

# SURVIVAL OF THE FITTEST

Increase neuronal cell survival by more than 50%  
with the B-27 Plus Neuronal Culture System



# The standard in neuronal cell culture just got better

Since 1993, Gibco™ B-27™ Supplement and Neurobasal™ Medium has been the trusted standard for a variety of neuronal culture applications, with citations in more than 11,000 publications. However, as the desire for more reliable and biologically relevant models has increased, so too has the necessity for a media system that can maintain and mature functional neurons at optimal densities over longer periods *in vitro*. To meet this need, we have developed the Gibco™ B-27™ Plus Neuronal Culture System. It features an optimized formulation, upgraded manufacturing process, and more stringent quality control for raw materials and final product. These improvements enable:

- Increased neuronal survival by more than 50%—the highest in the industry
- Accelerated neurite outgrowth
- Improved electrophysiological activity and maturation of neurons

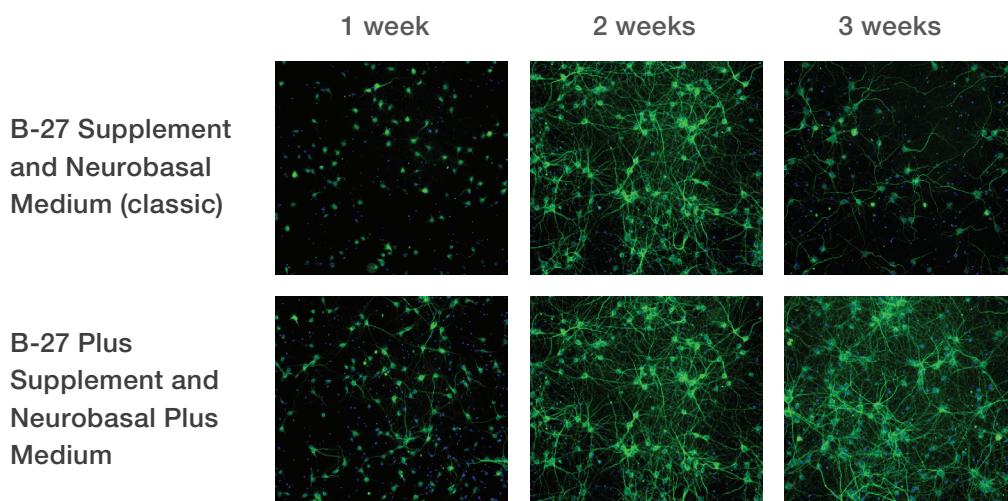
## The B-27 Plus Neuronal Culture System includes:

- Gibco™ B-27™ Plus Supplement (50X), 10 mL
- Gibco™ Neurobasal™ Plus Medium, 500 mL

Seamlessly transition from other neuronal cell culture systems, such as classic Neurobasal Medium with B-27 Supplement, in maintenance, maturation, and differentiation protocols with no change to your current workflow.



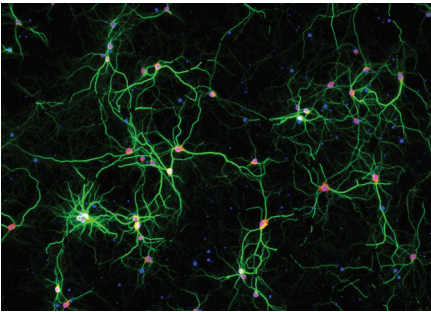
## Increases neuronal survival by more than 50% compared to the classic products



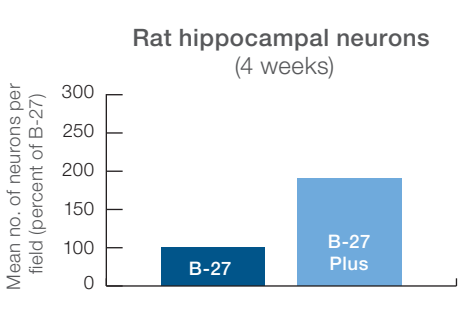
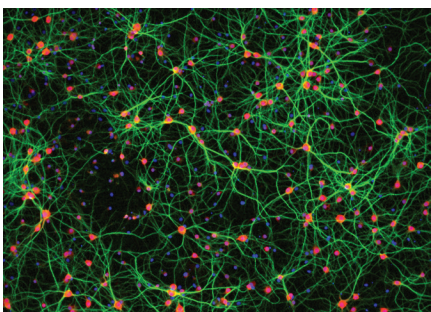
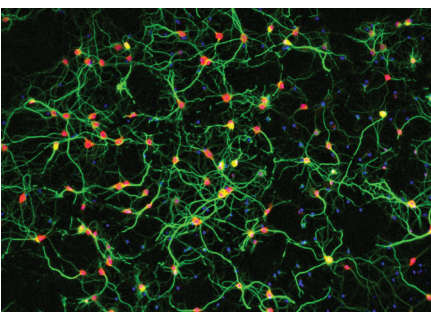
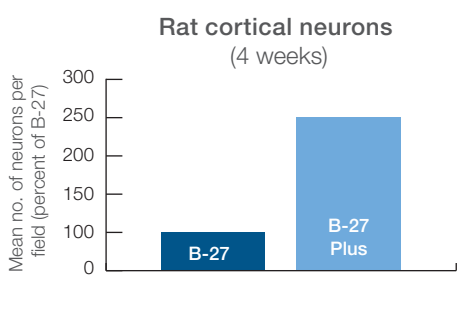
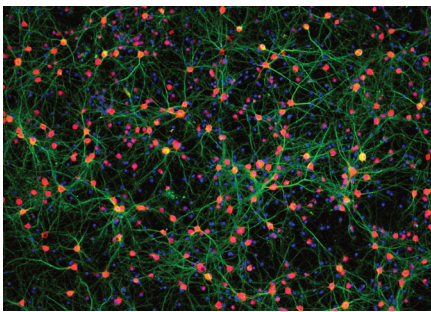
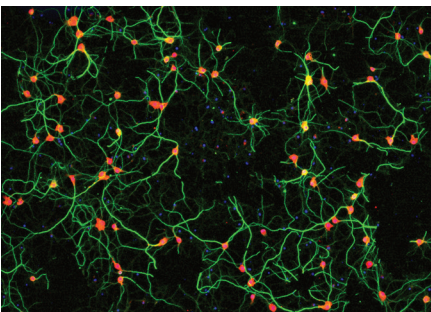
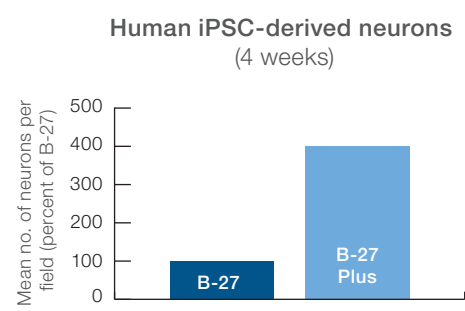
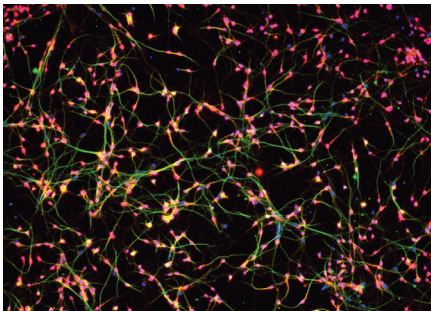
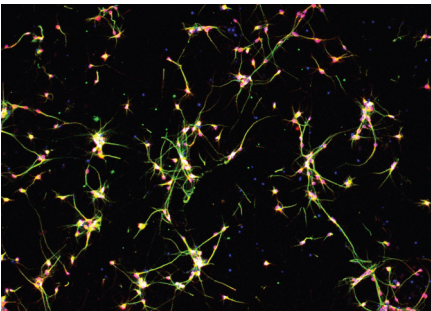
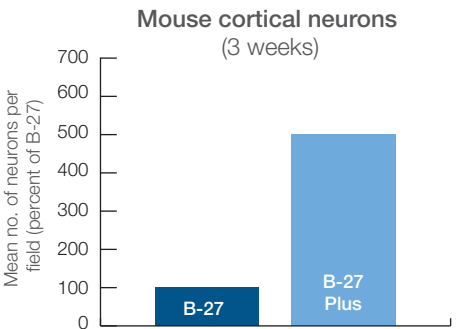
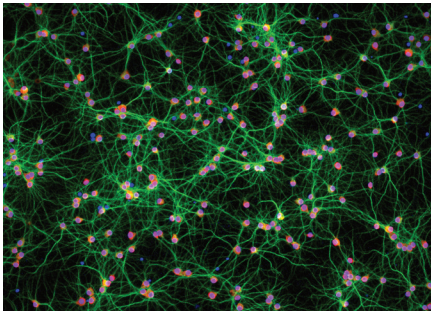
**Figure 1. B-27 Plus Neuronal Culture System maintains higher neuronal survival rates compared to classic B-27 Supplement and Neurobasal Medium.** Gibco™ Primary Rat Cortex Neurons (Cat. No. A10840) were cultured for up to 3 weeks in the listed media system and then immunostained for MAP2 on days 7, 14, and 21.

# Superior viability of primary rodent and human iPSC-derived neurons

B-27 Supplement and Neurobasal Medium



B-27 Plus Supplement and Neurobasal Plus Medium

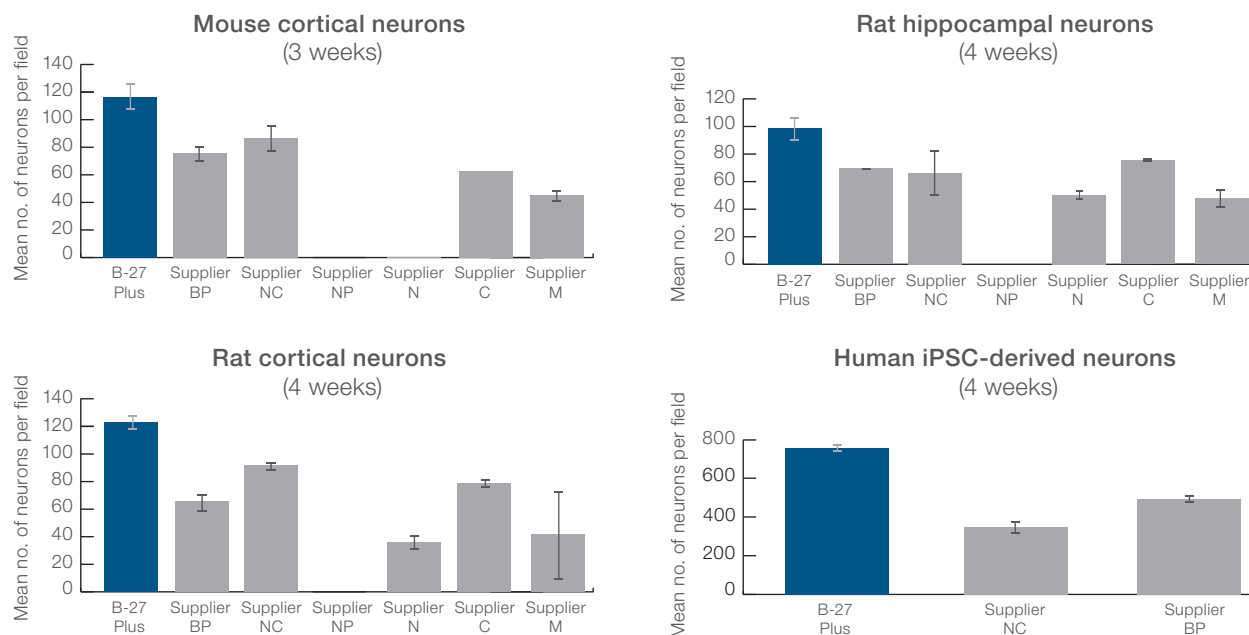


MAP2 HuC/D DAPI

Figure 2. The B-27 Plus Neuronal Culture System enables superior cultures of primary rodent and induced pluripotent stem cell (iPSC)-derived neurons compared to B-27 Supplement and Neurobasal Medium. Cryopreserved neurons were cultured for 3–4 weeks in the listed media system. Neurons were immunostained at the indicated time points for the neuronal dendritic marker MAP2 (green) and the neuronal cell body marker HuC/D (red). Nuclei were counterstained with DAPI (blue).

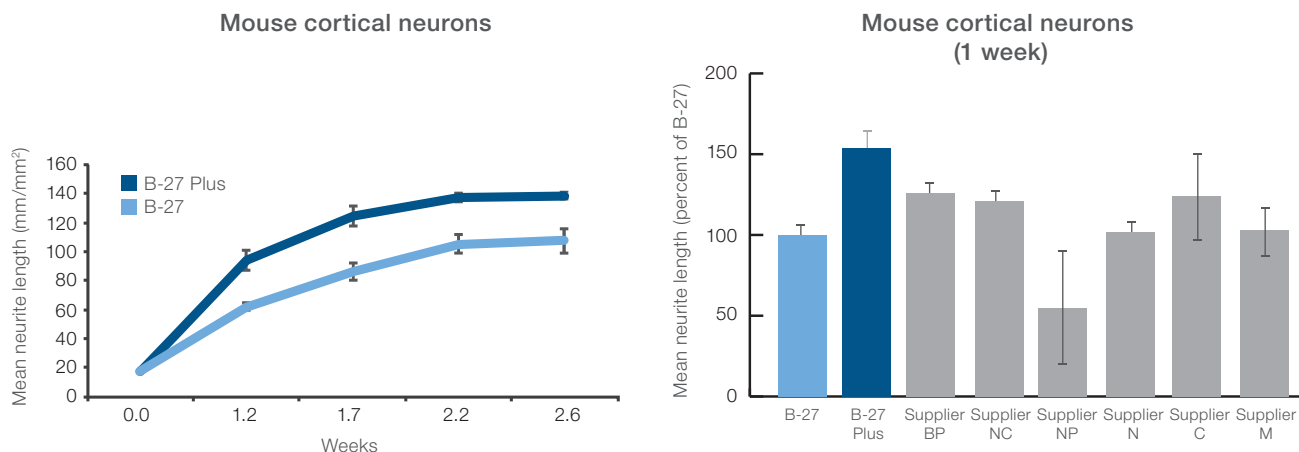
# Improved neuronal survival in long-term cultures compared to alternative media systems

## Neuronal survival at 3 and 4 weeks



**Figure 3. The B-27 Plus Neuronal Culture System achieves the highest neuronal survival in long-term cultures compared to other commercially available serum-free neuronal media systems.** Primary Rat Cortex Neurons (Cat. No. A10840), Gibco™ Primary Rat Hippocampus Neurons (Cat. No. A10841), Gibco™ Primary Mouse Cortical Neurons (Cat. No. A15586), and human iPSC-derived neurons that had been matured for 9 days before cryopreservation were thawed in classic Neurobasal Medium with B-27 Supplement. The neurons were plated onto poly-D-lysine-coated 96-well plates and maintained for 3–4 weeks in the B-27 Plus Neuronal Culture System and alternative serum-free supplemented media systems following the suppliers' recommended protocols. Neuronal survival was quantitated by immunofluorescent labeling using Invitrogen™ HuC/HuD Monoclonal Antibody (Cat. No. A21271) at 3 or 4 weeks. Data shown are from one of three experiments, with each run showing that B-27 Plus Neuronal Culture System enables the highest neuronal cell survival among the alternative systems.

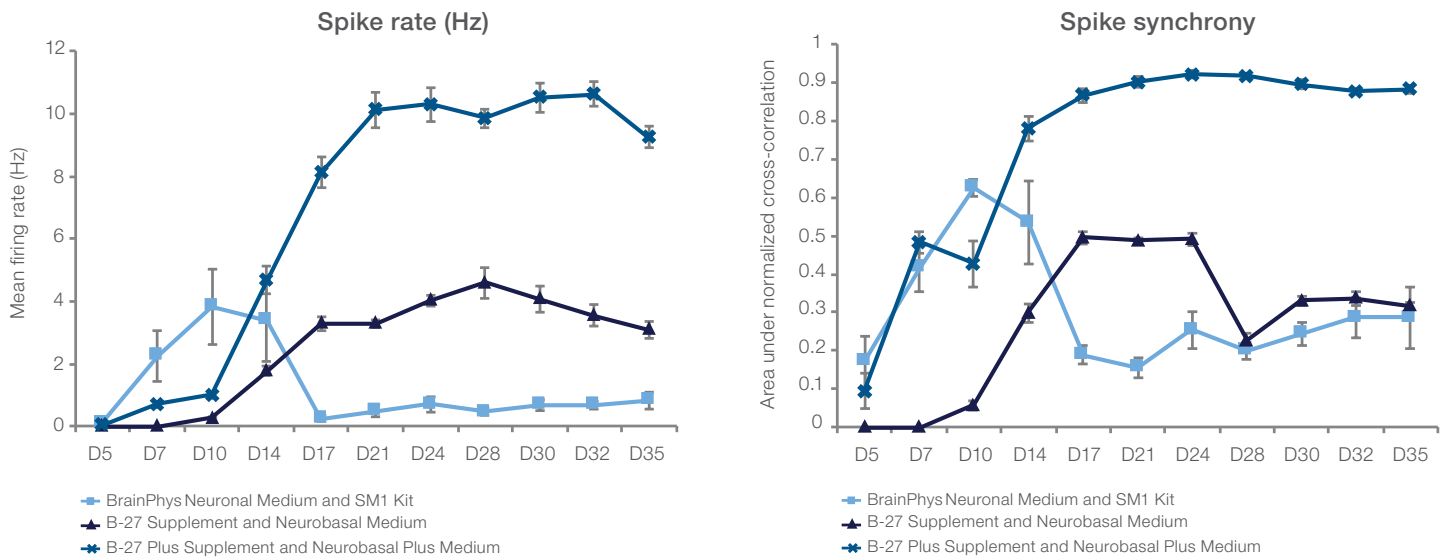
## Accelerates and increases neurite outgrowth



**Figure 4. Acceleration of neurite outgrowth compared to B-27 Supplement and Neurobasal Medium and alternative media systems.** Primary Mouse Cortical Neurons (Cat. No. A15586) were thawed in classic Neurobasal Medium with B-27 Supplement and plated onto poly-D-lysine-coated 96-well plates. Neurons were maintained for ~3 weeks in Neurobasal Medium with B-27 Supplement, Neurobasal Plus Medium with B-27 Plus Supplement, and other commercially available serum-free neuronal media systems following the suppliers' recommended protocols. Neurite outgrowth was quantitated using differential interference contrast images taken at the time points specified. The B-27 Plus Neuronal Culture System significantly accelerates neurite outgrowth over the first few weeks compared to classic Neurobasal Medium with B-27 Supplement. Data shown are from one of three experiments, with each run showing that after 1 week, the B-27 Plus Neuronal Culture System enabled the highest mean neurite length compared to all alternative systems.

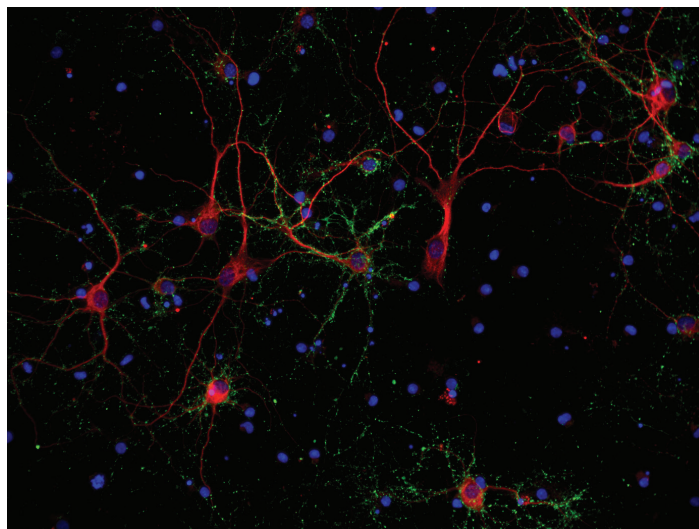


# Improves electrophysiological activity and maturity

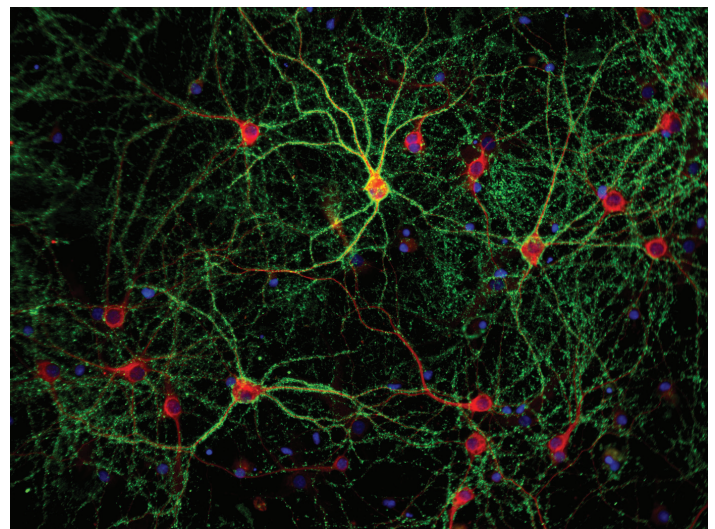


**Figure 5. Improvement of electrophysiological activity compared to BrainPhys Neuronal Medium and SM1 Kit.** Primary Rat Cortex Neurons (Cat. No. A10840) were plated on to 48-well multi-electrode array (MEA) plates. Cells were cultured for 35 days in the listed media system following the suppliers' recommended protocols. Spontaneous electrophysiological activity was recorded throughout with the Axion BioSystems™ Maestro™ MEA platform. Data shown are from one of four experiments, with each run showing similar trends. The B-27 Plus Neuronal Culture System enabled more consistent, stable, and highly synchronized spontaneous electrophysiological activity over time.

## B-27 Supplement and Neurobasal Medium



## B-27 Plus Supplement and Neurobasal Plus Medium



**Figure 6. Enhancement of neuronal maturation with B-27 Plus Supplement and Neurobasal Plus Medium compared to that with B-27 Supplement and Neurobasal Medium.** Primary Rat Cortex Neurons (Cat. No. A10840) at day 22 were stained for the dendritic marker MAP2 (red) and synapsin 1/2 to label presynaptic terminals (green). Neurons maintained in the B-27 Plus Neuronal Culture System had significantly higher synaptic-positive puncta.



## Recommended applications and ordering information

B-27 Plus Supplement and Neurobasal Plus Medium come in the same formats and will work in the same protocols as the classic B-27 and Neurobasal products. Get advice on how to successfully transition to

the B-27 Plus Neuronal Culture System using our newly updated protocol guide. We recommend using the Plus versions and other combinations of medium and supplement in the following applications:

Application	Recommended supplement(s)	Recommended basal medium
Maintenance and maturation of prenatal primary neurons	B-27 Plus Supplement	Neurobasal Plus Medium
Differentiation, maintenance, and maturation of stem cell-derived neurons	B-27 Plus Supplement and CultureOne Supplement	Neurobasal Plus Medium
Electrophysiology studies	B-27 Plus Supplement	Neurobasal Plus Medium
Expansion of neural stem cells	StemPro NSC SFM Supplement	KnockOut DMEM/F-12 Basal Medium
Maintenance and maturation of postnatal and adult brain neurons	B-27 Supplement	Neurobasal-A Medium
Studies of insulin secretion or insulin receptors	B-27 Supplement minus insulin	Neurobasal-A Medium
Studies of oxidative stress and damage, apoptosis, or where free-radical damage to neurons occurs	B-27 Supplement minus antioxidants	Neurobasal Medium
Product	Quantity	Cat. No.
B-27 Plus Neuronal Culture System		A3653401
B-27 Plus Supplement (50X)	10 mL	A3582801
Neurobasal Plus Medium	500 mL	A3582901



Find out more at [thermofisher.com/b27plus](https://thermofisher.com/b27plus)

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