

# Multispectral imaging: Development and applications for snapshot MSI instrumentation

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**Abstract** While there are a wide variety of commercially available MSI imaging techniques and instruments, the community has long desired a simultaneous multichannel imaging sensor. We will describe the design and utility of exactly such a system as there has recently been a snapshot multispectral imaging system released to the market. Here, we will focus on the development path from standard research-grade spectrometer to filter wheel-based multispectral imager leading to the simultaneous multichannel “snapshot” MSI camera. Representative data from each modality will be presented to illustrate the progression of instrumentation. For the purposes of this paper, two similar looking samples, one biological in nature and one synthetic in nature will provide suitable objects for illustration.

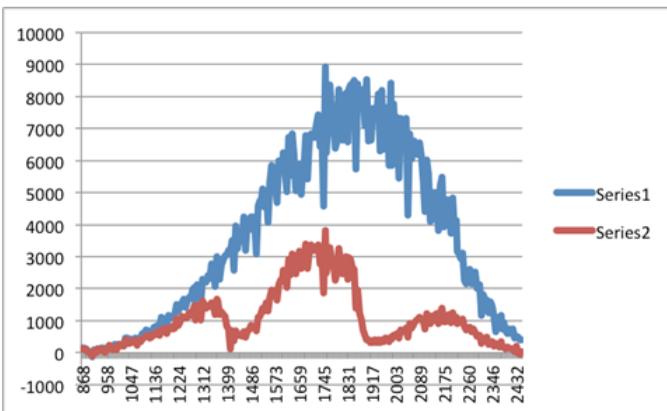
## 1 Introduction

Every multispectral imaging application is unique and the wavelength ranges of interest run from the UV through short-wave infra red (SWIR). The particular regions that will be useful for one application will be dictated by the material properties of the components that will be used for imaging. The first step to successful multispectral imaging will always be accurate identification of relevant wavelengths to utilize for imaging. For many applications, these proof of concept parameters are already known or are proprietary knowledge. Filters to be utilized in either a wheel camera or on a patterned dichroic filter array sensor can

typically be designed with 5nm to 100nm FWHM bandpasses. Therefore, the approach described here utilizing three different instruments in the experimental pathway is a valid one for commercial, industrial and academic applications.

## 2 A non-imaging start

For initial studies to better define wavelengths of interest for a system, a research spectrometer can be a very useful tool. As mentioned above, we will present data from two samples, one biological and one synthetic. The initial measurements on these roses are shown in Fig. 9.1, done with an Ocean Optics NIRQuest IR spectrometer. The differences in the reflectance measurements obtained from the roses are significant, though perhaps not what one would consider diagnostic. However, as you see from the data obtained in the NIRQuest, it hints at the fact there will be some interesting spectral differences between the two samples.



**Figure 9.1:** Raw spectral traces obtained from Ocean Optics NIRQuest IR spectrometer operating in reflectance mode, 400nm-2200nm data. Series 1 represents reflectance data from synthetic flower, Series 2 is from a natural flower.

### 3 Seeing multispectrally

With spectral data now in hand, we can move on to an imaging method that will utilize the information obtained on the two samples in the NIRQuest. The spectrometer data shows some potentially significant differences in the spectral behavior of the two samples and we will attempt to use that in the next step on our pathway. The SpectroCam from PIXELTEQ (Fig. 9.2) is a multispectral imaging system with sensitivity from the UV through the SWIR regions. A SWIR camera was utilized here with six filters in order to take the next steps towards a snapshot MSI system. The screenshot in Fig. 9.3 below demonstrates the raw image output from SpectroCam.



**Figure 9.2:** The PIXELTEQ SpectroCam design.

Spectrally specific image data can be very powerful information and provide essential proof-of-concept data before progressing to a snapshot MSI system. In this example, you can see clear visual differences in the two samples. Contrast between the biological sample as compared to the non-biological are clearly shown in filters 3-5, especially filter 5. Filter 5 here corresponds to a wavelength range of approximately 1500nm–1600nm, which matches an area of the spectral reflectance data (Fig. 9.4) indicating significant differences between the real and synthetic flowers. A larger image from filter 5 is shown below and demonstrates how an imaging system perceives the spectral differences in the 1500nm wavelength range. Important for next section to note that in a wheel-based camera, each image is collected sequentially as the filters move and cover the sensor in succession.



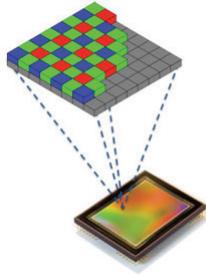
**Figure 9.3:** SpectroCam by PIXELTEQ live view screenshot. The six channels refresh constantly to provide a live view of the samples. Filters with approximately 100nm bandwidths spanning from 1050nm to 1700nm were utilized.



**Figure 9.4:** Image from filter 5 in PIXELTEQ SpectroCam MSI camera, corresponding to 1500nm-1600nm. Note the real rose on the left shows stronger signal (indicated by greater pixel intensities) in the 1500-1600nm range.

## 4 Real-time snapshot multispectral imaging system

After proving concept and imaging criteria for the system, it may be advantageous to migrate to a real-time snapshot MSI system. The primary advantage of a snapshot multispectral system is that all spectral channels are imaged simultaneously and therefore, imaging can be close to video rate in all channels. For this method, a custom sensor is created using proprietary technology [1–3]. However, the classic Bayer pattern sensor is a good illustration here to recall how typical imaging sensors are created.



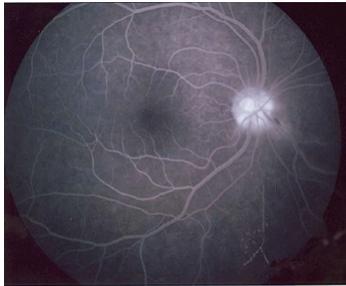
**Figure 9.5:** Classic Bayer pattern RGB sensor. Illustrative of how a dichroic filter array can be created directly on an imaging sensor.

With a custom patterned dichroic filter array sensor in place, integrated in an instrument called PixelCam, we were then able to produce the images shown below in representative screen shot, Fig. 9.6. Three channels were utilized in this demonstration camera, however, the biological matter clearly shows distinctive spectral properties when compared to the synthetic rose. That channel includes the 1500nm-1600nm range that was illustrated in the SpectroCam data. It is important to note the individual spectral channels are successfully de-mosaiced in the software and each channel provides an image of the subject matter. The images can be assigned a pseudo-color as well and reassembled into the image shown in the lower right panel of the software screen shot.



## 5 Downstream applications

The true utility and power of multispectral imaging systems is best illustrated by the wide range of applications already successfully implemented or are currently under development. To mention just a few here, MSI imaging has been a very powerful tool for: art and archaeology analysis, forensics, industrial inspection, agricultural analysis and water quality monitoring. Perhaps the most interesting however, are the potential applications for MSI systems in the biomedical sciences. As a case study in the biomedical world, consider the emerging applications both in research and in clinical use for indocyanine green (ICG) as a contrast agent for imaging. Currently growing in popularity, ICG is FDA approved for use in humans and research animals and specifically labels vascular tissues: arteries and veins. It has been successfully used for retinal angiography (Fig. 9.8) as well as in-vivo animal research.



**Figure 9.8:** Retinal angiography.

A SpectroCam wheel camera or perhaps even better yet, a PixelCam can be very useful tools for this type of imaging. The specific fluorescence emission from the ICG can be shown along with other channels (such as RGB) in order to better analyze and present image data. This particular application could be relevant for imaging during surgery, to analyze wound healing after surgery or to assess hepatic function or other circulatory system functioning in real-time. Applications are currently under development for this type of imaging and provide just one example of the potential for MSI imaging in the biomedical sciences.

## References

1. J. D. Jason M. Eichenholz, "Ultracompact fully integrated megapixel multispectral imager," ser. SPIE Proceedings, vol. 7218, 2009.
2. G. Overton, "Multispectral imaging: Filter-based multispectral imager has low complexity and cost," *Laser Focus World*, 2009.
3. J. D. Jason M. Eichenholz, "From science experiments to commercial products," *Advanced Imaging Magazine*, 2011.