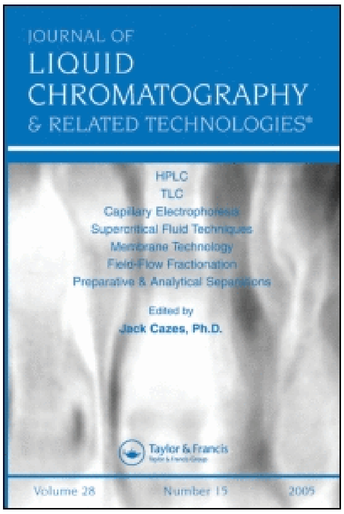

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Software for Microprocessor Controlled HPLC: Data Acquisition and Display

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SOFTWARE FOR MICROPROCESSOR CONTROLLED HPLC: DATA ACQUISITION AND DISPLAY

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ABSTRACT

Programs are described for the direct computer-assisted acquisition and display of chromatographic data from conventional Hplc equipment. The programs are written for a dual-microprocessor Hplc controller and provide the necessary utility routines to calibrate the instruments, and to acquire, display, and store chromatographic data in real-time. The data acquisition routines use data filtering techniques suitable for real-time data acquisition which remove spurious noise and ripple associated with detector analog outputs. The results of chromatographies indicate only

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This is the third paper in this series (see ref. 7 and 8 for first two papers).

Address all correspondence to RPS. Inquire RPS for availability of complete program listings on floppy disk or hard copy (Tel: 316-689-3120). Program listings are also described in the dissertation of D. B. Smoll, it can be loaned from Ablah Library, The Wichita State University, Wichita, KS 67208.

Abbreviations: A/D, analog-to-digital; D/A, digital-to-analog; HPLC, Hplc, High-performance Liquid Chromatography; DHC, data handling microcomputer; see the text for other abbreviations.

Note: Apple is a registered trademark of Apple Computer, Inc. and Explorer-85 is a registered trademark of Newtronics Research and Development, Ltd.

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negligible differences between data acquired via the computer equipment and data acquired directly from Hplc detectors. The stored data can be retrieved for further computer-assisted analysis.

INTRODUCTION

The chromatography of complex mixtures produces chromatograms which are difficult to analyze. Retrieving information from these chromatograms is time consuming to decipher by manual methods. Moreover, due to imprecision in manual measurement techniques, a significant degree of subjectivity is introduced into the analysis. However, the introduction of computers for experimental data analysis can eliminate many of these difficulties and offer fast and accurate methods for complete analysis of these chromatograms (1-6). Though several computer-assisted chromatography systems are available commercially, they often are not versatile enough to permit use of existing equipment, and therefore, the investigator ends up purchasing several hardware and software Hplc components. Furthermore, they often lack the flexibility desired for specific, unique research needs of the investigator. For example, the "Rapid Scan Detector" (LKB-Produkter AB, Bromma, Sweden, model 2140) requires a software package (LKB 2140-250 wavescan) to access and store the data in an IBM PC computer. However, this software still fails to integrate the peaks and process the data. For this processing, another software package (Nelson Analytical, Inc., CA, model 3000) is required.

This paper is the third paper in a series dealing with microcomputer-assisted Hplc. Earlier articles describe the microprocessor hardware, interface hardware, and operating system software used to acquire and analyze real-time chromatographic data and to provide an operator-interface (7,8). The objective of this work is to facilitate microcomputer-assisted analysis of chromatographic data using existing chromatography detectors, strip-chart recorders, and pumps. This paper deals with the software to acquire, filter, and display chromatographic data. The programs described herein are executed entirely in real-time.

The first step in this effort is the acquisition and entry of the chromatographic data into the computer. The hardware, as described in the previous papers, is designed specifically to simplify the procedure by reducing the programming necessary to service the interfaces and automating data converter functions. Hence, data entry is a straightforward procedure since data is read directly from the converters when the conversion is completed (8). However, sufficient software control is maintained over the interfaces to provide programming flexibility. This was essential to ensure versatility of the data-handling microcomputer (DHC) for future applications.

The use of conventional detectors necessitates filtering of the raw data to remove noise and other interferences (9). The direct connection of detectors to recorders, in the past, has not required any signal conditioning since the dampened response of most recorders serves as a built-in filter. However, the response time of the electronic hardware used here, being much faster, requires some signal conditioning to eliminate undesirable electrical signal components (10). Although, use of such filters was restricted in order not to limit application of the interface or affect the accuracy of the monitoring device.

A more flexible alternative to hardwired signal conditioning is software filtering of the data, though care must be exercised to maintain the transparency of the computer to the data collection process. Weighting of the collected data provides a criterion for the rejection of spurious noise and instabilities (11). The method of weighted "boxcar" averaging provides a means to reject electical spikes and average out noise without adversely affecting the data (12). However, this method is only applicable to instability within the data set to be averaged.

In our application, the major component of electrical instability within a data set is AC ripple. Therefore, selection of the appropriate timing interval for data conversion scheduling is able to provide a reproducible and reliable average data value (13). Instability between data points is screened by defining a minimum interval for legitimate peaks. Peaks encountered that require less than this interval are interpreted as electrical instability and they are averaged out over the interval. This method of filtering out noise has the additional advantage of being more applicable to "real-time", machine-language program where data is collected at short intervals.

MATERIALS AND METHODS

A. Hplc Equipment

The Hplc equipment consisted of conventional Hplc equipment interfaced to a dual microprocessor system as outlined in Scheme I of the previous paper (8). The columns (0.4 X 25 cm, reversed-phase C-18, Rainin Microsorb) were enclosed in a water jacket and maintained at a constant temperature using a constant temperature bath (Haake). The sample was applied using an injector equipped with a calibrated sample loop. The column was eluted with a microprocessor-controlled solvent delivery system capable of generating complex elution gradients (Perkin-Elmer Series 4 Liquid Chromatography pump). All solvents were mixed and gradients were generated using computer-assisted programs for this pump.

Effluent absorbance was monitored at 254 nm using an absorbance monitor (ISCO, Lincoln, NB, model UA-5 with Type-6 optical unit) and also simultaneously at 280 nm using another monitor (Altex Instuments, CA, model 152) connected in tandom. The former (ISCO) monitor was equipped with a micro-flow cell of 19 microliter volume (series 0080-012), and the latter (Altex) monitor with a flow cell of 20 microliter volume. Each absorbance monitor could be connected directly to a separate strip-chart recorder or to the data-handling microcomputer (DHC), specifically designed for our needs (7,8).

Effluent from the column (after passing through the UV monitors) was collected with a fraction collector (ISCO, model 1200 PUP) for peak characterization, etc.

B. Buffers and Solvent

Buffers were made from analytical grade chemicals, and absorbance of buffer solutions was checked with a spectrophotometer at 254 and 280 nm. After filtering with a filter of 0.22 micrometer - pore diameter (Millipore Corp.), they were stored at 4°C until used. Similarily, glass-distilled water was filtered, and both buffers and distilled water were sparged with helium gas for 10-15 min just prior to use.

Organic solvents used in reversed-phase chromatography were Hplc grade and were used directly without filtering but were sparged with Helium prior to use.

C. Gradient Programs

Gradient programs used for this work are shown in Table I. Reversed-phase columns were equilibrated routinely with a mixture of 75.0% Methanol and 25.0% water. The columns were cooled to ambient temperature and then stored overnight in this solvent at the end of an experiment.

D. Computer Equipment and Operating System

The Data Handling microcomputer (DHC) consisted of two interconnected computers (Apple II computer and Explorer-85 microcontroller) with peripherals and interfaces. The Turnkey and System Supervisor programs were used as the operating system for the two

TABLE I: ELUTION GRADIENT PROGRAMS for Hplc PUMP

SECTOR	TIME (min)	FLOW RATE (ml/min)	RESEVOIR(%) A	B	C	GRADIENT
EQUIL		1.5	100.0	0.0	0.0	
1	15.0	1.5	95.0	3.0	2.0	linear
2	15.0	1.5	85.0	9.0	6.0	linear
3	10.0	1.5	75.0	15.0	10.0	concave
4	10.0	1.5	25.0	45.0	13.0	concave
5	10.0	1.5	0.0	60.0	40.0	concave
6	5.0	1.5	0.0	60.0	50.0	

Resevoirs: A: 0.25 M Ammonium Acetate, pH 6.00; B: Distilled Water; C: Acetonitrile computers. These two programs provided the operator interface and automated the loading and running of the Explorer programs as described in detail in an earlier paper (8).

DATA ACQUISITION SOFTWARE DESIGN

The Turnkey Program provided the operator interface with all Explorer programs as mentioned above. The System Supervisor Operating program was loaded automatically along with a "patched" version of Apple DOS 3.3 by the Turnkey program. The System Supervisor routines were then used to initialize the Explorer, load and run Explorer programs, and retrieve data from the Explorer. Programs and data stored on floppy disks were retrieved using embedded DOS commands of the Turnkey program.

A. Description of Chromatography Monitoring Programs

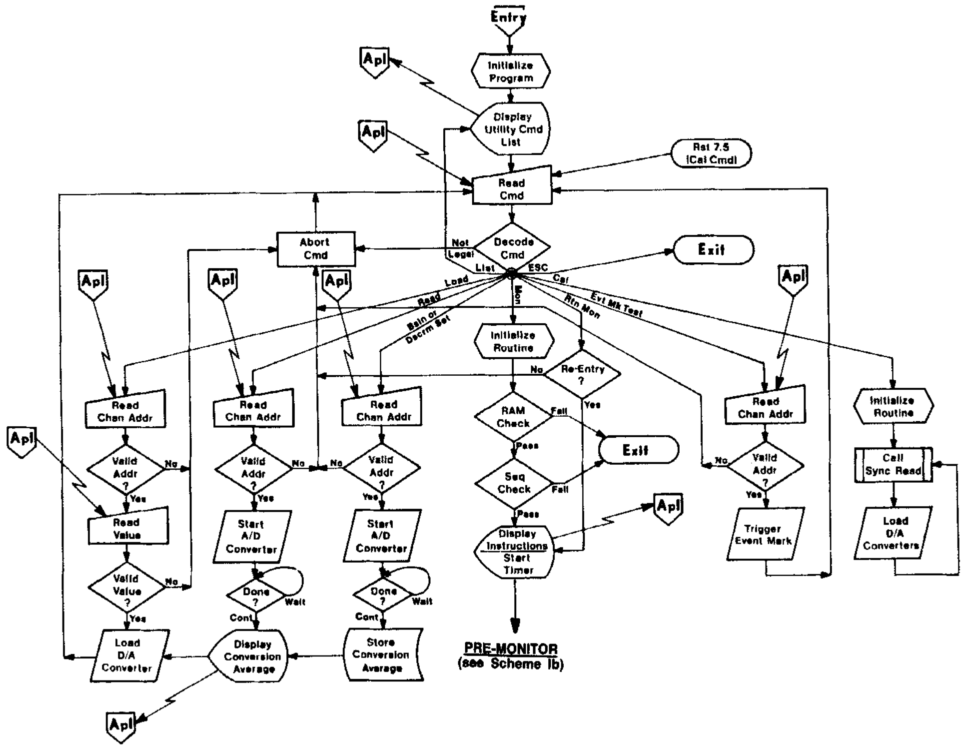
Two machine-language programs residing in the Explorer were used to monitor the output of the two monitors (ISCO and Altex). Both programs consisted of modular utility command routines individually called through a command decode mainline program. This limited structure allowed random scheduling of the utility commands.

Two programs were required to handle different levels and types of noise in the output signal of the two monitors. The program "FOLLOWER" was used normally with the ISCO monitor, and was written to average out the low-level 60 Hz ripple. This program computed the digital value of the monitor reading as an average of 256 conversions made over a 66.67-msec period. The program "NOISE DAMPER", on the other hand, was used with the Altex monitor to remove the attenuation-dependent 104 Hz ripple on the chopper inputs of its amplifiers. However, it could also handle 60 Hz ripple, and therefore, could be used with either monitor. This was achieved by computing the digital value as an average of 256 conversions over a 116.17-msec period and using a subroutine which dampens abrupt changes in data values.

The two programs were identical, as shown in Scheme I of this paper, except for the above-mentioned differences. Both programs were designed to record and display two A/D analog input channels simultaneously (referred to as Channel 0 and Channel 1). Data values for these channels were stored in two 4K-RAM storage buffers ($1000-$1FFF for channel 0 and $2000-$2FFF for channel 1) after the digital value was computed without any further processing. The pointers and counters were saved in the Explorer between $0F00-$0F3F. Data values for the absorbance monitor readings were otuput through the D/A converters to strip-chart recorders as they were computed, with the exception of "NOISE DAMPER". The display was 5 sec behind the readings because of the noise-handling in this program.

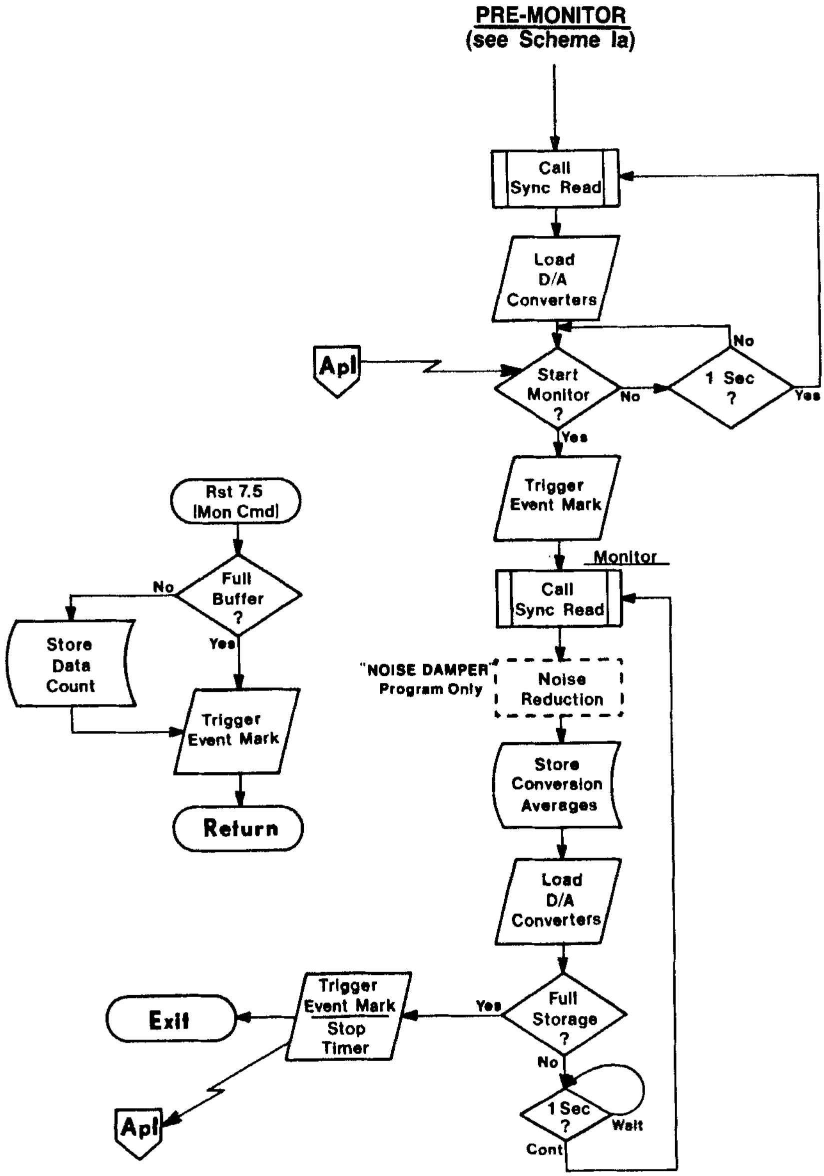
1. Utility Commands

A list of the utility commands was displayed when the program was entered. Commands could be entered only in response to a prompt. Consequently, commands had to be terminated before a new command could be issued. Only valid commands were decoded, and other entries resulted in a default, designated by a question mark, which aborted the current command.

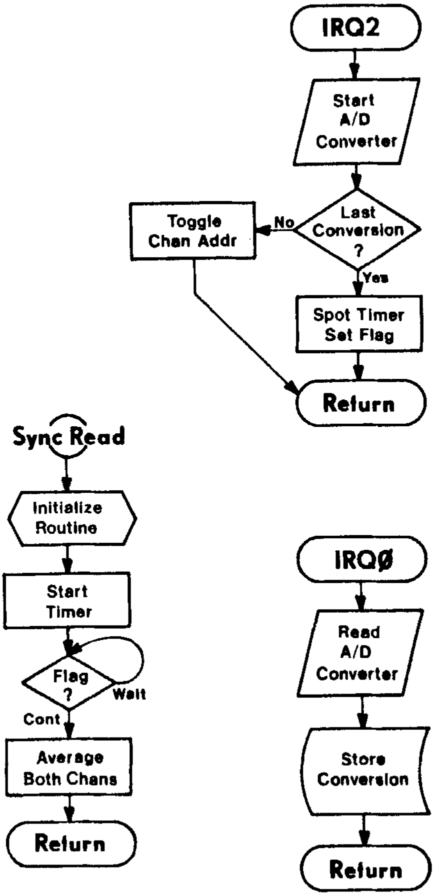


Scheme 1a. Flow Diagram of Data Acquisition Programs: Utility Commands. Utility functions are provided for equipment calibration, testing, and entry into the data acquisition routines.

The utility commands provided the means to enter the software routines necessary to perform an experiment. Strip-chart recorders and UV monitors could be calibrated through commands which directly loaded D/A converters, and read data directly from A/D converters. These calibration commands were used routinely at the begining of each experiment to check instrument calibration and to monitor continuously equilibration and cleaning of Hplc columns.



Scheme 1b. Flow Diagram of Data Acquisition Programs: Hplc Monitoring Routines. The data acquisition routine (monitor) is entered through the pre-monitor. The principal function of the routine is to acquire and save a digitalized version of the chromatogram.



Scheme 1c. Flow Diagram of Data Acquisition Programs: Interrupt Handler Routines and Dual A/D Synchronized Read Routines. The synchronized read routine minimizes the offset between the reading of the two A/D converter channels during chromatographic data digitalization.

The monitor command was entered through a pre-monitor after completion of set-up of the experiment. The data acquisition monitor routine was the only routine in which chromatographic data was saved. After completion of the chromatography, the monitor routine was exited and the data transfered to the Apple computer for further processing.

2. Pre-Monitor Routine

This section of the program was entered directly upon execution of the Return-to-Monitor or Monitor commands. This routine performed not only diagnostics to ensure proper operation of the computer, but also initialization of the Monitor routine before attempting to save the chromatographic data. Because of this pre-initialization of the Monitor routine, immediate entry into the data acquisition monitor routine was possible on injection of the sample.

The Pre-Monitor routine was intended to be a temporary state of the program which waited for the completion of the set-up procedures of Hplc (i.e., sample loading and column equilibration). Digital values for UV monitor readings were computed and displayed via strip-chart recorders as they were made at a 1 Hz rate. The program could remain in this section indefinitely, but no data value was saved while in this section.

3. Monitor Routine

This section of the Monitor command was entered immediately from the "Pre-Monitor" by pressing any key on the Apple computer keyboard (ESC key was used most often). Event-mark instructions were issued to the strip-chart recorder on both channels. Digital values continued to be computed at a 1 Hz rate as in the pre-monitor, but were saved in consecutive-memory locations in the data-storage buffers of the Explorer. Data-storage pointers and data count were updated as each data value was computed.

The program remained in this section of the Monitor command until either an Explorer "RESET" terminated it or a "Data Count Overflow" was encountered. The former was executed by pressing the "R" key of the Explorer. It resulted in no loss of data, but the Explorer had to be reinitialized before data could be transferred to the Apple computer.

"Data Count Overflow" termination of the Monitor command was executed when the data-storage buffers became full, i.e., after 1 hr 8 min 16 sec. No data values were lost, and the program was exited to the Explorer System Monitor for data transfer to the Apple computer.

4. Marking Buffer

The data count was saved in the Marking Buffer when the "I" key of the Explorer was pressed without disrupting the data-collection process. Event-mark instructions were issued to the strip-chart recorder to mark the position of the data count. The Marking Buffer resided at $0F20 - $0F2F in the Explorer and could save a maximum of eight marks. After it was full, the data count was no longer saved but event marks were still issued to the recorder. This procedure was used to record the data count at which the elution gradient changed.

5. Diagnostics

Diagnostics performed on entry into the pre-monitor consisted of two routines which checked all memory addresses from $0F3C to $2FFF in the Explorer (twice) when the Monitor command was used (this did not apply to the Return-to-Monitor command). The first routine, RAM Check, tested all bits for viable RAM. If this test was passed, the program proceeded to a second diagnostic routine, Sequence Check. This latter diagnostic tested all combinations of address lines of this memory for valid addressing.

If either test failed, the message "Failed at $XXXX" was displayed, where $XXXX indicated the memory address where the diagnostic found an error. The Monitor command was aborted, and program control was transferred to the system monitor for further investigation.

B. Description of Turnkey Operation

All Explorer programs were loaded and data saved using the Turnkey Program. After automatic initialization of the Explorer, the selected Explorer program was retieved from floppy disk and loaded into the Explorer by the Turnkey Program. Computer control was then transferred to the System Supervisor program and the Explorer program was entered. All Explorer progam utility commands were issued from the Apple keyboard directly to the Explorer through the monitor mode of the System Supervisor. (The DOS or EOS command mode of the Supervisor were rarely used since the Turnkey provided a simplified use of these routines.) After completion of the chromatography, the Explorer program was exited and control was returned to the Turnkey by performing a machine-language "RTS". The data was transferred from the Explorer through the Turnkey and saved on floppy disk for future retrieval.

Another chromatography experiment could then be run by re-entering the Supervisor and Explorer program and repeating the above procedure. A new Explorer program was loaded into the Explorer if necessary. Neither the Supervisor nor the Turnkey needed to be reloaded since the Turnkey contained all necessary functions and it was never exited.

RESULTS

A. INSTRUMENT EVALUATION

1. Evaluation of data regeneration by microcomputers

To examine true reproduction, acquisition, and retrieval of the chromatographic data using data-handling microcomputers (DHC), the following experiments were carried out. A complex mixture of solutes consisting of the four major, and several minor, modified ribonucleosides was applied to a RP-Hplc column. The separation was recorded simultaneously at 254 and 280 nm by a strip-chart recorder and also by DHC connected in parallel to the monitor output. After reproduction of the chromatogram from the DHC, the two chromatograms (Figure 1) were examined for congruity by superimposing them. Differences in peak width and retention time were within 0.5% of the corresponding values for the chromatogram produced directly from the monitor. These small differences (±0.5%) are well within tolerence limits of the strip-chart recorder speed

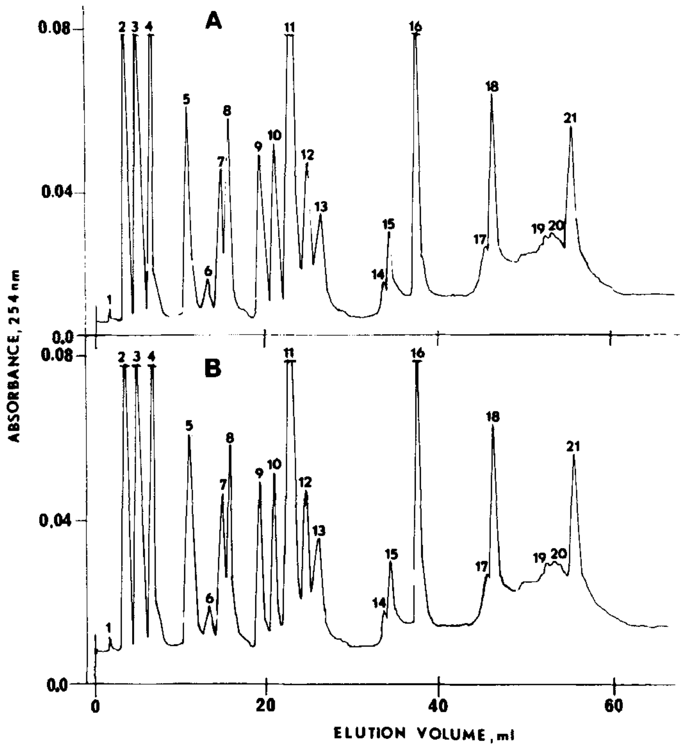


Figure 1. Hplc separation of a complex mixture of ribonucleosides on a C-18 reversed-phase column. Separation was carried out with programed gradients (see Table I). Panel A, data acquired directly from the Hplc detector. Panel B, data displayed from Data Handling Microcomputers (DHC) after its stroage in the DHC memory and filtering the data (i.e., removal of noise, ect.). No significant differences between the two chromatograpms can be noted

(±1.5%). Peak heights displayed by DHC (Table II) are in total agreement with the direct recording for nine peaks, reduced for seven peaks (0.6-1.4%), and incresed for the remaining five peaks (0.3-1.7%). The DHC data vary for most components within a range of -0.05 cm to +0.08 cm which is within the resolution of ±0.10 cm for a 10-inch (25.5 cm) vertical display. (The resolution is a

TABLE II
COMPARISON OF PEAK HEIGHTS RECORDED BY TWO DIFFERENT METHODS IN CHROMATOGRAPHY OF A COMPLEX MIXTURE OF RIBONUCLEOSIDES

Peak No.	Ribonucleosides	Peak Height in cm Method A	Method B	Different from Md.B (%)
1	"Breakthrough"	0.51	0.51	0.0
2	Pseudouridine	24.30	24.00	-1.25
3	Cytidine	12.01	11.86	-1.26
4	Uridine	17.30	17.35	0.29
5	5-Methylcytidine	8.36	8.36	0.0
6	1-Methyluridine	1.65	1.65	0.0
7	2'-O-Methylcytidine	6.25	6.25	0.0
8	Guanosine, 5-Methyluridine	7.95	7.95	0.0
9	7-Methylguanosine	6.60	6.55	-0.76
10	2'-O-Methyluridine	7.01	6.96	-0.72
11	2'-O-MeGuo, 1-MeGuo	18.06	18.21	0.82
12	Adenosine	6.15	6.20	0.81
13	2-Dimethylguanosine	4.29	4.29	0.0
14	2'-O-Methyladenosine	1.52	1.52	0.0
15	2-Methyladenosine	3.61	3.56	-1.40
16	6-Methyladenosine	12.24	12.19	-0.41
17	Unknown	2.87	2.92	1.71
18	6-Dimethyladenosine	8.91	8.86	-0.56
19	Unknown	3.38	3.38	0.0
20	Unknown	2.97	2.97	0.0
21	Unknown	7.51	7.53	0.93

Method A, signal from monitor recorded directly by a strip-chart recorder.

Method B, signals acquired by Data Handling Microcomputer from monitor were sent to the strip-chart recorder using a recording speed same as in Method A.

consequence of the step function of the digital converters; it is based on a full scale of 25.5 cm).

Peaks, exceeding this resolution (peaks 2, 3, and 11), were carefully examined. The apex of each peak was characterized by at least three points of the same value. Hence, the peak-height differences could not result from the program read-cycle, which was repeated every one second. Deviations in peak heights were further examined by approximation of the error caused in integration. The error amounted to losses of only 0.21% and 0.06% for peaks 2 and 3 respectively, and a gain of 0.17% for peak 11 in their peak-area integration.

DISCUSSION

As the complexity of a chromatogram increases so does the burden placed on the investigator to analyze it. As a consequence, manual routine analysis of complex chromatograms is not practical without adversely affecting the productivity of the investigator. To solve this problem, we have interfaced a typical Hplc system to a dual-microprocessor controlled data-handling system (DHC). This paper deals with the prerequisites for microcomputer-assisted analysis of chromatographic data, that is, acquisition, storage, and retrieval of the data. Subsequent papers will deal with the processing of the data collected.

A. Hardware and Software for DHC

The hardware for the DHC is designed to facilitate real-time acquisition of chromatographic data. The coupling of the Explorer to the Apple computer provides the necessary interfacibility and processing throughput required for real-time controlling and monitoring of the Hplc equipment. This extends the capabilities of the DHC beyond conventional computer peripherals, e.g., disk drives and printers. The hardwiring of required interface procedures -- such as initiation and EOC resetting of converters -- reduces the complexity of the protocol for these procedures and frees CPU time. As a result, the Explorer is able to acquire, average, and display the data with sufficient time left in a 1 Hz cycle to perform any necessary noise reduction. Similarily, the interrupt system and the synchronized timers provide the necessary event timing and communication to the CPU with minimal CPU intervention.

The software for the DHC is designed to provide real-time data collection and display, storage, and retrieval of chromatographic data. The software consists of interactive programs which can be divided into two categories: operating system programs and application programs. The operating system programs, System Supervisor and Turnkey, are described in detail in a previous paper (8). For this work, however, they have been expanded to automate the loading and running of the Explorer programs. The use of a user-friendly language (BASIC) for the Turnkey programs makes such modifications less time consuming and disruptive to existing programming.

The application programs consist of the data acquisition programs written for the Explorer. They are written to be as unobtrusive to the operation of the Hplc equipment as possible. Although such added complexity to the operation of the equipment is not entirely avoidable since the DHC and the Hplc equipment must be manually synchronized for certain operations, and the calibration of the instruments must be performed through the DHC. The normal experimental procedures for Hplc, however, are maintained.

B. Accuracy of Acquisition and Display of Data

1. Data resolution

The programs written for diagnostics and assay of interface performance verify reliability and accuracy of the hardware components of DHC. To establish accuracy of data display from D/A converters, calibration procedures are performed at the beginning of each experiment. Thus, reproduction of the chromatogram by DHC results in a qualified evaluation of accuracy maintained by A/D converters and programs during the digital-conversion process. The chromatogram obtained directly from the detectors as compared with that acquired and reproduced by DHC is shown in Table II and Figure 1. The results indicate that both accuracy and reproducibility of data are maintained by DHC within resolution limits of the display for most peaks (±1.0 mm). Differences between the two methods (excluding peaks 2, 3, and 11) occur with a standard deviation of only 0.039 around a mean of -0.001. This indicates consistently accurate data conversion and processing by the monitoring programs.

To further enhance resolution of data (i.e., to reduce tolerance limits of data) with the use of the exsisting 8-bit A/D converters, it is possible to revise the programming and upgrade the D/A converter interface. The signal-averaging routine used in the digital-conversion process sums 256 8-bit values to generate a 16-bit total which is divided by 256 by truncating the least-significant byte. Hence, a portion of the least-significant byte of the 16-bit total can be used to increase the number of bits stored and displayed.

2. Error in data collection

Only three peaks exceed tolerance limits (peaks 2, 3, and 11 in Table I). Differences in these peaks cannot be caused by the A/D converter or by monitor instability, since conversion error for the A/D converters was 0.5 of the least-significant bit (14) and monitor instability would have appeared in both versions of the chromatograms recorded simultaneously. However, to test the "worst case" situation, the "NOISE DAMPER" program was used for this chromatography. It uses a subroutine which can alter the value of the stored data in order to eliminate monitor instability. This subroutine averages readings when a slope-reversal occurs within a four-second period, thus accounting for three identical values at the apex of these peaks.

Interestingly, evaluation of apparent error caused in the peak integration indicates that the error is rather insignificant: -0.21, -0.06, and +0.17% for peaks 2, 3, and 11 respectively. Moreover, peaks eluted very early in the chromatogram and having appreciable absorbance (for example, peaks 2 and 3) tend to be affected by the above signal-averaging routine, since they have the steepest slope and are the narrowest.

CONCLUSIONS

The hardware interfaces, described in previous papers, and the software, described here, are capable of producing data appropriate for further processing by the DHC. The noise filtering employed by the programs is adequate to remove the AC ripple and spurious noise found in the analog outputs of Hplc detectors. The resulting data is reliable and reproducible, and is well within the expected tolerance limits of the detectors and strip-chart recorders. As a consequence, further processing of this data can also be expected to produce equally reliable and reproducible results.

The programs have demonstrated their ability to function reliably. Both Explorer programs as well as the System Supervisor and the Turnkey Operating System have been in use for more than two years without any problems. All utility commands and routines are used constantly, and they are free of "bugs". The unobtrusive nature of the operation of the DHC and its software makes data collection by the DHC easy to learn and use. The DHC has been used satisfactorily in our laboratory for data collection and routine monitoring of Hplc for over two years.

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REFERENCES

1. Long, E. C., Managing laboratory data on a desktop personal computer: part three, Amer. Lab., **16** (10), 96, (1984).

2. Long, E. C., Managing laboratory data on a desktop personal computer: part two, Amer. Lab., **16** (9), 112, (1984).

3. Walling, P. L., Gentille, T. E. and Gudat, A. E., Amer. Lab., **14** (2), 23, (1982).

4. Harder, M. E. and Koski, P. A., Amer. Lab., **15** (9), 27, (1983).

5. Tehrani, A. Y., Automating an existing analytical chromatograph for preparative HPLC, Amer. Lab., **17** (2), 43, (1985).

6. Li, J., Hillier, E. and Cotter, R., Computerized HPLC detection in pharmaceutical research, Amer. Lab., **7** (2), 93, (1985).

7. Singhal, R. P. and Smoll, D. B, Hardware and software for microprocessor controlled Hplc: Interfacing of converters (A/D and D/A) for data acquisition and display, J. Liquid Chromatogr., this volume, (1986).

8. Smoll, D. B. and Singhal, R. P., Hardware for microprocessor controlled HPLC: interfacing of an interval timer and interrupt controller to the S100 Bus System, J. Liquid Chromatogr., **6** (11), 2095, (1983).

9. Malmstadt, H. V., Enke, C. G., and Crouch, S. R., in Electronics and Instrumentation for Scientists, The Benjamin/Cummings Publishing Company, Inc., Reading, (1981), pp. 409.

10. Cassell, D. A., in Microcomputer and Modern Control Engineering, Reston Publishing Co., Inc., Reston., (1983), pp. 47.

11. Bevington, P. R., in Data Reduction and Error Analysis for the Physical Sciences, McGraw-Hill Book Co., New York, (1969), pp. 106.

12. Hutchens, D. E. and Hester, R. D., Computer assisted data acquisition and analysis for size exclusion chromatography, J. Liquid Chromatogr., **6** (6), 1037, (1983).

13. Malmstadt, H. V., Enke, C. G., and Crouch, S. R., in Electronics and Instrumentation for Scientists, The Benjamin/Cummings Publishing Company, Inc., Reading, (1981), pp. 406.

14. National Semiconductor's Linear Databook, National Semiconductor Corp., Santa Clara, (1980), 8.45.