

Supplementary Data

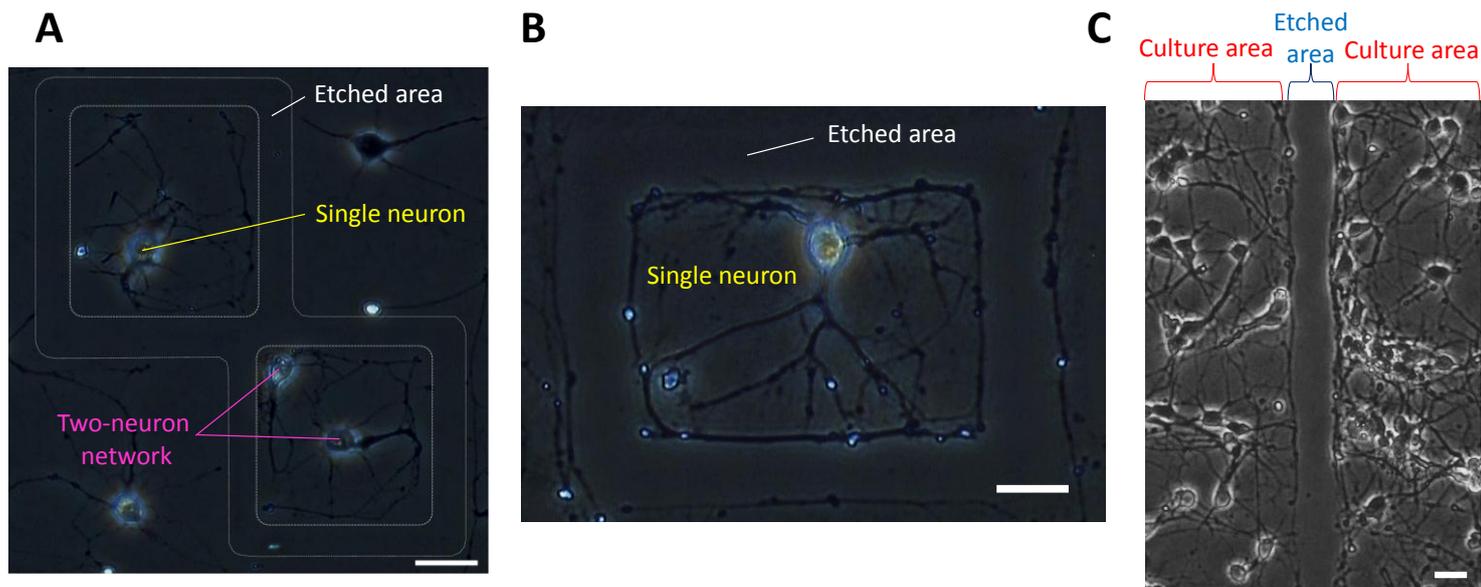


Figure S1. Control of cultivable area using collagen gel photothermal etching. Neurons and neurites did not grow into the etched area. (A) Isolated single neuron and two-neuron network at 7DIV. (B) Isolated neuron at 14 DIV. (C) A lot of neurons and neurites at 21 DIV did not grow into etched straight area.

Movie S1. Collagen gel photothermal etching during cultivation. Collagen gel around the target neuron at 2

DIV was etched by laser irradiation and scanning under microscopic observation.

Movie S2. Isolated single neuron at 11 DIV. Single neuron elongated neurites into 10 μ m thick 3D collagen

gel. This movie was taken by shifting the focus to the Z axis.

Movie S3. Isolated two-neuron network at 5DIV. Two neurons elongated neurites into 3D collagen gel and

generated connections. This movie was taken by shifting the focus to the Z axis.

Movie S4. Photothermal etching to control the direction of neurite elongation and the position of synaptic

connection.

Movie S5 [Ca²⁺]_i imaging of isolated single neuron. [Ca²⁺]_i responses of a Oregon green 488 BAPTA-1

loaded isolated neurons at 8 days of culture. The movie comprises 240 images collected at 0.06-s intervals.

Movie S6 $[Ca^{2+}]_i$ imaging of isolated six neurons network. $[Ca^{2+}]_i$ responses of a Oregon green 488

BAPTA-1 loaded isolated neurons at 8 days of culture. The movie comprises 500 images collected at 0.06-s intervals.