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Polarization Standards for Confocal Microscopy

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"Fluorescence polarization standard for near IR spectroscopy and microscopy" Applied Optics 47(33) 6257 (2008)

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Discovery

A stretched polyvinyl alcohol (PVA) film with embedded fluorescence dyes that can be used for determining an instrumental correction factor (G-factor) in polarization measurements. Such films can be conveniently used as high polarization standards and have applications to near infrared (NIR) imaging microscopy, confocal microscopy and other spectroscopy applications and assays involving fluorescence anisotropy.

Features

- Stretched polymer films are produced with inexpensive and commercially available components, and simple chemistry.
- ➤ The fluorescent dye LDS 798 (styryl 11) has a very wide visible spectrum (450 nm to 730 nm). The chromophore can be efficiently orientated in an anisotropic environment. Dye concentrations are optimized to avoid crystallization.
- Using oriented dye-dope films; the investigator does not need to know either anisotropy values or stretching ratios to estimate the instrumental G-factor.

Benefits

- \triangleright Easy to produce thin (~100 μ m) and stable oriented PVA films.
- ➤ The LDS 798 dye spectrum affords very convenient wavelengths for laser diode excitations at 635 nm and 650 nm, which are common with today's microscopy systems.
- The strong single electronic transition in the wide (visible-MIR) range of wavelengths, its large Stoke's shift and its highly efficient orientation of the dye molecule under stretching conditions make LDS 798 dye an ideal fluorescence standard.
- Provides a more general and accurate approach to determine G-factor values. Useful for finding the G-factor in any spectroscopy/microscopy instrumentation, especially in the case of high numerical aperture objectives that distort polarization.

Opportunities

- Simple methods do not exist to test and calibrate optical paths for many confocal microscope configurations.
- Value-added technology for instrumentation manufacturers.

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