Highly Sensitive TOF Mass Spectrometry Allows for New User-Friendly Flow Modulator Design for GCxGC

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Overview

- Flow modulation provides a lower cost alternative to thermal modulation.
- Flow modulation exhibits lower sensitivity than thermal modulation due to splitting or low duty cycle.
- New highly sensitive Time-of-Flight (TOF) mass spectrometers (MS), when coupled to flow modulation, yield performance equivalent to previous generation TOFs coupled with thermal modulation.
- Diverting mode flow modulation provides truly user-friendly GCxGC.

Introduction

Flow modulation has been of growing interest for GCxGC users. Not only those looking to adopt the technique, but also those seeking to avoid the cost of cryogens, which is required for best performance with thermal modulation. To date, flow modulation technologies have revolved around differential flow modulation methods, such as reverse fill flush modulators.[1,2] These types of modulators have some performance drawbacks as well as challenges when a user tries to optimize the instrumental method. These drawbacks include relatively broad chromatographic peak widths when compared to thermal modulation, lower sensitivity when coupled to MS detectors due to the need to split the high flow rates of carrier gas prior to the MS, and the need to manage multiple flow rates and make multiple connections during set up and optimization. Seeley et al. published on a low duty cycle flow modulator that avoids many drawbacks of differential flow modulation.[3] Advances in benchtop TOF mass spectrometers [4,5] help the low duty cycle flow modulator overcome its primary drawback; sensitivity. The extremely sensitive detector brings sensitivity on par, if not better than, the previous generation of widely used TOF-MS coupled to thermal modulation. The low duty cycle flow modulator offers "thermal modulator-like" performance without the need for cryogens, and in an easy-to-use package.

Methods

The LECO FLUX™ diverting flow modulator was coupled to a Pegasus® BT 4D benchtop TOF mass spectrometer. For comparison, a LECO *Pegasus* 4D-C was used to collect thermal modulation data.

Figure 1. (Left) Pegasus BT 4D with FLUX flow modulator; (Right) The inside of the GC oven with the FLUX modulator.

The diverting flow modulator is comprised of two fittings (a cross and a tee), as well as a three-way valve outside of the GC oven to direct an auxiliary gas flow to the modulator (Figure 2). The modulator has two states, divert and inject. In the modulator's divert state the auxiliary gas opposes the effluent from the primary column (sending it to exhaust), while supplying the flow to the secondary column. Actuating the valve sets the modulator in the inject state, and the auxiliary flow is sent straight to exhaust allowing the primary column effluent to be injected onto the secondary column. The modulator cycles continuously between these two states during an analysis to yield a 2D separation.

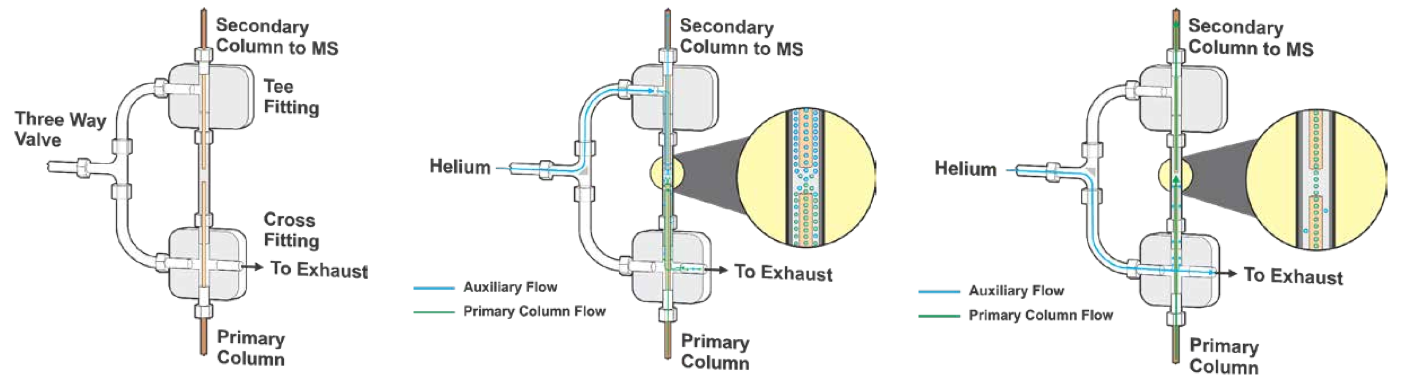

Figure 2. A schematic for the FLUX diverting flow modulator comprised of a three-way valve, two fittings, and a connecting tube between the fittings (left); the diverting mode state (center); and the injecting mode state (right).

Standards and Conditions

Column Configurations

OFN and Grob Mixture Data

Primary Column:	Restek Rxi-5 MS 30 m x 0.25 mm x 0.25 µm
Secondary Column:	Restek Rxi-17Sil MS 0.75 m x 0.10 mm x 0.10 µm

Kirby Fuel Oil Sample Data

Primary Column:	Restek Rxi-1 MS 60 m x 0.25 mm x 0.25 µm
Secondary Column:	Trajan/SGE BPX50 1.25 m x 0.10 mm x 0.10 µm

Experimental Conditions

OFN

GC Method		*MS Method*	
Column 1 Flow Rate:	1.4 mL/min	Ion Source Temp:	250 °C
Inlet Split Ratio:	100:1	Acquisition Rate:	200 sp/s
Inlet Temp:	250 °C	M/Z Range:	45–350
Oven Ramp:	60 °C for 0.5 min, then ramping to 180 °C at 60 °C/min; 2.60 min hold before ramping to 250 °C at 60 °C/min	Electron Energy:	70 eV
Transfer Line Temp:	250 °C		
Secondary Oven Offset Temp:	+5 °C		
FLUX Modulation Period:	0.6 s		
FLUX Inject Duration:	0.05 s		
Thermal Modulation Period:	1.5 s		
Thermal Hot Jet/Cold Jet:	0.45/0.30 s		
Thermal Modulator Offset Temp:	+15 °C		

Grob Mix

GC Method		*MS Method*	
Column 1 Flow Rate:	1.4 mL/min	Ion Source Temp:	250 °C
Inlet Split Ratio:	100:1	Acquisition Rate:	200 sp/s
Inlet Temp:	250 °C	M/Z Range:	45–350
Oven Ramp:	40 °C for 0.5 min, before ramping to 250 °C at 20 °C/min	Electron Energy:	70 eV
Transfer Line Temp:	250 °C		
Secondary Oven Offset Temp:	+5 °C		
FLUX Modulation Period:	0.6 s		
FLUX Inject Duration:	0.05 s		
Thermal Modulation Period:	1.5 s		
Thermal Hot Jet/Cold Jet:	0.45 s/0.30 s		

Intermediate Fuel Oil, Kirby

GC Method		*MS Method*	
Column 1 Flow Rate:	1.0 mL/min	Ion Source Temp:	220 °C (thermal) 250 °C *(FLUX)*
Inlet:	splitless	Acquisition Rate:	50 sp/s (thermal)100 sp/s *(FLUX)*
Inlet Temp:	310 °C	M/Z Range:	40–600
Oven Ramp:	50 °C for 15 min, before ramping to 335 °C at 1.5 °C/min	Electron Energy:	70 eV
Transfer Line Temp:	320 °C	*1µL volume was used for all injections.*	
Secondary Oven Offset Temp:	+5 °C		
FLUX Modulation Period:	5 s		
FLUX Inject Duration:	0.08 s		
Thermal Modulation Period:	7.5 s		
Thermal Hot Jet/Cold Jet:	0.75 s/3.0 s		
Thermal Modulator Offset Temp:	+5 °C		

Results

The *Pegasus* BT 4D with *FLUX* was compared to the previous generation TOF, the *Pegasus* 4D-C with thermal modulation. The GCxGC detection limit for the *Pegasus* 4D-C is 1 pg of OFN on column. Figure 3 shows that the *Pegasus* BT 4D with *FLUX* has the same GCxGC detection limit, in spite of the low duty cycle modulator, due to the more sensitive BT TOF-MS. For 1D GC the *Pegasus* BT demonstrates an LOD of 20 fg of OFN on column.

In Figure 4, the Grob mixture runs under the same conditions. The flow modulator yielded slightly broader peak widths than the thermal modulator (but are still comparable). This means similar peak capacities are achieved. Table 1 compares the peak widths for a few components of the mixture. The *FLUX* flow modulator chromatographic performance is also on par with the quad jet thermal modulator. An additional benefit of low duty cycle flow modulation is the flow rates are similar to 1D GC and thermal modulation, making for easy method translation.

While the *Pegasus* BT 4D with *FLUX* provides comparable results to the *Pegasus* 4D-C, it also is extremely easy to set up and operate, providing a low-cost alternative to thermal modulation. Figure 5 shows how to set up the modulator and install columns. Only two connections are required to make the system fully operational (beyond the GC inlet connection and transfer line connection, which are required for 1D GC as well).

To install the primary column, thread the column through the 360 µm captured ferrule and nut, and insert up through the bottom of the modulator until it comes to a hard stop. Tighten the 360 µm nut using the red hand tool to finish the connection. Repeat this process for the secondary column, inserting it down through the top of the modulator.

Method development is also user-friendly, as column flows and heating rates can be carried over from 1D GC or thermal modulation GCxGC methods. The software controls the auxiliary gas flow, so the user needs only to input a second dimension time (modulation period) and inject duration. The inject duration has a drop down menu with three options for all around performance, higher sensitivity, or higher peak capacity (see Figure 6). The modulation period should be set to 1/3 of the peak width (produced from a 1D GC run). This ensures adequate sampling of the primary column effluent to maintain quantitative precision.[6] Figure 7 shows a complex sample comparison between the *Pegasus* 4D-C and *Pegasus* BT 4D with *FLUX*. An intermediate fuel oil sample known as Kirby was analyzed using the *Pegasus* 4D-C with thermal modulation by Robert Nelson and Chris Reddy from the Woods Hole Oceanographic Institution. The same sample was then analyzed using the *Pegasus* BT 4D with *FLUX* flow modulation. Similar separations are produced using the two systems with only minor changes to the methods. The volatile end is missing from the *Pegasus* BT 4D with *FLUX* data due to the sample being evaporated down prior to shipping for analysis with the *Pegasus* BT 4D with *FLUX* system.

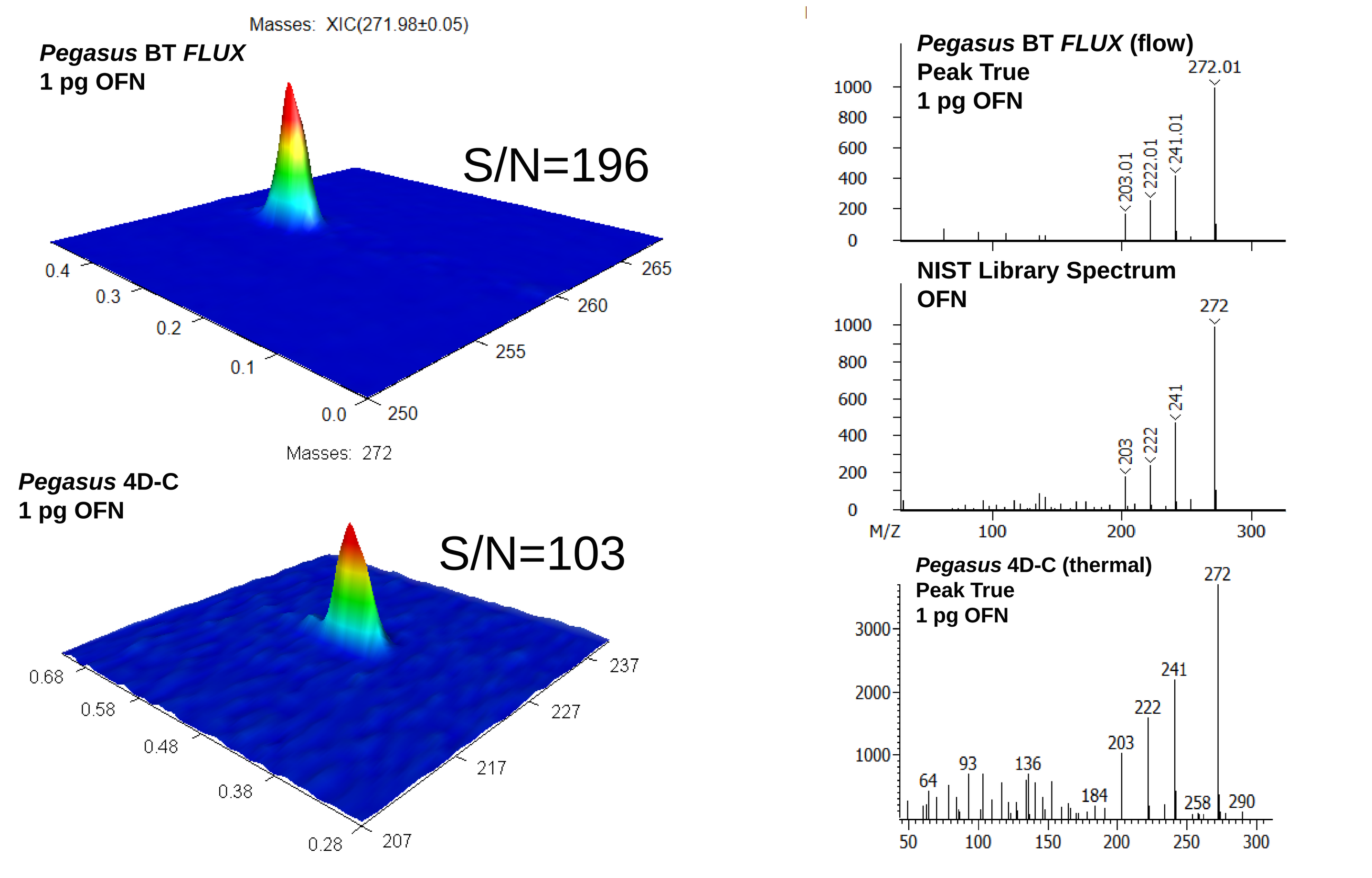

Figure 3. (Top Left) The XIC GCxGC surface plot for 1 pg of OFN on column collected using the Pegasus BT 4D with FLUX; (Bottom Left) The XIC GCxGC surface plot for 1 pg on column of OFN collected using the Pegasus 4D-C; (Top Right) The peak true mass spectrum for 1 pg OFN on the Pegasus BT 4D with FLUX; (Middle Right) A reference library mass spectrum of OFN for comparison; (Bottom Right) The mass spectrum for 1 pg of OFN on the Pegasus 4D-C.

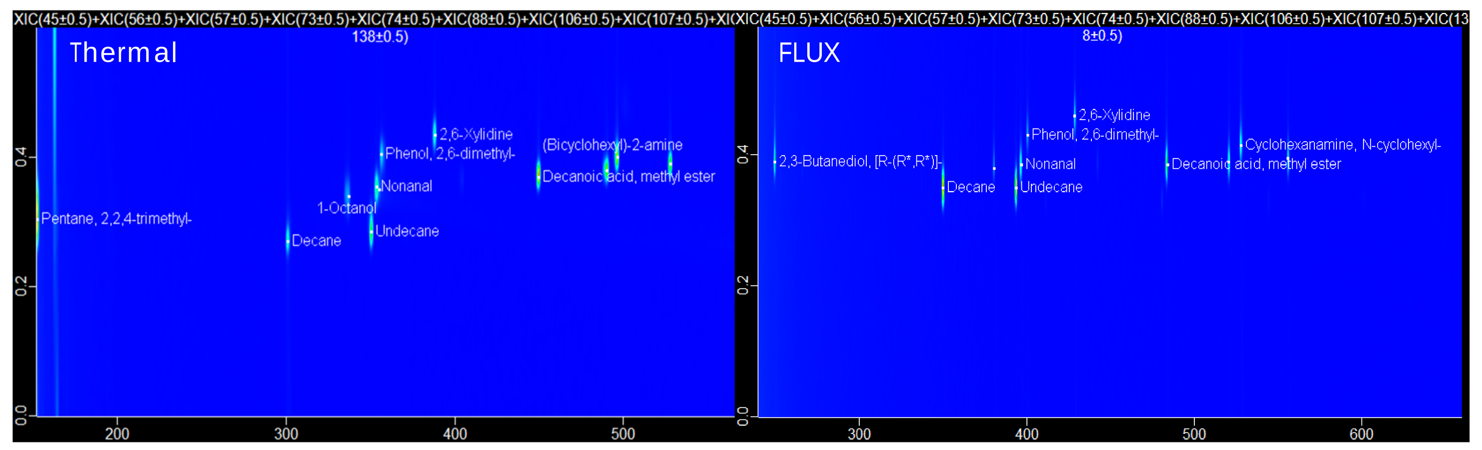

Figure 4. (Left) Grob mixture run using thermal modulation; (Right) FLUX flow modulator.

Table 1. Below, peaks full width at half height (FWHH) are compared between the FLUX flow modulator and thermal modulator for a few select analytes from the Grob mix.

Instrument	FWHH (msec)	Instrument	FWHH (msec)	Instrument	FWHH (msec)	Instrument	FWHH (msec)
Undecane		Dimethylaniline		Methyl Undecanoate		Dicyclohexylamine	
Thermal	39	Thermal	35	Thermal	27	Thermal	30
FLUX 1	41	FLUX 1	46	FLUX 1	43	FLUX 1	46
FLUX 2	41	FLUX 2	45	FLUX 2	42	FLUX 2	42
FLUX 3	37	FLUX 3	40	FLUX 3	38	FLUX 3	38

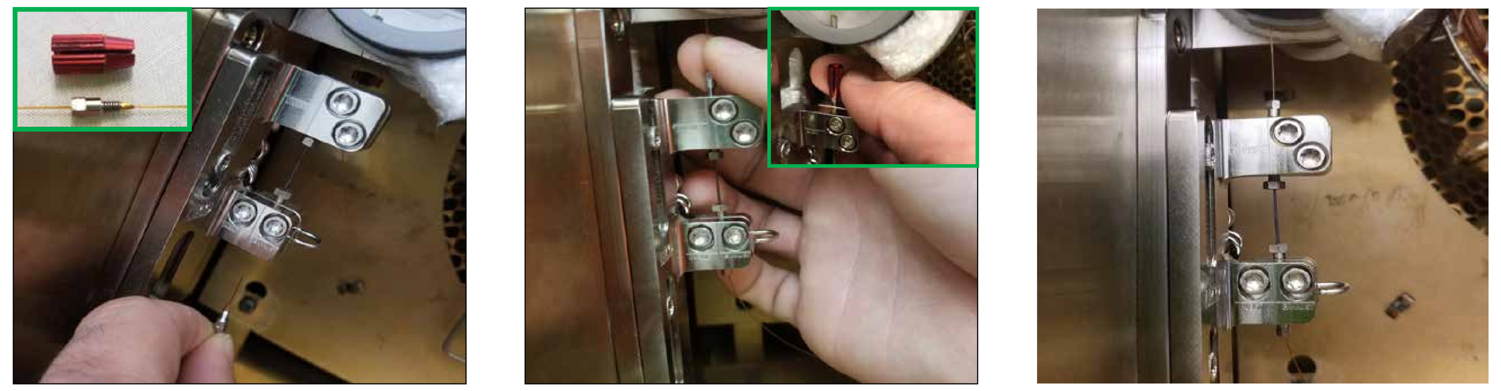

Figure 5. The installation of the columns procedure into the FLUX modulator. (Left) The primary column inserts through the bottom of the modulator until it comes to a hard stop. The captured nut (upper left inset) and ferrule is tightened using the provided hand tool (upper left inset). (Middle) The secondary column threads down through the top of the modulator until it encounters a hard stop, and like the primary column the nut with ferrule is tightened using the hand tool to provide a robust leak-free connection. (Right) The system is assembled and ready to run.

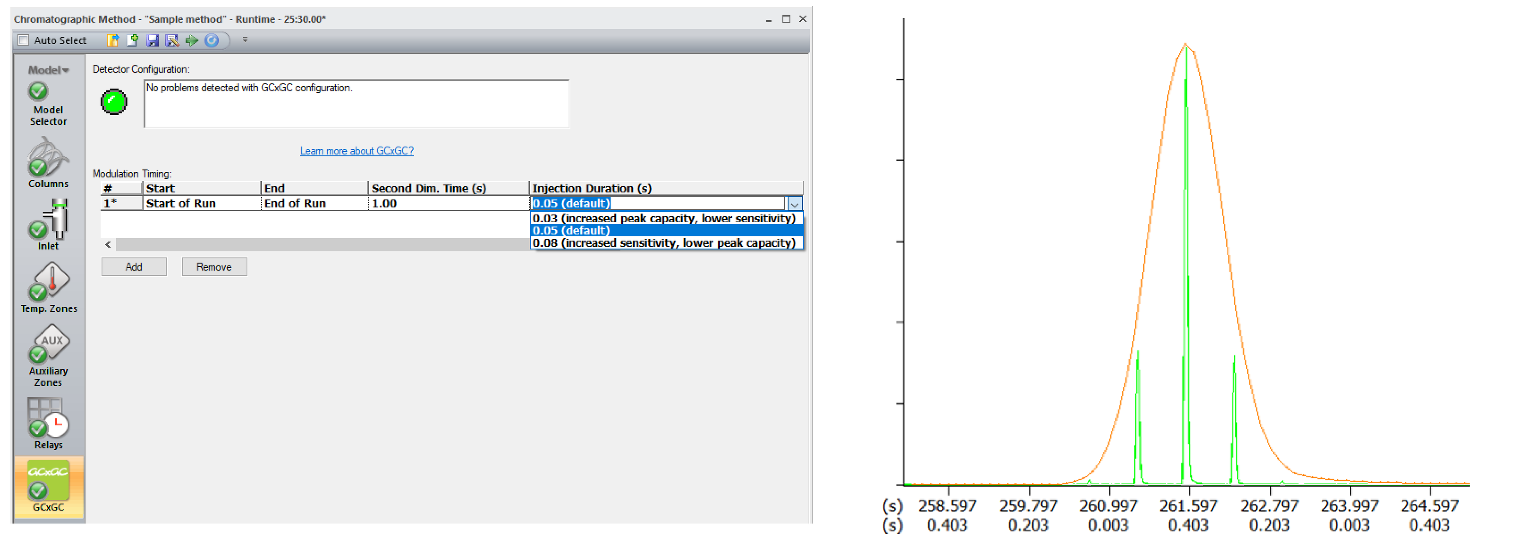

Figure 6. (Left) Method setup is simple for the FLUX modulator. The inject time for the modulator is selected from a drop down menu with three options: default, an option for increased sensitivity, and an option for increased peak capacity. The modulation period should be set to 1/3 of the peak width (FWHH) produced by a 1D GC run; (Right) This will sample the chromatographic peak eluting off the primary column (orange) at least 3 times (green), maintaining quantitative accuracy.

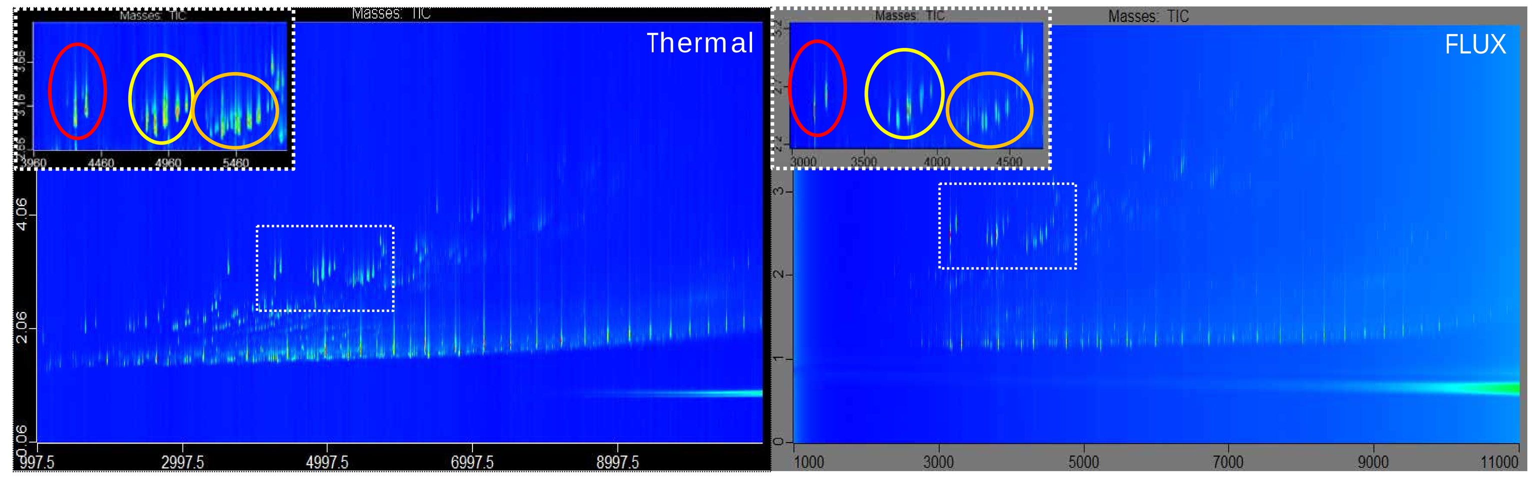

Figure 7. (Left) A contour plot acquired using the Pegasus 4D-C for the Kirby fuel oil sample analysis; (Right) A contour plot acquired using the Pegasus BT 4D with FLUX for the same sample. The contour plots are similar, except for the Pegasus BT 4D with FLUX some of the volatile analytes early in the run are missing. This is because the sample was shipped from the WHOI (MA) which acquired the thermal Pegasus 4D-C data, prior to shipping the solvent was evaporated off over night which also led to the loss of low boilers. The sample was later reconstituted and run via the Pegasus BT 4D with FLUX. A zoom-in of the alkyl naphthalenes (red), dialkyl napthalenes (yellow), and trialkyl napthalenes (orange) for the Pegasus 4D-C can be found in the upper left inset, and for the Pegasus BT 4D with FLUX in the upper right inset. Both instruments adequately separate out individual compounds which group together by class.

Conclusions

The *Pegasus* BT 4D TOF mass spectrometer offers a significant improvement in sensitivity over the previous generation of TOF-MS. This helps to overcome the primary drawbacks to diverting flow modulation, low duty cycle and a lack of cryo-focusing. The *Pegasus* BT 4D with *FLUX* flow modulator has been shown to provide equivalent sensitivity to the *Pegasus* 4D-C thermal modulator system, as well as comparable chromatographic performance for GCxGC. The system is easy to set up, and method development is also user-friendly providing a benefit over other differential flow based modulators. Therefore the *Pegasus* BT 4D with *FLUX* is a viable lower cost alternative to thermal modulation, and a user-friendly alternative to differential flow modulation based GCxGC-MS systems.

Acknowledgement

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