

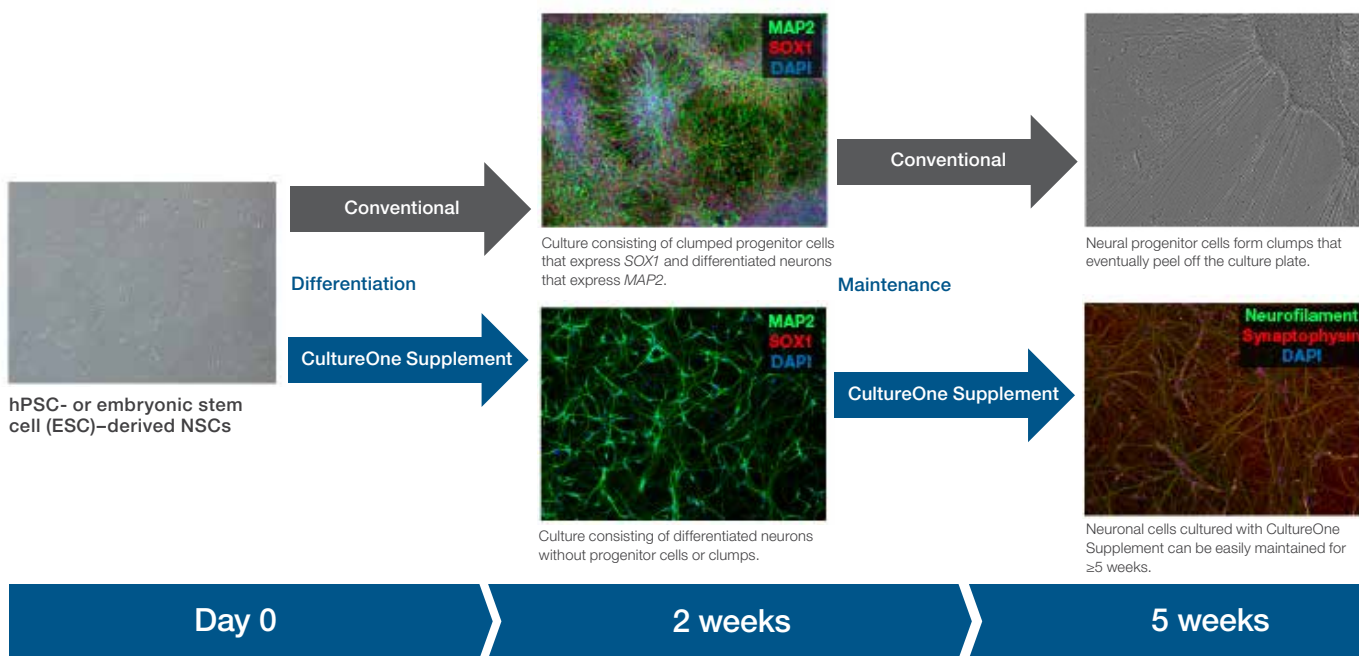
Improve neuronal cell culture

With CultureOne Supplement

Remove progenitor cells from hPSC-derived neuronal cell cultures and accelerate maturation for consistent downstream assays.

Neurons differentiated from human pluripotent stem cell (hPSC)-derived neural stem cells (NSCs) enable scientists to study human neural diseases in a larger and more diverse patient population than ever before. However, neuronal cell cultures are typically contaminated with proliferating neural progenitor cells (NPCs) that form clumps during and after differentiation. These contaminating progenitor cells make it nearly impossible to perform critical downstream assays with the mature neurons.

Gibco™ CultureOne™ Supplement can be added to any conventional neuronal differentiation medium to eliminate more than 75% of contaminating NPCs with minimal cell death and no effect on kinase-mediated pathways. The resulting cultures of evenly distributed neurons exhibit accelerated neuronal maturation and facilitate downstream analysis, and the cells can be maintained in culture for five weeks or more.



Neuronal purity in cell culture matters

CultureOne Supplement improves downstream assays

Differentiation with CultureOne Supplement facilitated imaging, mRNA expression, and electrophysiological analysis. After two weeks of differentiation with CultureOne Supplement, differentiated neurons (MAP2⁺) were evenly distributed with 75% fewer NPCs (SOX1⁺) and cell clumps than conventionally differentiated neurons (Figure 1A). The cell population also expressed more neuronal mRNA and less NPC mRNA (Figure 1C).

Accelerated maturation

Neurons differentiated from NSCs with CultureOne Supplement displayed a spike in cytosolic calcium when depolarized with KCl (Figure 1B), meaning they expressed significantly more voltage-gated calcium ion channels than neurons differentiated by the conventional method. Voltage-gated calcium ion channels are important markers of neuronal maturity and excitability. Longer neurites were also observed after two weeks of differentiation with CultureOne Supplement. Together, these results indicated that CultureOne Supplement accelerated neuronal maturation.

Ordering information

Description	Cat. No.
CultureOne Supplement (100X)	A3320201
B-27 Supplement (50X), serum free	17504044
B-27 Plus Supplement (50X)	A3582801
Neurobasal Medium	21103049
Neurobasal Plus Medium	A3582901

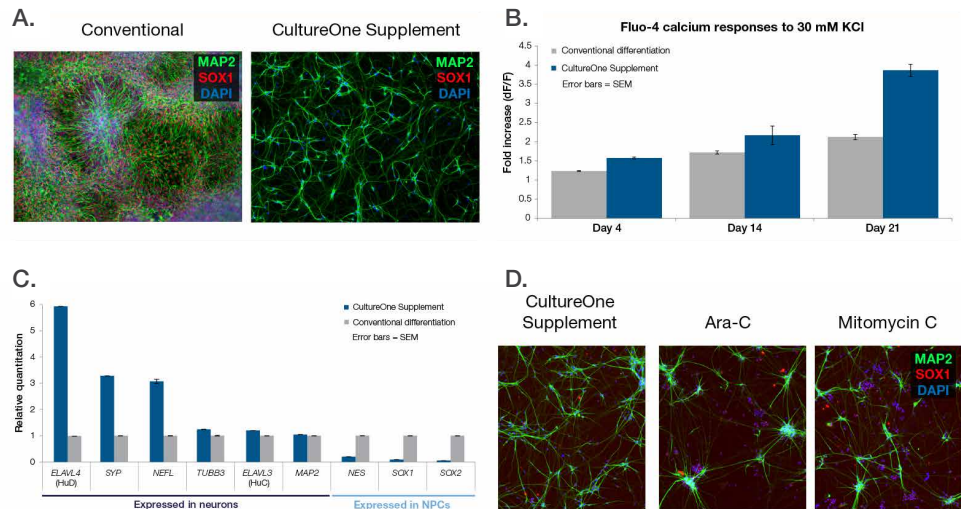


Figure 1. Comparison of neuronal differentiation and enrichment with and without CultureOne Supplement. (A) Imaging of differentiation on week 2. (B) Voltage-gated calcium channel activation. (C) qPCR analysis of mRNA expression on week 2. (D) Neuronal enrichment on week 2.

Superior enrichment

Neuronal cell cultures differentiated with CultureOne Supplement were compared to cultures that underwent antimetabolic enrichment with Ara-C or mitomycin C (Figure 1D). The cultures treated with CultureOne Supplement displayed better neurite outgrowth with less cell clumping and death. After two weeks, CultureOne Supplement yielded an enriched culture of neurons with approximately 60% of the starting NSC plating density. CultureOne Supplement can be paired with Gibco™ B-27™ Supplement or B-27™ Plus Supplement to further advance your neuronal differentiation experiments.