



(51) International Patent Classification:  
**G01N 30/32** (2006.01)

(21) International Application Number:  
PCT/US2009/005361

(22) International Filing Date:  
29 September 2009 (29.09.2009)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:  
61/194,625 29 September 2008 (29.09.2008) US

(71) Applicant (for all designated States except US): **THAR INSTRUMENTS, INC.** [US/US]; 575 Epsilon Drive, Pittsburgh, PA 15238 (US).

(72) Inventors; and

(75) Inventors/Applicants (for US only): **SIDHU, Harbaksh** [US/US]; 4657 Wellworth Court, Allison Park, PA 15101 (US). **WANG, Ziqiang** [US/US]; 116 Country Lane, Lansdale, PA 19446 (US). **WEBSTER, Michael** [US/US]; 4023 Ewalt Road, Gibsonsia, PA 15044 (US).

(74) Agents: **JANIUK, Anthony, J.** et al.; Waters Technologies Corporation, 34 Maple Street, Milford, MA 01757 (US).

(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

— without international search report and to be republished upon receipt of that report (Rule 48.2(g))

(54) Title: PARALLEL SCREENING SUPERCRITICAL FLUID CHROMATOGRAPHY

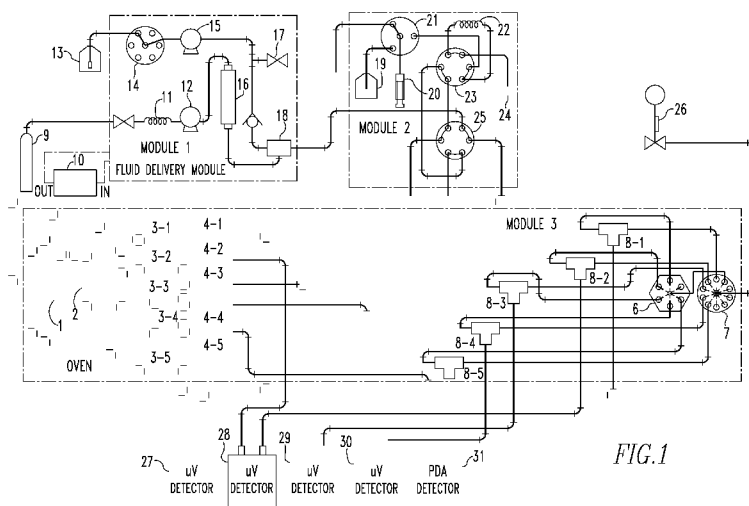


FIG.1

(57) Abstract: The invention provides an apparatus for supercritical fluid chromatography. The apparatus comprises a binary pump; an autosampler; a sampling valve; a first and second port switching valve; a first and second manifold; two or more channels, each having a check valve assembly, a separation column and one or more detectors operatively connected thereon; and a backpressure regulator. The apparatus also includes computer software and hardware to control distribution of fluid through the apparatus, including switching between a multi-channel mode or a single channel ode; 2) analyze data collected by the one or more detectors; and 3) optimize separation of analytes by controlling solvent combinations, concentration gradients, pressure and temperature. The apparatus excludes additional backpressure regulators or pumps on individual channels. Also provided is a method of screening a sample, using supercritical chromatography, using the above apparatus, where multiple samples can be screened simultaneously with parallel processing.

**PARALLEL SCREENING SUPERCRITICAL FLUID  
CHROMATOGRAPHY**

**CROSS-REFERENCE TO RELATED APPLICATIONS**

- 5 This application claims priority to provisional application Serial No. 61/194,625, filed September 29, 2008, incorporated by reference in its entirety.

**FIELD OF THE INVENTION**

- 10 This invention relates to an apparatus and method of parallel processing of two or more samples in supercritical fluid chromatography.

**BACKGROUND INFORMATION**

- Chromatography is a technique used to, among other things, separate component elements of a starting material. Within the general field of chromatography, there are several types, including normal phase and reverse phase. Supercritical fluid chromatography (SFC) is a high pressure method that typically operates near or above the critical point of the mobile phase fluid and offers significant speed advantage and resolution over traditional techniques such as high performance liquid chromatography (HPLC). SFC employs carbon dioxide or another compressible fluid as a mobile phase, sometimes with a co-solvent, to perform a chromatographic separation. SFC has a wide range of applicability and typically uses small particle sizes of 3-20 microns for column packing material and is for analytical to preparative scale applications because of the lower pressure drop. In HPLC applications pressure at the top of the column typically reaches up to several thousand or even tens of thousands psi but pressure at the bottom is reduced to ambient pressure, creating a significant pressure drop.
- 30

The growing role of supercritical fluid chromatography (SFC) for separations in such industries as pharmaceuticals and fine chemicals, along with the diversified nature of the compounds being used in such industries, requires new optimized method development methodology and systems for employing such new methodology. SFC is a normal phase chromatography technique and requires screening of solvents and columns for determining conditions for the separation. The purpose of a separation could be an analytical screen or an optimized method for isolation of compound in larger quantities. Currently, in the SFC process the mixture or compound is injected sequentially into a system that contains a binary pumping system for CO<sub>2</sub> and co-solvent, an autosampler for injections, a detector(s) for detecting the component elements of the starting mixture or compound and an automated back pressure regulator for pressure regulation. The user of the method and system repeats the injection in a sequential fashion by varying gradient conditions, columns, solvents, pressure and temperature until a desired result is obtained. It is highly desirable to obtain these results quickly and in an automated fashion. Once results are obtained, the results can be reviewed by the user and further optimized by selecting additional conditions to verify the results.

The sequential nature of current SFC processes and systems can make the process of optimization time-consuming. Therefore, there is a need for an effective method and apparatus for processing multiple mixtures and compounds in parallel instead of in sequence, which would significantly reduce the time and cost of processing by optimizing methods faster.

### SUMMARY OF THE INVENTION

The present invention is directed to an apparatus and methods of processing samples and mixtures in parallel mode, that overcomes some of the drawbacks experienced in the prior art of sequential  
5 processing.

Accordingly, in one aspect the present invention provides an apparatus for supercritical fluid chromatography, the apparatus comprising: a binary pump; an autosampler; a sampling valve; a first  
10 and second port switching valve; a first and second manifold; two or more channels, each having a check valve assembly, a separation column and one or more detectors operatively connected thereon; computer software and hardware to 1) control distribution of fluid through the apparatus, including switching between a multi-channel  
15 mode or a single channel mode; 2) analyze data collected by the one or more detectors; and 3) optimize separation of analytes by controlling solvent combinations, concentration gradients, pressure and temperature; and a backpressure regulator, wherein the apparatus excludes additional backpressure regulators or pumps on individual  
20 channels.

In an additional aspect, the present invention provides a method of screening a sample using supercritical fluid chromatography, the method comprising the steps of mixing a sample containing analytes  
25 of interest with a supercritical fluid and optionally, a suitable solvent, to create a mixed mobile phase; moving the mixed mobile phase along a flow path to an autosampler; injecting a single portion of the mixed mobile phase into the autosampler; moving the single portion along the flow path through a first column switch valve to a first manifold;  
30 dividing the single portion into two or more subportions and directing each of the two or more subportions to a separate channel for

separation; simultaneously moving each of the two or more subportions along each separate channel through a separation column and through one or more detectors; moving the two or more subportions through a second check valve and a second manifold to  
5 create a single exit stream; and moving the exit stream along the flow path to the backpressure regulator.

These and other aspects of the invention will become more readily apparent from the following detailed description, drawings, and  
10 appended claims.

#### BRIEF DESCRIPTION OF THE DRAWINGS

The invention is further illustrated by the following drawings in which:

Figure 1 is a diagram of one embodiment of the apparatus of the  
15 invention.

Figure 2 is a diagram of another embodiment of the apparatus of the invention.

Figure 3 is a diagram of an embodiment of the check valve assembly of the invention.

20

#### DETAILED DESCRIPTION OF PREFERRED EMBODIMENTS

As used herein in the specification and claims, including as used in the examples and unless otherwise expressly specified, all numbers may be read as if prefaced by the word "about", even if the term does  
25 not expressly appear. Also, any numerical range recited herein is intended to include all sub-ranges subsumed therein.

The present invention describes a method and apparatus for parallel supercritical fluid chromatography (SFC).

30

In an illustrated embodiment of the present invention, the method of parallel screening processes samples on 2 or more columns simultaneously within the same process.

5 The method of the illustrative embodiment includes simultaneously processing samples by supercritical fluid chromatography (SFC) on 5 channels on an analytical system (as shown in Figure 1). The number five is arbitrary, for ease of description; any number of channels can be designed into the system, as determined by the needs  
10 of the user. Parallel screening can be accomplished on any number of channels, such as 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, up to and including 96 channels, or even more channels, as needed. The method includes passing a portion of an original sample along a SFC flow path to a first channel,  
15 separating the sample portion into a profile of single peaks, and moving the peaks down the flow path. The pressure of the supercritical fluid in the flow stream is regulated by a backpressure regulator downstream. The method also includes passing the separated peaks down the path to an analyzer that is inserted in this  
20 same first channel flow path for peak characteristics determination.

The method further includes moving the flow stream, after it passes through the analyzer from this first channel, to a manifold (or valve/manifold) for merging with flow streams from other channels to  
25 recombine the samples for further analytical processing if any, such as splitting a portion of the sample to a mass spectrometry analyzer for molecular identification; or splitting a portion of the sample to an evaporative light scattering detector for confirmative quantitative analysis, before the samples pass to a backpressure regulator at the  
30 end of the flow path.

The parallel screening method of this illustrated embodiment further includes processing another portion of the sample along a second channel simultaneously with the processing of the first channel.

- 5 Processing on the second channel includes passing another portion of the original sample along a SFC flow path to a second channel, separating the sample into a profile of single peaks, and moving the peaks down the flow path. The pressure of the supercritical fluid in the flow stream of the second channel is also regulated by the same  
10 backpressure regulator downstream that is used for first channel. The method also includes passing the separated peaks down the flow path to an analyzer for peak characteristics determination. This analyzer, like the one used in the first channel, functions in a similar fashion, and is also fully dedicated to the use on this second channel  
15 only. However, the type of analyzer is not necessarily the same as the first one; it can be any type of analyzer, depending on the purpose of the design. The software is programmed to collect all data types of interest, depending on the type of detector or detectors used on a channel, for example including, but not limited to, absorptions under  
20 any specified ultra-violet wavelength (UV), full spectrum determination by photodiode array detector (PDA), light scattering detector (ELSD) and mass spectrometer (MS), flame ionization detector(FID), chemiluminescence nitrogen detector(CLND), corona aerosol detector(CAD), circular dichroism(CD) and other chiral detectors, and  
25 on-line infrared(IR) and nuclear magnetic resonance detectors(NMR).

The method further includes, after passing through the analyzer, moving down the flow stream from this second channel to the same manifold (or valve/manifold) used on first channel, for merging with  
30 flow streams from other channels including the first channel described above, to recombine the portioned samples for further

analytical processing, if desired, before passing the sample to backpressure regulator at the end of the flow path.

In the illustrative embodiment of the invention, the method of parallel  
5 screening includes processing a portion of the original sample along a third, fourth and fifth channel in a manner similar to the processes discussed above regarding to the first and second channels. In this embodiment, a dedicated analyzer is used for analyzing sample portions in each of corresponding channels before passing the  
10 samples to the same manifold (or valve/manifold) to recombine the samples with all other channels moving down stream for any further analytical processing, if any. The combined stream continues to move down the flow path to the same backpressure regulator.

15 In addition to the parallel screening apparatus and methods described above, in an additional embodiment the present invention also is directed to an apparatus and methods for processing samples and mixtures in the conventional sequential mode. In an illustrated embodiment of the present invention (as shown in Figure 2), a method  
20 of conventional sequential screening processes samples on 2 or more columns in a sequential manner within the same process.

The method of the illustrative embodiment includes sequentially processing samples by supercritical fluid chromatography (SFC) on 5 channels in an analytical system. As would be understood by one  
25 skilled in the art, the system and apparatus can be adapted to sequential screening on any number of channels, such as 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, up to and including 96 channels, or even more channels, depending on the needs of the user.

30



The method includes passing injected samples along the flow path to any single channel by a selective valve switch. Once the stream moves along the specific channel, flow control is realized through the combination of a switching valve and a directional valve so that the flow will only move along any single specific channel at one time. The method further separates the sample, and moves the sample down the flow path to an analyzer inserted in that channel for peak characteristics determination. After passing through the analyzer, the method moves the fluid stream through the manifold (or valve/manifold) and it continues downstream to a backpressure regulator. The pressure of the supercritical fluid in the channel is controlled by the backpressure regulator.

In this embodiment of the invention, the method of conventional sequential screening mode processes a sample through a first, second, third, fourth and fifth channel (or any number of channels) in a sequential way. On each channel the sample moves along the flow path, is separated via a separation column, is passed through a dedicated analyzer in that channel for determination, then moves down stream through the manifold (or valve/manifold) and passes on to the back pressure regulator. This apparatus and method enables the invention to possess both novel parallel processing along with a conventional sequential processing mode into one embodiment for various analytical applications.

Referring to Figure 1, in a parallel processing embodiment the apparatus comprises a binary pump for pumping a supercritical fluid such as carbon dioxide, and a modifier or co-solvent such as methanol, into the system. The injector injects the mixture into a line connected to a switching valve, which directs the flow into a valve manifold having check valves (directional valve). The valve manifold directs the flow (or a portion of the flow) into one or more separation

columns for separation. On each line is placed a separate detector, such as a UV detector as depicted in Figure 1. Any type of detector may be placed on any line. The flow is then directed to a second manifold which is connected to a second switching valve. A

5 backpressure regulator regulates the pressure of the entire system.

The analytical processing of samples by parallel channel screening, as well as focused optimizations within a single channel, is achieved by chromatography, and in particular by supercritical fluid chromatography (SFC), discussed in greater detail below.

10

The following reference numerals refer to the following components shown in Figures 1, 2 and 3:

Module 1. Binary fluid delivery module

Module 2. Autosampler

15 Module 3. Column oven

Items:

**1** port switching valve upstream;

**2** parallel screening manifold upstream;

20 **3-1** check valve assembly

**3-2** check valve assembly

**3-3** check valve assembly

**3-4** check valve assembly

**3-5**, check valve assembly

25 **4-1** channel 1;

**4-2** channel 2;

**4-3** channel 3;

**4-4** channel 4;

**4-5** channel 5;

30 **5-1** to **5-5** check valve;

**6** downstream manifold,

- 7** port switching valve downstream;
- 8-1** tee.
- 8-1** tee.
- 8-1** tee.
- 5 **8-1** tee.
- 8-5** tee.
- 9** CO<sub>2</sub> Supply (Customer Supplied)
- 10** Cooling Water Bath
- 11** Cooling Exchanger
- 10 **12** CO<sub>2</sub> Pump
- 13** Modifier Reservoir
- 14** Solvent Valve
- 15** Modifier Pump
- 16** Dampener Chamber
- 15 **17** Prime/ Purge Valve
- 18** Static Mixer
- 19** Buffer Solution
- 20** Syringe
- 21** Valve
- 20 **22** Sample Loop
- 23** Injection Valve Load Position
- 24** Sample
- 25** ISS Valve
- 26** ABPR Automated BPR
- 25 **27** UV Detector
- 28** UV Detector
- 29** UV Detector
- 30** UV Detector
- 31** UV Detector
- 30 **32** Screw
- 33** bracket

- 34** line to bottom of assembly
- 35** block
- 36** check valve
- 37** check valve housing
- 5 **38** line fittings(nut and ferrule)
- 39** line from manifold
- 40** line to channel/column

In Figure 1, the Module-1 box is the single binary fluid delivery  
10 module that delivers supercritical fluidic CO<sub>2</sub> through the CO<sub>2</sub> pump,  
and mixes it with a co-solvent that is pumped by the modifier pump.  
The CO<sub>2</sub> is supplied from either a stand-alone cylinder or a bulk  
supply line to a nominal pressure range by a boosting mechanism or  
something similar, to a pressure optimal for instrument operation,  
15 and pumped to the supercritical state. The co-solvent is chosen  
through the solvent selection valve from various types and  
combinations of chromatographic compatible solvents, and pumped  
by the modifier pump to the mixer for mixing with the supercritical  
CO<sub>2</sub>, inside **module-1**.

20

This mixed mobile phase moves along the flow path to reach **module-**  
**2**, the autosampler. In the autosampler, only one single injection of  
the sample is made, and the injected sample is switched into the flow  
stream by an injection valve. The method of injection can be an  
25 autosampler with sample handling capability, or a manual injection  
valve can be used. The illustrated embodiment uses an autosampler  
with plate handling capability.

Once the sample is injected, the mixed mobile stream passes along the  
30 flow path to reach **module-3**, the column oven, with an operational  
temperature range from 40°C to 90°C normally for a supercritical state

of the mobile phase. The oven also houses the valve system that is designed to realize the parallel (and/or single) screening mode. For the desired parallel screening mode, the stream first passes through column switch valve **1**, and moves along by designated port that leads to manifold **2**. Through manifold **2** the injected sample portion can be divided in multiple ways. One portion will move through check valve assembly **3-1** onto first channel **4-1**.

On channel **4-1**, an SFC column is installed to carry out the separation for this portion of sample for analysis and detection purposes. After the separation is achieved in this channel, the separated portion of the sample moves downstream to a dedicated detector in this channel, which is a UV detector in the illustrated embodiment. The characteristics of the separated sample are determined in the detector and the corresponding data is acquired and recorded by software along with specific channel information for identification purposes. Multiple detectors can be installed along this first channel (or along any channel) depending on the needs of the user.

After the sample portion passes through the detector, it moves through check valve **5-1**, manifold **6**, and to an automated backpressure regulator (ABPR) for the completion of process.

Within the sample time scale as the first portion is going through the first channel, a second portion of the sample divided at the manifold **2**, will pass through check valve assembly **3-2** onto the second channel **4-2**. On channel **4-2**, an SFC column is installed to carry out the separation for this portion of the sample for analysis and detection purposes. After the separation is achieved in this second channel, the separated portion of sample moves downstream to a dedicated detector

on this second channel, which is a UV detector in the illustrated embodiment. The characteristics of the separated sample are determined in the detector and the corresponding data is acquired and recorded by software along with specific channel information for  
5 identification purposes. Multiple detectors can be installed along this second channel according to user preferences.

After the second sample portion passes through the detector, it moves through the check valve **5-2** and manifold 6, where it recombines with  
10 the first portion into the same stream, and moves to the automated backpressure regulator (ABPR) for the completion of process. It is noted that multiple detectors as well as other types of apparatus such as a fraction collector, can also be installed at any point along the flow path, both before and optionally after the ABPR, to achieve various  
15 detection purposes.

In a similar manner, the 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> portions of the divided sample are simultaneously flowing through the corresponding 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> channels for analysis. In the illustrated embodiment, UV detectors are  
20 used on the 3<sup>rd</sup> and 4<sup>th</sup> channel, while the photodiode array detector (PDA) is used on the 5<sup>th</sup> channel. All portions of the original sample will pass through the detectors after separation, and after the check valves **5-3**, **5-4** and **5-5**, will recombine together at manifold 6 with aforementioned 1<sup>st</sup> and 2<sup>nd</sup> portions, and will continue to move down  
25 the flow path to the backpressure regulator.

Whilst this parallel screening mode is realized by the illustrated embodiment, the present invention also has the designed feature of single channel capability that enables sequential screening as well as  
30 a single channel operation mode for optimizing conditions or

experimenting on any one single channel for desired separation performance.

In the illustrated embodiment, if a single channel mode is desired, the  
5 designed valve system in **module-3** is used to achieve this feature.  
For example, if screening on channel 3 is desired, the column switch  
valve is programmed to a certain position such that the injected  
sample will only flow through directly to check valve assembly **3-3**,  
thus bypassing the manifold **2**. In this way the injected sample will  
10 only flow down channel **4-3** for analysis purposes, and will be  
prevented from flowing to any other direction in any other channels,  
by the directional mechanism of the valve system in **module-3**.  
For example, if channel 3 is selected, the 1<sup>st</sup> switch valve will rotate to  
the port that is directly connected to check valve 3-3, so the stream  
15 will flow directly to the tee point of 3-3, but not through the manifold  
2, because check valve 3-3 will only allow flow to go one direction, so  
the flow can not flow backwards to manifold 2, but instead will only be  
able to go down channel 4-3 and so on. In this way a single channel  
capacity is generated.

20  
In the same manner, each and every individual channel in the  
illustrated embodiment is also programmed to have a single channel  
screening mode. The sequential screening for sample analysis is  
therefore realized by injecting samples in the single channel mode one  
25 at a time with any desired combination of channels, as programmed  
through the controlling software.

In the sequential screening mode, it is noted that multiple detectors  
and other apparatus can be installed on any desired channel as well  
30 for specific application purposes, just as they are used in the parallel

screening mode.

In the methods of the present invention, a valve system, together with the other hardware and software, enables the simultaneous screening  
5 on all channels with columns.

The valve assembly is a combination of a check valve shown in Figure 3 and a mixing tee, both currently manufactured by TharSFC. The valve system, by which the parallel screening is realized, together with  
10 the other hardware and software, also enables a single channel screening mode where any user-defined preferred single channel can be used, without the need to use all other channels at any specified time and or occurrence of particular events.

15 The software used in the methods of the present invention is programmed with the capability of screening multiple co-solvents, gradient conditions, temperature and pressure conditions sequentially with each injection of a sample. Additionally, the software can be used to program either gradient or isocratic screening conditions  
20 simultaneously onto all the channels for optimization of the separation.

The software evaluates each run based on user-specified criteria for separation performance on each channel, and adds additional runs to  
25 the sequence by identifying the channel with the best column performance and then switching the valves to run in single channel mode with optimized conditions based on those criteria. For example, a user may select a desired peak count and separation factors that the software will use to automatically determine which channel performs  
30 the best, and then manipulate the hardware to run repeated



separations on that single channel for optimization. Then, the user would simply load the hardware with samples and start the process.

The software is also designed to acquire all data types for any specific sample simultaneously from a combination of various characteristics determinations, such as, but not limited to, absorptions under any specified ultra-violet wavelength(UV), full spectrum determination by photodiode array detector(PDA), light scattering detector(ELSD) and mass spectrometer (MS). The software provides a single data package output for all of the data types acquired, in the form of a report to the user for decision making. This report includes all necessary information to describe the selected channels and conditions so that the user may quickly determine what channels and conditions were successful, and to what degree, so that candidates may be selected quickly and efficiently for optimization. In a different embodiment, a mass spectrometer (MS) can be placed where all streams join together prior to the back pressure regulator. In this embodiment, when screening is performed on all channels, the MS data (spectra) generated is the combined output of all channels. The software through additional processing attempts to deconvolute the data and associate spectra to each channel based on data generated by the detector on the specific channel.

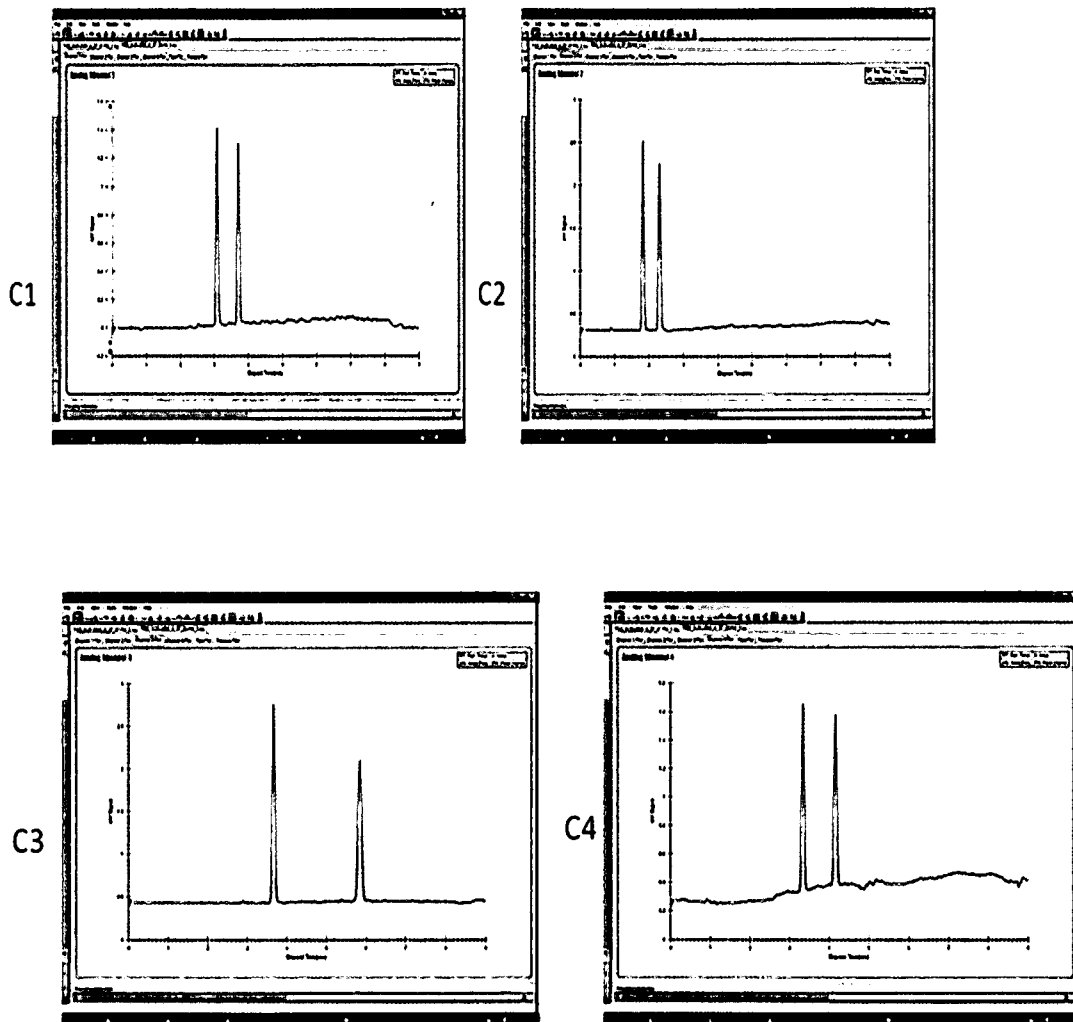
25

### EXAMPLES

Below are examples of the apparatus and method according to the present invention. These examples are intended to illustrate the invention and should not be construed as limiting the invention in any way.

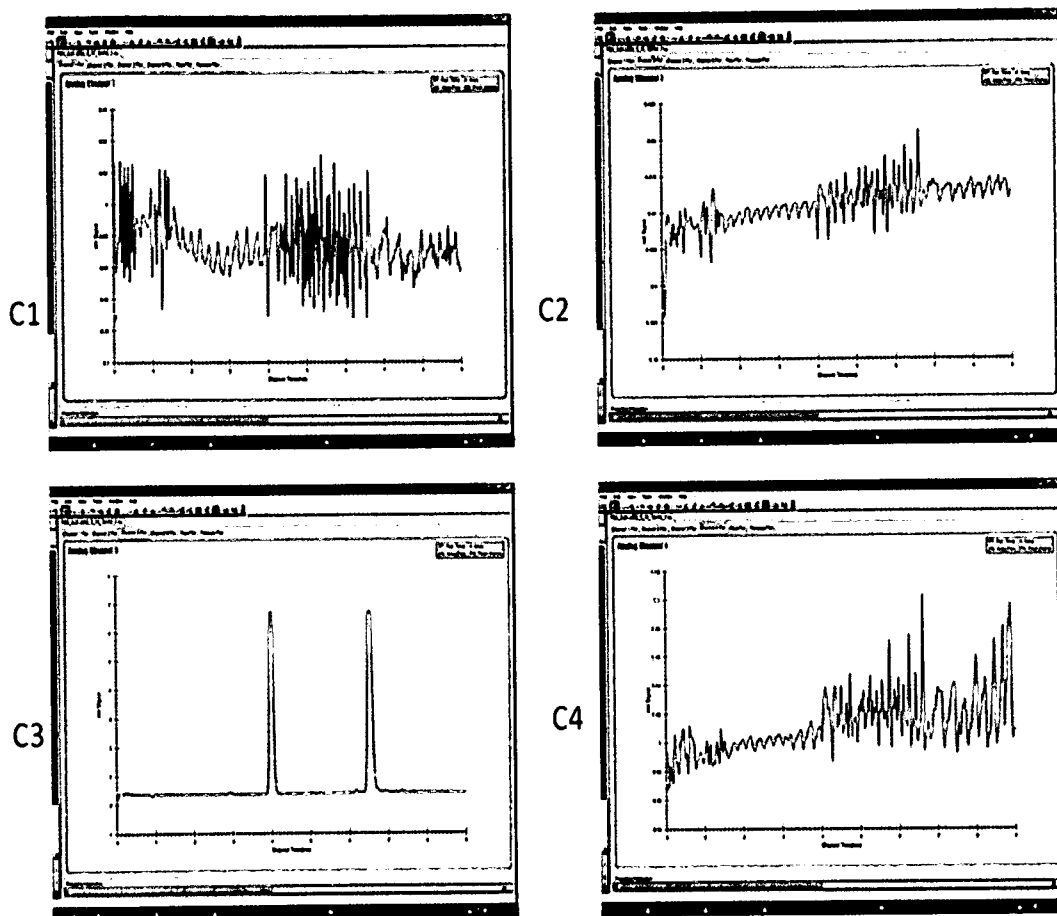
30

In Example 1, chromatograms are shown from one embodiment of the present invention where multiple detectors are placed on all channels for simultaneously parallel screening. In this example, four UV detectors were used to analyze a single injection of trans-stilbene oxide (TSO) in gradient flow mode. C1-C4 are columns on each channel.



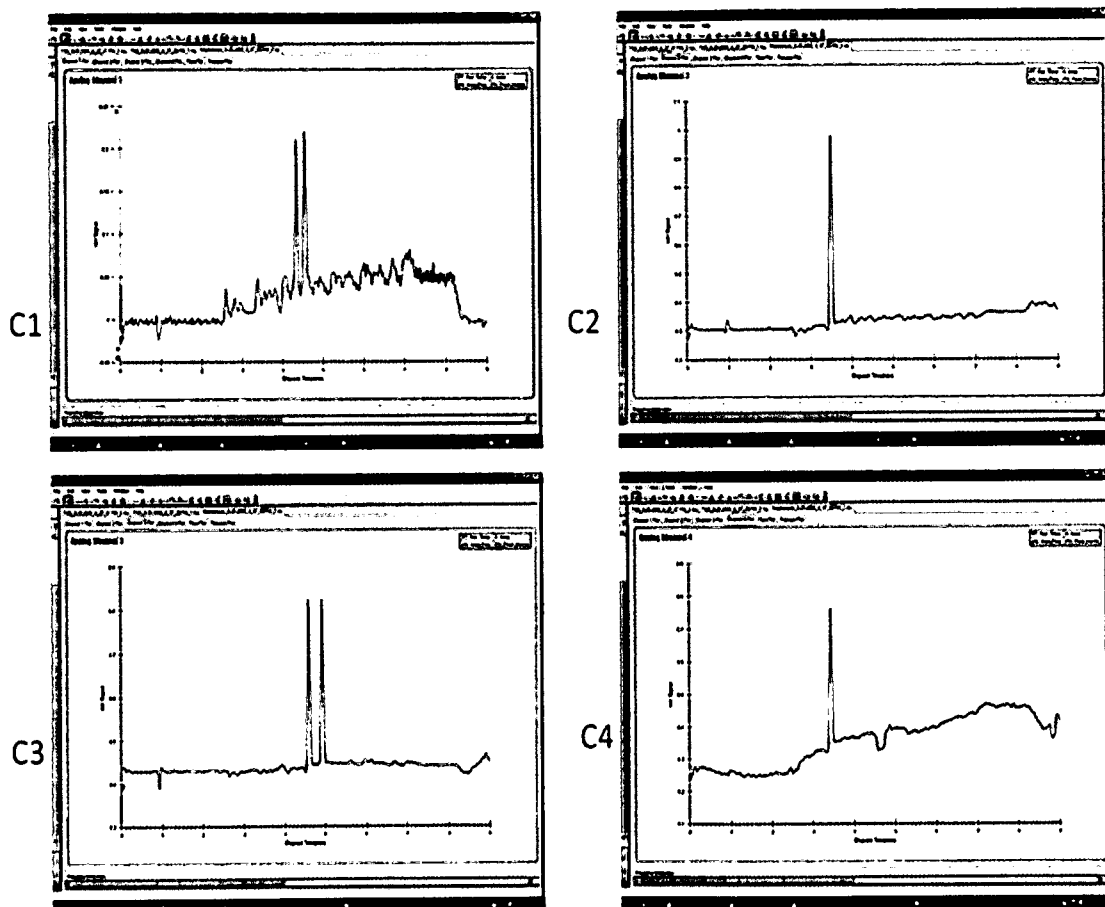
Figures 1a - 1d

Example 2 shows chromatograms from one embodiment of the present invention where a single channel is employed using general gradient mode on channel 3. The compound is TSO.



Figures 2a – 2d

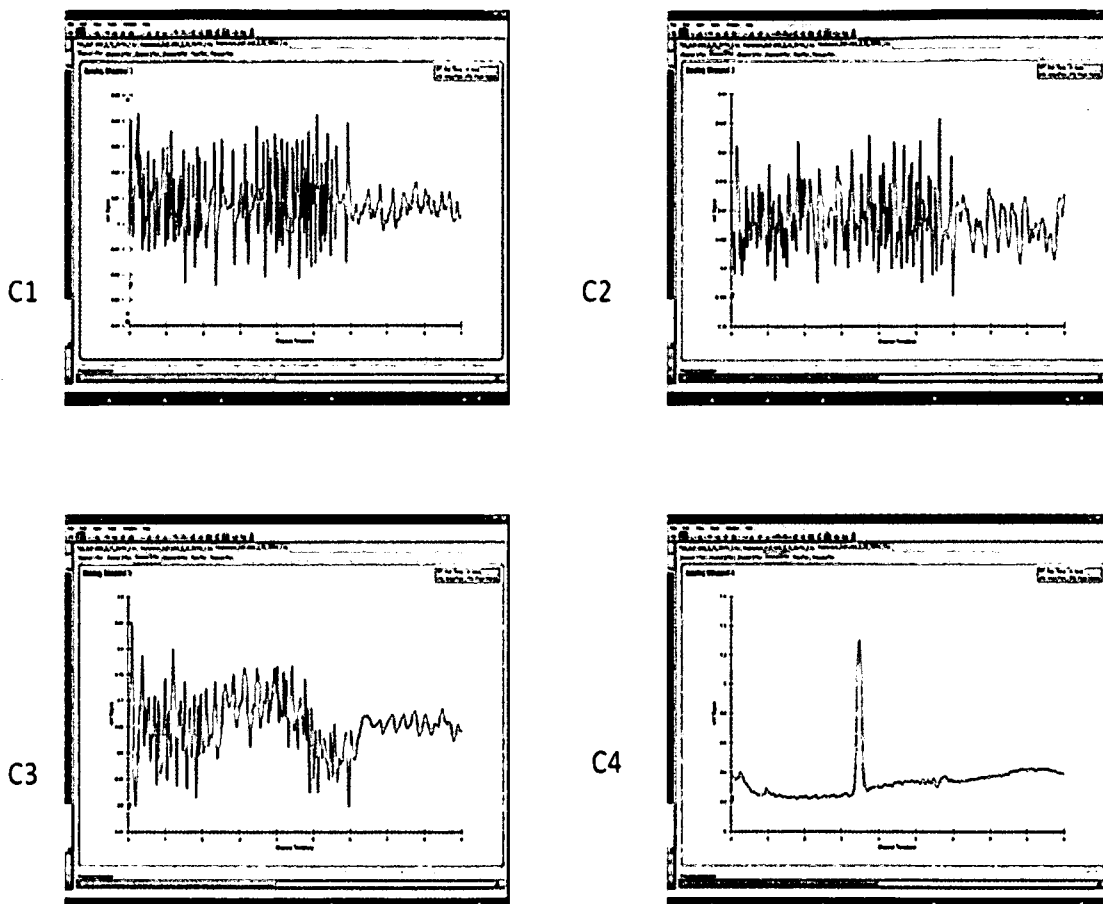
- In Example 3 below are chromatograms from one embodiment of the present invention where multiple detectors can be placed on all channels for parallel screening of a second sample. In this example, four UV detectors were used to analyze a single injection of
- 5 Mephenesin in gradient flow mode. C1-C4 are columns on each channel.



Figures 3a – 3d

In Example 4 below are chromatograms from one embodiment of the present invention where a single channel is employed using general gradient mode on channel 4. The compound is Mephenesin is C1-C4 are columns on each channel.

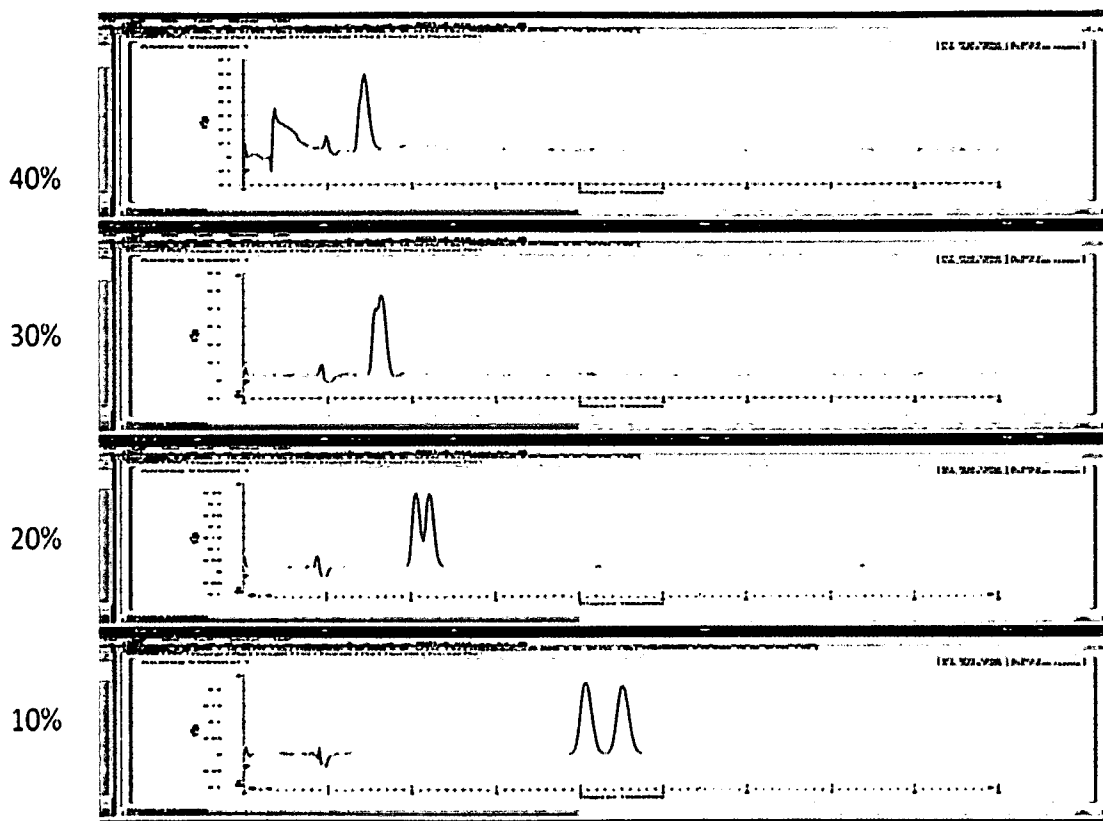
5



10

Figures 4a – 4d

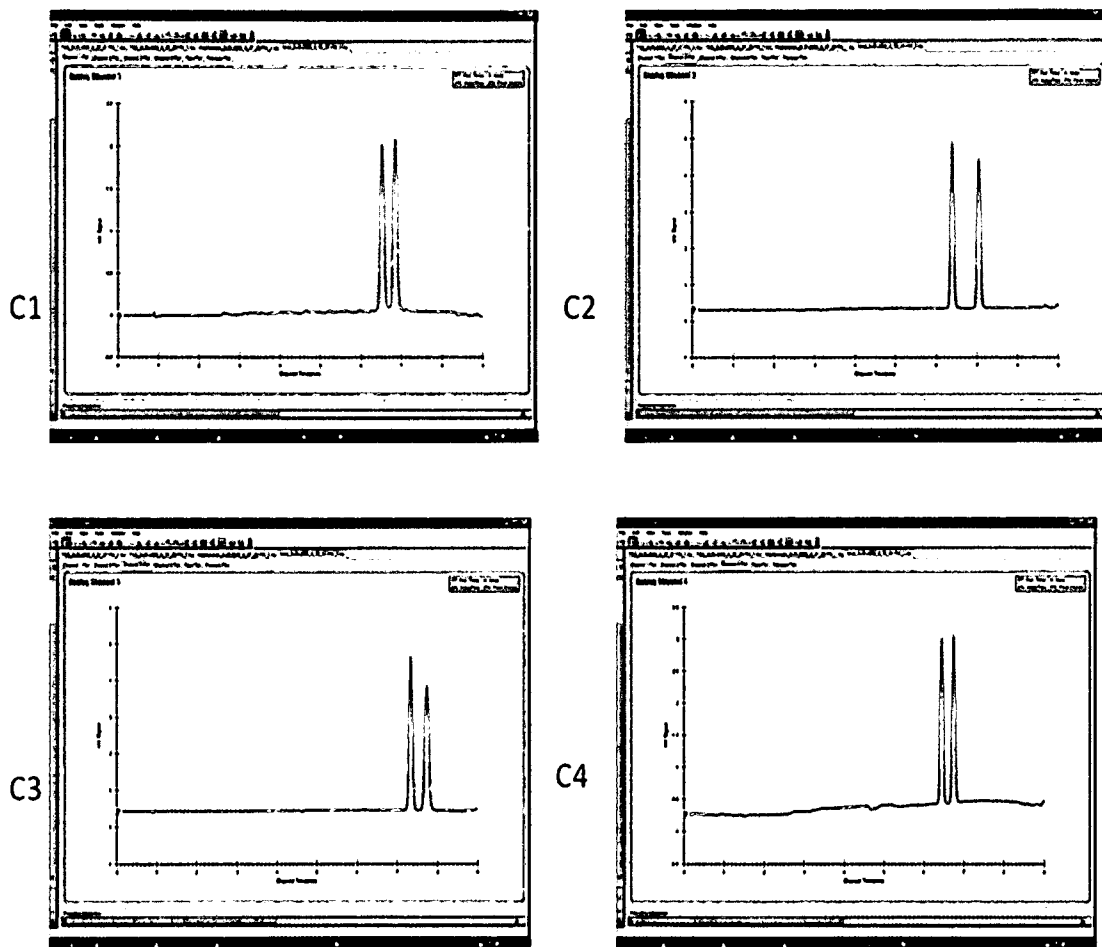
In one embodiment the second step of method development is to run a focused gradient. Below in Example 5 are chromatograms from running a focused gradient on single channel with sample Mephenesin. The results are evaluated and isocratic sequential  
5 injections are made on the single channel with column C1 to obtain an optimized method.



Figures 5a – 5d

Below in Example 6 are chromatograms from a third example of method development on the compound binol. All four channels are used to quickly develop an optimized method for screening binol. C1-C4 are columns on each channel.

5

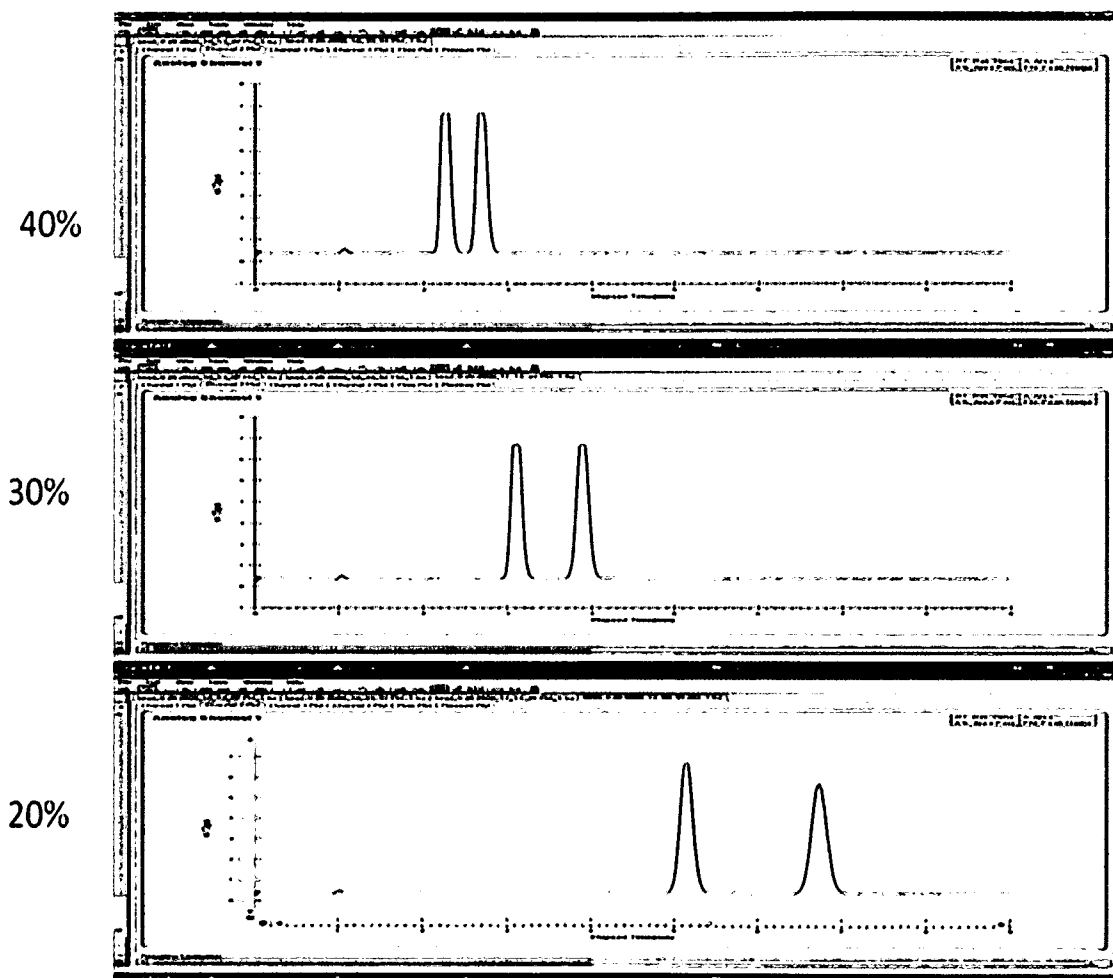


10

Figures 6a – 6d

Continued below are chromatograms for the second step from the same example (binol) method development. Results were obtained by running a focused gradient on binol. The results are evaluated and isocratic sequential injections are made on the single channel with column C2 to obtain an optimized method.

5



Figures 6e - 6g



From the foregoing it will be appreciated that, although specific embodiments of the invention have been described herein for purposes of illustration, various modifications may be made without  
5 deviating from the spirit, and scope of the invention. Accordingly, the invention is not limited except as by the appended claims.

What is Claimed Is:

1. An apparatus for supercritical fluid chromatography, the apparatus comprising:

a binary pump;

an autosampler;

a sampling valve;

a first and second port switching valve;

a first and second manifold;

two or more channels, each having a check valve assembly, a separation column and one or more detectors operatively connected thereon;

computer software and hardware to

1) control distribution of fluid through the apparatus, including switching between a multi-channel mode or a single channel mode;

2) analyze data collected by the one or more detectors;

and 3) optimize separation of analytes by controlling solvent combinations, concentration gradients, pressure and temperature;

and

a backpressure regulator,

wherein the apparatus excludes additional backpressure regulators or pumps on individual channels.

2. The apparatus of claim 1, wherein the apparatus switches between multi-channel mode and single channel mode using only software, without any physical configuration change by a user.

3. The apparatus of claim 1, further including a software capability of optimizing separations based on obtained results.
4. The apparatus of claim 1, wherein the number of channels is greater than 8.
5. The apparatus of claim 1, wherein the number of channels is greater than 24.
6. The apparatus of claim 1, wherein the software is programmed to provide simultaneous parallel screening of a single sample through the two or more channels.
7. The apparatus of claim 1, wherein the software is programmed to provide optional sequential screening through one or more channels.
8. A method of screening a sample using supercritical fluid chromatography, the method comprising the steps of
  - mixing a sample containing analytes of interest with a supercritical fluid and optionally, a suitable solvent, to create a mixed mobile phase;
  - moving the mixed mobile phase along a flow path to an autosampler;
  - injecting a single portion of the mixed mobile phase into the autosampler;
  - moving the single portion along the flow path through a first column switch valve to a first manifold;
  - dividing the single portion into two or more subportions and directing each of the two or more subportions to a separate channel for separation;

simultaneously moving each of the two or more subportions along each separate channel through a separation column and through one or more detectors;

moving the two or more subportions through a second check valve and a second manifold to create a single exit stream; and

moving the exit stream along the flow path to the backpressure regulator.

9. The method of claim 8, further comprising providing additional analytical processing of the exit stream prior to exit at the backpressure regulator.

10. The method of claim 8, wherein the one or more detectors is selected from the group consisting of ultra-violet wavelength (UV), photodiode array detector (PDA), light scattering detector (ELSD) and mass spectrometer (MS). Flame ionization detector, chemiluminescence nitrogen detector, corona aerosol detector, circular dichroism and other chiral detectors, and on-line infrared and nuclear magnetic resonance detectors.

11. The method of claim 8, wherein at least one channel has two or more detectors thereon.

12. The method of claim 8, wherein multiple co-solvents, gradient conditions, temperature and pressure conditions are sequentially screened with each injection.

13. The method of claim 8, wherein either gradient or isocratic screening conditions are simultaneously set on any of the two or more channels for optimization.

14. The method of claim 8, wherein software evaluates each run based on user-specified criteria for separation performance on each channel and adds additional runs to the sequence by identifying the channel with the best column performance and then switching the valves to run in single channel mode with optimized conditions based on those criteria.

15. The method of claim 8, wherein the software acquires all data types for any specific sample simultaneously.

16. The method of claim 15, wherein the data type is data generated by a detector selected from the group consisting of an ultra-violet wavelength (UV) detector, full spectrum determination by photodiode array detector(PDA), light scattering detector(ELSD), mass spectrometer (MS), flame ionization detector, chemiluminescence nitrogen detector, corona aerosol detector, circular dichroism and other chiral detectors, on-line infrared and nuclear magnetic resonance detectors, and combinations of any of these.

17. The method of claim 15, wherein all data types acquired for any specific sample are included in a single data report to a user for decision making.

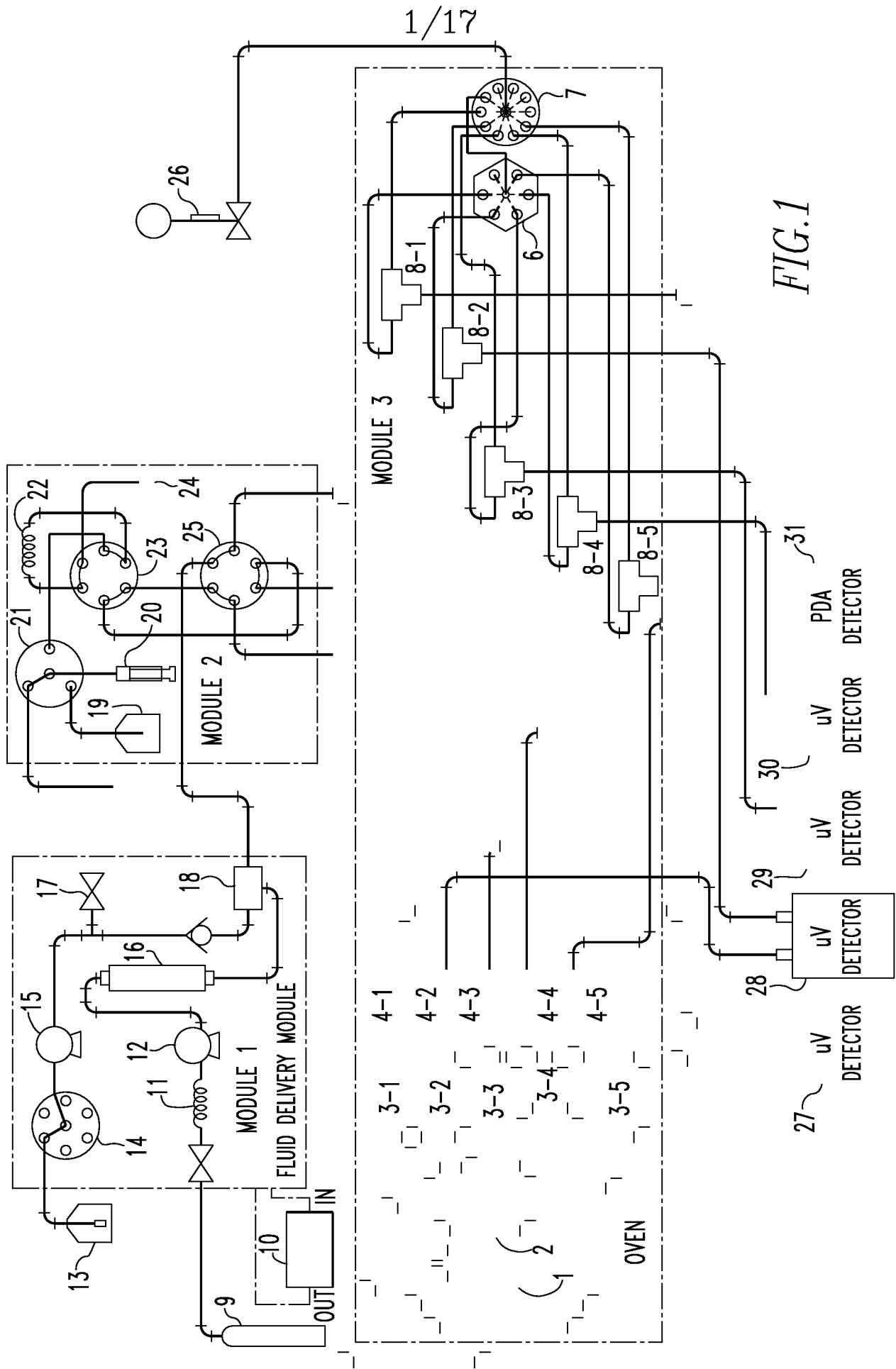


FIG. 1

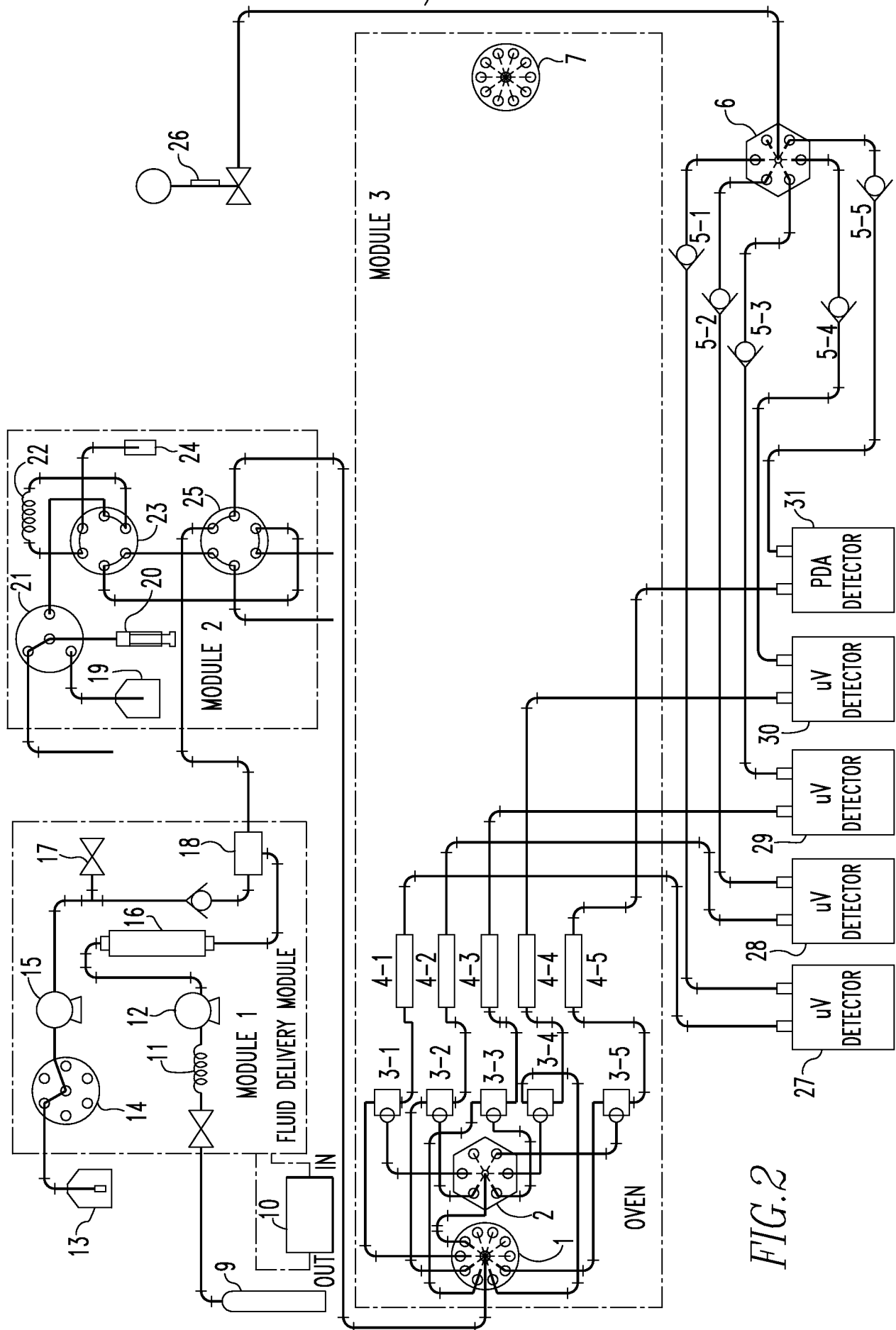
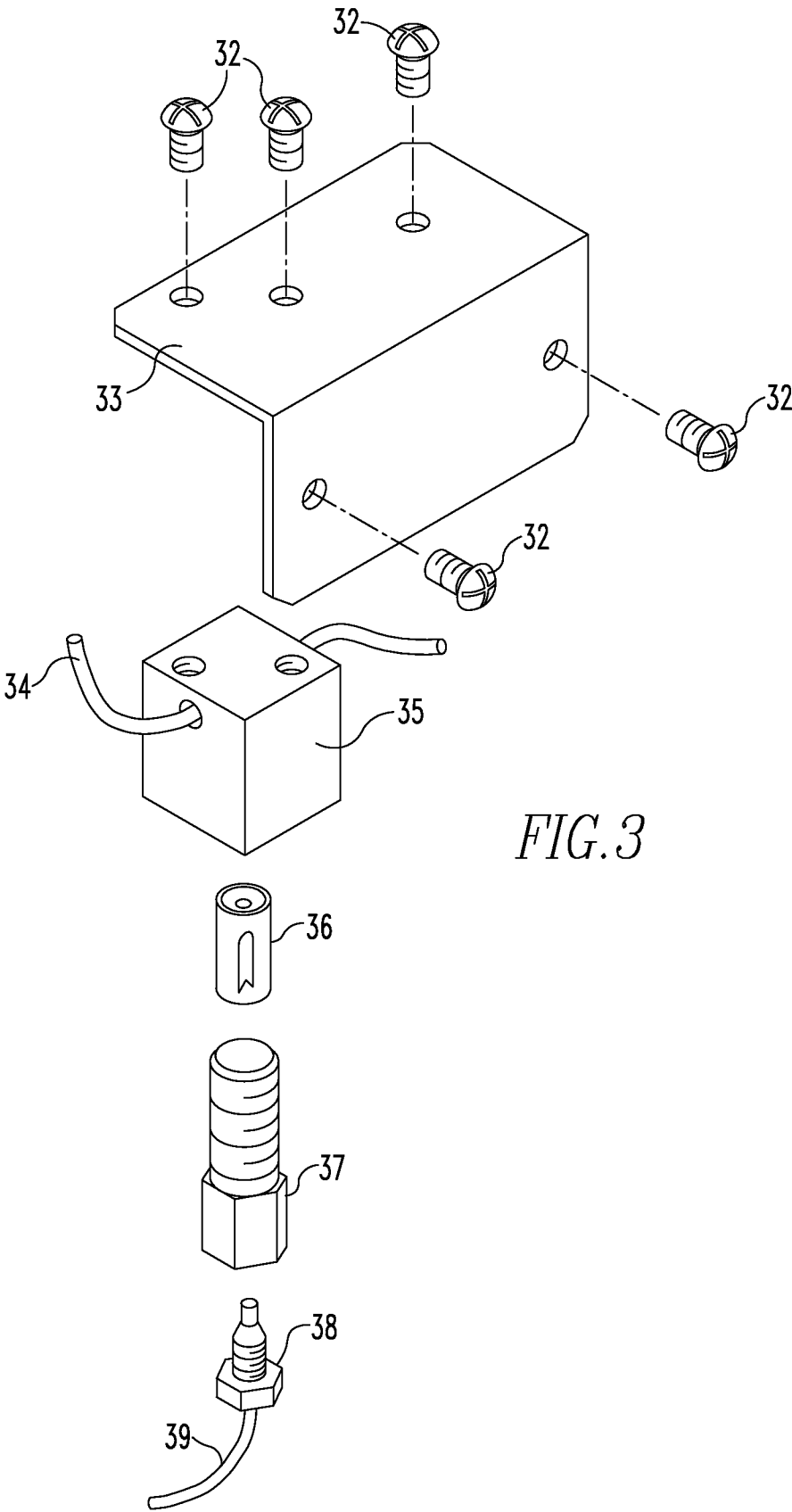


FIG. 2

3/17





4/17

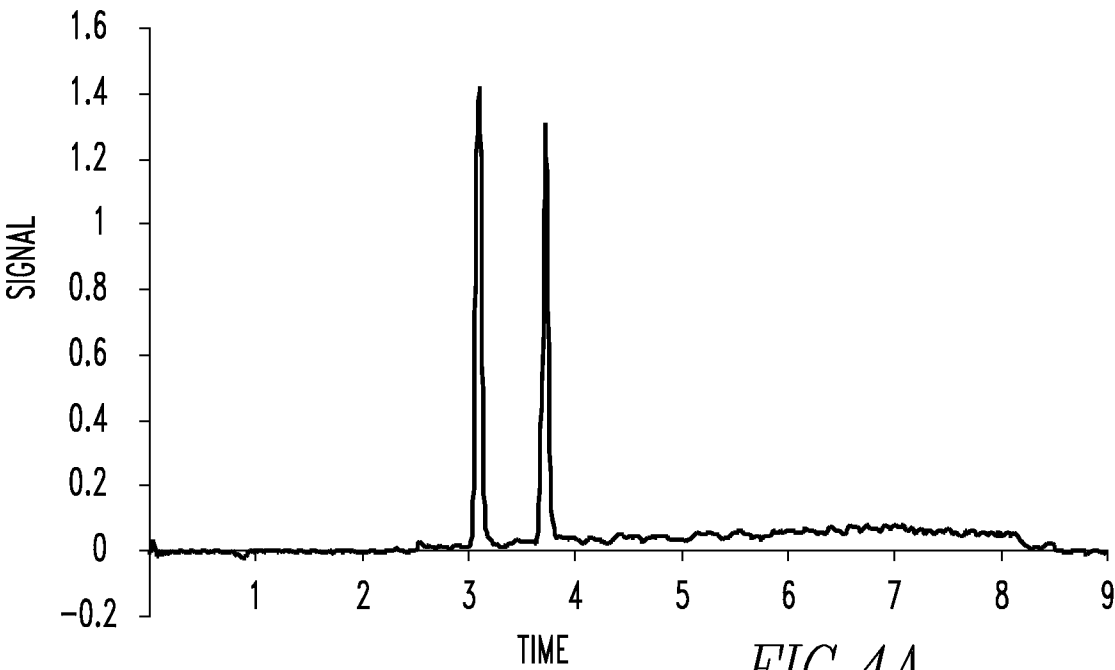


FIG. 4A

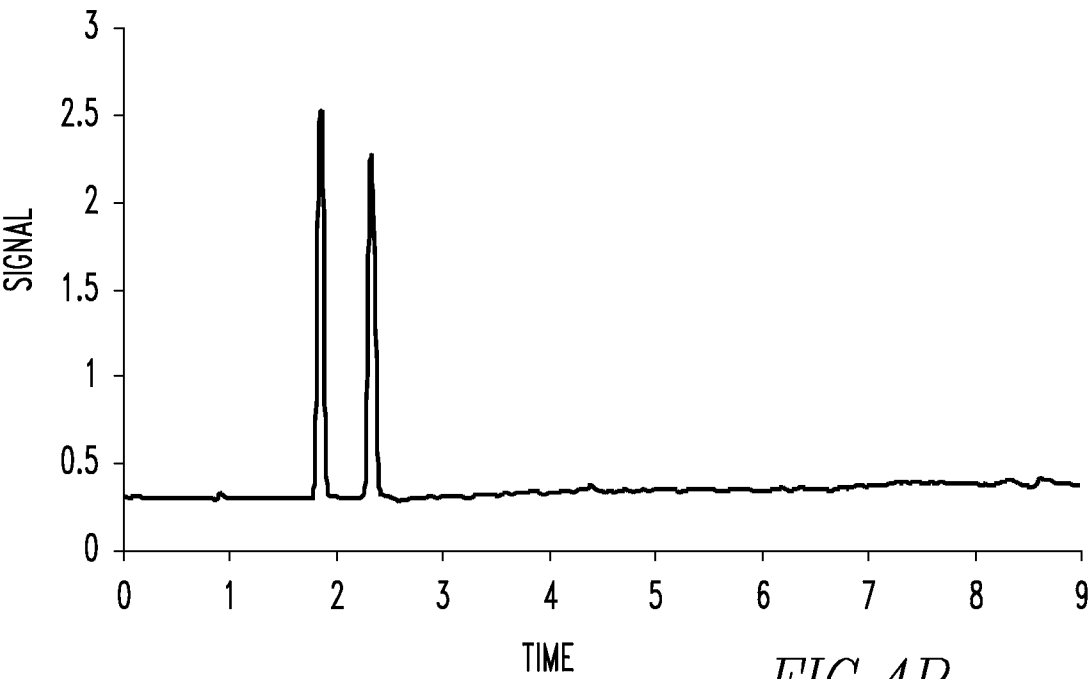


FIG. 4B

5/17

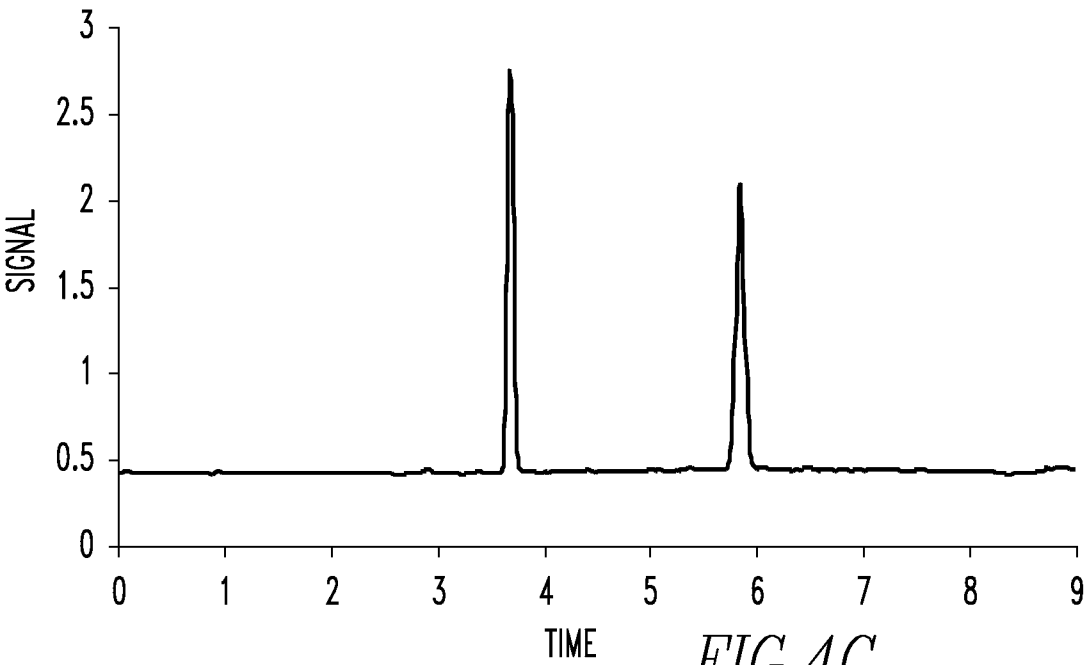


FIG. 4C

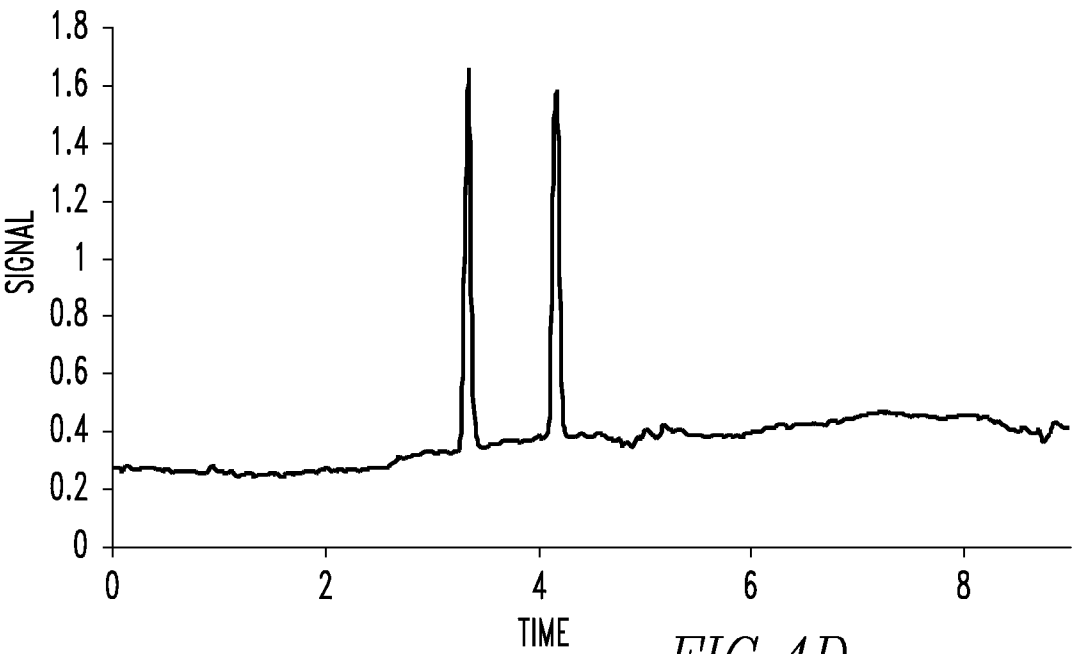


FIG. 4D

6/17

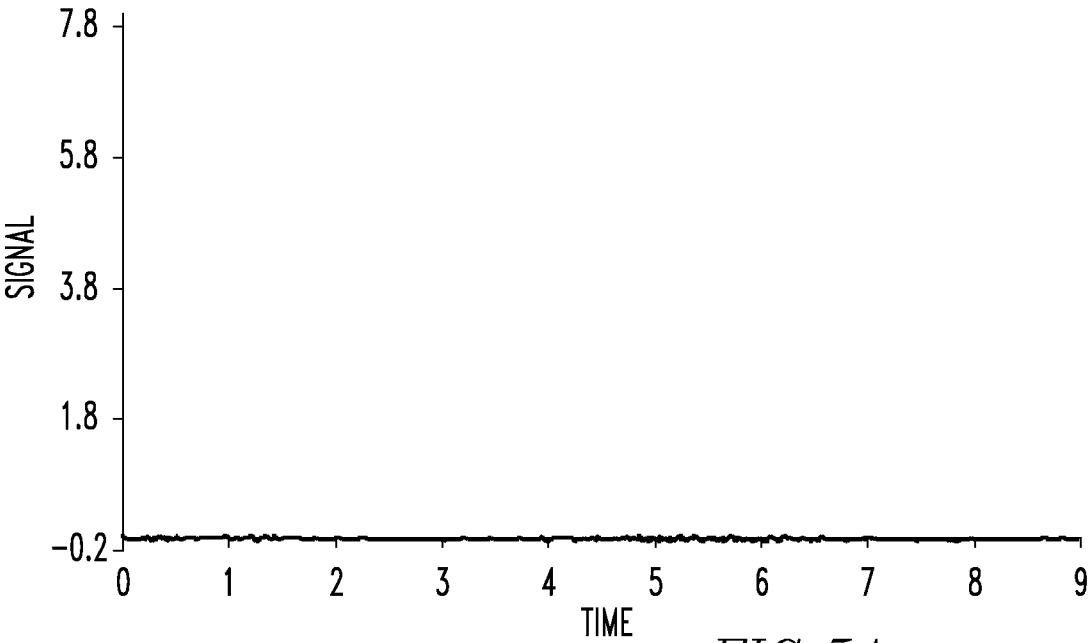


FIG. 5A

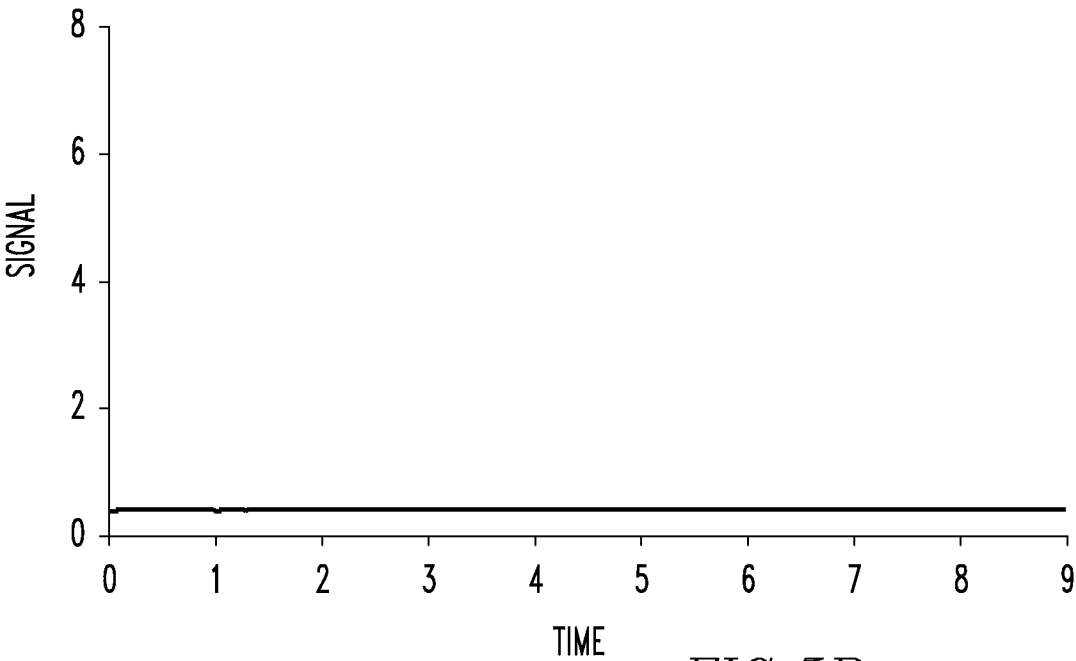


FIG. 5B

7/17

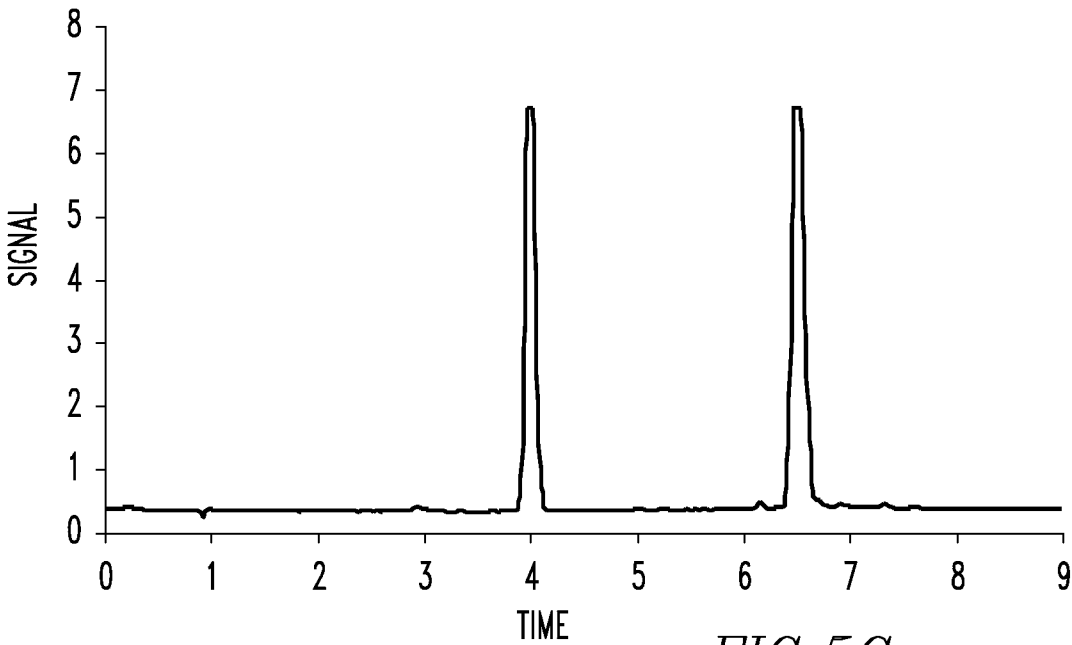


FIG.5C

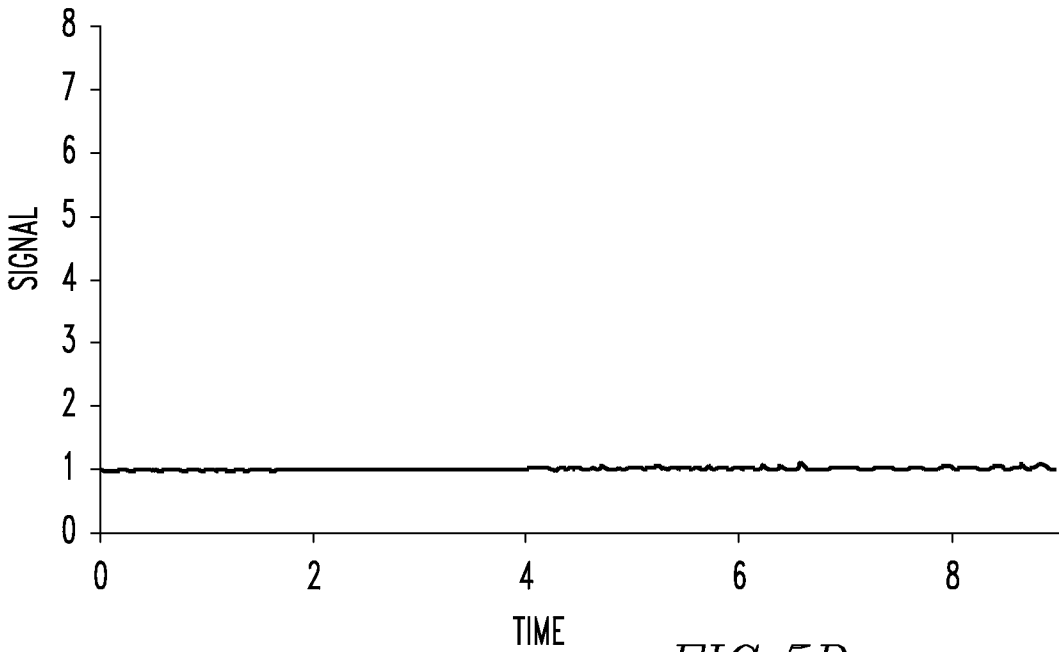
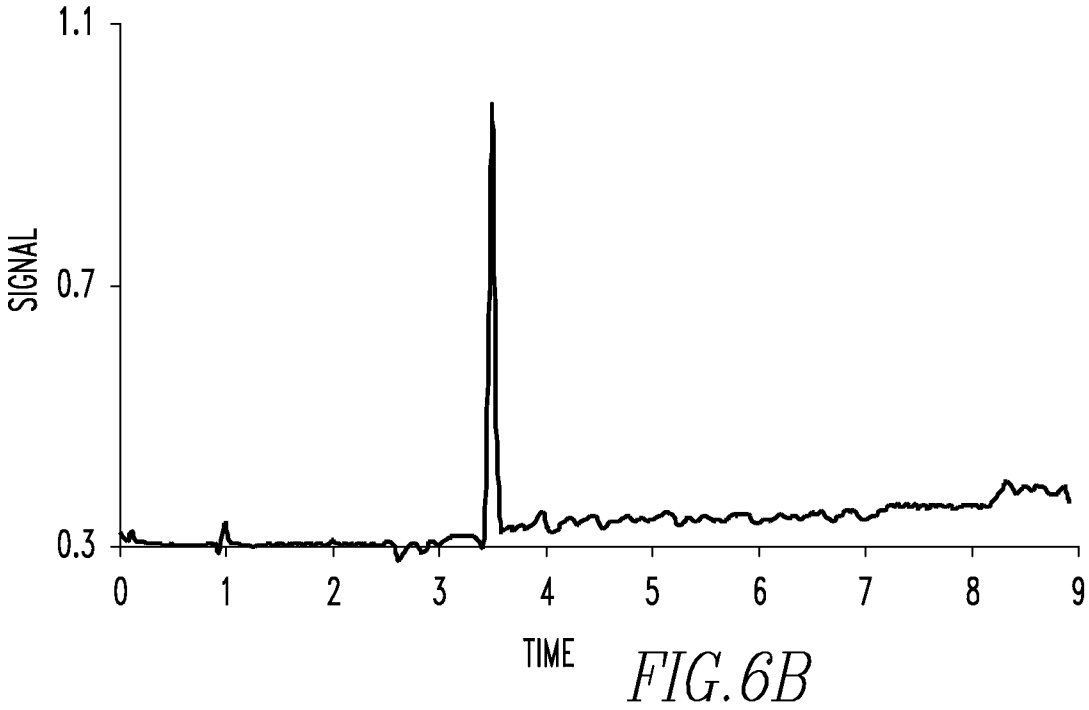
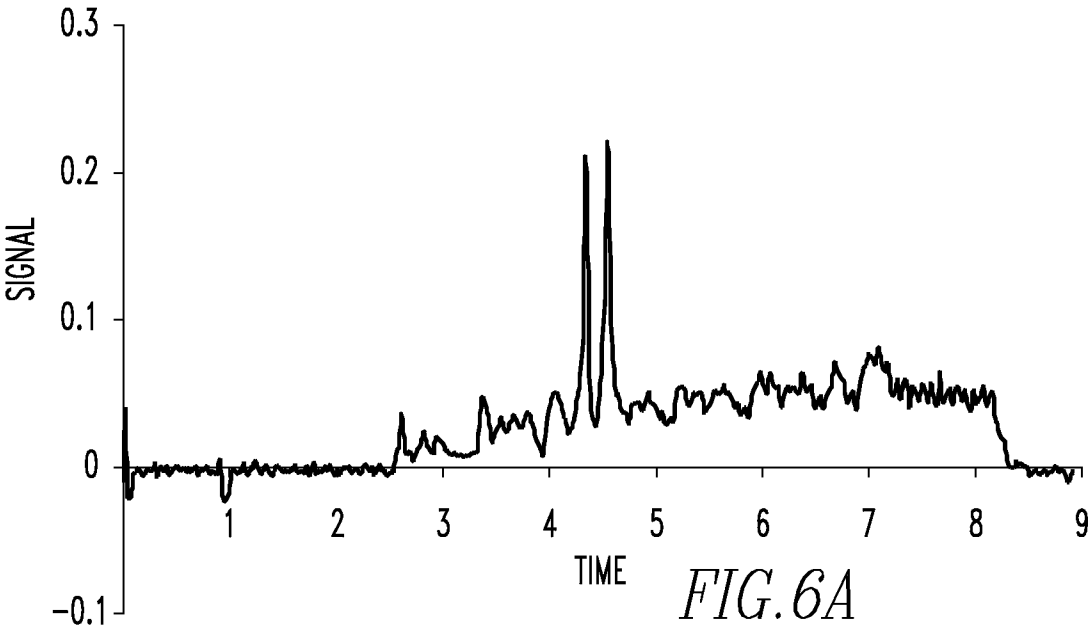


FIG.5D

8/17



9/17

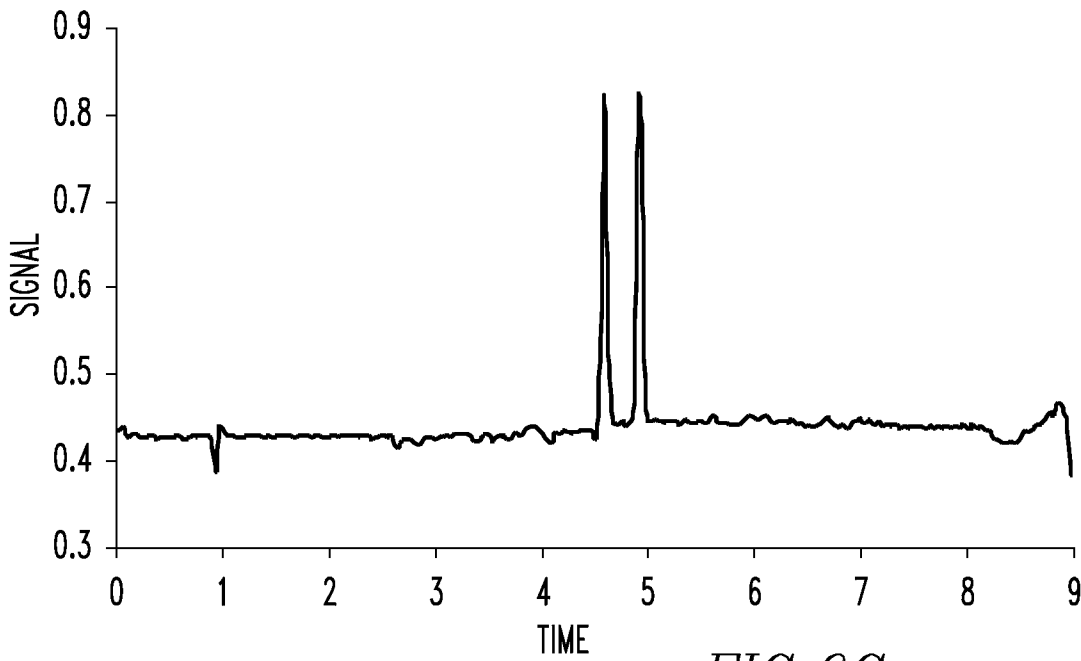


FIG. 6C

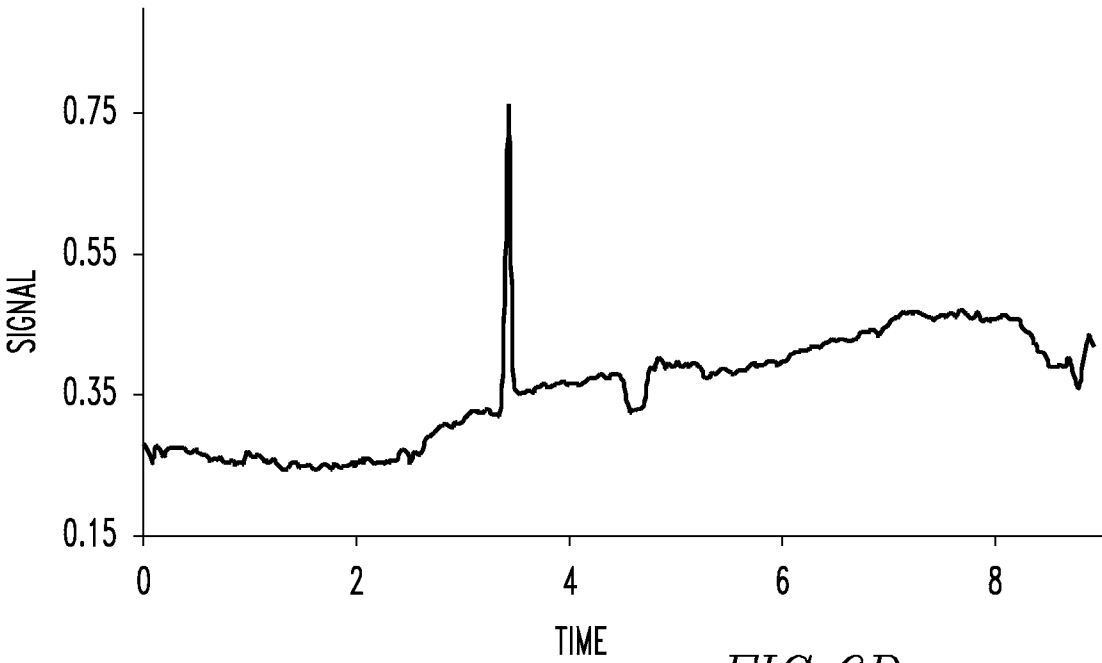
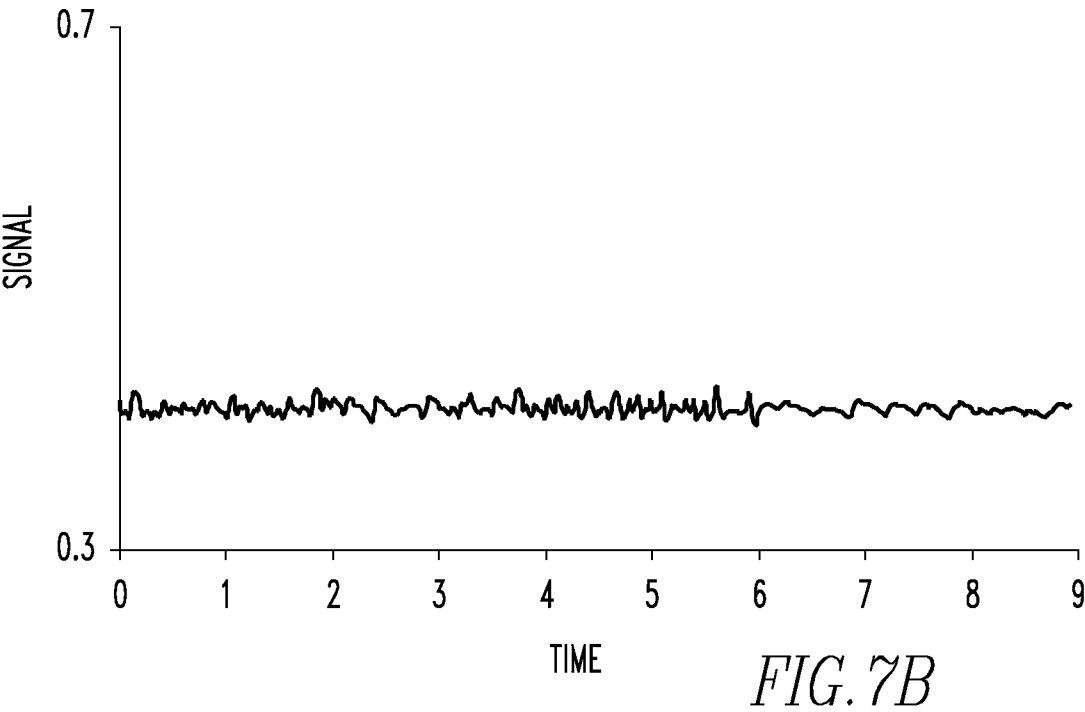
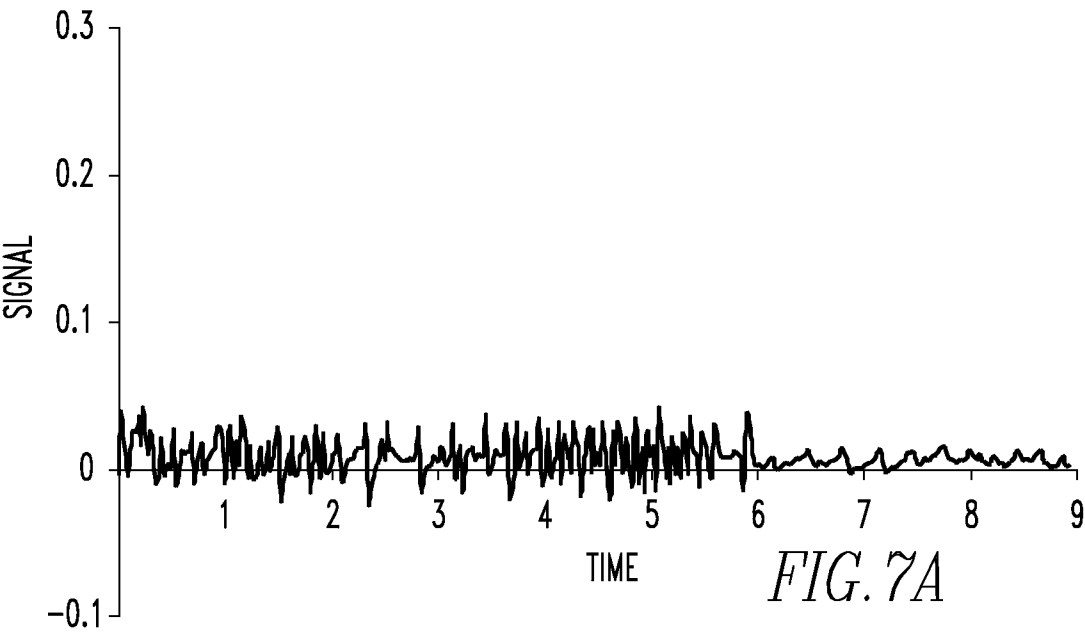


FIG. 6D

10/17



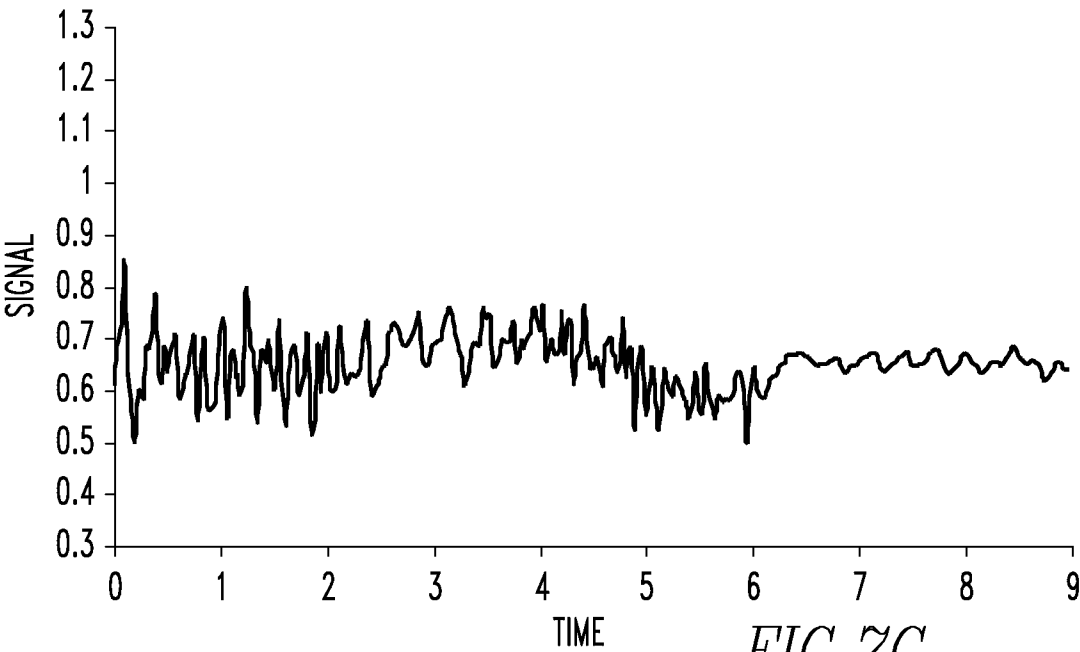


FIG. 7C

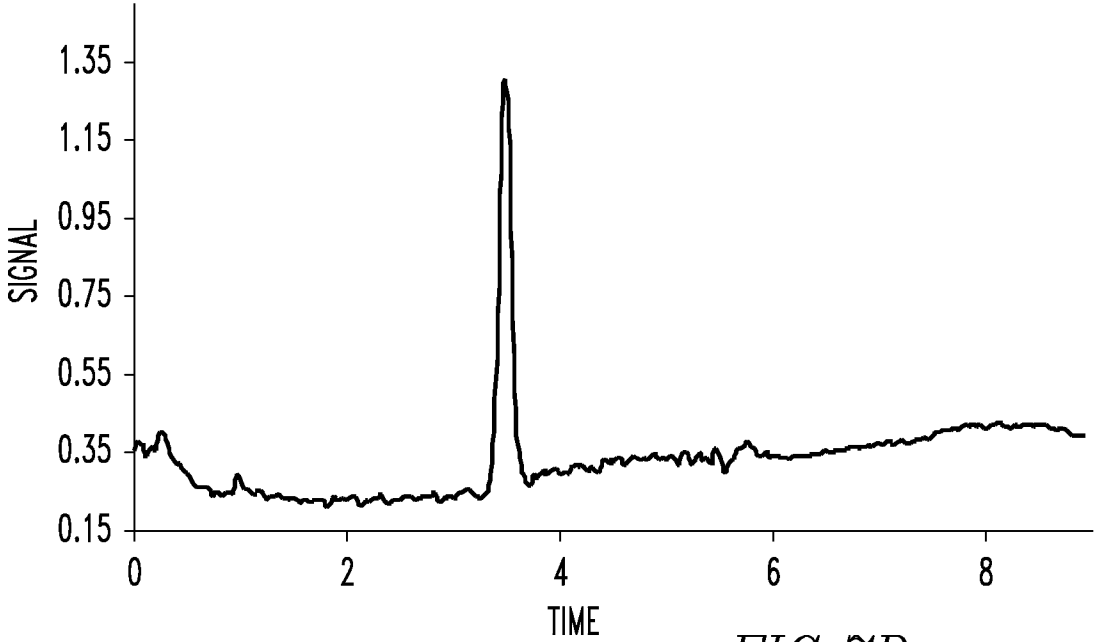
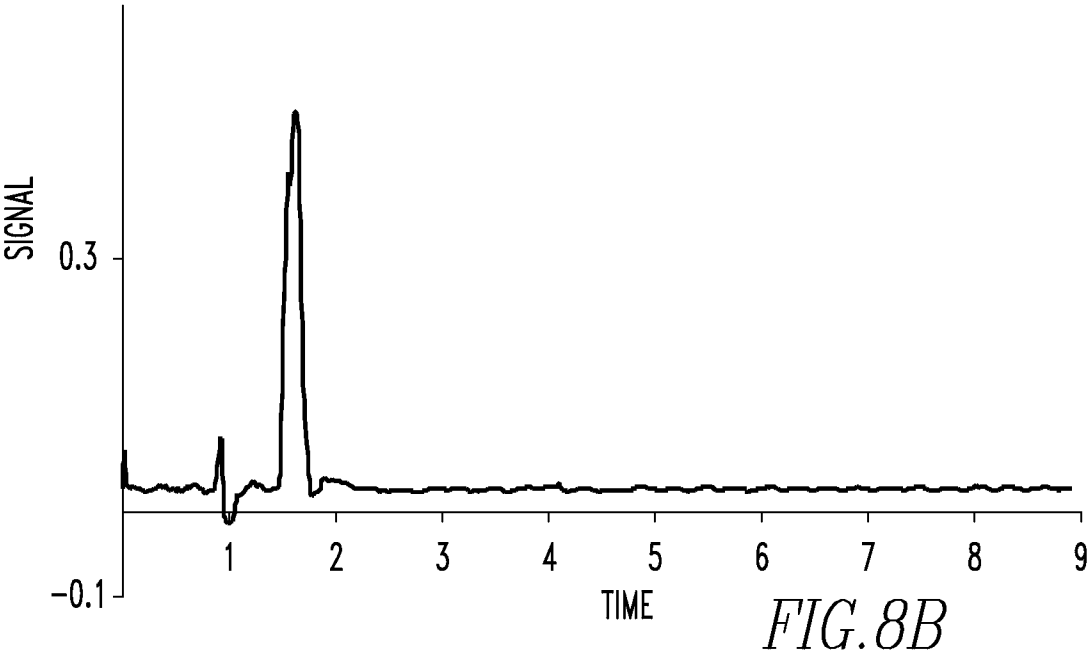
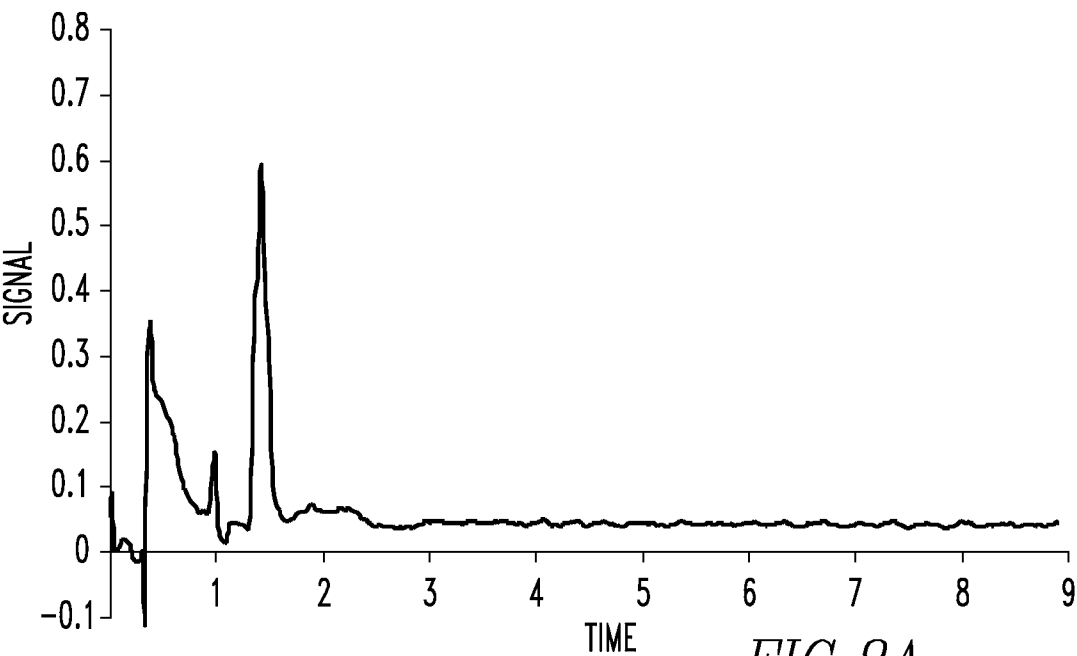
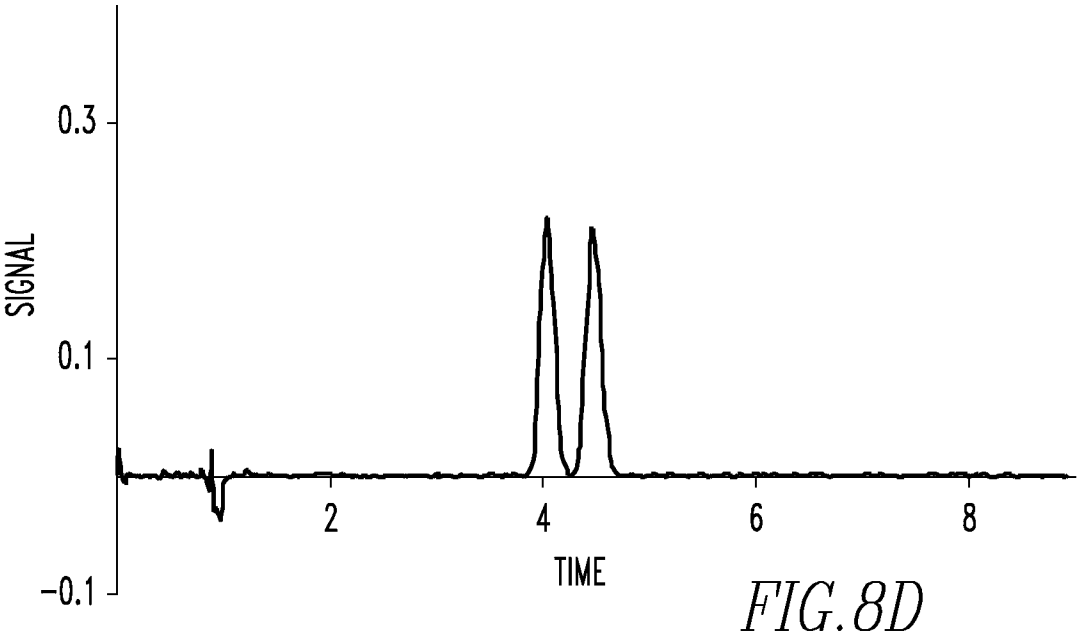
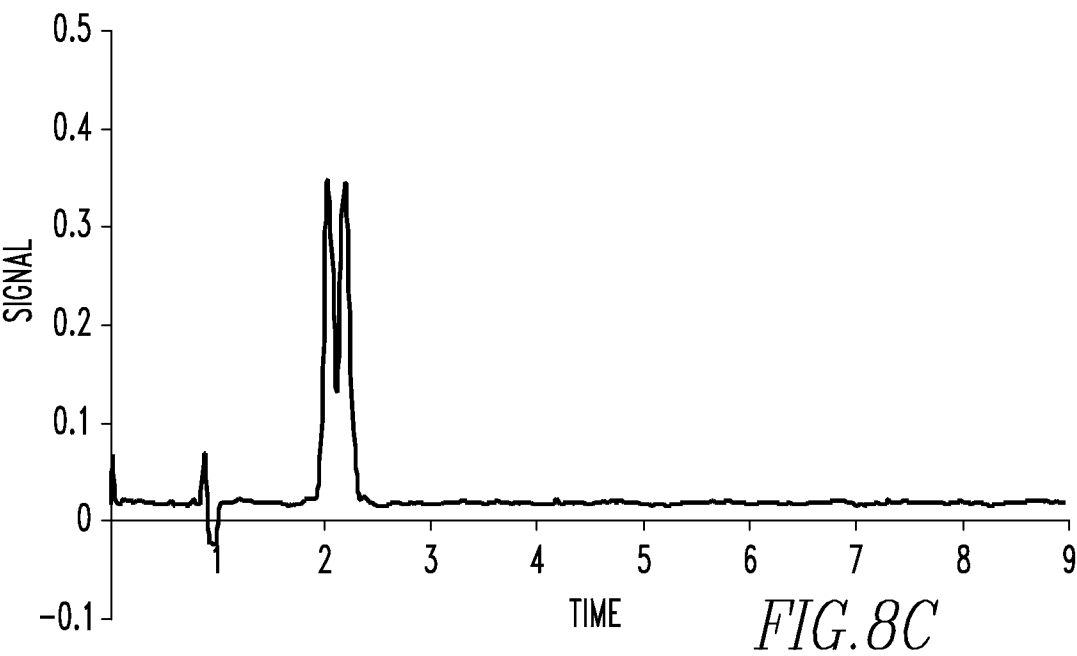


FIG. 7D

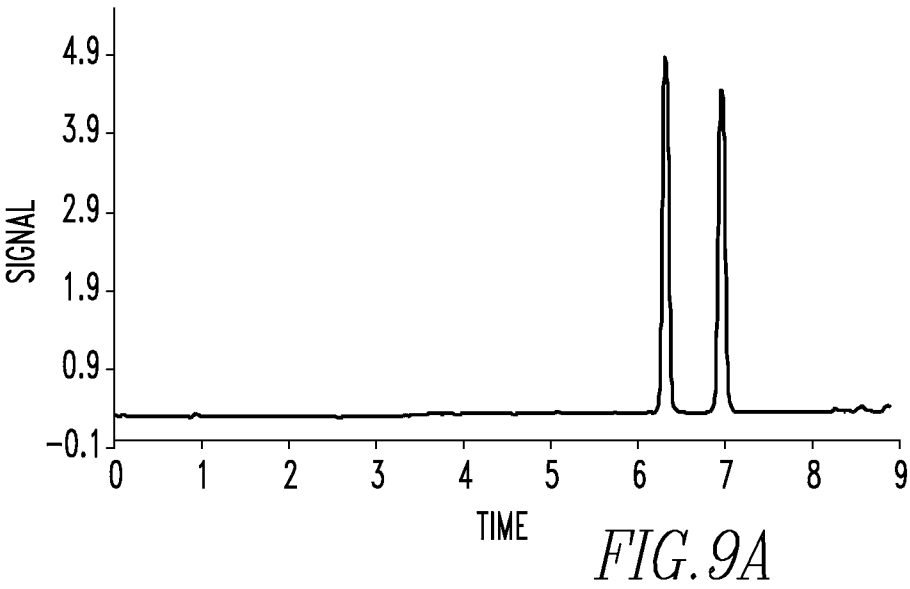
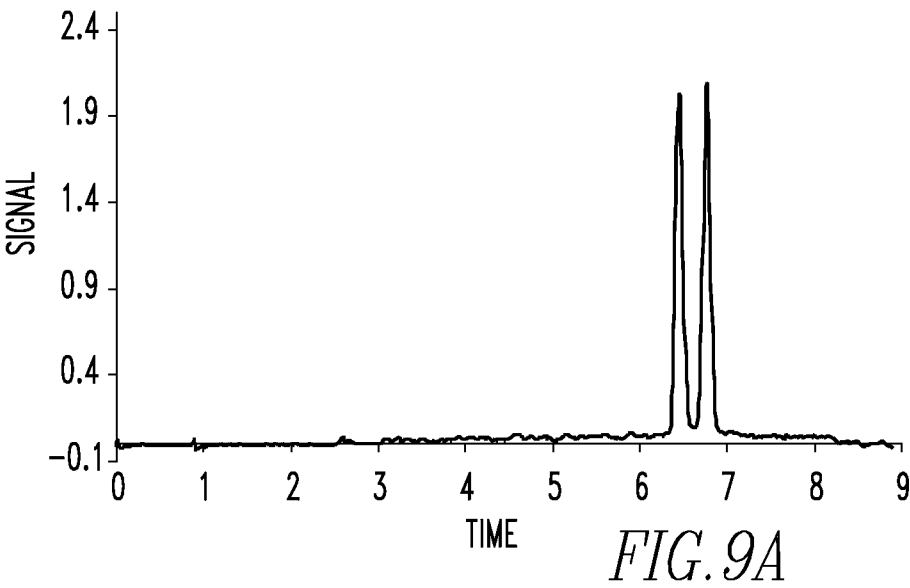


12/17





14/17



15/17

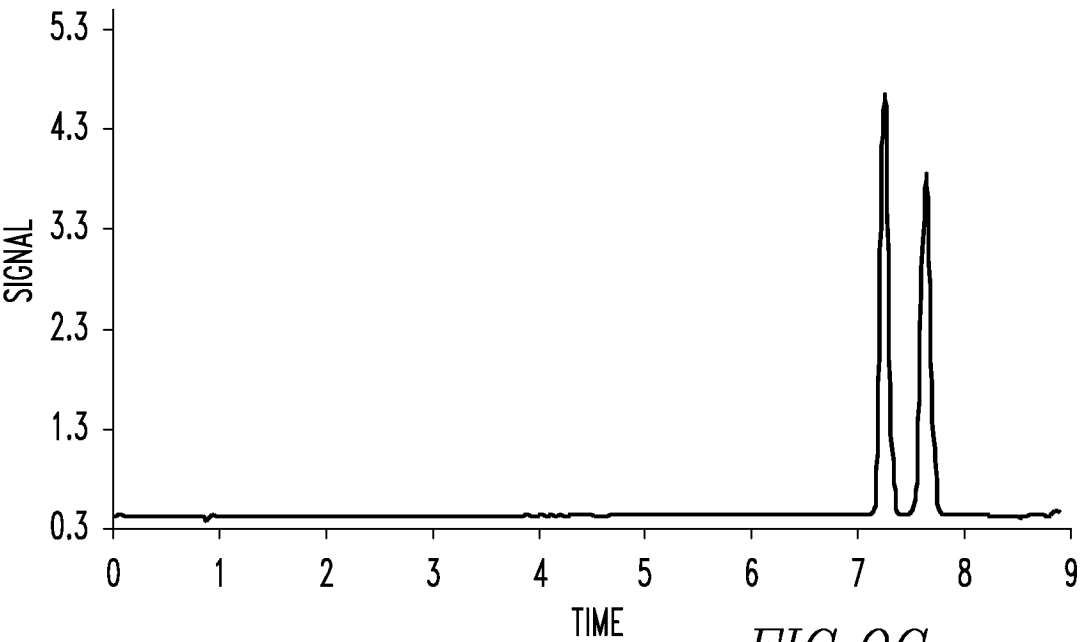


FIG. 9C

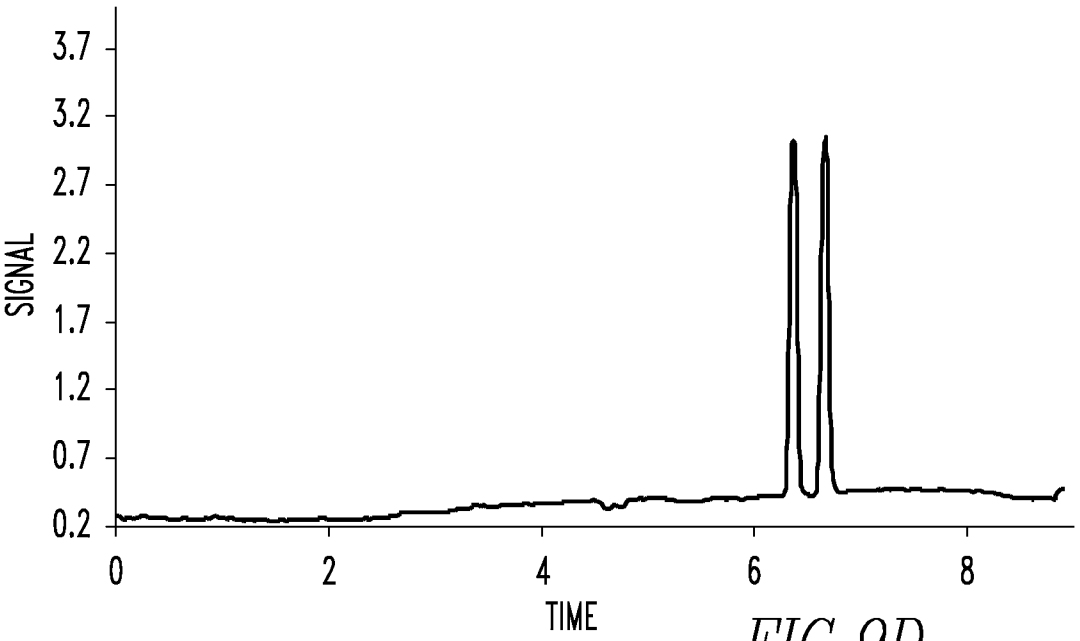


FIG. 9D

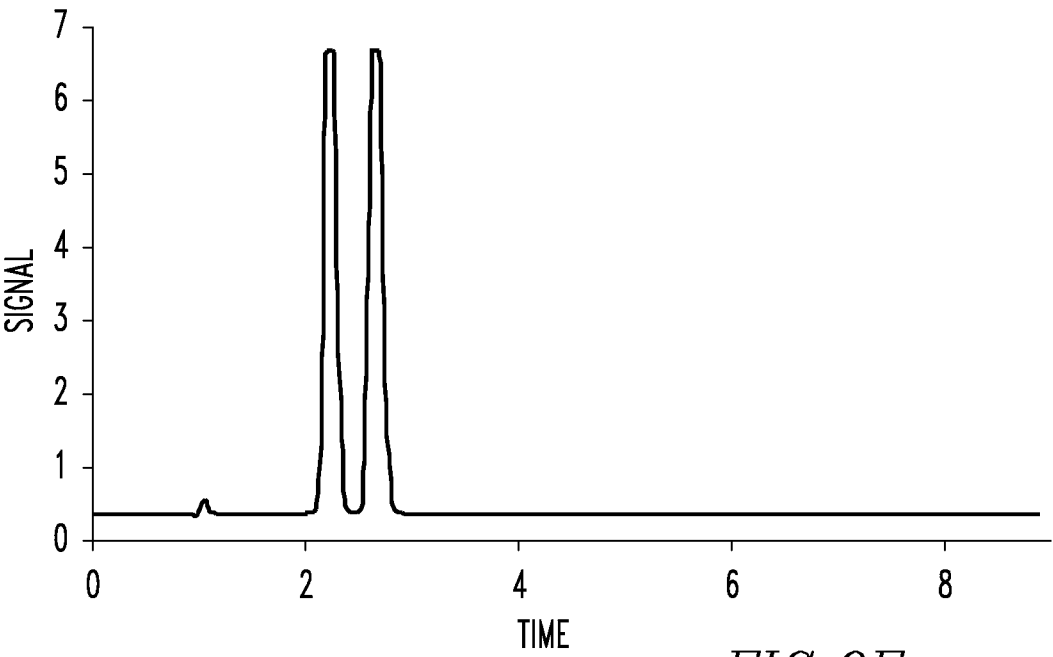


FIG. 9E

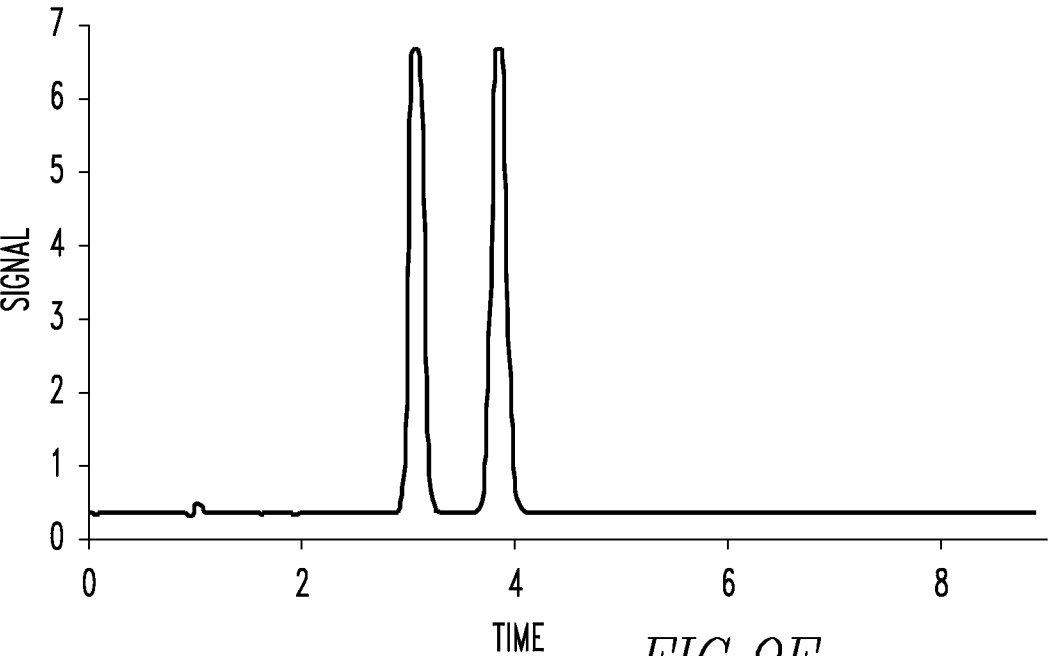


FIG. 9F

