ABSTRACT

Quantum dots (Qdots) have several advantages compared to other probes for single molecule imaging. These include enhanced brightness and photostability as well as in some cases smaller size. Perhaps the major advantage of using Qdots for single molecule imaging is the possibility of simultaneous imaging of multiple species at fast repetition rates over long periods of time. With this in mind, we have begun assembling a microscopy system eventually capable of imaging multiple colors of single quantum dots at high repetition rates over long periods of time in cells and substrate-supported planar membranes. With our current system, which consists of an Olympus IX81 microscope equipped with a 100 W Hg arc lamp for excitation and an electron-multiplied CCD (Andor DV887-EC3) for detection, we can image single Qdots with 100 µs signal integration or at rates up to about 250 Hz. These results are however very dependent on the particular emission color characteristics of the Qdots, as we find that certain quantum dot colors are dimmer and/or primarily in a non-fluorescent state. Here we present data on the intensity and on/off characteristics of a variety of Qdots. We also give examples of single molecule imaging with Qdots for tracking membrane proteins in cells.

RESULTS AND FUTURE EXPERIMENTS

In this work, the data presented is limited to streptavidin-Qdots conjugated from Zinprogen. These materials were previously sold by Quantum Dot Corporation. In order to identify the best Qdots for SPT, we imaged single Qdots that had been non-specifically immobilised on a glass coverslip. For these measurements stock quantum dots (~1 µM) were diluted in 1:1 in 50 mM sodium borate, pH 8.2 with 1% BSA. Imaging specimens were prepared by applying 65 ml of a dilute solution of streptavidin-Qdots to a sample chamber consisting of a 22 x 22-mm No. 1 1/2 glass coverslip.

REFERENCES