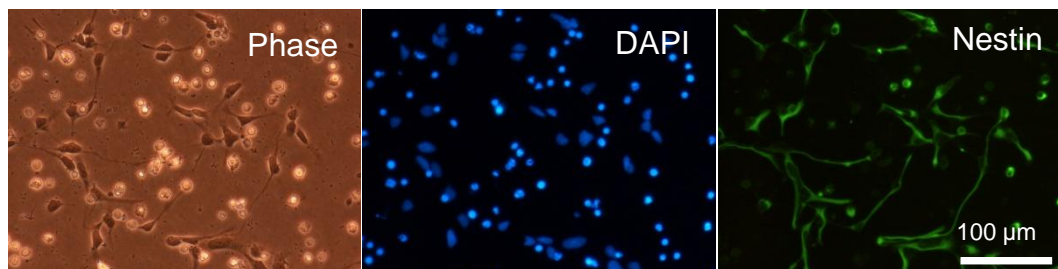


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Viability: 71.7% (measured by trypan blue staining)

## Immunostaining images of the NPCs cultured for 1 day

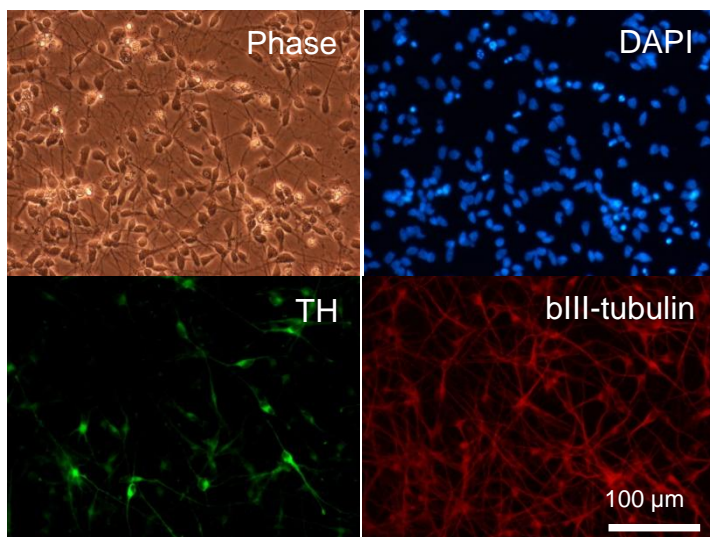


Primary antibody  
Anti-nestin (1:750)  
Millipore, AB5922

Secondary antibody  
Alexa<sup>®</sup> 488 anti-rabbit IgG (1:750)  
Invitrogen, A11008

Nestin-positive cells are obtained after replating the NPCs.  
Nestin-positive cells are 95.0% of total cell.

## Immunostaining images of the NPCs cultured for 14 days



Primary antibody  
Anti-βIII-tubulin (1:750)  
Sigma, T8660

Anti-tyrosine hydroxylase (TH) (1:250)  
Epitomics, 2266-1

Secondary antibody  
Alexa<sup>®</sup> 546 anti-mouse IgG (1:750)  
Invitrogen, A21123

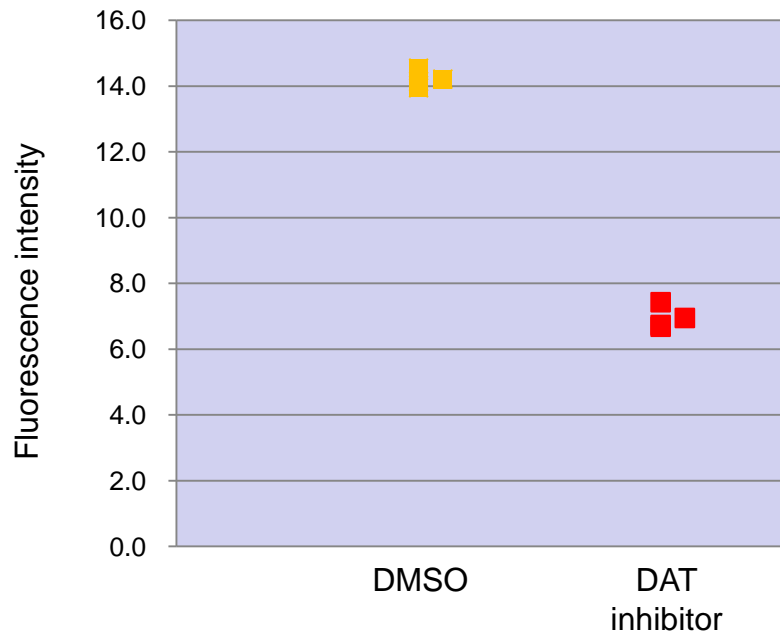
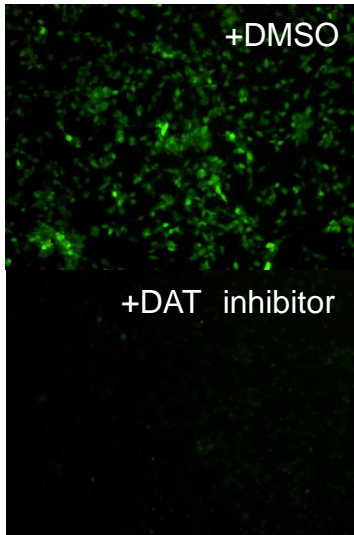
Alexa<sup>®</sup> 488 anti-rabbit IgG (1:750)  
Invitrogen, A11008

Dopaminergic neurons expressing both βIII-tubulin (neuron marker) and TH (dopaminergic neuron marker) are obtained after a 14-day culture.  
βIII-tubulin positive cells are 79.4% of total cell.  
TH positive cells are 33.2% of βIII-tubulin positive cell.

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## Dopamine uptake by dopaminergic neurons derived from human iPS cells



Dopaminergic neurons derived from human iPS cells were treated with dimethyl sulfoxide (DMSO) or a dopamine transporter (DAT) inhibitor (GBR12909, 20  $\mu$ M). Dopamine uptake was measured by using the Neurotransmitter Transporter Uptake Assay Kit (Molecular Devices). The fluorescence intensity in the control was reduced by DAT inhibitor treatment. These results indicate the functional expression of the neurotransmitter transporter. Average is shown as X. n=1

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