United States Patent [19]

Annino et al.

[11] **3,717,028**

[45] **Feb. 20, 1973**

[54] CHROMATOGRAPHIC PEAK SELECTOR

[75] Inventors: **Raymond Annino**, Plainville; **William E. Jordan**, Foxboro; **William E. Earle**, North Easton, all of Mass.

[73] Assignee: **The Foxboro Company**, Foxboro, Mass.

[22] Filed: **June 2, 1969**

[21] Appl. No.: **829,682**

[52] **U.S. Cl.** **73/23.1**

[51] **Int. Cl.** **G01n 31/08**

[58] **Field of Search** 73/23.1, 23

[56] **References Cited**

UNITED STATES PATENTS

3,559,455	2/1971	Karasek	73/23.1
3,126,731	3/1964	Armstrong	73/23.1
3,177,138	4/1965	Larrison	73/23.1 X
3,301,482	1/1967	Bullen	236/6 X
3,318,149	11/1970	Varadi	73/23.1

OTHER PUBLICATIONS

Schupp "Gas Chromatography" pp. 313–316 Library No. QD 251 W44 Vol. 13 1968.

Hamilton Paper pp. 10–11 "Application of Fisher Model 25...to Respiratory... Determinations" Aug. 1961.

Keuleman "Gas Chromatography" pp. 32–34 1957 Library No. QD 271 K53.

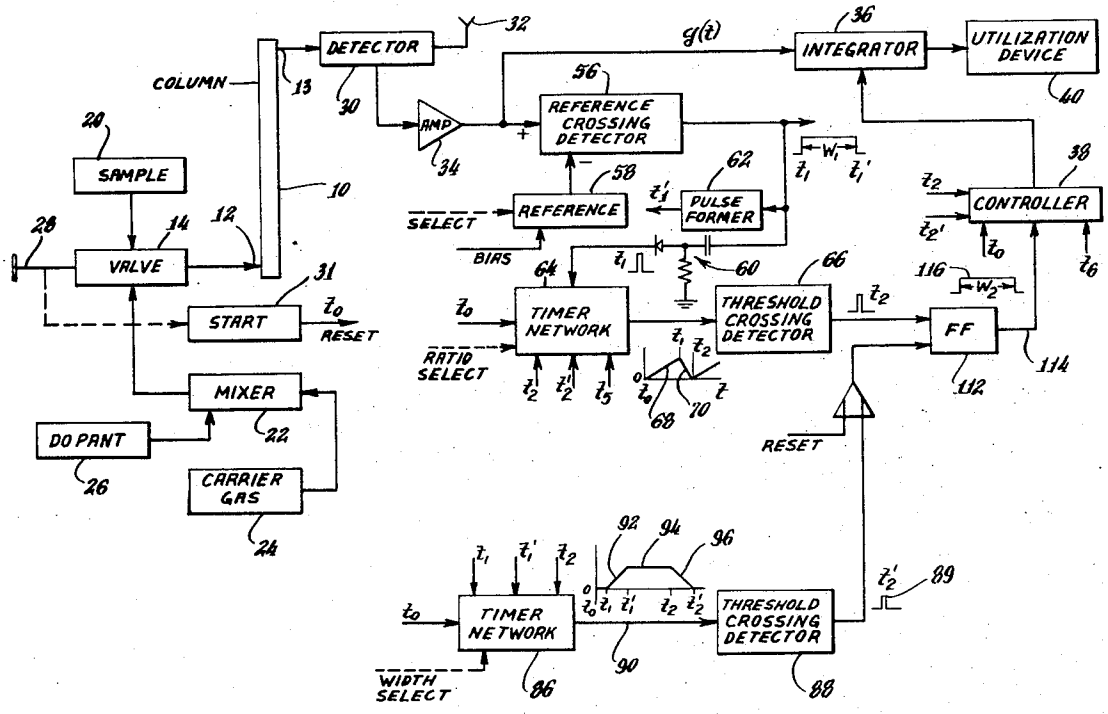
Tandy et al. "Analysis of Gas–Liquid Chromatograms" by a Punched Card Technique", 1961, from Analytical Chem. Vol. 33 No. 6 pp. 665–669.

Primary Examiner—Richard C. Queisser
Assistant Examiner—C. E. Snee, III
Attorney—Bryan, Parmelee, Johnson and Bollinger

[57] **ABSTRACT**

A chromatographic apparatus and technique are described wherein a peak of interest in the output chromatogram is selected for integration by a timing circuit controlled by a preceding reference peak. The timing function starts when the sample mixture is injected into the column, and the time of appearance of the reference peak is used as the basis for computing a predicted time for appearance of the peak of interest. A similar technique is used to compute a predicted width of the peak of interest in order to control the period of integration.

33 Claims, 15 Drawing Figures



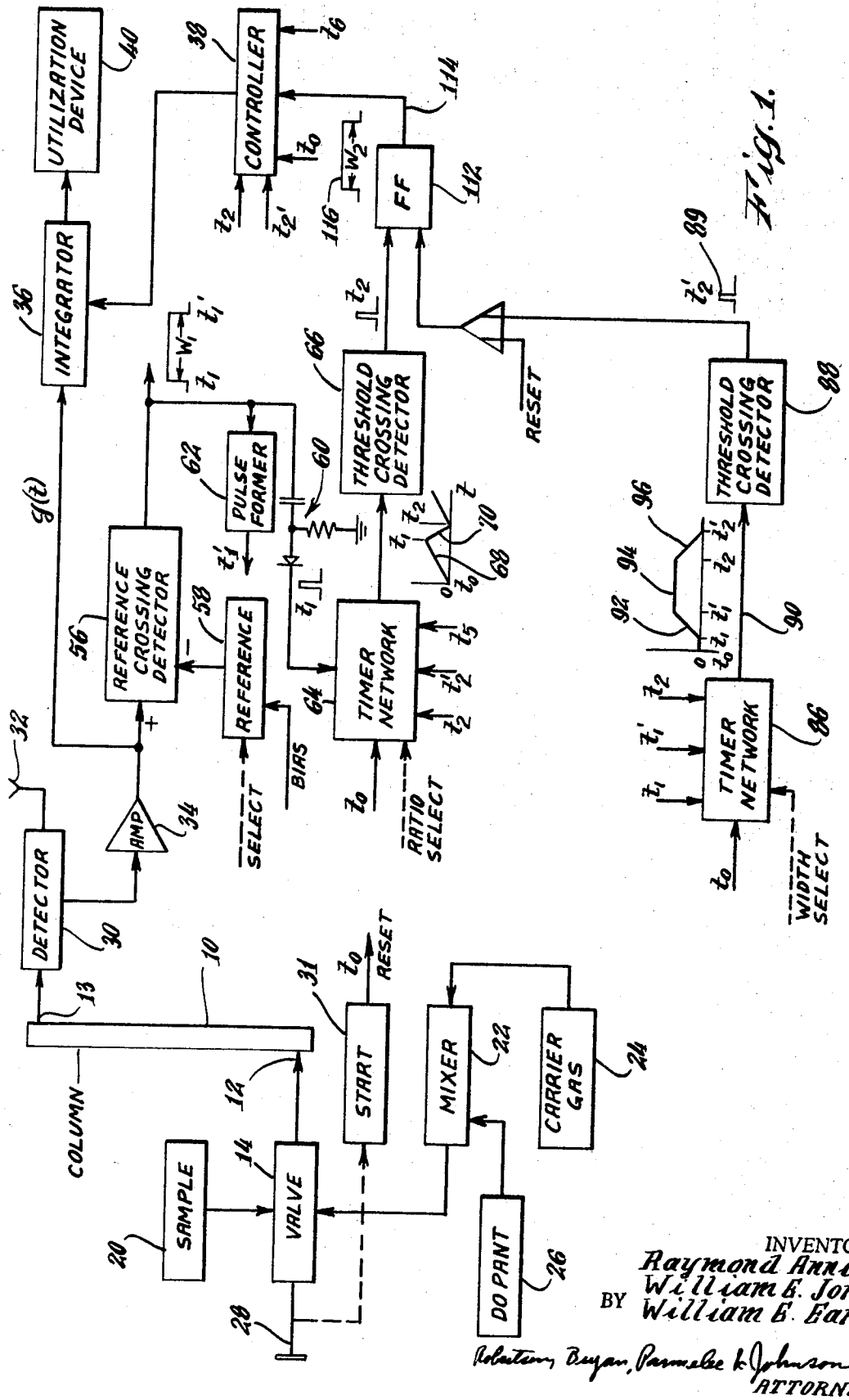
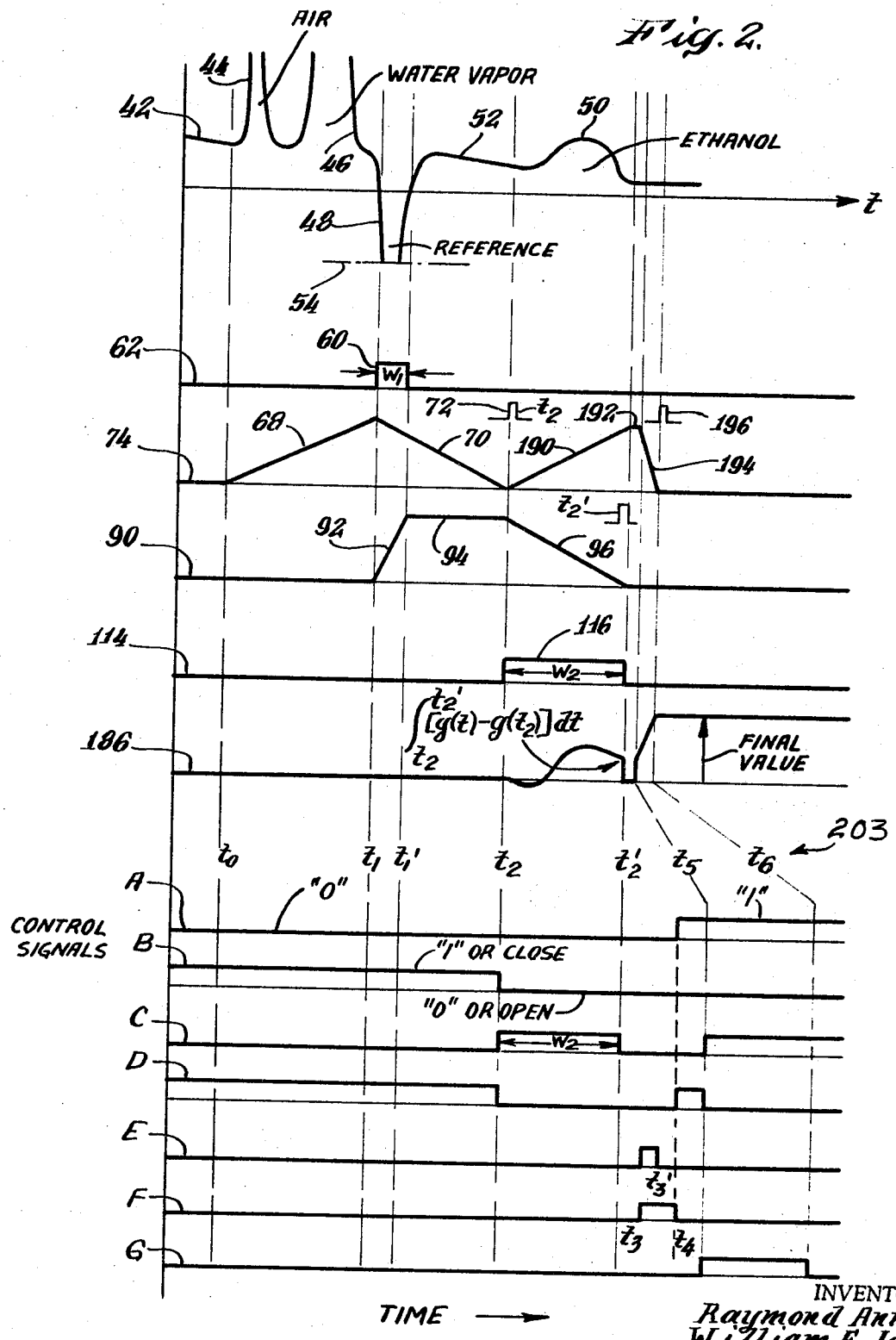


Fig. 1.

INVENTORS.
Raymond Annino
William E. Jordan
William E. Earle
BY
Robertson, Bryan, Parmelee & Johnson
ATTORNEYS.

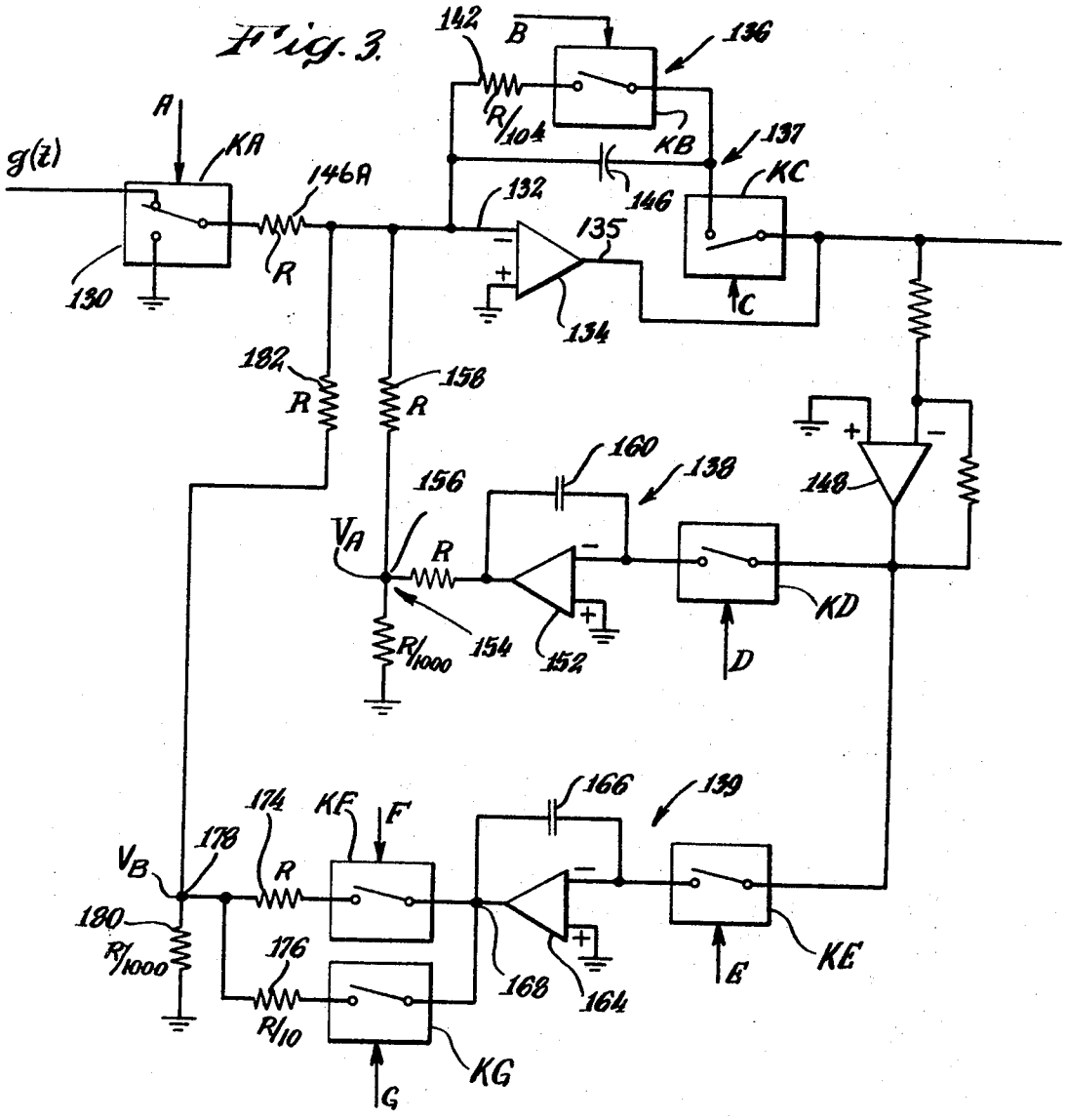
Fig. 2.



INVENTORS.
Raymond Annino
William E. Jordan
William E. Earle

BY
Robertson, Bryan, Pennebaker & Johnson
ATTORNEYS.

Fig. 3.



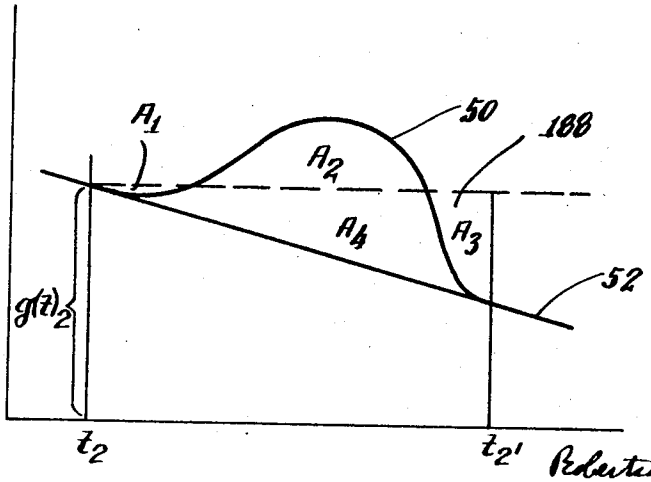


Fig. 8.

INVENTORS.
Raymond Annino
William B. Jordan
William E. Earle
BY
Robertson, Bogan, Pamerlee & Johnson
ATTORNEYS.

Fig. 4A.

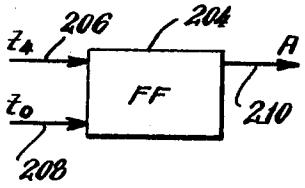
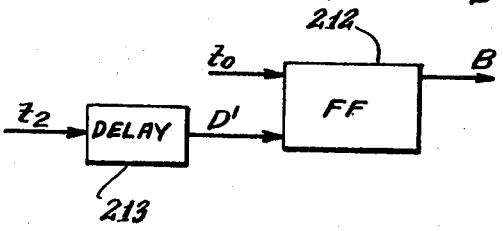


Fig. 4B.



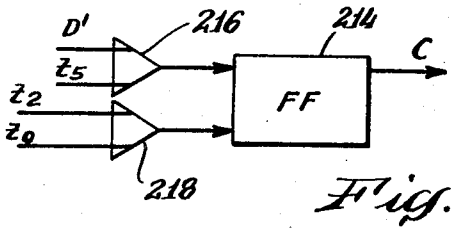


Fig. 4C.

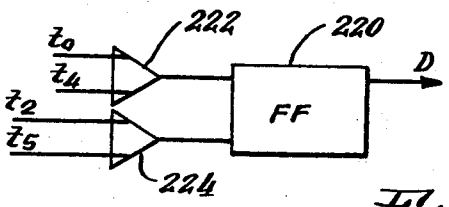


Fig. 4D

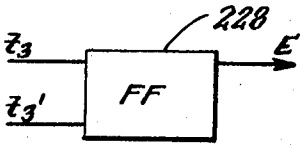


Fig. 4E.

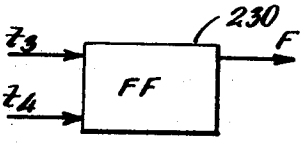


Fig. 4F.

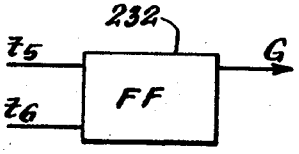


Fig. 4G.

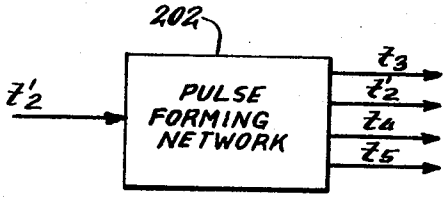


Fig. 5A.

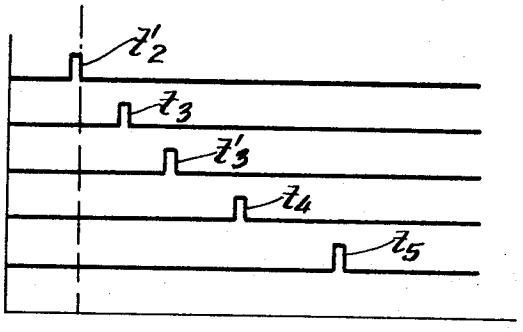


Fig. 5B.

INVENTORS.
Raymond Annino
William E. Jordan
BY William E. Earle

Robertson, Brown, Pamelee & Johnson
ATTORNEYS.

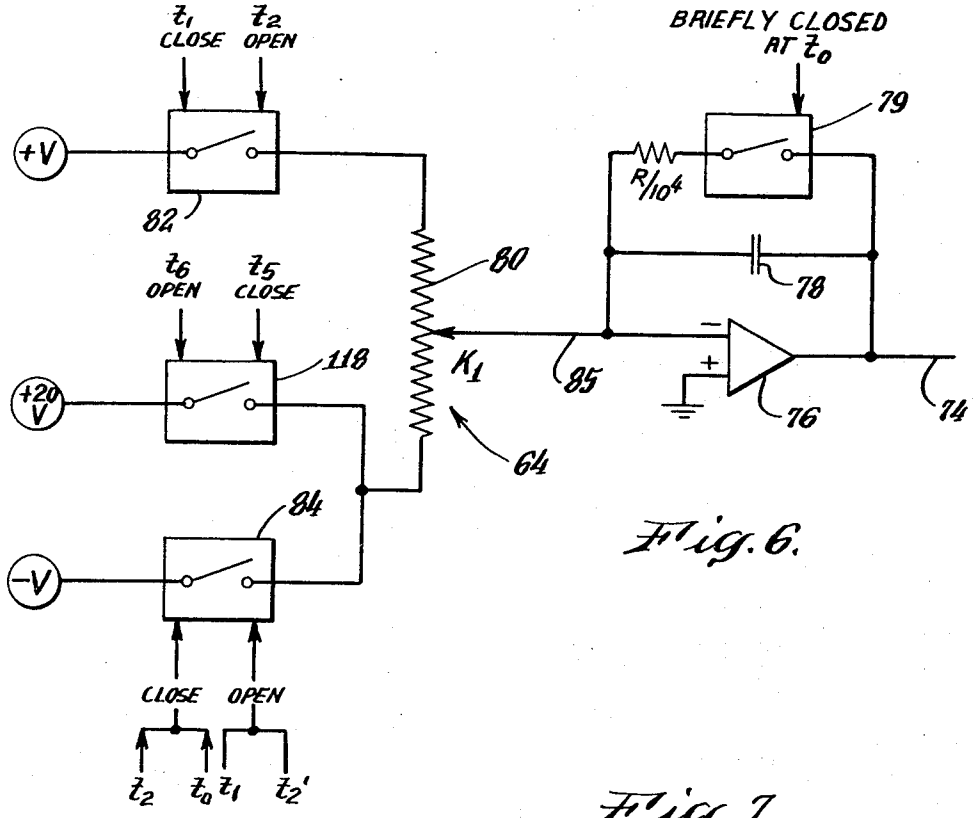
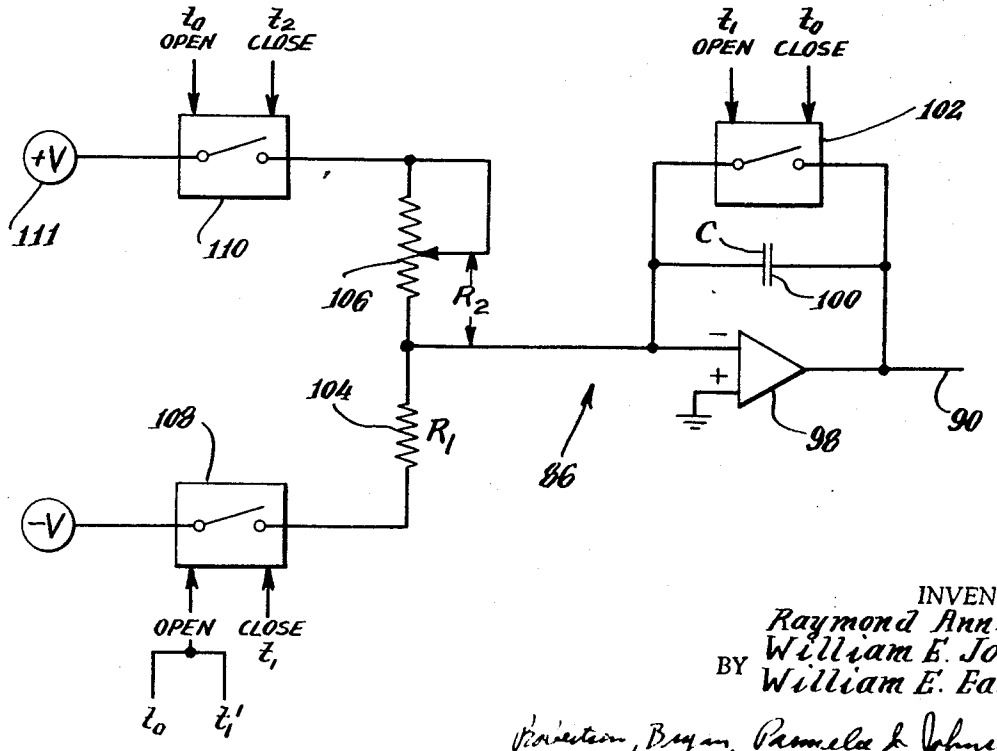


Fig. 6.

Fig. 7.



INVENTORS.
Raymond Annino
BY William E. Jordan
William E. Earle
Pennie, Bryan, Pennie & Johnson
ATTORNEYS.

CHROMATOGRAPHIC PEAK SELECTOR

This invention relates in general to the field of chromatography. More particularly, this invention relates to improved means and techniques for determining with high precision the concentration of a component of a sample mixture injected into a chromatographic column.

Chromatography is a physical method of separation wherein a mixture of two or more components to be separated is percolated through a column containing a large surface area stationary bed of material. The stationary bed in the column is adapted to retain the components of the mixture for differing periods of time. Thus the individual components emerge at different times from the column. A detector placed at the output of the column provides an electrical signal in the form of a series of peaks each representing the concentration of a respective component. A typical detector is a well known thermal conductivity cell.

The stationary material in the column may be either in solid or liquid form while the mobile mixture may be liquid or gaseous. Hence, four fundamental forms of combination are possible for the mixture and column material. These combinations are respectively gas-solid, gas-liquid, liquid-solid and liquid-liquid.

In one chromatography technique, a continuously flowing carrier gas, such as helium, is supplied to a column packed with an inert material such as Celite which has been coated with a high boiling point organic liquid such as dinonyl phtalate or silicone oil. A sample of the mixture to be analyzed is inserted in the carrier gas to be entrained thereby through the column. Components in the sample will have different degrees of affinity for the column materials and their retention times in the column thus will differ. Accordingly, the components in the sample will elute separately and at different times from the column.

Any variation in the composition of the gas exiting from the column caused by an eluted component from the sample will cause an unbalancing of a detector circuit at the column exit. The degree of unbalance is proportional to the concentration of the component. The detector sends a corresponding signal to a utilization device, frequently a strip chart recorder. The recorder thus draws a series of peaks which usually appear at least approximately symmetrical. The concentration of any component may be obtained by measuring the corresponding peak height, but for the most reliable and accurate results the peak area should be measured.

One serious problem in using the integration technique has been the difficulty of determining accurately when the integration should begin and end. Prior methods for accomplishing this have usually depended on a time base generator and a signal selector with adjustable controls to operate the selector at fixed points in time. Obviously, errors in the measurement will result if the integrating period used is too short for the particular peak of interest, or if the period begins too soon or ends too late so as to include a part of a preceding or succeeding peak in the integration results.

Although the length of time required for any particular component to pass through a column will be known approximately from previous calibration data, the exact time is influenced by factors such as temperature, column loading, which changes with usage, and flow rate. Prior equipment typically included control equipment to maintain the temperature and flow rate set at predetermined fixed levels, so that the analysis would always be conducted under the same conditions for which the original calibration was established. Such control equipment not only had to be quite elaborate and expensive, but it did not solve the problem of long-term variations in column loading.

In a preferred embodiment of the invention to be described hereinbelow in detail, there is provided a chromatographic instrument particularly designed to measure the concentration of a single component of an injected sample. Such an instrument is especially useful in process control applications. Another application is that of measuring the concentration of alcohol in a person's breath, in order to determine, for police purposes, whether he is intoxicated. In any such applications, the measurement must be consistently accurate. Moreover, the apparatus must be relatively simple and automatic in operation, yet it must be relatively inexpensive to manufacture.

In the described embodiment, special means are provided for accurately determining automatically the period during which the peak of interest is to be integrated. This determination is effective over a wide range of temperature, flow rate and column loading conditions prevailing during any particular analysis run. Thus, there is no need for highly elaborate control equipment for eliminating long-term drift of temperature and flow rate.

In accordance with one aspect of this feature, a reference component is supplied to the column in such fashion as to produce a reference peak prior to the peak developed by the component of interest. This reference peak has a unique characteristic permitting it to be distinguished from the other peaks automatically. Based on a measurement of the time difference between the appearance of this reference peak and the initial peak to appear in the detector output, a computation is made predicting the time of appearance of the peak of interest. Specifically, it can be shown that there will be a constant ratio between the times of appearance of the reference peak and the peak of interest, over changing conditions of flow rate and temperature. Similarly, the ratio of the width of the reference peak to the width of the peak of interest will remain substantially constant, so that by measuring the width of the reference peak, it becomes possible to automatically compute the width of the peak of interest, i.e., the length of time for the integrating action.

In the preferred embodiment, the apparatus employs a method which might be termed "plateau elution chromatography" as a means for developing a reference peak having a unique, readily recognizable characteristic. In this method, the carrier gas is purposely contaminated (doped) with a component which can dissolve in the stationary liquid phase or which can be adsorbed on the active solid support. Provided that the dopant is not present in the sample or, if present, is in lower concentration than in the carrier gas, it will be found that a reference peak will be produced at a retention time characteristic of the particular dopant and having a polarity opposite to the other peaks produced, e.g., the reference peak will be "negative" if the other peaks are considered to be "positive." Thus, the reference peak can be recognized and distinguished.

The time necessary for the reference component to pass through the column as well as the width of its peak are measured parameters used to generate a "time window" during which the detector output is identified as providing the peak of a component of interest to be analyzed. For the duration of this window the detector output is integrated to provide an accurate measure of the area of the peak of interest. The results of this integration represent the concentration of the component being analyzed.

As noted above, the time interval between an initial peak (such as air in the sample) and the reference peak will bear a constant ratio to, i.e., will be directly proportional to, the time interval between that initial peak and any selected peak of interest over changing conditions of temperature, flow or quantity of stationary phase. In some cases the initial peak occurs almost immediately after the sample is injected into the column, and as a practical matter it can be considered that the time between injection and appearance of the reference peak will be directly proportional to the time between injection and the appearance of the peak of interest. If the reference peak appears at a measured time t_1 with a time width of W_1, then the time t_2 for appearance of the peak of interest relates as

$$t_1/t_2 = K_1$$

and the time width W_2 of the peak of interest relates as

$$W_2/W_1 = K_2$$

These ratio relationships have been found to hold true over significant temperature and flow rate variations.

Based on these relationships, means are provided in the disclosed embodiment to compute automatically the starting and ending times for the period W_2 during which the integration of the peak of interest takes place. The integrating apparatus also includes special means to eliminate errors which might otherwise occur due to drift of the base line during integration.

Accordingly, it is an object of this invention to provide improved means and techniques for chromatographic analysis. It is a more specific object to effect an accurate integration of a signal peak produced by chromatographic methods. Other objects, aspects and advantages of the invention will be understood from the following description of a preferred embodiment of the invention considered together with the drawings wherein:

FIG. 1 is a schematic diagram of a gas chromatography instrument and system in accordance with the invention;

FIG. 2 is a waveform diagram illustrating operating waveforms of voltages at different points of the diagram of FIG. 1;

FIG. 3 is a circuit diagram of an integrator used to measure the area of the signal peak;

FIGS. 4A – 4G shows a network employed to generate timing signals for the operation of the integrator of FIG. 3;

FIGS. 5*a* and 5*b* show a timing diagram illustrating the operation of the network shown in FIG. 4;

FIG. 6 is a circuit diagram of a timer used in the embodiment of FIG. 1;

FIG. 7 is a circuit diagram of a timer used in the embodiment of FIG. 1; and

FIG. 8 is an enlarged portion of a signal waveform shown in FIG. 2.

With reference now to FIG. 1, a chromatography apparatus of the elution-partition type is shown in block diagram form. This apparatus includes a column **10** containing a suitable known material for the separation of components of a gaseous mixture. The column inlet port **12** is connected to the outlet port of a sample valve **14** which may be of known construction. The valve **14** has one input port **16** coupled to a supply of gaseous material **20** to derive a sample therefrom for analysis. A second input port **18** is coupled to a mixer **22** supplying a carrier gas, such as helium, and a dopant gas such as acetaldehyde, in a concentration of approximately 0.5 percent. The mixer **22** has input ports coupled to supplies of carrier gas **24** and dopant gas **26**. Alternately the dopant can be premixed and one tank of carrier gas used.

The valve **14** is provided with an actuating lever **28** which may be manually or automatically controlled. The actuation of the lever **28** introduces a preselected quantity of the sample into the carrier gas stream for passage to the column **10**. At the same time of the lever actuation a pulse t_0 is generated from a start network **31**. The sample gas inserted at the inlet port **12** of the column **10** eventually elutes from a column exit port **13** with the components separated by the column. The eluted material passes through a thermal conductivity cell **30** for the detection of the separated components and the production of electrical signals representative thereof. After passage through detector **30** the carrier gas stream and separated components are vented at an exhaust port **32**.

The output of detector **30** is amplified in amplifier **34** to provide a suitable signal $g(t)$ for analysis. The output of this amplifier is applied to an integrator **36** which operates under control of a timing controller **38**. The output of the integrator is applied to a utilization device **40** such as a strip chart recorder, meter or other display device.

In the chromatographic apparatus of FIG. 1, the carrier gas is doped with a preselected material, not present in the sample (or present in much smaller concentration), to cause the detector **30** to produce a reference peak having an opposite polarity from the peaks produced by other components in the sample. The reference peak in effect represents a "vacancy" in the sample, and may be unambiguously detected because of its opposite polarity characteristic. Although in the preferred embodiment the reference peak actually represents a missing component in the sample, it can be considered to have been produced by what can broadly be termed a reference component in the sample.

The waveform **42** in FIG. 2 illustrates a chromatography signal $g(t)$ representative of a person's breath containing ethanol. Several of these peaks are of such large amplitude that they are shown in truncated form. It should be understood, however, that these peaks are in practice essentially symmetrical and continuous.

A first peak **44** is produced by air in the sample, a second peak **46** by water vapor. The third peak **48** is the reference peak with an opposite polarity, and represents the absence of the preselected material used to dope the carrier gas. A fourth peak **50** represents the ethanol present in the sample. As can be seen from waveform **42**, the peaks **44**, **46** and **48** are large compared with the ethanol peak **50** but this is to be ex-

pected in view of the small amount of ethanol present. THe waveform **42** further has a base line **52** which as illustrated may drift downwardly or upwardly during the operation of the instrument.

The chromatograph waveform **42** commenced at a time t_0 corresponding with the time of insertion of the sample into the carrier gas stream. The air and water peaks **44–46** are detected first, followed by the reference peak **48** at time t_1. The appearance of the reference peak must be uniquely detected, in order to permit a measure of the time t_1 required for the vacancy to pass through the column, as well as its duration or "width" W_1 equal to $t_1' - t_1$. From this information a "-window pulse" is subsequently generated commencing at time t_2 and terminating at time t_2'.

Returning to FIG. **1**, the start pulse t_0 is directed to a first timer unit **64** provided to measure the time required for development of the reference peak. This timer basically consists of a ramp generator the output of which, as shown by waveform **74** (FIG. **2**) increases linearly with time while the reference component is in the column. In order to sense the appearance of the reference peak, the output of amplifier **34** is coupled to a reference crossing detector network **56** to which also is applied a reference signal from a reference source **58**. The reference signal voltage is selected to correspond with the threshold level **54** of FIG. **2**, and the network **56** produces a predetermined output signal whenever the measurement signal is below that threshold level. Thus, as the reference peak is produced by the chromatographic detector **30**, the crossing detector produces a pulse of width W_1, i.e., pulse **60** in waveform **62**.

The threshold level **54** is located quite low (near base line **52**) on the reference peak **48** to reduce the influence of amplitude changes of the reference peak on the measured duration W_1. The reference voltage may be manually adjusted, but generally remains fixed for any particular dopant. However, it may be desirable to provide means for automatically eliminating any net base line offset signal at the start of each cycle so as to maintain a constant threshold detection level relative to the base line of the signal from the detector **30**. Such offset can be introduced by a suitable bias which can automatically be made of the correct value to zeroize the output of detector **30**.

The leading edge of the reference peak pulse **60** is applied to the timer **64** to cause it to reverse its direction of ramp action, i.e., to sweep downward. The slope of the downward sweep is prefixed for the component of interest. Thus when the timer output returns to zero, it will at that instant identify a second time t_2 which is proportionally related to t_1 by a predetermined proportionality factor K_1.

The output of this timer unit **64** is applied to a second threshold crossing detector **66**. When the downward ramp **70** reaches zero volts, this threshold crossing detector produces a short output pulse **72**. By selecting the slopes of the upward and downward going ramps, the output pulse **72** is made to occur at the desired time t_2 signifying the start of the peak of interest. The pulse **72** is used to start the integration function for the peak of interest.

In order to determine the width of W_2 of the integration window following time t_2, means are provided to measure the width W_1 of the reference peak. For this purpose, there is provided a second timer unit **86** which, like the first timer **64**, makes use of a ramp generator for making time measurements. This second timer is started by the pulse t_1, and produces a linear upsweeping signal shown in waveform **90**. The subsequent pulse t_1' at the end of the reference peak stops the ramp action, and the signal level thus built up is stored for subsequent use in fixing the width of the integration window for the peak of interest. Thus, at the start of that window, time t_2, the ramp action is reversed to produce a downwardly sweeping signal form along **96**. When the signal crosses zero volts, this is detected by a crossing detector **66** which thereupon produces a pulse t_2' signifying the end of the period of integration. The slope of the downward ramp is pre-set at a fixed ratio with respect to the slope of the up ramp. Therefore, the period of integration will always be directly related to the width of the reference peak.

FIG. **6** illustrates details of the timer network **64**. An operational amplifier **76** is provided with positive and negative inputs and a negative feedback capacitor **78** to provide an integration function at its output **74**. A potentiometer **80** has its end terminals selectively coupled to positive and negative supply voltages through switches **82–84** to provide the respective upward and downward ramps **68–70**. Switches **82** and **84** may be electronic elements such as gate insulated field effect transistors. As shown, switch **84** is closed at t_0 and opened at t_1, and switch **82** is closed at t_1 and opened at t_2. The voltage at output **74** thus swings upwardly and downwardly at a slope determined by the setting of potentiometer **80** with the result that the downward ramp reaches zero volts at a time t_2 related to t_1 by the ratio of $t_1/t_2 = K_1$ where K_1 is selected in accordance with the setting of the wiper arm **85** of the potentiometer. Switch **79** is closed at the beginning of the t_0 pulse and opened at the end of the pulse to assure that at time t_0 the voltage across the capacitor **78** is zero.

FIG. **7** illustrates a detailed circuit which will produce the waveform from network **86**. Thus an operational amplifier **98** having positive and negative input terminals with positive input grounded is provided with a negative feedback capacitor **100** having a capacitance C. A switch **102** shorts capacitor **100** to provide initial voltage control (zero) over the capacitor stored voltage. The negative input terminal is coupled to the junction of a fixed resistor **104** having a resistance R_1, and a variable resistor **106** having a resistance R_2. The resistors **104** and **106** are connected respectively to negative and positive voltage sources through switches **108** and **110** respectively. The switches **108** and **110** may be electronic and are actuated under control of timing pulses as labelled in FIG. **7**.

In the operation of the timer circuit **86**, and with reference to the waveform **90** in FIG. **2**, switch **102** is closed at time t_0 and opened at time t_1. At time t_1 switch **108** is closed thereby permitting the output of operational amplifier to rise, charging capacitor **100** at a slope determined by $(+V.)/R_1C$ V represents the supply voltage. At time t_1' (W_1 later), switch **108** is opened and the output on capacitor **100** is held at level **94** since there is no discharge path. At time t_2 switch **110** is closed, thereby forcing a downward sloping along ramp **96** with a slope determined by

$(-V.)/R_2C$

When the ramp **96** crosses zero volts the detector **88** issues the pulse **89** which is representative of the time t_2' when the pulse W_2 terminates.

Pulses t_2 and t_2' respectively set and reset a flip flop **112** to provide at its output **114** a wide pulse **116** representative of the desired integration window W_2.

The width W_2 of the window pulse **116** can be varied by selecting the position of the wiper arm of the variable resistor **106** in FIG. 7. This varies the resistance R_2 and correspondingly alters the constant K_2 in the relationship

$$W_2/W_1 = K_2$$

In this manner the proper constants may be dialed into the instrument by varying the potentiometer **80** (FIG. **6**), for K_1, and **106** (FIG. 7) for K_2.

In a typical application of the instrument of FIG. 1 for the detection and integration of the ethanol peak **50**, K_1 was 0.45 and K_2 was 3.74.

Integration of the ethanol peak **50** proceeds as may be best understood with reference to FIG. **8** which shows the peaks on an enlarged scale. In general, the procedure is as follows: At time t_2 the function g (t) is sampled and the voltage g (t_2) of the base line **52** is stored. Thereafter integration proceeds on a signal which is the difference between g (t) and g (t_2). The area thus integrated equals $(-A_1 + A_2 - A_3)$. At the end of the integration at time t_2', the base line **52** is sampled again and the difference between the base line voltages at times t_2 and t_2' is stored. Thereafter the area within the triangle formed by areas $A_1 + A_4 + A_3$ is determined by integrating the stored difference signal and dividing the result by two. The triangle area is effectively added to the previously integrated function to yield the net area of the peak $A_2 + A_4$.

FIG. 3 illustrates details of the integrator **36**. The detector output signals g (t) from the amplifier **34** are coupled through a switch KA to the negative input **132** of an operational amplifier **134** having its positive input grounded. This amplifier includes a feedback capacitor **146** which, in conjunction with input resistor **146A** provides linear integrating action. During the initial part of the cycle, i.e., prior to integration, capacitor **146** is effectively by-passed by a resistor **142** so that amplifier **134** serves a normal amplifying function.

The operation of the integrator **36** is dependent upon the setting of the several switches. The following description of its operation relies upon a preselected terminology for the control signals A, B, C, D, E, F, and G which respectively control the state of switches KA, KB, KC, KD, KE, KF and KG. A "**0**" indicates that the switch is in its normal state, as drawn in FIG. 3, and a "**1**" indicates the switch is in its alternate position. The control signals for the switches are generated as shown in FIG. **4**. The switches may be conventional electronic gates. The timing relationships of the control signals A through G may be observed from FIG. **2** where a positive level represents "**1**," and a zero level represents "**0**."

Prior to integration, the switches are in the condition

A	B	C	D	E	F	G
0	1	0	1	0	0	0

During this condition, the voltage is applied to input **132** of amplifier **134** which acts as a straight amplifier. Its output is inverted by a second amplifier **148** and directed through a feedback loop **138** including a third amplifier **152**. The net voltage at the input to amplifier **134** is maintained at zero by negative feedback action, so that the voltage at junction **156** will be equal to $-g$ (t). A voltage divider preferably is used in the output circuit of amplifier **152**, e.g., to provide a ratio of 1,000:1, so that the voltage at the amplifier output actually will be $-1{,}000 \times g$ (t). This voltage is stored in a memory capacitor **160** connected around amplifier **152**. It may be noted that this feedback action prior to integration effectively zeroizes the integrator, i.e., it stores a signal value representing not only base line drift level, but also including a component responsive to any amplifier offset voltage.

At time t_2 switch KD is opened, and the signal at the output of amplifier **152** is $-1{,}000 \times g(t_2)$. This value is stored in capacitor **160** since there are no current paths available to discharge capacitor **160**. Thus, $V_A = -g(t_2)$.

KC is now closed and KB is opened so that the control signals are

A	B	C	D	E	F	G
0	0	1	0	0	0	0

The feedback current around amplifier **134** through capacitor **146** is

$$\frac{g(t) - g(t_2),}{R}$$

where R is the resistance of resistors **146A** and **158**. This current develops a voltage across the capacitor **146** (of capacitance C) equal to

$$\frac{1}{RC}\int_{t_2}^{t}\Big(g(t) - g(t_2)\Big)dt.$$

Since the input of amplifier **134** is maintained at ground potential (the + input is grounded), the voltage at output **135** of amplifier **134** will be equal to the voltage across capacitor **146**. The waveform **186** in FIG. **2** illustrates the voltage at the output **135** of amplifier **134**.

At the time t_2', the end of the integration period W_2, the switch KC is opened to stop further integration, and the charge on the capacitor **146** remains fixed at that level while certain control functions are effected to prepare for a base line correction. To this end, switches KE and KF are closed to bring in operation another feedback loop **139** including a second feedback amplifier **164** having a memory capacitor **166**. The control signals are thus in the following states:

A	B	C	D	E	F	G
0	0	0	0	1	1	0

Amplifier **134** still requires, because of its grounded positive input, a zero input voltage. This requirement drives the junction **178** to a voltage $V_B = -g(t_2') + g(t_2)$. After this voltage has been established, switch KE is opened so that the memory capacitor **166** stores the voltage $1{,}000\,(g(t_2) - g(t_2'))$, i.e., a voltage representing the difference between the base line level at the start of integration and the base line level at the end of

integration. This difference voltage thus reflects the amount of base line drift which has occurred during integration.

The next step in preparing for the base line compensation is to zeroize feedback loop **138**. For this purpose, switch KD is closed and switch KA is opened. This effectively causes junction **156** to return to zero since all other inputs to amplifier **134** are now equal to zero. Thus any amplifier offset voltages are substantially eliminated. The switch signals at this time are as follows:

A	B	C	D	E	F	G
1	0	0	1	0	0	0

Immediately after discharge of memory capacitor **160**, the switch KD is opened to disconnect the second feedback path **138** from the integrating amplifier **134**. The integrating capacitor **146** however retains the accumulated integral.

At this time the base line correction is commenced by applying to the input of the integrator amplifier **134** a voltage derived from memory capacitor **166** and carrying out a further integration for a period of time proportional to the period of original integration. As previously explained, the capacitor **166** stores a voltage representative of the difference between the base line levels at times t_2 and t_2'. Graphically, the correction required is to remove the area enclosed by the right-angled triangle indicated at **188** in FIG. **8** from the area originally integrated for the time period W_2. Hence, after the storage in capacitor **166** of the difference voltage $g(t_2) - g(t_2')$ and the effective discharge of capacitor **160**, switches KC and KG are closed to connect the stored signal of memory capacitor **166** to the input of the integrating amplifier **134**. The charge on the capacitor **146** thereby is augmented (or diminished) at a rate proportional to the amount of base line drift which occurred during integration. The control signals at this time are:

A	B	C	D	E	F	G
1	0	1	0	0	0	1

The time period of this compensating integration is determined by the time period of the original integration "window." However, it will be apparent that the compensation time period need not be the same as that of the original integration, providing that a corresponding change is made in the level of voltage applied to the input of the integrating amplifier during the selected time period. In one embodiment, the input voltage level was selected to be equal to 10 times the actual amount of base line voltage drift. This multiplication factor was introduced by a suitable voltage divider in the output circuit of amplifier **164**, specifically by selecting the resistor **176** to be **100** times the size of resistor **180**. Thus the voltage at junction **178**, with switch KG closed, will be .01 times the voltage on memory capacitor **166**; since this capacitor voltage was 1,000 times the base line drift voltage, the net output voltage will be 10 times the base line drift voltage.

With a voltage multiplication factor of 10, the time period for the compensation integration must be one-twentieth of the original period of integration, in order to produce the required amount of correction as discussed above in connection with FIG. **8**. This reintegration time period is controlled, in the present embodiment, by the same timer unit **64** previously used to fix the time t_2, i.e., the start of the original integration window.

With further reference to FIG. **6**, therefore, at time t_2 switch **84** was again closed to cause the output of timer **64** to ramp up as indicated by waveform **74**, at line **190**. The switch **84** is opened at the end of the integration (t_2'), and the timer thereby stores a signal level **192** corresponding to the period of integration (W_2).

Upon the commencement of the base line correction, at time t_5, a switch **118** (FIG. **6**) is closed connecting the negative input of the amplifier **76** to a positive voltage source **122**. This source has a voltage which is greater than that of the positive voltage source **111** by a factor of 20, determined by the desired shortening of the time for the base line correction.

The closure of switch **118** drives the output of amplifier **76** to ground along ramp **194** (FIG. **2**). Upon reaching zero volts output, the threshold crossing detector **66** issues a pulse **196** at a time t_6. The time period $t_5 - t_6$ for base line correction is proportionally related to the integration window W_2. This proportional dependence upon the period W_2 may be appreciated from the fact that the duration of W_2 determines the level **192** and, consequently, the time for the subsequent downward ramp **194** to reach zero volts.

Returning to FIG. **3**, the only input to amplifier **134** during the time period $t_5 - t_6$ is the difference voltage 10 $(g(t_2) - g(t_2'))$ at the output of amplifier **164**. The base line correction integration proceeds with this voltage for the time period $t_5 - t_6$ as indicated by waveform **186**. Upon the occurrence of the t_6 pulse **196**, the switch KG is opened to stop integration, and the amplifier output **135** at time t_6 represents the area of the ethanol peak **50**. This final output may be recorded on a suitable strip chart instrument or the like.

FIGS. 4A through 4G and FIGS. 5A and 5B illustrate a circuit for generating the control signals A through G. These control signals are produced by using timing pulses such as t_0, t_1, t_1', t_2, t_3, t_3', t_4, t_5 and t_6. The generation of these timing pulses has been described previously except for t_3, t_3', t_4 and t_5. With initial reference, therefore, to FIGS. 5A and 5B the timing pulse t_2' is shown connected to a network **202** from which pulses t_3, t_3', t_4 and t_5 are generated in the order as shown in the timing diagram of FIG. **5B**. These pulses may be produced by conventional delay circuits and single shot multivibrators as are well known in the art. The pulses t_3, t_3', t_4 and t_5 occur right after the integration window when the voltage waveform **74** in FIG. **2** is held to level **192**. Since this appears as a small time span, the control signals effective during this time period are shown on an enlarged time scale as is indicated at **203** in FIG. **2**.

FIG. **4A** includes a flip flop **204** having a set input **206** and a reset input **208**. The output **210** provides the desired control signal A for the control of switch KA in FIG. **3**. The reset signal for flip flop **204** is the pulse t_0 and the flip flop is set by timing pulse t_4 as shown by the A waveform in FIG. **2**.

FIG. **4B** includes a flip flop **212** which is set by pulse t_0 and reset by a pulse D' generated from a delay circuit **213** actuated by timing pulse t_2. The output of flip flop **212** is the control signal B having a waveform as shown

in FIG. 2. The delay of the resetting of flip flop **212** avoids unwanted transients as various switches in the integrator are actuated.

FIG. 4C includes a flip flop **214** set by a pulse from an OR circuit **216** driven by timing pulses D' and t_5. The flip flop **214** is reset by a pulse from OR circuit **218** coupled to timing signals t_2' and t_0. The output of flip flop **214** is the control signal C whose waveform appears in FIG. 2.

FIG. 4D illustrates a flip flop **220**, which is set by an OR circuit **222** coupled to timing pulses t_0 and t_4 and is reset by the OR circuit **224** coupled to timing pulses t_2 and t_5. The output of the flip flop **220** is the control signal D whose waveform appears in FIG. 2.

FIG. 4E shows a flip flop **228** set by timing pulse t_3 and reset by t_3'. The output of the flip flop **228** is the control signal E shown in FIG. 2. In a similar manner the flip flops **230** and **232** provide at their outputs control signals F and G whose waveforms are shown in FIG. 2. The flip flop **230** is set by t_3 and reset by t_4. The flip flop **232** is set by t_5 and reset by t_6.

The utilization device **40** may be actuated at the completion of the base line correction to record the final integrated output from the amplifier **134** at time t_6. Such synchronized recordation may be accomplished by coupling the t_6 pulse to a strip chart recorder whose pen is momentarily enabled by pulse t_6 to record the output **135** of amplifier **134**.

Although a preferred embodiment of this invention has been described hereinabove in detail, it is desired to emphasize that this is for the purpose of illustrating the invention, and should not be considered as limiting thereof except as required by the prior art, it being understood that many variations within the scope of this invention can be devised by those skilled in this art to meet requirements of specific applications. For example, although the instrument described herein provides a measure of a single component of a sample mixture, it is apparent that the same techniques can be used for measuring multiple components. Other modifications will be apparent.

We claim:

1. A chromatographic apparatus comprising a column through which a sample mixture may be directed and including material adapted to separate the individual components of the mixture so that they elute from the column in predetermined sequence to be sensed by a detector which produces an output in the form of a peak-type signal responsive to the concentration of the component in the sample mixture; the improvement in said apparatus for identifying the time when a selected component of interest will elute from the column so as to permit making an accurate measurement of the signal produced thereby, comprising:

means to introduce into the column a substance to develop in said output a reference peak occurring prior to the peak of interest and having a uniquely distinguishable characteristic, the rates of passage through said column of said substance and said selected component having a predetermined relationship;

means to detect said distinguishable characteristic;

means to measure the time between a predetermined initial event and the detection of said reference peak; and

means responsive to said measured time for computing in accordance with said predetermined relationship a corresponding time when said peak of interest will be developed.

2. Apparatus as claimed in claim **1**, wherein said measuring means comprises an electronic ramp generator adapted to produce a linearly varying signal until said reference peak is detected, whereby the signal level at that time represents the measured time.

3. Apparatus as claimed in claim **2**, wherein said responsive means includes means for reversing said ramp generator so that it ramps back to its initial condition;

means for detecting the time when the ramp signal reaches its initial condition and for starting the integration of the peak of interest.

4. The method of chromatographic analysis comprising the steps of:

operating a chromatographic column to produce a uniquely recognizable reference peak the time of appearance of which bears a certain relationship to the time of appearance of the component of interest;

determining the time period between a predetermined initial condition and the appearance of the reference peak;

computing a second time, subsequent to and based both on said time period and said certain relationship, following which the component of interest will appear; and

producing a signal responsive to said component of interest which appears after the computed second time.

5. The method of claim **4**, wherein the computation of said second time includes the step of multiplying the first time period by a predetermined constant.

6. The method of claim **4**, including the steps of:

measuring the width of the reference peak;

performing a mathematical operation on said measured width to compute a predicted period of time for the width of the peak of interest; and

integrating the signal produced by the component of interest during the computed time period thereof.

7. A chromatograph apparatus comprising;

chromatographic column means adapted for separating components in a sample mixture driven therethrough by a fluid carrier stream including a preselected reference component having a uniquely recognizable elution characteristic upon separation by the column means, the time of appearance of said reference component bearing a predetermined relationship to the time of appearance of a desired mixture component;

detecting means for sensing the emergence of components separated by the column means and for producing signals representative thereof;

means for recognizing the detected signal representative of the preselected reference component and for producing a timing signal representative of the time for the reference component to pass through the column means; and

means effectively controlled by the timing signal and said predetermined relationship for selecting the detected signal of said desired mixture component to be analyzed and for producing a signal corresponding therewith for analysis.

8. The apparatus as claimed in claim 7 and further including:
means actuated by the timing signal for integrating the mixture component detected signal for a time period related to the timing signal.

9. The apparatus as claimed in claim 8, wherein the timing signal producing means further includes:
means actuated by the detected reference component signal for generating a first pulse signal having a width related to the width of the detected reference component signal; and
means for generating a second pulse signal having a width bearing a first preselected ratio to the width of the first pulse and applying the second pulse signal to the integrating means to determine integration of the desired mixture component signal.

10. The apparatus as claimed in claim 9, wherein the second pulse signal generating means further includes:
means for initiating the second pulse signal at a time bearing a preselected ratio to the time for the reference component to pass through the column means.

11. A chromatographic apparatus comprising:
chromatographic column means for separating components in a sample mixture entrained therethrough by a carrier fluid stream;
detecting means for sensing components eluting from the column means and for producing electrical peak signals representative thereof;
means for introducing a reference fluid into the column means to identify the time when a selected component in the sample mixture will elute from the column, said reference fluid being selected to produce a uniquely recognizable reference peak signal from the detecting means at a time bearing a predetermined relationship to the time of elution of said selected component;
means based on said predetermined relationship, and actuated by the reference peak signal, for generating a control signal substantially coincident in time with the elution of the selected component from the column means; and
means actuated by the control signal for analyzing the peak of the selected component.

12. The apparatus as claimed in claim 11, wherein the reference fluid introducing means introduces the reference fluid into the carrier fluid to produce a recognizable vacancy reference peak signal from the detecting means representative of the relative vacancy in the sample mixture of the reference fluid in comparison with the carrier fluid.

13. A method of analyzing a component in a sample mixture introduced to a chromatographic column, comprising the steps of:
measuring the time required for the elution of a uniquely recognizable reference component from the chromatographic column;
computing the start time of a time period the start of which bears a predetermined relationship with the measured reference component elution time, with the computed start time being subsequent to the elution of the reference component and identifying a time after which a desired component in the mixture will elute from the column; and
producing a signal responsive to the column output after the computed time.

14. The method as claimed in claim 13, wherein said signal is produced by integrating an electrical signal representative of the column output.

15. A method of determining the concentration of a component in the breath of a person with a gas chromatographic column comprising the steps of:
inserting a preselected reference component having a recognizable elution characteristic into a carrier gas stream coupled to the chromatographic column;
introducing a sample of the breath from a person into the carrier gas for entrainment thereby through the column;
detecting separated components emerging from the column;
measuring the elution time and the duration of the reference component;
defining in response to the measured elution time and duration a time period when there will emerge from the column a desired component the elution time and duration of which have a predetermined relationship with said reference component elution time and duration;
generating an electrical signal representative of the separated desired breath component; and
integrating the electrical signal for said defined time period.

16. A method of determining the concentration of a component in a mixture with a chromatographic column comprising the steps of:
measuring the time required for the passage of a uniquely recognizable reference component through the chromatographic column to a detecting station;
selecting a desired component emerging from the column for analysis during a time period the start time of which bears a predetermined ratio to the measured time for passage of said reference component; and
producing a measurement signal responsive to the column output during said time period.

17. A method of selecting a component in a mixture for analysis from a chromatographic column comprising the steps of:
inserting a reference component having a uniquely recognizable elution characteristic into a carrier fluid stream coupled to a chromatographic column;
introducing a sample of the substance to be analyzed into the carrier fluid stream;
detecting components separately emerging from the column;
recognizing the detected reference component characteristic;
determining the time period required for the reference component to pass through the column; and
selecting a desired separated mixture component following the reference component at a subsequent time period the start time of which is determined in preselected relationship with said determined time period for passage through the column of the reference component.

18. The method as claimed in claim 17, wherein said preselected relationship is a predetermined ratio between the passage time of the reference component and the time duration between the introduction of said

sample and the start time of said subsequent time period.

19. The method as claimed in claim 18, wherein the selecting step further includes the steps of:

measuring the time width of the reference component after passage through the column; and

selecting the desired component for analysis for a time period duration bearing a predetermined ratio to the measured time width of the reference component.

20. In chromatographic analysis of a fluid sample wherein the sample is passed through a chromatographic column to produce at the column output time-separated component-peak elutions the amplitudes of which are detected for analysis purposes;

the improved method of selecting a particular component for analysis which comprises:

controlling the input to the column at a start time to produce in the output thereof a reference peak which precedes the component to be selected and which has a unique detectable output signal characteristic distinguishing such reference peak from the peak of any other component in the sample, the time of appearance of said reference peak and of said particular component being related by a certain known function;

producing a first signal responsive to the detection of said reference peak and indicating the time between said start time and the time of detection of said reference peak;

producing a second signal bearing a predetermined relationship to said first signal in accordance with said certain known function to identify the start of a subsequent time period during which the desired component is at the column output; and

selecting for analysis the column output signal appearing during the time period identified by said second signal.

21. For use with chromatographic apparatus of the type including a column through which a fluid sample is to be passed for the purpose of separating on a time scale the components of said fluid sample for individual detection by a detecting apparatus;

that improvement for automatically selecting a particular component for analysis which comprises:

control means to control the input to the column to produce in the output a reference peak preceding the particular component to be selected and having a unique characteristic distinguishing such reference peak from the output signal produced by any other component in the fluid sample;

recognition means operable with said detecting apparatus to recognize said unique characteristic;

means to produce a first signal indicative of the time of passage through said column of said reference peak;

computation means responsive to said first signal and operable to produce a selection signal identifying on said time scale a subsequent time period the start of which bears a predetermined relationship to the time of detection of said reference peak; and

selection means operable by said selection signal to produce an output signal responsive to the output of said column during said subsequent time period.

22. Apparatus as set forth in claim 21, including means for producing a width signal responsive to the width of the reference peak;

said computation means including means responsive to said width signal for controlling the duration of said subsequent time period in accordance with a predetermined relationship to said width signal.

23. For use with on-line chromatographic apparatus of the type including a column through which a fluid sample is to be passed for the purpose of separating on a time scale the components of said fluid sample for individual detection by a detecting apparatus;

that improvement for automatically selecting a particular component for analysis which comprises:

control means to control the input to the column to produce in the output a reference peak preceding the particular component to be selected and having a unique characteristic distinguishing such reference peak from the output signal produced by any other component in the fluid sample;

recognition means operable with said detecting apparatus to recognize said unique characteristic while the material is eluting from the column;

means under the control of said recognition means to produce a first signal indicative of the time of passage through said column of said reference peak;

computation means responsive to said first signal while said fluid sample continues to elute from said column and operable to produce a selection signal identifying on said time scale a subsequent time period the start of which bears a predetermined relationship to the preceding time of detection of said reference peak; and

selection means operable during said subsequent time period to produce an output signal responsive to the output of said column while the component of interest is being detected.

24. In chromatographic analysis of a fluid sample wherein the sample is passed through a chromatographic column to produce at the column output time-separated component-peak elutions;

the improved method of selecting a particular component for analysis which comprises;

controlling the input to the column at a start time to produce in the output thereof a reference peak which precedes the component to be selected, said reference peak having a unique detectable output characteristic distinguishing the reference peak from the peak of any component in the sample and appearing in the column output at a time which is related to the time of appearance of the component to be selected in accordance with a predetermined relationship;

producing a first signal responsive to the detection of said reference peak and which serves to indicate the time between said start time and the time of detection of said reference peak;

producing a second signal responsive to said first signal and in accordance with said predetermined relationship to identify a subsequent time after which the desired component appears at the column output; and

performing in response to said second signal an operation the function of which is related to the

appearance of said desired component at the column output.

25. The method of claim 24 carried out in an on-line chromatographic analysis wherein said first signal is produced when said reference peak is detected and said second signal is produced while portions of the sample subsequent to said reference peak are eluting from the column.

26. In chromatographic analysis of a fluid sample wherein the sample is passed through a chromatographic column to produce at the column output time-separated component-peak elutions;

the improved method of selecting a particular component-peak which comprises:

controlling the input to the column at a start time to produce in the output thereof a reference peak which precedes the component peak to be selected and which has a unique detectable output signal characteristic distinguishing such reference peak from the peak of any other component in the sample, the times of appearance of said reference peak and said component peak having a predetermined relationship;

producing a first signal responsive to the detection of said reference peak and representative of the time between said start time and the time of detection of said reference peak;

producing a second signal responsive to said first signal and in accordance with said predetermined relationship to identify the selected component-peak signal; and

performing in response to said second signal an operation related to said desired component.

27. The method of claim 26, wherein said operation comprises producing a peak signal responsive to the detection of said selected component;

zeroizing an integrator at the time of said second signal; and

applying said peak signal to said zeroized integrator.

28. The method of claim 26, including:

producing a third signal representative of the time duration of said reference peak; and

performing a mathematical operation on said third signal to compute a predicted time period following said start time and during which the desired component will be available for sensing.

29. The method of claim 28, including the step of integrating the selected component-peak signal throughout said computed time period.

30. The method of claim 29, including the step of zeroizing an integrator at the time of said second signal; and

applying said selected component-peak signal to said zeroized integrator.

31. For use with chromatographic apparatus of the type including a column through which a fluid sample is to be passed for the purpose of separating on a time scale the components of said fluid sample for individual detection by a detecting apparatus;

that improvement for automatically selecting a particular component for analysis which comprises:

control means to control the input to the column to produce in the output a reference peak preceding the particular component to be selected and having a unique characteristic distinguishing such reference peak from the output signal produced by another component in the fluid sample;

recognition means operable with said detecting apparatus to recognize said unique characteristic;

means to produce a first signal indicative of the time of passage through said column of said reference peak;

computation means responsive to said first signal and operable therewith to produce a selection signal identifying on said time scale the start of a subsequent time period with the start time bearing a predetermined relationship to the time of detection of said reference peak;

an integrator under the control of said selection signal and operable to produce an output signal corresponding to the time-integration of the selected component peak; and

means responsive to said selection signal for zeroizing said integrator prior to the integration of said selected component peak.

32. In chromatographic equipment of the type including a column through which a fluid sample is to be passed for the purpose of separating on a time scale component peaks of said fluid sample;

that improvement for use in selecting a particular component comprising:

control apparatus for controlling the input to the column to produce in the output thereof a reference peak preceding the peak of the desired component to be selected and having a unique characteristic distinguishing such reference peak from the output signal produced by any other component in the fluid sample, the times of appearance of said reference peak and the peak desired to be selected being related in accordance with a predetermined function;

a detector responsive to the column output and arranged to recognize said unique characteristic;

a signal-producing device coupled to said detector to produce a first signal representative of the time of appearance of said reference peak;

computation apparatus responsive to the time represented by said first signal and operable to produce in accordance with said predetermined function a selection signal identifying the desired component; and

an output element under the control of said selection signal to produce a function related to the appearance of said desired component.

33. In a chromatographic process wherein a sample mixture is introduced to a chromatographic column for separation of components thereof, the method comprising the steps of:

measuring the time required for the elution of a uniquely recognizable reference component from the chromatographic column;

computing the start time of a time period the start of which bears a predetermined relationship to the measured reference component elution time, with the computed start time being subsequent to the elution of the reference component and identifying a time after which a particular component of the sample mixture will elute from the column;

producing a signal indicating said computed start time; and

performing in response to said signal a function related to the appearance of said particular component.

* * * * *