

A Compact Field-portable Computational Multispectral Microscope using Integrated Raspberry Pi

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Abstract

In this paper, we present a compact field-portable computational multispectral microscopy system powered by an integrated Raspberry Pi computer. The optical components of the low-cost, compact microscopy system consists of a cemented achromatic doublet lens placed on top of a Raspberry Pi camera module, used to capture magnified broadband microscopy imaging acquisitions at three broadband channels in the visible spectrum based on the Bayer pattern color filter array. The captured broadband microscopy acquisitions are then processed by a numerical spectral demultiplexer, which demultiplexes the magnified broadband acquisitions to generate a set of narrowband microscopy images. A Helianthus stem was imaged using the proposed computational multispectral microscopy system, and demonstrated to be able to produce narrowband microscopy images at 16 different wavelengths with a single acquisition without the need for wavelength scanning, which makes it well-suited for fast microscopy imaging applications such as the study of transient phenomena.

1 Introduction

Multispectral microscopy systems enable specimens on the micrometer and nanometer scale to be observed at different wavelengths of light. A major advantage of multispectral microscopy systems is that they can provide additional information regarding the imaged specimen, since light-matter interaction can be unique for a given specimen across multiple wavelengths. However, most multispectral microscopy systems capture imaging data at different wavelengths in a sequential manner, those limiting their efficacy for capturing transient phenomena with live specimens *in-vivo*. For example, Brydegaard *et al.* utilized sequential LEDs [1] to illuminate a sample to achieve multispectral microscopy imaging. In addition, such multispectral microscopy systems often comprise a large number of optical elements, thus increasing the cost and complexity of the system [2].

Motivated to tackle the challenges associated with multispectral microscopy, we present a fully-integrated, compact, field-portable computational multispectral microscopy system powered by an integrated Raspberry Pi computer. Extending significantly beyond our preliminary findings in computational microscopy [3], the presented multispectral microscopy system is capable of producing a set of narrowband microscopy images at 16 different wavelengths with a single acquisition using a Raspberry Pi camera module without the need for wavelength scanning, which makes it well-suited for fast microscopy imaging applications such as the study of live specimens.

In Section 2 of this paper will present the hardware and software components of the presented computational multispectral microscopy system. Next, in Section 3 we will present the experimental setup and discuss the results. Finally, in Section 4 we will discuss our conclusions and future work.

2 Computational Multispectral Microscopy System

In Section 2.1, we will discuss the design of the microscope chassis, the integrated Raspberry Pi computer, as well as the Raspberry Pi camera module that constitutes the hardware subsystem. In Section 2.2, we will discuss the software subsystem for producing narrowband microscopy images at different wavelengths.

2.1 Hardware Subsystem

In order to design the computational multispectral microscopy system as a compact, low-cost, field-portable device, the hardware subsystem is broken into five different components, each serving a specific purpose, as seen in Fig. 1.

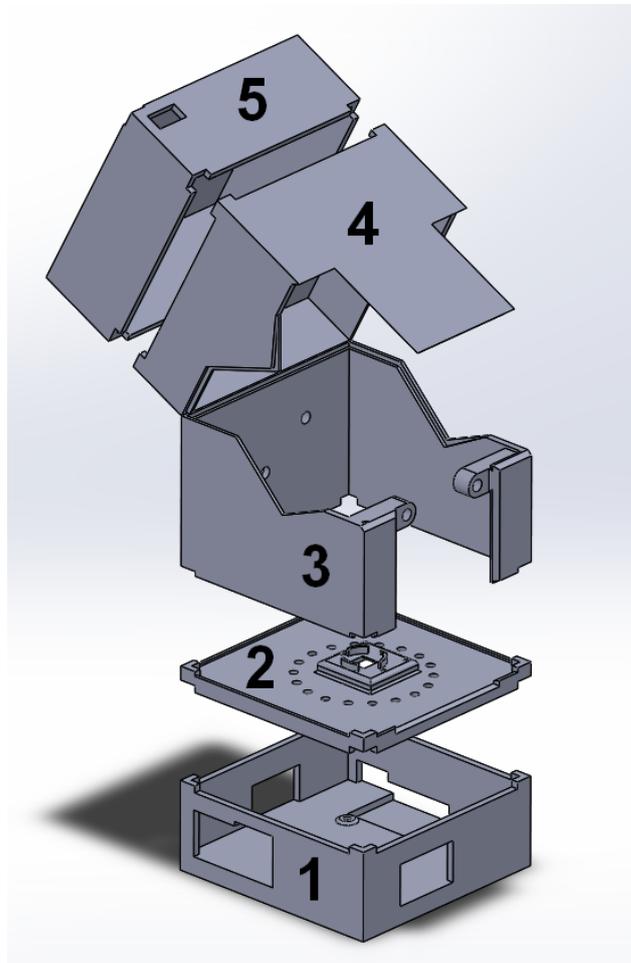


Fig. 1: A 3D CAD view of the computational multispectral microscopy system with five components. The Raspberry Pi computer is housed in component 1, which is the base of the microscope chassis while the camera module and lens is housed in component 2. Component 3 is used to move the microscope sample vertically to ensure the specimen being imaged is in focus. Finally, component 4 is used to block out ambient light if needed, and component 5 can be used to house additional electronics.

The first component houses the Raspberry Pi computer and is the base of the microscopy system. The Raspberry Pi camera module is then mounted in the second component of the system. In addition, an achromatic doublet lens with a focal length of 10 mm and a diameter of 8 mm [4] is placed on top of the Raspberry Pi camera module in the second component. This lens will provide the magnification required for microscopy applications. The third component of the chassis controls the vertical displacement of the slide above the lens which allows for fine-tuned focusing of a specimen. The fourth component is a cover to block any ambient light from entering the system or any stray light within the system and can also house the LED light sources. Finally, the fifth component can house any additional electronics which are needed to control the LED in the fourth component.

2.2 Software Subsystem

Given the magnified broadband microscopy imaging acquisitions at three broadband channels in the visible spectrum captured by the Bayer pattern color filter array in the Raspberry Pi camera module, the software subsystem then processes the acquisitions to produce a set of narrowband microscopy images at a number of specific

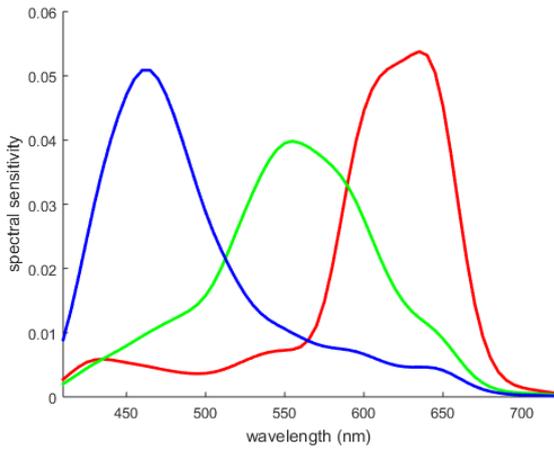


Fig. 2: The spectral sensitivity of the Raspberry Pi camera module was measured using a monochromator. This data was used to train a spectral demultiplexer which can take the broadband acquisitions and demultiplex them into narrowband images.

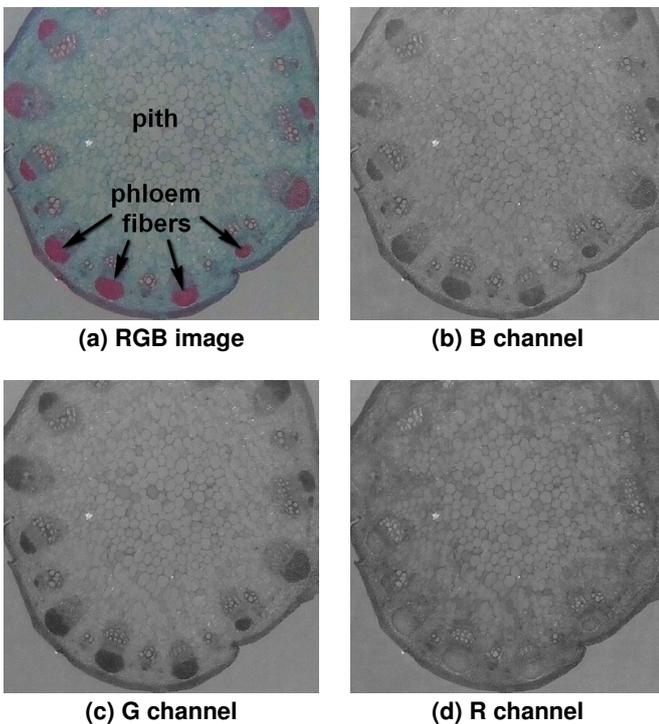


Fig. 3: (a) The RGB image captured by the Raspberry Pi highlighting the phloem fibers and the pith; (b-d) Broadband acquisitions (R,G,B).

wavelengths. This is accomplished with a numerical spectral demultiplexer [5], calibrated based on the spectral sensitivity of the Raspberry Pi, as seen in Fig. 2. The calibrated numerical spectral demultiplexer used in this microscopy system is based on a random forest model that was trained with 10,000 training pairs produced using a calibrated forward model. The output of the computational multispectral microscopy system is a set of 16 narrowband microscopy images from 410 nm - 710 nm with 20 nm spacing.

3 Results & Discussion

To demonstrate the efficacy of the presented computational multispectral microscopy system, a sample of the cross-section of a Helianthus (Sunflower) stem was imaged using the microscopy system, as seen in Fig. 3(a). The three broadband acquisitions are shown in Fig. 3(b)-(d), while the sixteen narrowband microscopy images produced by the computational multispectral microscopy system are shown in Fig. 4(a)-(p).

A number of observations can be made from the multispectral microscopy imagery produced by the computational multispectral

microscopy system. The first observation is that the phloem fibers (see arrows) on the perimeter of the Helianthus stem have a higher relative response in 410 nm, 450 nm, 650 nm narrowband images, and have a lower relative response in the 490 nm, 530 nm, and 570 nm images, which matches the expected spectral composition of pink hue of the fibers. On the other hand, the strong contrast found in the narrowband images is not present in the red and blue broadband acquisitions, due to the fact that these acquisitions integrate over a wide spectrum range.

The second observation is that the pith (center area) of the Helianthus stem has a lower relative response in the 410 nm and 560 nm images, and has a higher relative response in the 490 nm, 530 nm, and 570 nm narrowband images. This also matches the expected spectral distribution of the green hue of the pith. Both of these observations illustrate the efficacy of the computational multispectral microscopy system and its ability to obtain multispectral microscopy imagery of a specimen from a single acquisition. Furthermore, this illustrates that the use of multispectral microscopy imagery can provide additional information and insights about the imaged specimen, and provide better delineation between different parts of a specimen.

4 Conclusion

In this paper, a compact field-portable computational multispectral microscopy system powered by an integrated Raspberry Pi was demonstrated. The efficacy of the proposed low-cost, compact microscopy system was demonstrated by imaging a Helianthus stem sample, producing narrowband microscopy images at 16 different wavelengths with a single acquisition without the need for wavelength scanning. Future work involves the integration of more powerful microscope objectives to boost the resolution capabilities of the system for applications where higher spatial resolution may be required.

Acknowledgments

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References

- [1] Brydegaard, M., Guan, Z., and Svanberg, S., "Broad-band multispectral microscope for imaging transmission spectroscopy employing an array of light-emitting diodes," *American Journal of Physics* **77**(2), 104–110 (2009).
- [2] Hiraoka, Y., Shimi, T., and Haraguchi, T., "Multispectral imaging fluorescence microscopy for living cells.," *Cell structure and function* **27**(5), 367–374 (2002).
- [3] Deglint, J., Kazemzadeh, F., Wong, A., and Clausi, D., "Numerical spectral demultiplexing microscopy of measurements from an anatomical specimen," *Vision Letters* **1**(1) (2015).
- [4] Deglint, J., Kazemzadeh, F., Shafiee, M. J., Li, E., Khodadad, I., Saini, S. S., Wong, A., and Clausi, D. A., "Virtual spectral multiplexing for applications in in-situ imaging microscopy of transient phenomena," in [*SPIE Optics and Photonics*], (August 2015).
- [5] Deglint, J., Kazemzadeh, F., Cho, D., Clausi, D. A., and Wong, A., "Numerical demultiplexing of color image sensor measurements via non-linear random forest modeling," *Scientific Reports* **6**(28665) (2015).

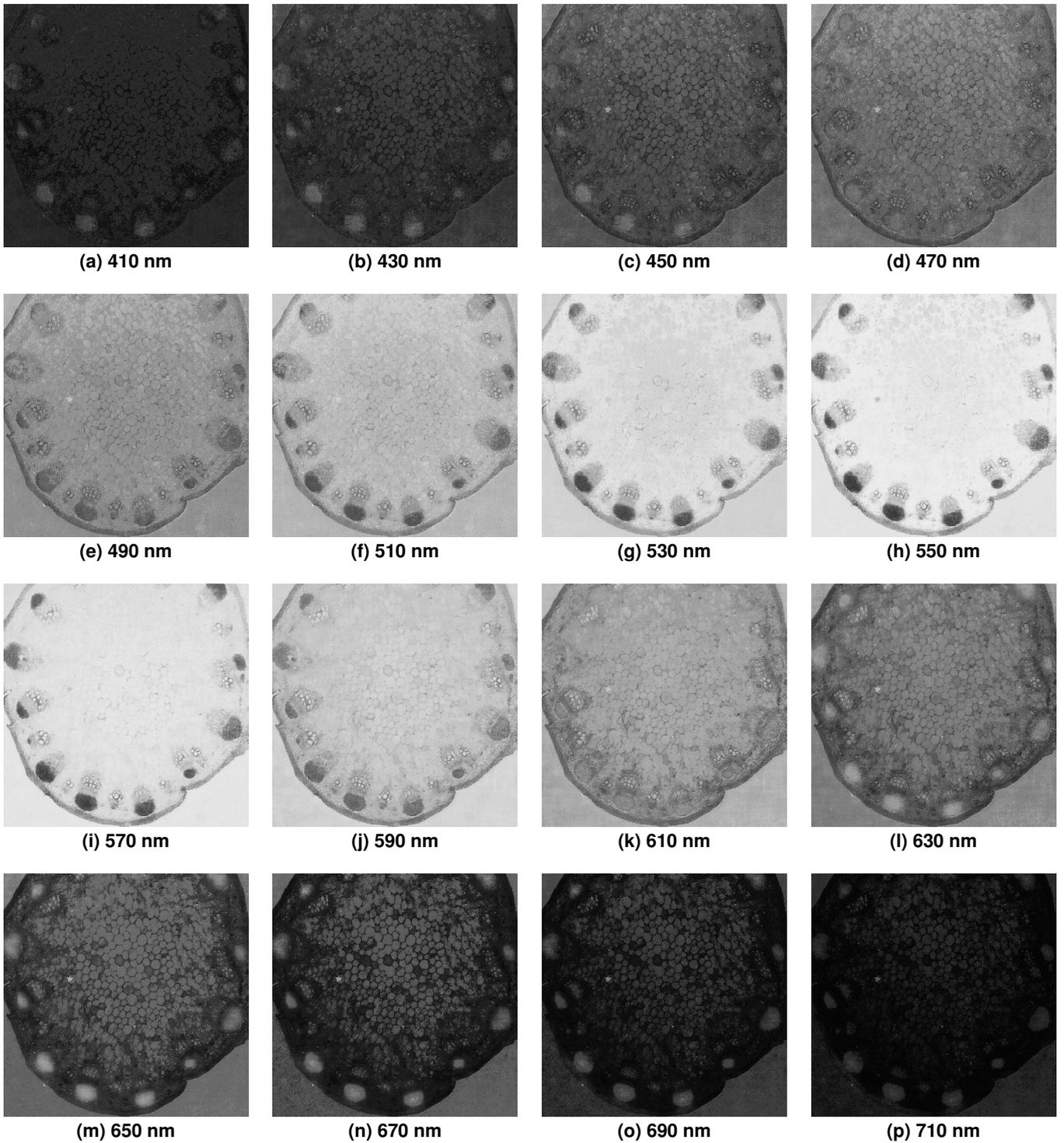


Fig. 4: (a-p) Narrowband images produced by the computational multispectral microscopy system at the sixteen wavelengths for a cross-sectional image of a *Helianthus* stem. It can be observed that the use of multispectral microscopy imagery can provide better delineation between different parts of a specimen compared to the broadband RGB images.