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Using Dual Gate Subtraction to Enhance the Selectivity of a Pulsed Flame Photometric Detector (PFPD)

Application Note 11271299

Keywords
Dual Gates
Pesticides
PFPD
PFPDView
Phosphorus
Sulfur
WinPulse

Introduction

Pulsed flame photometric detector (PFPD) operation is based on a propagating flame that terminates within a glass combustor. The gas phase reactions produced by the propagating flame result in light emissions with specific luminescence wavelengths and lifetimes. The differences in specific emission lifetimes combined with the kinetics of the propagating flame allow both time and wavelength information to be used to improve the selectivity of the PFPD. Emission signals are dependent on the flame composition within the PFPD's combustor (which is determined by the relative flow rates of hydrogen, air, carrier gas, and analytes) and the spectral sensitivity of the optical filter and photomultiplier tube (PMT).

OI Analytical's Model 5380 PFPD provides two independent channels of digitized output data. The integration gate for each data channel is set by using the WinPulse software to specify the start and stop times of each gate within the detector's 25-msec time domain.

The emission times of specific heteroatoms often overlap (e.g., the tail end of the phosphorus (P) emission overlaps with the initial phase of the delayed sulfur (S) emission). The selectivity for the delayed, extended S emission relative to the earlier P emission can be enhanced by simply moving the start S gate to a point beyond the interfering P emission. This improvement in the selectivity between phosphorus and sulfur will result in a small decrease in the sulfur sensitivity. Eliminating the S emission from the P emission with simple adjustment of gate settings is less easy because the S emission overlaps with a significant portion of the phosphorus emission; however, the Model 5380 PFPD and WinPulse software allow the use of the powerful "dual gate" enhanced mode of operation to significantly increase phosphorus selectivity relative to sulfur (and visa versa).

This technique can be used to enhance the interheteroatom selectivity among many of the 28 elements that can be analyzed with the PFPD, but it is not necessarily needed for many PFPD applications. If the chromatographic retention times of the sulfur- and phosphorus-containing compounds differ sufficiently to provide selectivity for the compounds of interest, then it is unnecessary to use the dual gate mode except for identification purposes. The greatest value of this technique will occur when the matrix complexity or insufficient sample resolution results in insufficient selectivity between the specific heteroatoms being analyzed. This technique may also be useful when analyzing compounds containing both phosphorus and sulfur (e.g., some organophosphorus pesticides).

Using the Dual Gate Option in WinPulse

The basic premise for using dual gates is that the observed PFPD signal is the sum of the independent emission time profiles of all emitting species (e.g., the S and P emission profiles). Successful application of the dual gate technique requires sufficiently large differences in the emission time domain of each species to allow discrete gates to be specified. Each gate incorporates a segment of the time domain of one emitting species only (see Figure 1). Note that within the phosphorus gate (time domain) the P emission dominates, while the S gate contains mainly S emission. Specifying these gates results in an overlap between the two elements. An additional requirement for the successful application of the dual gate technique is that gate conditions, gas composition, spectral filters, and PMT conditions are held constant.

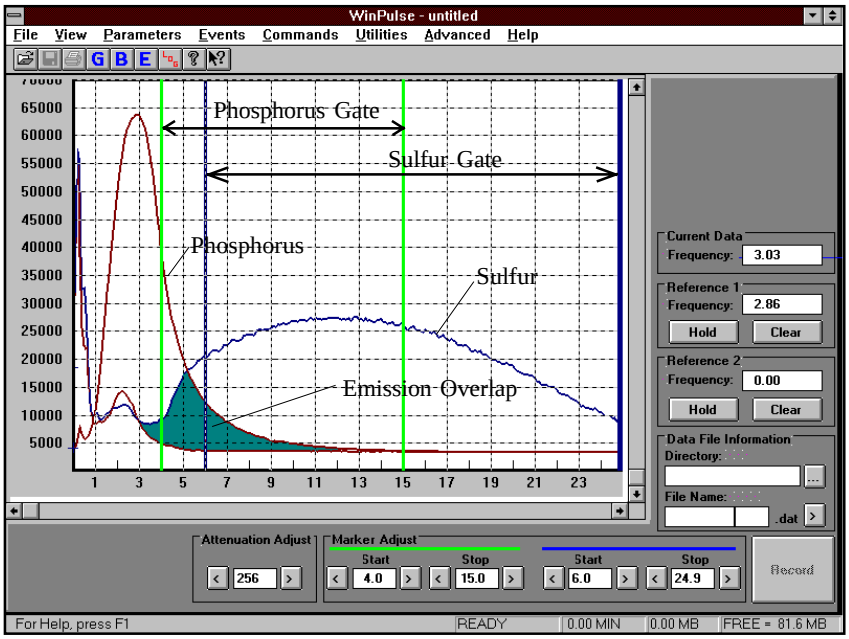

Figure 1. Phosphorus and Sulfur Emissions

As shown in Figure 2, the PFPD operational software WinPulse allows the operator to specify two gates, A and B, for each mode. In order to optimize detectivity, Gate A is set to incorporate as much as possible of the emission time domain of the species of interest (i.e., phosphorus in the P mode) while minimizing the emission time domains of interfering species (e.g., carbon and sulfur in the P mode).

To use the dual gate technique to eliminate interference between two species, reciprocal gate assignments are entered for each of the two modes specified in the Gate Parameters window of the WinPulse software (see Figure 2). To accomplish this, Gate A is assigned to the species of interest, and Gate B is assigned to the interfering species. For example, Gate A start and stop times (4–15 msec) for the P mode are specified to optimize phosphorus response, while Gate A start and stop times (6–24.9 msec) for the S mode are set to optimize the sulfur response. Conversely, Gate B in each mode specifies the optimal gate settings for the interfering species (i.e., sulfur in the P mode and phosphorus in the S mode).

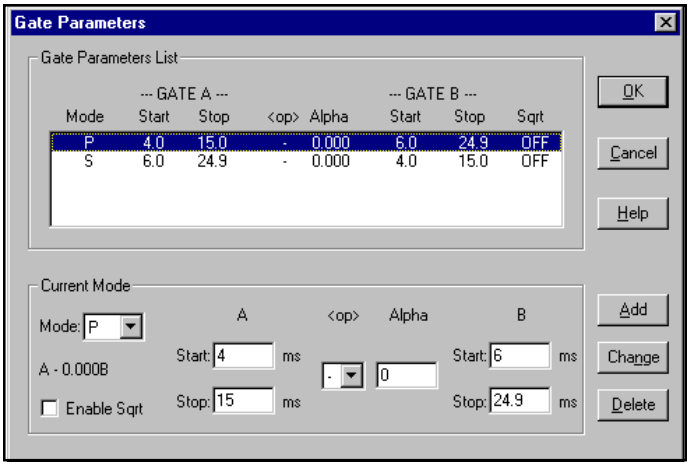

Figure 2. Gate Parameters Window Showing Gate Settings for Phosphorus (P) and Sulfur (S)

Note: Appropriate gate settings will depend on the application. There is substantial overlap between the 4–15 msec phosphorus gate and the 6–24.9 msec sulfur gate settings shown in Figure 2. Such large overlaps may be unnecessary or even undesirable if a substantial segment of one species lies beyond the emission time profile of another species (e.g., the sulfur emission extends well beyond the phosphorus response).

Since the Model 5380 PFPD provides two separate signal output channels, the P mode can be assigned to Channel 1 and the S mode to Channel 2, as shown in Figure 2. If the dual gate mode is not used, the Alpha values for both gates are set to zero. A chromatogram derived using zero Alpha values for a test standard containing tributyl phosphate (TBP) (P only) and tetrahydrothiophene (THT) (S only) is shown in Figure 3. In this example, using Alpha values set at zero did not produce optimal interheteroatom selectivity between phosphorus and sulfur. The secondary peak in each chromatogram is caused by the interfering species (i.e., overlap of the P and S emissions). In this example, the GC column could suffice to provide the required selectivity to identify and quantitate the peaks of interest. However, co-elution of the peaks of interest or the matrix-related interference peaks would benefit from the use of the dual gate mode.

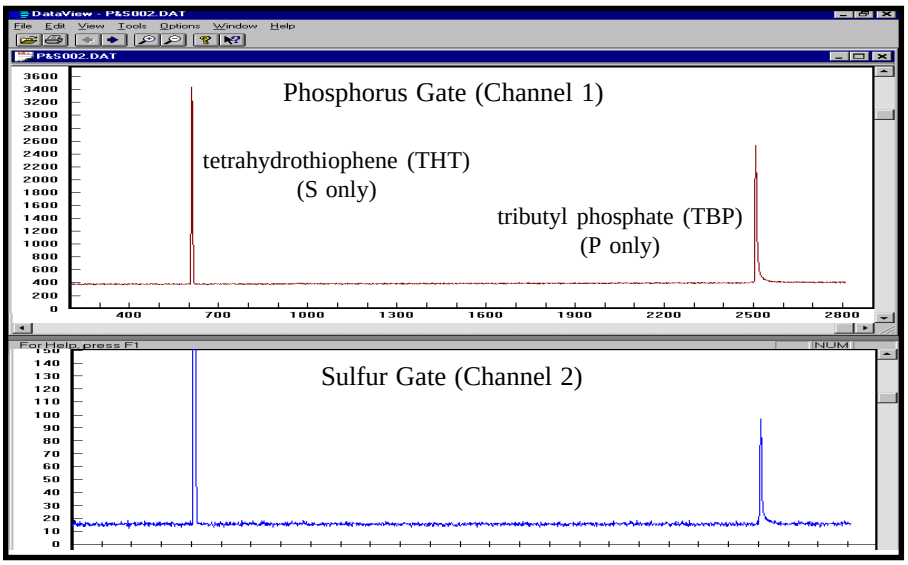

Figure 3. Chromatograms from P and S Gates Using TBP/THT Standard

Empirical Estimation of Gate B Scaling Factors

In order to minimize or eliminate this observed overlap (peak) caused by the interfering species, an optimal Alpha value (Gate B scaling factor) must be specified for each mode (or channel). Optimal Alpha values can be determined in three ways: (1) through GC iteration (see Amirav and Jing, 1995); (2) by empirical calculation (estimating or trial and error); or (3) through post-run data iteration using OI Analytical's PFPDView software program.

Empirical estimation of appropriate Alpha values for detecting both phosphorus and sulfur using the Channel 1 and Channel 2 signal outputs requires the following steps:

1. Go to the Parameters menu and click on Gates/Mode. This accesses the Gate Parameter window. Specify start and stop times for Gate A and Gate B in each mode (see Figure 2 for example of gate settings for P and S modes). Set the operator (<op>) sign to "–" and the Alpha value for each mode to zero.

2. Go to the Parameters menu and click on Board/Channel. This accesses the Board/Channel Parameters window. Assign the P mode to Channel 1 and the S mode to Channel 2. Set appropriate zero offsets for both channels (zero offsets do not need be the same), and set the same attenuation value for each channel.

3. Ensure that a suitable filter for detecting **both** species of interest is installed in the PMT assembly (e.g. WG 345 for phosphorus and sulfur, see OI Analytical Application Note #1166).

4. Configure the GC for a test run to analyze a suitable test standard containing both phosphorus and sulfur. Ensure that each compound incorporated in the test standard contains only one of the heteroatoms of interest. It is important that the heteroatom concentrations in the test standard compounds be similar to the anticipated concentrations of these species in the substances that are to be subsequently analyzed. If this is not the case, estimated Alpha values may have to be adjusted in order to subtract the emission from interfering species.

Note: Alpha values should be set using concentrations similar to the expected values of analyte and interference to avoid positive or negative baseline excursions. The PFPD signal must be within the proper range, avoiding electronic saturation.

5. Conduct a GC run of the test standard and obtain a chromatogram for both the Channel 1 and Channel 2 output. Figure 3 shows the chromatograms obtained from the two output channels using a TBP/THT test standard. In this case, the sulfur-containing compound is the first peak and the phosphorus-containing compound is the second.

6. Using the two chromatograms, select two segments that represent the two cases shown in Figure 3. Case 1: Small sulfur peak (S_S) in Channel 1 chromatogram with corresponding large sulfur peak (S_L) in Channel 2 chromatogram; measure the heights of the two peaks above the average baseline signal. Case 2: Large phosphorus peak (P_L) in Channel 1 chromatogram with corresponding small phosphorus peak (P_S) in Channel 2 chromatogram; measure the heights of the two peaks above the average baseline signal.

7. The Alpha value for the P and S modes can be obtained using the values obtained from case 1 and 2. The P mode Alpha value (α_p) is derived by dividing the height of the small sulfur peak (S_S) by the large sulfur peak (S_L).

$$\alpha_P = S_S / S_L \quad \underline{\text{or}}$$

$$\alpha_p = \text{average S response in P gate/average P response in S gate}$$

Similarly, the Alpha value for the S mode (α_S) is derived by dividing the height of the small phosphorus peak (P_S) by the height of the large phosphorus (P_L) peak

$$\alpha_S = P_S / P_L$$

The TBP/THT standard used to generate the two chromatograms shown in Figure 3 produced the following results:

$$S_S = 24, 159; \quad S_L = 65{,}350; \quad \alpha_P = 0.370$$

$$P_S = 1{,}076; \quad P_L = 23{,}729; \quad \alpha_S = 0.045$$

After the Alpha values are calculated, they can be included in the P and S mode specifications using the Gate Parameters window (see Figure 4). The "–" operator value (<op>) signifies that the product of the Alpha and the Gate B integration value is subtracted from the Gate A value. The effect of incorporating appropriate Alpha values for each mode is shown in Figure 5.

Figure 5 shows that the corrected response of each gate results in the disappearance of the unwanted peak. It should be noted that if the calculated Alpha values insufficiently compensate to eliminate the unwanted peak or if there is overcompensation (noted by a negative baseline deflection in the corresponding chromatogram), the Alpha values entered in the Gate Parameters window will have to be adjusted to eliminate the desired peak.

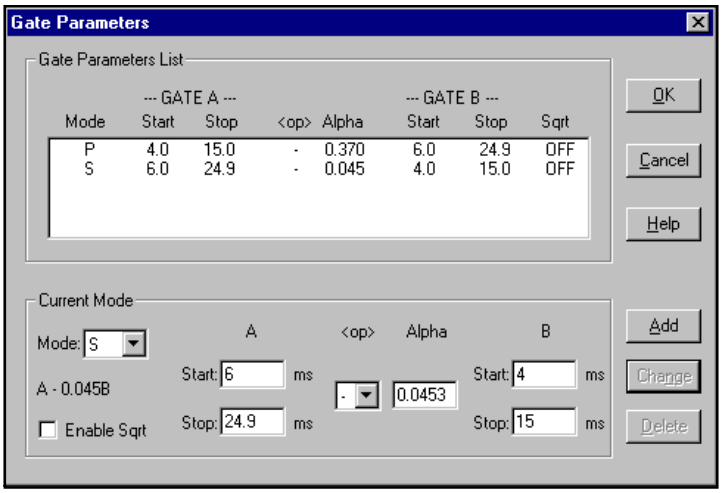

Figure 4. Gate Parameters Values for the Phosphorus (P) and Sulfur (S) Mode Including Alpha Values

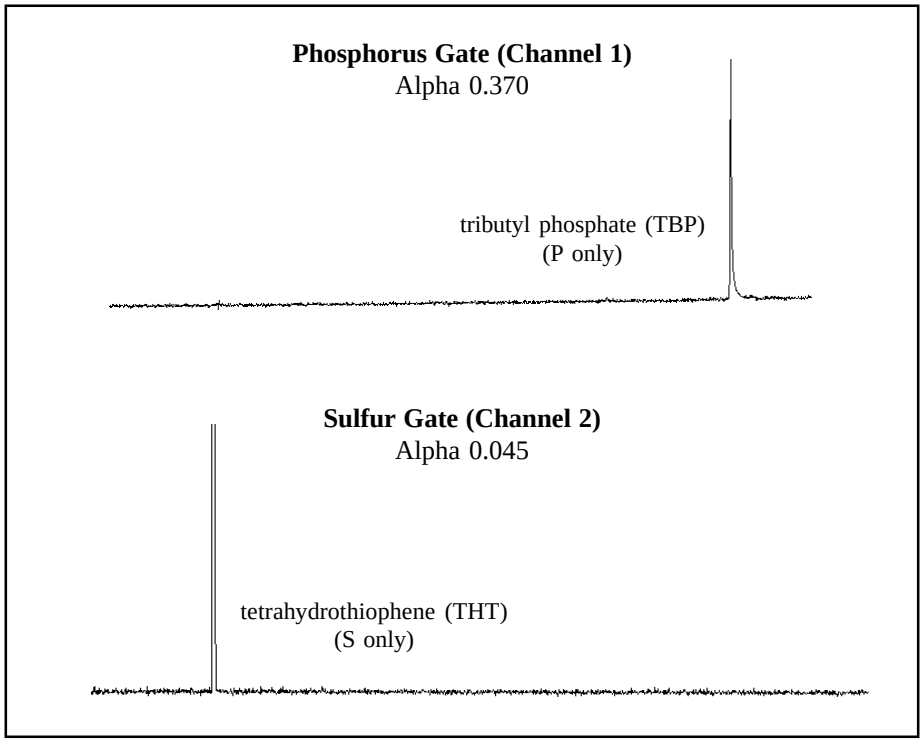

Figure 5. Chromatograms Derived Using Calculated Alpha Values

Using PFPDView and Dual Gate Subtraction to Eliminate Matrix Interferences in Pesticide Analysis

OI Analytical's optional PFPDView software greatly facilitates post-run iteration to estimate appropriate Alpha values. This program allows post analysis observation of the emission waveform at any point during the chromatographic run, and it allows the operator to determine how changes to any gate parameter (not just dual gate effects) will affect the chromatogram. All of this optimization process can be done after running just a single chromatographic analysis with the Model 5380 PFPD. This section describes an example of using PFPDView to quickly and easily obtain an appropriate Alpha value in an application involving the elimination of sulfur matrix interference when analyzing for phosphorus pesticides in an onion extract. Additionally, it is noted how PFPDView can be used to obtain some structural (heteroatom) information on unknown compounds by observing (post-run) the emission profile.

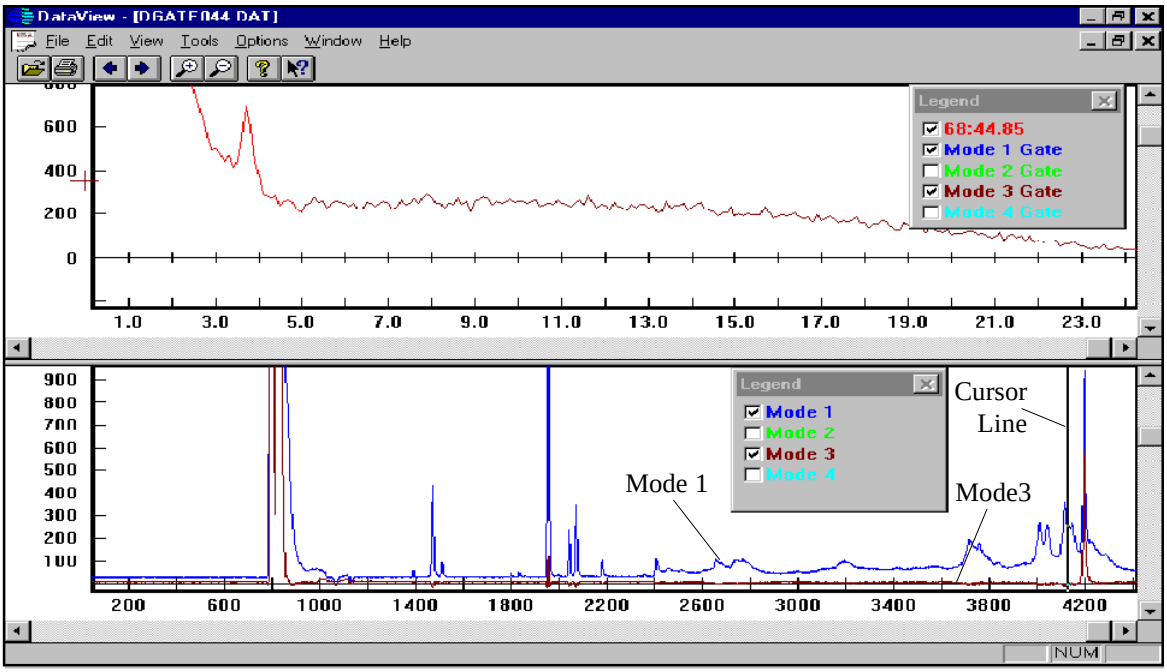

Figure 6. Chromatograms with Marker on S Peak (Mode 1 (α_p = 0) and Mode 3 (α_p = 1.5))

Since there can be considerable interference between sulfur- and phosphorus-containing compounds in gas chromatography, eliminating sulfur response may be critical for selectively detecting low concentrations of phosphorus pesticides. Due to its high matrix background of sulfur-containing compounds, onion juice was selected to demonstrate the effectiveness of the dual gate technique for eliminating unwanted matrix interferences.

Equipment

A Hewlett-Packard (HP) 6890 GC equipped with a Model 5380 PFPD was used in this study. The PFPD was configured with an R1924 photomultiplier tube and a GG 495 filter (optimum for phosphorus detection). Data analysis was performed using HP ChemStation software. Two identical P modes were specified using the WinPulse software program. In order to optimize the phosphorus output, the start and stop times of Gate A were set to 5 and 10 msec, respectively, for each mode. The start and stop times for Gate B were set at 10 and 24.9 msec, respectively, to include the segment of the sulfur emission profile where the P emission intensity is low. Using the WinPulse Board/Channel Parameters window, the first P mode was allocated to Channel 1 of the HP ChemStation software and the second to Channel 2. This setup provided two chromatograms simultaneously for each analysis.

Sample

An onion juice extract was obtained by homogenizing and liquefying an onion sample in a blender and filtering the slurry through filter paper. The sample filtrate was extracted with 0.5 mL of hexane for every 2.5 mL of the filtrate juice and decanted the supernatant. A 20-µL subsample of the supernatant was then spiked with 20 pg of malathion. A 1-µL sample of this spiked onion juice extract was injected into a split/splitless injector of the GC (in the splitless mode). This extract produced the pair of chromatograms as shown in Figures 6 and 7.

PFPDView Software - Post Run Optimization of PFPD Parameters for Dual Gate Operation

The PFPD pulses were saved as a data file using WinPulse. WinPulse data files are imported into the OI Analytical PFPDView program (seen in Figures 6 and 7), which enables the operator to modify many of the original PFPD parameter values in order to determine (without having to rerun the analysis) the effect of such modifications on the resulting chromatograms.

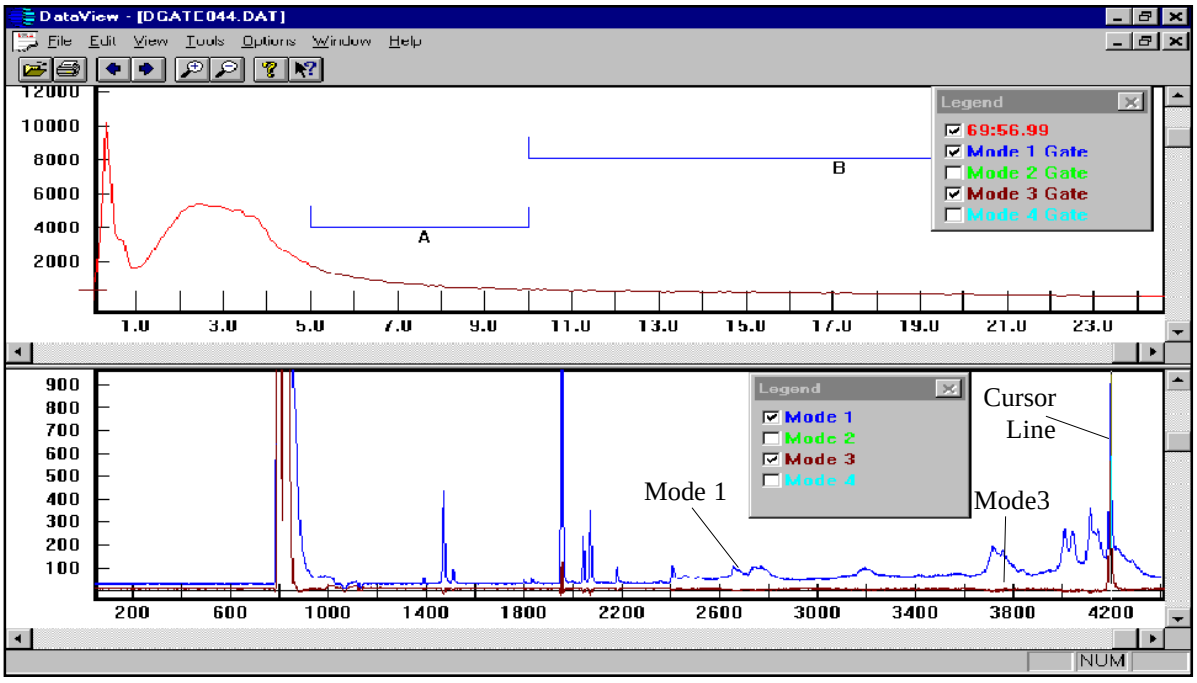

Figure 7. Chromatograms with Marker on P Peak (Mode 1 (α_p = 0) and Mode 3 (α_p = 1.5))

PFPDView also allows the operator to observe the emission time profiles of any peak produced during the analysis. This can provide the operator basic structural information about known or unknown compounds pertaining to a peak on the resulting chromatogram (i.e., based on the emission profile, it indicates if the compound contains sulfur and/or phosphorus). For example, by placing the cursor on the area around 4100 in the chromatogram displayed in the bottom of the PFPDView screen (Figure 6), the upper display shows the actual emission profile for that pulse. The delay and shape of the profile clearly indicate that this area contains S (noted by the characteristic S emission from 5 to 24 msec), and P is not observed. The cursor can be moved throughout the chromatogram to show each pulse that contributed to the chromatogram. Moving the cursor shows this area of the chromatogram to be dominated by the background S matrix, as expected from an onion extract.

Moving the cursor line to the peak at 4200 (Figure 7) displays a waveform now dominated by a P emission with minimal S contribution (note scaling difference from Figure 6). (S emissions from the matrix peaks can be observed by zooming in on the S-gate area of the waveform.) Moving the cursor to the other major peaks in the resulting chromatogram clearly shows these to be peaks containing only sulfur.

In this application, the sulfur response across Gate A was reduced by adjusting the Alpha value in the Gate Parameters window. Using PFPDView, small adjustments are made and the resulting chromatogram is observed until the optimum Alpha value is determined. In this example, an Alpha value of 1.5 is found to eliminate the observed sulfur response. This parameter is entered into the Gate Parameters window in WinPulse and saved to memory for future analyses. Once this parameter is established and entered, WinPulse can be exited because it is not needed to operate the PFPD since all entered parameter values are stored in the detector controller's memory.

Results

Figure 6 shows the PFPDView screen from this particular analysis. In the lower panel of this figure, Mode 1 higher trace) represents the chromatogram derived using Alpha = 0, and Mode 3 (lower trace) represents the chromatogram derived using Alpha = 1.5. By comparing the mode 1 and 3 chromatograms in Figure 6, it is clear that using an Alpha value of 1.5 eliminates all the sulfur peaks except for the large peak at approximately 3 minutes and a small peak at 8.8 minutes. All of this work is done after running a single analysis and optimizing using the PFPDView program. Modified chromatograms can then be saved in PFPDView in AIA format, which can be imported into virtually all data handling software programs (e.g., ChemStation) for reintegration and analysis under the new gate conditions.

The pair of chromatograms that was obtained by rerunning the same analysis using the new gate settings, Alpha values of zero for Channel 1 and 1.5 for Channel 2, and a 1-µL splitless injection of the spiked onion juice extract is shown in Figure 8. Comparison of the two chromatograms confirmed that all of the sulfur response peaks (with the exception of those at 3 and 8.8 minutes) disappeared when Alpha = 1.5 is used. By contrast, the phosphorus peak at 17.2 minutes remained relatively unaffected. The improved selectivity between the S matrix background and the phosphorus-containing pesticide is readily observed by comparing the two chromatograms, which were collected simultaneously on the two channel PFPD.

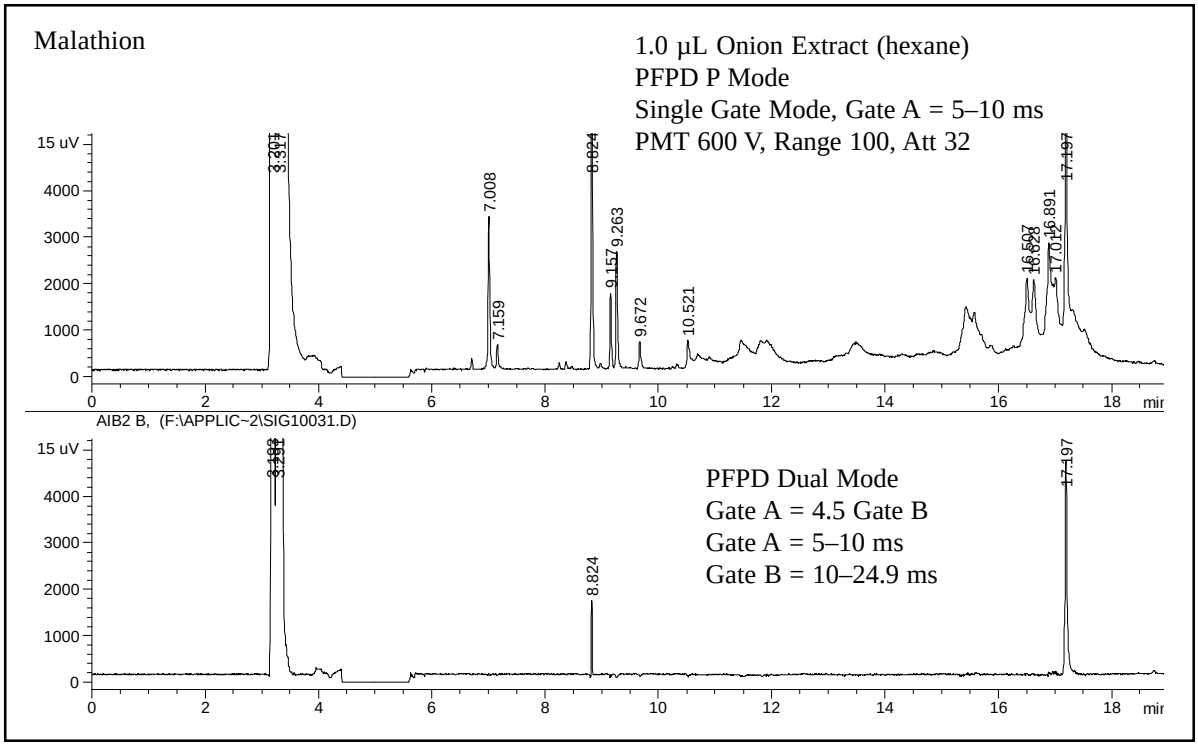

Figure 8. Onion Extract Chromatograms with α_p = 0 in Top Panel and α_p = 1.5 in Bottom Panel

Conclusion

This application demonstrates the analytical power of using the PFPD's dual gate technique to increase the selectivity for specific heteroatoms (interheteroatom selectivity), even in the presence of substantial interference from other heteroatoms. Although this capability is not useful for every combination of elements detectable with the PFPD, for many applications it can offer additional capabilities with the PFPD that are well beyond what can be done with standard flame photometric detectors. The use of WinPulse and PFPDView to conduct post-run manipulations of the raw data greatly increases the ease and speed of optimizing the PFPD detector parameters. This coupled with the improved sensitivity, selectivity, and lower gas consumption versus the FPD makes the PFPD the preferred detector for pesticide residue analysis.

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