

## CPL Spectrum Measurement of Green Fluorescent Protein (GFP)

### Introduction

Circularly polarized luminescence (CPL) measures the difference in the emission intensities of left- and right-handed circularly polarized light of chiral compounds. Unlike CD, CPL provides information on the excited state properties of chiral molecules.

Green fluorescence protein (GFP) is a protein that emits green fluorescence when exposed to ultraviolet light and has been shown to have chiroptical properties<sup>1</sup>. This application note illustrates the CPL and CD spectra of GFP obtained with the CPL-300 and J-1500, respectively.



**CPL-300**

Circularly Polarized Luminescence Spectrophotometer

### Keywords

J-1500, Circular dichroism, CPL-300, Circularly polarized luminescence, Protein structure, Fluorescence, Biochemistry

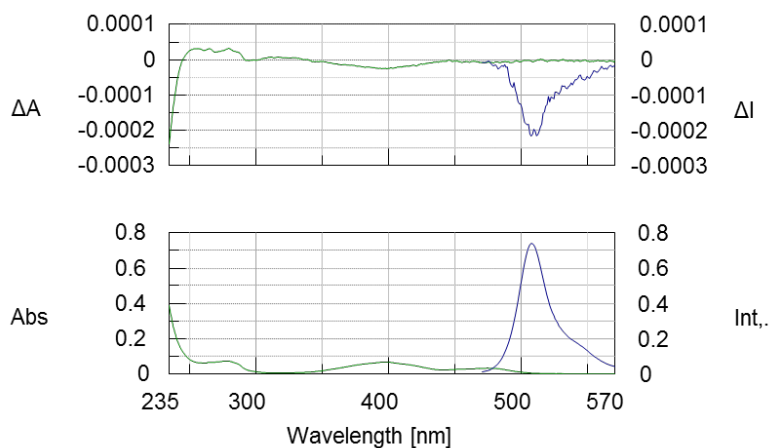
### Experimental

Measurement Conditions CPL-300 Circularly Polarized Luminescence			
Excitation Wavelength	399 nm	Excitation Bandwidth	12 nm
Data Interval	1 nm	Response	16 seconds
Emission Bandwidth	8 nm	Scan Speed	10 nm/min
Accumulations	36	Path Length	10 mm

Measurement Conditions J-1500 Circular Dichroism Spectrophotometer			
Data Pitch	0.2 nm	D.I.T.	1 sec
Bandwidth	1 nm	Scan Speed	200 nm/min
Accumulations	16	Path Length	20 mm

## Results

CD and absorption spectra (Figure 1, green) of a 0.03 mg/mL GFP solution was measured in the near-UV and visible regions. The GFP chromophore is observed in the visible region at 400 nm and the absorption bands due to the aromatic amino acid residues are observed in the near-UV region between 250 and 300 nm. The CPL and fluorescence spectra were also obtained and show in blue in Figure 1. The characteristic fluorescent and CPL spectra of GFP are observed between 470-570 nm.



**Figure 1.** CPL (blue) and CD (green) spectra (top) of green fluorescent protein. The absorption (green) and fluorescence (blue) spectra are shown at the bottom.

## References

1. Hiromasa Goto, Isao Sawada, and Nobuhiko Nomura, International Journal of Polymeric Materials, 2010, 59, 786-792.