

## INSTRUMENT DESCRIPTION

## Chronos

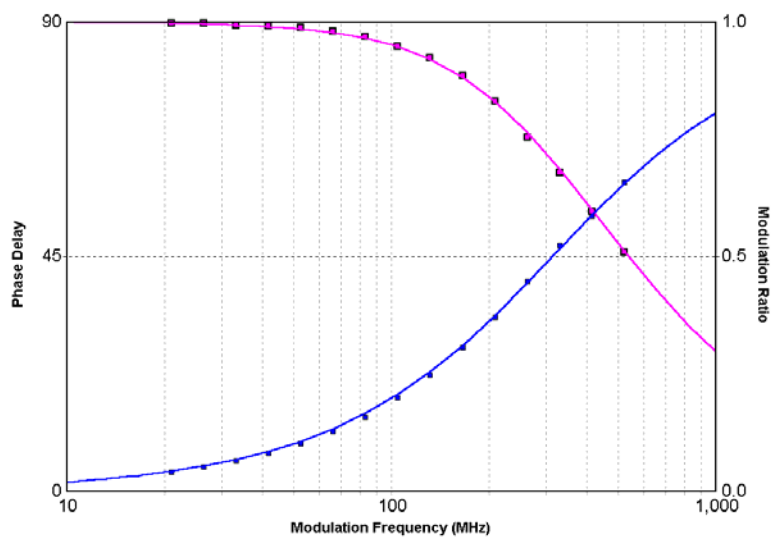
The **Chronos** is the first, frequency-domain fluorescence lifetime instrument based on the use of laser diodes and light emitting diodes (LED) as the sources for excitation. This powerful tool provides all the benefits of a full lifetime fluorometer at an affordable price. The **Chronos** has been enhanced for those users operating at standard wavelengths of excitation typically at a set region. The **Chronos** is a fully-automated instrument using a Windows based software program and can be upgraded to steady-state acquisition.

### INSTRUMENT FEATURES

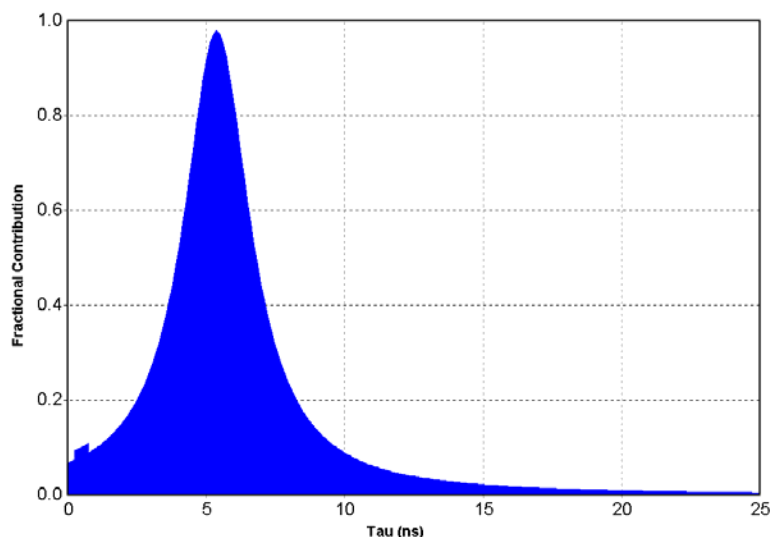
- Compact and portable
- Interchangeable modules for each wavelength
- Full hardware automation
- Full acquisition automation
- Speed – measurements from complex decays are acquired in less than one minute

### MEASUREMENTS

- Lifetime determination of multi-exponential decays
- Lifetime analysis using continuous distributions
- Rotational motions of molecules
- Phase and modulation resolved kinetics
- Phase and modulation resolved spectra
- Lifetime millisecond kinetics
- Time-resolved spectra



**Figure 1.** The plot shows the phase and demodulation data of indocynine green (ICG) in water acquired with excitation at 786 nm using a laser diode. Fluorescence was collected through a high Fluorescence was collected through a high pass filter RG830. A single decay time of 510 ps is determined.



**Figure 2.** Phase and demodulation data of Human Serum Albumin (HSA). Excitation was at 300 nm using the LEDs; emission was collected through a WG320 high-pass filter. Lifetime analysis is best using a Lorentzian distribution (center at 5.4 ns, width = 2.9 ns, fractional distribution = 98%) and a second discrete component ( $\tau = 0.51$  ns and fractional contribution = 0.02 %).

## ANALYSIS

The decay times of fluorophores in mixtures can be determined using multi-exponential, non-exponential and lifetime distribution models. The rotational correlation times of up to three independent species in complex environments is investigated assuming either isotropic, anisotropic or hindered rotators models. The evolution of time-resolved spectra is displayed both in 3D plots and by plotting the center of gravity of the spectrum at selected intervals after excitation. Phase and modulation resolved spectra and millisecond lifetime kinetics separate the spectral and time behavior of up to three independent species.

## LIGHT SOURCES

The light source utilized in the **Chronos** is typically either a laser diode or a Light emitting diode (LED). The light source modules are easily interchangeable when a user requires operation at other excitation wavelengths. Other light sources can be easily interfaced to the instrument to cover an unprecedented wavelength range.

LIGHT EMITTING DIODES		
Center wavelength (nm)	FWHM (nm)	Modulation frequency (MHz)
280	12	DC - 300
300	12	DC - 300
370	12	DC - 250
460	12	DC - 110
470	23	DC - 140
485	20	DC - 140
520	22	DC - 140

LASER DIODES	
Center wavelength (nm)	Modulation frequency (MHz)
370	0.1 - 400
405	0.1 - 650
450	0.1 - 600
635	0.5 - 1200
690	0.5 - 550
782	0.5 - 530
830	0.5 - 600

## ***Power at the push of a button . . .***

### **HARDWARE**

Adjustments to **Chronos** hardware parameters are handled via software. Automation includes control of the following: four shutters, rotating sample compartment, two stirrers, and the voltage of the photomultiplier tubes. The **Chronos** houses additional ports on the instrument bench for control of up to five additional devices. Other devices (temperature bath, titrator, high pressure pump) can be interfaced through the RS232 port of the computer and controlled from via ISS data acquisition software.

### **SOFTWARE**

The *Vinci Multidimensional Fluorescence Spectroscopy Analysis* software for Windows 98/ME/XP controls the instrument automation and its interface to computer-controlled external devices. *Vinci* includes routines for the automatic acquisition of unidimensional data files (excitation and emission spectra; polarization and anisotropy spectra; synchronous luminescence spectra; slow and fast kinetics studies) and multidimensional data files (intensity and polarization versus excitation and emission wavelengths, time, temperature, lifetime). Raw data are stored in ASCII format along with the experimental parameters. The analysis portion of the software includes data manipulation (operations between spectra, smoothing, correction, derivative and integration). The graphical display allows for 2D and 3D plots as well color display of user defined functions with zooming and rotation capabilities, statistical operators, and plot export to popular formats (metafile, bitmap).

### **UPGRADE TO STEADY-STATE OPERATION**

The **Chronos** can be upgraded for steady-state fluorescence in photon counting mode, or analog mode of acquisition, to perform the following measurements:

- Excitation and emission spectra
- Polarization (anisotropy) measurements
- Synchronous luminescence spectra
- Intensity measurements at fixed wavelengths
- Dual wavelength excitation or emission measurements

For more information please call (217) 359-8681  
or visit our website at **[www.iss.com](http://www.iss.com)**



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