SMARTPHONE HEMOCYTOMETER DESIGN DOCUMENT

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1 Introduction

1.1 Objective

White blood cell counts and classification are critical measurements in medical settings as it alerts doctors to autoimmune diseases, immune deficiencies, and blood disorders and helps monitor the effectiveness of chemotherapy and radiation therapy for cancer. Most of the doctors are using hemocytometers with microscopes to differentiate and count the number of white blood cells in a counting chamber, which is both bulky and inconvenient. In some developed countries, this task is performed by means of an automated blood cell counter. But neither method is practical in terms of cost and convenience. [Ose+09]

The goal of our project is to design and build a portable smartphone hemocytometer that enables testing and documentation at the point of care. Using the smartphone's camera as the microscope, the project creates an automatic platform for taking panoramic microscopic images of the blood sample. On the electrical subsystem side, the smartphone hemocytometer consists of a power supply module that provides power for all devices in the circuit, a motion module of high precision linear actuator to drive the blood smear on a motion stage, a microcontroller as the control module, and a communication module that serves as a bridge between the smartphone and the microcontroller. On the software side, an Android application will be devised to communicate with the micro-controller and to take photo. This innovation has a large advantage in efficiency because it will cut doctors' time sending the blood sample back and forth for lab work. It also has advantage in complexity in operation because it saves doctors from operating complicated optical microscopes and in availability thanks to the wide popularity of smartphones. The project also provides a foundation for future development so that eventually doctors can run a machine learning identification algorithm on the hemocytometer to differentiate the five different types of white blood cells and complete said important item from blood test that would have taken hours.

1.2 Background

The innovation of a 3D-printed smartphone microscope lens by Pacific Northwest National Laboratory (PNNL) testifies to the feasibility of turning a smartphone into a microscope. [Hut+15] From this research detecting Bacillus Anthracis, which is a spore around 1 micron in length, the researchers were able to identify 50 to 5000 spores in 3 to 5 hours. This research demonstrates that the lens has high enough resolution to identify blood cells which has larger dimensions and that it is still slow to identify the cells manually. Our project will provide an automated system to establish a stabilized and accurate motion system to coordinate with the smartphone in order to best utilize the clarity of the PNNL microscope lens.

Although the clip on lens that turns phone cameras into microscopes has been on the market for a while, there is no automated system that provides an integrated solution. To our knowledge, there is no smartphone-based microscope that can take a panoramic image of a blood cell sample. The non smart phone based portable microscope usually costs more than $600. Our product will be the first to create a low cost solution to address the needs of many emergency care physicians, veterinarians, and more.

1.3 High-Level Requirement

There are three major requirements for our design:

1. Mechanical Resolution

   Since we need to take high magnification images, the field of view of one frame is very small. We need to pick a stepper motor that has the step precision to move the sample within the field of view of one frame. Based on our calculation below, the field of view for 40X magnification is around 100um * 100um. We need to make sure when the motor moves, there is no gap between frames of the white blood cell images so that they can be stitched to a panorama photo.

2. Motion Control and Stability

   The microcontroller should give precise control to the stepper motor. It needs to provide high frequency (up to 16MHz) PWM and digital signals to the motor driver to control the steps and direction of the linear movement. It should also guarantee enough buffer time (at least 0.5s) between frames to ensure the motion stage has finish the movement when the phone is taking photo.

3. Visual Quality

   We need to fix the phone camera's focus length so that it does not need to refocus on the blood smear sample during the lateral movement.

2 Design

2.1 Block Diagram and Physical Diagram

2.1.1 Block Diagram

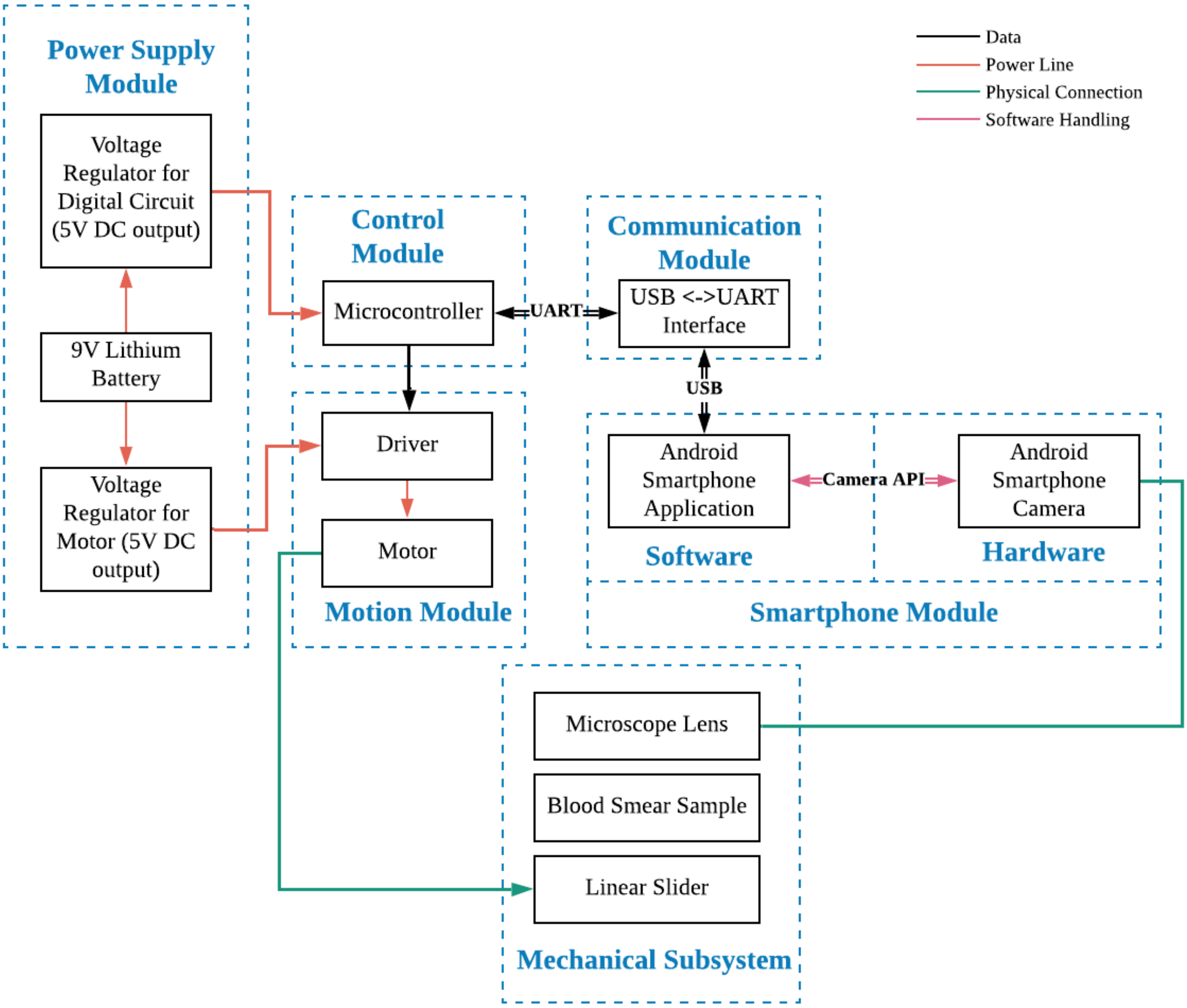


Figure 1: Block Diagram

2.1.2 Physical Diagram

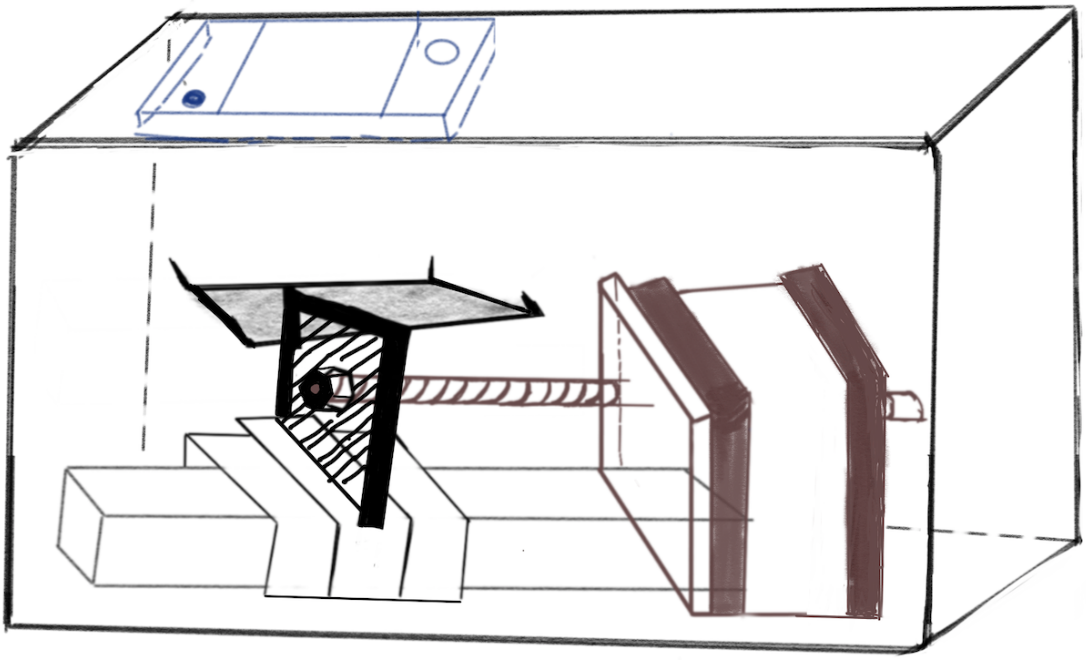


Figure 2: Physical Diagram

2.2 Functional Overview

2.2.1 Mechanical Subsystem

Ball Microscope Clip-on

Below is the mechanical design for the clip-on. We will print it using the 3D printer and then add a ball lens into the plastic housing. It is attached to the smartphone camera for 40X magnification.

Linear Slider

We plan to purchase a ball bearing slider. The linear motor will push the slider to move the blood smear sample laterally.

Blood Smear Sample

The blood smear samples are all prepared by Prof Cunningham.

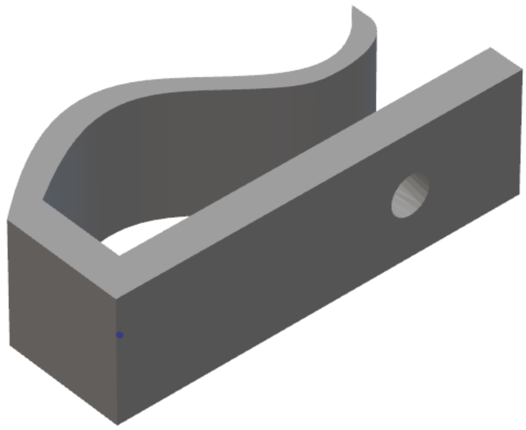


Figure 3: Microscope Clip-on [19b]

2.2.2 Electrical Subsystem

The electrical subsystem of the Smartphone Hemocytometer consists of 5 main modules: Power Supply Module, Control Module, Communication Module, Display Module and Motion Module. We will integrate them on the PCB.

Power Supply Module
Input: 9V DC battery voltage.
Output: 3.3V and 5V DC voltage.
The power supply module provides power to the motion module (motor and motor driver) and other digital circuit components.
Essential Components:

1. TPS76933 3.3 Volt Linear Regulator by Texas Instrument
2. L78S 5 Volt 2 Amp Linear Regulator by ST

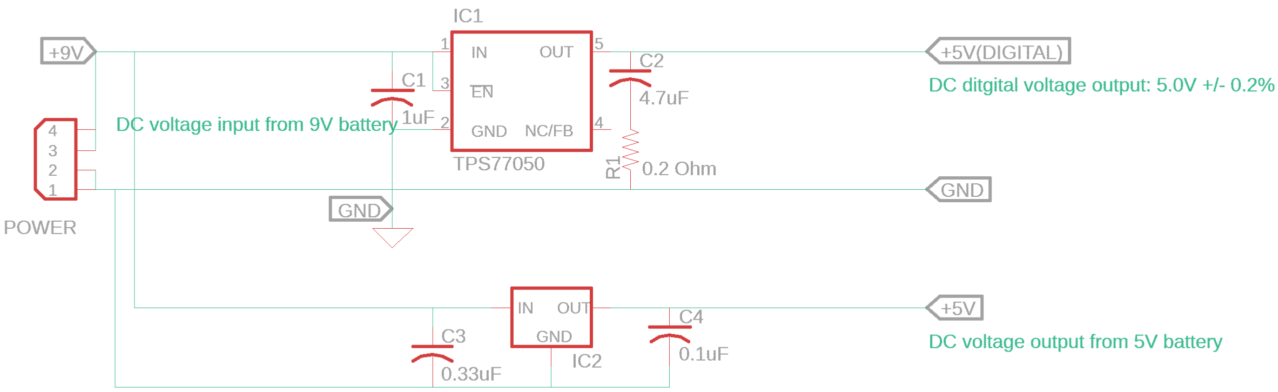


Figure 4: Power Supply Module Schematics

Pin	Function	Connection	Voltage
+9V	The main power input from 9V battery.	Connect from the battery to both regulators.	9V DC
+5V	Supply power to the motor.	Connect from the output of 5V linear regulator and extend to motion module.	5V DC
+5V (dgtl)	Act as the main power for digital circuit. Provide upto 100mV of current.	Connect From Pin 5 from linear regulator, extend to other modules.	5V DC
GND	Act as the ground voltage reference.	Connect from Pin 1,2 from the Power Connector to other modules.	0V DC

Table 1: Pin Layout Power Supply Module

Requirement	Verification
Able to convert voltage ranging from 8.7V to 9.2V to 5±0.3V and 3.3±0.1V.	Provide 9V input voltage from DC power supply to the component. The output voltage should be 5±0.3V and 3.3±0.1V.
Able to provide a max of 100 mA current to power the digital circuits.	Use a clamp-on amp meter to measure the current consumption when the device is working.
Able to provide a max of 1.3 A current to power the motor.	Use a clamp-on amp meter to measure the current consumption on digital circuit power line and motor power line when the device is working.

Table 2: R and V for Power Supply Module

Control Module

The control module contains a microcontroller responsible for controlling the motion module and display module, and communicating with the smartphone. It is powered by 5V DC from the voltage regulator in the power supply module.

Essential Components:

1. ATmega328P Microcontroller

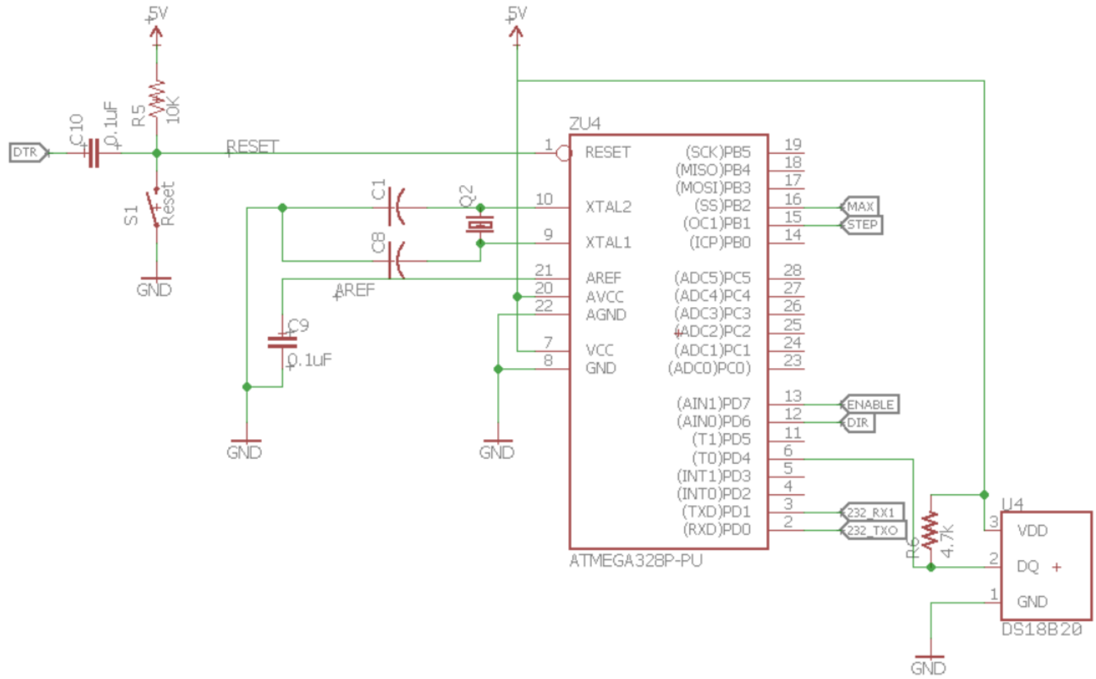


Figure 5: Control Module Schematic

ATmega328P Microcontroller

Power Input:

5V DC, operating range(1.8V - 5.5V)

Data Input:

UART RX signals from the USB-UART interface in the communication module.

Data Outputs:

Control signals and high frequency PWM signal to the motor driver in

motion module.
Control signals to the LEDs to show device status.
UART TX signals to the USB-UART interface in the communication module.

Description:
The ATmega328P is a low power, CMOS 8-bit microcontrollers based on the AVR® enhanced RISC architecture.It has frequency up to 20MHz, and contains 32KB ISP flash memory with read-while-write capabilities, 1024B EEPROM and 2KB SRAM. It has a total of 32 pins, 23 general purpose I/O lines, 32 general purpose working registers, three flexible timer/counters with compare modes. It also supports serial programmable USART. [18]
For our design, we will use a 5V output voltage regulator combined with the battery unit to supply power to the microcontroller. It can control the linear stepper motor driver using its high-speed digital outputs and timer outputs, communicate with the software in smartphone module through the USB-UART interface, and drive the LEDs to show device status.

Requirement	Verification
Must have at least 10 digital output pins for motor driver control.	Check the datasheet and pin layout to make sure the chip contains enough GPIO pins.
Able to output correct digital control signals to the stepper motor driver.	Connect the GPIO pins to an oscilloscope and check if the outputs' voltage levels match with what we specifies in the software.
Able to output 16MHz frequency PWM signal.	Connect the output pin to an oscilloscope and check the duty cycle and frequency of the waveform.
Able to both receive and transmit over UART.	Connect the UART communication pins with a SparkFun FTDI Basic Breakout board. Then connect the board to a PC terminal via USB cables. Use the Arduino software to check if we can both successfully receive and transmit UART signals.
Able to use the digital output pins to drive LEDs for showing device status.	Connect the specific digital output pins to digital multimeter and check the voltage output levels.

Table 3: R and V for Control Module

Communication Module

The communication module is responsible for building up a bidirectional bridge between the microcontroller and smartphone software. The software will send USB control signals to the embedded software on the microcontroller. The signals will be converted to UART through this interface and send to the microcontroller. For the other direction, the UART signals sent out from the microcontroller are also converted to USB to send to the smartphone.

Essential Components:

1. FT232R USB UART IC

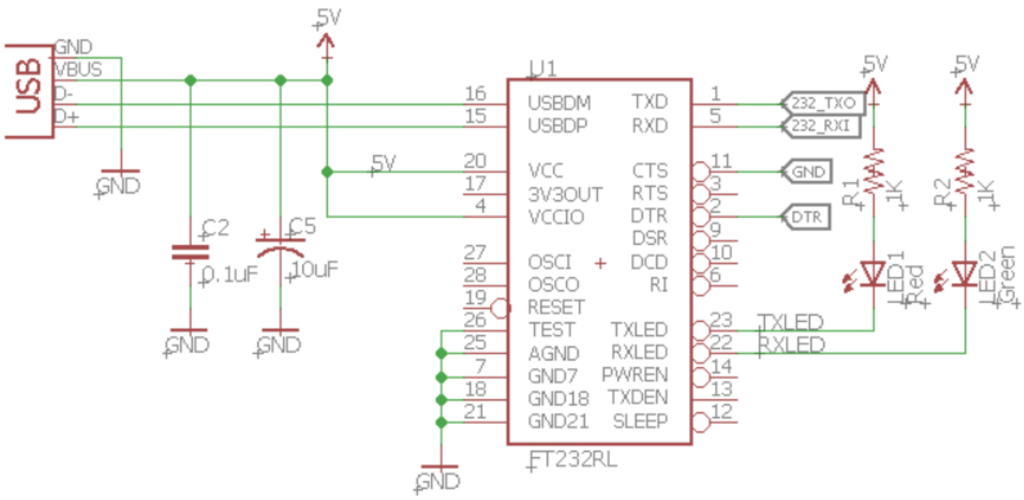


Figure 6: Communication Module Schematic

FT232R USB UART IC

Power Input:

5V DC, operating range(1.8V - 5.5V)

Data Inputs:

UART TX signals from the microcontroller in the control module.

USB signals received from the smartphone.

Data Outputs:
UART RX signals converted from USB signals to the microcontroller in the control module.
USB signals converted from UART TX signals to the smartphone.

Description:
This chip is a highly-integrated solution for USB - UART conversion that has USB transceiver, oscillator, and Universal Asynchronous Receiver/Transmitter (UART) in packages. This IC supports 5V operating supply voltage and high data transfer rate, and compatible with USB 2.0 full speed. [19a]

Requirement	Verification
Able to convert USB to asynchronous serial data and vise versa.	Check datasheet and try to connect it with the microcontroller and PC terminal to verify the data transmission accuracy.
Able to support Serial Frames with 5, 6, 7, 8, or 9 Data Bits and 1 or 2 Stop Bits.	Check datasheet.
Able to support data transfer rates from 2400 to 1M bps.	Check datasheet and try different baud rates when testing with microcontroller and PC terminal.
Have driver support for USB communication with Android.	Check datasheet and download the specific Android driver from FTDI's website. Install the driver and try sending and receiving back small data packages.

Table 4: R and V for Communication Module

Motion Module

Input: Digital control signals, 3.3 Volt digital voltage input(Vcc), and 5 Volt load supply voltage input(VC+).

Output: 4 Motor control signal pins labelled as A,B,C,D.

Communication: Logic high and low The Motion Module receives the digital control signal from Control Module and drives the motor. The input and output are labelled in green boxes. The connector is illustrated on the top right. A test LED circuit is added to verify the motion of the motor.

Essential Components:

1. A3932 Bipolar Motor Driver with DMOS logic

2. Dual Motion 43000 Series Linear/Rotary Actuators by Haydon Kerk Motion Solution.

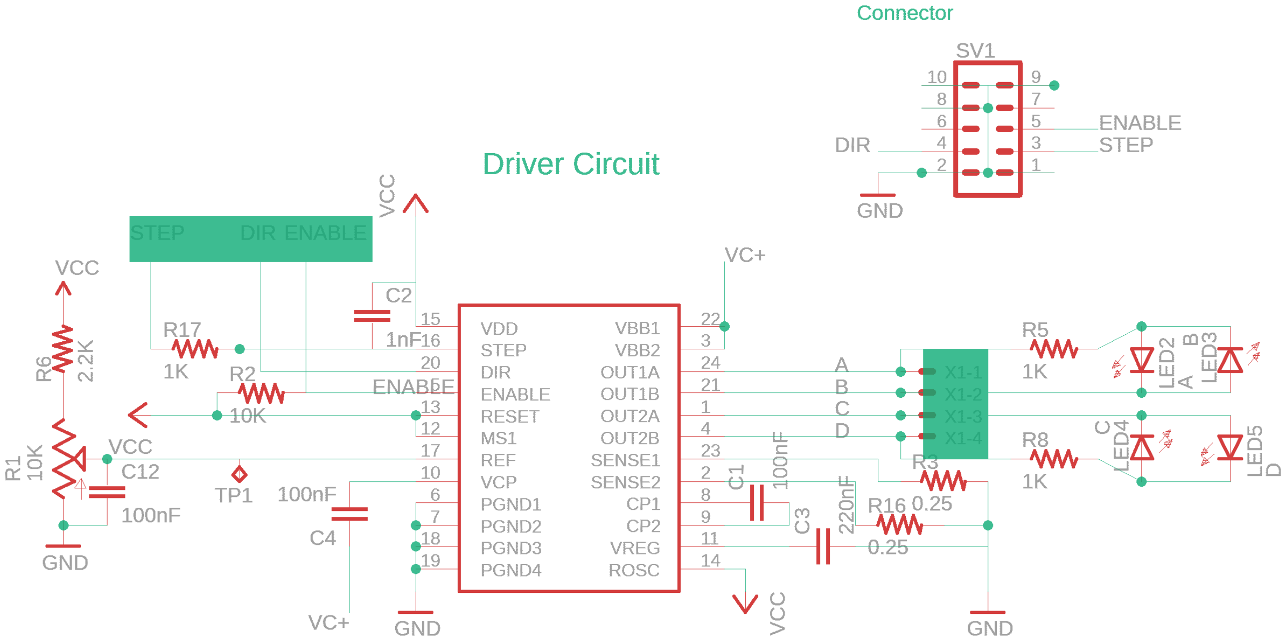
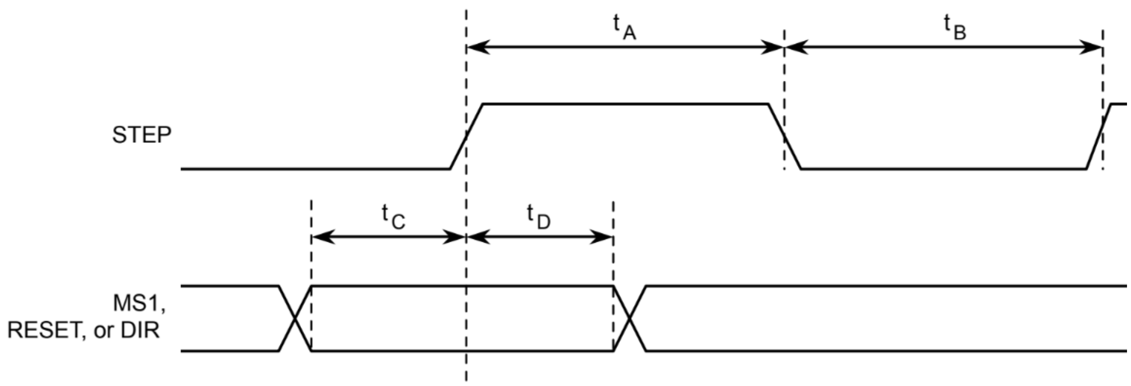


Figure 7: Motion Module Schematic



Time Duration	Symbol	Typ.	Unit
STEP minimum, HIGH pulse width	t_A	1	μs
STEP minimum, LOW pulse width	t_B	1	μs
Setup time, input change to STEP	t_C	200	ns
Hold time, input change to STEP	t_D	200	ns

Figure 8: Digital Input Scheme

Pin	Function	Connection	Data/Voltage
STEP	A low-to-high transition on the STEP input sequences the translator and advances the motor by one increment.	Connect from control module digital input to Pin 16 on the motor driver.	Logic high and low sequence. Communication Scheme Below.
ENABLE	This signal turns on or off all of the DMOS outputs(A,B,C,D). When set to a logic high, the outputs are disabled.	Connect from control module digital input to Pin 5 on the motor driver.	Logic high and low sequence.
DIR	This determines the direction of rotation of the motor. When low, the direction will be clock-wise and when high, counterclockwise. Changes to this input do not take effect until the next STEP rising edge.	Connect from control module digital input to Pin 5 on the motor driver.	Logic high and low sequence. Communication Scheme Below.
A,B,C,D	The DMOS outputs regulate the operating point of the motor.	Connect from the output pins on the driver to the bipolar motor connection.	Typically 0V or 5V output with max of 1.3 A current.
REF	The REF pin combined with the selection of RSx determines the maximum value of current limiting.	Connects the voltage dividing resistor circuit and Pin 17 on the driver.	Voltage between 0V to 5V

Table 5: Pin Layout Power Supply Module

1. The DOS outputs are able to drive motor at 5.0V±0.3V and supply upto 1.3A and does not fluctuate beyond this range.	Use clamp-on Amp Meter and Volt Meter to determine the operating point for the duration of 5 full forward movement and back.
2. The motor doesn't fluctuate for more than 5um.	1. Use Smartphone Microscope Camera to monitor if there is recoil. 2. Use Amp Meter and Oscilloscope to track if there is backward current.
3. Shut off load power supply in less than 1us to ensure precision	Use Amp Meter and Oscilloscope to track the shut off transition time.
4. The output has low ripple current, at lease 20dB smaller than DC current.	Use the Volt Meter and the Oscilloscope to monitor output voltage and its spectrum.

Table 6: R and V for Motion Module

2.2.3 Software Subsystem

We propose to build two applications - a high-level application that runs on a smartphone and a low-level application that runs on a microcontroller. The android application will take a series of photos of different regions of the blood sample, and stitch these photos together into a panorama and display the panorama on the phone. The microcontroller software will control the motor movement and communicate with the android application regarding when to move the motor and when the application should take a photo. The FSM and the flowchart are shown below.

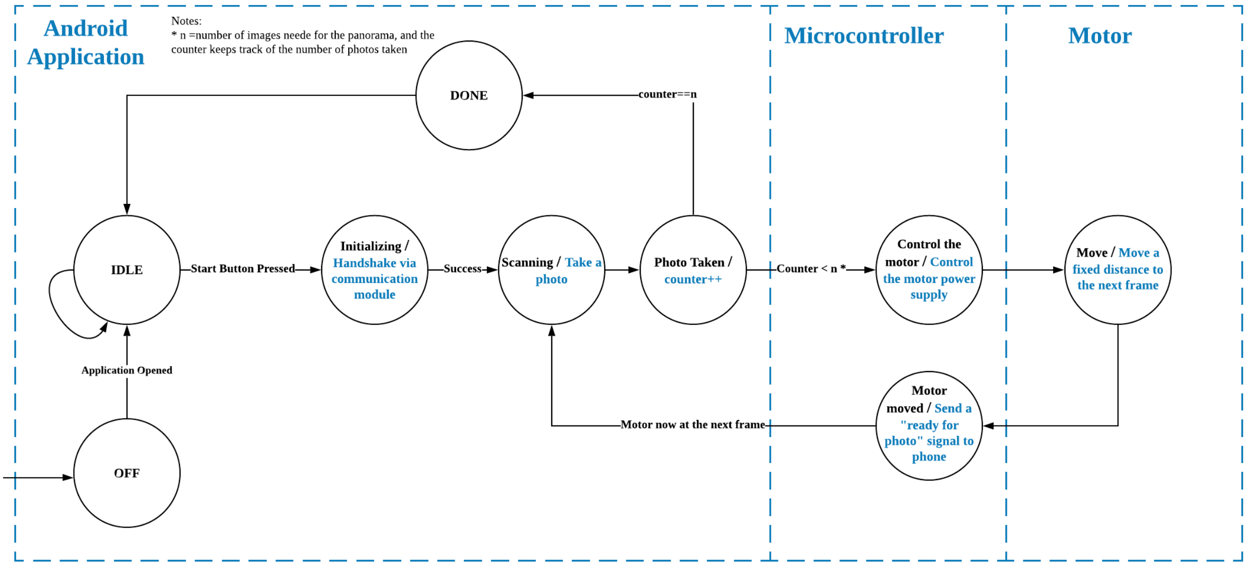


Figure 9: FSM of Software Operation

00	Android Start
01	Android Ready for photo
11	Error
02	Android Done
12	MCU Start
22	MCU Moving to next frame
32	MCU Done

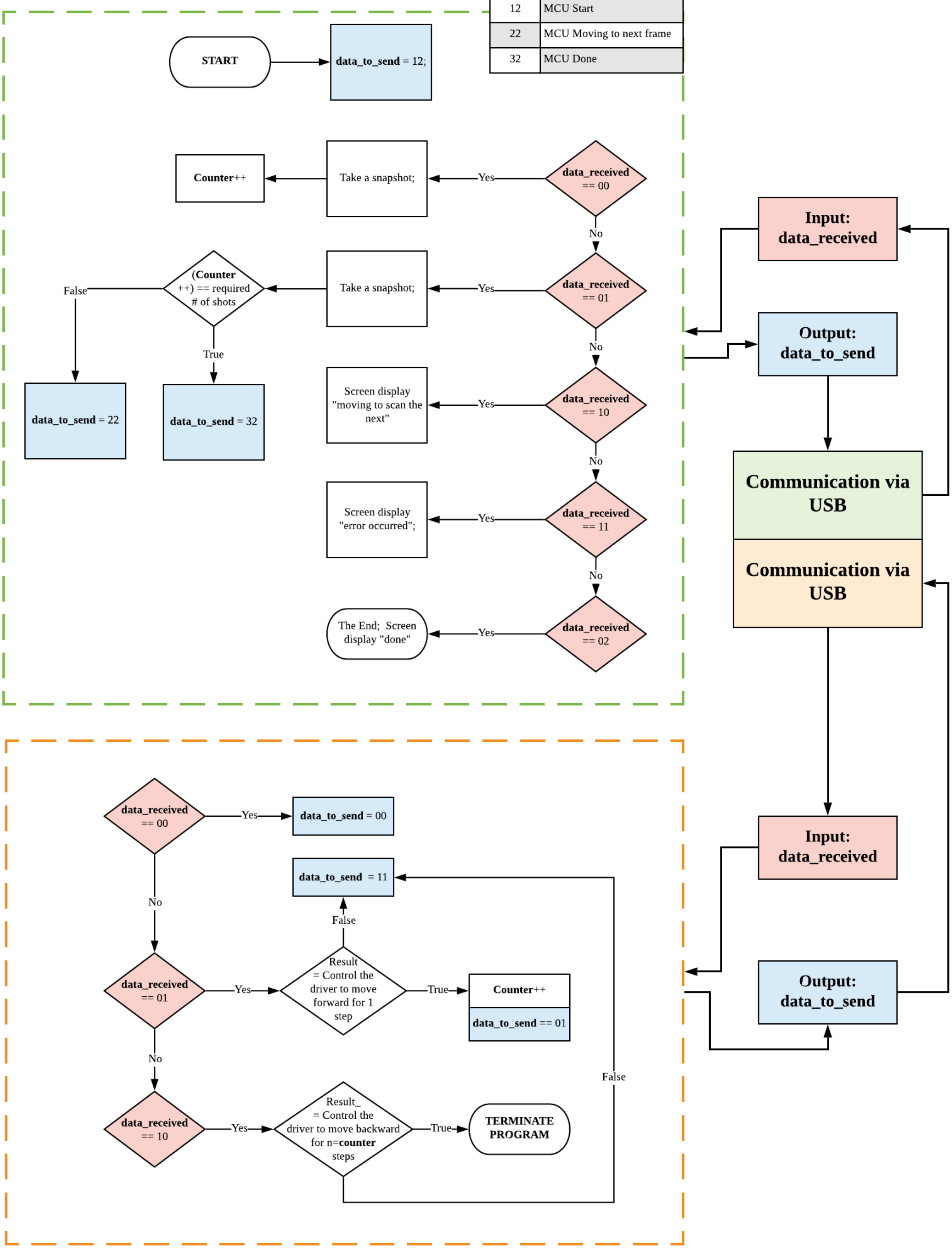


Figure 10: Software Flowchart

Android Application

Once the user opens the application, the FSM would go to the idle state and waits for the user to click "start". After the user clicks that “start” button on the screen, the mobile application will start checking its connection with the microcontroller by handshaking with the microcontroller via communication module. If success, the phone will take the first photo and send a “next frame” signal to the microcontroller. After the motor moves to the next frame, the microcontroller will send a “ready for photo” signal to the phone, and the phone will take another photo. This process repeats until a required number of photos are taken. The application will then stitch the images together into a panorama utilizing OpenCV4Android SDK package.

1. Communication Unit: In order to communicate with the microcontroller, we will use android.hardware.usb API, and the flowchart is shown below.

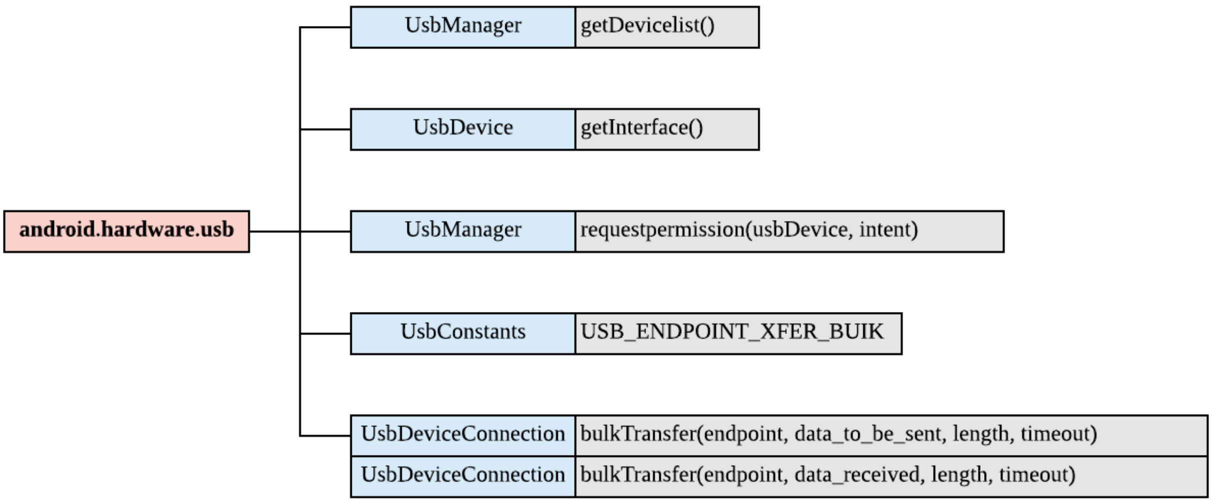


Figure 11: Communication Flowchart

2. Camera Unit: In this unit, the input comes from the communication unit. Once received the “ready for photo” signal, we would initialize the process of image capturing. We intend to use the android.hardware API, the continuous-auto-focus mode in the camera class allows us to only do auto-focus at the first time rather than auto-focus on every image. The Flash-Mode-On function allows us to keep the flashlight on when the

phone is taking snapshot, thus satisfying the lighting condition that's required for seeing white blood cells.

Microcontroller Software

The micro-controller software sends control signals to the motor driver to drive the linear stepper motor go a fixed distance after it receives the "next frame" signal from the android application. It will return the "ready for photo" signal after the motor completes its action.

Requirement	Verification
the smartphone camera should be able to take snapshot with flashlight on	we will write a function utilizing Flash-Mode-On function, and to test it on the white blood sample to see if the phone can always take snapshots with flashlight and to also check if the lighting condition is enough for manually counting the differentiated cells
the micro controller should be able to control the motor driver to make the motor move a fixed distance	we will write a function in the microcontroller that controls the motor driver to move the motor to a fixed distance.
Once the microcontroller is connected to the android phone, the android application should be able to find the microcontroller device, and to send and receive messages	we will write a function in the android application that identifies the device connected from the device list, requests permission from the user, and initializes the transfer channel. Then we will test the function by “buck-Transfer” x66 data segment to the microcontroller, and at the same time we'll write a function in the microcontroller to send x88 to the android application if it receives the x66 data from android.
the camera should be able to continuously take snapshots without having to auto-focus every time when it takes a photo	we will write a function that utilizes the continuous auto focus mode from the android.hardware API. Fixing the postion of the phone, we will slowly slide the sample while continuously taking photos to see if the camera only does auto-focus at the first time.
the application should be able to de-noise and enhance the blood cell image, as well as to stitch images that has no gap/some overlap between	we will find a blood cell image database online, and we will try out different de-noise filters and enhancement algorithms. We will measure our result by matching corresponding white blood cell features using machine learning model, and to find the most optimized filter and algorithm

Table 7: R and V for Software Subsystem

2.3 Supporting Material

2.4 Tolerance Analysis

Power Consumption
Idle mode:
0.75 uA - ATMega328p microcontroller [18]
70 uA - FT232R USB UART IC
7 mA - A3982 driver

Working Mode:
14 mA - ATMega328p microcontroller [18]
15 mA - FT232R USB UART IC
12 mA - A3982 Driver

Delay
Motor Driver Module: Delay from receiving:

$$D_{total} = D_{PWMGENERATOR} + D_{DMOS} = t_{crossoverdead} + t_{LowtoHigh} = 450ns + 200ns = 650ns. \quad (1)$$

Worst Case:

$$D_{total} = D_{PWMGENERATOR} + D_{DMOS} = t_{crossoverdead} + t_{LowtoHigh} = 800ns + 200ns = 1us. \quad (2)$$

3 Cost and Schedule

4 Ethics and Safety

The objective of our project is to simplify the process of blood cell differentiated counting such that doctors could diagnose within a shorter period of time, and this aligns with the IEEE Code of Ethics, #5 “to improve the understanding of technology, its appropriate application. . . ”[IEE].

There are several concerns regarding the safety and ethics of our project. The main safety hazard comes from the possibility of virus spreading via human blood sample. To mitigate the hazard, we will use horse blood as a sample of our senior design project to avoid the danger brought by human blood. The horse blood is extracted in a legal way by a veterinarian, and using horse blood wouldn't form any threat to one's health. After our senior design, the project might be dealing with human blood. Doing experiments with human blood involves dealing with biology safety level 2 hazard, and every individual involved in the project will be required to complete a safety level 2 training and to pass the safety quiz. In such way, people will be taught to appropriately dealing with blood sample, significantly reducing the possibility of getting virus, following the IEEE Code of Ethics, #9 "to avoid injuring others, their property. . . "[IEE]. Another safety concern comes from the power supply, and we will be responsible for ensuring the voltage and current fed into different electronic modules won't exceed their standard thresholds.

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