Lab Write-Up Week 10

Spectra and Filters

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Fluorescent Spectra Accuracy

The peaks of a fluorescent tube light emission were compared to the expected elemental peeks to examine the accuracy of the spectrometer.

Line	True Wavelength, nm	Measured Wavelength, nm
Hg I (neutral Hg)	404.7	404.45
Hg I (neutral Hg)	435.8	435.44
Terbium (Tb^{3+})	~543-544	543.97
Hg I (neutral Hg)	546.1	545.44
Europium (in Eu^{3+}:Y_2O_3)	~611	611.85

The measured wavelength of light is offset from the expected wavelength by a linear factor.

$$\lambda_{Measured} = 0.9971\lambda_{true} + 0.9055nm$$

IR Emission

The emission measured by the spectrometer in the IR spectrum, above 700nm, differs greatly from the expected emissions from a body radiating at 2800K. This is due to the quantum efficiency of the CCD camera used inside the Spectrometer. The response the maximum at ~550nm and drops off as wavelength increases. The measured spectra is the product of the Black Body and the CCD response.

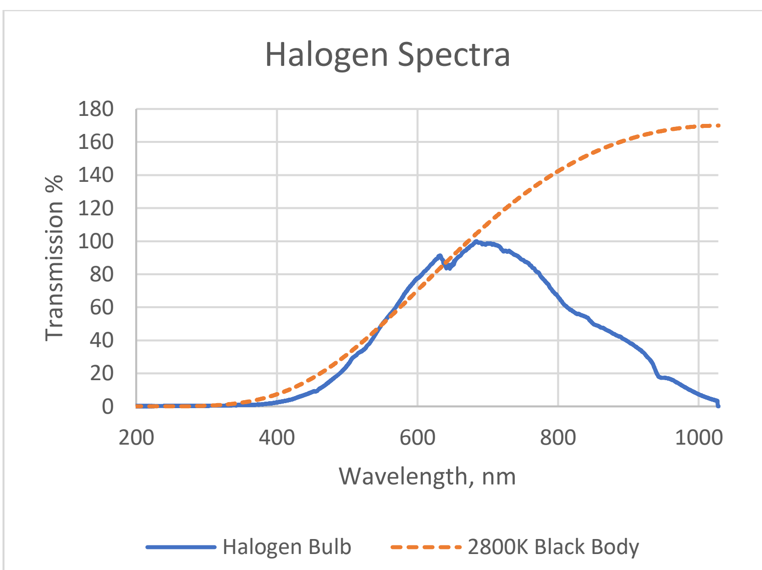


Figure 1. Halogen Bulb spectra and expected spectra.

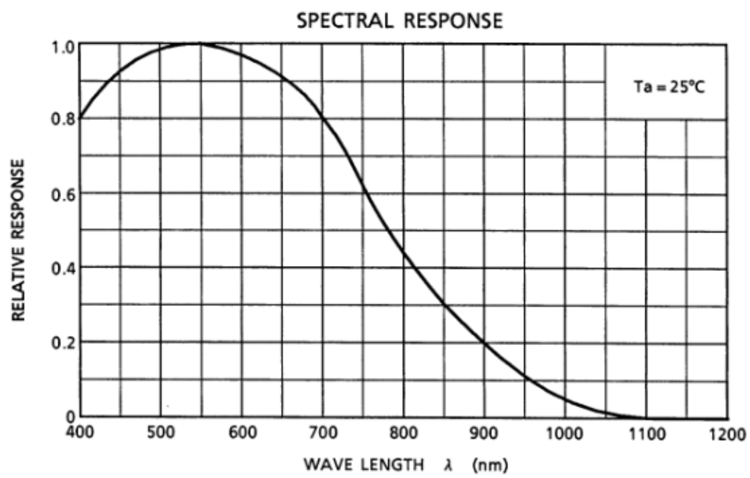
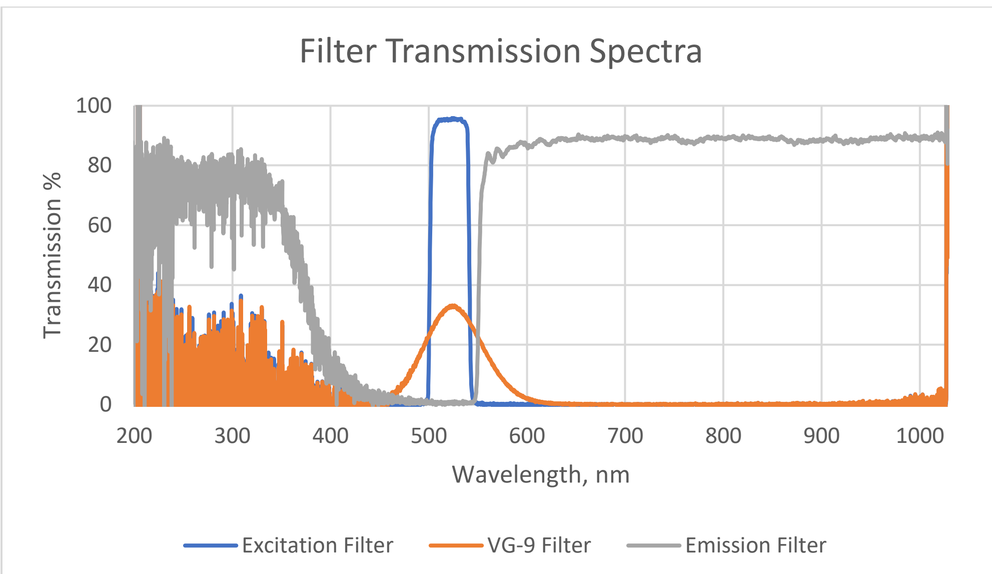


Figure 2. Spectral response of the CCD used inside the spectrometer.

Filter Transmission

The transmission of filters were measure with a spectrometer.



Using the measured spectrum, the total transmission of the Excitation filter and Emission filter in series, as well as the Excitation filter and VG-9 filter in series were calculated by taking the product of the transmission.

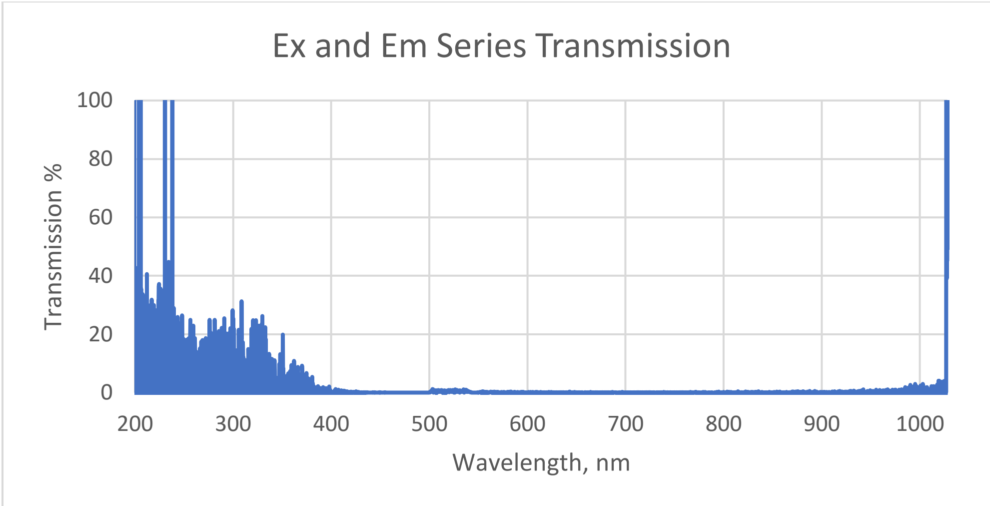


Figure 3. Transmission through Excitation and Emission filter, light is completely blocked by the filters.

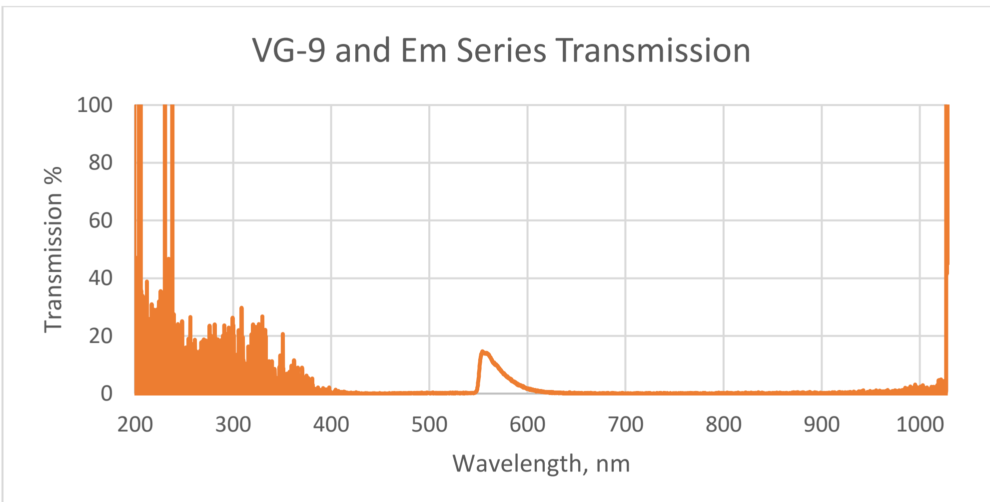
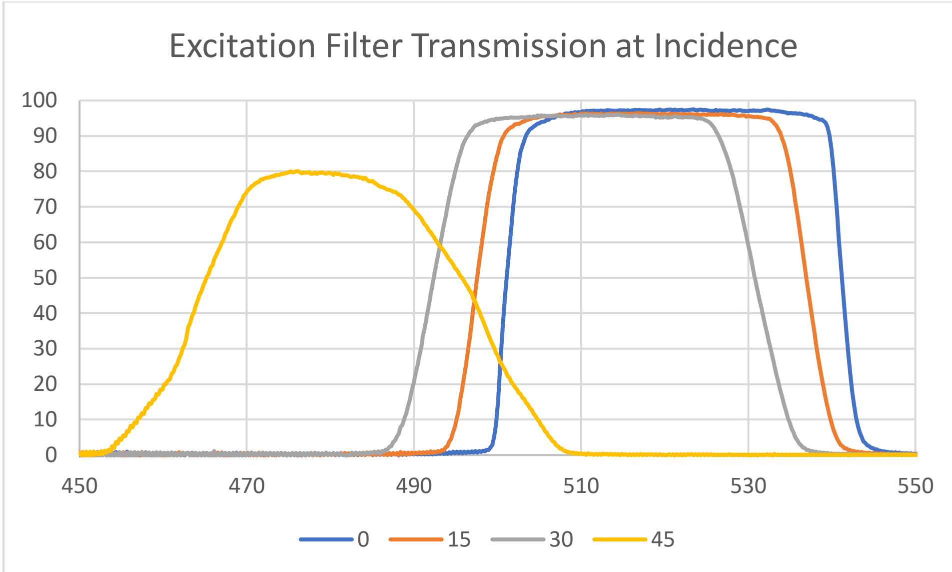


Figure 4. Transmission through VG-9 and Emission filter. Light is transmitted between 550-600nm.

Using the VG-9 color filter instead of the Excitation Filter allows light to pass through both filters when in series. This light will be much brighter than the light emitted by fluorescence, such that the image will be in brightfield.

Interference Filter Incidence Angle

The transmission of light as the Excitation filter was angled to be at different angles was measured.



As the angle of light increases the FWHM of the Transmission band shrinks. The center wavelength also becomes shorter in what is known as 'blueshift'.

It would not be a good idea to place the Excitation filter between the Condenser and the Objective. The maximum angle of light passing through the filter is related to the NA. Light passing through the Condenser will be bent sharply to focus at the sample plane, which will cause a large angle of incidence of light near the outer edges and normal incidence for light on the optical axis.

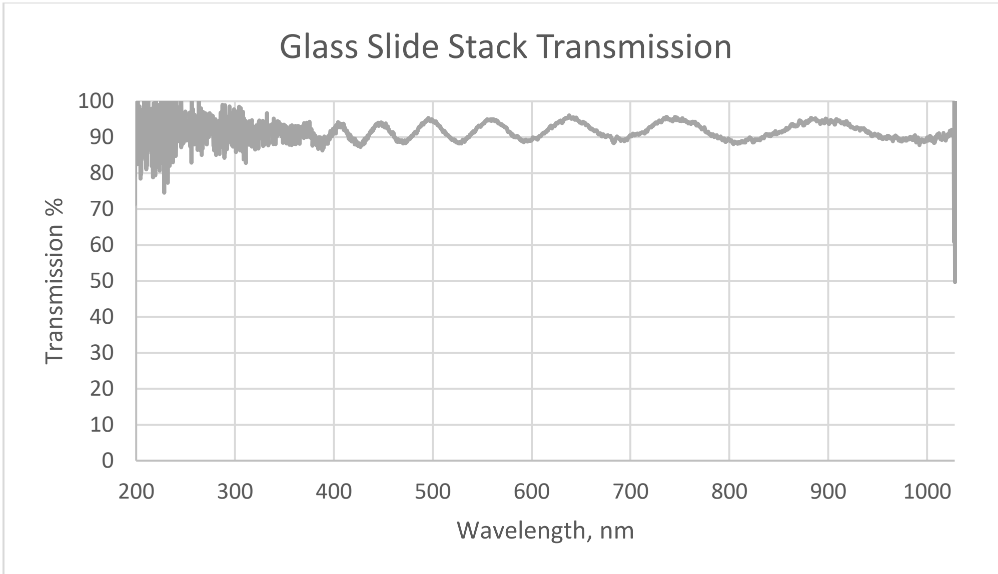
If would be worse if the emission filter were rotated 15deg versus if the excitation filter were rotated 15deg. Rotating the lens causes the transmission to blue shift. As fluorescence emits at a longer wavelength, the emission filter is always transmitting at a longer wavelength than the excitation filter. Angling the emission filter at a large enough angle will cause the transmission bands of both filters to overlap thus ruining the imaging.

If only the Excitation filter were rotated 15deg, then the total transmission would decrease as the bandpass of each filter grows further apart.

If only the Emission filter were rotated 15deg, then the total transmission would increase as the bandpass of each filter overlaps more.

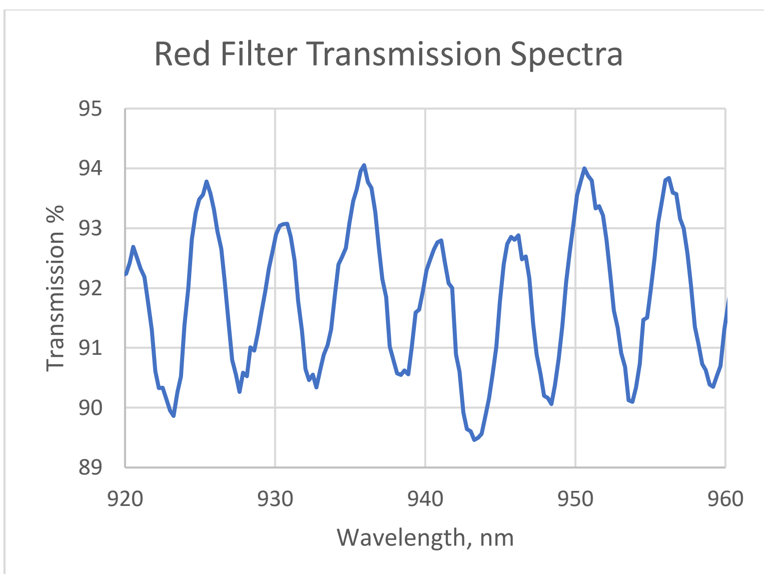
Interference Effects

Two glass slides were places in front of the spectrometer to show the interference fringes that occur.



Where the round-trip distance between the two plates is an integer multiple of wavelength, constructive interference occurs: with destructive interference occurring as half intervals. This causes the oscillations shown in the transmission spectrum, known as etalon fringes.

A red plastic spectrum of a red plastic Roscolux filter was also taken. Light reflects off both side of the filter, creating etalon fringes that can be used to measure the thickness of the filter.



The thickness of the filter can be calculated by: $d = \frac{m\,\lambda_{vacuum}}{2n}$ where $n_{cellophane} \approx 1.47$

The wavelength of several adjacent peeks were taken, and the wavenumber of the first peek was found by ensuring the calculated distance for each peak was equal to each other.

Peak wavelengths	Fringe Number	distance(nm)
925.43	4	57918
930.8	3	57938
935.93	2	57939
941.07	1	57937
945.96	0	57916

The first m_0 was found to be 180, and as such the thickness of the filter was found to be approximately 58um.

Relative Intensity

The spectra meter is calibrated against a Planck Black-Body graph in order to more accurately measures the intensity at different wavelengths.

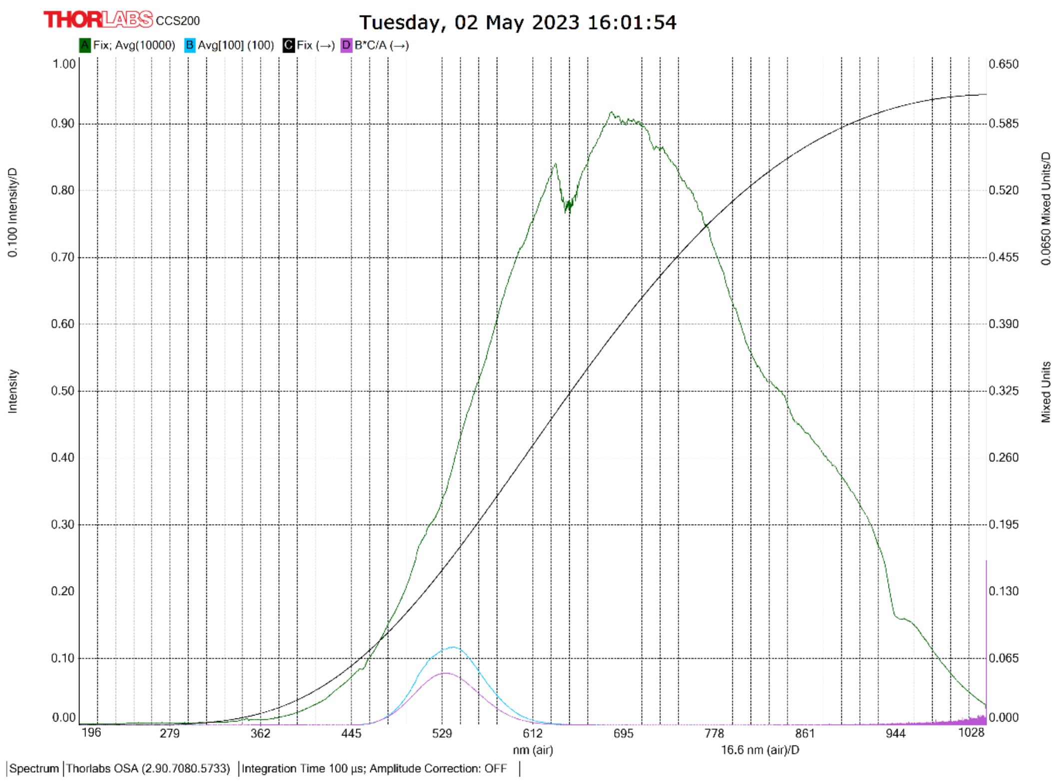


Figure 5. Green: Raw Spectrometer reference of Halogen Bulb. Cyan: Raw spectrometer reading of transmission through VG-9 Filter. Black: Graph of Planck Black-Body for 2800K object. Purple: Calibrated Spectrum of the VG-9 Filter.