



Optimization of a waveguide-mode sensing chip for an ultraviolet near-field illumination biosensor

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Abstract: A waveguide-mode sensor with a planar sensing chip, consisting of two waveguiding layers and a glass substrate, is a promising candidate for a near-field illumination biosensor. Aiming at using fluorescent labeling induced by ultraviolet light, we optimize the structure of a waveguide-mode sensing chip, based on the mechanism for enhancing ultraviolet near-field light revealed by numerical calculations. Candidates of optimal materials are also presented. The chip optimized as above should be able to enhance the intensity of ultraviolet near-field light 25 times as high as an Al surface plasmon resonance sensing chip.

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1. Introduction

Near-field light is generated when light is totally internally reflected at the interface between two substances such as a glass substrate and liquid. As the penetration depth or the illumination space of the near-field light is limited, total internal reflection fluorescence (TIRF) microscopy utilizing near-field light is more suitable for observing a substance put on the surface of a substrate than other techniques such as epi-illumination microscopy and confocal microscopy [1]. Therefore, TIRF microscopy is used, for example, for a cell imaging [1,2] and for detecting fluorescent-labeled protein [3] in an aqueous buffer solution. Here, the electric field strength of the near-field light can be enhanced by surface plasmon resonance (SPR) [4–7]. Therefore, surface plasmon enhanced TIRF microscopy can excite fluorescent molecules more effectively than conventional TIRF microscopy [8–10].

Many fluorescent labels adequate for the above fluorescence have been developed in recent years. Their excitation wavelengths range from UV to near-infrared. Especially, fluorescent labels using quantum dots have attracted much attention, since they can emit bright fluorescence with a large Stokes shift and have a high resistance against bleaching [11].

Aiming at developing methods to excite fluorescent labels more efficiently, much research has been carried out. However, most research focuses on SPR in a visible region using a metal thin film such as Au and Ag [12,13]. Compared to the visible region, research has scarcely been done for the UV region. Although attempts to enhance SPR using Al thin layers have been reported [14,15], their enhancement ratios of electric field are lower than those for the visible region. Moreover, Al is oxidized easily in air, which makes the electric field enhancement by SPR smaller, although Al oxide has an advantage of preventing fluorescence quenching by the Al layer [15].

A waveguide structure is another candidate to induce evanescent field with enhanced electric field. Figure 1 shows a schematic configuration of a waveguide-mode (WM) sensor [16–21]. The sensing chip is composed of two waveguiding layers such as Si and SiO₂ on a SiO₂ substrate. When light with a proper wavelength illuminates the sensing chip at an appropriate incident angle, it can be propagated within the layers. The WM resonance can be excited by both p- and s-polarized lights, but the sensitivity of WM resonance is higher for s-polarized light than for p-polarized light [16]. Therefore, s-polarized light is assumed in the following chapters in this study.

From this viewpoint, we optimize the structure of a WM sensing chip to realize high-efficiency UV near-field illumination. We analyze the relationship between the refractive index and the optimal thickness for the layers theoretically. In addition, we calculate the normalized electric field strength at the interface between the sensing chip and the buffer using a transfer matrix method [22,23] and select optimal materials for the layers based on the calculation results.

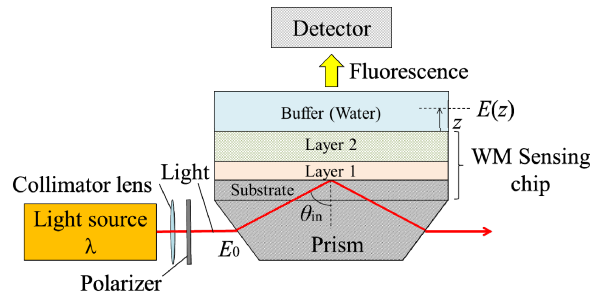


Fig. 1. Optical arrangement of a WM sensor in the Kretschmann configuration (not to scale).

2. Theory

The WM sensing chip has the configuration and optical arrangement shown in Fig. 1. Figure 2(a) shows a schematic of the cross section of the sensing chip and a buffer solution. On the Cartesian coordinate system shown on the right, the interface between the sensing chip and the buffer with analytes is set to $z = 0$. When light, which is a coherent plane wave, illuminates the sensing chip from the side of a substrate via a prism, the electric field intensity in the sensing chip is approximated one-dimensionally as a function of z [23]. When $E(z)$ denotes the electric field of light at position z on the light incident surface, which is perpendicular to the interface between the chip and the buffer, the normalized electric field strength squared $N(z)$ [23] is defined as

$$N(z) = \left| \frac{E(z)}{E_0} \right|^2, \quad (1)$$

where E_0 denotes the electric field of incident light before the light illuminates the prism. Fluorescent-labeled analytes show strong fluorescence if $N(0)$ is large [8]. Therefore, the structure of the sensing chip must be optimized to maximize $N(0)$.

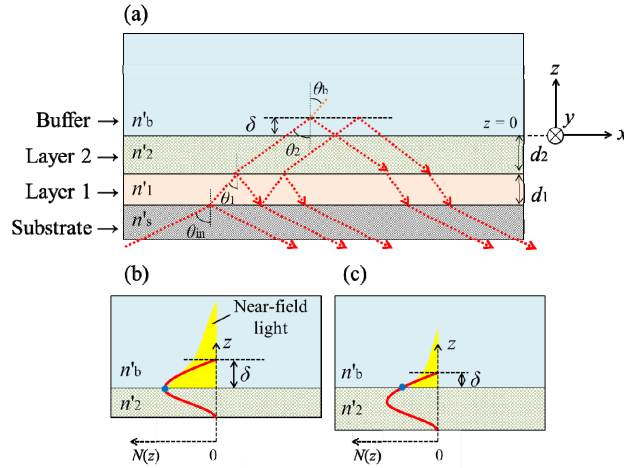


Fig. 2. Schematic views of the cross-sectional structure of the WM sensing chip. The directions of the light reflection and transmission (a) and the change in value of $N(z)$ in the chip when $\theta = \theta_c$ (b) or $\theta > \theta_c$ (c), θ_c : critical angle of total reflection (not to scale). n'_s, n'_1, n'_2 , and n'_b denote the complex refractive indices of the substrate, the layer 1, the layer 2, and the buffer, respectively. Yellow areas in (b) and (c) are conceptual images of near-field light.

Light is reflected and transmitted in the sensing chip as shown in Fig. 2(a), depending on the values of complex refractive indices of the substrate, the layer 1, the layer 2, and the buffer. Here, complex refractive index n' of a substance is expressed as $n' = n - ik$, where the real part n is the refractive index in a narrow sense and k is the extinction coefficient that represents the absorption of light. When k of a substance is large, light is much absorbed in the layer and the intensity of reflected light is decreased. Thus, the values of k of the layers 1 and 2 are assumed to be zero in this chapter.

In Fig. 2(a), the equation,

$$n_s \sin \theta_m = n_1 \sin \theta_1 = n_2 \sin \theta_2, \quad (2)$$

is satisfied according to Snell's law. Here, n_s, n_1 , and n_2 respectively represent the refractive indices of the substrate, the layer 1, and the layer 2, while θ_m, θ_1 , and θ_2 respectively denote the incident angles to the layer 1, to the layer 2, and to the buffer. When materials of the prism and the substrate are assumed to be SiO_2 as in our previous work [16–21], θ_m agrees with that to the substrate.

Total internal reflection occurs when θ_2 satisfies the relation

$$\theta_2 \geq \sin^{-1} \left(\frac{n_b}{n_2} \right) = \theta_c, \quad (3)$$

at the interface between the layer 2 and the buffer. Here, θ_c is the critical angle for total internal reflection and n_b is the refractive index of the buffer. In this case, near-field light is generated and the position where light is totally reflected is slightly inside the buffer as shown in Fig. 2(a).

The energy reflectivity R at the interface between the layer 2 and the buffer is expressed as [24]

$$R = \left(\frac{\sin(\theta_2 - \theta_b)}{\sin(\theta_2 + \theta_b)} \right)^2 = \exp(j2\Phi), \quad (4)$$

$$\Phi = 2 \tan^{-1} \left[\frac{\sqrt{(n_2 \sin \theta_2)^2 - n_b^2}}{n_2 \cos \theta_2} \right], \quad (5)$$

where θ_b denotes the complex angle of refraction in the buffer. Here, Φ in Eq. (5) is the Goos-Hänchen shift of the near-field light.

In Fig. 2(a), the distance δ from the surface of the chip to the position, at which light is totally reflected, is expressed as [24]

$$\delta = \frac{\lambda(\pi - \Phi)}{4\pi n_2 \cos \theta_2}. \quad (6)$$

When θ_2 is equal to the critical angle θ_c , $\Phi = 0$ is satisfied from Eq. (5). Then, the distance δ becomes $\delta = \lambda/(4n_2 \cos \theta_2)$ from Eq. (6). In this case, the maximum light intensity appears at the interface between the layer 2 and the buffer. That is, the maximum of $N(z)$ is $N(0)$ as shown in Fig. 2(b). On the other hand, when θ_2 becomes larger than θ_c , Φ becomes larger and δ becomes smaller. That is to say, the position of the maximum of $N(z)$ shifts toward the inside of the layer 2 and $N(0)$ becomes smaller, as shown in Fig. 2(c). This means that θ_2 should be as close to θ_c as possible but slightly larger because analytes in the buffer or temperature of the buffer may increase n_b in an actual system.

Next, the thicknesses of the layers are optimized. In order to increase $N(0)$, the thicknesses of the layers must be set so that the phase of the light reflected at the interface between the substrate and the layer 1, that of the light reflected at the interface between the layer 1 and the layer 2, and that of the light reflected at the position of the distance δ from the surface of the chip are synchronized. Here, when light transmitted through a medium is reflected by a medium with a higher n , its phase is shifted by π whereas when light is reflected by a medium with a lower n , the phase shift does not occur. In case of the WM sensing chips [16–21], which satisfy $n_s < n_1$ and $n_1 > n_2$, the first reflection at the interface between the substrate and the layer 1 causes a phase-shift of π . On the contrary, the second reflection at the interface between the layer 1 and the layer 2 and the third reflection at the distance δ from the surface of the chip do not cause the phase shift.

For the thickness d_1 of the layer 1, the optical path difference between the two lights reflected at the first interface and the second interface is $2d_1 \cos \theta_1$. Therefore, the most suitable d_1 can be determined by the equation,

$$d_1 = \frac{\left(m_1 + \frac{1}{2}\right)\lambda}{2n_1 \cos \theta_1}, \quad (7)$$

where m_1 is zero or a positive integer.

For the thickness d_2 of the layer 2, when we assume a virtual thickness,

$$D = d_2 + \delta, \quad (8)$$

the suitable thickness is determined by the equation,

$$D = \frac{m_2 \lambda}{2n_2 \cos \theta_2}, \quad (9)$$

where m_2 is a positive integer. If we set $m_1 = 0$ and $m_2 = 1$, we obtain

$$d_1 = \frac{\lambda}{4n_1 \cos \theta_1}, \quad (10a)$$

and

$$D = \frac{\lambda}{2n_2 \cos \theta_2}. \quad (10b)$$

When d_1 and d_2 satisfy the conditions calculated from Eqs. (10a) and (10b) and θ_2 is set to be slightly larger than θ_c , $N(z)$ changes as shown in the solid red line in Fig. 3.

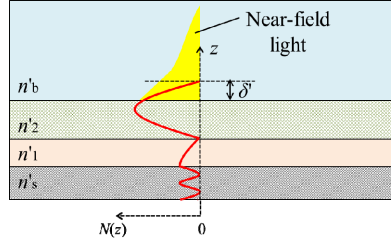


Fig. 3. Change in value of $N(z)$ in the WM sensing chip when the real part of n'_1 is higher than those of n'_s and n'_2 . Yellow area is a conceptual image of near-field light.

3. Optimization of the structure and materials for a WM sensing chip

The materials of the layers 1 and 2 of the WM sensing chip are optimized to enhance UV near-field light effectively. Born and Wolf calculated reflection and transmission coefficients in a multilayered medium using a numerical method called “transfer matrix method” for solving the Maxwell equations [22]. Using software based on a similar transfer matrix method, the electric field distribution in a multilayered medium with parallel boundaries of layers can be calculated [23]. Assuming that the incident light, which is a coherent plane wave, illuminates the multilayered medium, $N(z)$ in Eq. (1) is calculated by multiplying 2×2 characteristic matrices of the multilayers. The phase of the reflected light can also be calculated by inputting the complex refractive index of each layer in the matrix [23].

In this chapter, $N(z)$ of a WM sensing chip was calculated by the software under the following conditions. The incident light with a wavelength λ of 375 nm is assumed to be s-polarized and both the prism and the glass substrate are assumed to be made of SiO_2 as mentioned in Chapter 1 and 2. As a buffer solution to be dropped on a chip, water ($n_w = 1.354$, $k_w = 0.000$ [25]) is assumed for simplicity. From Eqs. (2) and (3), θ_c is calculated to be 66.8° for a SiO_2 substrate with $n_s = 1.473$ and $k_s = 0.000$ [25]. Based on this calculation, θ_{in} is set as 67.0° .

As a first step, the layer 2 is assumed to be SiO_2 and the layer 1 is optimized as for its d_1 , n_1 , and k_1 . Namely, $N(0)$ at the interface between the sensing chip and the water is calculated as a function of n_1 and k_1 , where d_1 is optimized to give the maximum value of $N(0)$ for given values of n_1 and k_1 .

Figure 4(a) shows the results of the maximum value of $N(0)$, which is referred to as $N_M(0)$. Note that the ranges of abscissa and ordinate of Fig. 4(a) are chosen by taking account of complex refractive indices of possible applicable materials shown in the figure. The values of $N_M(0)$ are indicated by the colors shown on the right in a logarithmic scale. That is, warm colors represent high values of $N_M(0)$. As shown in Fig. 4(a), for realizing high values of $N_M(0)$, n_1 should be as high as possible, while k_1 should be as low as possible. The values of n_1 and k_1 at the wavelength of 375 nm of several candidates for the layer 1, ITO (i), diamond (ii), SiC (iii), TiO_2 (iv), ZnO (v), and Si (vi) [25–28], are also shown in Fig. 4(a). Furthermore, in Fig. 4(b), the values of $N_M(0)$ calculated for the above candidates of layer 1 are shown, together with the corresponding thickness d_1 [nm] in parentheses that yields each value of $N_M(0)$. In Fig. 4(b), it is clear that d_1 agrees with the value calculated from Eq. (10a). The thickness of the SiO_2 layer 2 is 176 nm in all cases of (i) – (vi), which also agrees with the value calculated from Eq. (10b). As for Si (vi), which is used as the layer 1 for the

WM sensors [16–21], $N_M(0)$ is only 20.3 even though it is as thin as 14 nm. Therefore, Si is not suitable for near-field illumination in a UV region. TiO_2 (iv) increases $N_M(0)$ to 138 which is 6.8 times as high as that of Si.

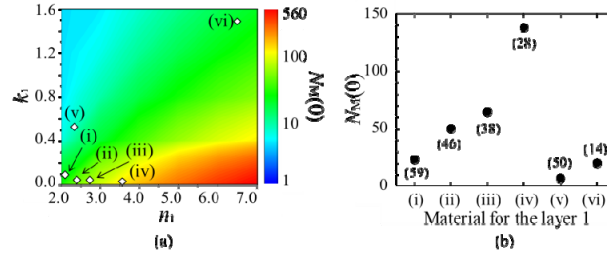


Fig. 4. (a) Calculation results of $N_M(0)$ as a function of n_1 and k_1 . White squares stand for the values of ITO (i), diamond (ii), SiC (iii), TiO_2 (iv), ZnO (v), and Si (vi). The layer 2 is assumed to be SiO_2 . (b) Values of $N_M(0)$ for the candidates (i) – (vi). Numerals in parentheses are the optimized thicknesses d_1 [nm] that yield these values of $N_M(0)$.

Next, the material is optimized for the layer 2. In this case, we want to maximize $N_M(0)$, on the condition that the layer 1 is TiO_2 . The results are shown in Fig. 5. In Fig. 5(a), it is clearly indicated that n_2 and k_2 should be as low as possible. Although not shown here, $N_M(0)$ is not enhanced for $n_2 < 1.356$ because the light is totally reflected between the layers 1 and 2 and it does not reach the interface between the layer 2 and the water. The values of n_2 and k_2 of several materials, namely, MgF_2 (vii), CaF (viii), SiO_2 (ix), PMMA (x), and BK7 (xi) are plotted in Fig. 5(a) [25,29,30]. In Fig. 5(b), the values of $N_M(0)$ in addition to the corresponding optimized values of d_2 are shown. It is clear that d_2 agrees with the value calculated from Eq. (10)b). The thickness of the TiO_2 layer 1 is always 28 nm, which also agrees with the value calculated from Eq. (10)a). MgF_2 (vii) can increase $N_M(0)$ most effectively in (vii) – (xi) to 387.

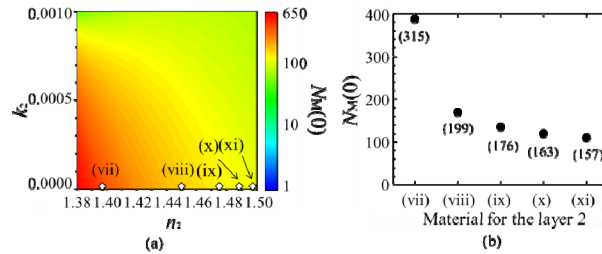


Fig. 5. (a) Calculation results of $N_M(0)$ as a function of n_2 and k_2 and for MgF_2 (vii), CaF (viii), SiO_2 (ix), PMMA (x), and BK7 (xi). The layer 1 is assumed to be TiO_2 . (b) Values of $N_M(0)$ and the corresponding optimized values of d_2 [nm] in parentheses.

As shown in Figs. 4 and 5, the combination of materials, which can yield the highest $N_M(0)$, are TiO_2 28 nm thick for the layer 1 and MgF_2 315 nm thick for the layer 2. Therefore, the distribution of $N(z)$ is calculated for the sensing chip composed of the 28-nm TiO_2 layer 1 and the 315-nm MgF_2 layer 2 as shown in Fig. 6. Here, the solid black vertical line drawn at $z = 0$ shows the interface between the layer 2 and the water. Furthermore, $z < -343$ (broken blue line), $-343 < z < -315$ (dotted orange line), $-315 < z < 0$, and $z > 0$ show the SiO_2 prism and the SiO_2 substrate, the TiO_2 layer, the MgF_2 layer, and the water, respectively. The distance δ from the surface of the chip to the position where light is totally reflected is calculated to be 239 nm from Eq. (6), which is shown by the broken black line.

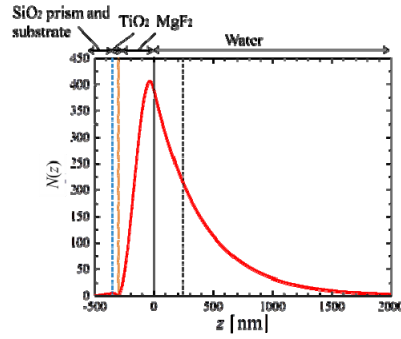


Fig. 6. Normalized electric field strength squared $N(z)$ in the WM sensing chip composed of the 28-nm TiO_2 layer and the 315-nm MgF_2 layer as a function of distance z from the interface between the MgF_2 layer and the water. The interfaces between the substrate and the TiO_2 layer, the TiO_2 layer and the MgF_2 layer, and the MgF_2 layer and the water are indicated by the broken blue line, the dotted orange line, and the solid black line, respectively. The position where the light is totally reflected is shown by the broken black line.

To evaluate the allowable ranges of the thicknesses of the TiO_2 and MgF_2 layers, $N(0)$ is shown in Fig. 7 as a function of thicknesses of the two layers. It is clear that $N(0)$ can be enhanced similarly even if the thickness of each layer varies by a few nm.

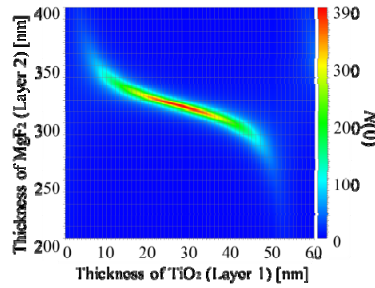


Fig. 7. $N(0)$, calculated as a function of thicknesses of the TiO_2 layer and the MgF_2 layer.

4. Comparison of the electric field enhancement effect with an Al SPR sensing chip

An SPR sensing chip is commonly used to obtain enhanced UV near-field light. In this chapter, the electric field enhancement effect is compared between the above-mentioned optimized $\text{TiO}_2/\text{MgF}_2$ WM sensing chip and an SPR sensing chip. It is well known that UV light is enhanced significantly by an Al SPR [15]. Therefore, we calculate the value of N at the interface between the Al layer of the SPR sensing chip and water, $N_{\text{sp}}(0)$, when p-polarized light at a wavelength λ of 375 nm illuminates the chip. Here, the prism and the substrate of the sensing chip are assumed to be SiO_2 . We use $n_{\text{Al}} = 0.347$ and $k_{\text{Al}} = 4.535$ as the values of the real and imaginary parts of the complex refractive index of Al, based on our measurement result obtained using a spectroscopic ellipsometer (VASE, J.A. Woollam). For calculations, thickness of Al, d_{Al} , is set to be in a range from 5 to 50 nm. Since the incident angle to the Al layer, θ_{sp} , which can excite SPR, depends on d_{Al} , θ_{sp} is optimized to maximize $N_{\text{sp}}(0)$.

Figure 8 shows calculation result of $N_{\text{spm}}(0)$, which is the maximum value of $N_{\text{sp}}(0)$ (solid red line, left axis), and θ_{sp} (broken blue line, right axis) as a function of d_{Al} . It is indicated that we can obtain the highest $N_{\text{spm}}(0)$ of 15.5 at θ_{sp} of 73.5° when d_{Al} is 20 nm. This value of $N_{\text{spm}}(0)$ is about 1/25 of the highest $N_{\text{M}}(0)$ of 387 obtained for the optimized $\text{TiO}_2/\text{MgF}_2$ WM sensing chip.

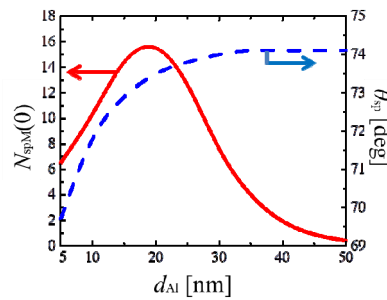


Fig. 8. Calculation results of $N_{\text{spm}}(0)$ (solid red) obtainable for the Al SPR sensing chip and the incident angle to the Al layer θ_{sp} (broken blue) as a function of d_{Al} .

There have been several papers that attempted to enhance the electric field strength of UV light by SPR. In many cases, Al nanoparticles [31,32] or Al nanostructured films [33] were used, but they required sophisticated skills for fabrication. In addition, they would not be suitable for near-field illumination since hot spots, where an enhanced electrical field is induced by the nanostructures, are limited.

The optimized WM sensing chip with appropriate thicknesses of TiO_2 and MgF_2 can enhance $N(0)$ by 387 times theoretically. This enhancement factor is 25 times that of the SPR sensing chip, which is the most important advantage of our WM sensing chip. In the WM sensing chip, the light is transmitted through the layers 1 and 2 after it was reflected at the interfaces between the substrate and the layer 1 and between the layers 1 and 2, if the phase matching condition is fulfilled. If the reflection occurs more, the electric field is enhanced more due to the near-field effect. This is the main reason for the high electric-field enhancement of our chip. Furthermore, since the chip is composed solely of inert inorganic dielectrics, quenching of fluorescence would not occur.

We have already reported that a WM sensing chip with layers of Si and SiO_2 can enhance near-field light in a visible region in a similar manner to what is expected by numerical calculations [16–21]. The present research reveals that the enhancement mechanism of our chip. Moreover, it reports numerically optimized structures and materials for our chip usable in an UV region. Experimental evidence supporting the calculations developed and the mechanism proposed in this paper will appear in the near future. Highly effective and stable UV near-field illumination would be realized, by applying the above-mentioned optimization to a sensing chip of a WM sensor.

5. Conclusions

The structure and materials of a WM sensing chip, which can effectively enhance the electric field strength in the UV region, was optimized for high-sensitivity detection of fluorescent-labeled materials. Based on calculation results using a transfer matrix method, we found that the WM sensing chip composed of a layer with a high refractive index and a low extinction coefficient and a layer with a low refractive index and a low extinction coefficient is the best. The thicknesses of the two waveguiding layers are set so that reflected lights have the same phase. The WM sensing chip composed of such optimized TiO_2 and MgF_2 layers can enhance the intensity of UV near-field light at a wavelength of 375 nm more efficiently than an Al SPR sensing chip. The optimized WM sensing chip would realize high-efficiency UV near-field illumination.

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