The creation of a laboratory instrument quality monitoring system with AdaSAGE

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ABSTRACT

Two existing Ada tools AdaSAGE and AYACC were combined to produce a system that parses International Society for Analytical Cytology, ISAC, Flow Cytometry Standard 2.0 files and stores the data in AdaSAGE tables. There are significant differences in the way manufacturers interpret and conform to Flow Cytometry Standard 2.0. AdaSAGE is employed to analyze and plot the data from multiple experiments. This data is used to assess the stability of flow cytometers. The initial release will be for DOS. The utilization of AdaSAGE, which is a flexible database tool, will facilitate subsequent development of other products.

The software engineer, whose previous professional experience was with C and C++, had very few problems with Ada syntax. The interface to the compiler and other tools was immature compared to those available for C++. The DOS text based user interface environment provided by AdaSAGE limited the functionality of the user interface. However, the present DOS 386 program can be directly ported to the newly released version of AdaSAGE for Microsoft Windows 95. Ada's strong type checking and package structure have significantly facilitated the development of the product.

Keywords: Software, Ada, Medical Device, Regulated, Good Manufacturing Practices, GMP, Flow Cytometry, Analytical Cytology. Database

1. INTRODUCTION

Clinical use of flow cytometry instrumentation requires that the measurement system be demonstrably proven to provide reproducible results. The Phoenix Flow, Flow Cytometry Quality Control program, FCQC, is a means to detect and monitor drift in the instrument settings. Stability in the data indicate that the results produced by the instrument have clinical validity. Conversely, instability indicates the necessity for intervention by either the service personnel or the primary operator.

1.1. Flow Cytometry Standard:

The International Society of Analytical Cytology, has created and maintained a data standard for Flow Cytometry Standard (FCS)[1]. This permits third parties to develop data analysis and other software that can work with the files created by commercial flow cytometers and cell sorters. The FCS is presently in its second revision (FCS 2.0)[2] and is scheduled to be revised (FCS 3.0). The present version of the FCQC program reads FCS 2.0 data. Unfortunately, FCS 2.0 is not a rigorous standard, and there is no validation suite to test compliance with the standard. FCS 2.0 includes in the same file both text in ASCII format and data, which can be in one or more of the following formats: ASCII, binary integer, or floating point. Any order of the bytes is allowed as long as it is specified by the value of the $BYTEORD key word. The file can have three types of text sections. It must start with a text HEADER which includes pointers to the beginning and end of two other ASCII formatted sections, TEXT and ANALYSIS. The HEADER also contains the byte addresses of the beginning and end of one or more DATA parts. Additional user defined parts can also be included. Lamentably, no provision for registering and adding these user defined parts was included in this standard, nor subsequently enacted by its sponsor, the International Society for Analytical Cytology. For instance, the TEXT part contains KEYWORDS and associated VALUES. "Keywords and their values may be of any length (number of bytes) except that they must contain at least one character (no nulls)." The standard FCS keywords begin with the $ character; user-defined, (mostly vendor-defined) keywords begin with some other character. The ANALYSIS part is typically appended after the data is acquired.

Table 1 shows the layout of a simple HEADER.

Table 1

FCS 2.0 HEADER

Version	Text Begin	Text End	Data Begin	Data End	Analysis Begin	Analysis End
FCS2.0	128	552	640	2687	2688	2900

2. Materials and Methods

The FCQC software is hosted on an Intel 386 or better PC running MS DOS 5.0 or latter. This software can be hosted on either a separate computer or on the computer which is part of the flow cytometer. In the latter case, the flow cytometry software will **not** be running simultaneously with the FCQC program. These are totally independent systems, which will only communicate by having the FCQC program read the Flow Cytometry Standard Files produced by the flow cytometer. The FCQC program does not produce data that can be read by a flow cytometer.

3. Software Development Methodology

3.1. Tools:

Both Ada[3] and a tailored version of Mil-STD-498[4] were employed. Since the FCQC project absolutely depended on being able to read FCS 2.0 data, this technologically challenging (highest risk) step was done first. This was accomplished by employing an already existing parser, AYAC[5,6] and an expert (JTS) on its use. Only the text data was used from the FCS files. This data was reformatted and stored with the AdaSAGE[7,8] database manager. The capacity to manipulate the binary data was deliberately omitted In order to minimize the regulatory burden[9].

3.2. Storyboarding:

The Ada_Med software design methodology of Iterative storyboarding[10,11] is based on the spiral model of software development[12]. The design of the human interface was rapidly prototyped by first developing the screens. The system was then implemented with the AdaSAGE screen generator and subjected to a detailed walk-through, which resulted in significant changes. It should be noted that the concept for the product was created by Phoenix Flow's management, who are not software engineers. The use of storyboarding permitted them to control the project and thus produce software which embodied their concepts.

3.3. Software Methodology:

PC software for DOS was written in Ada (ActivAda for Intel 386 and above, Thomson Software Products, San Diego, CA). The AYAC parser and the AdaSAGE database and screen generator employed for the FCQC program are reusable components. The use of AdaSAGE, which is a portable, relational database, will permit new features to be added easily and ports to other operating systems. The development process employed good software engineering practices including the use of a portable package for numerical types and organizing the software components into cohesive packages, which exhibit minimal coupling.

AdaSAGE is an Ada component; however, the AdaSAGE database and screen definitions, which are table driven, are not directly linked with the Ada code. Communication is by passing strings, such as field names, between the Ada program and AdaSAGE. The contents of the fields are passed as memory records, which can then have the values of their elements passed to Ada variables. Both the Ada source code and the AdaSAGE definition files had to be cross-referenced. The strings that were employed to communicate between the Ada code and AdaSAGE were stored as appropriately named Ada constants. The AdaSAGE database and screen definition forms include an alias field, which can be up to 312 characters in length, which is more than sufficient to included the name of the Ada package where the object is used. AdaSAGE generates reports, which describe the databases and the screens. The aforementioned alias field is included in these reports.

The FCQC screens shown below were captured with Hijaak Pro 3.0 under DOS (Inset Systems Inc., Brookfield CT.) and then imported into Hijaak Pro 3.0 under Microsoft® Windows 95. Whereever possible, the black and white pixels were reversed and the colored pixels were changed to black. Most of the screens have been cropped to show just the form.

4. Description of the FCQC Program

The FCQC program is divided into two parts. The setup procedure, which is run infrequently, requires knowledge of a PC's file system and should be performed by supervisory personnel, such as the flow cytometry laboratory managers. The actual use of the system for automated data entry (reading of the FCS 2.0 files) and report generation was designed to be very user friendly for ease of use by the cytometer operator.

4.1. Setup

The menu bar of the Setup program includes: File, Cytometer, Lot Number, Lab_Name, Supervisor Name, Printer, and Help. At present, the file menu is only used to exit the program. Each cytometer must be named and if there is more than one, the user selects one.

ADD CYTOMETER

NAME: FILE TYPE:
AMACIII   STANDARD FCS 1.0/2.0   NEW FILE TYPE

RAW DATA FILES DIRECTORY:
C:\QC\DATA   CHOOSE DATA DIR

ARCHIVE DIRECTORY
D:\QC\ARCHIVE   CHOOSE ARCH DIR

MANUFACTURE
Newport Instruments

MODEL: SERVICE PERSON NAME:
1.0   C. K. Becker

SERIAL NUMBER   SERVICE PHONE: EXT:
0000001   (619) 453-5095

OK   CANCEL

Figure 1a, Screen to add a cytometer. This is a redrawing of the original DOS screen, which did not reproduce well.

Two data paths are specified. The first is the directory that contains the raw data, and the second is the directory to be used for archiving, which should not be on the same disk drive as the original data. A network or removable drive can be specified.

The lot number entered in Figure 1b is that of the test particles employed to standardize the cytometer. The standard deviation obtained from the initial number of measurements is displayed on the quality control graphs (please see section 4.2.). The data entered in Figure 1c are appended to the Cytometer service text file. There are three additional menus in the Setup program. The first to enter the name and address of the laboratory. The second is to enter the supervisor's name. And the third to set up the printer.

```
ADD LOT NUMBER

CYTOMETER NAME:
 AMACIIIS

DATAPOINTS FOR STATISTICS:
 20

NEW LOT NUMBER:
 001

      [ OK ]          [ CANCEL ]
```

```
LOG SERVICE EVENT

CYTOMETER NAME:
AMACIII

DATAPOINTS FOR STATSTICS:
20

SERVICE NOTE:
This is a test.

      [ OK ]          [ CANCEL ]
```
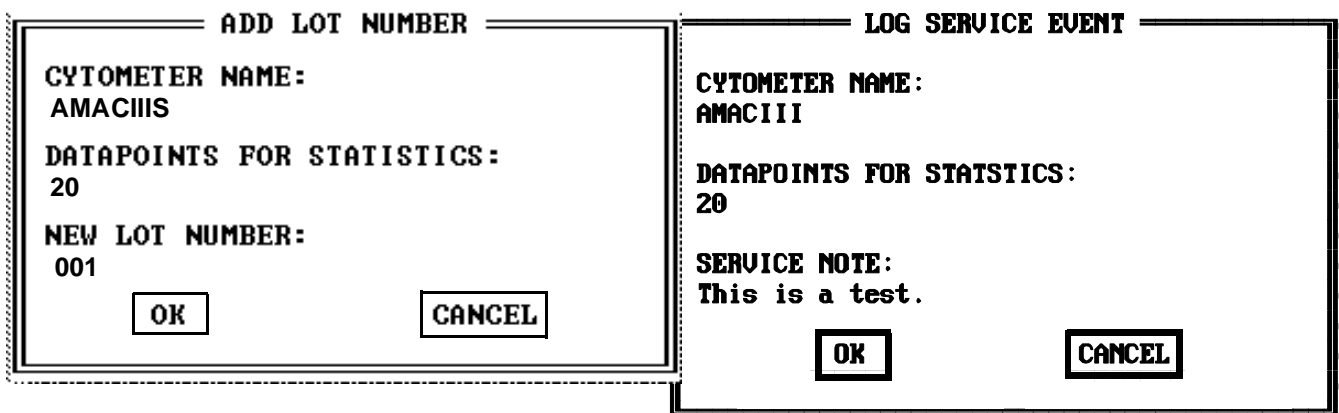
Figure 1b (left), Screen to specify the lot number and the initial number of data points to use for the standard deviation calculations. This screen was captured from the Windows 95 version of AdaSAGE.

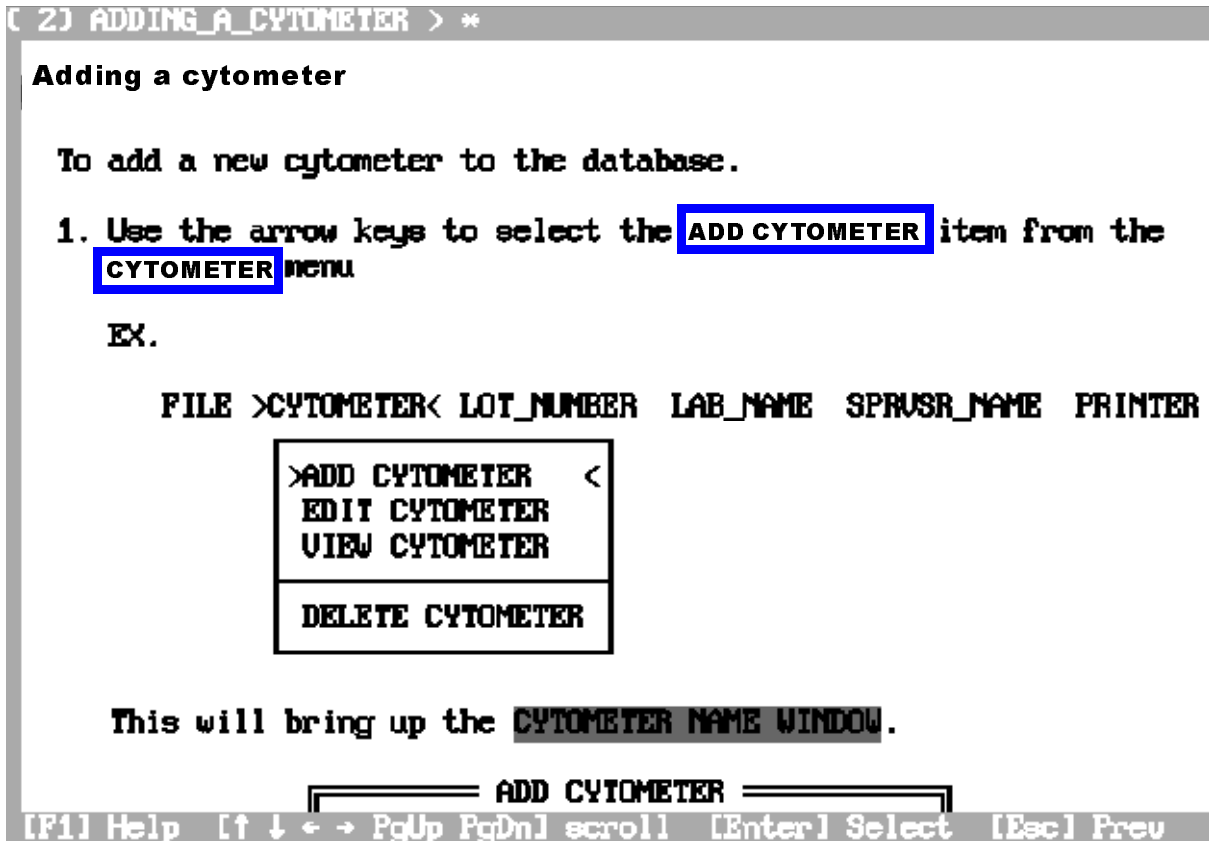Figure 1c (right), Screen to record servicing the cytometer.

Figure 1d,   Second level Help hypertext screen. The ADDING_A_CYTOMETER text block
was selected which resulted in this screen.

The setup program includes an extensive hypertext help system. The user scrolls through the help items (not shown) and selects a phrase. Figure 1d shows the screen that appears after Adding a cytometer has been selected. Only the top two thirds is shown. The first level help screen includes an alphabetic index of the subjects, which is also keyed to the hypertext help screens.

4.2. Flow Cytometry Quality Control Program

The first screen has four pull down menus: File, QC Reports, Comments, and Help. The File menu has two options, Add QC File and Exit. The user can either add single files or read in a complete directory. As is shown in Figure 2a, the path for the data files must be specified and the operator has to determine whether to add the files one by one or to add all of the new files in the directory. The parsing of the files and storage of the data into the AdaSAGE relational database is the most resource intensive part of the FCQC program.

```
FILE  QC_REPORTS  COMMENTS  HELP
Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow !!
Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow !!
Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow !!
Phoenix Flow Phoen┌══════════ ADD QC FILE ══════════┐ow Phoenix Flow !!
Phoenix Flow Phoen│                                 │ow Phoenix Flow !!
Phoenix Flow Phoen│ CYTOMETER:                      │ow Phoenix Flow !!
Phoenix Flow Phoen│ AMACIIIS                        │ow Phoenix Flow !!
Phoenix Flow Phoen│                                 │ow Phoenix Flow !!
Phoenix Flow Phoen│ DIRECTORY:                      │ow Phoenix Flow !!
Phoenix Flow Phoen│ C:\QC\TESTDATA                  │ow Phoenix Flow !!
Phoenix Flow Phoen│                                 │ow Phoenix Flow !!
Phoenix Flow Phoen│ FILE NAME:                      │ow Phoenix Flow !!
Phoenix Flow Phoen│ 00000575.H01    │SELECT FILE│   │ow Phoenix Flow !!
Phoenix Flow Phoen│                                 │ow Phoenix Flow !!
Phoenix Flow Phoen│ ADD ALL NEW FILES: Y            │ow Phoenix Flow !!
Phoenix Flow Phoen│                                 │ow Phoenix Flow !!
Phoenix Flow Phoen│  │ADD QC FILE│    │CANCEL│       │ow Phoenix Flow !!
Phoenix Flow Phoen│                                 │ow Phoenix Flow !!
Phoenix Flow Phoen└═════════════════════════════════┘ow Phoenix Flow !!
Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow Phoenix Flow !!
Phoenix Flo┌─────────────────────────────────────────────────┐
Phoenix Flo│CYTOMETER:  AMACIIIS    LOT NUMBER: 05_DEC_1995  │
Phoenix Flo└─────────────────────────────────────────────────┘
```

Figure 2a, ADD QC FILE Menu. Since ADD ALL NEW FILES is set to YES, all of the files in C:\QC\TESTDATA will be processed.

    After the data has been stored, the user can create reports. The first step is to pull down the QC REPORTS menu and select (Figure 2b) the length of the period. Time periods are measured from the present to the past. The content of the graph is determined (Figure 2c) by selecting the parameter to serve as the ordinate, adding a comment, and choosing if the graph will be displayed on the screen or be printed.

QC_REPORTS

```
DAILY SUMMARY
15 DAY
CURRENT MONTH
30 DAY
60 DAY
90 DAY
180 DAY
360 DAY
ONE YEAR
```

QC REPORT

START DATE:
1995/11/05

PARAMETER:
MEAN   CHANGE PARAM

ADD COMMENT

DISPLAY   PRINT

EXIT

Figure 2b, Left, QC REPORT PERIOD Menu.

Figure 2c, Right, QC REPORT Parameter, Comment, and Display Menu.

QC REPORT

```
START DA
  1995/11    PARAMETER:
             VOLTAGE
PARAMETE     GAIN
  VOLTAGE    MEAN          ARAM
             STD
             CV
             HALF CV
             PEAK CHANNEL

    DIS                    T
             <ENTER> = SELECT
             <ESC>   = CANCEL
```

START DATE: 12/11/1994
END DATE: 12/06/1995

DATE: 12/06/1995   TIME: 16:16:47

FL4 VOLTAGE

```
V   360
O   353
L
T   346
A
G   339
E
    332
      0   95  190  285  380
    DAYS FROM START DATE
```
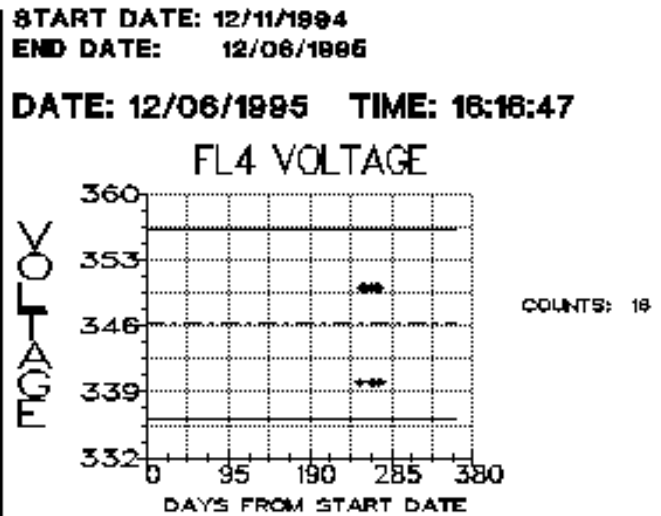
COUNTS: 16

Figure 2d, Left, QC REPORT Parameter selection.

Figure 2e, Right, Display of one of the six graphs. One year's (sixteen) measurements of the FL4 PMT voltage are graphed.

The following parameters can be monitored: laser power in milliwatts, laser current in amperes, voltages of all photomultipliers. Gains of all photomultiplier and other parameter detection amplifiers, sheath air pressure, test bead light scattering or fluorescence. The results are sorted by: cytometer and Lot Number. The means, peak channels, standard deviations (STD), and coefficients of variation(CVs), of the measured parameters are calculated and plotted. The results for each day, usually the means and CVs. are plotted against time. The peri-

ods for the graphs are: 5, 15, 30, 60, 90, 180, and 365 days. The positive and negative standard deviation for a preset number of initial measurements, usually the first 20, are presented as horizontal lines in Figure 2e.

4.3. Customer Support:

Ada has a very powerful and efficient exception handler. When an exception is raised, all error messages are first written to a log file and then the text produced by the exception is displayed on the screen. The log file, if the user so desires, can be sent to Phoenix Flow.

Since both Ada and AdaSAGE have range checking, out of range data can be detected and not accepted by the FCQC program. If possible, the FCQC queries the operator for instructions as whether to continue without that data point; and if that is not possible, the program exits.

The present Ada package structure and future use of the new child libraries will facilitate the addition of new capabilities to the present program and reuse of present software components in future products. The portability of Ada and AdaSAGE will permit this DOS implementation to be followed by a Microsoft Windows, and possibly X Windows and Macintosh implementations.

5. CONCLUSION

This project has demonstrated the utility of the use of storyboarding to prototype and specify a software product. Ada can be employed for main stream, commercial software products. Although the software engineer for this project (R.R.) had no previous experience with Ada, he had no serious difficulties with the language. The problems he had with the Ada language were not related to its syntax or design. The readability of his code was significantly enhanced by the use of Ada.

The extremely powerful tools supplied with the Thomson compiler; unfortunately, were not as user friendly as those supplied by vendors of compilers for other languages. The library management tools, which were overkill for our single programer project, will be replaced in the new Thomson Ada 95 compiler. AdaSAGE comprises both a good relational database and tool-set. However, its formal documentation is not at the same level as equivalent commercial products. This is to a large extent offset by the source code being provided with AdaSAGE. Our first experience with the AdaSAGE Windows binding indicates, as shown in Figure 1b, that the esthetics of the screens has been significantly improved, and that porting the FCQC program to Windows will require minimal effort.

6. ACKNOWLEDGMENTS

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