Portable Fluorescence Sensor for Lyme Disease Antibody Detection

Sponsor:

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Motivation

- Create Demonstration of Everix thin filter technology
- Create a portable fluorescence sensing device that can be used outside the laboratory environment
- Problems with traditional fluorescent sensing devices
  - Low Portability
  - High Cost

Goals & Objectives

- Create a device which accurately and precisely excites fluorophores of particular interest that are attached Lyme disease antibodies
  - Select an ailment and Fluorescent marker to detect
  - Determine the excitation and emission wavelength of the fluorescent marker
  - Find an illumination source with a peak emission wavelength equal to the excitation wavelength of the chosen fluorophore
- Create a device which accurately and precisely measures the concentration of fluorescent emission from the fluorophores
  - Photodetector chosen with a high sensitivity within the fluorescence emission wavelength range
  - Optical filter chosen to isolate the fluorescent emission wavelength, cutting off the excitation light
- Compact design with reduced weight and bulk compared to other fluorescent sensing devices
  - Compact optical design through use of angled illumination reflection and detection
  - Compact circuit design
- Portable design for use in the field outside of the lab
  - Portable power supply/long battery life
  - Re-chargeable battery
- Visual display of sample concentration of fluorophores representing a particular ailment detected through florescence
  - Display with a high enough pixel count to display the decimal quantities with units of molar concentration

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Specifications & Requirements

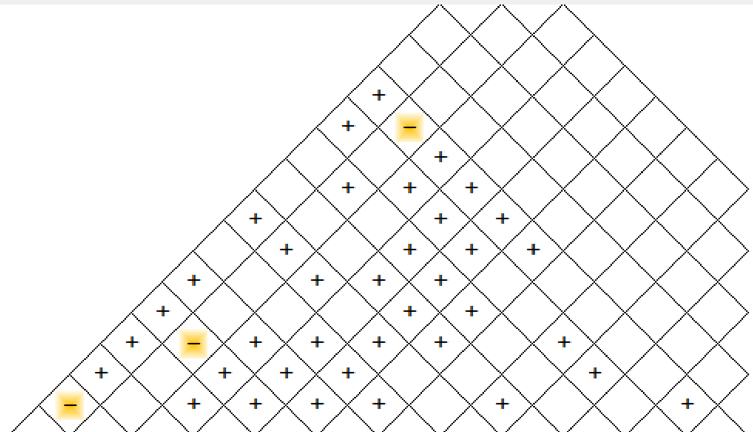
- The long pass optical filter shall be curved such that fluorescent light emission and light excitation light reflected off the sample will enter the filter at a normal angle of incidence
- The LED shall emit a spectrum of light with the highest intensity peak centered within the desired excitation wavelength range (~460nm for fluorescein sodium salt)
- The long pass optical filter shall only allow light of the fluorescent emission wavelength of around 515nm to pass through and cut off wavelengths of light emitted from the illumination source of around 465 nm
- The optical system will have a spacing between optics that allows for a compact design of the optical system
- The optical system shall have a limit of detection (LOD) <5 picomolar (pM) for fluorescence emission detection

House of Quality

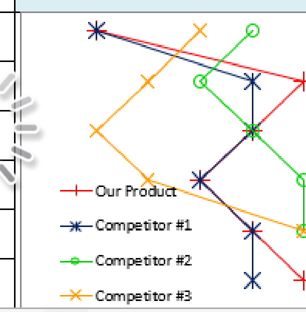
Correlations	
Positive	+
Negative	–
No Correlation	

Relationships	
Strong	●
Moderate	○
Weak	▽

Direction of Improvement	
Maximize	▲
Target	◇
Minimize	▼

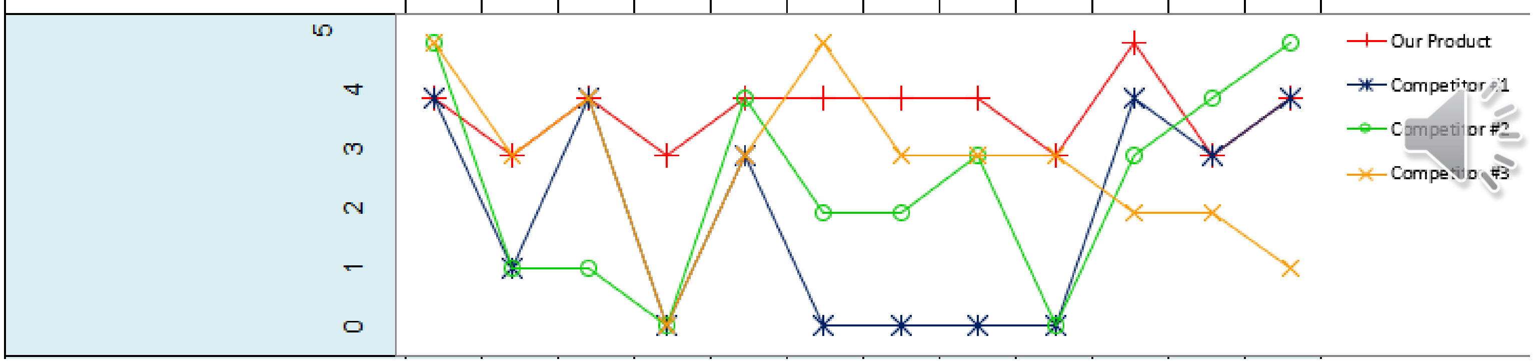


		Column #	1	2	3	4	5	6	7	8	9	10	11	12	Customer Competitive Assesment					
		Direction of Improvement	▲	▼	▲	◇	▲	◇	◇	▲	▲	▼	▼	▼						
Category	Weight	Customer Requirements (Explicit and Implicit) / Engineering Requirements	Florescence detection accuracy	Optical System focal length	Fluorophore Excitation accuracy	Effective Optical Filter Switching	Accurate Display of Fluorescence Data	Effective user input to parameter adjustment	Simple GUI Operation	Accurate fluorescence calculation software	Accurate Fluorescence calibration software	Dimensions	Power use	Cost	Our Product	Portable Fluorescent Sensor for on-site detection of microalgae	Low-Cost, Portable Smart Instrument for Detecting Colorectal	Fluobeam	0 1 2 3 4 5	Row #
Safety	2	Blue LED light emission within safe intensity	○	▽	●		▽	○			▽		●	○	1	1	4	3		1
Everix	8	Compact size	○	▽	▽	●	▽	▽			▽	●	●	●	5	4	3	2		2
	4	Portability	▽	▽	▽	○	▽	○	○	▽	▽	●	●	○	4	4	4	1		3
	4	Low cost		○	○	●	○	○		▽		●	●	●	3	3	5	2		4
	8	Accurate fluorescence detection	●	▽	●	○	▽	▽				○	▽	▽	4	4	5	5		5
	4	Battery Life				●							●	●	5	4				6

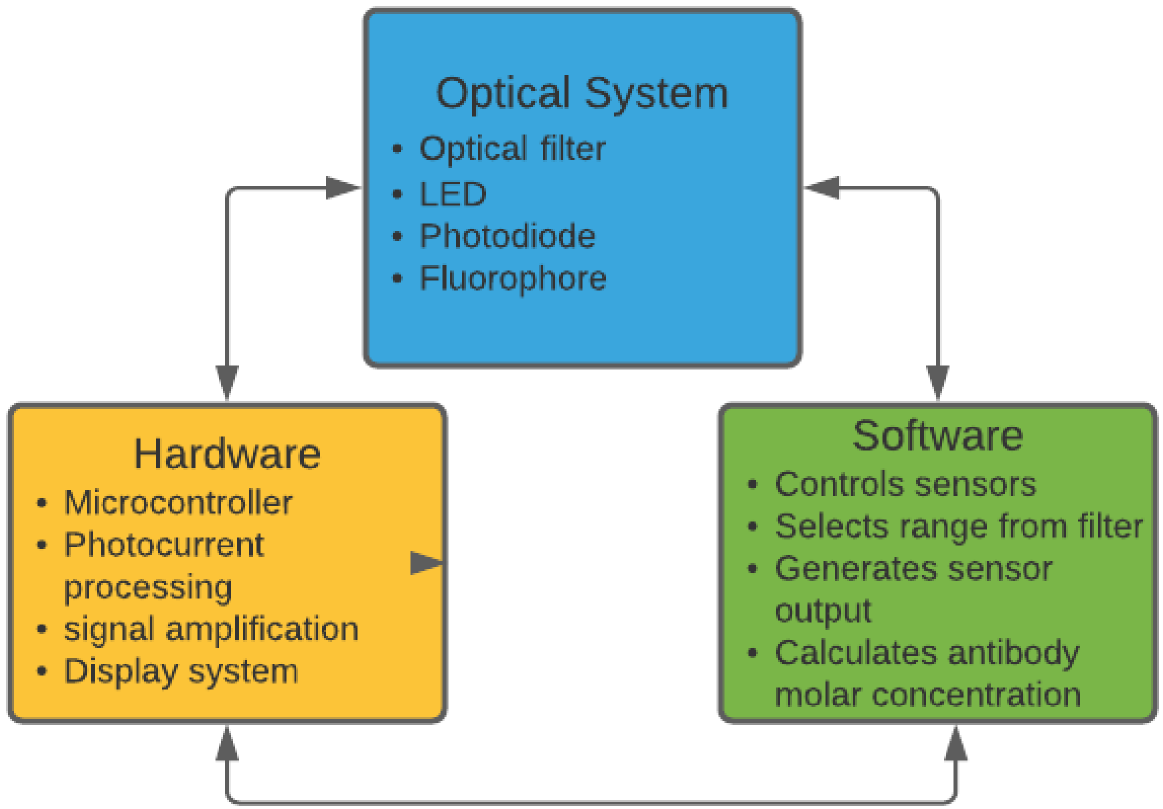


House of Quality

Target	Florescence detection accuracy	Optical System focal length	Fluorophore Excitation accuracy	Effective Optical Filter Switching	Accurate Display of Fluorescence Data	Effective user input to parameter adjustment	Simple GUI Operation	Accurate fluorescence calculation software	Accurate Fluorescence calibration software	Dimensions	Power use	Cost
Max Relationship	9	3	9	9	3	3	3	1	1	9	9	9
Technical Importance Rating	10600	3400	11400	18000	3400	4600	1200	800	1400	16800	20600	17000
Relative Weight	10%	3%	10%	16%	3%	4%	1%	1%	1%	15%	19%	16%
Weight Chart												
Our Product	4	3	4	3	4	4	4	4	3	5	3	4
Portable Fluorescent Sensor for on-site detection of microalgae	4	1	4	0	3	0	0	0	0	4	3	4
Low-Cost, Portable Smart Instrument for Detecting Colorectal Cancer Cells	5	1	1	0	4	2	2	3	0	3	4	5
Fluobeam	5	3	4	0	3	5	3	3	3	2	2	1



Overall Block Diagram



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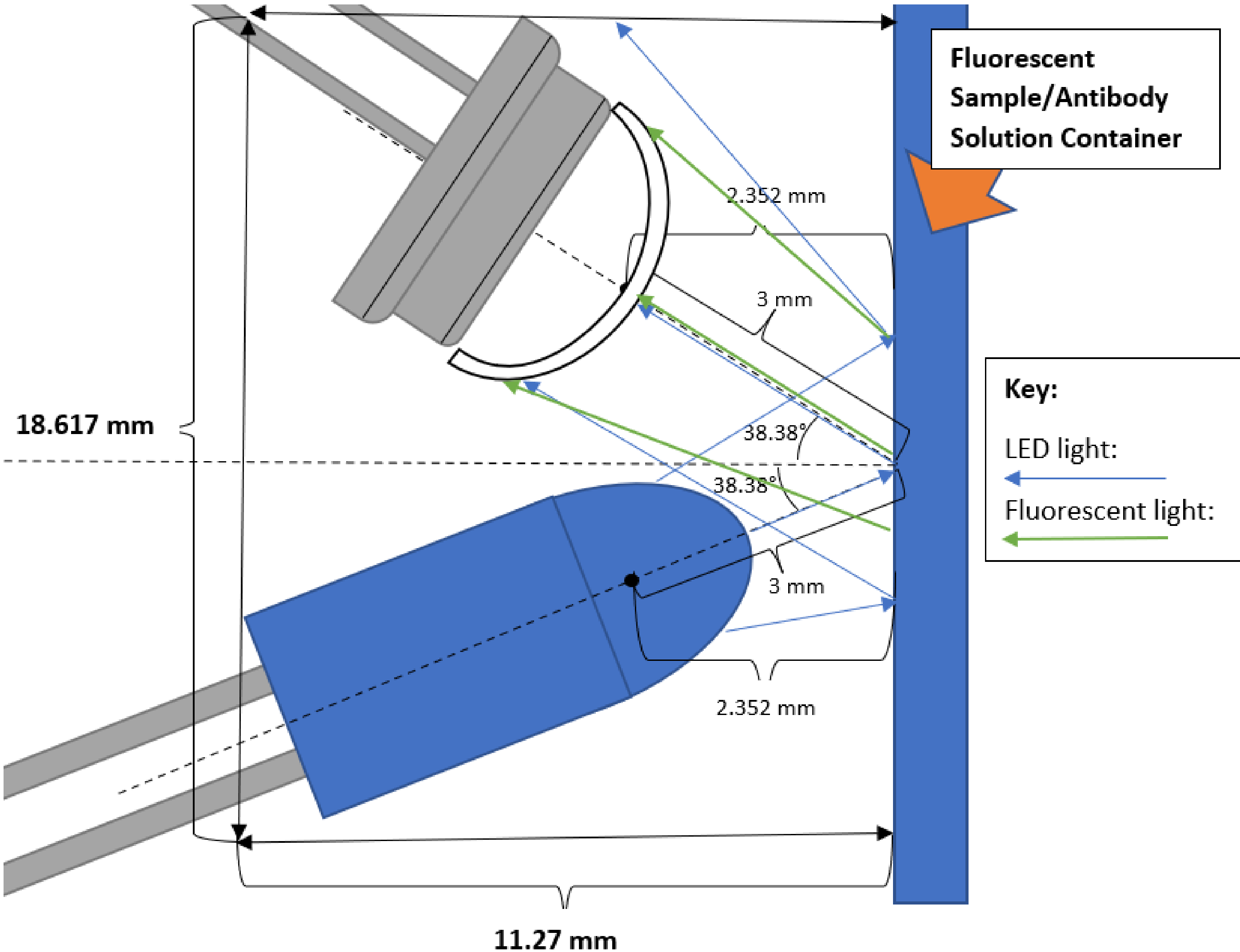
Optical System Design Diagram



Explain Design Approach

- Reflection based fluorescence detection
  - Higher signal detection from fluorescent light emission
  - Compact optical system due to angled illumination
  - Flexibility in size/shape of the sample container compared to transmission design

Optical Lens Considerations

- Edmund Optics' Uncoated, Plano-Convex Lens 6.0mm Dia. x 6.0mm FL
  - Plano-convex lens type for effective collimation
  - Lens diameter equal to 6 mm diameter photodiode window
  - Focal length of 6 mm for a compact optical system size
  - Small F/# = 1 to collect as much light as possible
  - Everix curved optical filter
    - No need for the light to be collimated
    - Condensed optical system without lens

Optical Filter considerations

- Optical Filter
  - Longpass filter
  - Cut-off light wavelengths below 500 nm
  - Unwanted LED illumination leakage at non-normal angles
  - Everix Thin filter decreasing size of optic system
  - Filter curved for normal incidence at all angles of incident light
    - Concave
    - Convex

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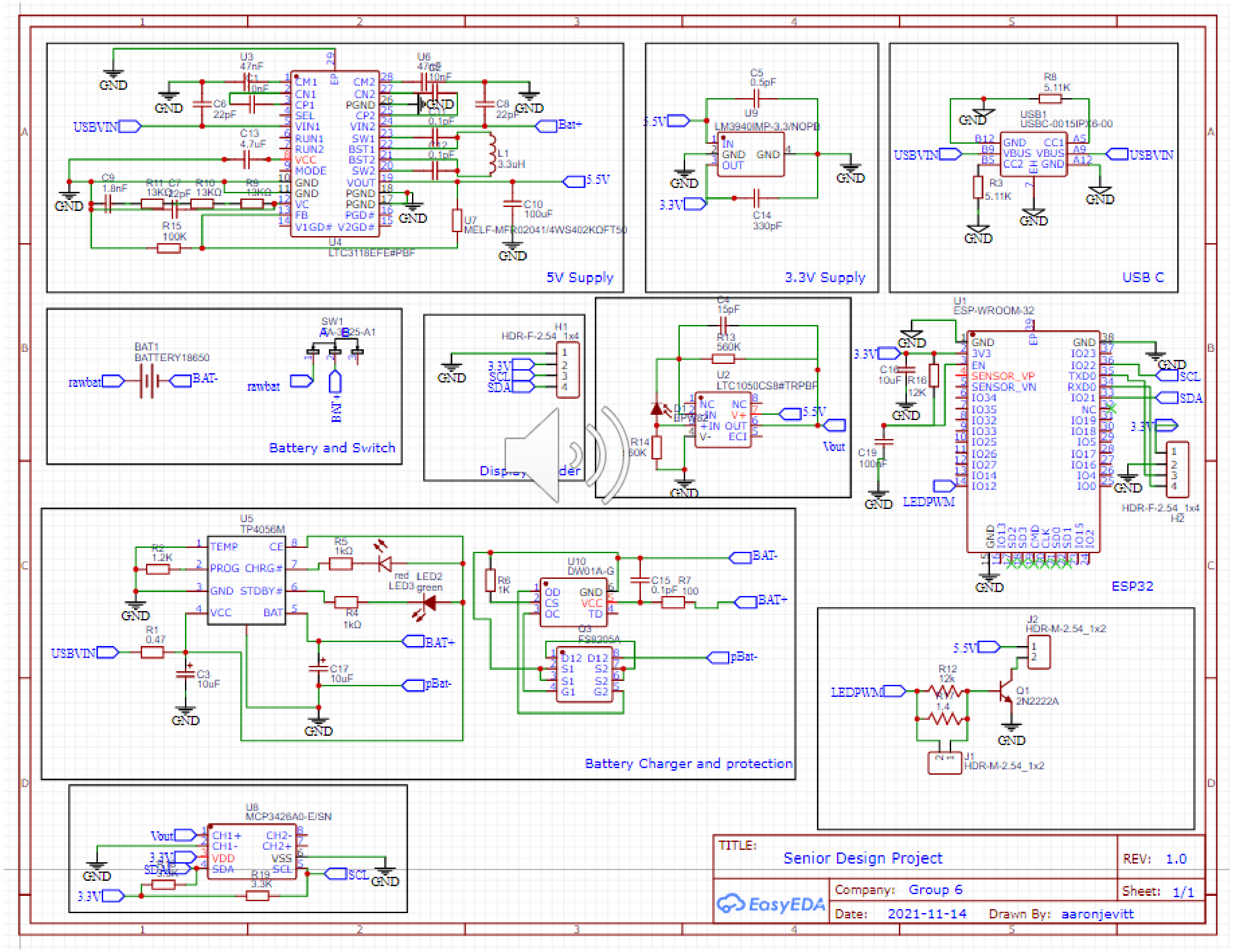
Overall Optical Component Decision

- LED excitation source
  - Peak LED emission wavelength 474nm near fluorophore excitation
- Photodiode detector
  - Linear signal response
  - High responsivity
  - Large active area
- Lack of lens
  - Decrease optical system size
  - Collimation of reflected and fluorescent light not needed due to use of a curved optical filter
- Optical filter
  - Curved to decrease reflected LED light leakage
  - 500nm wavelength cut off chosen to isolate fluorescent light signal

Electrical Design Overview

- The electrical design will be composed of a 3.3V and 5V Regulators, ESP32, Analog to Digital converter, Op-Amp for photodiode, rechargeable battery, battery charge charger IC and battery protection IC
- Powered by 5V USB Type C connector
- Considerations:
  - Size
  - Battery Life
  - Photodiode Measurement Precision

Electrical Design Schematic



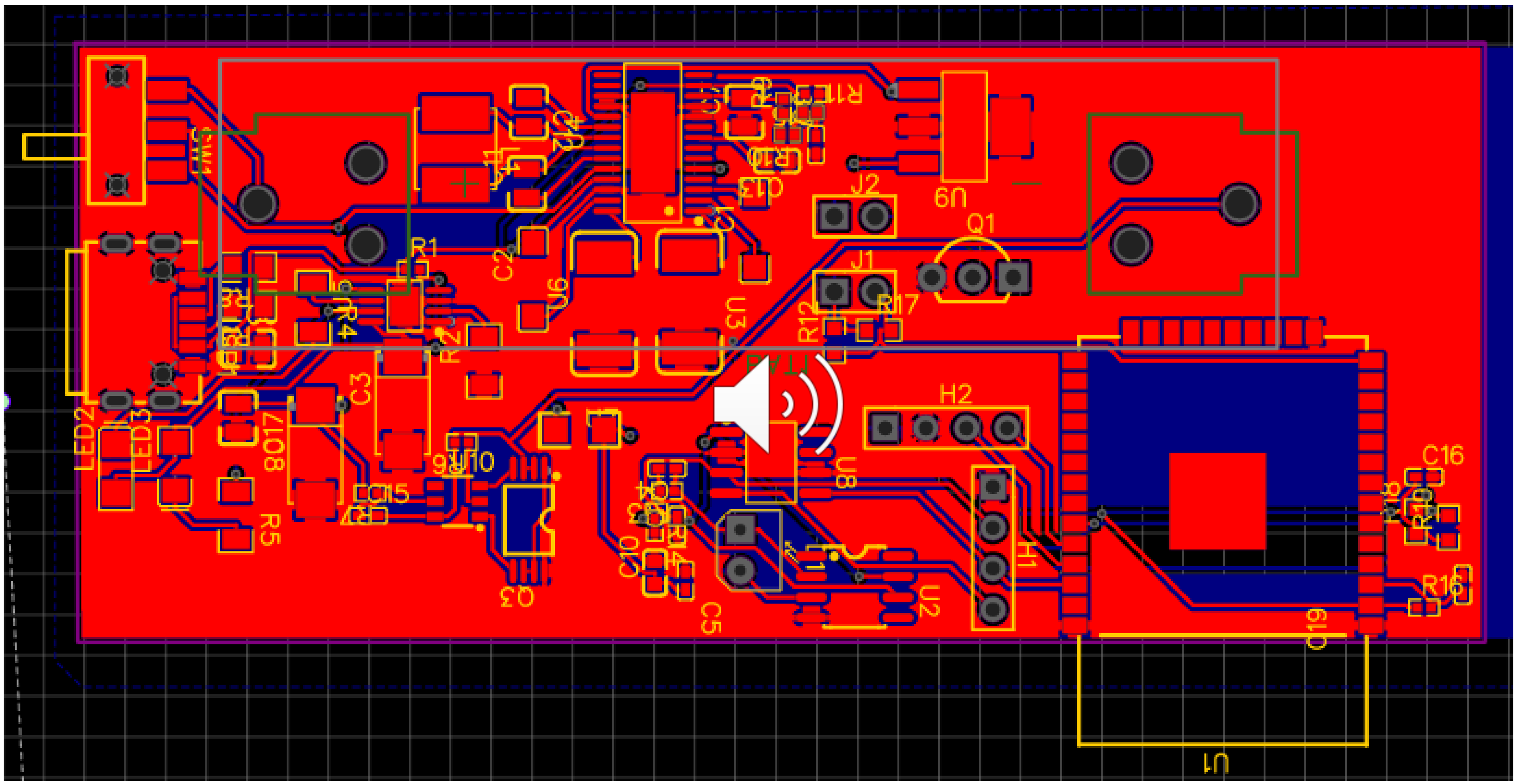
Microcontroller Considerations

- Considerations
  - WiFi Communication
  - I2C Bus
  - SPI Bus
  - Non-Volatile Flash Memory
- We chose the ESP32 because it has a Wifi Transceiver, the required communication buses, and a built in Flash Memory Chip.

Photodiode Amplifier Considerations

- Considerations
  - Small photocurrent
  - Minimize dark current to reduce error
  - Create voltage signal that can be fed to analog to digital converter
- We chose the LTC1050 op-amp configured as a transimpedance amplifier because of its low drift, offset voltage of <5 µV, and availability.

Electrical System PCB Layout



Dimensions: 37mm x 88 mm

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Electrical Design Standards

- This project uses the following standards:
  - WiFi
  - SPI
  - I2C

Device Software Design Overview

- Necessary Functionality
  - WiFi client and access point
  - Web Server
  - ADC interfacing via I2C
  - Display interfacing via SPI
- The ESP32 will be programmed using Arduino IDE due to library availability for major components like WiFi, Display, I2C, and SPI

Web GUI Design Overview

- Necessary Functionality
  - Menu for configuring device
  - Home Page for initiating sampling, and viewing most recent sample
  - Viewer for sample database
- The Web GUI will be programmed in HTML using Javascript to facilitate the transfer of data to and from the device

Outstanding Software Design

- Sampling routine is still yet to be determined
  - Through testing, the team will determine the number of measurements that will be required to average and ensure a consistent reading.
- Web Client Interface
  - Data protocol still yet to be decided
    - Web Socket, simple http requests

Design Constraints

- Budget
- Safety
- Fluorophore (fluorescent marker)
- Size
- Battery-Powered

Standards related to our project

- Safety
  - Low blue light radiance value
  - Safe battery storage
- Fluorescence spectroscopy standards
  - Qualitative measurement
  - Quantitative measurement
- Electrical housing standards
  - Protect electrical hardware from outer environment

Successes & Difficulties

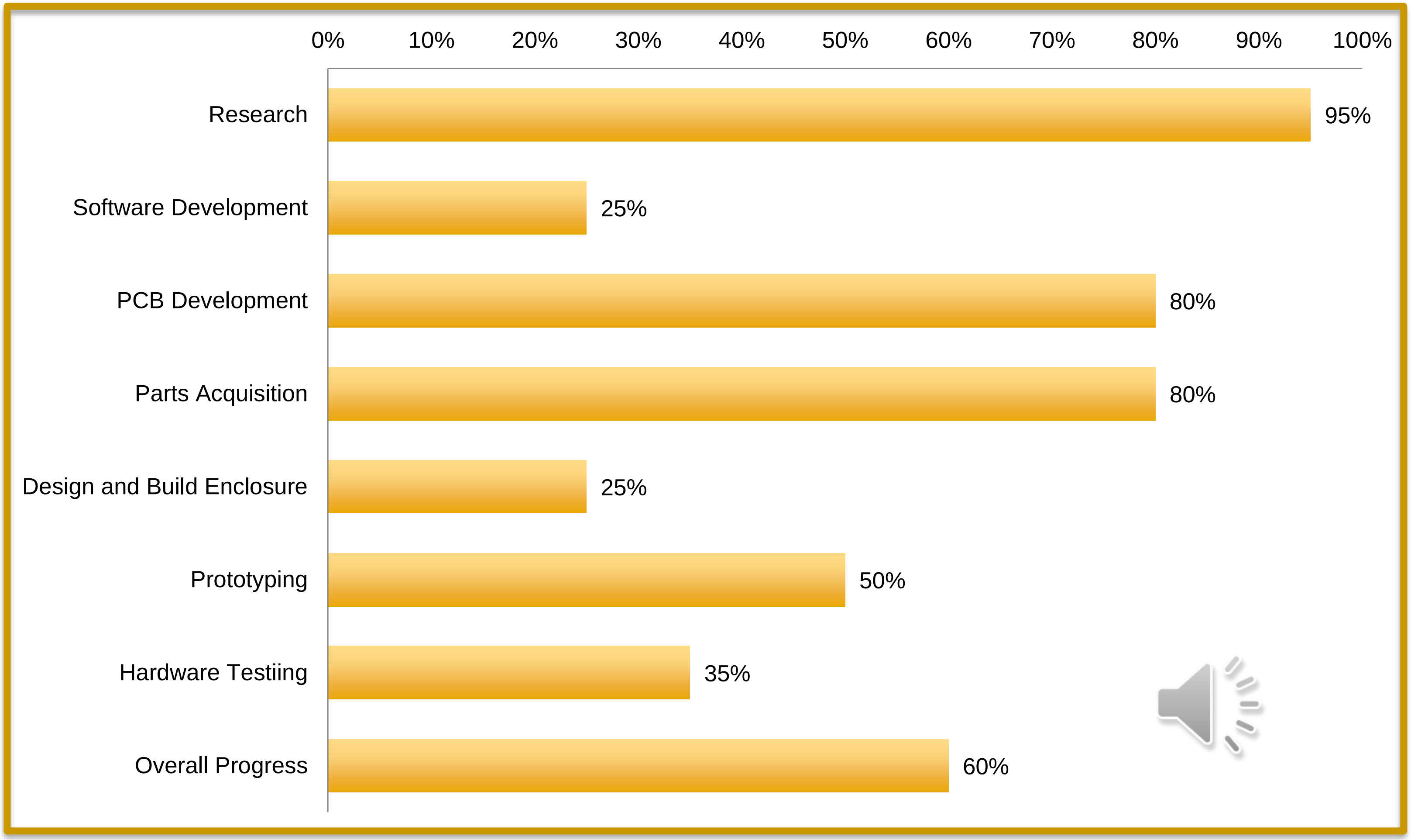
- Difficulties
  - Complex ray trace due to angled detection & illumination system
  - Difficulty ordering photodiode
  - Complex design of curved optical filter
- Success
  - Senior design 1 optical demo

Budget & Financing – Deliverable 2

	Costs for Fluorescence Sensor				
	Item	**Quantity**	**Price/Unit**	**Projected Cost**	**Actual Cost**
1	LED	1	$4.992	$4.992	$4.992
2	**Photodiode**	**1**	**$48.71**	**$48.71**	**$48.71**
3	Optical Filter	1	$400.00	$400	FREE
4	Fluorescein	100g	$30.5	$30.5	$30.5
5	Microcontroller	1	$ 4	$ 4	FREE
6	PCB	1	$2.04	$10.02	$10.02
7	**Display**	**1**	**$3.00**	**$3.00**	**$3.00**
8	**Custom Enclosure**	**1**	**$20.00**	**$20.00**	**$20.00**
9	Circuit Components	1	$ 75	$ 75	
				$596.222	**$196.222**

Team Budget	$200.00
Sponsorship	$1,000.00

Current Progress



Final Steps

	11-Feb	18-Feb	25-Feb	4-Mar	11-Mar	18-Mar	25-Mar	1-Apr	8-Apr	15-Apr	22-Apr
PCB Testing	■										
Optical System Testing	■	■	■								
Develop Software	■	■	■								
Assemble/Test PCBs		■									
Order Revised PCB			■								
Fabricate Enclosure				■							
Integrated Testing/ Calibration				■	■						
Final Assembly						■					
Final Device Testing							■				

References

- https://www.everixopticalfilters.com/

Questions about our project?