Supporting Information for:

Microfabrication and In Vivo Performance of a Microdialysis Probe with Embedded Membrane

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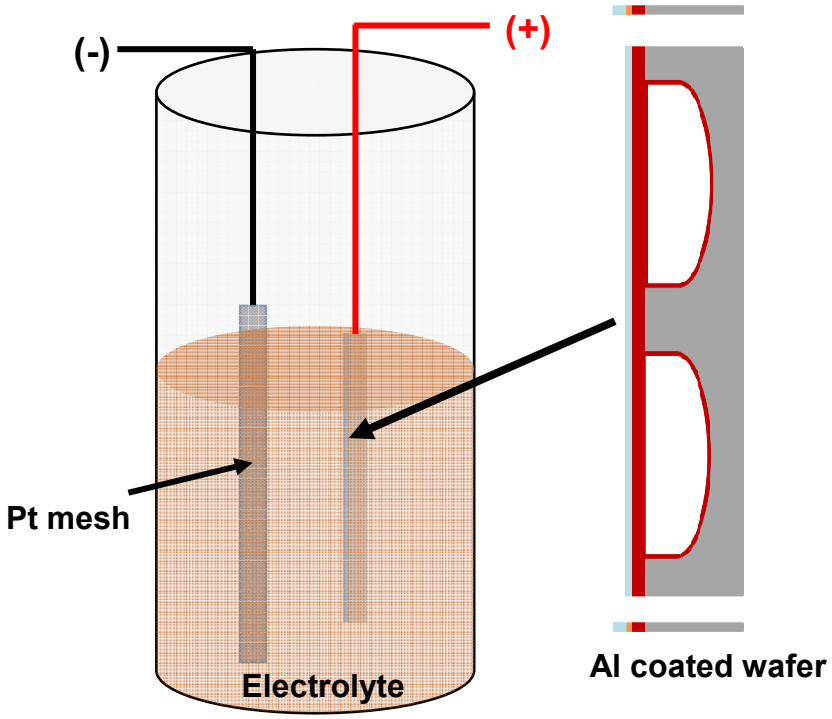
Figure S-1 Overall setup of AAO process. After deposition of 400 nm Al layer over the wafer, the Al coating was anodized at 60 V in 0.3 M oxalic acid for 15 min at 15 °C. Platinum mesh was used as a counter electrode, which was aligned parallel to the Al coated wafer. The device wafer was facing toward and 1inch apart from the platinum mesh.



Power supply	60 V
(-)	Platinum mesh
(+)	400 nm Al coated wafer
Electrolyte	0.3 M Oxalic acid
Temperature	15 °C
Time	15 min

Figure S-2 Fabrication of Probe Holder. The probe holder has guides to connect probe ports and capillaries. Probe holders were fabricated from a silicon wafer with growth of a 1 μm silicon dioxide layer. The 2 mm wide rectangle for a guide of probe connection ports was patterned by lithography and SiO_2 layer was removed by BHF. A second lithography step was performed to pattern 360 μm wide trenches for a guide of capillaries and DRIE etched 50 μm deep. Photoresist on the wafer was removed by positive resist stripper (PRS 2000; Avantor Performance Materials, PA) and 40 μm deep trenches and rectangles were additionally etched by DRIE. To connect fluidic capillaries to microchannels in a probe, a union capillary of 1 cm length with 180 μm ID and 360 μm OD was glued on the guiding trenches of a probe holder with epoxy gel resin. (A) Probe ports were inserted into union capillaries and 12 cm length of 100 μm ID and 150 μm OD fluidic capillaries were joined to inlet and outlet probe ports through the union capillary. Finally, all assembled devices were sealed with epoxy gel resin. The probe glued to the holder as shown in (B).

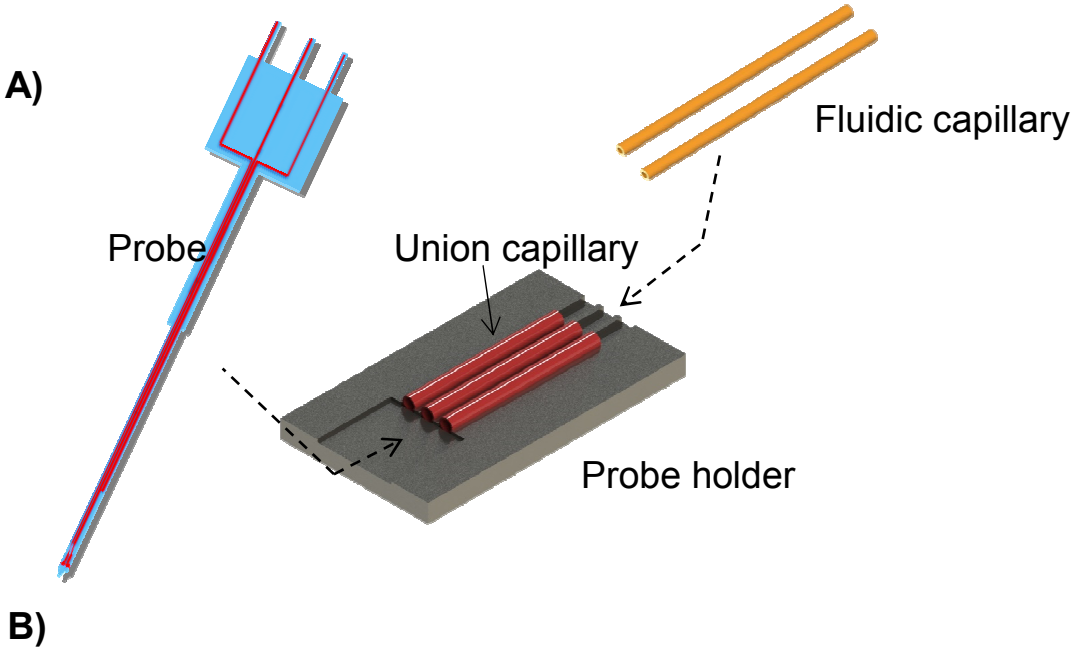


Figure S-3 Determination of the dynamic response time of the microfabricated probe to step change in sampled concentration, and the relationship between perfusion flow rate and relative recovery. (A) Illustration of experimental setup. Probe was inserted into a solution of water and then switched to a solution of 50 μM fluorescein as the sample. Fluorescence was monitored downstream on the outlet capillary. (B) Trace shows representative example for detection fluorescent change with time beginning at switch. Delay time is 14.5 min, which corresponds to the internal volume of ~1400 nL at the perfusion rate of 100 nL/min. Inset shows the 10% to 90% response time. Average was 47 ± 2 s (n = 7). (C) Scatter plot shows an inverse relationship between flow rate and relative recovery from the microfabricated probe sampling.

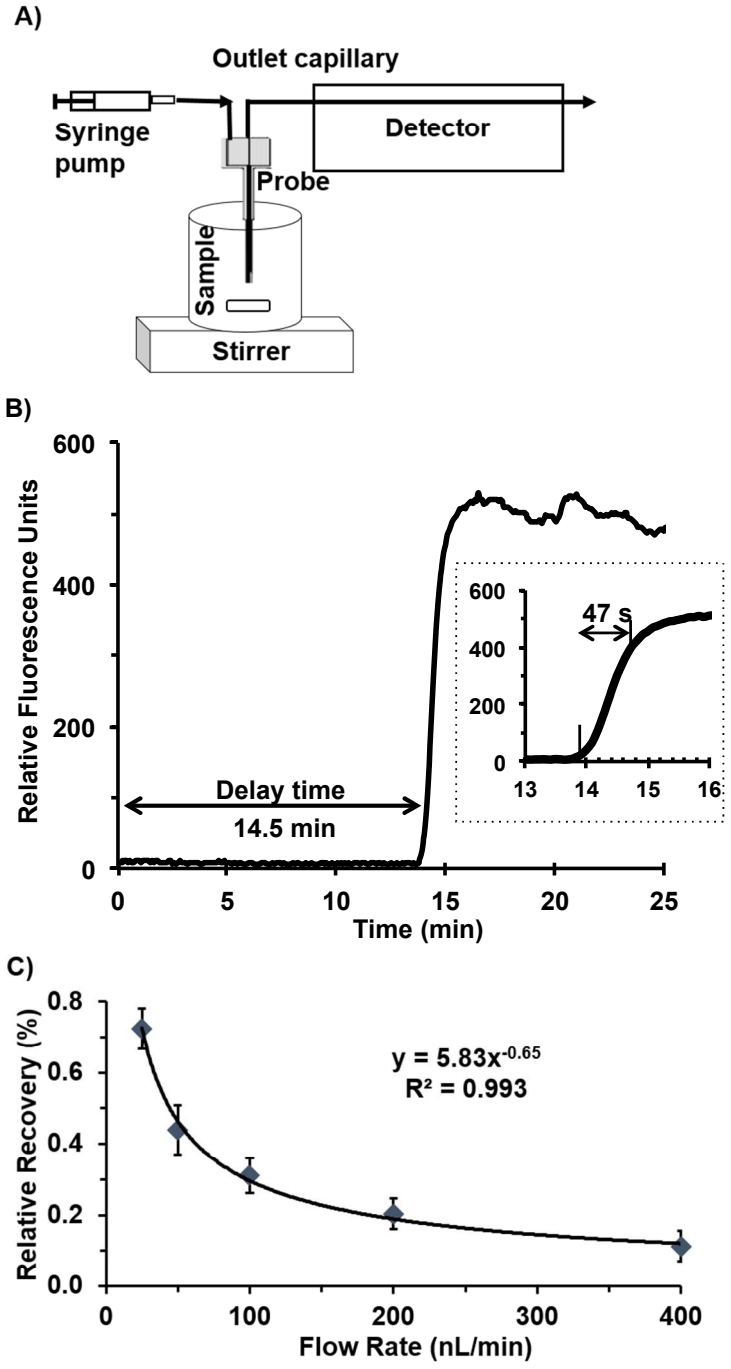


Figure S-4 Comsol modeling and simulation of 1 mM dopamine diffusing through the membrane of microfabricated probe. The microfabricated probe was constructed in COMSOL Multiphysics 4.4 (Burlington, MA) to model its recovery. The fabricated probe has semi-circular fluidic channels; but for ease of modeling, the channels were designed as 60 µm x 24 µm rectangles which gave the same cross-sectional area as the actual probes. The total length of the channels was 8 mm in a 'U' shape like the actual probe. The porous membrane was designed as a polysilicon rectangle, 5 µm x 60 µm, overlaid the channels. A large box was connected to the external boundary of the membrane region for simulating the probe in a well-stirred solution.

The 'Free and Porous Media Flow' and 'Species Transport in Porous Media' physics models were applied to the channel and membrane regions. Navier-Stokes equations modeled the fluid (water) in the open channel with a flow rate of 100 nL/min. Fluid movement in the porous region was defined by the Brinkman equation and a permeability of 50 nm^2 was used. The porosity variable was modified to match experimental data. For the transport of chemical species, the diffusion coefficient of dopamine, 6 $x10^{-10}$ m^2/s was used†. The exterior probe volume used 'Laminar Flow' and 'Transport of Diluted Species' physics with a flow rate of 10 mL/min to simulate a well-stirred solution. Images below show cross section of probe at the middle and end of the probe color coded for DA concentration. A) Recovery for probe with 30% porosity and B) for 0.9% porosity.

† Gerhardt, G.; Adams, R. N. *Anal. Chem.* **1982**, *54* (14), 2618–2620.

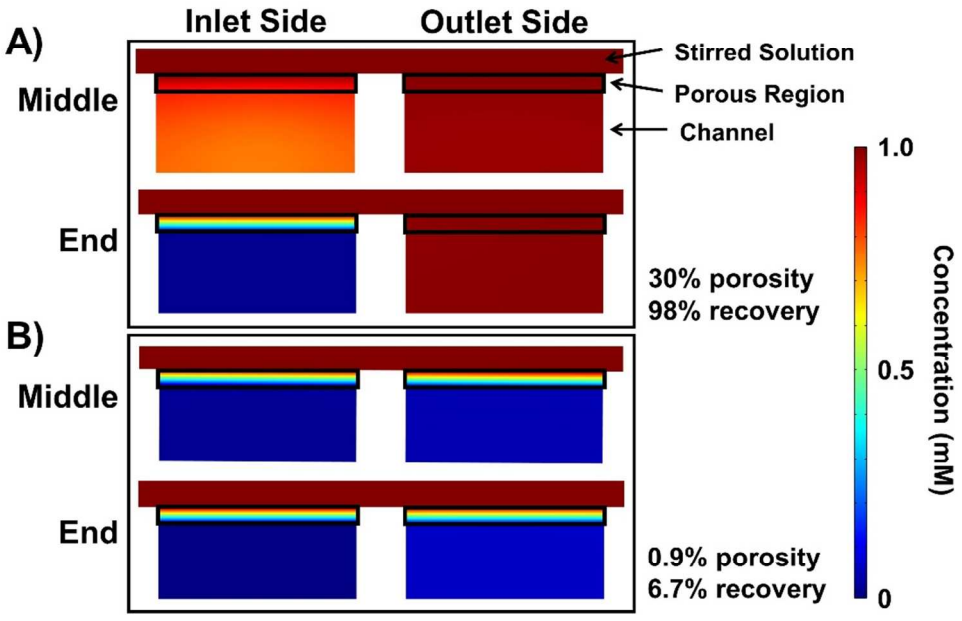


Figure S-5 Histology indicating probe placements and smaller probe tract by μFab sampling. A) The brain was implanted bilaterally by a conventional dialysis probe (left) and the microfabricated probe (right). The brain was cut into 40 μm coronal sections and Nissl stained to show overall brain structure with probe tracts (boxes with dashed line). B) Dotted lines indicates tracts overlaid on a rat brain atlas diagram, corresponding to (A).

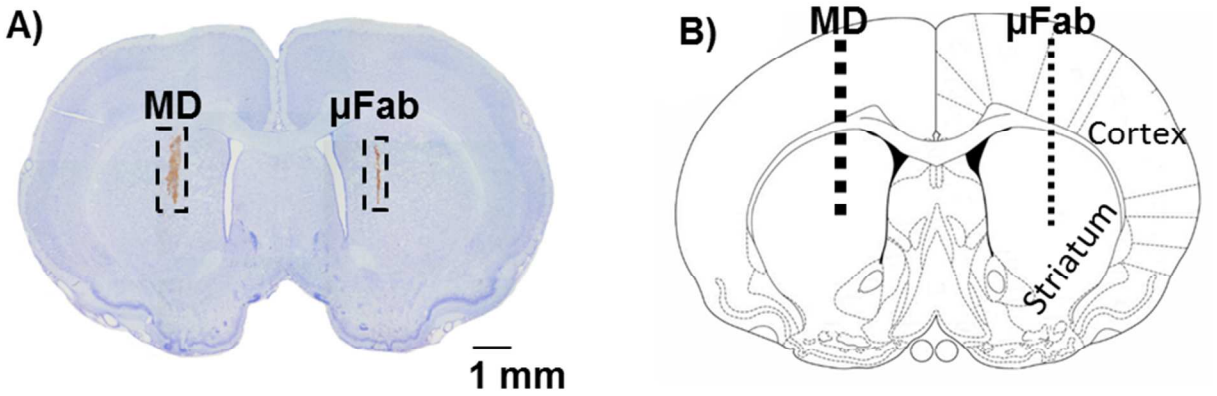


Table S-1. Examples of materials and fabrication processes of semi-permeable membranes on a microfluidic devices.

Device type	*Device material*	*Membrane material*	*Membrane fabrication Process*	*Ref.*
Probe	Polyimide	Polyimide	Ion irradiation and chemical etching by sodium hypochlorite[30]	[21]
	Silicon	Silicon	Sacrificial oxide spacer layer[22] or permeable polysilicon	[10]
	Parylene	Cellulose acetate	Spin coating and drying	[20]
Biocapsule	Silicon	Silicon	Sacrificial oxide spacer layer	[22]
Microchip	Glass	Zwitterionic polymer	laser-patterning techniques[27]	[28]
	PDMS	Polycarbonate	Oxygen plasma	[29]
	PDMS	Silicon- silicon nitride	Thermal annealing	[24]
	Silicon	Silicon	Electrochemical etching	[25]

Table S-2. In vitro recovery of selected neurochemicals at flow rate of 100 nL/min. The results from the microfabricated probes (μFab) are compared with the conventional dialysis probes (MD). Recovery values are given as mean ± 1 standard deviation for different probes (n = 4 probes).

Analyte (tested concentration)	μFab % Recovery	MD % Recovery
Acetylcholine (500 nM)	7 ± 1.2	105 ± 6
Choline (50 μM)	6 ± 1.1	99 ± 12
3,4-Dihydroxyphenylacetic acid (10 μM)	2 ± 0.6	92 ± 5
3,4-Dihydroxyphenylalanine (1 μM)	3 ± 0.5	94 ± 9
Dopamine (500 nM)	5 ± 0.9	100 ± 3
γ-Aminobutyric acid (10 μM)	3 ± 1.2	109 ± 3
Glucose (1 mM)	3 ± 0.6	95 ± 10
Glutamate (10 μM)	21 ± 7.7	106 ± 10
Glutamine (500 μM)	2 ± 0.5	105 ± 6
Histamine (1 μM)	5 ± 1.0	109 ± 8
5-Hydroxyindoleacetic acid (10 μM)	4 ± 0.5	99 ± 3
Homovanillic acid (10 μM)	3 ± 1.1	105 ± 4
3-Methoxytyramine (500 nM)	4 ± 0.8	95 ± 2
Phenylalanine (10 μM)	4 ± 2.1	115 ± 6
Serine (50 μM)	3 ± 0.9	111 ± 20
Serotonin (500 nM)	4 ± 0.6	103 ± 4
Taurine (50 μM)	4 ± 0.6	110 ± 4
Tyrosine (10 μM)	4 ± 1.3	105 ± 7