Digital Fluorescence Technique for Biomedical Applications

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Abstract

We are investigating an optoelectronic imaging technique for localizing fluorescence probes in biologic specimens and experimental animals analyze the uptake, localization, and significance of these agents in biological structures.

A prototype, digital optical imaging system consisting of a white light illuminator with an optical fiber light guide, a custom designed wavelength-selective filter module, and a highly sensitive CCD imager was developed in our laboratory. For acquiring "digital fluorescence" images, the fluorescent probe is excited by the external light source with a selected wavelength. The fluorescent light emitted is then guided to the photosensitive surface of the detector through a dichroic mirror and a series of optical filters corresponding to the emission wavelength of the optical probe. The electronic output of the detector is then routed to a computer for analysis, storage and display.

Our prototype system acquires digital fluorescent images in 1K x 1K format with a 2.5cm x 2.5cm field of view and 0.024mm x 0.024mm pixel size. Spatial resolution of the digital fluorescent system is limited primarily by the detector pixel pitch. The system offers14bit digitization and the dynamic range is larger than 12 bits. Experiments with experimental animals, microtome specimens and corresponding quantitative analysis demonstrate a high sensitivity for our prototype.

The digital fluorescent imaging device was built with state-of-the-art, yet currently available optoelectronic technologies. It can be applied to cellular, molecular, histologic, embryologic and pharmacologic and other biomedical applications. Clinically feasible devices for tracking various optical probes are under development currently.

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