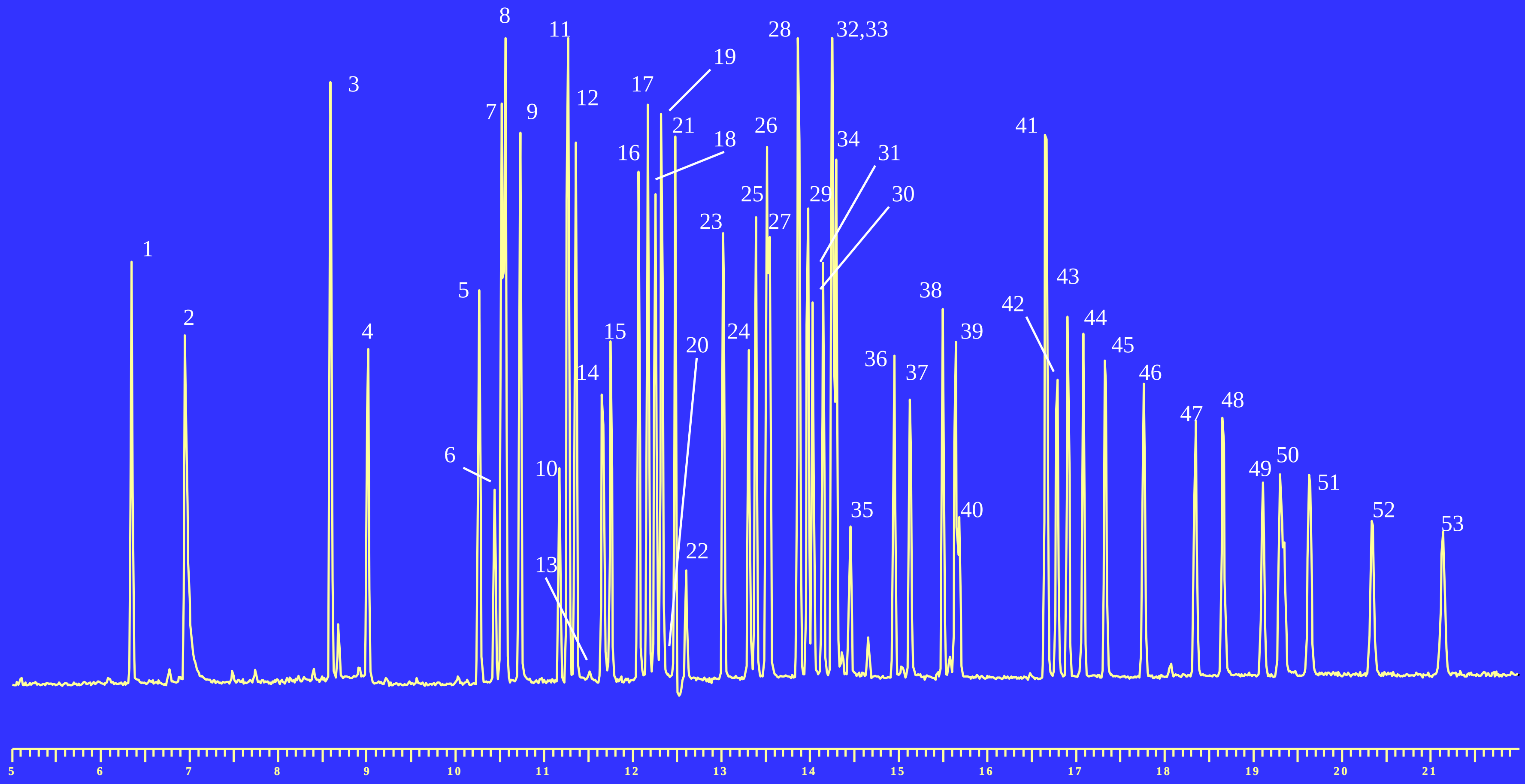
The analysis of organophosphorus pesticides requires a primary and secondary column for positively identified compounds using selective detectors.

The Rtx-OPPesticides2 column is an excellent choice for the primary column, resolving all 53 compounds except for one pair within 22 minutes.

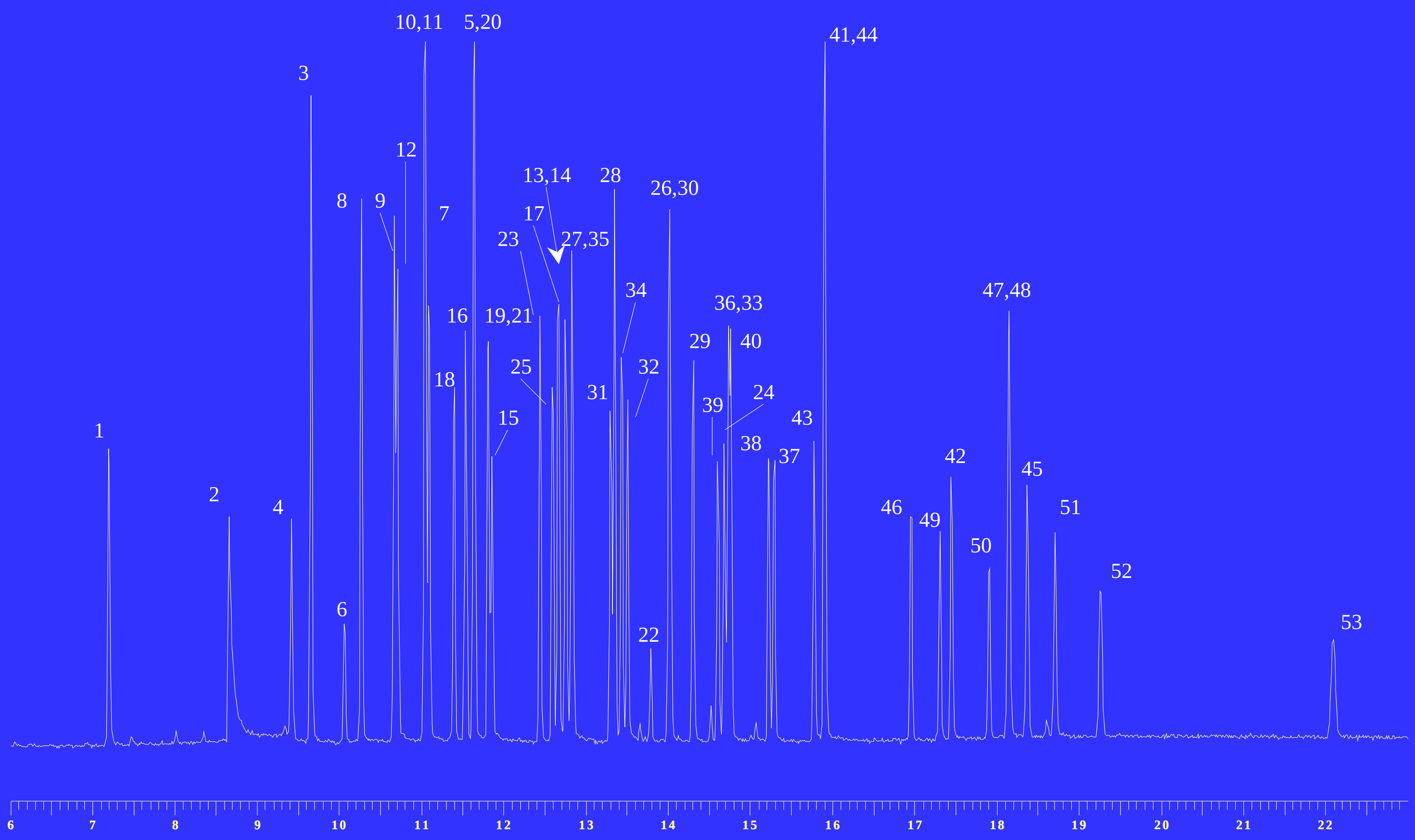
The Rtx-OPPesticides column is a good choice for a second column offering the fewest number of coelutions in the short analysis time of 22 minutes, under the same run conditions as the Rtx-OPPesticides2 column.

Following are the chromatographic results of 53 organophosphorus compounds run under optimized conditions for the Rtx-OPPesticides2 column.

Rtx-OPPesticides2 column
(30m x 0.32mm ID, 0.5um film)



Rtx-OPPesticides column
(30m x 0.32mm ID, 0.5um film)



Organophosphorus Pesticides by FPD Peak IDs

PK#	Compound	PK#	Compound	PK#	Compound
1	dichlorvos	20	fonophos	38	stirofos
2	hexamethylphosphoramide	21	disulfoton	39	tokuthion
3	mevinphos	22	phosphamidon isomer (breakdown product)	40	merphos oxon (breakdown product)
4	trichlorfon				
5	TEPP	23	dichlorofenthion	41	ethion
6	demeton-o	24	phosphamidon	42	fensulfothion
7	tributyl phosphate (SS)	25	chlorpyrifos methyl	43	bolstar
8	thionazin	26	parathion-methyl	44	carbophenothion
9	ethoprop	27	ronnel	45	famphur
10	naled	28	aspon	46	triphenyl phosphate (SS)
11	sulfotepp	29	fenitrothion	47	EPN
12	phorate	30	malathion	48	phosmet
13	dicrotophos	31	chlorpyrifos	49	leptophos
14	monocrotophos	32	trichloronate	50	tri-o-cresyl phosphate
15	demeton-s	33	parathion-ethyl	51	azinphos-methyl
16	terbufos	34	fenthion	52	azinphos-ethyl
17	dimethoate	35	merphos	53	coumaphos
18	diazinon	36	chlorfenvinphos		
19	dioxathion	37	crotoxyphos		

Organophosphorus Pesticides by FPD Run Conditions

GC oven: 80°C(0.5min)@12°C/min to 280°C(10min)

Injector: 200°C splitless, purge off time 1min,

4mm single gooseneck Siltek sleeve

Detector: Agilent FPD 250°C

DT @80C = 0.98min for the Rtx-OPPesticides

= 1.03min for the Rtx-OPPesticides2

Carrier gas: helium

Columns:
RTX-OPPesticides, cat# 11239 30m x 0.32mm ID, 0.5um film

RTX-OPPesticides2, cat# 11241 30m x 0.32mm ID, 0.32um film

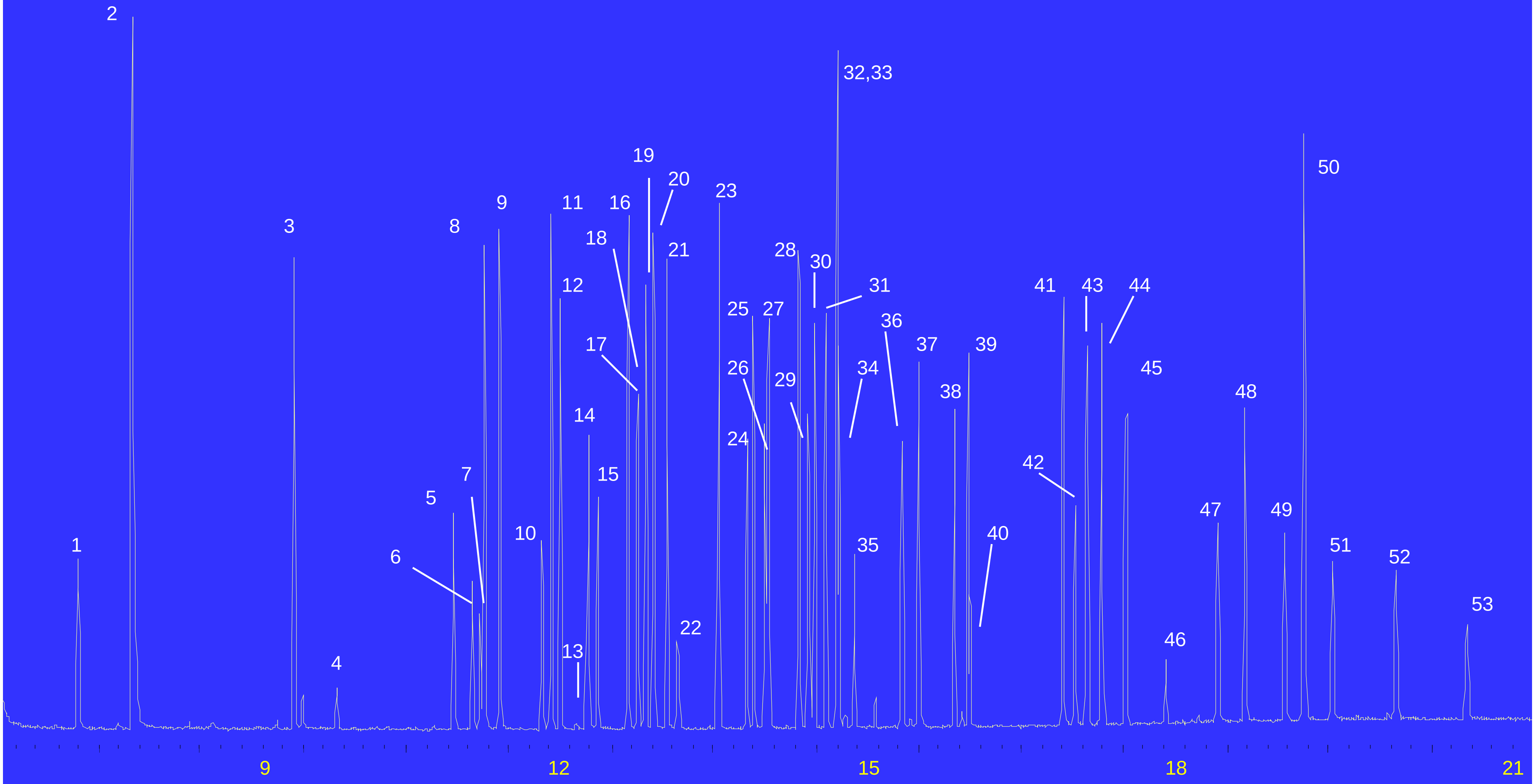
Standard Conc.: 100 ng/ml

Analysis OP Pesticides by GC/MS

Polymers designed for selective detectors, like the NPD or FPD, must maximize resolution between analytes for identification and quantitation. Due to the high selectivity for the modeled compounds, these columns are also a good choice for GC/MS analysis. Although MS detectors can be used for resolving compounds that chromatographically coelute, it is best to minimize coelutions.

The Rtx-OPPesticides2 column is optimized for resolution of all 53 OP Pesticides and is made with a low bleed polymer. This makes the column an excellent choice for GC/MS analysis of organophosphorus pesticides. The following chromatogram shows an analysis time of <21 minutes and only 2 pairs of coeluting compounds. These coeluting compounds have different mass spectra, allowing them to be identified and quantitated.

GC/MS analysis using Rtx-OPPesticides2 column 30m x 0.25mmID, 0.25um film



Compound List for Rtx-OPPesticides2 GC/MS

GC oven: 80°C (0.5min) @ 12°C/min to 280°C (15min)

Injector: 225°C splitless, hold time 1min, 4mm single gooseneck Siltek sleeve

Detector: Agilent 5971A MSD
full scan 50-550AMU

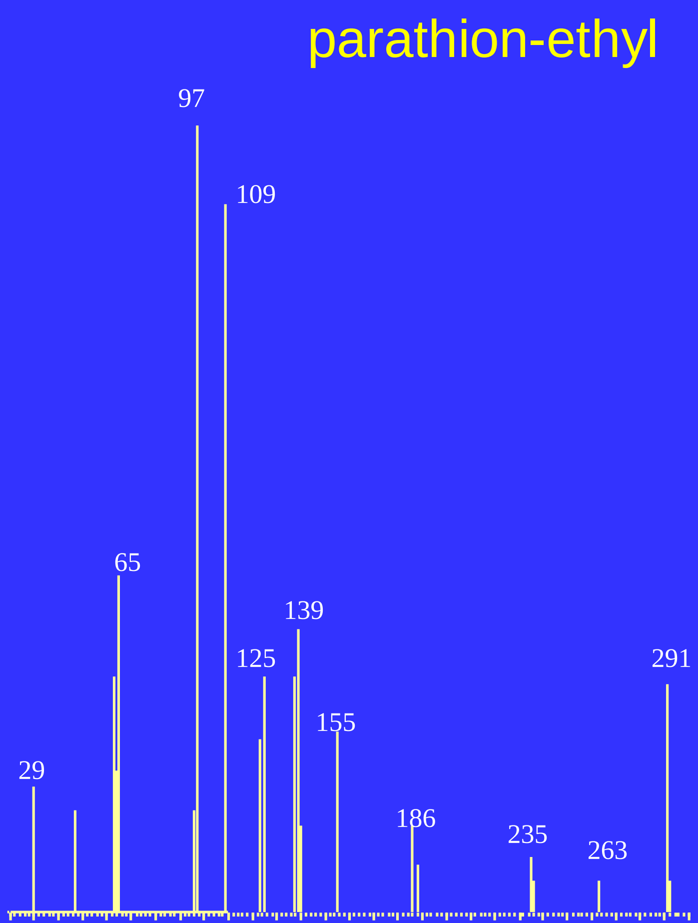
DT @80°C=1.44min, helium carrier

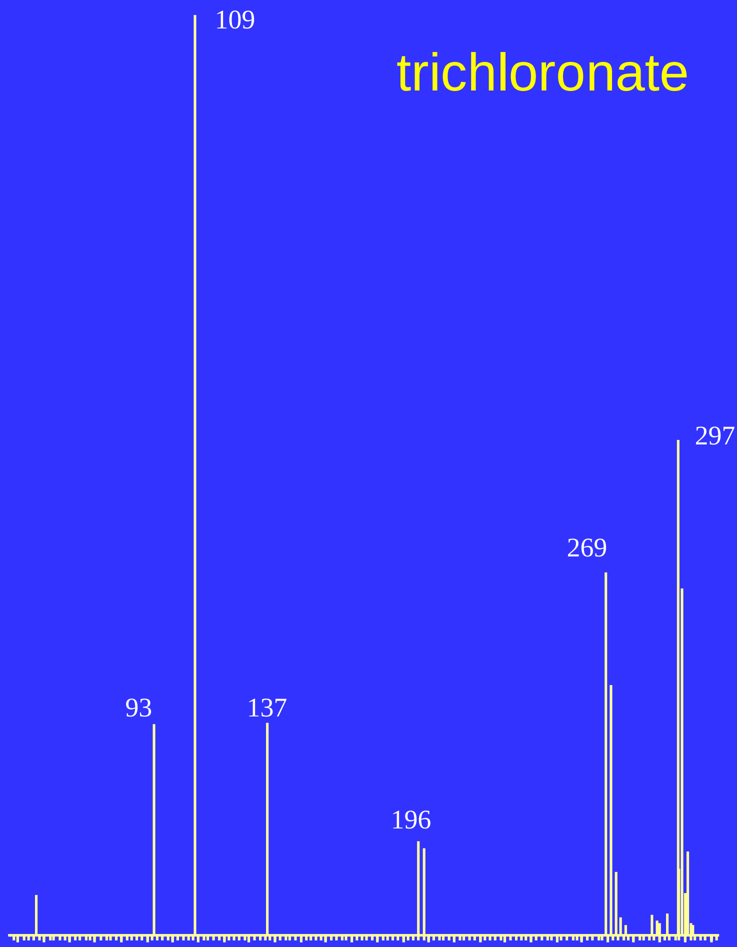
Column: Rtx-OPPesticides2, cat# 11243
30m x 0.25mmID, 0.25µm

cpd #	compound	RT		cpd#	compound	RT
1	dichlorvos	6.81		28	aspon	13.85
2	HMPA	7.33		29	fenitrothion	13.94
3	mevinphos	8.92		30	malathion	13.99
4	trichlorfon	9.33		31	chlorpyrifos	14.10
5	TEPP	10.48		32	parathion-ethyl	14.20
6	demeton-o	10.66		33	trichloronate	14.20
7	thionazin	10.77		34	fenthion	14.24
8	TBP	10.73		35	merphos	14.39
9	ethoprop	10.93		36	chlorfenvinphos	14.85
10	naled	11.34		37	crotoxyphos	15.00
11	sulfotepp	11.43		38	stirofos	15.35
12	phorate	11.51		39	tokuthion	15.48
13	dicrotophos			40	merphos oxid prod	15.52
14	monocrotophos	11.79		41	ethion	16.42
15	demeton-s	11.88		42	fensulfothion	16.53
16	terbufos	12.18		43	bolstar	16.65
17	dimethoate	12.27		44	carbophenothion	16.80
18	diazinon	12.34		45	famphur	17.03
19	dioxathion			46	TPP	17.41
20	fonophos	12.42		47	EPN	17.92
21	disulfoton	12.57		48	phosmet	18.19
22	phosph isomer	12.67		49	leptophos	18.58
23	dichlorofenthion	13.06		50	TOCP	18.76
24	phosphamidon	13.33		51	azinphos-methyl	19.04
25	chlorpyrifos methyl	13.40		52	azinphos-ethyl	19.66
26	parathion-methyl	13.52		53	coumaphos	20.35
27	ronnel	13.55				

Organophosphorus Pesticides MS Spectra

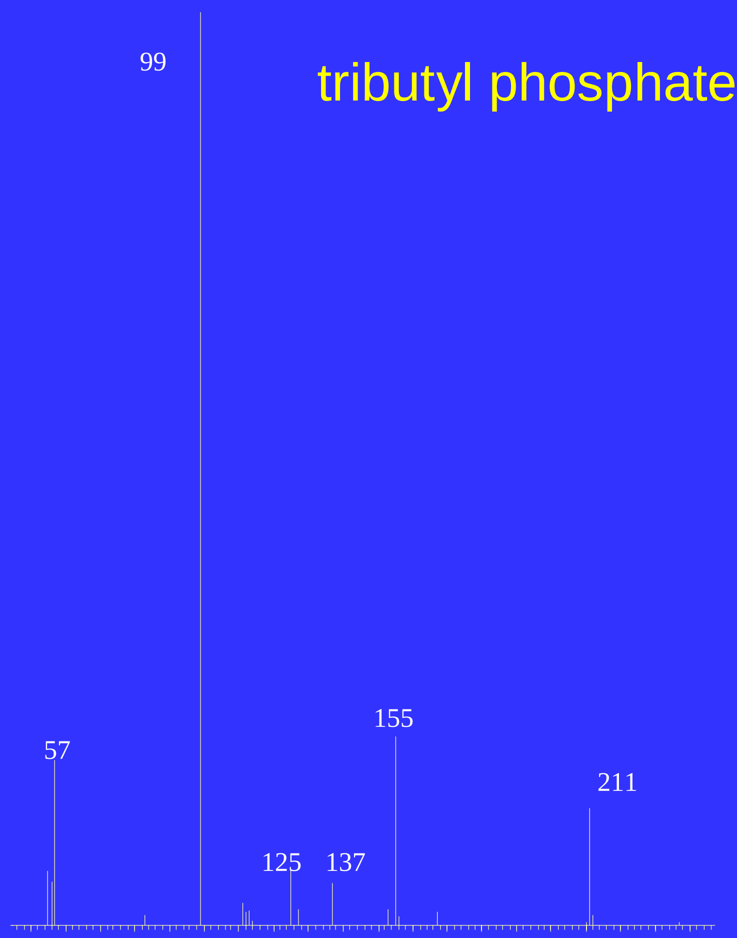
Rtx-OPPesticides2
(30m x 0.25mm, 0.25µm)

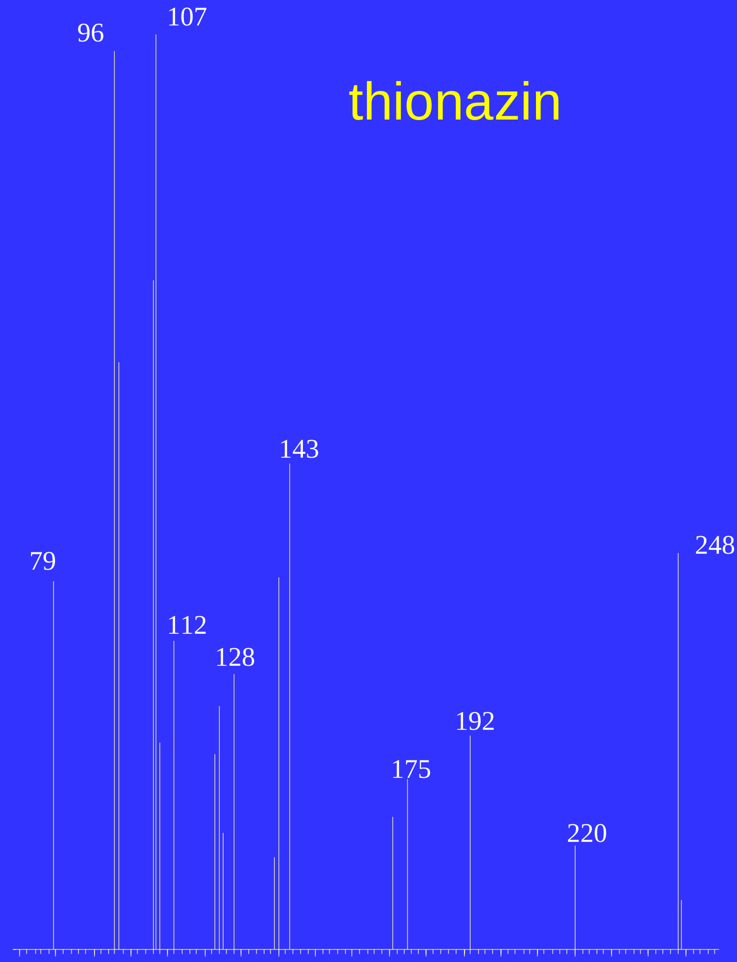




Organophosphorus Pesticides MS Spectra

Rtx-OPPesticides2
(30m x 0.25mm, 0.25µm)





Summary

- The Rtx-OPPesticides and Rtx-OPPesticides2 columns are the optimal dual column pair for USEPA 8141A.

- The Rtx-OPPesticides2 column provides the best separation of organophosphorus pesticides by GC/MS due to the low bleed of this phase and separation of compounds with similar spectra.

- Using computer assisted stationary phase development, the phases have been optimized for these separations, resulting in faster analysis times.