Abstract

Time-resolved fluorescence measurements enable the study of structure of molecular systems and dynamical processes inside them. This is possible because of a very high sensitivity of fluorescence lifetime to the physical and chemical properties of micro-environment in which fluorophores are situated. However, proper detection of the fluorescence lifetime is a challenging task, due to the fact that the fluorescence decay time of commonly used fluorophores lies in a nanosecond range. This puts strict requirements on the parameters of the fluorescence detectors.

The features of single-photon avalanche diodes (SPAD) make these optical detectors a good alternative to conventional photomultiplier tubes and micro-channel plates. CMOS technology allows cointegration of a SPAD and electronic circuits on the same substrate and provides advantages in time resolution and noise characteristics. Monolithic integration of signal processing circuits and detectors on the same chip allows using such detectors without additional external hardware.

New SPAD sensors with improved characteristics are produced every year. However, the designers consider various performance metrics while the importance of each particular detector characteristic depends on its application. Therefore, the validation and optimization of SPAD characteristics should be performed in a close connection with the analysis of a specific system, wherein this detector will be used.

This work was aimed at developing of a model able to describe a typical *fluorescence experiment with SPAD-based detector. The model simulates all essential parts of the fluorescence experiment starting from the light emission, through photo-physical processes occurring inside a bio-sample, to a detector itself and read-out electronics.*

The ability of the developed model to simulate various light sources (laser and micro-LED), fluorescence measurement techniques (time-correlated single photon counting and time-gating) was verified. The simulated results were in good agreement with the experimental data and the model proved its flexibility. Furthermore, the model provided the explanation of the distortions in experimental fluorescent curves measured under a very high ambient light when pile-up effects appear. Finally, a set of virtual experiments were established to investigate the influence of noisy pixels in SPAD array on a lifetime estimation and to study the feasibility of time-filtering instead of conventional optical filtering. Simulation results are in good agreement with data available in literature.

Keywords [single photon avalanche diode, fluorescence decay, simulation modelling, time-correlated single photon counting, time-gating]