第78回応用物理学会秋季学術講演会 講演予稿集 (2017 福岡国際会議場)

Deep-UV Surface Plasmon for Bio-Imaging

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1. Introduction

Surface plasmon resonance (SPR) has attracted considerable attention in relation to optical biosensors, solar cells, holography, and other applications, because it can enhance the electric field of incident light by several factors of ten.12 Its application to bioimaging has also been extensively investigated. Fluorescence imaging has been widely used to analyze the dynamic behavior of cellular components. Recently, deep-ultraviolet surface plasmon resonance (DUV-SPR) has also been investigated [1-3]. It can potentially be used in combination with photoelectron emission and Raman scattering measurements because of the high photon energy associated with DUV light. In the present study, DUV-SPR was used for the simultaneous excitation of enhanced multicolor fluorescence from several types of quantum dots. This technique was also applied to high-sensitivity observation of dye-labeled cells.

2. General Instructions

DUV-SPR enhanced fluorescence imaging was used to observe dye-labeled HeLa cells cultured on the aluminum film. As a reference, cells were also cultured on a slide glass. The mitochondria, actin, and nuclei in the cells were stained with Mito Tracker (red), ATTO 488 (green), and DAPI (40 ,6-diamidino-2-phenylindole) (blue), respectively. These fluorophores are commonly used for staining specific organelles in cells. Their emission wavelengths are 599, 523, and 461 nm, respectively. The laser power was 2 mW, and the CCD exposure time was 1 s.

Figure 1(a) shows a DUV-SPR enhanced fluorescence image of stained HeLa cells on aluminum. Individual organelles can be clearly observed. On the other hand, Fig. 1(b) is a fluorescence image of HeLa cells on the glass slide, with no aluminum film present. In the absence of a DUV-SPR effect, the intensity is seen to be extremely low. Figures 1(c) and 1(d) show intensity profiles along the dashed lines in Figs. 1(a) and 1(b), respectively. The fluorescence intensity is about three times higher in the DUV-SPR enhanced image. Figures 1(e)–1(g) show images band-pass filtered at 575–625 nm, 510–550 nm, and 442–492 nm, respectively. The fine particles around the cell nucleus in Fig. 1(e) represent mitochondria, the fibrous structures in Fig. 1(f) represent actin, and the central cells in Fig. 1(g) represent nuclei. Thus, highly sensitive imaging of dye-labeled HeLa cells can be achieved using DUV-SPR.

3. Conclusions

In conclusion, theoretical and experimental investigations were carried out into DUV-SPR excitation on an aluminum film. Numerical calculations were first performed to optimize the aluminum thickness and the incident angle of the light. Experimental measurements demonstrated fluorescence from stained HeLa cells. DUV-SPR led to a significant increase in the fluorescence intensity. In the stained cells, mitochondria, actin, and cell nuclei could be simultaneously observed.

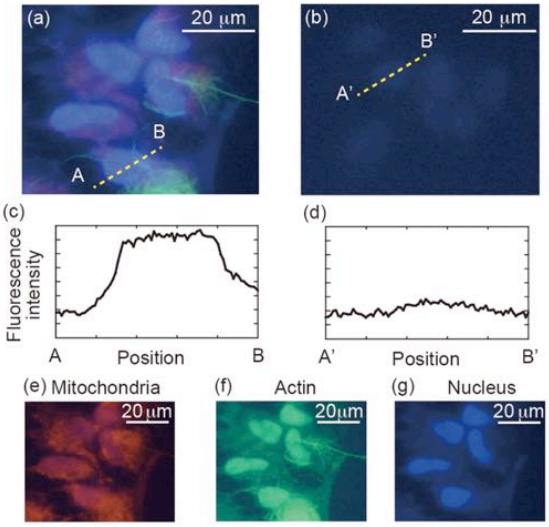

Fig. 1. Observation results of HeLa cells with DUV-SPR

References

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