## **Supplementary Information for:**

# Color-encoded Microcarriers Based on Nano Silicon Dioxide Film for Multiplexed Immunoassays

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#### Principle of color encoded method

The reflectivity of the nano film was calculated assuming a constant refractive index of the  $SiO_2$  layer of 1.45. The two variables that remain are the wavelength of the incident beam and the thickness of the oxide layer, which thus directly determine the reflectivity. The reflection coefficient of the  $SiO_2$  film on an Si substrate can be approximated by Airy's formulas:<sup>1</sup>

$$r = \frac{r_1 + r_2 e^{-2i\phi_{12}}}{1 + r_1 r_2 e^{-2i\phi_{12}}} \tag{1}$$

Where  $r_1$ ,  $r_2$  are the Fresnel reflection coefficients of the air/SiO<sub>2</sub> and SiO<sub>2</sub>/Si interface, respectively, which can be calculated by Fresnel's formulas:

For s polarized light

$$r_{s} = \frac{n_{i}\cos\theta_{i} - n_{t}\cos\theta_{t}}{n_{i}\cos\theta_{i} + n_{t}\cos\theta_{t}}$$
(2)

For p polarized light

$$r_p = \frac{n_i \cos \theta_t - n_t \cos \theta_i}{n_i \cos \theta_t + n_t \cos \theta_i}$$
(3)

Here  $n_i$  and  $n_i$  are the refractive indices of the incident and refractive media, respectively.  $\theta_i$  and  $\theta_i$  are the incident and refractive angles, respectively.  $\phi$  is the optical phase difference between the two reflections,  $r_1$  and  $r_2$ . For each wavelength,  $\phi$  is given by:

$$\phi = \frac{4\pi n_t d\cos\theta_t}{\lambda} \tag{4}$$

For the silicon dioxide layer,  $n_t$  is equal to 1.45; d is the physical thickness of the oxide film. Finally, the reflectivity of the nano-film can be written as:

$$R(\lambda) = \frac{1}{2} \left( r_s^2 + r_p^2 \right) \tag{5}$$

The reflective spectrum of the microcarriers with nano-films can be written as:

$$P(\lambda) = S(\lambda) \cdot R(\lambda) \tag{6}$$

 $S(\lambda)$  is the spectrum of the optical source, which is measured using a spectrometer (Maya, Ocean Optics, Dunedin, FL). Using equations (1)-(6), the reflective spectrum  $P(\lambda)$  from 380 nm to 780 nm for each oxide thickness could be calculated by a program in Matlab 2010a.

To calculate the RGB values corresponding to the respective reflective spectrum of the microcarrier with a certain SiO<sub>2</sub> film, the reflective spectrum  $P(\lambda)$  was converted into RGB space by the classic RGB model proposed by G. Wald in 1964.<sup>2</sup>

### Atomic Force Microscopy (AFM) measurements

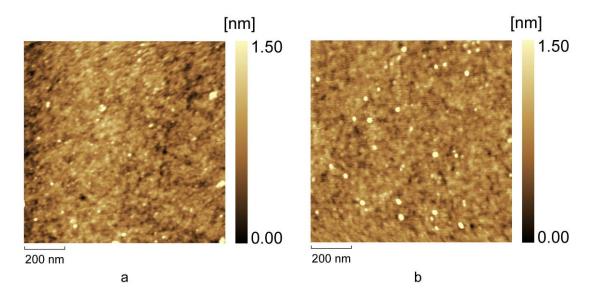


Figure S1. AFM micrographs of SiO<sub>2</sub> layer with thicknesses of 267.2 nm (a) and 360.7 nm (b).

#### References

- 1. P. Yeh, *Optical waves in layered media*, John Wiley, New York, 2005.
- 2. G. Wald, Science, 1964, 145, 1007-1016.