Electronic Supplementary Information

Dendrimer Matrix for Performance Enhancement of Evanescent Wave Absorption based Fiber-Optic Biosensor

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Fig. S-1 Calibration curve of FITC molecules in tri-sodium phosphate buffer solution.
S-1: Algorithm used for average height calculation and peak detection for AFM images in MATLAB 6.5.

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Main Algorithm
Start
Read the image and multiply by conversion factor
Detect peaks using the peak detection algorithm
Calculate mean and standard deviation of all the pixel values
Define pixel value ranges between mean-std and mean+std in steps of 0.2^std
Collect the peaks that fall in each range and display
Stop

Peak Detection Algorithm
Start
Read the image and multiply by conversion factor
Scan each pixel of the image
For each of the pixel of interest, locate all the surrounding neighboring pixels and form a 3x3 matrix with the pixel of interest in the center
Are all the neighboring pixel values less than the pixel of interest?
No
The pixel of interest is not a peak
Stop
Yes
Scan the next pixel
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S-2: Surface area calculation for sensor matrix

Since, there was no significant change in average height of bare glass (1.83 ± 0.18 nm), silane (1.85 ± 0.26 nm) and CDI (1.83 ± 0.18 nm) coated surfaces, it was assumed that the surface area is nearly same (surface area for 1 um × 1 um of sensor surface = 1 µm²) for these three surfaces. In contrast, average height was found 3.65 ± 0.43 nm for dendrimer matrix due to globular shape of dendrimer. Due to this, there was a definite increase in surface area of dendrimer matrix. The surface area was calculated by assuming all the features having ellipsoid shape as shown in Fig. S-2.
The dimensions $a$ and $b$ were taken as radius of dendrimer molecule which is nearly 2.25 nm as reported in literature [1,2]. The peak height, i.e. $h$, was calculated by image analysis using MATLAB. Thereafter, the surface area ($S$) of half ellipsoid (only the upper half ellipsoid will be available for bioconjugation) was calculated as per given formula.

$$S \approx 2\pi\left[\left(\frac{a^p b^p + a^p h^p + b^p h^p}{3}\right)^{1/p}\right]$$

where $p = 1.6075$

Finally, total area of the sensor surface was calculated by the following formula:

Total surface area = surface area of bare glass surface + area of all half ellipsoid features – surface area of ellipsoidal circle (i.e. area beneath the dendrimer molecules)

For example, assume a 1 um $\times$ 1 um dimension of glass surface on which dendrimer was immobilized. Now, assume $n$ peaks were detected by the peak detection algorithm in MATLAB in the height range from Avg height – SD to Avg height + SD. Let the heights of these peaks be $h_1, h_2, \ldots, h_n$.

$$S_n \approx 2\pi\left[\left(\frac{a^p b^p + a^p h_1^p + b^p h_n^p}{3}\right)^{1/p}\right]$$

$$\Sigma S = S_1 + S_2 + S_3, \ldots, S_n$$

Total surface area = $1 + \Sigma S - n \pi a b$
**Fig. S-3** Absorbance spectra of FITC-HIgG antibody solution (conc. = 100 µg/mL) exhibiting the excitation peak at 495 nm.

**Fig. S-4** Time-resolved absorbance response obtained from rabbit IgG antibodies coated silanized (●) and dendrimerized (■) sensor matrices when incubated with nonspecific analyte, i.e. FITC-GaHIgG (conc. = 10 µg/mL).
References: