

Supporting Information

Quantitative Reflection Imaging of Fixed *Aplysia* *Californica* Pedal Ganglion Neurons on Nanostructured Plasmonic

*An-Phong Le[†], Somi Kang[‡], Lucas B. Thompson[†], Stanislav S. Rubakhin^{†,||}, Jonathan V.
Sweedler^{†,||}, John A. Rogers^{†,‡}, Ralph G. Nuzzo^{*,†,‡}*

[†] Department of Chemistry, University of Illinois at Urbana-Champaign, Urbana, Illinois 61801,
United States of America

[‡] Department of Materials Science and Engineering, University of Illinois at Urbana-Champaign,
Urbana, Illinois 61801, United States of America

^{||} Beckman Institute for Advanced Science and Technology, University of Illinois at Urbana-
Champaign, Urbana, Illinois 61801, United States of America

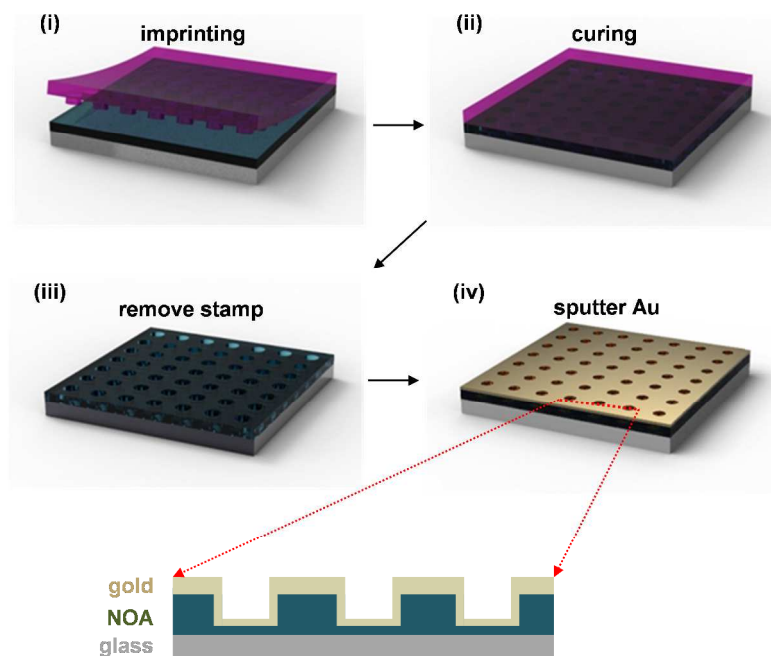


Figure S1. Schemes of the soft nanoimprint lithography process: (i) A nanostructured PDMS stamp is cast from a square nanohole array SOG master and used to imprint into an NOA prepolymer on a glass slide. (ii) The stamp is pressed into the NOA prepolymer and cured under ultraviolet light. (iii) The stamp is removed, leaving a replica of the square nanohole array from the original substrate. (iv) 32 nm of gold is sputtered onto the patterned NOA surface to create the plasmonic crystal.

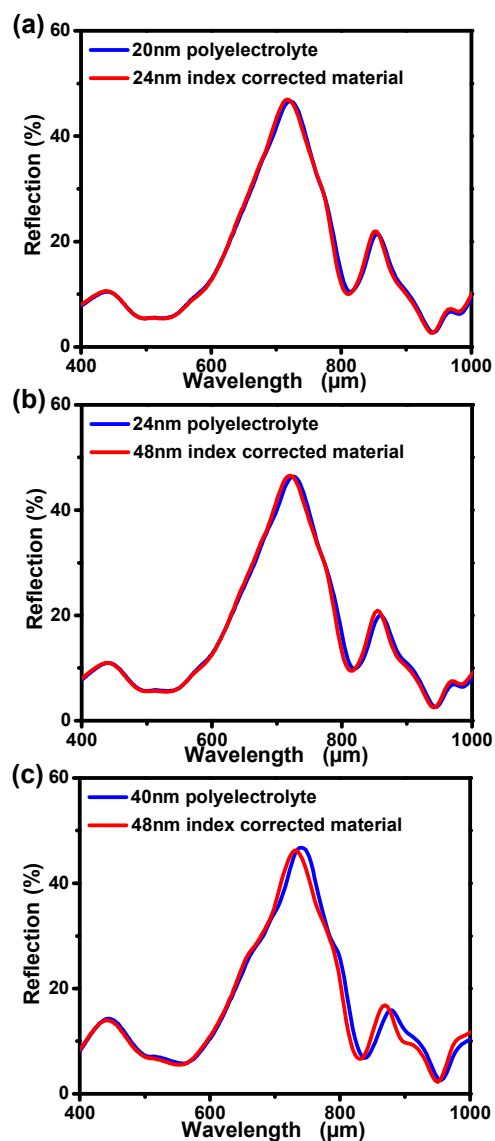


Figure S2. Finite-difference time-domain (FDTD) simulated reflection spectra for pairs of optically equivalent polyelectrolyte and index-corrected material for the fixed cell conformally covering a 30 nm of gold coated plasmonic crystal: (a) 20 nm polyelectrolyte and 24 nm index-corrected material; (b) 24 nm polyelectrolyte and 28 nm index-corrected material; (c) 40 nm polyelectrolyte and 48 nm index-corrected material.

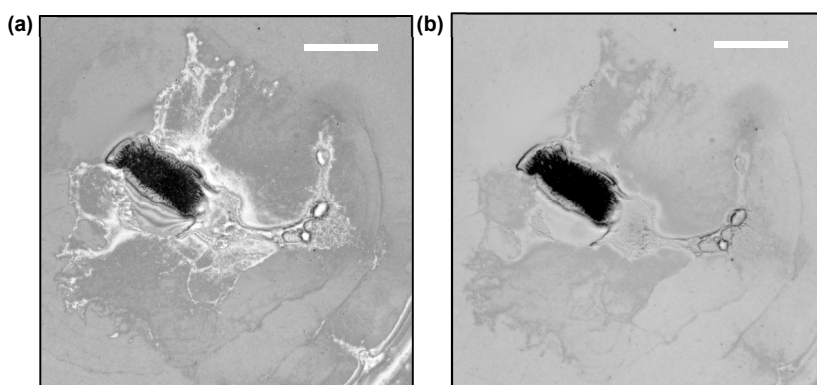


Figure S3. Reflection images of an *Aplysia* neuron cultured on a flat gold surface acquired using (a) 500-550 nm bandpass filter and (b) 525-1000 nm bandpass filter. The scale bar on each image corresponds to 100 μm . The rectangular shape of cell body on the figure represents the presence of some glia stack on the left top corner of the neuron surface.

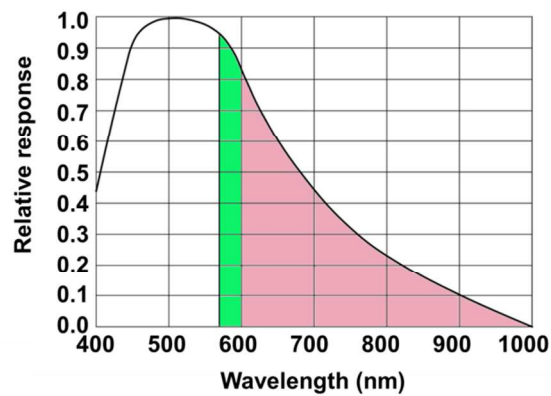


Figure S4. Spectral sensitivity of CCD camera over the 400-1000 nm wavelength range. The green region highlights the 570-600 nm wavelength range, while the green and pink regions combined represent the 570-1000 nm wavelength range. (Sony ICX085AL 2/3-inch Progressive Scan CCD Image Sensor with Square Pixel for B/W Cameras, Datasheet No. E95Z10C73; Sony Corp., <http://www.alldatasheet.com/datasheet-pdf/pdf/47409/SONY/ICX085AL.html>)

Quantitative Thickness Estimation Using Film Interference (Newton's Ring)

Thin film interference arises from the difference in path length from the reflection of light between two surfaces. Constructive interference of light rays reflected from both surfaces produces bright fringes, while dark fringes appear where destructive interference occurs.

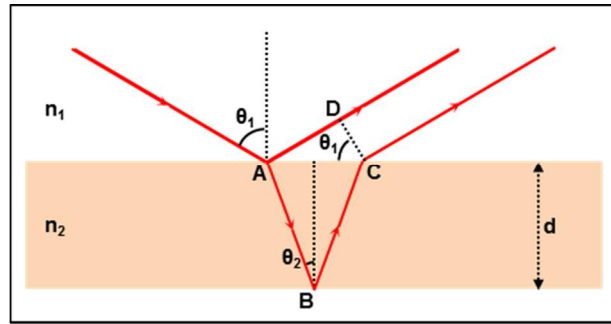


Figure S5. Ray diagram for thin film interference from light reflected from two adjacent surfaces.

Figure S5 illustrates the path length difference originating from reflections of light propagating through a medium with refractive index n_1 and reflecting from two surfaces – the top and bottom surfaces of a thin film with a thickness d and a refractive index n_2 . The optical path difference (OPD) is given by the expression below:

$$OPD = n_2 (\overline{AB} + \overline{BC}) - n_1 (\overline{AD})$$

where $\overline{AB} = \overline{BC} = \frac{d}{\cos(\theta_2)}$ and $\overline{AD} = 2n_2 d \tan \theta_2 \sin \theta_1$

The OPD equation can be simplified using Snell's law ($n_1 \sin \theta_1 = n_2 \sin \theta_2$) to

$$OPD = 2n_2 d \cos \theta_2$$

At the bright fringe, the optical path difference must be an integer multiple of the wavelength ($m\lambda$, where m is an integer).

$$m\lambda = 2n_2d \cos \theta_2$$

$$d = \frac{m\lambda}{2n_2 \cos \theta_2}$$

Figure 5b was acquired using a 500-550 nm bandpass filter and the incident angle of light (θ_1) ranges from 0° to 23.6° due to the numerical aperture of the objective lens (NA = 0.40).

Therefore, the thickness of the cellular film ($n_2 = 1.50$) at the bright fringes can be roughly estimated:

		$\lambda = 500 \text{ nm}$		$\lambda = 550 \text{ nm}$	
		0°	23.6°	0°	23.6°
Thickness of protein	m=1	167 nm	173 nm	183 nm	190 nm
	m=2	333 nm	346 nm	366 nm	380 nm
	m=3	500 nm	519 nm	549 nm	571 nm