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# Ready-to-use kit Neuronal-LSB3i (x1000)

## **Cocktail Formulation:**

LSB: 0.1 mM LDN-193189 and 10 mM SB431542 in DMSO solution 3i: 3 mM CHIR99021, 10 mM SU5402 and 10 mM DAPT in DMSO solution

## **Effective Concentration in Cell Culture:**

LSB: 0.1 µM LDN-193189 and 10 µM SB431542 3i: 3 µM CHIR99021, 10 µM SU5402 and 10 µM DAPT

Purity:  $\geq 98\%$  for each compound Storage: 4°C for 3 month. -20°C for 6 month.

Recently published in *Nature Biotechnology* (1) by Chambers et al, LSB3i is a combination of five smallmolecule pathway inhibitors that significantly improves the conventionally slow and inefficient neuronal differentiation of human ESCs or iPSCs. Under the LSB3i treatment, human ESCs or iPSCs very rapidly differentiate into functional neurons with over 75% efficiency within 10 day of differentiation. The resulting neurons exhibit typical markers and functional properties of human nociceptors, including tetrodotoxin (TTX)-resistant, SCN10A-dependent sodium currents and response to nociceptive stimuli, such as ATP and capsaicin. This convenient, robust, and scalable generation of functional human nociceptors from human ESCs/iPSCs using LSB3i provides a platform for basic research, disease modeling, and drug discovery related to pain studies.

XcessBio's Neuronal-LSB3i contains two combinations/components: LSB (combination of LDN193189/BMP inhibitor and SB431542/TGF $\beta$  inhibitor) and 3i (combination of CHIR99021/GSK3 inhibitor, SU5402/FGF-VEGF-PDGF inhibitor, DAPT/Notch inhibitor).

### How to Use:

**In vitro:** Simply add/dilute the received LSB3i stock solutions from each vial at 1:1000 into your hESC/iPSC differentiation basal media to make the LSB and 3i induction media. LSB is the first 2-small-molecules combination used on day 0-5; and 3i is the second 3-small-molecules combination used on day 2-10.

#### **Reference:**

- 1. Chambers SM, et al. Combined small-molecule inhibition accelerates developmental timing and converts human pluripotent stem cells into nociceptors. (2012) Nature Biotechnology. In press.
- Sun, L. et al. Design, synthesis, and evaluations of substituted 3-[(3- or 4-carboxyethylpyrrol-2yl)methylidenyl]indolin-2-ones as inhibitors of VEGF, FGF, and PDGF receptor tyrosine kinases.(1999) J. Med. Chem. 42, 5120–5130.
- Bennett, C.N. et al. Regulation of Wnt signaling during adipogenesis. (2002) J. Biol. Chem. 277, 30998–31004.
- 4. Dovey, H.F. et al. Functional gamma-secretase inhibitors reduce beta-amyloid peptide levels in brain. (2001) J. Neurochem. 76, 173–181.

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