Computer Control of a Charge-Coupled Device in a Prototype Blood Analysis Machine

by

Laura E. Mendyke

Submitted to the Department of Electrical Engineering and Computer Science
in partial fulfillment of the requirements for the degree of
Master of Science in Electrical Engineering

at the

MASSACHUSETTS INSTITUTE OF TECHNOLOGY

May 1994

© Laura E. Mendyke, MCMXCIV. All rights reserved.

The author hereby grants to MIT permission to reproduce and distribute publicly paper and electronic copies of this thesis document in whole or in part, and to grant others the right to do so.
Computer Control of a Charge-Coupled Device in a Prototype Blood Analysis Machine

by

Laura E. Mendyke

Submitted to the Department of Electrical Engineering and Computer Science on May 13, 1994, in partial fulfillment of the requirements for the degree of Master of Science in Electrical Engineering

Abstract

This thesis details the implementation of a charge-coupled device (CCD) in a prototype blood analysis machine. In the machine, a blood tube is centrifuged to separate the blood into bands. Then the tube is illuminated, and the tube image is focused on the linear CCD, which contains a single row of optical elements called pixels. The pixels collect and store charge proportional to intensity of the light, and a "picture" of the tube image is output from the CCD chip as a series of analog voltage levels. This information is digitized and transferred to a PC, where it may be analyzed to determine the width of the blood bands, and thus obtain cell counts and present possible diagnoses. In addition, the CCD is used to read a bar code associated with the blood tube to support a patient identification system.

The CCD which was best suited for this application was selected. Circuit boards were constructed to send the CCD the required control signals, process the output of the CCD, and interface the CCD with the computer. Software was written to test and demonstrate the operation of the CCD, and data was acquired and analyzed.

This research was performed in association with Becton Dickinson, Inc., and the New Products Program at M.I.T.

Thesis Supervisor: David M. Otten
Title: Research Engineer
Acknowledgments

Thanks to Dave Otten for putting me on the project team, for providing a lot of technical help, for contributing a lot of software and electronics to the machine, and for being patient with me when I was confused, which was most of the time.

Thanks to Ben Linder for providing technical help, and for doing the layout of the CCD board. Thanks to Shin John Choi for designing the barcode, and to David Barrett for developing the software to read it.

Thanks to the rest of the MIT Project team for being dedicated and fun to work with: Gwen Barrett, Amy Battles, Don Lee, Karon McLean, Nanette Palmer, Andres Pieczanski, John Sieh, and Ming Wu.

Thanks to Mike Rosen for being our fearless leader for the first half of the project, and to Woodie Flowers for coming to our rescue during the second half.

Thanks to Brad Thomas from Becton Dickinson for believing in us, communicating with us, providing help and support, and in general being a great liason to BD.

Thanks to Mike Eurice, Paul Gaidis, and Ed Skevington from BD for providing help with and information about the BD electronics and software.

Thanks to Rudy Rodriguez from BD, and Will Durfee and Woodie Flowers from the MIT New Products Program for providing this opportunity.

Thanks to Wayne Hagman for taking me on as a research assistant for one term while I was writing this thesis and didn’t have time to do much actual research.

Thanks to my parents for supporting me, believing in me, and loving me.

And last, but certainly not least, thanks to my husband Michael for always being there for me, for putting up with me when I get frustrated, and for being understanding, caring, loving, and generally wonderful.

Research for this thesis was conducted within the MIT Laboratory for Electronic and Electromagnetic Systems.
To my husband, Michael
Contents

1 The MIT New Products Program

1.1 Project Team ........................................ 14
1.2 Project Definition .................................. 15
1.3 Funding ................................................. 15
1.4 Degree Program ....................................... 16
1.5 New Product Development Process ..................... 16
1.6 Benefits ................................................ 17
1.7 Communication ........................................ 18
1.8 Invention Ownership and Patent Rights .................. 18
1.9 Confidentiality ....................................... 19
1.10 Stanley Works ....................................... 19
1.11 Becton Dickinson .................................. 19
   1.11.1 MIT Project Team ............................... 20
   1.11.2 Communication ................................. 21
   1.11.3 Process ..................................... 21
   1.11.4 Market Research ............................... 22
   1.11.5 Results ..................................... 22
   1.11.6 Equipment and Facilities .................... 24

2 Hematology Analysis .................................. 25

2.1 Blood Analysis Technology ............................ 25
   2.1.1 Early Blood Analysis ............................ 25
   2.1.2 Impedance Counting ............................. 26
2.1.3 Quantitative Buffy Coat Analysis

2.2 Blood Tubes

2.2.1 E-Z-Prep QBC® Tubes
  2.2.1.1 Venous Tube
  2.2.1.2 Capillary Tube

2.2.2 The QBC® Vac-Q-Tube

3 QBCE Walkaway Prototype

3.1 Machine Specifications
  3.1.1 QBCC Autoread
  3.1.2 QBCC Walkaway

3.2 Machine Concepts

3.3 Low-End Market

3.4 Patient Identification
  3.4.1 Implementation
  3.4.2 Usage

3.5 Movement of Tubes Through the Walkaway System
  3.5.1 Centrifuge
  3.5.2 Optical Reading Station
  3.5.3 Results

3.6 User Interface Hardware

3.7 Alpha Prototype

4 Optics

4.1 Alternatives
  4.1.1 Photomultiplier Tube
  4.1.2 Photodetector
  4.1.3 CCD

4.2 Preliminary Optical Designs and Testing
  4.2.1 CCD Imager
<table>
<thead>
<tr>
<th>Section</th>
<th>Title</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>4.2.1.1</td>
<td>Calibration Tube Scans</td>
<td>52</td>
</tr>
<tr>
<td>4.2.1.2</td>
<td>Tube Bar Code Scans</td>
<td>53</td>
</tr>
<tr>
<td>4.2.1.3</td>
<td>Blood Tube Scans</td>
<td>54</td>
</tr>
<tr>
<td>4.2.2</td>
<td>Scanning Mirror</td>
<td>57</td>
</tr>
<tr>
<td>4.2.3</td>
<td>Fiberscan</td>
<td>58</td>
</tr>
<tr>
<td>4.3</td>
<td>Conclusions</td>
<td>61</td>
</tr>
<tr>
<td>5</td>
<td>CCD selection</td>
<td>64</td>
</tr>
<tr>
<td>6</td>
<td>Implementation of CCD Boards</td>
<td>75</td>
</tr>
<tr>
<td>6.1</td>
<td>CCD Printed Circuit Board</td>
<td>75</td>
</tr>
<tr>
<td>6.1.1</td>
<td>Analog-to-Digital Converter</td>
<td>77</td>
</tr>
<tr>
<td>6.1.2</td>
<td>Operational Amplifier</td>
<td>79</td>
</tr>
<tr>
<td>6.1.3</td>
<td>Buffers</td>
<td>80</td>
</tr>
<tr>
<td>6.1.4</td>
<td>Voltage Regulators</td>
<td>81</td>
</tr>
<tr>
<td>6.2</td>
<td>CCD Computer Interface Board</td>
<td>81</td>
</tr>
<tr>
<td>6.2.1</td>
<td>JDR PCB</td>
<td>81</td>
</tr>
<tr>
<td>6.2.2</td>
<td>Counters</td>
<td>83</td>
</tr>
<tr>
<td>6.2.3</td>
<td>SRAM</td>
<td>83</td>
</tr>
<tr>
<td>6.2.4</td>
<td>Buffers</td>
<td>83</td>
</tr>
<tr>
<td>6.2.5</td>
<td>Logic Gates and Registers</td>
<td>84</td>
</tr>
<tr>
<td>7</td>
<td>Operation of CCD Boards</td>
<td>85</td>
</tr>
<tr>
<td>7.1</td>
<td>CCD Inputs</td>
<td>86</td>
</tr>
<tr>
<td>7.1.1</td>
<td>Voltage Sources</td>
<td>86</td>
</tr>
<tr>
<td>7.1.2</td>
<td>Integration Time</td>
<td>87</td>
</tr>
<tr>
<td>7.1.3</td>
<td>CCD Clock</td>
<td>88</td>
</tr>
<tr>
<td>7.2</td>
<td>CCD Output</td>
<td>89</td>
</tr>
<tr>
<td>7.2.1</td>
<td>Processing</td>
<td>89</td>
</tr>
<tr>
<td>7.2.2</td>
<td>Storage and Retrieval</td>
<td>91</td>
</tr>
<tr>
<td>7.3</td>
<td>Communication</td>
<td>93</td>
</tr>
<tr>
<td>Section</td>
<td>Title</td>
<td>Page</td>
</tr>
<tr>
<td>---------</td>
<td>-------------------------------------------------</td>
<td>------</td>
</tr>
<tr>
<td>A.2.2</td>
<td>Patient ID Specifications</td>
<td>135</td>
</tr>
<tr>
<td>A.3</td>
<td>Software Interpretation of the Bar Code</td>
<td>136</td>
</tr>
<tr>
<td>A.4</td>
<td>Data</td>
<td>136</td>
</tr>
<tr>
<td>B</td>
<td>Band Length Calculation</td>
<td>139</td>
</tr>
<tr>
<td>B.1</td>
<td>QBC® Autoread</td>
<td>139</td>
</tr>
<tr>
<td>B.1.1</td>
<td>Tube Data</td>
<td>139</td>
</tr>
<tr>
<td>B.1.2</td>
<td>Software Algorithms</td>
<td>140</td>
</tr>
<tr>
<td>B.2</td>
<td>QBC® Walkaway Prototype</td>
<td>143</td>
</tr>
</tbody>
</table>
List of Figures

2-1 Venous E-Z-Prep Tube .......................................... 28
2-2 Capillary E-Z-Prep Tube .......................................... 28

3-1 Preliminary Prototype Machine ..................................... 35
3-2 E-Z-Prep Tube with Flag ........................................ 36
3-3 Prototype Carousels .................................................. 37
3-4 Prototype Machine Interior (Front View) ................................ 39
3-5 Prototype Machine Interior (Back View) ................................ 40
3-6 Prototype Machine Interior (View of Gripper Arm) ................................ 41
3-7 Blood Bands in a Venous E-Z-Prep Tube ................................ 44
3-8 Blood Bands in a Capillary E-Z-Prep Tube ................................ 44
3-9 Alpha Prototype of the QBC® Walkaway Machine ................. 45

4-1 CCD Imager ................................................................. 51
4-2 Test CCD Data from Green Stripe Calibration Tube ....................... 53
4-3 Test CCD Data from Black-and-White Calibration Tube ....................... 54
4-4 Test CCD Data from Tube Bar Code ....................................... 55
4-5 Test CCD Scan of a Normal Blood Tube (Red Fluorescence) ...................... 56
4-6 Test CCD Scan of a Normal Blood Tube (Green Fluorescence) ...................... 56
4-7 Scanning Mirror Design .................................................. 59
4-8 Fiberscan Design .......................................................... 60
4-9 Preliminary Prototype Optical Configuration .................................. 62
4-10 Alpha Prototype Optical Configuration ...................................... 63
5-1 Drawing of Sony ILX508 7926-Pixel CCD ............................. 73
5-2 Dimensions of Sony ILX508 7926-Pixel CCD .......................... 74

6-1 Block Diagram of the CCD Boards ........................................ 76
6-2 Schematic of the CCD PCB .................................................. 78
6-3 Optical Components of the Prototype System ............................ 79
6-4 Schematic of the CCD Computer Interface Board .......................... 82

7-1 CCD PCB Voltage Sources ..................................................... 86
7-2 Sony ILX508 7926-Pixel CCD Timing Diagram .............................. 90
7-3 Operational Amplifier Circuit ............................................... 91
7-4 Timing Diagram (Start) ....................................................... 100
7-5 Timing Diagram (Middle) .................................................... 101
7-6 Timing Diagram (End) ....................................................... 102

9-1 CCD Dark Output ............................................................. 112
9-2 CCD Dark Reference Pixel Output ......................................... 113
9-3 QBC® Walkaway Prototype Scan of a Green Stripe Calibration Tube (Green Fluorescence) ......................................................... 114
9-4 QBC® Walkaway Prototype Scan of a Green Stripe Calibration Tube (Green Fluorescence, Expansion) ................................................. 115
9-5 QBC® Walkaway Prototype Scan of a Green Stripe Calibration Tube (Transmission) ................................................................. 116
9-6 QBC® Walkaway Prototype Scan of a Black and White Calibration Tube ................................................................. 117
9-7 QBC® Walkaway Prototype Scan of a Black and White Calibration Tube (Expansion) ................................................................. 117
9-8 QBC® Walkaway Prototype Scan of the Bar Code Calrod .................. 118
9-9 QBC® Walkaway Prototype Scan of the Bar Code Calrod (Expansion) 119
9-10 QBC® Walkaway Prototype Scan of a Human Hair (Bar Code Background) ................................................................. 120
9-11 QBC® Walkaway Prototype Scan of a Human Hair (Level Background) 120
9-12 QBC® Autoread Cap Scan of a Normal Blood Tube .......................... 121
9-13 QBC® Autoread Plasma Scan of a Normal Blood Tube ........ 122
9-14 QBC® Autoread Transmission Scan of a Normal Blood Tube ..... 122
9-15 QBC® Autoread Red Fluorescence Scan of a Normal Blood Tube .. 123
9-16 QBC® Autoread Green Fluorescence Scan of a Normal Blood Tube . 123
9-17 Walkaway Transmission Scan of a Normal Blood Tube .......... 124
9-18 Walkaway Red Fluorescence Scan of a Normal Blood Tube ....... 125
9-19 Walkaway Green Fluorescence Scan of a Normal Blood Tube ...... 125
9-20 Walkaway Red Fluorescence Scan of a Normal Blood Tube (Expansion) 126
9-21 Walkaway Green Fluorescence Scan of a Normal Blood Tube (Expansion) 126
9-22 Walkaway Red Fluorescence Scan of a Normal Blood Tube (Better-Defined Buffy Coat Region) .......................... 127
9-23 Walkaway Green Fluorescence Scan of a Normal Blood Tube (Better-Defined Buffy Coat Region) .......................... 128
9-24 Walkaway Red Fluorescence Scan of a Normal Blood Tube (Expansion, Better-Defined Buffy Coat Region) .......................... 129
9-25 Walkaway Green Fluorescence Scan of a Normal Blood Tube (Expansion, Better-Defined Buffy Coat Region) .......................... 130
9-26 QBC® Walkaway Prototype Red Fluorescence Scan of an Altered Blood Tube .......................................................... 131
9-27 QBC® Walkaway Prototype Green Fluorescence Scan of an Altered Blood Tube .......................................................... 132
9-28 QBC® Autoread Red Fluorescence Scan of an Altered Blood Tube . 132
9-29 QBC® Autoread Green Fluorescence Scan of an Altered Blood Tube . 133
A-1 CCD Scan of a E-Z-Prep Flag Bar Code ............................. 137
A-2 CCD Scan of a Vac-Q-Tube Flag Bar Code ............................. 138
B-1 Packed Cell Layers in a QBC® Capillary Tube ...................... 141
B-2 Packed Cell Layers in a QBC® Venous Tube ....................... 142
List of Tables

3.1 QBC® Autoread Cell Count Parameters .................. 43
4.1 “Hand-calculated” CCD Imager and Autoread Cell Counts ........ 57
5.1 CCD Comparison Table: Physical Characteristics ................ 65
5.2 CCD Comparison Table: Electrical Characteristics ................ 66
5.3 CCD Comparison Table: Noise Characteristics ................ 67
5.4 CCD Comparison Table: Test Data ........................... 68
5.5 CCD Comparison Table: Ordering Information ................. 68
7.1 Communication Signals between the CCD PCB and the CCD Computer Interface Board ........................................ 94
7.2 Computer Bus Signals used on the CCD Computer Interface Board .... 96
7.3 Computer Port Addresses used in the CCD Subsystem ............ 96
9.1 QBC® Autoread Results for a Normal Blood Tube .................. 124
Chapter 1

The MIT New Products Program

Research for this thesis was conducted under MIT's New Products Program (NPP). Within this program, initiated in September, 1991[6], students and faculty from engineering and management disciplines are brought together with industrial sponsors to develop new products for sponsoring companies.

1.1 Project Team

An interdisciplinary project team involves faculty and students in the departments of Mechanical Engineering and Electrical Engineering and Computer Science, and the Sloan School of Management. Nominally, 3 Master's-level students and 3 faculty advisors, representing each of the three disciplines, form the project team. One faculty member serves as the principal investigator of the project, and is responsible for the completion of the project[6]. A Ph.D. student performs related research supervised by principal investigator[6]. Each of the core team of Master's-level students selects an aspect of the product prototype development as a thesis project; this helps to ensure that the project will be carried through to completion and archived in the thesis documents. Engineers and managers from the sponsoring company keep in constant contact with the MIT group and are considered part of the project team.

In addition to the four students directly funded by the project, the project also may attract undergraduates and additional graduate students who have independent
support and are looking for design experience or a thesis project[4].

1.2 Project Definition

Contact is made with a potential sponsoring company through the Office of the Dean for Engineering, the Industrial Liaison Program, the Technology and Licensing Office, and through individual faculty members. Representatives of the NPP visit the company to formally present and discuss the program[6].

The sponsoring company defines the general form of the prototype product, and cooperates with the selected MIT project team to work out the details of the product[6]. In order to use the NPP to their best advantage, the sponsoring company must give careful consideration to selecting a project. An incremental upgrade to an existing product may be more effectively done within the company, as many of the tools and designs are already available. However, with a new technology or new design style, collaboration with MIT offers many advantages to the company[4].

The length of the development cycle is set at two years, which is the length of the typical engineering Master’s degree program. If the estimated development time of the product is more than two years, the sponsoring company may divide the design responsibilities between MIT and the company engineers to reduce the scope of the project[4]. An incomplete or hurriedly-built product is not beneficial either to MIT or to the sponsoring company.

Prototype products developed by the MIT project team are not sidelines or pure research projects. Products developed through the NPP are expected, both by MIT and by the sponsoring companies, to be put on the market soon after the completion of the prototype by the MIT project team[4].

1.3 Funding

The project is funded by the sponsoring company as a sponsored research contract with the principal investigator of the project team[4]. The sponsoring company pro-
vides full-time support for four graduate students in the form of Research Assistantships (RAs), and part-time support for three faculty supervisors. The company also contributes materials, equipment, and supplies related to the development of the prototype product.

Within MIT, the Dean of Engineering, the Technology Licensing Office, and the Associate Provost for the Arts have allotted resources to the NPP. Further support for the New Products Program comes from endowment gifts donated to support faculty chairs and graduate fellowships.

1.4 Degree Program

The NPP is compatible with existing graduate study and research programs, so there are no special admissions requirements for the program. Master's-level students enter MIT graduate school through the standard admissions process, and must complete the course work and required by the department. The students are funded as research assistants to work on the project, and their Master's theses provide documentation and a design record for the product.

1.5 New Product Development Process

An overall objective of the NPP is to research methods of improving the new product development process. The different projects pursued by NPP teams have the common result of adding to a knowledge base of the design process. The research of the Ph.D. student funded by the sponsoring company is generally tangential to the development of the prototype product; the Ph.D. student works with the rest of the team to study general aspects of the design process. These may include the coordination of productive "brainstorming" sessions; the organization of product and market segment information; the prototyping of user interfaces; and the communication between remote design groups working on the same project. This information is useful to the sponsoring company as well as to the NPP.
1.6 Benefits

It is felt by the coordinators of the NPP that everyone involved—faculty, students, and the industrial sponsor—benefits from participation in the program.

Over its two-year span, the project covers marketing, detailing and manufacturing aspects of product design—a wider spectrum than may be taught in a classroom or found in a pure research project[4]. Students involved with the program gain real world product design experience as well as an MIT technical education. Mechanical Engineering Professor Michael Rosen states that MIT is “changing the paradigm for developing a product.”[11] With traditional design methods, each group handles a narrowly-focused piece of the project without consideration to how the pieces fit together. In the New Products Program, each student concentrates on one area of expertise, but all members of the project team participate in every aspect of the design, from start to finish: market and customer need identification, detailed product definition, concept development and selection, overall design, detailed design, and manufacturing considerations[4].

The opportunity to test theoretical concepts on real design projects is also an important benefit for faculty members of the team. Aspects of the project may also be used as examples in an engineering design or marketing class. However, the program may be more appealing to tenured professors. Tenure, and promotion for junior faculty, is generally based on research publications leading to widespread recognition, which is not a primary outcome of NPP participation[4].

The sponsoring company receives a new product prototype at the end of the project. In addition, the company benefits from the technical expertise available at MIT which may lead to new process insights. Some companies may also perceive the cooperative effort as a means to establish good relations with MIT and improve their ability to recruit MIT graduates for employment[4].
1.7 Communication

The success of the program depends on good communication between MIT and the sponsoring company to create the product prototype[4]. Each side of the collaboration has important contributions to make. The company provides market and customer information, feedback about proposed designs, technical knowledge about its existing products, and resources for use in building and testing the prototype product. The MIT faculty organize the project and facilitate communication between the project contributors, adding their own technical expertise and providing references to other resources at MIT. The MIT graduate students provide fresh viewpoints, intelligent thought, and hard work[4]. If possible, communication should involve company engineers periodically participating in design meetings and working sessions at MIT, and members of the MIT team visiting the company’s engineering laboratories.

Communication between all members of the MIT project team is also critical. However, it is often difficult to accomplish, because of the varying office locations and busy schedules of faculty and students. Short meetings scheduled once or twice a week and supplemented by communication via electronic mail (E-mail) are effective in keeping all team members fully informed[4]. E-mail is also useful in keeping a record of the design process. Engineers from the sponsoring company may be included in the group’s E-mail “mailing list,” so that they may see and contribute to design discussions and answer technical questions.

1.8 Invention Ownership and Patent Rights

In the creative design environment fostered by the NPP, it is likely that original, patentable ideas will be generated. The NPP policy on invention is straightforward: “Company invents, company owns; MIT invents, MIT owns; joint invention, joint ownership.”[4] If the sponsoring company uses a patented design owned by MIT in the final product, exclusive licensing rights are given to the sponsoring company, who agrees to pay MIT a royalty based on product sales. Within MIT, the distribution
of these royalties is covered by a formula which applies to all revenue-generating inventions[4]. An equitable agreement must be arrived at jointly by the company and MIT.

1.9 Confidentiality

Generally, a company wants to restrict or control the flow of information about a project under development. In contrast, universities are interested in free exchange of information. A compromise must be made in the interests of the NPP. Only the most general information about the projects in the program is released to publicize the program[4]. In addition, students' theses are held back from general distribution until the company authorizes the release of information.

1.10 Stanley Works

When the NPP was started in September, 1991, two projects were initiated[4]. The first project to commence was sponsored by Stanley Works of New Britain, Connecticut[4], and involved a new product for powered nailing[6]. Professor Karl Ulrich was the principal investigator for this project, and Professors Will Durfee and Woodie Flowers were co-supervisors. The technological expertise and original thinking of the MIT project group led to a new approach to the tool, which should give a competitive advantage to Stanley when the product is released. Stanley may use the technology to create an entire family of new products[4].

1.11 Becton Dickinson

The second NPP project was sponsored by Primary Care Division of Becton, Dickinson and Company (BD) in Baltimore, Maryland[6]. The proposed product was a new-generation blood analysis machine which combined functionalities of current BD products[4].
1.11.1 MIT Project Team

At the beginning of the project, Mechanical Engineering Professor Michael Rosen was named principal investigator. The supervisors were Principal Research Engineer David Otten in the Department of Electrical Engineering and Computer Science, and Professor William Qualls in the Sloan School of Management. The Ph.D. student in Mechanical Engineering working under Professor Rosen was Karon MacLean; her research involved rapid prototyping of product user interfaces[6]. At BD, Brad Thomas was named senior engineer in charge of the project, and stayed in close contact with the MIT group to determine the product specifications.

The other students on the project team included one sponsored Master’s student from each of the departments of Mechanical Engineering and Electrical Engineering and Computer Science, and one from the Sloan School of Management; three Mechanical Engineering Master’s students and one Sloan School graduate student with independent funding; two undergraduate students in Mechanical Engineering; and one undergraduate student in Mechanical Engineering working with Ms. MacLean on the user interface design. As students involved in the project graduated or moved on over the two year period of the project, the composition of the team changed considerably; however, the size and apportionment of the team remained essentially constant.

In the summer of 1993, principal investigator Dr. Rosen made the decision to accept a position at the University of Tennessee. Although he still remained in contact with the team and continued to supervise his students’ work, a new principal investigator was needed for the project. Professor Woodie Flowers was named to the position. As a coordinator of the NPP and a supervisor for the Stanley Works project, Professor Flowers already had a working knowledge of the project format, and led the team very effectively.
1.11.2 Communication

The team held weekly meetings and communicated through an E-mail mailing list. During the design phase of the project, the weekly meetings often became brainstorming sessions for new designs; later, the meetings were used primarily to give progress reports on each subsystem of the project to the group.

The E-mail forum was used primarily to ask questions and schedule working meetings for sections of the team. All of the team members were on the mailing list, including Brad Thomas of BD, and several MIT faculty members involved in the NPP[1]. In general, any communication among members of the group was directed at the entire mailing list, so that everyone would be aware of current problems, results, and ideas, and could offer comments and suggestions. Much of this E-mail was archived in a computer locker created for the group, to provide a reference and a history of the project. Since it was often difficult, due to schedule differences, for team members to meet in person, E-mail proved to be a valuable resource.

Approximately every six months, a delegation of officials from BD visited MIT for a formal presentation of the project status. Each member of the MIT team would present one of the subsystems of the product, and a discussion session would be held afterward. Brad Thomas visited more frequently, occasionally bringing a small group of project engineers. These visits took the form of “working meetings” rather than formal presentations, and were beneficial to the project.

1.11.3 Process

All of the students on the team for the marketing phase of the project received some exposure to customer opinion through participation in customer interviews.

The design phase of the project involved many brainstorming sessions. Concepts from tiny hand-held systems to huge machines which could process more than a hundred samples at a time were generated. Each subsystem—patient ID, human interface, blood separator, material transport, and tube scanner—was carefully discussed using customer information and technical data, and building tradeoff charts[1].
There were no strictly demarcated "phases" of the project. The design process continued throughout the project; however, there was a shift from a heavy emphasis on design at the beginning of the project to a heavy emphasis on building and testing at the end of the project. Market research also continued through a good part of the project.

1.11.4 Market Research

The original project specifications called for a high-end machine which would be used in hospital laboratories and large physician practices. However, new market research caused a shift in direction in February, 1993[1]; customer opinion was probably influenced by concern over the new presidential administration's health care policy. The previous design concepts, focusing on high throughput with a priority interrupt capability, were discarded, and new designs of a smaller, simpler, and less expensive nature were developed. However, the optical subsystem of the machine remained largely unaffected by this change in priorities.

1.11.5 Results

This project was one of the first tests of the New Products Program. On the whole, it was successful; however, this project did have some problems which were caused partly by inexperience with the new program, and partly by unforeseeable events.

- The major setback of the project was the fact that the target market segment changed more than halfway through the project, and a large amount of design work was abandoned.

- The scope of the project was more complex than any of the original negotiators realized; it was a lot of work for a student group of this size to complete in the required time, even if the original product specifications had remained constant.

- Although the NPP was initiated in September, 1991, a design team was not formed for this project until November, 1991. Late in the term, it was more
difficult to recruit incoming graduate students, and the goal of gathering a core
team of graduate students who would stay with the project for the entire two-
year duration was not met. Two of the Master's students on the project team
graduated before the end, and another one was not hired until several months
into the project. The turnover rate for undergraduates on the team was even
higher. Only one student was involved with the project for its entire duration.

- The turnover was not limited to students, as the principal investigator, Professor Rosen, left the project about halfway through the two-year span, and was replaced by Professor Flowers.

The MIT project ended in November, 1993. Due to the problems listed above,
the final form of the project was an "alpha" prototype, which is functional but not in the exact form of a finished product, instead of a production prototype, which is ready to be put into production. BD still needs to put a significant amount of work on the product, and it consequently won't be out on the market for another couple of years. A BD product team is currently working on evaluating the MIT design, and they have indicated that they plan to carry the product through to completion.

Despite the minor setbacks, the BD officials agree that the project has been a success and that they would consider collaboration with MIT again. One BD engineer noted that if the MIT team hadn't been working on the product, nothing would have been accomplished on it in the past two years, as no BD engineers were available to work on the the product at that time; by that measure, BD has a two-year head start on the product. However, the BD engineers will be picking up somebody else's design; they weren't closely involved with the design phase of the project, and they won't be as immediately familiar with the MIT design as they would have been with their own concept.

A BD newsletter quotes Rudy Rodriguez, the Vice President of Research and Development at BD's Primary Care Division, as saying about the project: "MIT wanted a real-life project they could bring in-house, not an academic exercise. Becton Dickinson wanted access to the best minds and brightest new ideas in product
engineering. This partnership gives us access to all the intellectual resources of MIT, from all disciplines. It’s a win-win situation.”[3]

Two mechanical design concepts which came out of the project are in the process of being patented by MIT.

1.11.6 Equipment and Facilities

All work on this project was done in the Laboratory for Electromagnetic and Electronic Systems at MIT. Standard equipment such as oscilloscopes, power supplies, function generators, logic probes, and some discrete and IC components, were available in the lab. A 486DX 33 MHz PC-AT was loaned to the project by BD. Special purpose components were selected and ordered from outside vendors. Funding for these purchases was provided by BD.
Chapter 2

Hematology Analysis

2.1 Blood Analysis Technology

Two primary technologies exist for blood analysis: quantitative buffy coat analysis and impedance counting methods.

2.1.1 Early Blood Analysis

In 1852, physician Karl Vierordt obtained good results with a method of diluting blood and counting the cells under a microscope[9]. Although accurate, this method was extremely time-consuming.

In the 1920s, centrifuged blood tubes—tubes which had been spun at high speed so that the different blood cells separated into layers by density—were used to aid in the diagnosis of many diseases[9]. Looking at the relative amounts and the appearance of packed, or centrifuged, red blood cells, white blood cells and plasma in the blood could give important insights into a patient’s health, even though the technology was far from definitive. However, until recently, accurate optical measurement of cell layers in a centrifuged tube was not feasible.
2.1.2 Impedance Counting

Impedance counting analyzers make up the bulk of the hemotology analysis machines currently on the market[1]. In these machines, blood is diluted with chemicals and passed through a tiny wire ring. The current induced in the wire is proportional to the cell volume, and the cell type and number of cells may be determined[9].

Impedance counting instruments are extremely accurate. However, they are also very expensive and require considerable daily maintenance. Due to their complexity, they must be operated by a skilled technician[1]. These factors make such a machine impractical for a small physician practice which requires only a small number of analyses per day; such practices generally send out their blood samples to larger laboratories. BD’s customer surveys have shown that if it were possible to get accurate results in-house using a smaller, low-maintenance, inexpensive machine, many physician practices would be interested in purchasing the machine.

2.1.3 Quantitative Buffy Coat Analysis

Quantitative Buffy Coat (QBC®) Analysis is a patented method for hematology analysis, developed in the early 1980’s by Stephen Wardlaw and Robert Levine[9]. A narrow cylindrical tube about three inches long is filled with blood and centrifuged so that the blood cells separate into layers, or bands, according to the density of the cells. The percentages and absolute counts of the different cell types in the blood are determined by measuring the length of each band.

Important diagnostic information is contained in the white cells of the buffy coat, which comprises about one percent of the total blood count, and forms a narrow region between the dense red blood cells and the lighter plasma in a centrifuged tube[1]. So that the lengths of the bands formed by the different white blood cell types may be measured more accurately, the region is expanded to about ten times its original length by insterting a plastic float into the tube. The float is of the same density as the buffy coat and of a slightly thinner diameter than the inside of the tube[9]. Even with the float expansion, the length of the buffy coat is only a small fraction of the
tube length. The machine's optical equipment must be sensitive enough to measure the band lengths within this narrow region accurately.

QBC® machines are inexpensive, easy to operate, and require only occasional maintenance. One drawback of the QBC® technology is that for good results the blood tubes must be centrifuged immediately, and may only be held for 4 hours, and only in a cool, dark area, before optically processing, or reading them.

Since with the QBC® method the blood never leaves the tube, it is a cleaner and safer alternative to impedance-counting machines. Small physician practices can save time and money by using a QBC® machine rather than performing manual blood counts under a microscope or sending samples to an outside laboratory.

2.2 Blood Tubes

There are two different sizes of QBC® blood tubes: the E-Z-Prep and the Vac-Q-Tube. Each tube is coated with dry anticoagulants to prevent blood clots, and with acridine orange to cause certain blood cells to fluoresce at specific wavelengths of excitation light. Once prepared, a QBC® blood tube should be centrifuged immediately. If a centrifuged tube cannot be analyzed immediately, it should be stored in a cool, dark area for no more than 4 hours.

2.2.1 E-Z-Prep QBC® Tubes

E-Z-Prep tubes are 3 inches long and 0.088 inches in diameter[16]. There are two different types of E-Z-Prep tubes: the capillary tube, and the venous tube. Each of these has different markings and preparation techniques.

2.2.1.1 Venous Tube

The venous blood tube is shown in Figure 2-1. To fill the venous tube, the venous blood is first drawn into a large collection tube called a VACUTAINER®. Venous tubes are filled from the VACUTAINER® using a pipetter. The venous tube is filled so that the blood level is within ±1 mm of the fill line. A closure is seated on the end
of the tube after filling, and a float is inserted, bringing the blood level to between the two indicator lines. The nominal fill volume of the venous tube is 111.1 μL[2].

2.2.1.2 Capillary Tube

The capillary blood tube, shown in Figure 2-2, is filled directly from a finger puncture. The capillary action of the narrow tube draws drops of blood from the finger surface into the tube. The capillary tubes are filled from the end of the tube nearest the two fill lines, until the blood level is between these two lines. A closure is seated on the other end of the tube after filling, and a float is inserted in the tube, bringing the blood level from the closure to between the two fill lines. The fill volume of the capillary tube is between 55 and 65 μL[16].

2.2.2 The QBC® Vac-Q-Tube

The Vac-Q-Tube is a new blood tube being developed by Becton Dickinson. It is a smaller, standalone version of the VACUTAINER®, which will be processed by newer QBC® machines.

The Vac-Q-Tube necessarily will have a larger diameter (3-4 mm[16]) than the E-Z-Prep tube, so that the vacuum force of the evacuated tube pulls a sufficient amount of blood into the tube. The length of the Vac-Q-Tube will be close to that of the E-Z-Prep tubes, and the fill volume of the Vac-Q-Tube will be about 500μL.

This tube is an appealing alternative for blood collection, with considerably less
exposure to blood-borne pathogens than either pipetting or finger puncturing. The new Vac-Q-Tube used with the clean, in-tube QBC® blood analysis technique hopefully will result in safer health care.

However, since the Vac-Q-Tube requires a larger amount of blood, the E-Z-Prep may be a better choice for seriously ill patients or very young children. Furthermore, if several blood samples are needed for different tests, filling venous tubes by pipetting from a single VACUTAINER® may be a more desirable option.
Chapter 3

QBC® Walkaway Prototype

This chapter discusses the development of the prototype product concept, and describes the overall operation of the machine.

3.1 Machine Specifications

3.1.1 QBC® Autoread

In the determination of specifications for the prototype product, the QBC® Autoread, the latest in BD’s line of QBC® hematology instruments, was often used as a benchmark.

The Autoread processes, or reads, one centrifuged blood tube at a time; the centrifuge is a separate machine[2]. A medical technician must centrifuge a circular tray, or carousel, of blood tubes, then transfer the tubes one at a time into the Autoread for analysis. Centrifugation takes about 5 minutes, and reading one tube in the Autoread takes about one minute, so a considerable portion of the technician’s time is wasted standing by the instrument and waiting for the next step.

When a tube is inserted into the Autoread, it is transported to an optical reading station. The tube is illuminated through a slit and moved along its longitudinal axis past a single photodetector, which outputs a signal corresponding to the intensity of the illuminated portion of the tube which is imaged onto the photodetector[2].
3.1.2 QBC® Walkaway

The proposed new machine, the QBC® Walkaway, would process an entire carousel of tubes from centrifugation through reading without any further input from the medical technician. It would also be capable of processing a new and more convenient type of tube. The Walkaway system, incorporating centrifuge, reader, and transport mechanisms into a single machine, would be a marked improvement in convenience and capability over the Autoread.

A set of product specifications was determined by BD and the MIT project group. A partial list of the machine specifications follows:

- Fully automated; no user input required between carousel load for centrifuge and cell count output
- Capable of processing 1 tube in 5.5 minutes, and 10 tubes in 7 minutes
- Capable of processing two different tube sizes: the E-Z-Prep (including two different tube types, with different markings), and the Vac-Q-Tube, which is under development
- Capable of reading band lengths down to 0.002 inches with accuracy ±0.0005 inches
- Accuracy and reliability comparable to, or better than, the QBC® Autoread
- Reliable patient identification (ID) system

The focus of this thesis is the method chosen to read the centrifuged tubes in the prototype machine. It was decided to use a linear charge-coupled device (CCD), a linear array of optical sensing elements called pixels, instead of the single photodetector used in the Autoread. A currently-available CCD could maintain the resolution of the Autoread. Furthermore, CCD technology is rapidly improving, and it should be possible to upgrade the product easily. In addition to determining blood cell counts, reading a blood tube in the prototype machine includes interpreting a bar code associated with the tube, using the CCD, as part of a patient ID system.
This thesis details the process of integrating the CCD into the prototype machine. This process included the selection of a suitable CCD, and the design, implementation, and testing of hardware and software to control the CCD.

3.2 Machine Concepts

3.2.1 High-End Market

At the beginning of the design phase, the target market segment was high-end users: hospitals, laboratories, and large physician practices. These users needed a machine with a large tube capacity, with special features such as "STAT/INTERRUPT" to immediately stop the processing of one batch of tubes to allow another higher-priority batch to be run, and auto-disposal of tubes. At least two centrifuge rotors were needed in the high-end machines to yield the desired throughput of tubes.

One design, proposed by Amy Battles, was called the "Jukebox." The medical technician would stack filled carousels in the machine, and each carousel would automatically be transferred to one of the two centrifuge rotors as rotors became available. After centrifugation, the carousels would be moved to an optical reading station. The tubes could either automatically be disposed in an internal container, or retained and output with the carousel. This proposed design was ultimately rejected by BD as being too large.

Another design, proposed by Don Lee, was called the "Dual Rotor," and contained two centrifuge rotors and a single reading station. Both centrifuge rotors would be able to operate at once. When a centrifuge finished spinning, the carousel would automatically be moved to the optical reading station; if the reading station was occupied, the carousel would be held in the centrifuge until the reading station became free. The ratio of two centrifuges to one reading station was generally accepted as being optimal; two centrifuges would reduce the amount of time that a medical technician might have to wait for a free rotor to centrifuge a tray of tubes, but duplicating the optics would be unnecessary and expensive. This design was accepted
by BD, and work was begun on implementing the Dual Rotor concept.

Some intermediate designs which evolved during brainstorming sessions were not presented. One of these was a “starfish” configuration, proposed by principal investigator Dr. Rosen. This design would have a central reading station and several centrifuges around the outside. Tubes would be entered either one at a time through a bin or in carousels, and individual tube transport would take place around the circumference of the machine using a belt. The machine would automatically load tubes into carousels, ensuring an optimal tube distribution. When a carousel was filled, or after a certain amount of time had elapsed for a partially-filled carousel, it would be centrifuged. The tubes would then be transported individually to the reading station, where each tube would be analyzed. Although it is an interesting concept, this design was generally felt to be too large and complex for this particular project. In addition, there was also concern that tubes might break during the transport.

One design issue which generated considerable discussion in the project team was the loading of the machine. In a front-loading machine, the carousel of tubes would be inserted into a slot like a CD into a player; in a top-loading machine, a lid on the top of the machine would be opened, and the carousel would be placed on the rotor inside. A front-loading design would simplify some mechanical issues, and allow the machine to take up less vertical space. However, a top-loading design would allow the inside of the machine to be more accessible for cleaning. Eventually, a top-loading design, which appeared to be the user preference, was selected.

3.3 Low-End Market

Design was well underway for a high-end system based on the Dual Rotor concept when there was an abrupt change in focus. New market research had shown that high-end users were not sufficiently interested in the product. The new target market segment for the QBC® Walkaway was the low-end segment: smaller physician practices of 3–5 physicians.

This change in focus necessitated a change in direction of the design. The Dual
Rotor design was extensively revised, and a scaled-down version eventually became the basis for the single-rotor design that was implemented as the prototype.

One of the changes to the design was the elimination of the STAT capability. This feature had always been a subject of debate: if the centrifugation process is interrupted, the blood cannot be read or re-centrifuged, so the entire carousel of tubes must be discarded. In most cases, STAT means “within five minutes,” so there would be time for the current centrifugation to finish. An “INTERRUPT” feature, which would stop the process after completing the centrifuge cycle so that a new carousel could be inserted immediately, was implemented.

The auto-disposal feature was also eliminated. Medical technicians had liked this feature because it would minimize their contact with the blood tubes. However, if the machine reports an error or an unexpected result for a tube, the medical technician may want to visually inspect the tube for incorrect float insertion, wavy bands, or a cracked, chipped, or marked tube. Returning the tubes in the carousel allows them to be viewed by the medical technician if desired; otherwise, the carousel may just be tipped over a wastebasket.

The resulting prototype machine had one rotor, one read station, and no STAT or auto-dispose capability. This allowed the mechanical design was much simpler, with fewer motors (all of the movement in the system may be accomplished with three motors) and fewer degrees of freedom. The preliminary prototype of the machine, shown in Figure 3-1, was completed and presented on May 24, 1993.

3.4 Patient Identification

3.4.1 Implementation

A fundamental objective of the QBC® Walkaway system is the ability to analyze several tubes in a batch, without user input. A reliable patient ID system is essential to this process. Previous QBC® machines have left the responsibility of patient ID to the user: slots in the QBC II and World Class centrifuges are numbered, and
the medical technician must keep track of the slot number corresponding to each patient’s tube. The new Walkaway machine will have numbered centrifuge slots for convenience, but provision for a more rigorous patient ID system was desired.

During the design phase of the project, there was considerable discussion about how the patient ID system should be implemented. Associating a bar code with the tube was accepted as being the most universal and effective. The form of the bar code is described in Appendix A.

Initially it was thought that bar codes could be laser-etched directly on the tubes, but this proved to be too expensive, and the laser printing was not of high enough quality.

A bar code sticker which would be affixed directly on the tube by the medical technician was considered. A sheet containing one large and one small sticker with the same bar code would come with each tube. The larger bar code sticker would be attached to the patient’s medical records, and the smaller one would be fastened to the tube. However, in order to fit on the tube and not obscure too much information, the tube bar code sticker must be very narrow; this tiny sticker also must be aligned
with the tube in order to be read by the optics. This would be a difficult task for a busy medical technician wearing gloves.

The "flag" concept, shown in Figure 3-2, was developed to solve these problems. The flag is a flat moulded plastic part which caps both ends of the tube and extends off the side of the tube like a flag on a flagpole. A large bar code sticker will be attached to the flag, and a smaller sticker with the same bar code may be affixed to the patient's medical records and read with an external scanner.

The reverse side of the flag, which faces up when the tubes are placed in the carousel, will be writeable so that the patient's name (or other information) may be inscribed on it. This will enable the medical technician to find a tube belonging to a particular patient at a glance.

A prototype flag has been tested under centrifuge conditions without noticeable bending of the flag or leakage of the tube.

Two prototype carousels holding E-Z-Prep tubes and Vac-Q-Tube approximations with and without flags are shown in Figure 3-3.

### 3.4.2 Usage

There are several different ways in which the patient ID system may be used.
No patient ID: No flags would be used. The medical technician would be responsible for associating each patient’s name with the position number of that patient’s blood tube in the carousel. The output cell counts for each tube in the batch would be numbered in the same fashion (determined by the “home” position of the carousel).

Basic patient ID: Flags would be used. The bar code number would appear on the output cell counts. The medical technician would be responsible for matching that bar code with the corresponding patient’s name, either from the flag (if the name is written on the reverse side), or from the patient’s medical record, to which a bar code sticker should be affixed.

Enhanced patient ID: At some point during the machine cycle, the medical technician would scan the bar code on each patient’s medical record into the machine using an external bar code scanner. The technician would then enter the patient’s name, and other relevant information such as the patient’s age and gender. This information may be used in the software to make informative di-
agnoses, since the normal ranges for cell counts vary with these factors. The patient’s name would appear along with the bar code number on the results output.

The medical technician would not necessarily enter information about a patient while the machine is processing that batch of tubes. One alternative is that the system would store the information until the technician prompted it for the results by reading in the appropriate bar code with the external scanner. It would not be necessary to enter the patient’s name with this option; the next record to be printed out or displayed would be that of the selected patient. However, it would be beneficial to enter the patient’s name and other information.

When the patient’s name is linked to the corresponding results in the system, this opens up the possibility of creating a history file for each patient, in which all blood test results for the patient would be stored. This would be particularly useful if the QBC® machine were linked to a computer or interfaced to the hospital’s Laboratory Information System (LIS).

Top-of-the-line patient ID: It may be possible to install a bar code scanner in the printer which prints the output results. To obtain the results for a particular patient, the medical technician would load the patient’s medical record, with the attached bar code sticker, into the printer. The printer would read the bar code and print the patient’s cell counts on the same page. With a self-loading printer, the machine could have complete “walkaway” capability. All of the medical records from the current batch would be placed in the paper tray at the same time that the carousel of tubes was placed in the centrifuge. Without any user input, the results from each tube would be printed on the appropriate patient’s record after the batch was processed. (However, if diagnostic information is desired, some patient information would need to be entered into the machine before the results were printed.)
3.5 Movement of Tubes Through the Walkaway System

Two interior views of the prototype machine structure are shown in Figures 3-4 and 3-5. The following sections give an overview of the processing of blood tubes by this system.

3.5.1 Centrifuge

The medical technician initiates machine operation by placing a carousel holding up to ten tubes in the centrifuge rotor, closing the lid, and pressing a “start” button. After this point, the machine processes the tubes without any user input (although the sequence may be stopped immediately at any time by pressing an “abort” button).

The centrifuge is moved up into the lid to seal a “floating” (vibration isolated) cover[1]. In the QBC II and World Class centrifuges, the user must manually fix a cover in place. If the rotor cover is left off, tube breakage can occur. In the
Walkaway, which combines the centrifuge and optics in one machine, the problem of tube breakage is even more serious. If the centrifuge rotor is not sealed, the optics may be contaminated if tube breakage or leakage occurs. The built-in cover of the Walkaway prototype eliminates this possibility.

The centrifuge spins at 12,000 RPM for about 5 minutes, exerting a centrifugal force of 14,387 g at the rim. Because the centrifuge structure vibrated at the fundamental frequency of the centrifuge top speed, making the spin cycle extremely noisy, the prototype centrifuge was not brought to full speed in normal operation (tubes were spun in the World Class Centrifuge during testing). However the prototype centrifuge was tested at full speed, and with different manufacturing procedures and materials, the noise level will be reduced in the final product.

### 3.5.2 Optical Reading Station

When the centrifuge stops, the rotor moves down to disengage from the lid, and then transports the carousel horizontally to the optical reading station.
At the reading station, the carousel is scanned once to determine which tube types the carousel holds (there are different carousels for E-Z-Prep tubes and Vac-Q-Tubes, due to the different tube sizes), and which slots contain tubes. Two sensors positioned above the carousel scan the carousel and locate tubes using black-capped posts as optical references as the carousel is sequenced, or spun about its center in the horizontal plane.

After the initial scan, the carousel is sequenced so that the first tube is in position to be read. A “gripper arm” moves down, and a pair of “fingers” pick up the tube and bring it to the level of the optics. A photo of the prototype machine displaying the gripper arm is shown in Figure 3-6. Several different scans, or readings, are taken of the tube by the CCD, using different light sources and colored filters:

**Transmission** The light source for this scan is a long fluorescent bulb. The light shines through the blood tube, and the tube image is focused through a lens onto the CCD. The transmission reading is used to determine the tube type, the fill volume of the tube, and the position of the float in the tube.
Bar code  If a flag is attached to the tube, a bar code scan is taken using the transmission lamp. The tube is indexed, or rotated about its longitudinal axis, until the bar code on the flag is visible to the CCD. The resulting scan data is used in the software to determine the bar code number associated with the tube.

Fluorescence  The light source for these scans is a linear fiberoptic array positioned directly above the blood tube, at an angle of 90° to the CCD. Light from the fluorescence lamp passes through a blue filter (480 nm) before illuminating the tube. A blue light source, such as a blue light-emitting diode (LED), was considered for the fluorescence light source; however, blue LEDs are not commonly available yet.

The light from the tube passes through another filter before being focused on the CCD. This filter serves to clarify certain features in the blood. At this time, two different filters are used to view the tube image: red (620 nm center) and green (540 nm). However, some planned future assays will require different wavelengths of light. A linear variable filter, which incorporates wavelengths varying over the visible spectrum, is used in the prototype. To add more fluorescence readings with different filter wavelengths, only the machine software must be updated.

The tube is indexed eight times, rotating 45° about its longitudinal axis each time, for each set of fluorescence scans. The indexing is done by a lead screw, which shifts the tube horizontally by a small amount for each rotation. At each tube position, one scan is taken at the red filter position, and one at the green filter position. In the software, the results of the eight sets of scans are averaged together to reduce the effects of wavy or skewed bands. One or more scans may be “unreadable” because of such anomalies as a blood clot in the tube, dirt on the tube surface, or the flag blocking the blood bands. At least four scans must be readable in order for results to be produced[2].

The fluorescence scans are used to determine the lengths of the bands of different
<table>
<thead>
<tr>
<th>Parameter</th>
<th>Unit</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>HCT</td>
<td>%</td>
<td>Hematocrit: percentage of red blood cell volume to total blood volume</td>
</tr>
<tr>
<td>HG</td>
<td>g/dL</td>
<td>Hemoglobin: percentage of hemoglobin in the cell makeup</td>
</tr>
<tr>
<td>MCHC</td>
<td>—</td>
<td>Mean corpuscular hemoglobin concentration: unitless ratio of hemoglobin/hematocrit</td>
</tr>
<tr>
<td>PLT</td>
<td>$10^9$/L</td>
<td>Platelet count</td>
</tr>
<tr>
<td>WBC</td>
<td>$10^9$/L</td>
<td>White blood cell count</td>
</tr>
<tr>
<td>GRANS</td>
<td>$10^9$/L</td>
<td>Granulocytes (absolute count)</td>
</tr>
<tr>
<td>GRANS %</td>
<td>%</td>
<td>Granulocytes (percent of WBC)</td>
</tr>
<tr>
<td>LYMPH/MONO</td>
<td>$10^9$/L</td>
<td>Lymphocytes and monocytes (absolute count)</td>
</tr>
<tr>
<td>LYMPH/MONO %</td>
<td>%</td>
<td>Lymphocytes and monocytes (percent of WBC)</td>
</tr>
</tbody>
</table>

Table 3.1: QBC® Autoread Cell Count Parameters

When a tube has been fully scanned, the arm places the tube back into the carousel. The carousel is then sequenced to the next tube position, and the process is repeated.

### 3.5.3 Results

After each tube in the carousel has been scanned, the rotor is moved back to the centrifuge position to return the carousel of tubes to the user.

The results of the tests, cell counts and diagnostic information, may be viewed on the screen or printed on a connected printer. The Walkaway will have the capability to calculate all of the Autoread parameters[2] shown in Table 3.1. The blood cell bands corresponding to these parameters in the capillary and venous tubes are shown in Figures 3-7 and 3-8.

Since the Walkaway will have a full-sized liquid crystal display (LCD) screen, there will be more options for displaying results. It will be possible, for instance, to display plots of the different scans, or to graph a series of results over time.
Figure 3-7: Blood Bands in a Venous E-Z-Prep Tube

Figure 3-8: Blood Bands in a Capillary E-Z-Prep Tube
3.6 User Interface Hardware

The prototype machine display is a Datalux flat screen, grayscale LCD monitor, and there is a compact keyboard for data entry. Both the display and the keyboard are connected to a 486/33 PC in the prototype; in the final product, an internal microprocessor will control the machine. An American Microsystems external bar code scanner is used to scan in tube bar codes; it is connected to the PC through the keyboard, making it appear that the numbers corresponding to scanned-in bar codes are entered from the computer keyboard.

3.7 Alpha Prototype

The alpha prototype of the QBC® Walkaway Machine was completed and presented on November 22, 1993. A foam core “skin” was constructed for the prototype to represent the form of the final machine. The finished prototype is shown in Figure 3-9.
Chapter 4

Optics

4.1 Alternatives

4.1.1 Photomultiplier Tube

A photomultiplier is a very sensitive optical detector for low light levels. A photosensitive cathode produces electrons when exposed to light, and this current is amplified through a progressive chain in the vacuum tube in which more electrons are generated in each stage. This type of detector is very good for detecting and amplifying images at low light levels, and has little noise.

However, there are a few drawbacks in this type of detector: it requires several inputs at high voltages to control the different stages of the amplifier; it is very sensitive to magnetic fields; and the optical sensitivity of the device is such that it may be destroyed if it is exposed to room light while powered on.

4.1.2 Photodetector

The QBC® Autoread uses a silicon photodetector[2] to capture optical tube data for processing. The tube is moved along its longitudinal axis, perpendicular to the light source. 5000 readings are taken by the photodetector along the length of the tube for transmission scans; other scans do not cover the full length of the tube, so fewer readings are taken with the same interval.
This method produces good data with little noise; since only one photodetector is used, there are no fabrication-related differences between readings taken at different points along the tube. However, there are some disadvantages. Motion is required for each scan. Higher tube resolution can be obtained by taking more readings along the tube, but this is limited by the optics and the accuracy of the motor moving the tube. In addition, moving the tube back and forth adds to the process time.

4.1.3 CCD

A CCD array is composed of a number of optical elements, called pixels, or photosites, which are generally $7\mu m$–$13\mu m$ on an edge. In a linear array, several thousand photosites are arranged in a line, yielding an optical detection area a few centimeters long and a few micrometers wide. When light shines on a pixel, free electrons are generated in the photosite. This charge is then transferred to an analog shift register and output from the CCD as an analog voltage[10]. The amount of time for which the CCD pixels collect charge is called the integration time.

The output voltage corresponding to each pixel is proportional to the intensity of the light shining on it, and to the integration time of the CCD\(^1\) If the illuminating light is too bright, or the integration time is too long, the output of the CCD saturates—reaches a maximum value—and does not contain any information. Similarly, if the integration time is too short, or the light levels are too dim, the CCD output is too low to detect any information. It is necessary to adjust the integration time or the intensity of the light source (or both) to position the CCD output in the center of the range.

Most CCDs use two banks of shift registers to transfer the collected charge from the photosites to the output. These registers are located on either side of the CCD array; the charge packets from adjacent photosites are transferred to opposite shift registers. The use of multiple shift registers helps to electrically isolate adjoining

---

\(^1\) There is some question as to whether increasing the integration time is exactly equivalent to increasing the intensity of the light source. It is possible that longer integration times provide more opportunity for noise to enter the system.
pixels and reduce charge leakage. However, there may be some slight fabrication differences between the shift registers which produces a built-in offset between the odd and even pixels. If separate output amplifiers are used, the gain of the amplifiers may be slightly different as well. Therefore, the offset between the odd and even pixels may be a function of the output voltage.

The CCD output always contains static noise, due to slight fabrication differences between individual pixels. The dark voltage level of the CCD is a function of the integration time, and may be measured in a calibration test and subtracted from the output during normal operation. However, the gain—the relationship between light intensity and generated charge—of the individual pixels may differ slightly, so that the noise increases with increased light intensity or longer integration times, and this is more difficult to correct. Charge leakage between pixels, or blooming, may also be a problem. Blooming is likely to happen when the CCD is at or near saturation. If one region of the CCD is saturated but another region is within normal light intensity levels, excess electrons from the saturated portion may “spill over” to non-saturated pixels, blurring and obscuring information in the non-saturated region. Some CCDs contain anti-blooming circuitry; excess charge in the photosites is diverted to a sink voltage ($V_{\text{sink}}$) instead of to the shift registers. This acts as a clipping circuit for the output [10].

To obtain the same resolution as the QBC© Autoread, a linear CCD array of at least 5000 pixels is needed. CCD arrays of this size are easily available, with a wide price range starting at about $150 for small quantities. CCDs with more than 6000 pixels are more expensive and harder to find. However, CCD technology is constantly being improved; there is a large demand for CCD components in the manufacture of optical devices such as scanners and cameras. If the Walkaway uses CCD technology, the development of better and higher-resolution CCDs should allow the machine to be easily upgraded.
4.2 Preliminary Optical Designs and Testing

During the summer of 1992, the project team breadboarded and tested three preliminary functional optical designs. Three of the core team members became design champions for the different designs; the design champion for a design was in charge of the testing and presentation of that design.

The mechanical components of the designs were machined from metal, and mounted onto an optics table. The optics table is a metal tabletop which has screw sockets at regular intervals, which facilitates the arrangement and alignment of optical equipment.

Each design used a different approach for reading blood tubes. Two of the designs used a CCD for light detection; the third used a photomultiplier tube. A 2591-pixel Texas Instruments CCD and the associated evaluation board used in these tests were loaned to MIT by BD. The evaluation board contained the control logic and clocks for the CCD; the output of the board was an analog voltage waveform, corresponding to the light intensity across the CCD, which could be viewed with an oscilloscope.

Since the 2591-pixel test CCD could not give the required resolution of 5000 points across the tube, the optics were configured to image about half of a tube onto the CCD. The optics focused on the float area of the tube; if the bands of the buffy coat can be detected and measured by the optics, measurement of the other, larger, bands should not be a problem.

BD also contributed a data acquisition package called DAPL, produced by Microstar Laboratories. The package included a PCB which was installed in one of the PC’s expansion bus slots. Digital or analog signals were sent to or read from the board through ribbon cables. DAPL’s software allows the user to create output trigger signals and voltage waveforms, read analog and digital data from the input into a file, and process and plot the data.

Although the CCD evaluation board and the DAPL package were useful in the preliminary testing, there were several considerations which prevented the use of a prepackaged CCD interface and data acquisition system in the final prototype.
It was found that the CCD evaluation board was too large to be integrated and aligned easily with the optics. The integration time was set manually, within a range of about 10 ms–250 ms, by adjusting a potentiometer on the board and measuring the resulting output on an oscilloscope; this would be unacceptable for the prototype product, in which all systems would be controlled from the computer. The data output of the board was an analog voltage waveform; this signal would travel through an off-board cable, probably introducing excess noise into the data, before it was converted to digital data and read into the computer.

The DAPL package was versatile, but it was not well-suited to this particular application. Most of the DAPL functions process data on the order of seconds or milliseconds; however, the output rate of the CCD is on the order of microseconds. Also, the CCD must be carefully controlled. For example, some early testing showed an unexpected amount of noise in the test CCD output. A trigger signal from the DAPL PCB was being used to clock out data from the CCD. Immediately following the trigger pulse, the DAPL board would read the CCD output, a voltage level corresponding to a single pixel. Immediately after the trigger pulse, though, there was some ringing in the output as it settled; a short time later, the output was much more stable. Delaying the data acquisition with respect to the trigger signal would greatly reduce the noise in the CCD output data; however, this was not supported by DAPL. A simple circuit to achieve the delay was built by Ben Linder.

4.2.1 CCD Imager

The CCD Imager, championed by Ben Linder and also worked on by Laura Mendyke, was a straightforward optical design. The blood tube was placed horizontally in a holder, and a lens focused the tube image on the linear CCD. The transmission lamp was a fluorescent bulb backed by a parabolic mirror, which was in a straight line with the tube, lens, and CCD. The fluorescence lamp was a linear fiberoptic array illuminated through a fiberoptic cable by a variable-intensity light source; the linear fiberoptic array illuminated the tube through a blue filter from the top, at a right angle to the lens and CCD. A diagram of the CCD Imager design is shown in Figure 4-1.
Figure 4-1: CCD Imager
Filters used in fluorescence readings were positioned between the lens the CCD. Single-wavelength filters were used for the preliminary testing; the red and green filters were added to or removed from the assembly as necessary.

In later tests, a linear diffraction grating replaced the multiple single-wavelength pass-through filters used in the fluorescence scans. A diffraction grating reflects light of different wavelengths from different points along its surface. However, the addition of the diffraction grating complicated the straight-line optical path of this design. Light from the fluorescence light source had to strike the diffraction grating at the correct angle and position to reflect the desired wavelength of light on the blood tube for each reading. In addition, the diffraction grating greatly reduced the light intensity of the fluorescence light source, which was already low.

Another problem was that the transmission lamp was much too bright for tube readings; it caused the CCD to saturate even with the shortest possible integration time. This problem was remedied by blocking much of the transmission light with a translucent piece of plastic in front of the bulb, but this gave a “fuzzy” quality to the light which blurred the tube image.

Conversely, the fluorescence lamp, light from which passed through a fiberoptic cable and two filters, needed to be set at maximum intensity in order to yield usable data, even with integration times as long as 100 ms. It was desirable to keep the integration times as short as possible, in order to reduce noise in the data.

DAPL plots of the CCD output were analyzed. Scans were taken of two BD calibration tubes and a tube bar code, as well as of blood sample tubes. In the output data, note that only the middle portion of each tube is focused on the CCD. A higher value of output data represents a brighter image, and a lower value a darker one.

4.2.1.1 Calibration Tube Scans

The green stripe calrod is the size of an E-Z-Prep tube, and has narrow grooves of width 0.004 in. which are filled with green coloring, etched around the circumference of the tube at intervals of 0.25 in. The black-and-white calrod is also the size of an
Figure 4-2: Test CCD Data from Green Stripe Calibration Tube

E-Z-Prep tube, and contains precise, alternating rods of black and white plastic. The length of each rod approximates the length of the blood cell band corresponding to its position in the tube. Data from these calibration tubes is shown in Figures 4-2 and 4-3.

4.2.1.2 Tube Bar Code Scans

Data was also taken of a clear bar code, approximately 0.75 in. × 0.25 in., attached to an E-Z-Prep tube. This test was done to evaluate the feasibility of reading a tube with a bar code printed on or affixed to the tube, before it was found that other considerations prevented this implementation. When the bar code was attached to a blood tube, it was determined that when the bar code faced the CCD, the reading of the bar code was not affected by the blood bands behind it, and when the bar code was on the other side of the tube, the reading of the blood bands was not affected by the bar code behind them. The bar code and the corresponding CCD data are shown in Figure 4-4.
4.2.1.3 Blood Tube Scans

Finally, data was taken of centrifuged blood tubes in transmission, green fluorescence, and red fluorescence lighting. Data from red and green fluorescence scans is shown in Figures 4-5 and 4-6. The float edge and buffy coat are clearly visible in these plots.

The CCD data taken with this optical configuration has many imperfections. The peaks in the green calrod data and the black-and-white calrod data should have flat tops and should all be the same height; it seems the tube image may not have been correctly focused on the CCD. The bar code data should also have clean edges and a uniform height, but the excessive noise in this data is probably primarily due to the poor print quality of the bar code. There is some rolloff in intensity at the ends of the CCD in transmission readings, which is partly due to the malfocused tube image and the edge of the lens. There is significant noise in the data, which is partially attributable to the blocking of the transmission light source, to the tube image focus, and to noise in the long cable carrying the analog CCD signal to the
Figure 4-4: Test CCD Data from Tube Bar Code
Figure 4-5: Test CCD Scan of a Normal Blood Tube (Red Fluorescence)

Figure 4-6: Test CCD Scan of a Normal Blood Tube (Green Fluorescence)
Table 4.1: “Hand-calculated” CCD Imager and Autoread Cell Counts

<table>
<thead>
<tr>
<th>Parameter</th>
<th>CCD Imager</th>
<th>Autoread</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total WBC</td>
<td>$7.21 \times 10^9$/L</td>
<td>$7.0 \times 10^9$/L</td>
</tr>
<tr>
<td>Granulocytes</td>
<td>$3.9 \times 10^9$/L</td>
<td>$4.8 \times 10^9$/L</td>
</tr>
<tr>
<td>% Granulocytes</td>
<td>54.1%</td>
<td>69%</td>
</tr>
<tr>
<td>Lymph/Mono</td>
<td>$3.31 \times 10^9$/L</td>
<td>$2.2 \times 10^9$/L</td>
</tr>
<tr>
<td>% Lymph/Mono</td>
<td>45.9%</td>
<td>31%</td>
</tr>
<tr>
<td>Platelets</td>
<td>$235.87 \times 10^9$/L</td>
<td>$233 \times 10^9$/L</td>
</tr>
</tbody>
</table>

However, even with these problems, the data was usable, and there were high expectations that much better data could be obtained using better lenses and light sources, a carefully-selected CCD, and an interface board specifically tailored for this project.

To further evaluate the success of this design, cell counts were estimated using CCD data from a blood tube in transmission, and red and green fluorescence lighting. The results are inexact, as the band lengths were measured “by hand,” by counting the number of pixels before the approximate band interface. The cell counts were calculated using the band length values and the QBC® Autoread algorithms discussed in Chapter 8. The same blood tube was tested in the Autoread for comparison; the “hand-calculated” cell counts and the Autoread outputs are shown together in Table 4.1.

4.2.2 Scanning Mirror

The Scanning Mirror, championed by Amy Battles and also contributed to by Ming Wu, also used a CCD for light detection, although the approach was different. The tube image was projected through a 12 μm (or smaller) slit onto a diffraction grating, which reflected and scattered the different wavelengths of light from the tube onto the linear CCD. The output of the CCD would then represent continuous color information for one point along the tube. This process would be repeated for every point along the tube; scans for each wavelength of interest could then be pieced together.
by selecting the data corresponding to the appropriate pixels from each of the CCD scans. The Scanning Mirror design is shown in Figure 4-7.

A major advantage of the Scanning Mirror design is that all of the wavelength information needed for present and future assays would be taken and stored. However, the design proved to be too complex for this machine. Data must be taken at intervals of 0.0005 inches over the 3-inch length of the tube to exhibit the desired resolution. Therefore, the CCD output must be read 6000 times for each tube, as opposed to once for each required wavelength; and it is unlikely that the number of wavelengths required will ever approach 6000. The time needed to read each tube would probably exceed an hour; not only is this an unacceptable waiting time for results, but as the blood tube is continuously exposed to light for this time, much of the information in the blood will be lost. The evaluation time for each tube would increase, as 6000 scans would be pieced together to compute the cell counts. The memory needed to store the information from 6000 CCD scans would exceed 10 Mb, which is unacceptably large.

Another concern with this design was that the resolution depends on the size of the slit through which the tube is focused on the diffraction grating. Upgrading the machine to a higher resolution depends on the precision with which a smaller slit may be manufactured. In addition, the alignment of this optical configuration was painstaking. Finally, it was found that much more light was collected from scans of the ends of the tube than of the center. This would make producing a single coherent tube reading, which would clearly show the relative intensities of the bands and the band interfaces, extremely difficult. Due to all of these complications, no usable data was collected with the Scanning Mirror design.

4.2.3 Fiberscan

The Fiberscan, championed by Don Lee, used a PMT for light collection. A parabolic mirror behind a blue halogen light source illuminated one end of a fiberoptic cable; the other end illuminated the blood tube. Another fiberoptic cable collected the light from the tube and projected onto a diffraction grating. Light was reflected from the
Figure 4-7: Scanning Mirror Design
diffraction grating to the PMT, which output a signal to the data acquisition circuitry. The tube was moved along its longitudinal axis, perpendicular to the ends of the two fiberoptic cables. The Fiberscan design is shown in Figure 4-8.

This design seemed simple and straightforward. However, the implementation required an unexpectedly large number of parts, including a scanning monochromator and controller, a precision rail and optical fiber carriage, an optical encoder, a photomultiplier tube, and a drive motor and controller. These parts increased to the projected cost of the machine. In addition, the large number of moving parts and degrees of freedom in the design would make the machine difficult to manufacture and assemble.

The optical path in the design included two fiberoptic cables, which greatly reduced the light intensity. In addition, the signal from the PMT in the test design was unacceptably noisy, although this was probably the fault of the implementation. Due to these difficulties, no usable data was obtained from this design.
4.3 Conclusions

The CCD Imager was found to be the most promising optical configuration of the three preliminary designs. The preliminary testing showed that a linear CCD design was feasible, and would have several advantages over a photodetector or a PMT design. When it was decided that a CCD would be used in the prototype, the process of selecting the CCD which would best suit the needs of the project was begun.

The optical configurations of the preliminary and alpha prototypes are shown for reference in Figures 4-9 and 4-10.
Figure 4-9: Preliminary Prototype Optical Configuration
Figure 4-10: Alpha Prototype Optical Configuration
Chapter 5

CCD selection

CCD chips from many different manufacturers were considered. The project budget restricted spending on the CCD to around or less than $1000, so many expensive, high-end CCDs were eliminated. In order to achieve the same tube resolution as the QBC® Autoread, a CCD having at least 5000 pixels was needed.

Comparison tables of important CCD characteristics were made for the seven CCDs evaluated which fit the price and resolution requirements[10, 17, 8, 12, 13, 14, 15]. Only preliminary data sheets were available for a few of the CCDs, so the comparison tables are incomplete for many characteristics.

The CCD comparison tables are divided into five parts: physical characteristics (Table 5.1), which includes the length and number of pixels; electrical characteristics (Table 5.2), which includes the saturation voltage and dynamic range; noise characteristics (Table 5.3), which includes the pixel non-uniformity; test data (Table 5.4), which specifies the conditions under which the electrical and noise characteristics were determined; and ordering information (Table 5.5), which includes the price and availability.

The terms used in the tables are defined as follows[10]:

**Antiblooming** Causes excess charge to be diverted to $V_{sink}$ instead of shift registers; acts as a “clipping circuit.”

**Channel offset** DC difference between the outputs of two analog shift registers
<table>
<thead>
<tr>
<th></th>
<th>LORAL CCD-191DC</th>
<th>THOM-SON TH 7508BCC</th>
<th>KODAK KLI-5001F</th>
<th>SONY ILX501</th>
<th>SONY ILX504B</th>
<th>SONY ILX506</th>
<th>SONY ILX508</th>
</tr>
</thead>
<tbody>
<tr>
<td># pixels</td>
<td>6000</td>
<td>5184</td>
<td>5000</td>
<td>5000</td>
<td>5000</td>
<td>5000</td>
<td>7926</td>
</tr>
<tr>
<td># dark ref. pixels</td>
<td></td>
<td>16</td>
<td></td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>80</td>
</tr>
<tr>
<td>size (µm)</td>
<td>10 × 10</td>
<td>7 × 7</td>
<td>7 × 7</td>
<td>7 × 7</td>
<td>7 × 7</td>
<td>7 × 7</td>
<td>7 × 7</td>
</tr>
<tr>
<td>pitch (µm)</td>
<td>10</td>
<td>7</td>
<td>7</td>
<td>7</td>
<td>7</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>length (cm)</td>
<td>6.0</td>
<td>3.6288</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
<td>5.5482</td>
</tr>
<tr>
<td># shift registers</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td># data output pins</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>window thickness (mm)</td>
<td>0.686</td>
<td>1.75*</td>
<td>0.75</td>
<td>0.8</td>
<td>0.8</td>
<td>0.8</td>
<td>0.8</td>
</tr>
<tr>
<td>window ind. of refraction</td>
<td></td>
<td></td>
<td></td>
<td>1.5</td>
<td>1.5</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>window options</td>
<td>AR coated</td>
<td>quartz</td>
<td>standard, multi-layer anti-reflection coated or photo-tic filtered</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*distance between external surface of window and top of photosensitive element.

Table 5.1: CCD Comparison Table: Physical Characteristics
<table>
<thead>
<tr>
<th></th>
<th>LORAL CCD-191DC</th>
<th>THOMSON TH7808BCC</th>
<th>KODAK KLI-5001F</th>
<th>SONY ILX501</th>
<th>SONY ILX504B</th>
<th>SONY ILX506</th>
<th>SONY ILX508</th>
</tr>
</thead>
<tbody>
<tr>
<td>supply (V)</td>
<td>17.0, 6.0, 2.0, -1.0</td>
<td>15.0, 13.0, 3.0, 1.8</td>
<td>15.0, 11.0, 5.0, 3.0, 0.75</td>
<td>9.0, 5.0</td>
<td>9.0, 5.0</td>
<td>9.0, 5.0</td>
<td>9.0, 5.0</td>
</tr>
</tbody>
</table>

# clocks  12  9  5  2  2  2  2

<table>
<thead>
<tr>
<th>range (Vp-p)</th>
<th>3.0</th>
<th>0.9</th>
<th>1.0</th>
<th>0.8</th>
<th>1.0</th>
<th>1.0</th>
<th>1.0</th>
</tr>
</thead>
<tbody>
<tr>
<td>DC offset (V)</td>
<td>10.0</td>
<td>10.0</td>
<td>9.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>max. data output (MHz)</td>
<td>5</td>
<td>16</td>
<td>25</td>
<td>25a</td>
<td>20 / 12a,b</td>
<td></td>
<td></td>
</tr>
<tr>
<td>output impedance (Ω)</td>
<td>1000</td>
<td>500</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>saturation voltage (V)</td>
<td>3.0</td>
<td>0.9</td>
<td>2.2</td>
<td>0.8</td>
<td>1.0</td>
<td>1.0</td>
<td>1.5</td>
</tr>
<tr>
<td>sat. equ. exp. (μJ/cm²)</td>
<td>0.5</td>
<td>0.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>dynamic rangec</td>
<td>3000</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>dynamic rangef</td>
<td>15000</td>
<td>6000</td>
<td>2000g,e</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>responsivity (V/μJ/cm²)</td>
<td>6.0</td>
<td>1.6</td>
<td>7.0</td>
<td>3.85g,h</td>
<td>8.75g,i</td>
<td>31.5, 73.5g,j,k</td>
<td>31.5, 73.5g,j,k</td>
</tr>
<tr>
<td>wavelengths (nm)</td>
<td>500–1000</td>
<td>400–1100</td>
<td>350–1000</td>
<td>400–1000</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>peak sensitivity (nm)</td>
<td>700</td>
<td>750</td>
<td>700</td>
<td>500</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

| antiblooming? | yes | no  | no  | no  | no  | no  | no  |

a max. input clock frequency
b 10 without external RS clocking, 6 with external RS
c with respect to peak-to-peak noise
d calculated from $V_{sat}(c^e) / D_{SNR}(c^e)$
e clocking and electronics noise is taken into account
f with respect to rms noise
g use conversion 1 μJ/cm² = 3.5 lx·s (valid for 2854 K light source at 700 nm)
h 1.1 V/lx·s
i 2.5 V/lx·s
j first measurement with 3200 K light source, second with 2854 K light source
k 9 V/lx·s / 21 V/lx·s

Table 5.2: CCD Comparison Table: Electrical Characteristics
<table>
<thead>
<tr>
<th></th>
<th>LORAL CCD-191DC</th>
<th>THOMSON TH7808BCC</th>
<th>KODAK KLI5001F</th>
<th>SONY ILX501</th>
<th>SONY ILX504B</th>
<th>SONY ILX506</th>
<th>SONY ILX508</th>
</tr>
</thead>
<tbody>
<tr>
<td>noise (mV&lt;sub&gt;p-p&lt;/sub&gt;)</td>
<td>1.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>noise (mV&lt;sub&gt;rms&lt;/sub&gt;)</td>
<td>0.15&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>noise equ. exp. (pJ/cm&lt;sup&gt;2&lt;/sup&gt;)</td>
<td>30</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DC channel offset (V)</td>
<td>0.5</td>
<td>0.2</td>
<td>0.025</td>
<td>0.015</td>
<td>0.001</td>
<td>0.001</td>
<td></td>
</tr>
<tr>
<td>AC gain mismatch (mV)</td>
<td>50</td>
<td>50</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CTE</td>
<td>0.999999</td>
<td>0.99998</td>
<td>0.99999</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TTE</td>
<td>0.951</td>
<td>0.95</td>
<td>0.95</td>
<td>0.92</td>
<td>0.92</td>
<td></td>
<td></td>
</tr>
<tr>
<td>dark current (pA)</td>
<td>0.01</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>dark signal (mV)</td>
<td>2</td>
<td>0.05</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>0.3</td>
<td></td>
</tr>
<tr>
<td>DSNU (mV&lt;sub&gt;p-p&lt;/sub&gt;)</td>
<td>2</td>
<td>0.02</td>
<td>0.24&lt;sup&gt;d,e&lt;/sup&gt;&lt;sup&gt;.,f&lt;/sup&gt;</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>PRNU (% V&lt;sub&gt;set,p-p&lt;/sub&gt;)</td>
<td>2</td>
<td>7</td>
<td>5</td>
<td>8&lt;sup&gt;g&lt;/sup&gt;</td>
<td>8&lt;sup&gt;g&lt;/sup&gt;</td>
<td>4&lt;sup&gt;h&lt;/sup&gt;</td>
<td>4&lt;sup&gt;h&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>in darkness  
<sup>b</sup>no kTC noise, reduced 1/f noise  
<sup>c</sup>entry for DSNU may belong in this space (in darkness)  
<sup>d</sup>150 e<sup>-</sup>  
<sup>e</sup>use conversion 1 e<sup>-</sup> = 1.6 $\mu$V  
<sup>f</sup>this entry may belong in space for noise (mV<sub>rms</sub>) (in darkness)  
<sup>g</sup>calculated with $\frac{V_{max}-V_{min}}{V_{set}}$  
<sup>h</sup>calculated with $\frac{(V_{max}-V_{min})/2}{V_{set}}$

Table 5.3: CCD Comparison Table: Noise Characteristics
<table>
<thead>
<tr>
<th></th>
<th>LORAL CCD-191DC</th>
<th>THOM-SON TH7808BCC</th>
<th>KODAK KLI-5001F</th>
<th>SONY ILX501</th>
<th>SONY ILX504B</th>
<th>SONY ILX506</th>
<th>SONY ILX508</th>
</tr>
</thead>
<tbody>
<tr>
<td>test exp. time (ms)</td>
<td>10</td>
<td>1</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>test light source</td>
<td>2854 K Tungsten filament lamp</td>
<td>2854 K Tungsten filament lamp</td>
<td>3200 K</td>
<td>3200 K</td>
<td>3200 K</td>
<td>3200 K, 2854 K</td>
<td>3200 K, 2854 K</td>
</tr>
<tr>
<td>test data output (MHz)</td>
<td>1.0</td>
<td>5</td>
<td>25</td>
<td>1, 6&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1, 2.5, 6&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>test filters</td>
<td>WBHM + 2 mm thick BG-38&lt;sup&gt;b&lt;/sup&gt;</td>
<td>WBHM + 2 mm thick BG-38&lt;sup&gt;b&lt;/sup&gt;</td>
<td>IR cut, CM-500S (t = 1.0 mm)</td>
<td>IR cut, CM-500S (t = 1.0 mm)</td>
<td>IR cut, CM-500S (t = 1.0 mm)</td>
<td>IR cut, CM-500S (t = 1.0 mm)</td>
<td></td>
</tr>
<tr>
<td>test wavelength (nm)</td>
<td>700</td>
<td>700</td>
<td>550</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>test operating temp. (°C)</td>
<td>25</td>
<td>25</td>
<td>23</td>
<td>25</td>
<td>25</td>
<td>25</td>
<td>25</td>
</tr>
</tbody>
</table>

<sup>a</sup>input clock frequency  
<sup>b</sup>limits frequency spectrum to 700 nm

Table 5.4: CCD Comparison Table: Test Data

<table>
<thead>
<tr>
<th></th>
<th>LORAL CCD-191DC</th>
<th>THOM-SON TH7808BCC</th>
<th>KODAK KLI-5001F</th>
<th>SONY ILX501</th>
<th>SONY ILX504B</th>
<th>SONY ILX506</th>
<th>SONY ILX508</th>
</tr>
</thead>
<tbody>
<tr>
<td>preliminary?</td>
<td>yes</td>
<td>yes</td>
<td>no</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>price (1)</td>
<td>$1600</td>
<td>$860</td>
<td>$500</td>
<td>$75</td>
<td>$80</td>
<td>$110</td>
<td>$680</td>
</tr>
<tr>
<td>price (1000)</td>
<td>$1000</td>
<td>~ $350</td>
<td>$153</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>price (1000, next year)</td>
<td>$700</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>lead time</td>
<td>2–3 days</td>
<td>4 weeks</td>
<td>2–30 days</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>evaluation board price</td>
<td>$4290</td>
<td>~ $4000</td>
<td>~ $4000</td>
<td>$4000&lt;sup&gt;a&lt;/sup&gt;</td>
<td>$4000&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
<td></td>
</tr>
<tr>
<td>eval. board schematic</td>
<td>yes</td>
<td>no</td>
<td>yes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<sup>a</sup>may not be available

Table 5.5: CCD Comparison Table: Ordering Information

68
under uniform illumination.

**AC gain mismatch** Difference between gain of two shift registers under uniform illumination.

**Charge transfer efficiency (CTE)** Percentage of valid charge information which is transferred between successive stages of the charge transport shift registers.

**Dark current** Current output from the CCD in the dark.

**Dark reference pixel** Pixel within the CCD array which is blocked from light during fabrication to provide a reference level for the CCD output in the dark.

**Dark signal** CCD output voltage signal in darkness. There are three sources of dark signal voltage: fixed pattern noise (which is constant and depends on slight fabrication differences between independent pixels); dark leakage current (which is caused by thermally generated electrons, and increases linearly with exposure time and approximately by a factor of 2 with every 7° C temperature increase); and thermodynamic noise (thermally generated or kTC noise, which is generated on diode diffusions, and depends on capacitance of the diode and the operating temperature).

**Dark signal non-uniformity (DSNU)** Difference of response levels of the most- and least-sensitive pixels in darkness.

**DC offset** Output DC offset at saturation.

**Dynamic range** Saturation equivalent exposure / temporal noise equivalent exposure (peak-to-peak or rms).

**Evaluation board** Prefabricated circuit board which can be used to test and control the CCD.

**Exposure level** Light intensity $\times$ integration time.

**Exposure time / integration time** Time during which charge is accumulated in the photosites.
**Lead time**  Time needed for the manufacturer to ship one CCD.

**Noise**  Ratio of saturation output voltage to rms or peak-to-peak noise in CDS (correlated double sampling) mode.

**Noise equivalent exposure**  Exposure level which will produce an output signal equivalent to the noise level of the output in darkness.

**Peak responsivity**  Wavelength of incident light for which the CCD output is at a maximum.

**Photoresponse non-uniformity (PRNU)**  Difference of response levels of the most- and least-sensitive pixels under uniform illumination (excluding the first and last elements of the array). Usually measured at 50% of $V_{sat}$.

**Pitch**  Center-to-center distance between adjacent pixels.

**Pixel**  Picture element (photosite).

**Range**  Output peak-to-peak voltage at saturation.

**Responsivity**  The output signal voltage per unit exposure for a specified wavelength of radiation: output voltage / exposure.

**Saturation voltage**  Maximum useable signal output voltage; charge transfer efficiency decreases sharply when the saturation output voltage is exceeded. Measured with respect to dark signal level.

**Saturation voltage equivalent exposure**  Minimum exposure level which will produce a output signal equivalent to saturation.

**Size**  Size (length $\times$ width) of each pixel.

**Total transfer efficiency (TTE)**  Charge transfer efficiency over the entire length of the charge transport registers.

**Wavelengths**  Range of wavelengths of incident light which produce an output from the CCD.
WBHM  Wide-band hot mirror.

Window  Thin glass covering over the CCD array.

For this application, the most desirable characteristics in a CCD were:

- Inexpensive
- Large number of pixels (high resolution)
- Simple interface (minimal number of clocks, outputs)
- Possible to distinguish bands at low light levels (sensitivity)
- Possible to detect both bright and dim bands in the same tube (dynamic range)
- Minimal charge leakage between pixels
- Minimal static noise due to differences between pixels

The following conclusions were made based on the CCD data:

- At less than $100 each, the 5000-pixel Sony CCDs were the cheapest of the CCDs, and the Loral 6000-pixel CCD for $1600 was the most expensive.
- The Sony ILX508, with 7926 7 μm pixels, had the most resolution of the CCDs studied, and was less than half the price of the 6000-pixel Loral CCD-191DC.
- The Sony CCDs had the simplest interface, in terms of number of clocks, voltage sources, and number of output pins.
- The responsivity of the Sony ILX506 and ILX508 was calculated to be so much higher than the responsivities of the other CCDs that it was suspected that the preliminary data sheets for these CCDs were inaccurate. This made choosing a CCD more difficult.
- The peak-to-peak saturation voltage of the Loral CCD was also much higher than for the other candidates. The higher peak-to-peak output would make it easier to detect small variances in the output voltage, and thus small changes in incident light levels.

- The Loral CCD had a much wider dynamic range than the Thomson and Kodak CCDs. Unfortunately, the dynamic range was not available for the Sony CCDs, so a full comparison could not be made.

- The noise of the CCD outputs, as determined by the DC channel offset, the charge transfer efficiencies, and the dark signal and photoresponse non-uniformities, seemed to be comparable for all the CCDs evaluated.

- Although it was not planned to purchase an evaluation board along with the CCD, it was hoped that it would be possible to obtain a schematic for the evaluation board, to serve as a model for controlling the CCD. Only Loral and Kodak offered the evaluation board schematic without purchase of the evaluation board. An evaluation board for the newer Sony CCDs, the ILX506 and ILX508, was not available at the time that the CCDs were planned to be purchased.

Since the Sony ILX508 had much more resolution than the other CCDs, for a reasonable price, it was decided to purchase this CCD. However, there was some delay at Sony that resulted in their revising the lead time to 6-8 weeks. This was unacceptable in the timeline of the project, and the order was cancelled.

The second choice was the Kodak KLI-5001F. The Loral CCD-191CD was too expensive without offering commensurate advantages, and the responsivity and dynamic range of the Thomson TH 7808BCC and the other Sony CCDs were not as good as the Kodak. An order was placed for two Kodak KLI-5001Fs.

However, this action proved to be premature. Brad Thomas of BD spoke to representatives at Sony and was successful in obtaining the 7926-pixel CCD in a matter of days, demonstrating the power a major company has in the business world.
which a university lacks. It was decided to keep the Kodak CCDs which had been ordered as backups in case the Sony CCDs were unusable for any reason. The Sony ILX508 again became the working CCD for the project, and work began on designing a controller/interface board for the CCD.

Figures 5-1 shows a drawing of the Sony CCD, and Figure 5-2 gives some dimensions of this CCD package[15].
Figure 5-2: Dimensions of Sony ILX508 7926-Pixel CCD
Chapter 6

Implementation of CCD Boards

Control of the CCD from the PC was accomplished using two circuit boards. The board which holds the CCD was implemented as a printed circuit board and is called the CCD PCB. The CCD PCB is controlled from a second board, called the CCD computer interface board, which fits into one of the expansion bus slots of the PC. The CCD computer interface board was wire-wrapped to facilitate slight changes and additions. A block diagram of the two boards is shown in Figure 6-1.

This chapter will describe the components chosen to implement these boards, and discuss the reasons for the choices. Chapter 7 will present the design and operation of the CCD boards. These two areas are difficult to separate, as the process of subsystem design and component selection often overlapped in this task.

6.1 CCD Printed Circuit Board

The CCD PCB would be part of the optical setup, and it would be necessary to easily adjust the board when aligning the optics. For this reason, the CCD PCB was designed to be compact, incorporating the minimum amount of hardware needed to convert the analog voltage output of the CCD to a digital signal. The analog-to-digital conversion is done on the PCB to help minimize the noise in the CCD output; driving an analog signal through the long ribbon cable connecting the two boards would degrade the signal. All of the analog signals in the system are contained on
Figure 6-1: Block Diagram of the CCD Boards
the CCD PCB; the printed circuit board implementation helps to reduce the noise on the board. All of the signals sent to and from the board are digital.

The layout of the CCD PCB was done by Benjamin Linder using PADS-PCB. The PCB is about three inches square. A schematic diagram of the CCD PCB is shown in Figure 6-2, and a photo of the PCB along with some other optical components is shown in Figure 6-3.

The components on the PCB consist of the Sony ILX508 CCD described in Chapter 5; an operational amplifier (op amp) circuit used to amplify and remove the DC offset from the CCD output; an analog-to-digital converter (ADC); a buffer for off-board signals; and two voltage regulators. Each of these components was carefully selected for use in the CCD interface. Several variables were considered in each selection, with price always being a factor.

### 6.1.1 Analog-to-Digital Converter

The primary considerations in choosing an ADC were as follows:

**Resolution** The QBC® Autoread used an ADC with 12-bit resolution, so the resolution of the ADC selected for the Walkaway prototype needed to be at least 12 bits. However, the time per conversion needed to be considered along with the desired resolution; higher resolution increases the conversion time.

**Conversion time** The conversion rate of the ADC determines the clock frequency of the system. The CCD can be clocked at a maximum frequency of 12.5 MHz; the corresponding clock period of 80 ns is faster than an ADC can perform a 12-bit conversion. It was important to get the fastest ADC available within the budget of the project to maximize the speed of the system.

**Track/Hold** The CCD output would need to be captured by a sample-and-hold (S/H) or track/hold (T/H) circuit to guarantee that the output would be stable during conversion. An external S/H or T/H component could be used; however, it would be convenient if this feature were included in the ADC.
Figure 6-2: Schematic of the CCD PCB
Noise  Low noise in the ADC output was desired.

Power  The power consumption of the ADC was considered, although it was not a determining factor in the decision.

The ADC selected was the Maxim MAX120 12-bit ADC, with a 1.6 μs conversion time and an on-chip track/hold with an acquisition time of 350 ns, resulting in a throughput of 500 ksamples/s. The output noise is ±1 bit, and the power dissipation is 210 mW, both of which are about average for an ADC of this size. The input range of the ADC is ±5 V, a factor which wasn’t considered in this decision, but which became important in the selection of an op amp.

6.1.2  Operational Amplifier

The primary considerations in choosing an op amp were as follows:

Slew rate  The slew rate is the number of volts per second attainable by the output.

It was important that this value be high; the output of the op amp must achieve
a full ±5 V swing well within the 2 µs conversion period of the ADC in order to take full advantage of the ADC resolution and speed.

**Settling time**  After reaching a final value, it was important that any ringing on the output settle quickly, to reduce noise in the data and provide a more accurate representation of the CCD output.

**Gain Bandwidth Product**  The gain-bandwidth product is generally a constant; a higher switching rate can be obtained by lowering the gain, and a higher gain can be obtained by slowing the switching rate. This application requires both a high gain (5–10) and a high bandwidth; the output of the CCD switches every 1 µs, and the output of the op amp must settle in a fraction of that time. Therefore, the gain-bandwidth product must be high. For a gain of 10 (the saturation output of the CCD is 1 V_{pp}, which is amplified to 10 V_{pp} to use the ±5 V input range of the ADC) and a bandwidth of 10 MHz (so that the output of the op amp settles within one-tenth of the time that the CCD output stays constant), the gain-bandwidth product would be 100 MHz.

The op amp selected was the Analog Devices AD744, which has a settling time of 500ns to 0.01% for a 10 V step, a 75V/µs slew rate, and a gain bandwidth of over 200 MHz with external decompensation (using an external capacitor across the op amp).

### 6.1.3 Buffers

The buffers are used to drive the output signals going through the ribbon cable to the CCD computer interface board, and to receive the clock signals coming from the CCD computer interface board. The CCD data sheets specified that the input clocks to the CCD should have a high voltage level of 5 V; since a TTL logic high level is generally around 3 V–3.5 V, CMOS buffers were used to provide a full voltage swing. The CMOS chips used were Texas Instruments 74HC541 buffers.
6.1.4 Voltage Regulators

The CCD required a 9 V and a 5 V voltage source. Standard 9 V voltage regulators exist, but are difficult to obtain. As the lead time for these devices was at least a month, it was decided to use instead a 5 V regulator with the ground input set by a voltage divider to bring the output up to 9 V. Texas Instruments μA78M05CKC 5 V voltage regulators were used. The circuit is shown in the Chapter 7 in Figure 7-1.

6.2 CCD Computer Interface Board

The logic to control the CCD was built on a board called the JDR-PR10, a JDR Microdevices prototype card for PC-AT computers. The card consists of a small PCB portion which buffers the data bus signals and decodes the I/O port addresses used by the board, and a larger breadboarding area. The components were placed in sockets which were wire-wrapped onto the board. In order to get the best connections possible, silver wire and sockets with gold-plated contacts and sleeves were used.

The CCD computer interface board contains an 8 MHz crystal oscillator from which the clock signals to the CCD and ADC are derived; counters to measure the integration time of the CCD; SRAM buffers and address counters to temporarily store the data from the CCD; output buffers to drive the ribbon cable connecting the CCD computer interface board to the CCD PCB, and to buffer the signals coming from the CCD PCB; and TTL logic gates, registers, and buffers. A schematic diagram of the CCD computer interface board is shown in Figure 6-4.

6.2.1 JDR PCB

The PCB portion of the JCR Microdevices prototype card contains two preprogrammed PAL (Programmable Array Logic) chips which decode the I/O port addresses and output control signals to the rest of the board. The other components on the PCB section of the JDR board are standard TTL 74LS one-way and two-way buffers which regulate signals going to and from the computer bus.
Figure 6-4: Schematic of the CCD Computer Interface Board
6.2.2 Counters

The counters used in the design are also Texas Instruments TTL 74LS series, although they are not as much in common use as the more popular '163 or '169 counters. Both counters used are eight-bit counters in a 16-pin package.

The counter used to measure the integration time is the 74LS592. It has eight load inputs and one RCO output. The inputs can be latched into an internal input register, and loaded from the register into an internal binary counter with a separate load signal.

The counter used to address the SRAM is the 74LS590. It has no load inputs (the counter is reset with the CLEAR input), and eight outputs (in addition to the RCO output) which reflect the internal state of the counter. The output registers of the counter are tri-stateable.

The eight bit counters with on-chip input or output registers reduced the number of chips needed in the design while preserving the functionality of the control system. This will be discussed further in Sections 7.1.2 and 7.2.2.

6.2.3 SRAM

The only constraints on the SRAM were that it must store 8 Kb of data, and that it must have a "reasonable" access time: less than 100 ns. The SRAM chosen was a Cypress Semiconductor CY7186-25PC. This SRAM was size 8 K × 8, with a 25 ns access time. Two 8-bit wide SRAM chips are needed to store the 12-bit output of the ADC; the high-order four bits of the second chip are not used.

6.2.4 Buffers

As on the CCD PCB, CMOS buffers are used on the CCD computer interface board to drive the clock signals to the CCD PCB through the ribbon cables, and to receive the data outputs from the CCD PCB. The CMOS chips used in this application were Texas Instruments 74HC244 buffers.
6.2.5 Logic Gates and Registers

As the project did not have access to a PAL programmer, all of the logic used in the system was implemented using standard gates. The 74LS series was chosen because of the lower power consumption and high availability. With a 500 kHz system clock, the timing of the system was not critical enough to require a faster gate.
Chapter 7

Operation of CCD Boards

In the prototype Walkaway machine the CCD is controlled from a PC, which allows for easy testing and modification of the control procedures. However, as the computer is also be used to control other subsystems of the machine, continuous monitoring of the CCD subsystem is not possible. The PC should be free, for instance, to move the linear filter while collected data is being read from the CCD.

The computer interface to the CCD was designed to minimize the amount of time that the computer actively controls the CCD. Parameters for a particular CCD scan would be loaded from the PC bus into registers on a controlling board before the start of a scan. The PC would then send a “START” signal to the CCD computer interface board, and the computer interface board would send the correct clock signals to the CCD. The CCD PCB would take scanned CCD data, process the data, and send the digitized data back to the computer interface board, where it would be stored in SRAM buffers. When these tasks were finished, the computer interface board would raise a “DAV” (Data AVailable) flag to indicate that there was new data in the SRAM. The computer would then read the status of this flag and retrieve the data at a convenient time.

The design of the CCD boards was developed primarily through study of the CCD inputs and outputs, to determine how the CCD should be controlled to achieve the desired sequences of operation of the subsystem.
7.1 CCD Inputs

The Sony CCD requires two voltage inputs, a 9 V source, and a 5 V source, and two input clock signals, called $\Phi_{\text{CLK}}$ and $\Phi_{\text{ROG}}[15]$. $\Phi_{\text{CLK}}$ defines the rate at which analog data is shifted out of the CCD, and $\Phi_{\text{ROG}}$ is an intermittent pulse which marks the beginning and end of the CCD integration time\(^1\).

7.1.1 Voltage Sources

It was decided to derive both the 9 V and 5 V voltage sources for the CCD from a 12 V source on the PC bus, which was carried to the CCD PCB through the ribbon cable. The 12 V voltage is regulated down to 9 V, and the 9 V source is then used as an input to a 5 V regulator, as shown in Figure 7-1. This arrangement prevents the 5 V input to the CCD from going higher than the 9 V input. To further ensure that this would be the case, a diode was back-biased between the two sources\(^2\).

Due to a miscommunication, the layout of the CCD PCB derived both the 9 V

\(^1\)It is convenient to say that the $\Phi_{\text{ROG}}$ pulse marks the beginning and end of the integration time. Actually, the CCD outputs data continuously. The $\Phi_{\text{ROG}}$ signal denotes divisions between integration times, but not the beginning and end of any one integration time. The CCD collects charge during the time that it is shifting out previous data.

\(^2\)One expensive CCD was damaged in testing due to insufficient precautions regarding the power supplies.
and 5 V sources from the 12 V source, necessitating a minor modification to the fabricated board.

### 7.1.2 Integration Time

In normal machine operation, each tube is scanned several times, using two light sources in different orientations with respect to the tube, and with filters of various wavelengths between the illuminated tube and the CCD. The light sources and filters greatly change the intensity of the tube image on the CCD. In order to ensure that the brighter images do not saturate the CCD output, and that the dimmer images are readable, it is necessary to vary the CCD integration time from reading to reading.

The prefabricated CCD evaluation board used in preliminary tests allowed variation of the integration time through the manual adjustment of a potentiometer. For the prototype, however, the integration time would be set from the computer.

The integration time control was implemented as follows: before the beginning of a scan, a number corresponding to the desired integration time is loaded from the computer bus into a register on the CCD computer interface board. When the board receives a START signal from the PC, the integration time number is loaded from the register into a counter and the counter begins to count at the same time as a $\Phi_{ROG}$ pulse is sent to the CCD, starting the integration time. When the counter asserts the carry-out (RCO) signal, another $\Phi_{ROG}$ pulse is sent to the CCD, ending the integration time period. Data is shifted out of the CCD, amplified, converted to digital, and stored in the SRAM at the system clock rate of 500 kHz. When all of the data from the scan has been stored, the DAV signal is enabled. The integration time number that was loaded into the register does not change; the same integration time will be used for the next scan if not explicitly changed from the computer.

The counter used to measure the integration time in the prototype is a 24-bit counter composed of three cascaded 8-bit counters; these counters have twice as many bits in a package the same size as a a standard 4-bit counter. In addition, the counters contain an internal input register, eliminating the need for a separate register to store the integration time loaded from the computer. This helps to reduce...
the number of chips needed in the implementation. However, the counters have no outputs except for a carry output, which makes it difficult to determine the state of the counter during testing and debugging.

The integration time counter is clocked at the system clock frequency of 500 kHz. The middle 16 bits of the counter are loaded from the PC bus; the low-order four bits of the counter are wired low, and the high-order four bits are wired high (and thus are not used). The number loaded into the counter is the maximum number that can be represented in the usable 20 bits of the counter, minus the number that, multiplied by the clock period of 2 $\mu$s, yields the desired integration time. The counter then increments until RCO is asserted, marking the end of the integration time. With this implementation, the integration time has a range of 0.032 ms–2097.15 ms, with a resolution of 0.032 ms. This was adequate for the purposes of the prototype.

The time needed for the CCD to shift out 8048 pixels of information at a clock frequency of 500 kHz is about 16 ms. Although in theory it is possible to specify an integration time of less than 16 ms, shorter integration times will result in faulty CCD data. One way to redesign the board to allow for shorter integration times would be to clock the CCD at a faster rate during the integration time, and switch to the 500 kHz clock rate, determined by the ADC and the op amp, when shifting data out of the CCD.

### 7.1.3 CCD Clock

The clock input to the CCD, $\Phi_{\text{CLK}}$, must exhibit a particular relationship to the $\Phi_{\text{ROG}}$ input. The minimum high time of the $\Phi_{\text{ROG}}$ pulse is 1200 ns. The $\Phi_{\text{CLK}}$ waveform

---

3Early in the design stage, the concern surfaced that it would not be possible to use integration times shorter than the amount of time needed to read the previous data out of the CCD. Some alternative design schemes which would allow shorter integration times were considered, but development of those designs was delayed until more information on the CCD could be obtained. It was hoped that the start of a new integration time would allow the new data to “overwrite” data from the previous integration time which was not shifted out of the CCD. However, information was not available from the manufacturing company on this mode of operation, and it was decided to implement the current design and modify it if necessary. Later testing showed that the data from the previous integration time stayed in the shift registers of the CCD until it was shifted out. However, by this time it had been determined that integration times shorter than 16 ms were not necessary for testing the CCD and the prototype machine, so the modification was never done.

---

88
must be high for a minimum of 200 ns before the rising edge \( \Phi_{\text{ROG}} \), and remain high for a minimum of 1200 ns after the falling edge of \( \Phi_{\text{ROG}} \).

Both the \( \Phi_{\text{CLK}} \) and \( \Phi_{\text{ROG}} \) signals are derived from the 500 kHz system clock. The \( \Phi_{\text{CLK}} \) frequency is normally 500 kHz. \( \Phi_{\text{ROG}} \) is output from a register clocked with an inverted system clock, so that it goes high on the falling edge of the system clock, and stays high for one 2 \( \mu \)s clock period. \( \Phi_{\text{CLK}} \) is then already high for 1 \( \mu \)s before the rising edge of \( \Phi_{\text{ROG}} \), and it is gated with the input to the \( \Phi_{\text{ROG}} \) register so that it stays high for one full clock period of 2 \( \mu \)s after \( \Phi_{\text{ROG}} \) goes low.

### 7.2 CCD Output

#### 7.2.1 Processing

The output of the Sony CCD is an analog voltage waveform which looks somewhat like a square wave. The top level of the waveform is a constant value to which the output returns between pixels. The output voltage corresponding to each pixel is a lower value; the difference between this value and the constant top level for a pixel is proportional to the light intensity seen by that pixel.

The CCD output contains information from 8042 pixels. 7926 of these contain information about the light levels seen by the CCD. Thirty-six pixels, eighteen at either end of the CCD, are "dummy pixels" which are present for isolation purposes but contain no information. Eighty pixels at one end of the CCD see an optical black input to give a "dark reference" output. A diagram from the Sony ILX508 CCD data sheets illustrating the clocking and output of the CCD is shown in Figure 7-2[15].

The output of the CCD is buffered through an NPN transistor in an emitter-follower configuration to prevent loading of the CCD output.

The maximum peak-to-peak voltage of the output waveform is 2 V (greater than the 1 V predicted by the preliminary data sheets), with a DC offset of about 3 V. The input range of the ADC used to convert the CCD output to a digital representation is \( \pm 5 \) V. To make full use of the 12-bit resolution of the ADC, an op amp circuit
Figure 7-2: Sony ILX508 7926-Pixel CCD Timing Diagram
amplifies and shifts the analog output of the CCD to about ±4.5 V. This circuit is shown in Figure 7-3.

The “start conversion” input to the ADC is the 500 kHz system clock; the output of the CCD and op amp circuit is continuously converted, whether or not the CCD output is of interest. However, the writing of the SRAM is controlled so that the output of the ADC is only stored in the SRAM after an explicit CCD scan.

In testing and optical alignment, it may also be useful to display the CCD output on an oscilloscope. The buffered analog output of the CCD may be used as a data input to an oscilloscope either before or after the op amp circuit. A START signal should be continuously sent to the CCD computer interface board from the computer at a rate determined by the integration time, to provides a repeating trigger signal which is aligned with the CCD output.

7.2.2 Storage and Retrieval

The data from one scan of the CCD is stored in an SRAM buffer until it is read by the computer. The SRAM is addressed by a 16-bit counter implemented with two eight-bit counters; these counters, which are the same physical size as common 4-bit counters and also incorporate an on-chip tri-state output register, help to reduce

---

4 Although one CCD scan consists of 8042 pieces of data, 8192 values are actually stored in the SRAM. Using a power of two makes some of the control logic simpler, and doesn’t significantly affect the timing.
the number of chips needed in the design. When the SRAM contains a complete new data set, DAV is asserted. The original intention was to implement a two-way communications flag which would be set by the CCD computer interface board when new data was stored in the SRAM, and reset by the computer when the new data was read, but it was decided that this would not be necessary. The cycle of scanning the CCD and storing the data is begun by the computer’s START signal to the CCD computer interface board. Simply requiring that the controlling software make sure that DAV is asserted before initiating another scan has the same effect as a two-way communication flag.

The DAV signal does not require extra gates or registers to implement. DAV is low when data is being written to the SRAM and high otherwise, serving as a “busy” signal. For a period corresponding to the integration time after START is asserted, DAV is still high, since the data in the SRAM from the previous scan is still valid. One possible pipelined implementation of the controlling software would be as follows:

1. The computer sends a START signal to the CCD computer interface board.
2. The computer reads the output data from the previous scan from the SRAM.
3. The computer checks that DAV is still high, verifying that the data just read is valid.
4. After the next rising edge of DAV, the computer sends another START signal.

This method is subject to the errors which may occur if the computer read time overlaps the SRAM write time. The algorithm used in the prototype control software is serial:

1. The computer sends a START signal to the CCD computer interface board.

---

*Since only 13 of the sixteen bits of the counter are used, the fourteenth bit of the counter is used as an RCO to signal the end of the counting. However, the outputs of the counter must be tri-stated when not in use, so that the computer may address the SRAM, but the RCO output must always be active. For this reason, a third counter was added to the arrangement. This new counter has the same inputs as the higher-order eight-bit counter, but the output is not tri-stated or used as an address input to the SRAM, and the sixth bit is used as the RCO for the SRAM address counters.*
2. After the next rising edge of DAV, the computer reads the output data from the SRAM.

In order to be sure that data has been written to the SRAM, it is necessary for the computer to detect the low cycle of the DAV signal (or determine that a sufficient period of time has passed).

The SRAM is implemented as a section of the PC memory. When the PC reads from an address in the range of the SRAM, an address decoder on the CCD computer interface board enables the SRAM output to be placed on the computer bus. The computer may also store values in the SRAM, making it possible to test the SRAM by storing values and reading them back. The memory address space used by the SRAM is the hexadecimal addresses 0xCC00-0xCCFF. To ensure that the computer doesn’t store anything in the memory used by the SRAM, the following line is included in the DOS PC’s CONFIG.SYS file: “DEVICE x = CC00-CCFF”.

7.3 Communication

A 40-pin ribbon cable connects the CCD computer interface board with the CCD PCB. Every even pin (corresponding to every other line in the cable) is grounded in order to reduce crosstalk between the signals.

The CCD PCB was originally designed for a 50-pin ribbon cable to allow for the possible addition of communication signals. Since no new signals were added, a 40-pin cable was used. The top 14 lines of the cable are unused and grounded, so the 40-pin connector is inserted into the bottom 40 holes of the connector socket.

In assembling the final prototype, it was found that the ribbon cable, which had been on the front (component side) of the PCB, obstructed the optics. The connector was removed and connected on the other side of the board. This necessitated reconnecting in reverse order all of the wires from the CCD computer interface board to the connector at the other end of the cable.

The signals on the odd lines of the cable connecting the CCD PCB and the CCD computer interface board are shown in Table 7.1. The input/output status of the
<table>
<thead>
<tr>
<th>Line</th>
<th>Signal</th>
<th>Status</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>$\Phi_{\text{ROG}}$</td>
<td>Output</td>
<td>Start/stop integration time pulse signal to CCD</td>
</tr>
<tr>
<td>3</td>
<td>CCDCLK</td>
<td>Output</td>
<td>Clock input to CCD (nominally 500 kHz)</td>
</tr>
<tr>
<td>5</td>
<td>CONVST</td>
<td>Output</td>
<td>Start convert signal to A/D converter</td>
</tr>
<tr>
<td>7</td>
<td>CLINK</td>
<td>Output</td>
<td>Clock input to A/D converter (8 kHz)</td>
</tr>
<tr>
<td>9</td>
<td>AD11</td>
<td>Input</td>
<td>MSB output from A/D converter</td>
</tr>
<tr>
<td>11</td>
<td>AD10</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>13</td>
<td>AD9</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>15</td>
<td>AD8</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>17</td>
<td>AD7</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>19</td>
<td>AD6</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>21</td>
<td>AD5</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>23</td>
<td>AD4</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>25</td>
<td>AD3</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>27</td>
<td>AD2</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>29</td>
<td>AD1</td>
<td>Input</td>
<td>Output from A/D converter</td>
</tr>
<tr>
<td>31</td>
<td>AD0</td>
<td>Input</td>
<td>LSB output from A/D converter</td>
</tr>
<tr>
<td>33</td>
<td>+12 V</td>
<td>Output</td>
<td>12-volt power for op amp</td>
</tr>
<tr>
<td>35</td>
<td>-12 V</td>
<td>Output</td>
<td>-12-volt power for op amp and A/D converter</td>
</tr>
<tr>
<td>37</td>
<td>GND</td>
<td>Output</td>
<td>Ground</td>
</tr>
<tr>
<td>39</td>
<td>GND</td>
<td>Output</td>
<td>Ground</td>
</tr>
</tbody>
</table>

Table 7.1: Communication Signals between the CCD PCB and the CCD Computer Interface Board

signals is with reference to the CCD computer interface board.

7.4 Interfacing with the Computer Bus

As described in Section 6.2.1, the CCD computer interface board is built on a JDR Microdevices prototype card consisting of a pre-fabricated PCB portion and a larger breadboarding area. The PCB portion buffers the bus signals and decodes the I/O port address through which the PC communicates to the board. The I/O port addresses used by the board are programmed into a PAL on the PCB section: hexadecimal addresses 0x300–0x30F[7]. Due to some conflicts of these addresses with the addressing of other boards interfaced with the computer, the CCD computer interface board was modified to respond to hexadecimal addresses 0x280–0x28F. Because
the project did not have access to a PAL programmer, this was done by externally inverting two of the address inputs to the address decode PAL.

The JDR board also conflicted with the other PC boards in the system through its assertion of the \texttt{IOCS16} computer bus signal. This signal must be asserted by the board whenever the board makes a sixteen-bit transfer over the bus using one of its I/O ports. However, with more than one I/O board in the computer, the signal coming from the board must be tri-stated so that other boards can control \texttt{IOCS16}. Similarly, the \texttt{MEMCS16} computer bus signal must be asserted whenever the SRAM on the board is addressed by the computer, putting a 16-bit value on the bus, and tri-stated otherwise. This signal is not provided on the JDR PCB, so it was implemented in logic gates in the breadboarding area.

The protoboard section of the JDR board contains power and ground planes which restrict placement of the components. As this was not clear from the board description, an early partial implementation of the CCD computer interface board had to be completely redone.

### 7.4.1 Bus Signals

The PC/AT bus signals which are used on this board are listed in Table 7.2[5].

### 7.4.2 Input/Output Port Addresses

The computer I/O port addresses used to communicate with the CCD computer interface board and the CCD PCB are shown in Table 7.3.

### 7.5 Control Operations

#### 7.5.1 Signals

The following signal names are used in describing the operation of the two CCD boards.

\textbf{SYSCLK} 500 kHz system clock
<table>
<thead>
<tr>
<th>Board</th>
<th>Bus</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>SA8</td>
<td>A23</td>
<td>Address bit which is inverted to change I/O port address space of board</td>
</tr>
<tr>
<td>SA7</td>
<td>A24</td>
<td>Address bit which is inverted to change I/O port address space of board</td>
</tr>
<tr>
<td>–12 V</td>
<td>B7</td>
<td>Output to CCD board to be used to power op amp and A/D converter</td>
</tr>
<tr>
<td>+12 V</td>
<td>B9</td>
<td>Output to CCD board to be used to power op amp</td>
</tr>
<tr>
<td>SMEMW</td>
<td>B11</td>
<td>Used to determine write enable signals to SRAM (memory write)</td>
</tr>
<tr>
<td>SMEMR</td>
<td>B12</td>
<td>Used to determine read enable signals to SRAM (memory read)</td>
</tr>
<tr>
<td>SBHE</td>
<td>C1</td>
<td>Used to determine read/write enable signals to SRAM (high byte enable)</td>
</tr>
<tr>
<td>MEMCS16</td>
<td>D1</td>
<td>Output to bus to indicate that a 16-bit memory transfer is in process</td>
</tr>
<tr>
<td>IOC16</td>
<td>D2</td>
<td>Output to bus to indicate that a 16-bit I/O transfer is in process</td>
</tr>
</tbody>
</table>

Table 7.2: Computer Bus Signals used on the CCD Computer Interface Board

<table>
<thead>
<tr>
<th>Port Address</th>
<th>Computer I/O</th>
<th>Function</th>
</tr>
</thead>
<tbody>
<tr>
<td>0x280–0x283</td>
<td>Output</td>
<td>Mark beginning of CCD integration time</td>
</tr>
<tr>
<td>0x284–0x287</td>
<td>Input</td>
<td>New CCD Data AVailable signal</td>
</tr>
<tr>
<td>0x288–0x28B</td>
<td>Output</td>
<td>Load integration time into counter</td>
</tr>
</tbody>
</table>

Table 7.3: Computer Port Addresses used in the CCD Subsystem
**LDST** Load signal from computer; loads LSB into register which holds START signal

**START** Starts sequence of operations when signal makes a transition from low to high

**INTLD** Load signal to integration time counter, produced by START transition from low to high; loads the integration time number from the input registers into the counting registers to prepare for counting off the integration time

**INTCNT** Count enable signal to the integration counter; tied through an inverter to **INTRCO** so that when the counter reaches its maximum value, it stops counting

**INTCTR** Internal state of integration time counter

**INTRCO** Carry out signal from the integration counter

**ISYNC** Pulse produced by transition of **INTRCO** from low to high, indicating that the integration time counter is done

**CLKSW** Signal (usually high) which is NANDed with the inverted system clock (500 kHz) to produce the CCD clock; keeps CCDCLK high during the $\Phi_{ROG}$ pulse

**CCDCLK** 500 kHz clock which is forced high for a few cycles around the $\Phi_{ROG}$ pulse

**$\Phi_{ROG}$** Pulse which marks the beginning and end of the CCD integration time

**CONVST** Convert start signal to A/D converter (8 MHz clock)

**VOUT** Output of CCD (on the timing diagrams, the analog output is represented by a low, shaded value during the time when the data is valid, and a high value between pixel outputs; this is intended to represent the form of the output)

**VO** Output of CCD after the op amp circuit
**Dout**  A/D converted CCD output

**Comp**  Output of comparator indicating whether the address on the computer bus matches the address of the SRAM

**Adr**  Signal which enables SRAM to be addressed from the computer bus

**Adrclr**  Clear signal to SRAM address register

**Adren**  Enables tri-state output buffers of SRAM address registers

**Adrcrt**  Output of SRAM address register

**Adrrco**  Carry-out of SRAM address counter (actually the 14th output bit)

**Async**  Pulse produced by Adrrco going from low to high

**Wrhi, Wrlo**  Write enable signals for high and low bytes of SRAM

**Rdh, Rdlo**  Read enable signals for high and low bytes of SRAM

**Bufhi, Buflo**  Output enable signals for high and low bytes of data from the board to the computer

**Dav**  Data Available signal to computer; low when data is being written to the SRAM, and high otherwise

**A0**  LSB address bit from the computer bus; low for a 16-bit transfer or a low byte transfer, and high for a high byte transfer

**SBHE**  Status signal from the computer bus; enabled (low) for a 16-bit or upper byte transfer, and disabled (high) otherwise.

**SMEMW**  Status signal from the computer bus; enabled when a memory write under 1 Mb is in progress

**SMEMR**  Status signal from the computer bus; enabled when a memory read under 1 Mb is in progress
7.5.2 Logic

Some of the signals on the CCD computer interface board are related through a set of logic equations which are implemented in standard TTL gates. These signals could also be implemented with a programmable logic array using the following equations:

\[
\begin{align*}
WRHI &= (DAV + SYSCLK) \cdot (SMEMW + COMP + SBHE) \\
WRLO &= (DAV + SYSCLK) \cdot (SMEMW + COMP + AO) \\
RDHI &= SMEMR + COMP + SBHE \\
RDLO &= SMEMR + COMP + A0 \\
BUFHI &= SMEMR \cdot SMEMW + COMP + SBHE \\
BUFHI &= SMEMR \cdot SMEMW + COMP + A0 \\
ADR &= SMEMR \cdot SMEMW + COMP
\end{align*}
\]

7.5.3 Timing

The timing diagrams in Figures 7-4, 7-5, and 7-6 illustrate the operation of the CCD subsystem. The sequential operation of the CCD computer interface board and of the CCD PCB are described below.

1. The computer loads the integration time count into the input registers of the integration time counter through I/O port CNTPORT (0x288).

2. The computer sends a LDST pulse (which loads the LSB of the computer bus into a register on the CCD computer interface board) through STPORT (0x280); if the LSB is 1 and the register previously contained a zero, START makes a transition from low to high.

3. START is synchronized to the 500 kHz system clock to produce a pulse, INTLD.

4. INTLD loads the integration time from the internal input register into the counting registers of the integration time counter.

5. INTLD causes CLKSW to go low and remain low for two clock cycles.
6. CCDCLK remains high while CLKSW is low (CCDCLK must remain high during a $\Phi_{ROG}$ pulse).

7. INTLD causes $\Phi_{ROG}$ to go high half a clock cycle later (the register is clocked with an inverted SYSCLK), and stay high for one inverted clock cycle (marking the beginning of the CCD integration time).

8. INTRCO goes high on the next clock cycle because the integration counter was loaded with a non-maximum value.

9. INTRCO going high causes INTCNT to go low and enable the integration time counter, which starts counting up.

10. The integration time counter reaches its maximum value, and INTRCO goes low.

11. INTRCO going low causes INTCNT to go high and disable the counter.
Figure 7-5: Timing Diagram (Middle)
Figure 7-6: Timing Diagram (End)
12. **INTRCO** going high produces ISYNC, which goes high one clock cycle later and stays high for one clock cycle.

13. ISYNC causes **CLKSW** to go low and remain low for two clock cycles.

14. CCDCLK remains high while **CLKSW** is low (CCDCLK must remain high during a \( \Phi_{ROG} \) pulse, and for a certain length of time before and after the pulse).

15. ISYNC causes \( \Phi_{ROG} \) to go high half a clock cycle later, and stay high for one inverted clock cycle (marking the end of the CCD integration time).

16. Through a series of flip-flops and gates, ISYNC produces **ADRCLR**, which goes low two clock cycles after ISYNC goes high, and which clears the SRAM address register counters.

17. When the address counters are cleared, ADRRCO goes high.

18. ADRRCO going low causes **ADRCNT** to go low, and enables the address registers to start incrementing the SRAM addresses.

19. ISYNC sets a latch which causes DAV to go low two clock cycles after ISYNC goes high, to indicate to the computer that data is being read into the SRAM.

20. DAV going low causes **ADREN** to go low and enable the output of the SRAM address registers.

21. DAV going low causes **WRHI** and **WRLO** to begin pulsing, and data from the CCD is written to the SRAM.

22. When the SRAM address register reaches the end of the CCD output data, ADRRCO goes high.

23. ADRRCO going high causes **ADRCNT** to go high, and the SRAM address registers stop incrementing.
24. ADRRCO going high produces ASYNC, which goes low one clock cycle later and stays low for one clock cycle.

25. ASYNC resets the latch to return DAV to a high state, indicating to the computer that there is new data in the SRAM.

26. DAV going high causes ADREN to go high and disable the output of the SRAM address registers.

27. DAV going high causes WRHI and WRLO to stop pulsing and remain high, and data from the CCD stops being written to the SRAM.

28. At a convenient time, data is read from the SRAM into the computer.

29. The computer sends a LDST signal to load a zero into the register which outputs the START signal, so that this value goes low in preparation for the next high transition.
Chapter 8

CCD Control Software

Software to control the CCD subsystem and analyze the CCD data was developed using Borland Turbo C. There are programs to perform the following functions:

- Test specific hardware interface functions
- Display the CCD output on an oscilloscope
- Read the CCD output data into a file
- Plot the CCD output data on the computer monitor
- Calculate the amount of dark pixel noise in the CCD output
- Interpret the CCD output of a bar code scan
- Format the CCD blood tube data for analysis by software algorithms used in the QBC® Autoread

8.1 Hardware Interface

The following programs are used in testing the CCD computer interface board:

COUNTER loads an unsigned 16-bit integer into the integration time counter on the CCD computer interface board.
READ reads the values stored in the SRAM, and writes them to standard output in hexadecimal notation. Since each data value consists of two bytes (16 bits), the memory address must be incremented by two to read successive data values.

WRITE writes an unsigned 16-bit integer to all addresses in the SRAM on the CCD computer interface board\(^1\). The same number is written to all locations in the SRAM.

### 8.2 CCD Control

The following programs are used to control the CCD.

START sends a “START” signal to the CCD board. A logic 1 is loaded into a latch on the CCD computer interface board. If the latch previously contained a 0, the “START” signal marks the beginning of an integration time which will last the length of time corresponding to the number last loaded into the integration time counter. Otherwise, no action occurs.

STOP sends a “STOP” signal to the CCD computer board. A logic 0 is loaded into a latch on the CCD computer board to ready it for a “START” signal. STOP may be called 2 \(\mu s\) after START is executed.

RUNCCD loads a value corresponding to a specified integration time into the integration time counter on the CCD computer interface board. The integration time in milliseconds is specified in the file INT.INP; this value is converted into an unsigned 16-bit number which is loaded into the middle bits of the 24-bit

---

\(^1\)In testing, it was found that the same values were not always read from the SRAM using READ as were written to the SRAM using WRITE. The problem appeared to be with the WRITE procedure. To test this, the input to the ADC was tied to a steady voltage (ground, 2.5 V, and 5 V), and a START signal was sent to the computer interface board to force the loading of the ADC output data into the SRAM. READ was used to look at the SRAM values, and it produced a steady output with not more than one bit of variance (which is attributable to noise in the analog-to-digital conversion. Writing to the SRAM from the computer bus may have been unpredictable because of bus and memory write timing issues. However, the other tests were a good indication that valid data was written to the SRAM by the ADC, and could reliably be read from the SRAM by the computer.
integration time counter. Then, the time for the integration time counter to count to its maximum value with a clock frequency of 500 kHz is the integration time, which is the closest possible approximation to the time specified in INT.INP. The program runs in a continuous loop, scanning the CCD using the specified integration time, and converting and storing the data. The data in the SRAM is overwritten with each scan, and is not stored in a file in the PC. The program is halted by typing “ESC.”

The repetitive nature of RUNCCD enables the output of the CCD to be viewed on an analog oscilloscope. The oscilloscope is triggered on a signal in the system with the same frequency as the scan rate of the CCD: the START input to the CCD computer interface board, which starts the scan process; \( \Phi_{ROG} \), which determines the integration time of the CCD; or the DAV output from the CCD computer interface board, which indicates when new data is available in the SRAM. The output of the CCD (taken at the input or output of the op amp circuit) is displayed on the scope.

### 8.3 Data Capture and Display

The following programs are used to read data from the SRAM and display it on the monitor.

**READCCD** reads the values stored in the SRAM and converts them from 2’s complement representation (which is the output of the ADC) to signed representation. READCCD can be used after exiting RUNCCD to capture the last set of data stored in the SRAM during execution. RUNCCD followed by READCCD has the same effect as READDATA.

**GRAPH** plots the contents of a specified data file. The data file should consist of a list of newline-separated floating-point numbers. The maximum number of points in the data file is 8192 (which is the number of values stored in the SRAM). The graph color is white. The values in the data files are assumed
to be y-values, which are plotted against their indices in the data file. No scale is printed on the graph. If GRAPH is called with the name of a file, it plots the data in that file. Otherwise, it plots the data in the default data file DATA.TMP. The x- and y-scaling may be changed by modifying the parameters in the input file PLOT.INP. The plot is held on the screen until "ESC" is hit. GRAPH uses the Turbo C library file GRAPHICS.LIB.

**READDATA** is called with an argument of a desired integration time in milliseconds. The program loads a 16-bit unsigned integer corresponding to this integration time into the integration time counter on the CCD computer interface board. The data from the CCD scan\(^2\) is converted into signed representation and stored in the specified file. If no filename is specified, the data is stored in the default file DATA.TMP.

**PLOTDATA** is used in conjunction with READDATA in normal operation of the prototype CCD subsystem. This program plots the contents of one or two data files on a fixed-scale graph. The data files should contain 8192 newline-separated floating-point values in the range \(-2048\) and \(+2047\), which is the normalized range of the ADC output. The data files are assumed to contain y-values, which are plotted against their indices in the file. Data from the first file is plotted in red; if a second data file is specified, the data from that file is plotted in green. If no files are specified in the command line, the default file DATA.TMP is used. No scale is printed on the plot. The plot is held on the screen until "ESC" is hit.

PLOTDATA uses the Turbo C library file GRAPHICS.LIB.

---

\(^2\)Actually, two scans are taken with the CCD. It was found that after the CCD had been sitting idle for a long period of time, one scan was not sufficient to shift out the accumulated charge. READDATA takes two scans in quick succession, which gives a valid reading.
8.4 Data Processing

The following programs are used to process and interpret the data output from the CCD.

**NOISE** calculates the noise, in millivolts, produced by the CCD. It is assumed that the input data to this program is taken while the CCD is completely blocked from light, so that the output should be flat except for dark noise variations. The data is adjusted for amplification in the op amp circuit so that the noise value produced reflects the noise in the unamplified output of the CCD. An example of the use of this program is shown in Section 9.1.1.

Since there is some expected offset between the odd and even pixels due to the two separate shift registers in the CCD, the noise for the odd and even pixels is found separately, and the larger of the two numbers is output. If there is minimal noise in the CCD circuit, the output noise value should be comparable to the DSNU (Dark Signal Non-Uniformity) of the CCD.

If NOISE is called with the name of a file, it finds the noise in the data in that file. Otherwise, it uses the default data file DATA.TMP.

**BARCODE** interprets the data from a CCD scan of a patient ID bar code, and outputs the ascii numbers corresponding to the bar code. If it is unable to interpret the bar code, it outputs some status information about the lines it was able to read. This program was written for the project by David Barrett. A detailed description of the bar code may be found in Appendix A.

**WRITEQ** reads a set of files corresponding to a complete set of CCD scans on a single tube, and writes them to an output file of a format which can be used by the Autoread software. A detailed description of the Autoread data software and file formats is given in Appendix B.
Chapter 9

QBC® Walkaway Prototype

Results

The results of testing done on the QBC® Walkaway prototype optical subsystem were promising.

- The Walkaway output contains more noise than the QBC® Autoread output, but the excess noise may be due to several factors: misalignment of the optics; misfocusing and rolloff of the lens; uneven lighting; the sockets on the CCD PCB; and the long cable connecting the two CCD boards. All of these situations should be corrected or improved in the final product. If the noise cannot be eliminated, much of it can be filtered out in software or in hardware.

- Although the ends of the tube are cut off in the scans, an optical lens adjustment in the Walkaway prototype will allow the entire tube to be focused on the CCD.

- The use of a CCD in the Walkaway is an improvement—at least for the mechanical subsystem design—over the single photodetector used in the Autoread, in front of which the tube was moved.

- The output data from the CCD is comparable to data from the Autoread, and if CCD quality and resolution continues to improve as it has in the past few years, the Walkaway will produce even more sensitive and accurate results when
the final machine is put on the market.

9.1 Test Data

9.1.1 CCD Dark Noise Calculation

The software program NOISE, described in Section 8.4, was used to estimate the noise in the CCD output data. With an input data file taken when the CCD was blocked from light, this program estimates the CCD output noise in volts, correcting for the amplification of the op amp. The odd and even pixel noise is calculated separately, to eliminate the offset error between shift registers, and the maximum of the two noise values is output.

Figure 9-1 shows the output of CCD with the pixel window blocked from light with black electrical tape. The CCD dark noise value given in the Sony data sheets uses an integration time of 10 ms; however, an integration time of 20 ms was used for this test, as the CCD output is unpredictable for integration times less than 16 ms in this control configuration.

The noise estimated by the program for this output was about 21.5 mV. The expected DSNU (dark signal non-uniformity) of the CCD, according to the data sheets, is 0.6 mV, and the maximum is 5 mV[15].

Some of this excess noise is attributable to the longer integration time, to the fact that the saturation output peak-to-peak voltage of this CCD is larger than specified in the data sheets (2 V compared to 1.5 V), and to noise in the op amp and in the ADC. In addition, the electrical tape was not completely successful in blocking the CCD from light; the output is higher than the dark reference pixels, and there is an upward trend in the digitized CCD output. However, these factors don't account for the observed digital output noise being an order of magnitude over the expected variance in the output. The measured noise at the analog output of the CCD was also on the order of 20 mV for a dark input and an integration time of 20 ms, although in this case the oscilloscope probes may have added to the noise.
As a reference, the DSNU was also computed for the 80 dark reference pixels of the CCD output. The dark reference pixel output for a randomly selected scan is shown in Figure 9-2.

Surprisingly, the noise in the dark reference pixels (during an illuminated scan) was greater than the noise in the output when the CCD was blocked from light. The NOISE program computed the DSNU for the dark reference pixels in this scan to be 27.0 mV.

Several experiments were tried to reduce the noise in the CCD output. A resistor/capacitor filter was added at the input of the op amp circuit, but this didn’t have a significant effect on the noise for values of RC which didn’t obliterate information. Several decoupling capacitors were added to the board with no noticeable effect. The ±12 V power supplies from the computer bus were a potential source of noise, so they were disconnected and the board was tested with an external power supply. However, the external supply did not appear to make a difference in the output noise, and it added an extra component to the optical configuration, so the board was reconnected.
to the computer bus supply voltages. It was thought that the 8 MHz clock signal (the start conversion signal to the ADC) on the CCD PCB might be a source of noise, as the frequency of this signal was 16 times faster than the system clock, but the noise at the CCD output does not appear to have a significant component at 8 or 16 Mhz.

It was finally decided to continue testing the optical subsystem with the CCD noise at the same level. This would determine if the CCD implementation was feasible even if the noise was inherent in the system, as well as allowing the testing process to move forward. It is possible that the CCD specifications underestimated the noise; the Sony ILX508 CCD chip is relatively new and had not been long on the market when one was ordered for the project.

9.1.2 Calibration Tube Readings

Three types of calibration tubes, commonly used in calibrating the QBC© Autoread, were used to test the Walkaway prototype optics.
9.1.2.1 Green Stripe Calibration Tube

The green stripe calrod, which was tested in the preliminary prototype described in Section 4.2.1.1, is the size of an E-Z-Prep tube, and is etched at 0.25 in. intervals around its circumference with 0.004 in. grooves which are filled with green coloring. The green stripe calrod may be viewed either under fluorescence lighting through a green filter, or with transmission lighting. This calibration tube is not generally used as a reference because there are imperfections in the grooves and the coloring which produce an unreliable output.

The green stripe calrod was scanned using fluorescence lighting with a green filter. This data is shown in Figure 9-3. The green filter causes the green stripes to appear brighter than the rest of the tube, so the stripes are the peaks in the data.

In an ideal scan of a green stripe calrod, the peaks would all be the same height, and the background would be flat. The illumination for this scan may have been uneven, and with the positioning and size of the lens, it was not possible to focus the
Green Stripe Calibration Tube

管子图像在CCD上，这样中心和两端的管子都在对焦。不过，这还是一个相当好的输出。

图9-4是图9-3的扩展图，显示了一个绿色条纹。输出峰的顶部是平坦的，两侧的斜率是陡峭的，
表明了透镜质量和CCD分辨率。

数据还采用了绿色条纹的校准管，使用透射光照明获取。在这组数据，如图9-5所示，
绿色条纹比管子的其他部分显得更暗，所以条纹是数据中的凹槽。

9.1.2.2 黑白校准管

黑色和白色校准管也在初步原型中进行了测试。此校准管的尺寸与E-Z-Prep管相同，
包含交替的黑和白棒，每根的长度约为血液带在其位置对应管子的长度。此管在读取

Figure 9-4: QBC® Walkaway Prototype Scan of a Green Stripe Calibration Tube (Green Fluorescence, Expansion)

tube image on the CCD so that the center and the ends of the tube were all in focus. Nevertheless, this is a reasonably good output.

Figure 9-4 is an expansion of the plot in Figure 9-3, showing one of the green stripes. The top of the output peak is flat and the slopes of the sides are steep, demonstrating the lens quality and the CCD resolution.

Data was also taken of the green stripe calrod using transmission lighting. In this data, shown in Figure 9-5, the green stripes appear darker than the rest of the tube, so the stripes are the dips in the data.

9.1.2.2 Black and White Calibration Tube

The black-and-white calrod was also tested in the preliminary prototype described in Section 4.2.1.1. This calibration tube is also the size of an E-Z-Prep tube, and contains alternating black and white rods, each of which is the approximate length of the blood band corresponding to its position in the tube. This tube is read with
transmission lighting, so that low output values correspond to the black rods and high output values correspond to white rods. The output from a scan of the black-and-white calrod is shown in Figure 9-6.

Again, in this calibration tube data, there is background noise and some rolloff at the ends of the tubes. However, the interfaces between the black and white “bands” in the tube are sharp, and the tops of the bands are flat. An expansion of the “buffy coat” area of the black and white calrod, which is represented by the three thin rods near the center of the tube, is shown in Figure 9-7. Good optical resolution is exhibited by the high-sloped sides and the flat tops.

9.1.2.3 Bar Code Calibration Tube

The bar code calrod is a recently-developed calibration rod which has a different form than the green stripe and black-and-white calrods. It is the same length as an E-Z-Prep tube, but the body of the calrod is only half-cylindrical, with a flat plane along
Figure 9-6: QBC® Walkaway Prototype Scan of a Black and White Calibration Tube

Figure 9-7: QBC® Walkaway Prototype Scan of a Black and White Calibration Tube (Expansion)
the longitudinal axis of the rod. The flat plane has a reflective surface, on which a
dense “bar code” image is printed. The bar code image is positioned at a 45° angle
to the fluorescence light source, and is read in green fluorescence lighting.

The bar code calrod is more precisely manufactured than the green stripe calrod.
The bar code consists of alternating black and green stripes of width 0.010 inches,
except for three 0.020 in. black stripes, two near the center and one near the end of
the bar code. Before the bar code, there is a wide green stripe, and after the bar code
on the other side, the calrod is black. Figure 9-8 shows the Walkaway prototype data
from this calrod.

Although it is not possible to see individual 0.010 in. bars in this plot, the three
0.020 in. black stripes are visible. Ideally, the heights of all the bars would be the
same; however, there is only slight rolloff due to the lens edge at the end of the bar
code.

A closer view of the calrod is shown in Figure 9-9. Two of the three 0.020 in.
black bars are shown in comparison with 0.010 in. black and green bars. This data
Figure 9-9: QBC® Walkaway Prototype Scan of the Bar Code Calrod (Expansion)

looks quite good; there is little noise, the edges of the bars are squared off, and the high and low values are constant and flat.

9.1.3 Hair Readings

A human hair, at about 0.002 in.–0.004 in. represents the approximate width of the smallest band that the Walkaway should be able to detect. A hair measured at 0.002 in. was placed against the background of the bar code calrod—once in the middle of a 0.010 in. green band, and once in the wide green band at the top of the calrod. The expanded outputs of these scans, taken in green fluorescence lighting, are shown in Figures 9-10 and 9-11. The “dip” in the data corresponding to the hair is unmistakeable in both plots, although it is better defined when the hair is in front of the level background.
Figure 9-10: QBC® Walkaway Prototype Scan of a Human Hair (Bar Code Background)

Figure 9-11: QBC® Walkaway Prototype Scan of a Human Hair (Level Background)
9.1.4 Blood Tubes

Since the purpose of this machine was to read and analyze blood tubes, much of the optical testing involved blood. Tests were done both with healthy blood and with blood which had been artificially altered to appear "sick"; i.e. one or more of the bands was abnormally small or non-existent.

9.1.4.1 Normal Blood

For reference, QBC® Autoread scans of normal blood tubes are shown in Figures 9-12–9-16. The different Autoread scans are described in Appendix B.1.1. Autoread results for this tube are given in Table 9.1

Figures 9-17–9-19 show the transmission and red and green fluorescence scans of the Walkaway prototype machine on a normal blood tube.

Expansions of the float area in the Walkaway fluorescence scans are shown in Figures 9-20 and 9-21, to better display the buffy coat.
Figure 9-13: QBC® Autoread Plasma Scan of a Normal Blood Tube

Figure 9-14: QBC® Autoread Transmission Scan of a Normal Blood Tube
Figure 9-15: QBC® Autoread Red Fluorescence Scan of a Normal Blood Tube

Figure 9-16: QBC® Autoread Green Fluorescence Scan of a Normal Blood Tube
<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hematocrit:</td>
<td>46.1 %</td>
</tr>
<tr>
<td>Hemoglobin:</td>
<td>15.9 g/dL</td>
</tr>
<tr>
<td>MCHC:</td>
<td>34.5 g/dL</td>
</tr>
<tr>
<td>Platelets:</td>
<td>$176 \times 10^9$/L</td>
</tr>
<tr>
<td>White cells:</td>
<td>$5.3 \times 10^9$/L</td>
</tr>
<tr>
<td>Granulocytes:</td>
<td>$64 %$</td>
</tr>
<tr>
<td>Lymphocytes/Monocytes:</td>
<td>$36 %$</td>
</tr>
</tbody>
</table>

Table 9.1: QBC® Autoread Results for a Normal Blood Tube

![Blood Tube (Transmission)](image)

Figure 9-17: Walkaway Transmission Scan of a Normal Blood Tube
Figure 9-18: Walkaway Red Fluorescence Scan of a Normal Blood Tube

Figure 9-19: Walkaway Green Fluorescence Scan of a Normal Blood Tube
Figure 9-20: Walkaway Red Fluorescence Scan of a Normal Blood Tube (Expansion)

Figure 9-21: Walkaway Green Fluorescence Scan of a Normal Blood Tube (Expansion)
The buffy coat regions in the Walkaway prototype scans shown in Figures 9-20 and 9-21 are not as well-defined as the Autoread scans shown in Figures 9-15 and 9-16. The Walkaway prototype optics have the resolution and sensitivity to produce good readings of the buffy coat; however the blood tube may have been held for too long before taking those particular scans.

Unfortunately, that set of prototype tube scans is one of the only saved sets which includes transmission data; only fluorescence data was obtained in most of the tests\(^1\). A better example of Walkaway fluorescence scan data is shown in Figures 9-22 and 9-23, with the float area expanded in Figures 9-24 and 9-25. The optical alignment of the system was slightly different for these scans; the cap end of the tube is visible, and the fluorescence lamp is not as bright.

Even though the CCD output is noisy and the lighting is slightly uneven, the

---

\(^1\) A color filter wheel was used to provide the red and green filter wavelengths in the preliminary prototype; however, as it was difficult to switch the filter wheel in and out of the optical path, transmission data was not generally taken.
CCD still produces a clear reading of the buffy coat bands in a blood tube. This is the most important requirement of the optics in the prototype machine.

9.1.4.2 Altered Blood

In other tests, several tubes of blood were chemically altered to appear abnormal. Expanded Walkaway prototype red and green fluorescence scans of a blood tube with a practically non-existent granulocyte band are shown in Figures 9-26 and 9-27.

For comparison, QBC® Autoread float scans of the same tube are shown in Figures 9-28–9-29.

9.2 Conclusions

The QBC® Walkaway will require a significant amount of work to become a finished product, but the indications are that it will be successful. The implementation of
Figure 9-24: Walkaway Red Fluorescence Scan of a Normal Blood Tube (Expansion, Better-Defined Buffy Coat Region)

the CCD was an important step, which will ultimately increase the resolution and upgradability of the QBC® machines.

Moreover, all of the students involved with the project acquired valuable experience in design and product development, and Becton Dickinson, Inc. gained a different perspective on their new product. Hopefully the New Products Program will continue to offer students and companies this rewarding opportunity.
Figure 9-25: Walkaway Green Fluorescence Scan of a Normal Blood Tube (Expansion, Better-Defined Buffy Coat Region)
Blood Tube (Red Fluorescence)

← CCD Output

Figure 9-26: QBC© Walkaway Prototype Red Fluorescence Scan of an Altered Blood Tube
Figure 9-27: QBC® Walkaway Prototype Green Fluorescence Scan of an Altered Blood Tube

Figure 9-28: QBC® Autoread Red Fluorescence Scan of an Altered Blood Tube
Figure 9-29: QBC® Autoread Green Fluorescence Scan of an Altered Blood Tube
Appendix A

Bar Code

A.1 Symbology

The bar code symbology used for the patient ID bar codes in the prototype QBC\textsuperscript® Walkaway machine is called “interleave 2 of 5.” Four types of bars are used: alternating black and white bars of two different widths.

Digits, or characters, are encoded in pairs. A sequence of 10 bars (5 black and 5 white) contains two digits, one encoded in the black bars, and one encoded in the white bars.

For each sequence of five bars of a single color, two of the bars are wide. Their placement among the three narrow bars determines the coded digit. The coding is only numeric.

A “start character” and a “stop character” mark the beginning and end of the bar code, and are used in determining the orientation of the bar code. The start character is 4 bars: two narrow black bars interleaved with two narrow white bars. The stop character is three bars: a wide black bar, a narrow white bar, and a narrow black bar.
A.2 Specifications

A.2.1 Optical Considerations

The bar code is placed on a plastic flag extending from the side of the tube. It is not possible to bring the flag image into focus on the CCD without the addition of an extra motor and a more complicated optical configuration. However, a sufficiently large unfocused bar code may still be “readable” by the optics. In order for a software program to interpret the bar code scan output, the narrow lines of the bar code must be wide enough to be distinct from each other in the output, and the wide lines must be wide enough to be distinguished from the narrow lines of the same color.

The prototype E-Z-Prep tube flag is long enough to hold a 2.58-inch bar code (the Vac-Q-Tube flag is long enough for a three-inch label). It was decided that 8 digits of code would sufficiently reduce the possibility of duplicate tubes being processed in the same day. Each eight-digit bar code contains 17 wide lines and 30 narrow lines, including the start and stop characters. After some preliminary testing, the widths of the lines were set at 0.032 inches for narrow lines, and 0.064 inches for wide lines, giving an overall bar code length of 2.048 inches.

A.2.2 Patient ID Specifications

An external bar code reader was purchased by the project to complete the prototype patient ID system. The bar code reader was an inexpensive, hand-held “pen,” American Microsystems model 1000/1002, with a computer keyboard interface. When the bar code reader is drawn across a bar code, the ascii characters corresponding to the code are sent to the computer’s standard input as though the characters were entered from the computer keyboard. The bar code reader is capable of interpreting seven different types of bar codes, including the interleaved 2 of 5 symbology.

In the patient ID system developed for the prototype Walkaway machine, each tube flag has a bar code label attached to it, and comes with another label with the same bar code which may be affixed to the patient’s records. The medical technician
may enter patient information, such as name, age, and gender, into the machine at any time before, during, or after the tube processing cycle. The medical technician may use the external hand-held scanner either to scan the bar code on the patient records, or to scan the bar code on the tube flag itself. Since the bar code on the tube flag is larger than standard bar codes, the external scanner is not as easy to use on the tube bar code; the scanner must be moved quickly across the bar code in order to compensate for the wider lines.

A.3 Software Interpretation of the Bar Code

A software program to interpret the CCD output of a bar code scan was written by David Barrett. This program takes as input a text file containing the converted output of the CCD, and outputs the numeric ascii translation of the corresponding bar code. If it is unable to interpret the barcode, it outputs some status information.

Unfortunately, this program was based on some early bar code scan outputs, and is not robust enough to adapt to later data after the bar code widths were changed slightly and some optical adjustments were made. The program uses a level algorithm to distinguish between black and white stripes; it may be improved by implementing a derivative algorithm so that the absolute value of the bar code data does not matter.

A.4 Data

Figure A-1 shows a prototype bar code which was used on an E-Z-Prep tube flag, along with the corresponding CCD output. Figure A-2 shows another prototype bar code used on a Vac-Q-Tube flag, along with the CCD output of this bar code scan. Low values in the plots correspond to black lines, and high values correspond to white lines. The plots show that even though the bar code image is out of focus on the CCD, the wide and narrow bars are easily distinguishable.
Flag Bar Code (E-Z-Prep Tube)

CCD Output

Figure A-1: CCD Scan of a E-Z-Prep Flag Bar Code
Flag Bar Code (Vac-Q-Tube)

\[\text{CCD Output}\]

![Graph showing a CCD scan of a Vac-Q-Tube Flag Bar Code]

01234566 (reversed)

Figure A-2: CCD Scan of a Vac-Q-Tube Flag Bar Code
Appendix B

Band Length Calculation

The QBC® Autoread software includes a set of algorithms to determine the counts and percentages of different types of cells in the blood from the tube scan output. In order to use this software, the prototype Walkaway tube data needed must be put into the same format as Autoread data.

B.1 QBC® Autoread

B.1.1 Tube Data

Like the prototype Walkaway machine, the Autoread uses two light sources, one for transmission readings, and one for fluorescence. The fluorescence light source is a miniature tungsten lamp with a 480 nm (blue) interference filter. The transmission light source is a 610 nm (red) LED[2].

The Autoread takes several scans of different regions of the tube. Because this machine physically moves the tube in front of a photodetector, it is possible for the Autoread to read only a relevant section of the tube each scan. The following scans are taken:

**Float**  The fluorescence lamp is used to scan the float area of the tube eight times, at 45-degree intervals around the circumference of the tube. Each tube position is scanned twice, using red and green filters, for a total of 16 data sets; each
data set consists of 1300 points. This data is used to calculate band lengths in the float region.

**Transmission**  One 5000-point scan covering the length of the tube is taken using the transmission lamp. This data is used to determine the tube type, float position, and closure type.

**Plasma**  The plasma region of the tube is scanned using the transmission lamp. The resulting data set consists of 3200 points. This data may be used to determine the fill volume of the tube.

**Cap**  The cap end of the tube is scanned using the transmission lamp. The resulting data set consists of 400 points. This data may be used to determine the cap type. (There are two different types of caps commonly used with QBC® tubes: a plastic cap which fits over the end of the tube, and a small rubber insert which fits inside the tube.)

The data from the photodetector is converted to digital format with a 12-bit ADC. The raw data ranges in value from 0–4095. A high value indicates a brightly illuminated image; a low value indicates a dark area.

The Autoread software stores all of the data sets corresponding to a single tube in one binary file; this type of file can store a large amount of information compactly. Besides the raw tube data, each binary file also contains scan information such as the tube and closure types, and calculated results such as cell counts.

### B.1.2 Software Algorithms

The scan data sets are processed using several numeric techniques:

- Gaussian filtering
- Median filtering
- Taking the derivative
Figure B-1: Packed Cell Layers in a QBC® Capillary Tube

- Normalization of red and green float scans
- Addition or subtraction of red and green float scans
- Ratioing the average values of two bands

The transmission and plasma scans are processed to determine the tube type, as well as whether the tube has been filled correctly and properly inserted into the machine. Each set of red and green fluorescence float scans taken at the same tube position is processed independently to find four band length values: \( L_2 \), \( L_3 \), \( L_4 \), and \( L_5 \). Length \( L_1 \) is found from the transmission scan and length \( L_6 \) is found from the plasma scan. The appearance of these six length values in the tube is shown in Figure B-1 for a capillary tube, and in Figure B-2 for a venous tube.

A float scan may be classified as “unreadable” for reasons such as a blood clot, a piece of dirt on the tube, or the blood tube having stored too long. If more than four of the eight float scan pairs are unreadable, no cell counts are found for the data. Otherwise, band lengths are computed for all of the readable sets of red and green
scans, using filtering to smooth the data and derivatives to locate band interface edges, and subtracting the red and green scans to clarify the bands within the buffy coat. The computed band lengths for all of the readable float scans are averaged together to produce normalized values for $L_2$, $L_3$, $L_4$, and $L_5$. This reduces the effects of wavy or tilted bands.

Since the tube, float, and nominal cell diameters are known, the six band lengths are used to determine counts and percentages of different cell types. The Autoread software formulas make use of specific band lengths to find values for each cell type:

- **Hematocrit:** $L_1$ and $L_2$
- **Hemoglobin:** $L_1$, $L_2$, $L_3$, $L_4$, and $L_5$
- **Platelets:** $L_5$
- **WBC:** $L_3$ and $L_4$
- **Granulocytes:** $L_3$
- **Lymphocytes/Monocytes:** $L_4$
**Hematocrit** is the percentage of the red cell volume to the total blood volume. The **hemoglobin** value is the percentage of hemoglobin in the cell makeup. Hemoglobin is calculated using the distance that the float sinks into the red blood cells to determine the density of the packed red cells. The RBC density can then be used to find the hemoglobin parameter.

The **lymphocytes** and **monocytes** are counted together, and are also referred to as **non-granulocytes**. Together, the **granulocytes** and non-granulocytes make up the white blood cells. The Autoread is a two-part differential analyzer, which means that it distinguishes between two types of white blood cells.

The Autoread outputs the following cell count and percentage values:

- Hematocrit (percentage)
- Hemoglobin (absolute measurement)
- Platelets (absolute count)
- White blood cells (absolute count)
- Granulocytes (percentage of the total white blood cell count, and absolute count)
- Non-granulocytes (percentage of the total white blood cell count, and absolute count)
- Mean corpuscular hemoglobin concentration (absolute measurement)

The **mean corpuscular hemoglobin concentration**, or MCHC, is a unitless ratio of hematocrit and hemoglobin.

**B.2 QBC® Walkaway Prototype**

The data obtained from the Walkaway prototype is quite different in format from Autoread data. CCD data read from the SRAM is converted in the computer to a
signed representation, with values ranging from $-2048$ to $+2047$. This data is stored in a specified file in an ascii list format. A different file is used for each tube scan: one transmission scan, eight fluorescence scans with a red filter, and eight fluorescence scans with a green filter. Since the Walkaway prototype uses a linear CCD to capture the tube image, it isn’t possible to scan only part of a tube.

As noted in Section 3.5.2, the tube is indexed by a lead screw which pushes the tube against a spring-loaded holder cup, so that the tube moves axially by a small amount with every 45-degree rotation during the fluorescence scans. Initially, provision was made to correct for the shift distance. However, since the Autoread processes each float scan separately and only averages the resulting band lengths, slight offsets between scans shouldn’t make a difference to the Autoread software.

The CCD allows for 7926-point precision along the length of the tube; however, the Autoread software only expects 5000 points. In order to use the Autoread software in the prototype machine, some of the CCD precision was sacrificed. The 7926 points were converted into 5000 by approximating the 7926-point file as a continuous, piece-wise linear function. At 5000 equally-spaced points within this function, the data was sampled, producing the 5000-point precision needed for the Autoread software. Subsets of this data corresponding to the different Autoread scans were taken, and the data sets were compounded into one binary file in the Autoread format.

However, there were other factors which make the conversion of prototype data into the Autoread format difficult:

- The tube magnification in the Walkaway optics is different than that of the Autoread, so that the Walkaway tube appears longer in the scan data. The transmission light source for the Walkaway is white light, which produces a different output than the red light used in the Autoread: the same features are present, but the relative levels are different, and in some transitions a falling edge replaces a rising edge (or a rising edge replaces a falling edge). The software will not find the same features in this data.

- Although the Walkaway and the Autoread data have the same maximum range
(the Walkaway data can easily be transposed up 2048 points to match the Autoread range of 0–4095), the levels of the data are significantly different, due to differences in the light sources, filters, amplifiers, and photodetectors. The Autoread software looks for some bands or peaks to be within certain level limits; the Walkaway data must be altered to match those specifications.

- The optical alignment of the prototype is slightly miscalculated; the tube and the CCD are too far from the lens, so that the ends of the tube image extend off the CCD. The Autoread software looks for the cap at one end of the tube, and the top fill line at the other end, to establish the fill volume and orientation of the tube. If these features are missing from the data, the software is unable to calculate band lengths.

Due to these obstacles, the software program written to translate the prototype Walkaway data into a format useable by the Autoread software was never implemented successfully. A partial solution translates Walkaway data into the Autoread format without correcting for the transmission, level, and tube-end problems. The data may then be plotted and processed using interactive Autoread software.
Bibliography


