Electronic Supplementary Information

Surface modification of polyvinyl alcohol sponge with functionalized boronic acid to develop porous materials for multicolor emission, chemical sensing and 3D cell culture

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Materials and methods

Unless otherwise described, reagents and solvents used for this study were commercially available and used as supplied. Polyvinyl alcohol (**PVA**, M_W = 89 000–99 000, >99% hydrolyzed) was purchased from Aldrich. Compounds **1a**, **1b**, **2a**, **2b**, **3a**, **3b** and **4** were synthesized in our previous reports.[1–3] The human cervical cancer (HeLa) cells were obtained from RIKEN Cell Bank. Glass-bottom dishes and 96-well plates were purchased from FPI and Thermo Scientific, respectively. All metal ions (Na^+, K^+, Mg^{2+}, Ca^{2+}, Fe^{3+}, Co^{2+}, Ni^{2+}, Cu^{2+}, Cd^{2+}, Hg^{2+}, Al^{3+}, and Pb^{2+} ion) were used as perchlorate salts. Aqueous solutions buffered to pH 7.0 were prepared using 4-(2- hydroxyethyl)-1-piperazine ethanesulfonic acid (HEPES, 5 mM). HeLa cells were cultured in Dulbecco's modified Eagle's medium (DMEM) supplemented with 10% fetal bovine serum (FBS, heat inactivated at 56 °C before use) and 1% penicillin/streptomycin/amphotericin B at 37 °C in a humidified atmosphere with 5% CO_2.

Field-emission scanning electron microscopy (FE-SEM) was conducted by a JEOL JSM-7500F (acceleration voltage: 5 kV). For FE-SEM measurements, specimens were coated with osmium by a Meiwafosis Neoc-Pro osmium coater. Solid-state ^{13}C cross-polarization magic-angle-spinning nuclear magnetic resonance (^{13}C CP/MAS NMR, 100 MHz) spectra were measured by a JEOL ECA 400 spectrometer. The spectrometer is equipped with a 3.2 mm MAS probehead capable of producing an MAS speed of 9 kHz and spectra were obtained using a ^1H–^{13}C CP with a contact time of 2 ms, an acquisition time of 50.9 ms, a relaxation delay of 5 s between scans. The chemical shifts were calibrated using adamantine (δ = 29.5 ppm) as an external standard relative to tetramethylsilane (TMS, δ = 0 ppm). Fourier transform infrared (FT-IR) spectra were recorded on a JASCO FT/IR-4100 spectrometer equipped with a diamond ATR crystal (JASCO PRO 450S). Digital photographs of samples were taken with a PENTAX *K-m* digital camera. Confocal laser scanning microscopy (CLSM) was conducted with a Nikon A1 confocal microscope system. UV-vis absorption spectra were recorded on a Shimadzu UV-3600 spectrophotometer and a 10 mm × 10 mm quart cell was used. Fluorescence spectra and quantum yields of luminescent sponges were obtained using a JASCO FP-8500 spectrofluorometer equipped with a substandard light source (JASCO ESC-842) and an integrating sphere (JASCO IL835). X-ray photoelectron spectra (XPS) were acquired using a JEOL JPS-9010MX. Fluorescence decay profiles were recorded by a fluorescence lifetime spectrometer (HAMAMATSU Quantaurus–Tau C11367). NMR spectra of solution samples were recorded on a Bruker AVANCE-500 spectrometer. TMS and 3-(trimethylsilyl)propionic-2,2,3,3-d_4 acid sodium salt were used as internal standards (δ = 0 ppm). All NMR spectra were recorded at 298 K. High-resolution mass spectra (HRMS) were measured by a Bruker micrOTOF mass spectrometer in an electrospray ionization (ESI) mode and Tuning-Mix was used as a reference.

Preparation of PVA sponge

Starting sponges were prepared by cross-linking of **PVA** with formaldehyde according to a reported literature procedure.[4,5] **PVA** (6.0 g) was added to water (54 mL) in a 300 mL beaker and the suspension was allowed to stand at 95 °C until complete dissolution using an oil bath. After complete dissolution, the beaker was removed from the oil bath. To the hot **PVA** solution were added aqueous formaldehyde solution (36-38%, 10 mL) and Triton X-100 (1.5 g). The resulting mixture was stirred (1000 rpm) for 25 min using a stirring bar to form a foam solution, then 50 wt% sulfuric acid solution (30 mL) was added to the solution over 2 min. The resulting foam solution was kept in an oven at 60 °C for 5 h. The obtained sponge was thoroughly washed with water and dried in an oven at 60 °C. The porosity of the sponge was determined to be 86 ± 12% by a water immersion method. ^{13}C CP MAS NMR (100 MHz): δ/ppm 40.0, 44.7, 68.2, 73.2, 87.5, 94.1. FT-IR (ATR): ν_{max}/cm^{-1} 1009sv, 1065s, 1129s, 1173s, 1241w, 1363w, 1392w, 1405w, 1432w, 1635w, 2786w, 2863w, 2914w, 2942w, 3478br.

Chemical modification of PVA sponges with boronic acids

Chemical modification of **PVA** sponges with boronic acid-appended fluorescent dyes was conducted by a soaking technique as follows: a piece of sponge (10.0 mg, 8 mm × 8 mm × 2 mm) was put in a 50 mL vial and a methanol solution (25 mL) of **1a** (2.0×10^{-5} M) was added. The sample was shaken at room temperature for 15 h by using a shaking apparatus. The resulting sponge was rinsed with methanol. For X-ray photoelectron spectroscopy (XPS) and attenuated total reflection Fourier transform infrared spectroscopy (ATR-FT-IR), methanol solutions of **1a** (1.5×10^{-4} M) were used. The adsorbed amounts of **1a**, **2a**, and **3a** were 11.4 mg/g, 1.9 mg/g and 15.6 mg/g, respectively. From these data and the amounts of the reactive diol moieties in **PVA** sponges calculated by Langmuir isotherm for **1a**, the dye-loading ratios of **PVA/1a**, **PVA/2a**, and **PVA/3a** against the reactive diol moieties in whole **PVA** sponges were also determined to be 10.0%, 1.6% and 15.8%, respectively.

Quantitative evaluation of the adsorption behavior of 1a

PVA sponges (4.0 mg, 6 mm × 5 mm × 2 mm) were immersed in methanol solutions of **1a** at various concentrations (0–3.2×10^{-4} M, 10 mL), respectively. The resulting samples were shaken at room temperature for 15 h using a shaking apparatus. Equilibrium concentrations of **1a** were determined from absorption intensities at 498 nm of the resulting solutions and the amounts of **1a** adsorbed were calculated from the equilibrium concentrations. The obtained adsorption isotherm was analyzed with an equation for a 1:1 stoichiometric binding mode between **1a** and the diol moieties of the **PVA** sponge. According to this assumption, the amount of **1a** adsorbed (q) can be expressed in the following equations:

$$q = \frac{q_{max} \times (K \times [\mathrm{dye}]_0 + K \times [\mathrm{diol}]_0 + 1 - \sqrt{(K \times [\mathrm{dye}]_0 + K \times [\mathrm{diol}]_0 + 1)^2 - 4 \times K^2 \times [\mathrm{diol}]_0 \times [\mathrm{dye}]_0})}{2 \times K \times [\mathrm{diol}]_0}$$

In the equations, q_{max}, K and $[dye]_0$ are the maximum adsorption capacity, the association constant and the initial concentration of **1a**, respectively. The value of $[diol]_0$ is defined as the initial apparent concentration of the diol moiety at the pore surface of the **PVA** sponge. The values of q_{max}, K and $[diol]_0$ were evaluated by a non-linear least-squares method.

Emission color tuning of PVA sponges

The emission color of the **PVA** sponge was tuned by varying the mole fractions of **1a**, **2a**, and **3a** with a total concentration of 2.0×10^{-5} M. Typically, to a methanol solution (25 mL) containing **1a** (1.6×10^{-6} M) and **3a** (18.4×10^{-6} M) in a 50 mL vial was added a **PVA** sponge (10 mg, 8 mm × 8 mm × 2 mm). The sample was shaken at room temperature for 2 h using a shaking apparatus. The resulting sponge was rinsed with methanol to afford a cyan-light-emissive **PVA** sponge (**PVA-C**). In a similar manner, **PVA-M**, **PVA-Y**, and **PVA-W** sponges exhibiting magenta-, yellow-, and white-light emission, respectively, were prepared using methanol solutions of dyes at the following concentrations: **PVA-M**: [**2a**] = 16.0×10^{-6} M and [**3a**] = 4.0×10^{-6} M, **PVA-Y**: [**1a**] = 19.2×10^{-6} M, [**2a**] = 0.8×10^{-6} M, **PVA-W**: [**1a**] = 2.4×10^{-6} M, [**2a**] = 1.6×10^{-6} M, [**3a**] = 16.0×10^{-6} M.

PVA sponges capable of the fluorescence detection of metal ions

A piece of **PVA** sponge (10.0 mg, 8 mm × 8 mm × 2 mm) was immersed in a methanol solution (25 mL) of **4** (2.0×10^{-5} M). The sample was shaken at room temperature for 2 h using a shaking apparatus and the resulting sponge was rinsed with methanol to afford a colorless sponge. The amount of **4** (λ_{max} = 307 nm and $\varepsilon = 6.3 \times 10^3$ M^{-1} cm^{-1} in methanol) adsorbed to the **PVA** sponge was determine to be 1.6×10^{-7} mol by measuring the absorption intensity at 307 nm of the resulting soaking solution. **PVA** sponges functionalized with **4** in the same manner (**PVA/4**) were immersed in aqueous solutions (5 mM HEPES buffer, pH = 7.0, 10 mL) of Na^+, K^+, Mg^{2+}, Ca^{2+}, Fe^{3+}, Co^{2+}, Ni^{2+}, Zn^{2+}, Cu^{2+}, Cd^{2+}, Hg^{2+}, Al^{3+} and Pb^{2+} ions ($[M^{n+}] = 3.0 \times 10^{-5}$ M), respectively. The resulting samples were shaken at room temperature for 30 min using a shaking apparatus and then fluorescence spectra of the resulting sponges were measured.

Synthesis of a boronic acid-appended lysine derivative

N^{α}, N^{ε}-Bis(*tert*-butoxycarbonyl)-L-lysine (0.76 g, 2.2 mmol) was dissolved in dry DMF (20 mL) under a nitrogen atmosphere at room temperature. To this solution was added 3-aminophenylboronic acid (0.27 g, 2.2 mmol), *O*-(benzotriazol-1-yl)-*N*, *N*, *N*', *N*'-tetramethyluronium hexafluorophosphate (HBTU) (0.91 g, 2.4 mmol) and *N*, *N*-diisopropylethylamine (DIPEA) (0.52 mL, 3.0 mmol) under a nitrogen atmosphere and the mixture was stirred at room temperature for 18 h. The reaction mixture was poured into a saturated aqueous ammonium chloride solution (100 mL). The resulting precipitate was collected and dried under vacuum. The crude product was chromatographed on silica gel

(wako gel C-300) using 2% methanol in dichloromethane as an eluent. In this way, 0.69 g of **6** was obtained as a precursor compound (70% yield). ^{1}H NMR (500 MHz, DMSO-d_6): δ/ppm 1.25-1.38 (m, 22H), 1.60 (m, 2H), 2.89 (m, 2H), 4.02 (m, 1H), 6.76 (t, J = 5.5 Hz, 1H), 6.92 (d, J = 8.2 Hz, 1H), 7.26 (d, J = 7.7 Hz, 1H), 7.47 (d, J = 7.3 Hz, 1H), 7.71 (d, J = 8.0 Hz, 1H), 7.83 (s, 1H), 8.00 (s, 2H), 9.82 (s, 1H). ^{13}C NMR (125 MHz, DMSO-d_6): δ/ppm 22.1, 28.22, 28.25, 29.3, 31.0, 55.0, 77.3, 78.0, 121.2, 125.2, 127.6, 129.0, 134.8, 138.2, 155.5, 155.6, 171.3. HRMS (ESI): m/z [**6** + Na]$^+$ calcd for $C_{22}H_{36}BN_3NaO_7$, 488.2543; found, 488.2534.

Compound **6** (0.27 g, 0.60 mmol) was dissolved in dry dichloromethane (5 mL) and the resulting solution was cooled using an ice bath. To this solution was added 0.5 mL of trifluoroacetic acid (TFA). The mixture was stirred for 18 h at room temperature. The resulting solution was concentrated under vacuum and co-evaporated several times with toluene to give a trifluoroacetic acid salt of **5** (0.28g) as a white solid (98% yield). ^{1}H NMR (500 MHz, D_2O): δ/ppm 1.54 (m, 2H), 1.74 (m, 2H), 2.03 (m, 2H), 3.01 (t, J = 7.7 Hz, 2H), 4.14 (t, J = 6.6 Hz, 1H), 7.49 (dd, J = 7.7, 7.7 Hz, 1H), 7.59 (ddd, J = 8.0, 2,3, 1.2 Hz, 1H), 7.64 (ddd, J = 7.6, 1.1, 1.1 Hz, 1H), 7.78 (dd, J = 1.4, 1.4 Hz, 1H). ^{13}C NMR (125 MHz, D_2O): δ/ppm 24.0, 29.1, 33.3, 41.7, 51.6, 56.3, 119.1 (q, $^{1}J_{CF}$ = 290 Hz), 126.9, 129.4, 131.7, 133.9, 136.3, 138.3, 165.7 (q, $^{2}J_{CF}$ = 35 Hz), 171.2. HRMS (ESI): m/z [**5** + H]$^+$ calcd for $C_{12}H_{21}BN_3O_3$, 266.1673; found, 266.1677.

Cell culture experiments

A piece of **PVA** sponge (10.0 mg, 8 mm × 8 mm × 2 mm) was immersed in a methanol solution (25 mL) containing **1a** (2.0×10^{-5} M) and a trifluoroacetate salt of boronic acid-appended lysine (**5**, 2.0×10^{-4} M) at room temperature. After shaking the sample for 2 h using a shaking apparatus, the sponge was taken from the solution and carefully rinsed with methanol, ethanol and water. **PVA** sponges functionalized with **1a** and a trifluoroacetate salt of **5** in the same manner were placed on a 96-well plate. A 200 μL portion of the cell suspension (5×10^5 cells/mL in phenol red-free D-MEM) was added to each well and the plate was incubated for one, three, and seven days at 37 °C with 5% CO_2. After the incubation, the sponges were rinsed with PBS and incubated with 1 μM Hoechst for 30 min. After washing the sponges with PBS, the sponges were placed on 14 mm glass-bottom dishes with phenol red-free medium.

References

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2. R. Nishiyabu, Y. Sugino and Y. Kubo, *Chem. Commun.*, 2013, **49**, 9869.
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4. H. G. Hammond, U. S. Patent, US 2668253, 1953.
5. Y. Pan, W. Wang, C. Peng, K. Shi, Y. Luo and X. Ji, *RSC Adv.*, 2014, **4**, 660.

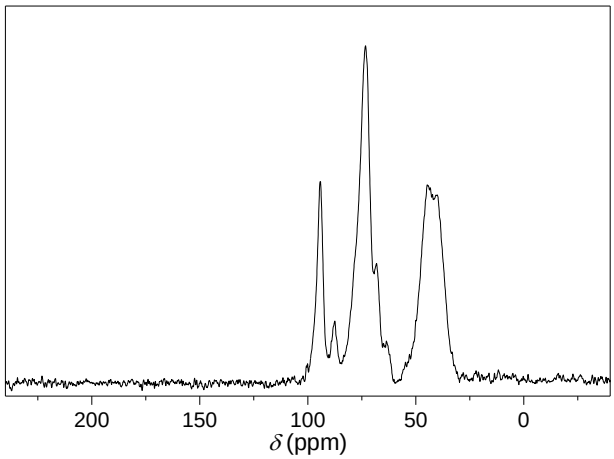


Fig. S1 Solid state ^{13}C CP MAS NMR spectrum of a formaldehyde cross-linked **PVA** sponge.

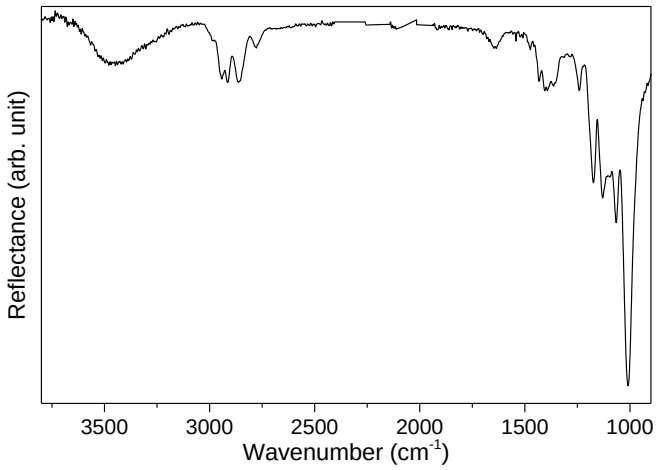
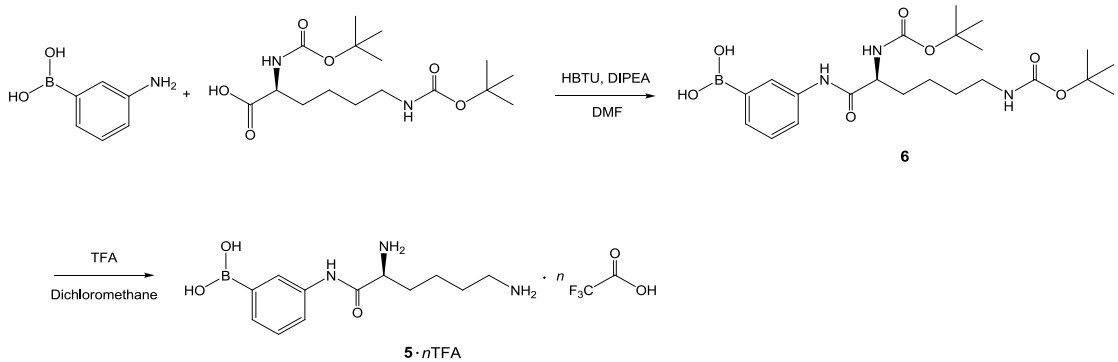


Fig. S2 FT-IR (ATR) spectrum of a formaldehyde cross-linked **PVA** sponge.



Scheme S1 Synthetic route to a boronic acid-appended L-lysine derivative.

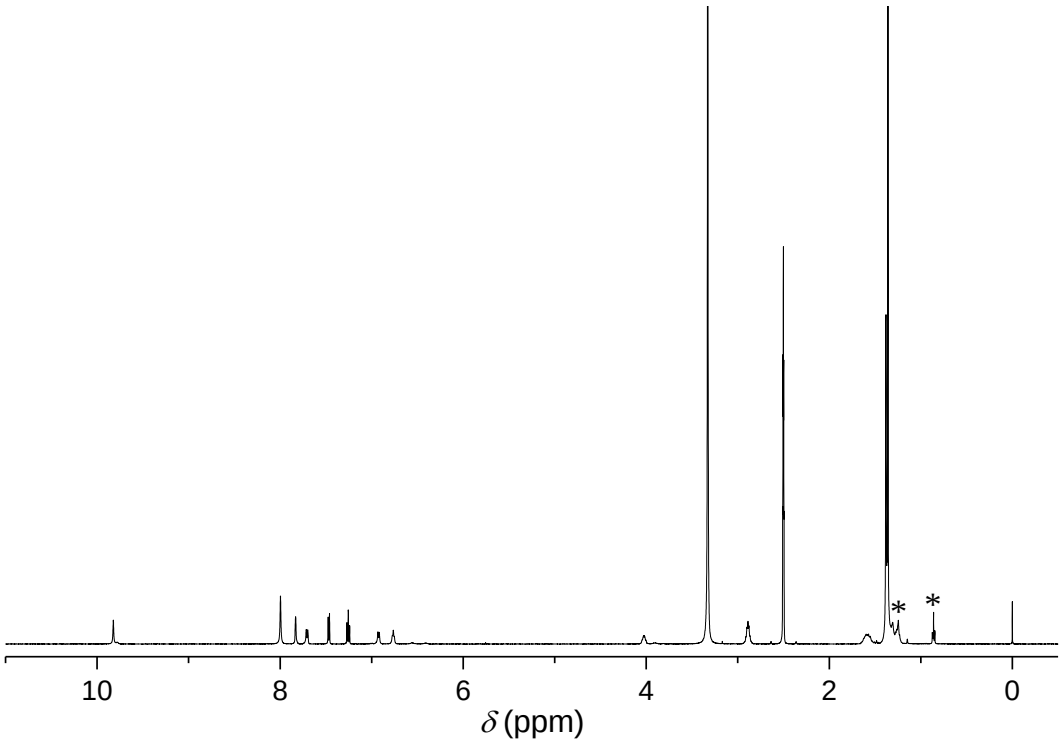


Fig. S3 ^{1}H NMR spectrum of **6** in DMSO-d_6. *Residual proton signals of solvent (*n*-hexane).

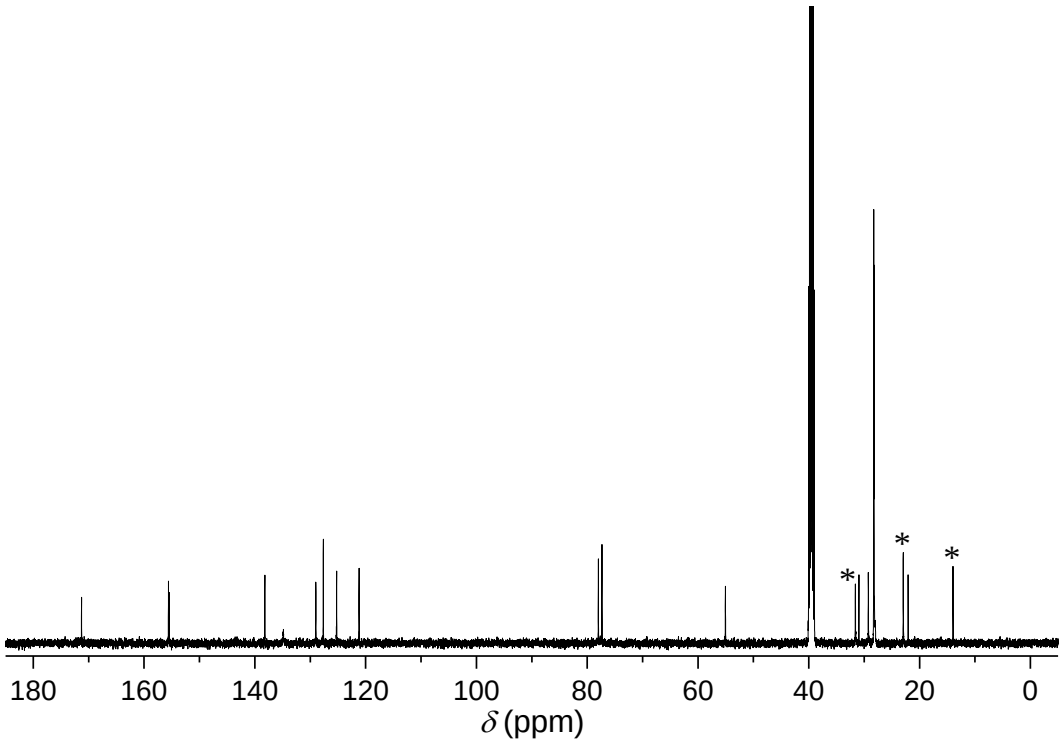


Fig. S4 ^{13}C NMR spectrum of **6** in DMSO-d_6. *Residual proton signals of solvent (*n*-hexane).

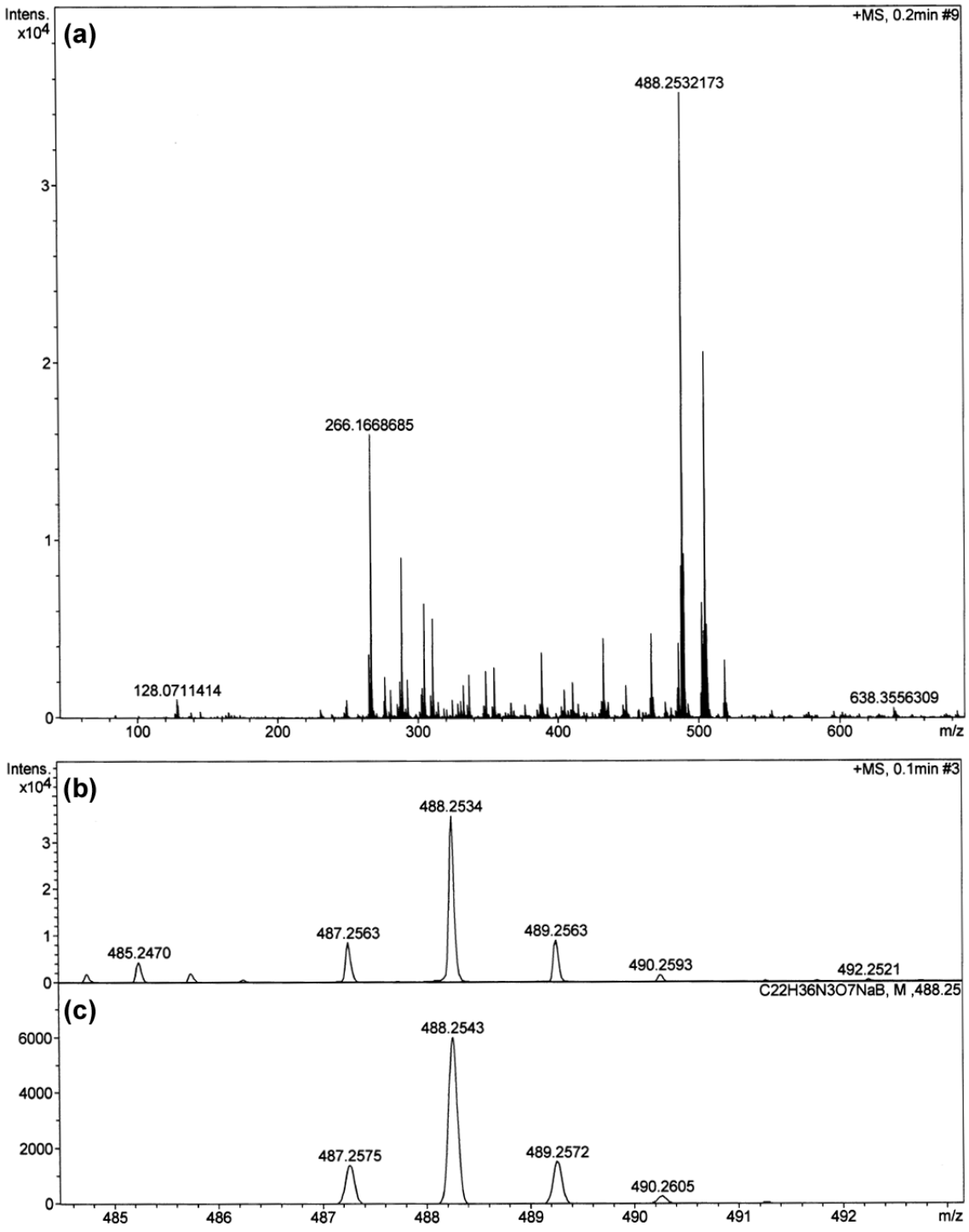


Fig. S5 (a) HRMS (ESI) of **6**. (b) Observed and (c) simulated isotope patterns of [**6** + Na]$^+$.

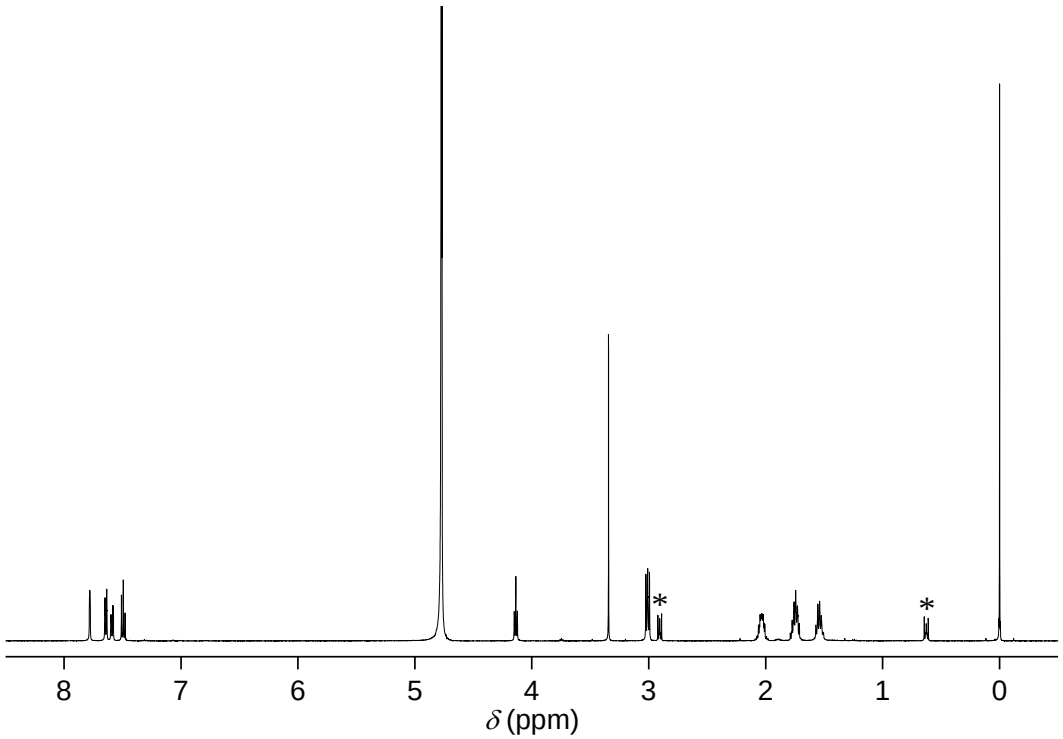


Fig. S6 ^{1}H NMR spectrum of trifluoroacetic acid salt of **5** in D_2O. *Residual proton signals of 3-(trimethylsilyl)propionic-2,2,3,3-d_4 acid sodium salt used as an internal standard.

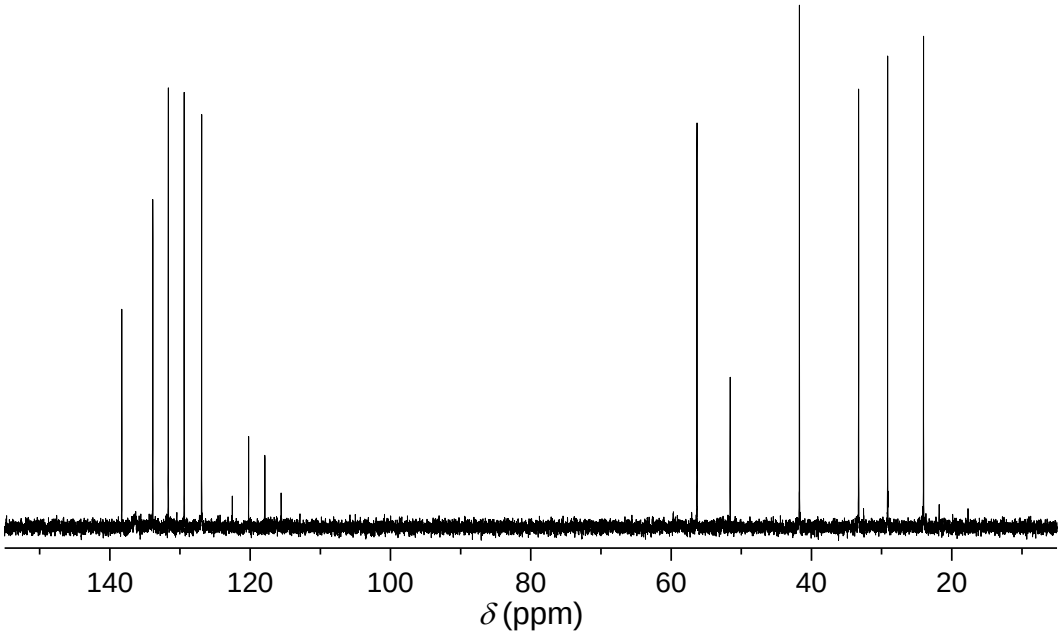


Fig. S7 ^{13}C NMR spectrum of trifluoroacetic acid salt of **5** in D_2O.

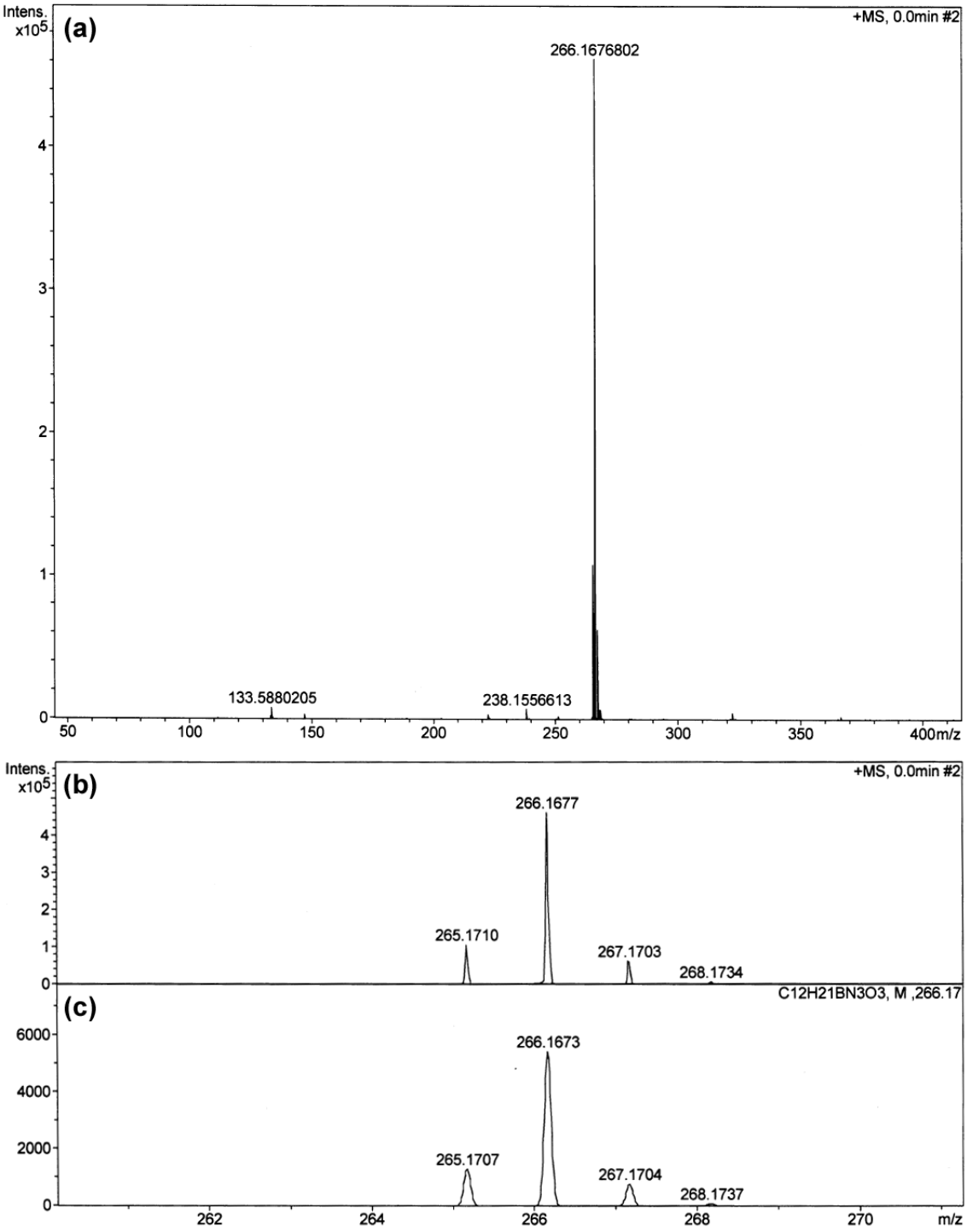


Fig. S8 (a) HRMS (ESI) of **5**. (b) The observed and (c) simulated isotope patterns of [**5** + H]$^+$.

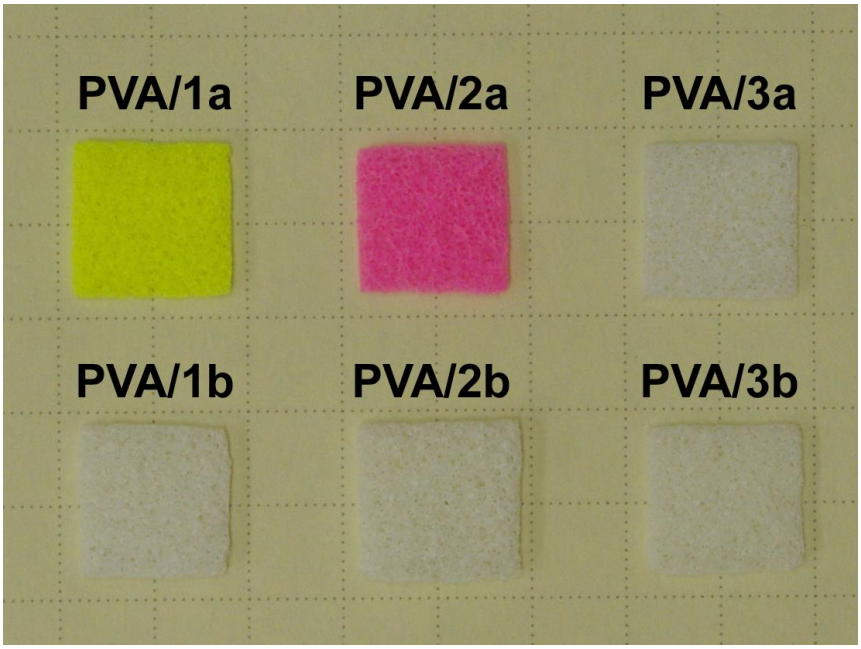


Fig. S9 Photograph of **PVA** sponges after the treatment with **1a** (**PVA/1a**), **2a** (**PVA/2a**), **3a** (**PVA/3a**), **1b** (**PVA/1b**), **2b** (**PVA/2b**) and **3b** (**PVA/3b**). The photograph was taken under ambient light.

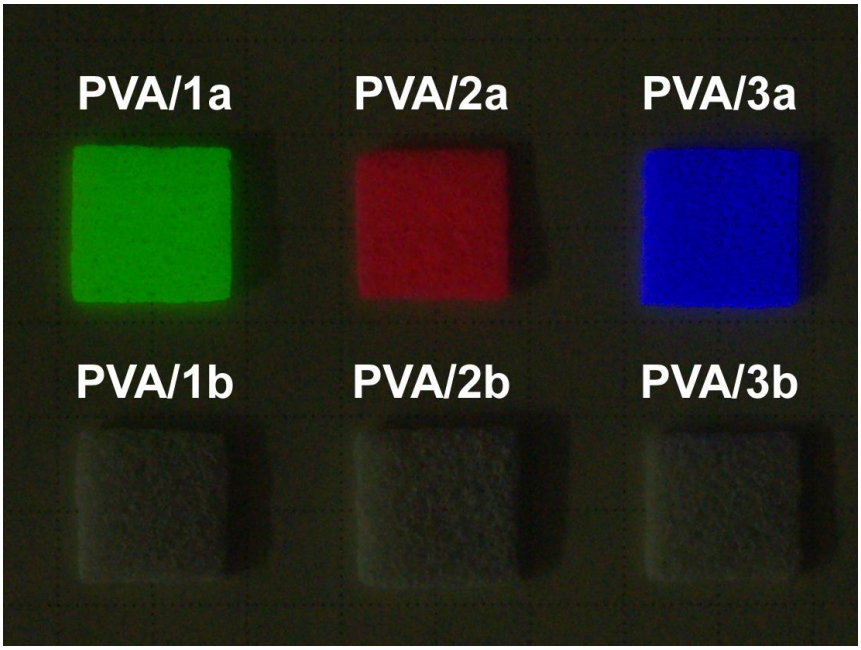


Fig. S10 Photograph of **PVA** sponges after the treatment with **1a** (**PVA/1a**), **2a** (**PVA/2a**), **3a** (**PVA/3a**), **1b** (**PVA/1b**), **2b** (**PVA/2b**) and **3b** (**PVA/3b**). The photograph was taken under UV light. λ_{ex} = 365 nm.

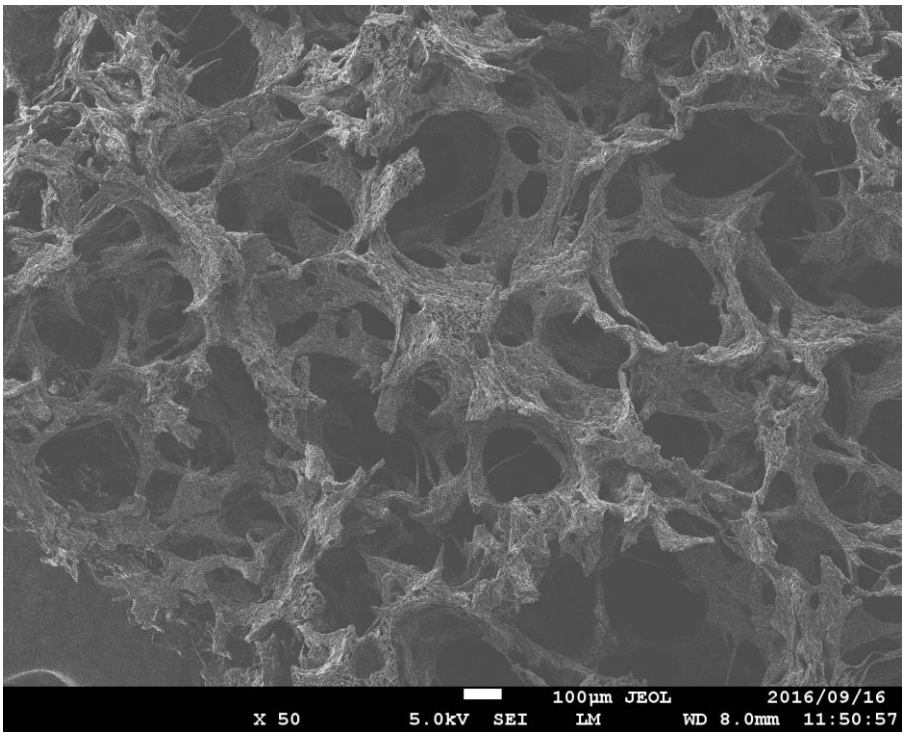


Fig. S11 FE-SEM image of **PVA/1a**.

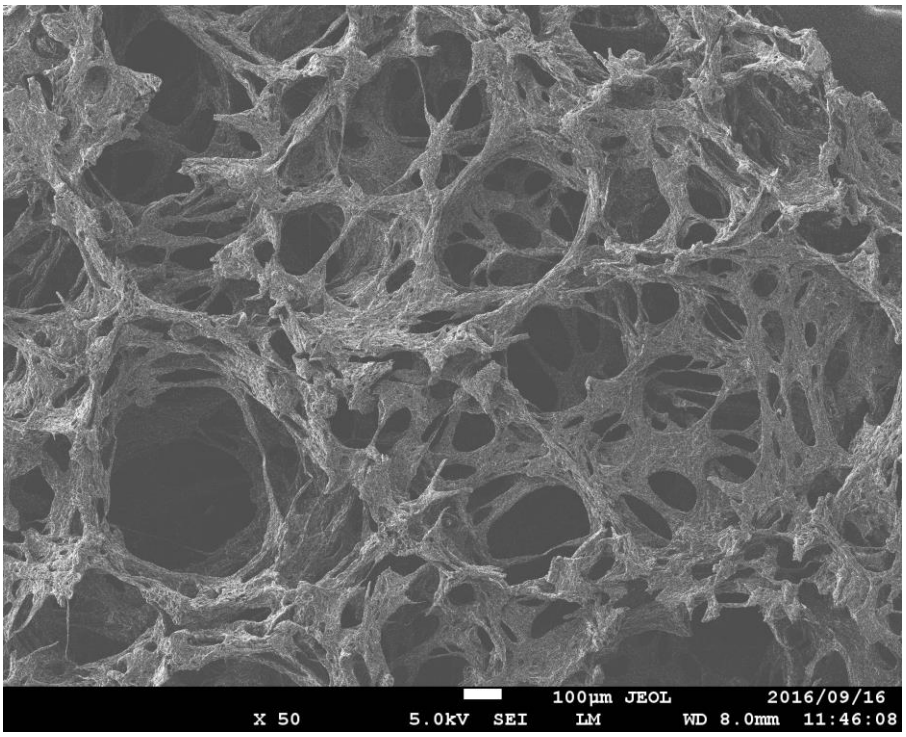


Fig. S12 FE-SEM image of **PVA/2a**.

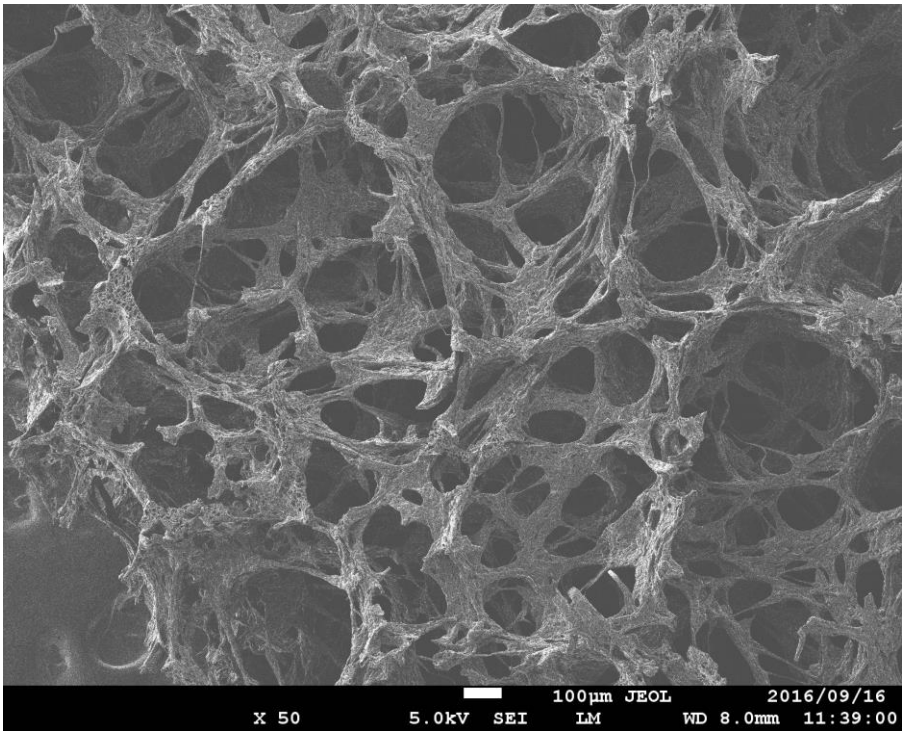


Fig. S13 FE-SEM image of **PVA/3a**.

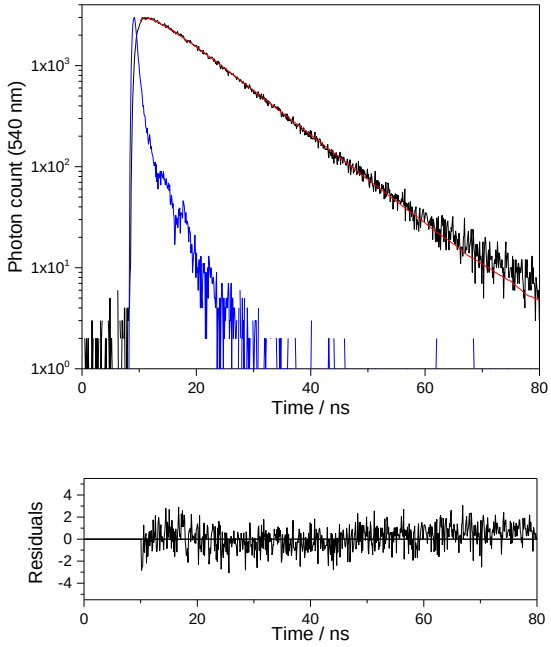


Fig. S14 Fluorescence decay profile (black) of **PVA/1a** (λ_{em} = 540 nm). The least-squares fit to a single exponential (red line) with a fluorescence lifetime of 9.7 ns. The decay curve (blue) represents the instrument response function. The bottom panel shows the weighted residuals of the fitted curve.

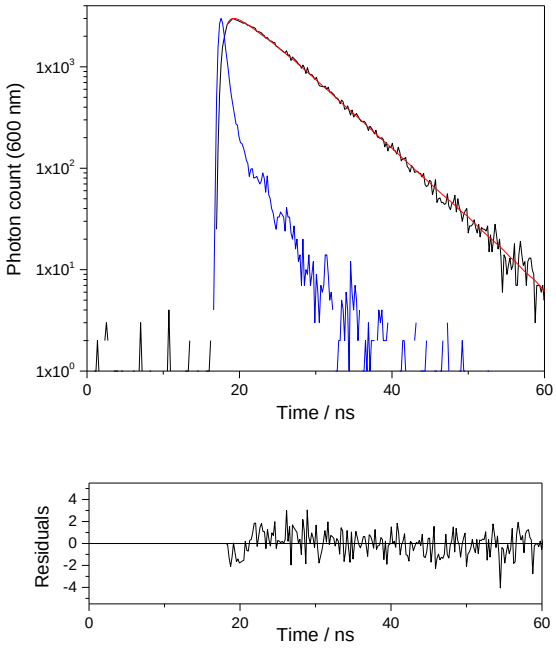


Fig. S15 Fluorescence decay profile (black) of **PVA/2a** (λ_{em} = 600 nm). The least-squares fit to a single exponential (red line) with a fluorescence lifetime of 6.0 ns. The decay curve (blue) represents the instrument response function. The bottom panel shows the weighted residuals of the fitted curve.

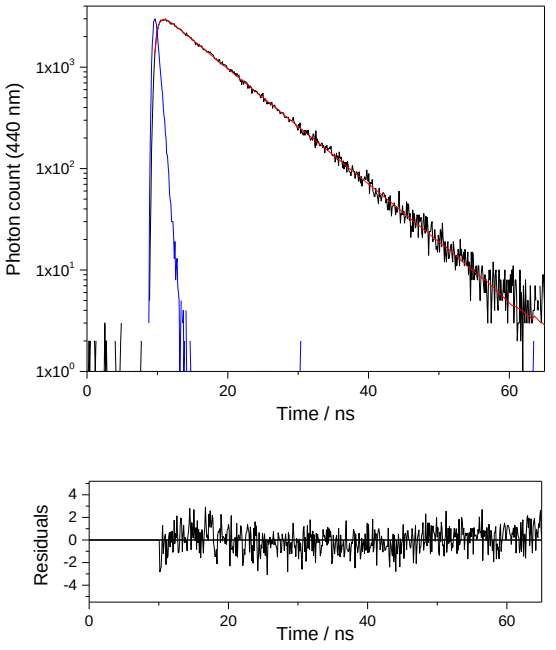


Fig. S16 Fluorescence decay profile (black) of **PVA/3a** (λ_{em} = 440 nm). The least-squares fit to a single exponential (red line) with a fluorescence lifetime of 7.6 ns. The decay curve (blue) represents the instrument response function. The bottom panel shows the weighted residuals of the fitted curve.

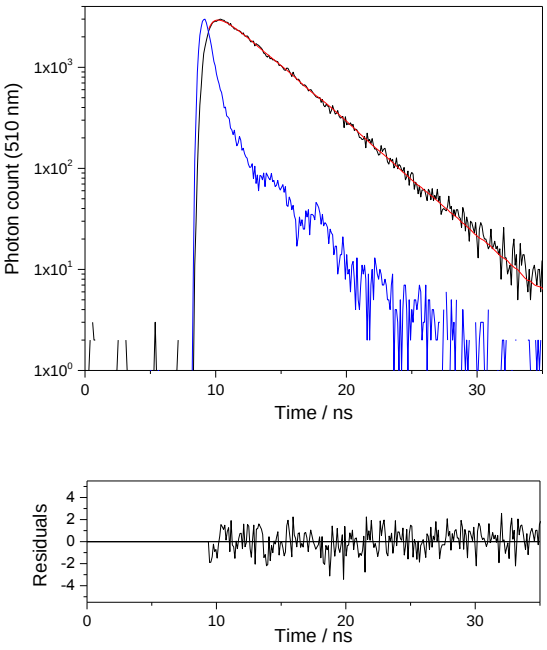


Fig. S17 Fluorescence decay profile (black) of **1a** (λ_{em} = 510 nm). The least-squares fit to a single exponential (red line) with a fluorescence lifetime of 3.1 ns. The decay curve (blue) represents the instrument response function. The bottom panel shows the weighted residuals of the fitted curve.

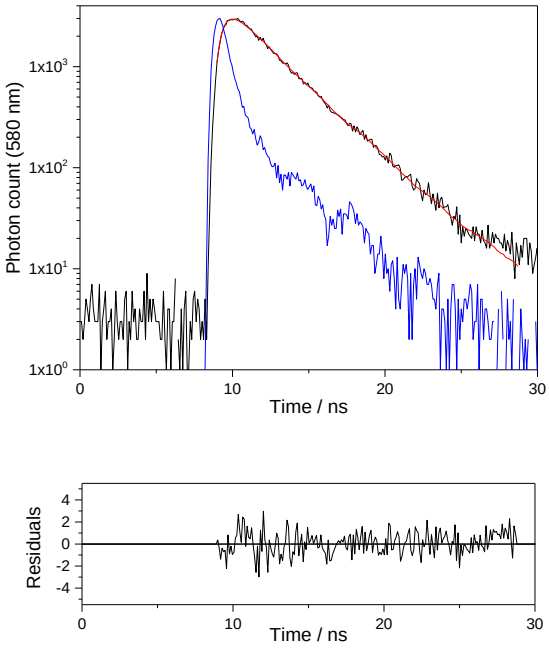


Fig. S18 Fluorescence decay profile (black) of **2a** (λ_{em} = 580 nm). The least-squares fit to a single exponential (red line) with a fluorescence lifetime of 2.2 ns. The decay curve (blue) represents the instrument response function. The bottom panel shows the weighted residuals of the fitted curve.

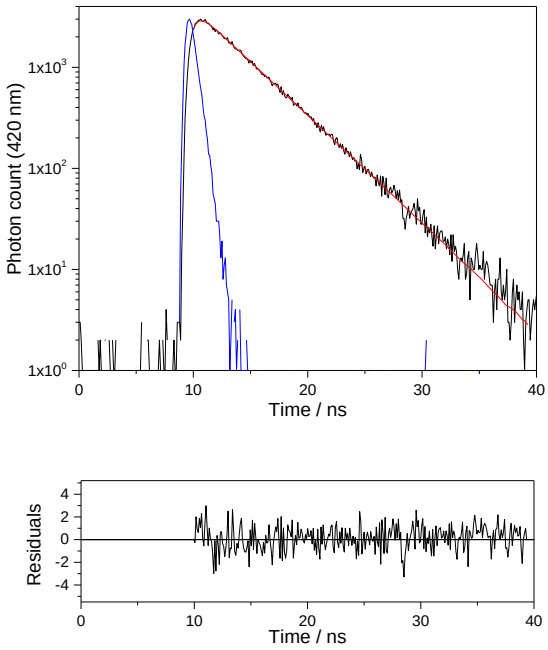


Fig. S19 Fluorescence decay profile (black) of **3a** (λ_{em} = 420 nm). The least-squares fit to a single exponential (red line) with a fluorescence lifetime of 4.1 ns. The decay curve (blue) represents the instrument response function. The bottom panel shows the weighted residuals of the fitted curve.

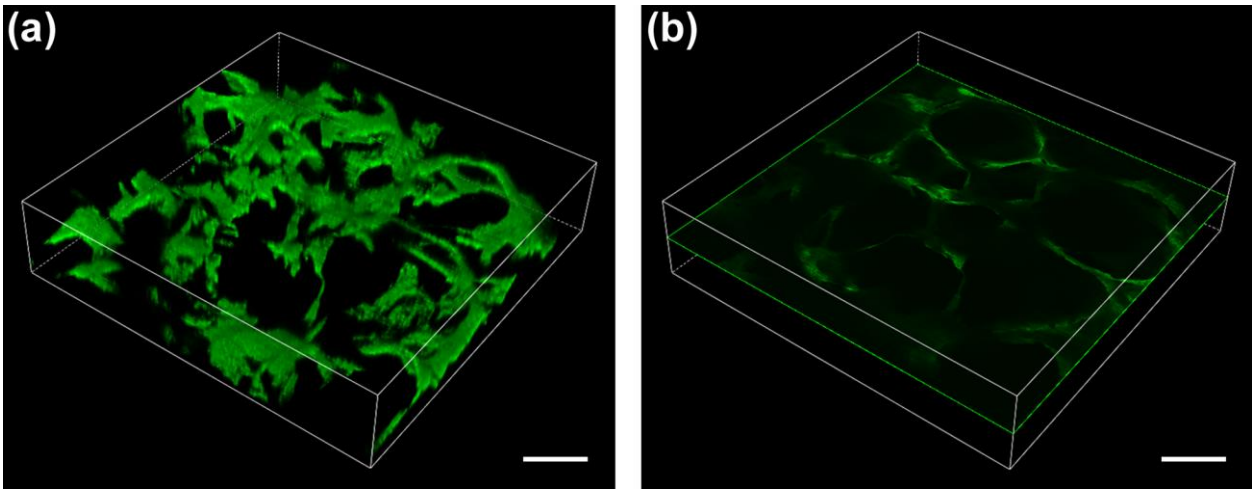


Fig. S20 (a) Three dimensional reconstructed fluorescence image of **PVA/1a** generated from the obtained CLSM images and (b) a cross-sectional image at a distance of 136 μm from the top image (λ_{ex} = 488 nm, λ_{em} = 500-550 nm). Scale bar: 200 μm.

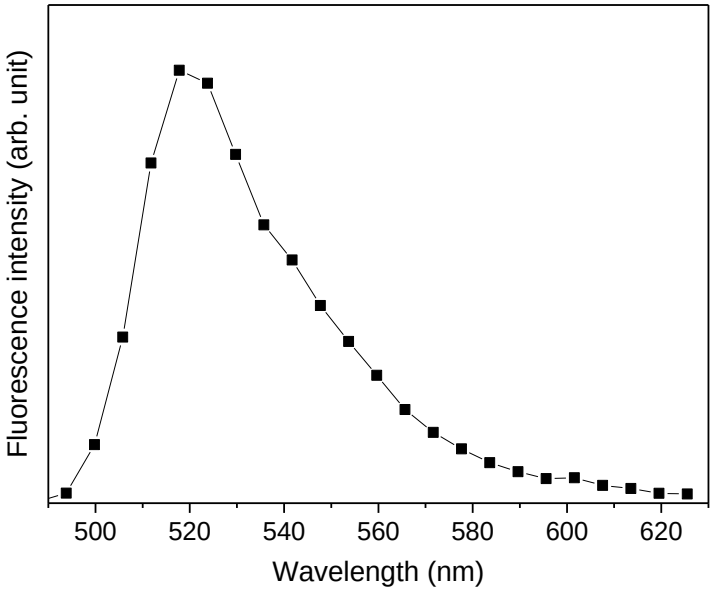


Fig. S21 Fluorescence spectrum of **PVA**/**1a** recorded by confocal laser scanning microscopy (λ_{ex} = 488 nm).

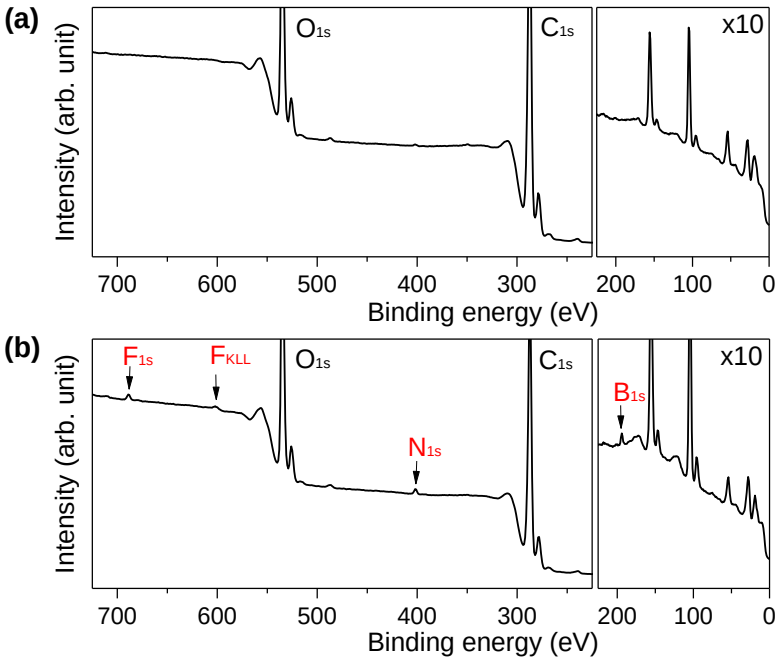


Fig. S22 XPS spectra of (a) a **PVA** sponge and (b) a **PVA** sponge functionalized with **1a**. Characteristic peaks of F_{1s} (686 eV), F_{KLL} (599 eV), N_{1s} (399 eV) and B_{1s} (191 eV) were detected in addition to peaks of C_{1s} (284 eV) and O_{1s} (532 eV) for the functionalized **PVA** sponge, while only peaks of C_{1s} (284 eV) and O_{1s} (532 eV) were detected in the parent **PVA** sponge.

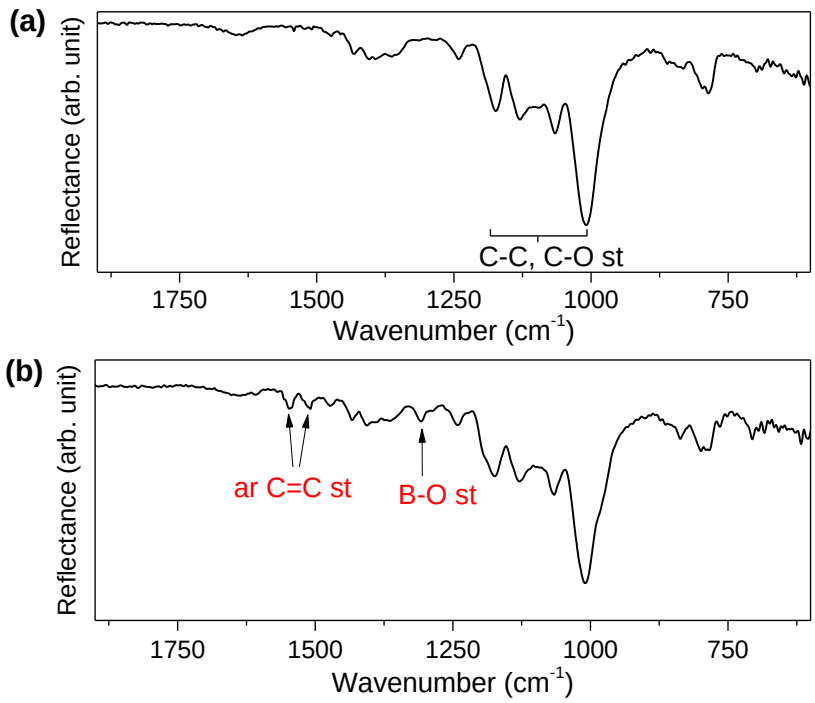


Fig. S23 ATR-FT-IR spectra of (a) a **PVA** sponge and (b) a **PVA** sponge functionalized with **1a**. Characteristic peaks assignable to aromatic C=C stretching at 1547 and 1509 cm^{-1}, and B−O stretching at 1307 cm^{-1} were observed in the functionalized **PVA** sponge in addition to the peaks of parent **PVA** sponge.

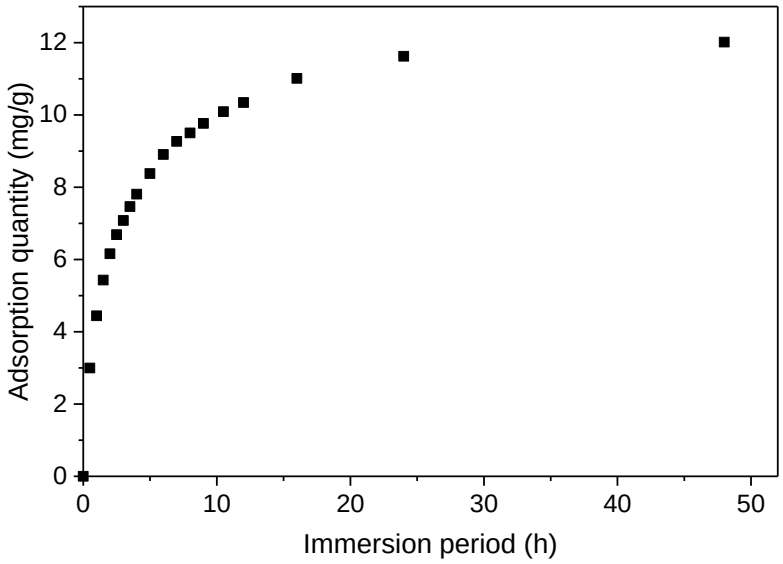


Fig. S24 Adsorption amounts of **1a** to a **PVA** sponge at different time. A **PVA** sponge (4 mg) was immersed in methanol solution (10 mL) of **1a** (2.0×10^{-5} M) at room temperature.

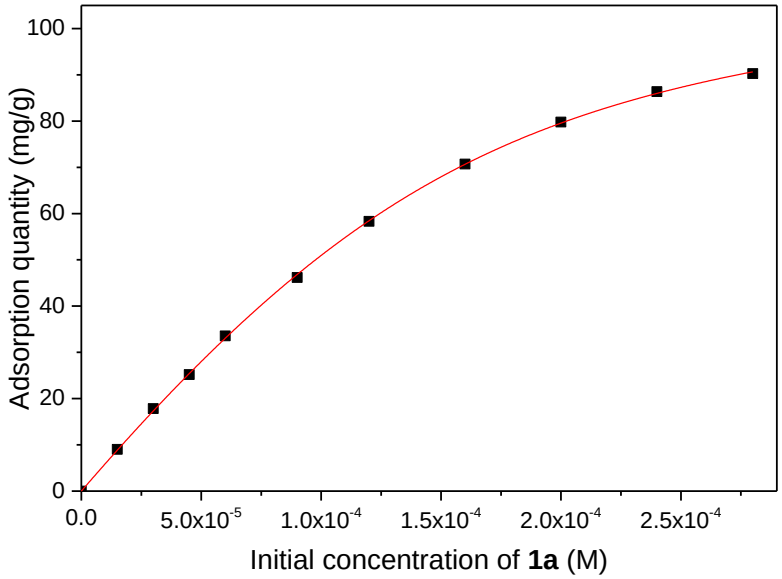


Fig. S25 Adsorption isotherm for **1a** to the **PVA** sponge. The solid line in red is the best fit to an equation on an assumption of a 1:1 stoichiometric binding of **1a** with the diol units of the **PVA** sponge.

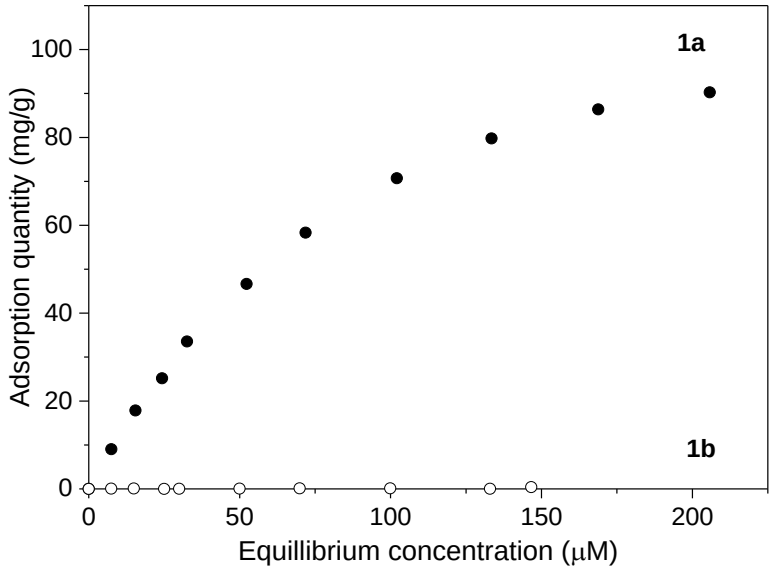


Fig. S26 Adsorption amounts of **1a** and **1b** to **PVA** sponges. **PVA** sponges (4 mg) were immersed in methanol solutions (10 mL) of **1a** and **1b** at different concentrations at room temperature for 15 h, respectively.

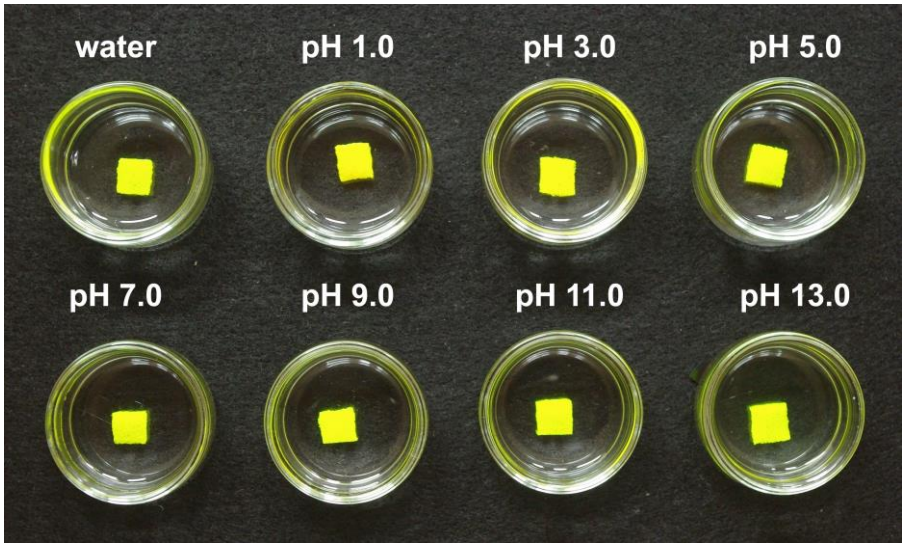


Fig. S27 Photograph of **PVA/1a** immersed in distilled water and aqueous solutions at different pH for 24 h. The photograph was taken under ambient light.

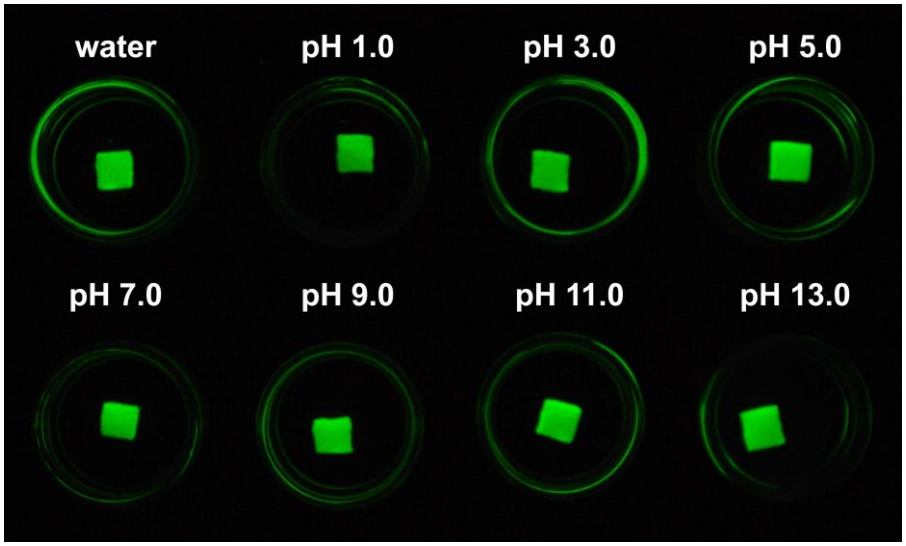


Fig. S28 Photograph of **PVA/1a** immersed in distilled water and aqueous solutions at different pH for 24 h. The photograph was taken under UV light. λ_{ex} = 365 nm.

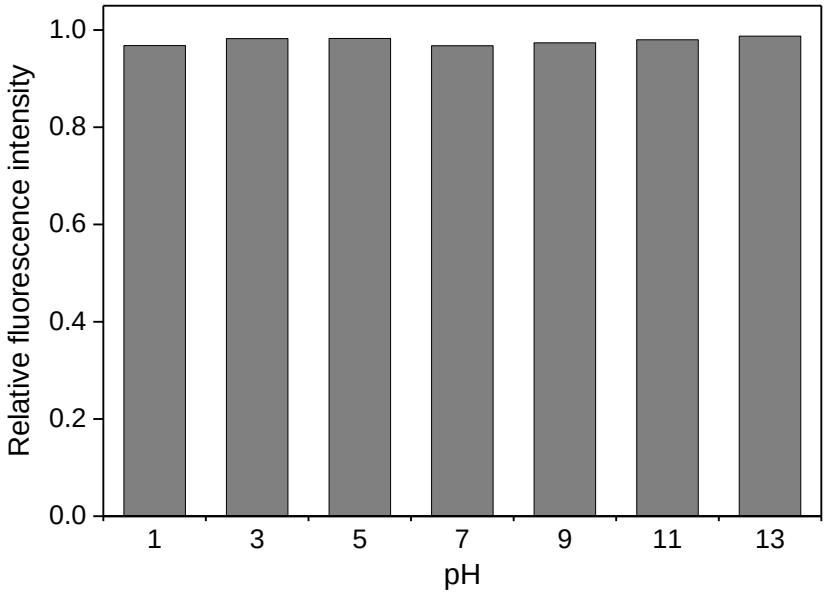


Fig. S29 Fluorescence intensities of **PVA/1a** immersed in aqueous solutions at different pH for 24 h. These data were normalized to the fluorescence intensity of **PVA/1a** in distilled water.

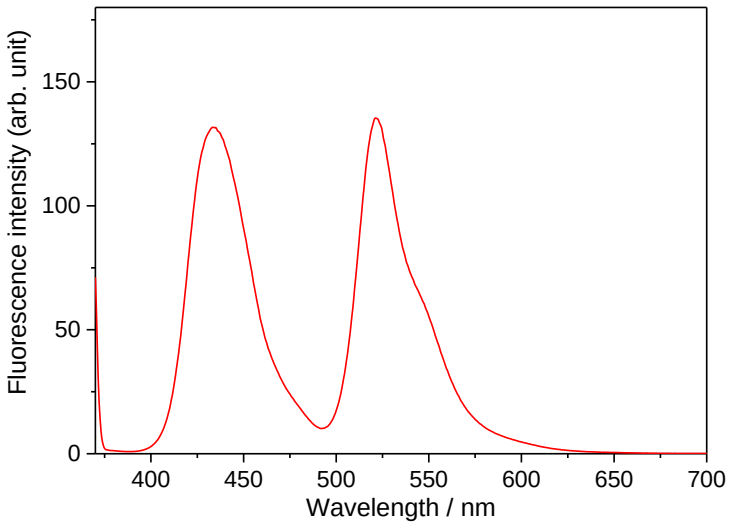


Fig. S30 Fluorescence spectrum of **PVA-C**. λ_{ex} = 365 nm. The amounts of immobilized **1a** and **3a** in **PVA-C** are 1.2 mg/g and 12.2 mg/g, respectively.

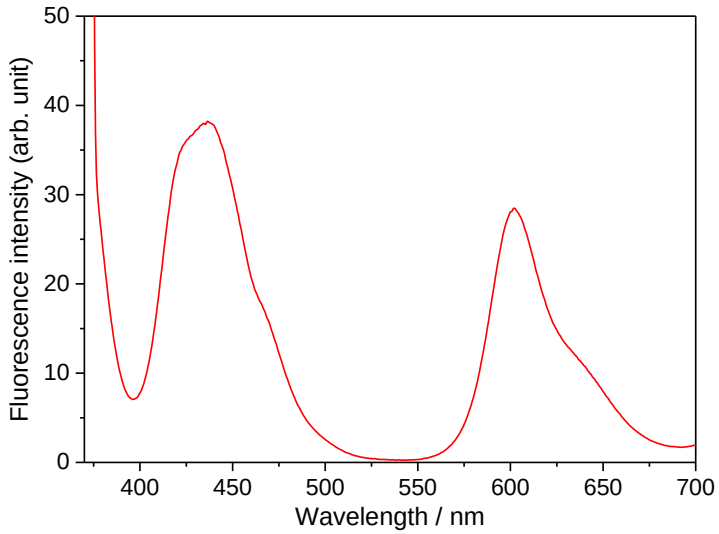


Fig. S31 Fluorescence spectrum of **PVA-M**. λ_{ex} = 365 nm. The amounts of immobilized **2a** and **3a** in **PVA-M** are 2.0 mg/g and 3.4 mg/g, respectively.

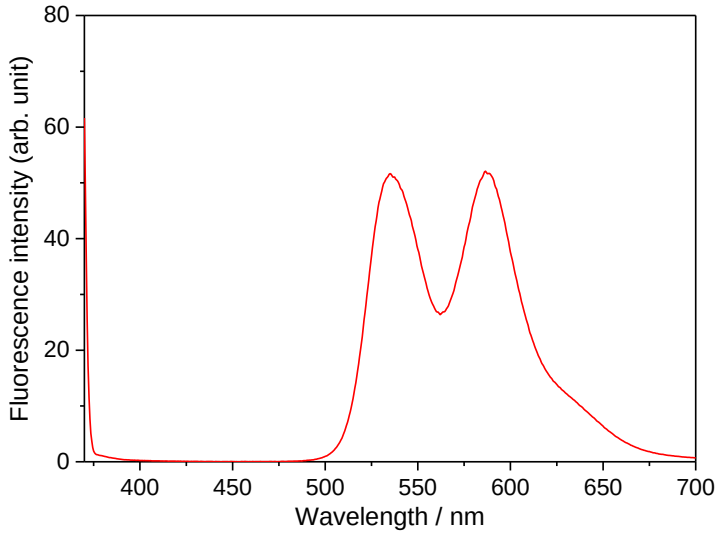


Fig. S32 Fluorescence spectrum of **PVA-Y**. λ_{ex} = 365 nm. The amounts of immobilized **1a** and **2a** in **PVA-Y** are 10.0 mg/g and 0.2 mg/g, respectively.

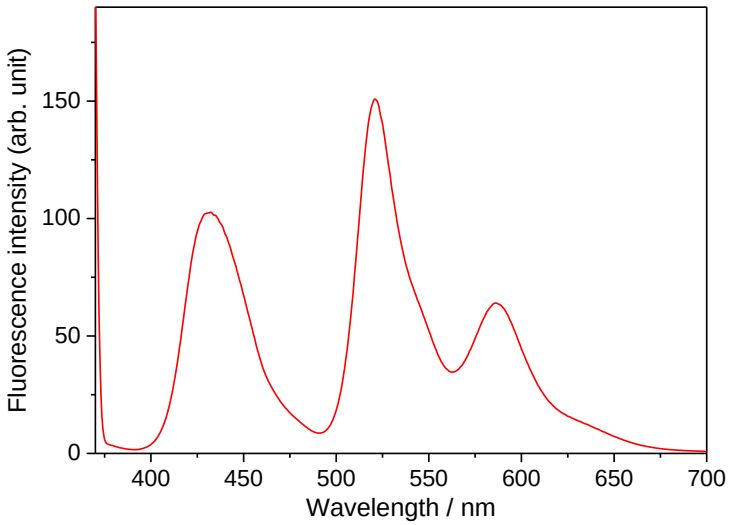


Fig. S33 Fluorescence spectrum of **PVA-W**. λ_{ex} = 365 nm. The amounts of immobilized **1a**, **2a** and **3a** in **PVA-W** are 0.6 mg/g, 0.1 mg/g and 8.6 mg/g, respectively.

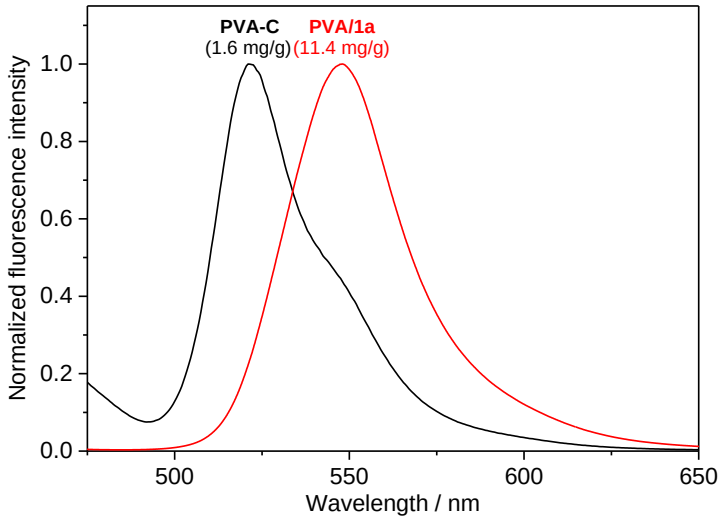


Fig. S34 Concentration-dependent fluorescence properties of the immobilized dyes for **1a**. Normalized fluorescence spectra of immobilized **1a** in **PVA-C** (λ_{em} = 521 nm, black) and **PVA/1a** (λ_{em} = 540 nm, red). The amounts of immobilized **1a** in **PVA-C** and **PVA/1a** are 1.2 mg/g and 11.4 mg/g, respectively. Immobilized **1a** in **PVA/1a** showed a fluorescence peak at 548 nm and immobilized **1a** in **PVA-C** showed a blue shifted fluorescence peak at 521 nm.

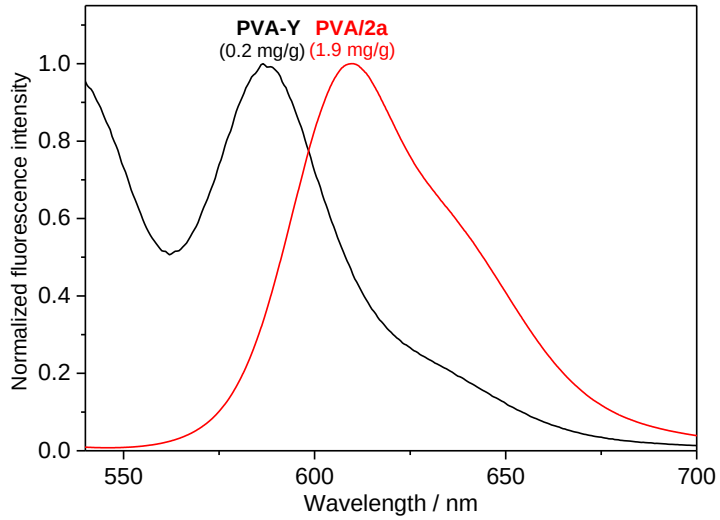


Fig. S35 Concentration-dependent fluorescence properties of the immobilized dyes for **2a** Normalized fluorescence spectra of immobilized **2a** in **PVA-Y** (λ_{em} = 587 nm, black) and **PVA/2a** (λ_{em} = 610 nm, red). The amounts of immobilized **2a** in **PVA-Y** and **PVA/2a** are 0.2 mg/g and 1.9 mg/g, respectively. Immobilized **2a** in **PVA/2a** showed a fluorescence peak at 610 nm and immobilized **2a** in **PVA-Y** showed a fluorescence peak at 587 nm

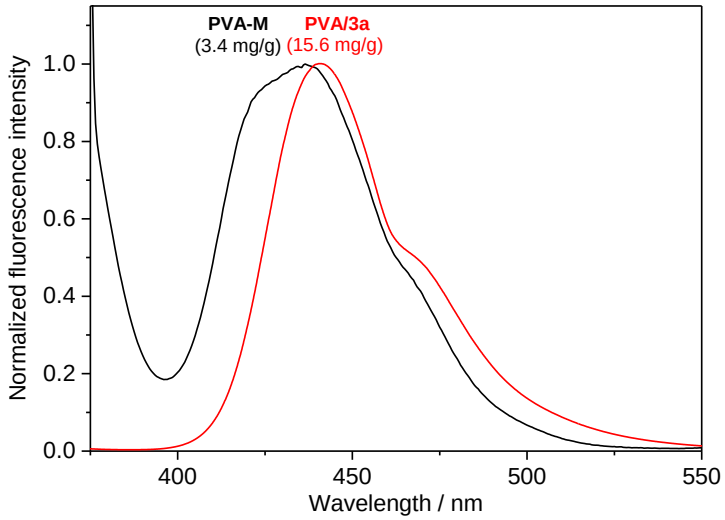


Fig. S36 Concentration-dependent fluorescence properties of the immobilized dyes for **3a** Normalized fluorescence spectra of immobilized **3a** in **PVA-M** (λ_{em} = 437 nm, black) and **PVA/3a** (λ_{em} = 400 nm, red). The amounts of immobilized **3a** in **PVA-M** and **PVA/3a** are 0.2 mg/g and 1.9 mg/g, respectively. Immobilized **3a** in **PVA/3a** showed a fluorescence peak at 440 nm. In contrast, immobilized **3a** in **PVA-M** showed a fluorescence peak at 437 nm with more distinct vibronic bands. These data indicated that the increase of the adsorbed amounts of dyes led to aggregation of dyes onto the sponge surface.

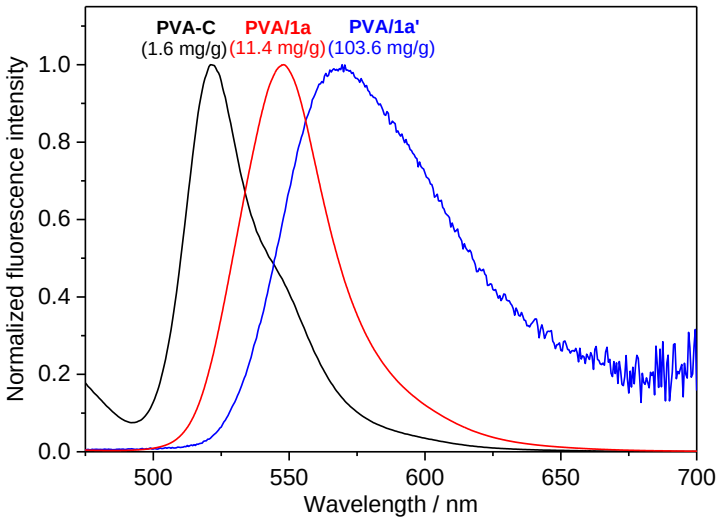


Fig. S37 Normalized fluorescence spectrum of a **PVA** sponge functionalized with **1a** at a nearly saturation point (**PVA/1a'**, the amount of immobilized **1a**: 103.6 mg/g, λ_{em} = 570 nm, fluorescence quantum yield: 5%, blue line). Normalized fluorescence spectra of **PVA-C** (the amount of immobilized **1a**: 1.6 mg/g, λ_{em} = 521 nm, black line) and **PVA/1a** (the amount of immobilized **1a**: 11.4 mg/g, λ_{em} = 540 nm, red line).

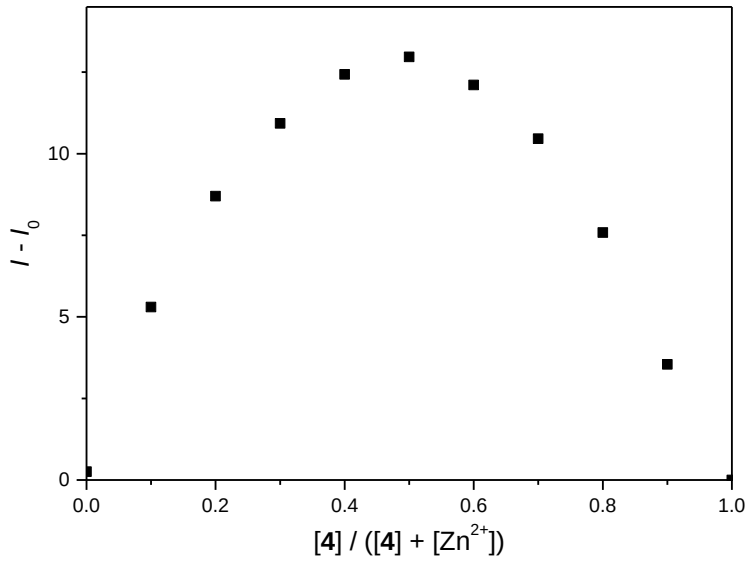


Fig. S38 Job's plot for complexation of **4** with Zn^{2+} ions in 5.0 mM HEPES buffer solution (2% DMSO, pH 7.0). Conditions: [**4**] + [Zn^{2+}] = 2.5 × 10^{-5} M, 25 °C, λ_{ex} = 325 nm, λ_{em} = 502 nm.[3]

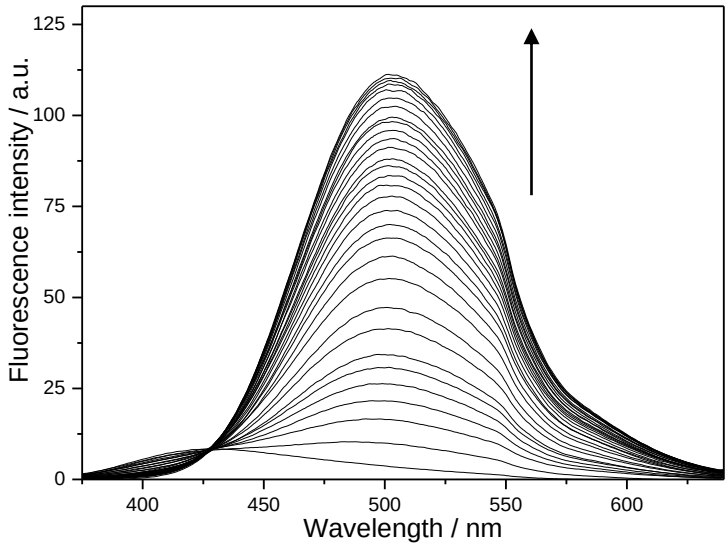


Fig. S39 Fluorescence spectra of **4** (2.5×10^{-5} M) in the presence of different concentrations of Zn^{2+} ions in 5.0 mM HEPES buffer solution (2% DMSO, pH 7.0). Conditions: 25 °C, λ_{ex} = 325 nm and λ_{em} = 502 nm.[3]

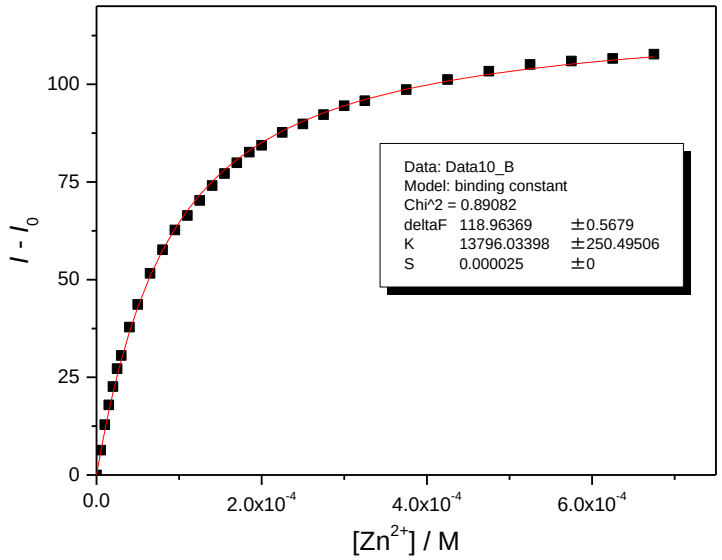


Fig. S40 Fluorescence titration data for complexation of **4** (2.5×10^{-5} M) with Zn^{2+} ions in 5.0 mM HEPES buffer solution (2% DMSO, pH 7.0). Conditions: 25 °C, λ_{ex} = 325 nm and λ_{em} = 502 nm.[3]

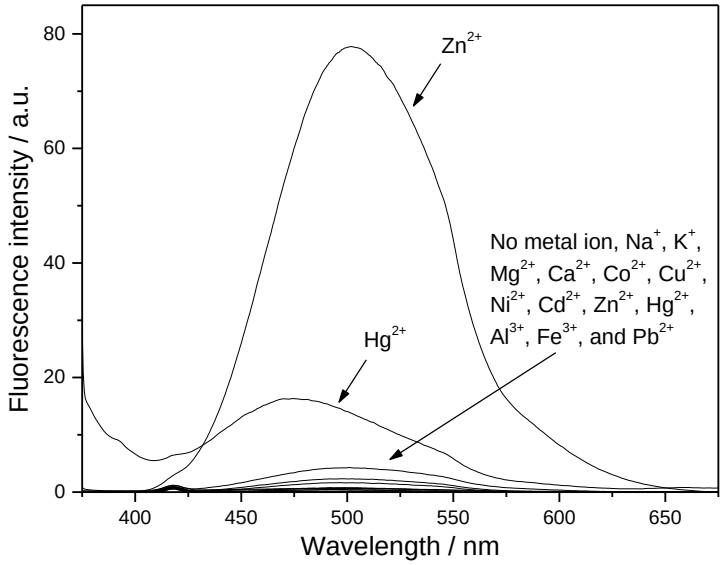


Fig. S41 Fluorescence spectra of **4** in the absence and presence of different metal ions (Na^+, K^+, Mg^{2+}, Ca^{2+}, Fe^{3+}, Co^{2+}, Ni^{2+}, Cu^{2+}, Zn^{2+}, Cd^{2+}, Hg^{2+}, Al^{3+} and Pb^{2+}) in 5.0 mM HEPES buffer solution (2% DMSO, pH 7.0). Conditions: [**4**] = [M^{n+}] = 5.0×10^{-5} M, 25 °C, λ_{ex} = 365 nm, λ_{em} = 502 nm.[3]

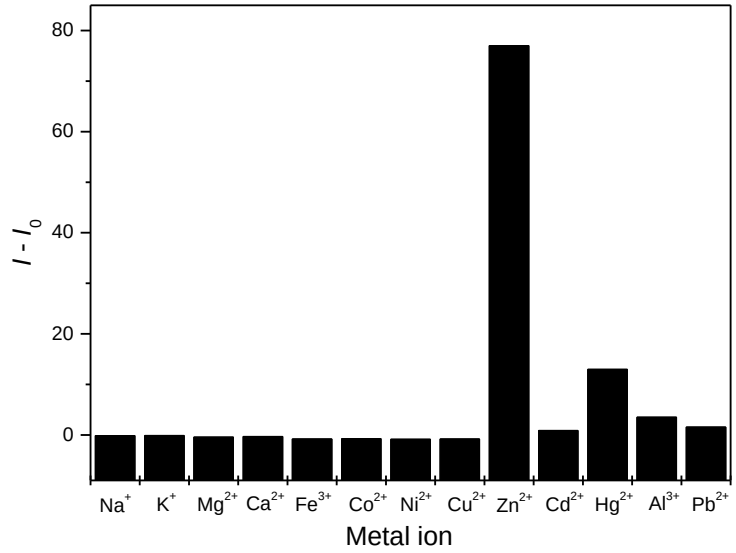


Fig. S42 Changes in the fluorescence intensities of 4 in the presence of different metal ions (Na^+, K^+, Mg^{2+}, Ca^{2+}, Fe^{3+}, Co^{2+}, Ni^{2+}, Cu^{2+}, Zn^{2+}, Cd^{2+}, Hg^{2+}, Al^{3+} and Pb^{2+}) in 5.0 mM HEPES buffer solution (2% DMSO, pH 7.0). Conditions: [**4**] = [M^{n+}] = 5.0×10^{-5} M, 25 °C, λ_{ex} = 365 nm, λ_{em} = 502 nm.[3]

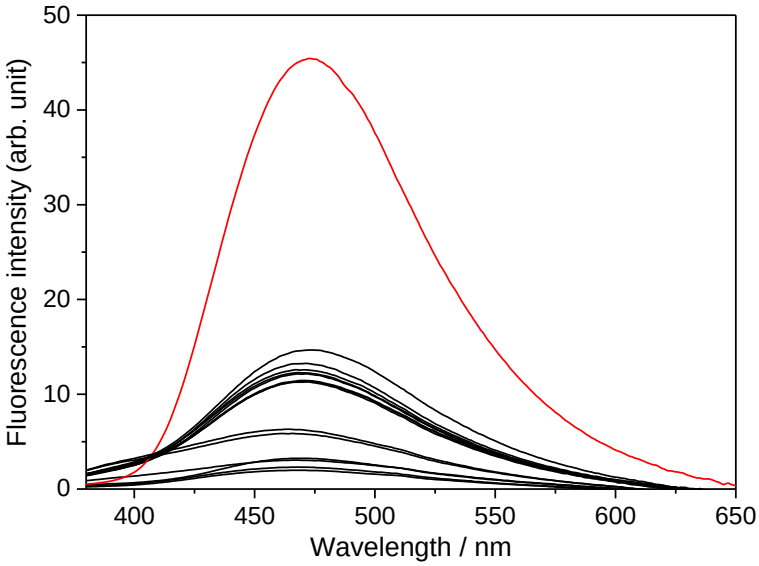


Fig. S43 Fluorescence spectra of **PVA/4** in aqueous solutions (10 mL) in the presence of Zn^{2+} ions (red line) and other metal ions (Na^{+}, K^{+}, Mg^{2+}, Ca^{2+}, Fe^{3+}, Co^{2+}, Ni^{2+}, Cu^{2+}, Cd^{2+}, Hg^{2+}, Al^{3+}, and Pb^{2+} ions, black lines). Immersion time: 30 min. Conditions: $[M^{n+}] = 3.0 \times 10^{-5}$ M, pH = 7.0, λ_{ex} = 365 nm.

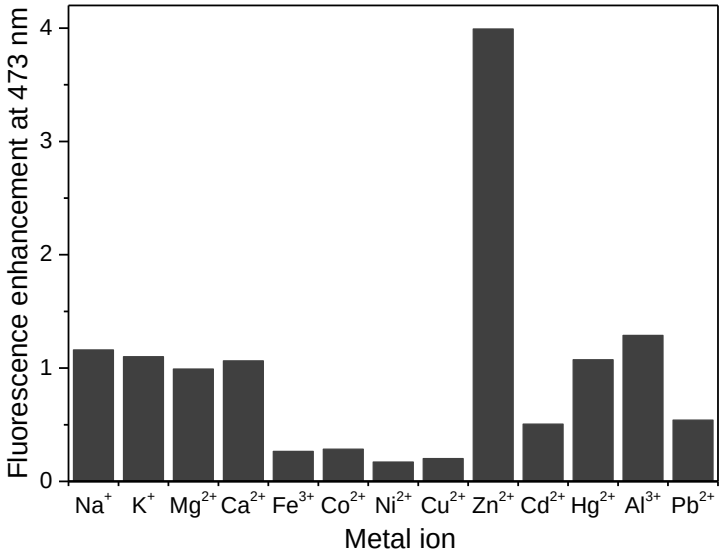


Fig. S44 Fluorescence enhancement of **PVA/4** in aqueous solutions (pH = 7.0, 10 mL) upon the addition of metal ions of Na^{+}, K^{+}, Mg^{2+}, Ca^{2+}, Fe^{3+}, Co^{2+}, Ni^{2+}, Cu^{2+}, Zn^{2+}, Cd^{2+}, Hg^{2+}, Al^{3+}, and Pb^{2+} ions ($[M^{n+}] = 3.0 \times 10^{-5}$ M) for 30 min. Immersion time: 30 min. λ_{ex} = 365 nm, λ_{em} = 473 nm.

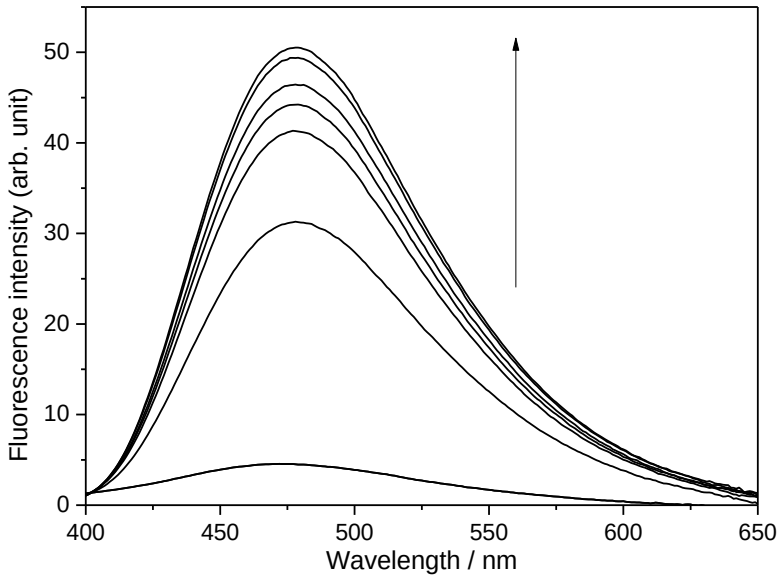


Fig. S45 Time-dependency (0, 5, 10, 15, 20, and 30 min) of fluorescence spectra of **PVA/4** after the addition of Zn^{2+} ions in aqueous solution (10 mL). Conditions: $[Zn^{2+}] = 1.5 \times 10^{-4}$ M, pH = 7.0, λ_{ex} = 365 nm.

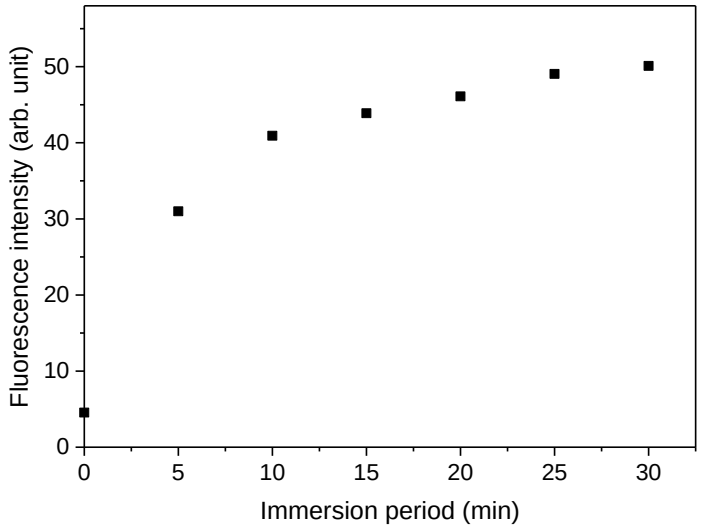


Fig. S46 Time-dependency in fluorescence intensity of **PVA/4** after the addition of Zn^{2+} ions in aqueous solution (10 mL). Conditions: $[Zn^{2+}] = 1.5 \times 10^{-4}$ M, pH = 7.0, λ_{ex} = 365 nm

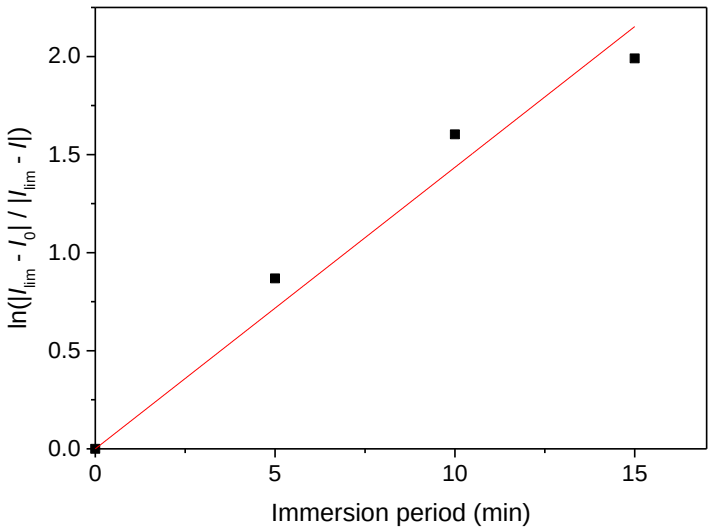


Fig. S47 Plots of ln(|I_{lim} − I_0| / |I_{lim} − I|) versus immersion period. **PVA/4** was immersed in aqueous solution (10 mL) in the presence of Zn^{2+} ions. Conditions: [Zn^{2+}] = 1.5 × 10^{-4} M, pH = 7.0, λ_{ex} = 365 nm and λ_{em} = 473 nm.

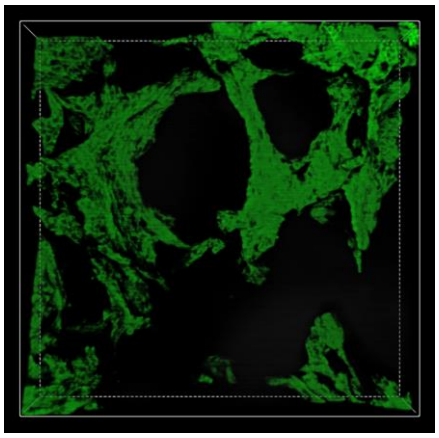


Fig. S48 CLSM image of **PVA /1a** incubated with HeLa cells for 3 days. The sample was stained with Hoechst. Conditions: λ_{ex} = 402 nm, λ_{em} = 425-475 nm (blue color) and λ_{ex} = 488 nm, λ_{em} = 500-550 nm (green color).

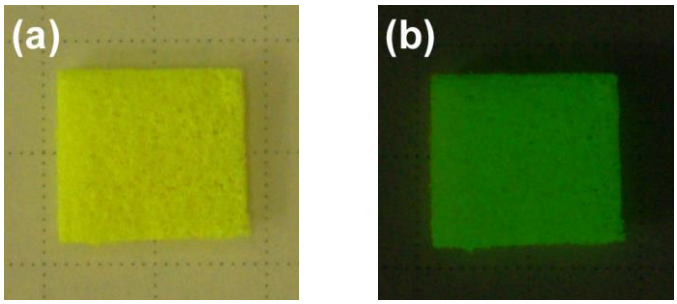


Fig. 49 Digital photographs of a **PVA** sponge co-functionalized with **1a** and boronic acid-appended L-lysine (**5**) trifluoroacetate salt. Photographs were taken under (a) ambient light and (b) UV light (λ_{ex} = 365 nm.), respectively.

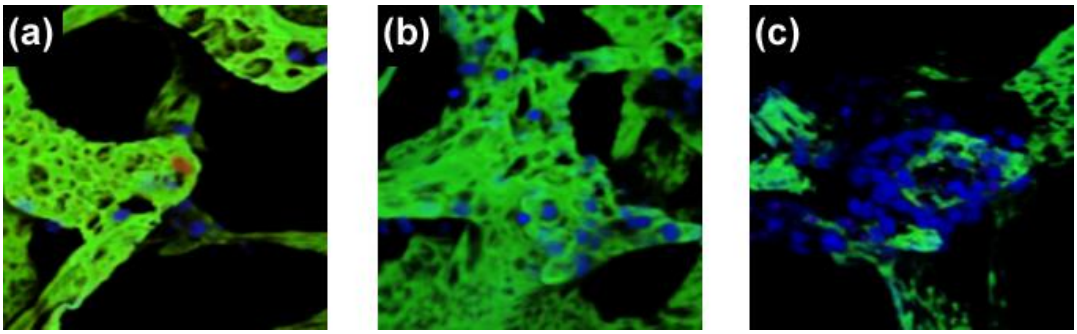


Fig. S50 CLSM images of HeLa cells cultured in the presence **PVA** sponge functionalized with **1a** and a boronic-acid appended lysine derivative at different periods; (a) 1 day, (b) 3 days and (c) 7 days. Hoechst (λ_{ex} = 402 nm indicated by blue color) and propidium iodide (λ_{ex} = 561 nm indicated by red color) were used for the detection of the living cells and the dead cells, respectively. Fluorescence from the sponge (λ_{ex} = 488 nm) are indicated by green color.