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High resolution imaging by electron-beam excitation assisted optical microscope with emission layer for a brighter nanometric light source

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Observations of biological specimens in nanometer range are one of the most promising techniques to improve nano-science and bioscience. We have developed an electron-beam excitation assisted (EXA) optical microscope [1] which has a few tens nanometer spatial resolution laterally in various surroundings like air or liquids. With an EXA microscope, we can observe optical constants like absorptions, refractive indexes, polarization properties, and their dynamic behaviors in nanometer scale.

Figure 1 shows schematic diagram of focal point in an EXA microscope. A nanometric light spot, as shown in Fig. 1, is formed in the luminescent film by irradiation of focused electron beam. The nanometric structures of specimens on the luminescent film are probed by the nanometric light source.

In the EXA microscope, stronger emission from the thin film is required to improve signal to noise ratio. We employed Si_3N_4 thin film as a separator of vacuum and air, and Si_3N_4 emits the light by electron beam irradiation. However, the intensity of the light from Si_3N_4 film was low for an observation of living cells with the EXA microscope.

We propose adding an emission layer on Si_3N_4 thin film for the brighter nanometric light source. This approach also makes it possible to select the wavelength. In this study, we chose Y_2O_3:RE as a luminescent film. Y_2O_3:RE is well known as a brighter luminescence emitter by irradiated electron beam. The wavelength of Y_2O_3 can be changed with rare-earth doping.

We chose Y_2O_3:Eu as a luminescent film. Y_2O_3:Eu film was deposited on a Si_3N_4 thin film with electron beam evaporation. The emission wavelength was 614 nm. The fluorescent intensity of the Y_2O_3:Eu film on the Si_3N_4 film was 135 times higher than that of only Si_3N_4 film.

We observed gold nanoparticles for the spot size estimation. The diameters of the gold nanoparticles are 98.8 ± 7.9 nm. In figure 2, (a) shows an optical image of the gold nanoparticles with EXA microscope, and (b) shows a corresponding back scattered electron image with scanning electron beam microscope (SEM). The distribution of gold nanoparticles observed with EXA microscope is well matched with that of the SEM observation. Figure 2(c) shows a light intensity of the line drawn in figure 2(a). The width of the gold nanoparticle was 93.7 nm. Because we observed 100 nm gold nanoparticles with the EXA microscope with Y_2O_3:Eu film, we could confirm that the resolution was higher than diffraction limit of the light.

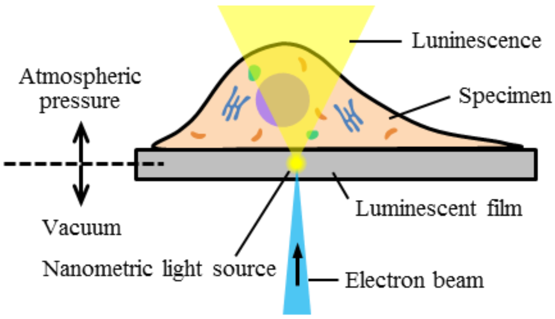

Figure 1: schematic diagrams of focal point in the EXA microscope. In this study, the luminescent film is Si_3N_4 thin film or Si_3N_4 + Y_2O_3:Eu thin film.

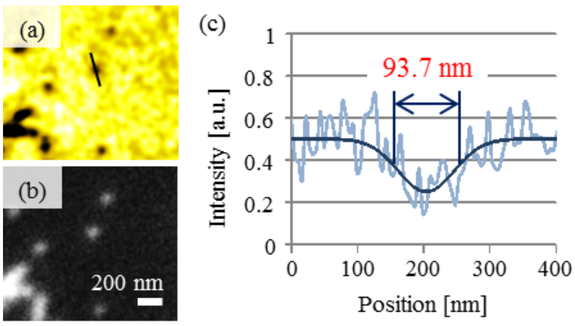

Figure 2: (a) Observation result of 98.8 ± 7.9 nm gold nanoparticles with the EXA microscope. (b) A back scattered electron image of gold nanoparticles with a SEM. The Scale bar is 200 nm. (c) A line profile of the line in (a).

[1] W. Inami *et al*, Optics Express, **18**, 12897-12902, (2010)

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