

CPL spectrum measurement of Green Fluorescent Protein (GFP)

Introduction

Circularly Polarized Luminescence (CPL) is the difference of circular polarized fluorescence intensity between right-handed and left-handed which is emitted from chiral compounds under excited condition. There are several main target samples and applications in CPL measurement as fluorescent chiral organic compound and chiral lanthanoid complex. And Biomacromolecule such as Green Fluorescent Protein (GFP) is also different target sample. GFP is a fluorescent protein which isolated from *Aequorea victoria*, the CPL spectra of the protein have already been reported, which indicates that living jelly fish may emit the circular polarized light ¹⁾. And Mantis shrimp is known to perceive circular polarized light and this detail mechanism is expected to be elucidated in future related with the research progress.

This application note shows CPL measurement results about GFP fluorescence protein by using CPL-300.

Keyword: CPL, Green Fluorescent Protein, Biomolecule

Measurement condition

[CPL-300: Circular polarized light luminescent measurement system]

Excitation wavelength:	399 nm	Measurement Range:	450 - 570 nm
Excitation bandwidth:	12 nm	Fluorescence bandwidth:	8 nm
Scan speed:	10 nm/min	Response:	16 sec
Data acquisition interval:	1 nm	Accumulation:	36 times
Optical Path Length:	10 mm		

[J-1500: Circular dichroism spectrometer]

Measurement Range:	235 - 570 nm	Bandwidth:	1 nm
Scan speed:	200 nm/min	Response:	1 sec
Data acquisition interval:	0.2 nm	Accumulation:	16 time
Optical Path Length:	20 mm		

Measurement Results

CD and absorption spectra of GFP solution (0.03 mg/mL) was measured in the wavelength range from near UV to visible. The band of GFP fluorescent chromophore is observed in visible region. Additionally, in the near-UV region, the band of side chain of amino-acid aromatic is observed. (Fig. 1, green line) And then, CPL and fluorescence spectra of the solution are measured using CPL-300. Characteristic fluorescent and CPL spectra of GFP are observed in the wavelength range from 470 nm to 570 nm. (Fig. 1, blue line)

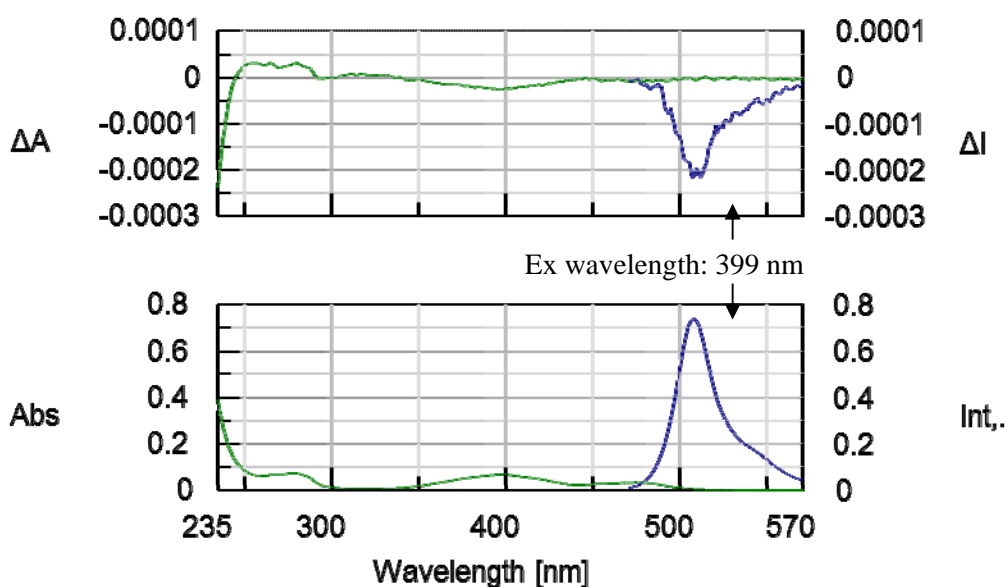


Fig. 1 Top: CPL/CD spectra, Bottom: Fluorescence/Absorption spectra of GFP solution

— : CD/Absorption spectra
— : CPL/Fluorescence spectra

<Reference>

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