

FLUORESCENCE LIFETIME MICROSCOPY OF GREEN TO RED EMITTING FLUORESCENT DYES USING A NEW LASER

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The new laser system is a pulsed tunable laser that ranges from 488 to 640 nm. Its narrow bandwidth provides precise and noise free excitation. The repetition rate and pulse width make it an ideal excitation source for FLIM. The spectral range allows using it for measuring the lifetime of dyes that could not be evaluated using microscopy techniques before. It is fully integrated into the LSM 710 hardware and software providing both flexibility in detection and excitation without any compromises to image quality. It can be used in combination with standard FLIM equipment and retro-fitted to existing systems.

We present lifetime data from selected dyes and combination of dyes which are typically excited with blue, green or red lasers. The data have been acquired using TCSPC electronics from B&H with a detector type PMZ 100-1. For specimen with multiple labels using green, orange and red Alexa Fluor dyes coupled to antibodies the system provides lifetime data that allow for identification of the different labels. Thus, this new laser source together with pulsed diodes in the near UV covers the whole visible spectrum for FLIM experiments or applications using new FRET dyes.