

# CNS-3D Organoids

Brain organoids enable preclinical safety assessments and efficacy evaluations that deliver critical translational data earlier in the pipeline.

## Overview

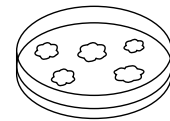
CNS-3D Organoids are a high-throughput *in vitro* human model for investigating neurotoxicity, seizure liability, synaptogenesis, neurogenesis, excitotoxicity, ion channel activity, and neuropharmacology. These brain organoids support multiplexed, high-content analysis across diverse therapeutic modalities, including small molecules, siRNAs, antisense oligonucleotides (ASOs), adeno-associated viruses (AAVs), and antibody-based therapeutics.

## CNS-3D Organoids

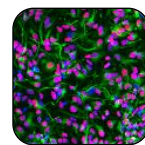
CNS-3D Organoids are derived from donor-induced pluripotent stem cells (iPSCs) that are differentiated into neural progenitor cells (NPCs). These NPCs self-organize into a balanced 50:50 co-culture of neurons and astrocytes, closely recapitulating key features of the human brain. The organoids grow to diameters of 500–800  $\mu\text{m}$  and develop complex, functional neural networks that exhibit spontaneous, electrically driven activity. This coordinated network behavior can be captured in real time using high-throughput functional assessment platforms such as FLIPR. Validated through transcriptomic analysis and subjected to a rigorous quality control process, CNS-3D Organoids deliver consistent, physiologically relevant performance across production lots, ensuring reliability for research and therapeutic screening applications.

**Schematic of the CNS-3D organoid generation procedure.** Induced pluripotent stem cells (iPSCs) are expanded before being derived into neural progenitor cells (NPCs), which is then cryopreserved to serve as the starting material for each CNS-3D organoid production batch. Over the course of 6-weeks, organoids grow to diameters between 500–800 $\mu\text{m}$ , differentiate into both neurons and astrocytes, and exhibit spontaneous coordinated network activity. Shown above are waveforms measured on FLIPR (Molecular Devices).

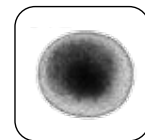
iPSC Adaption  
& Expansion



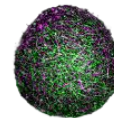
NPC Derivation  
& Biobanking



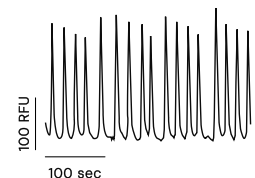
Growth



Organoid  
Characterization

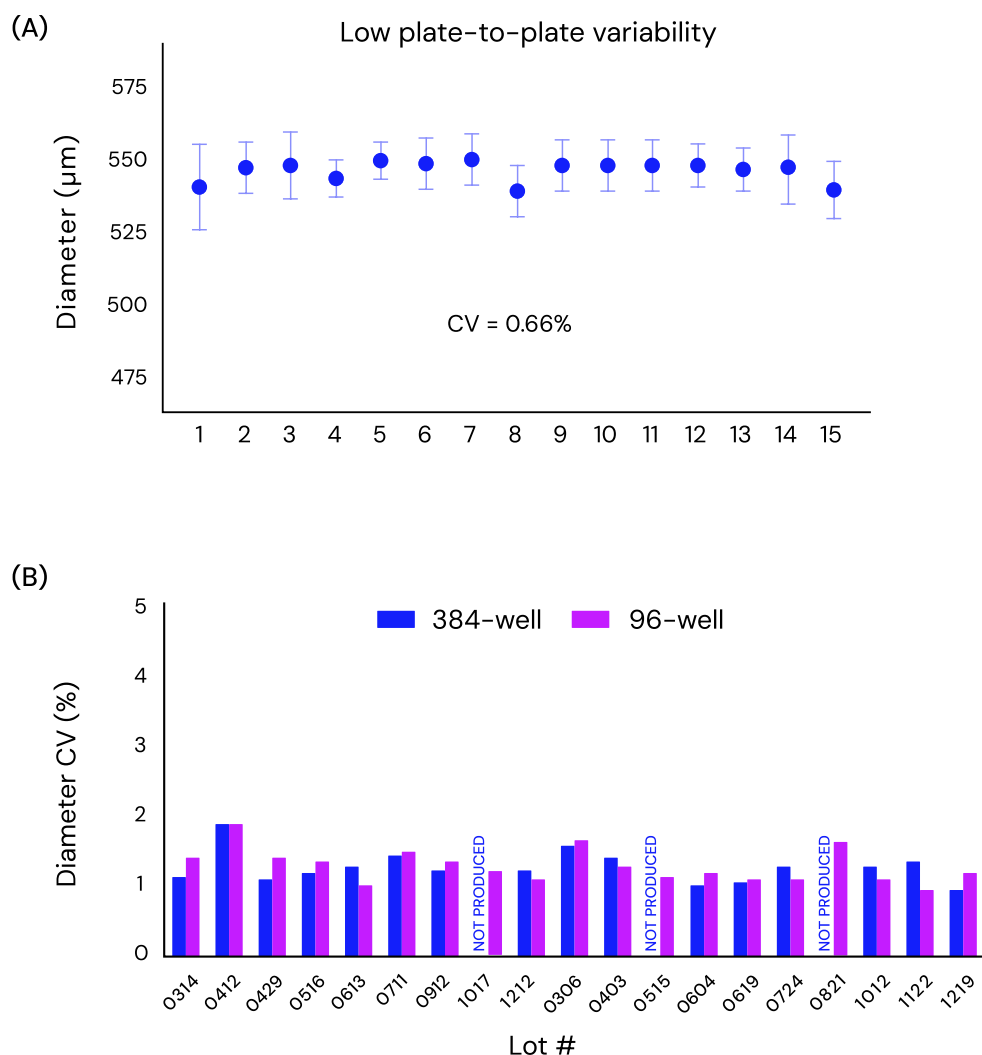


Spontaneous  
Activity



## Reproducibility & Stability

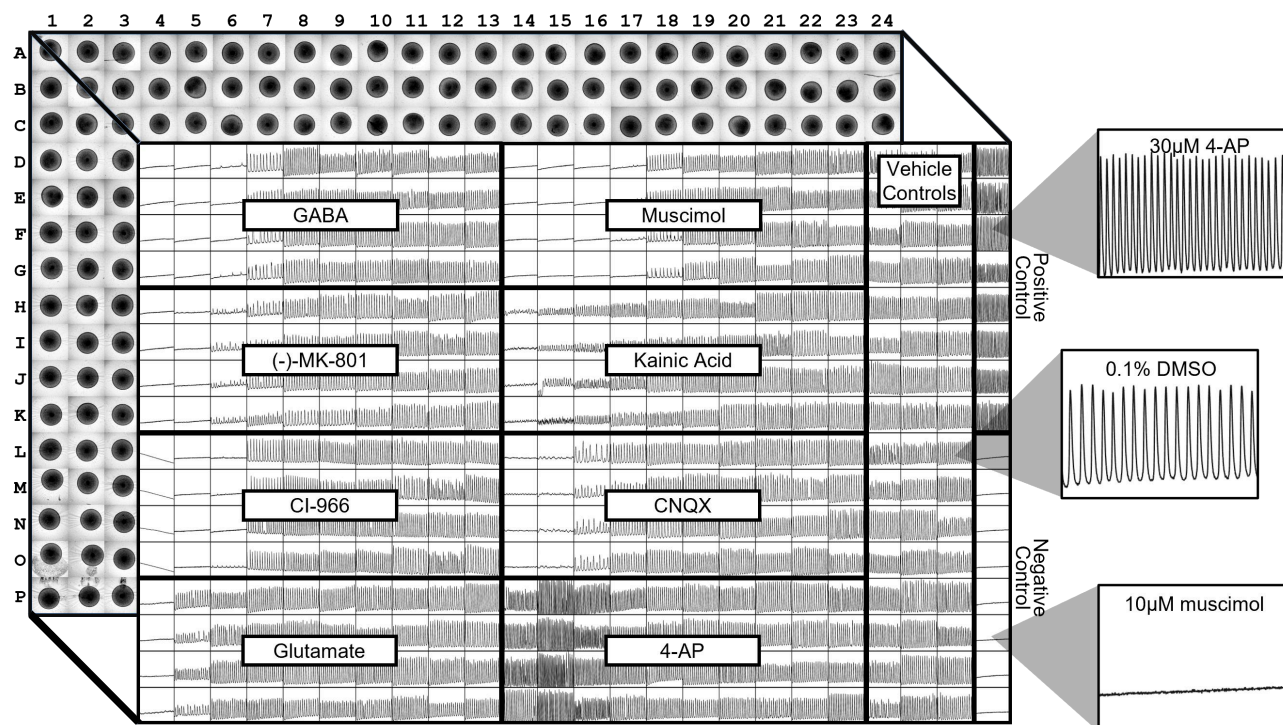
CNS-3D Organoids demonstrate consistent performance, reducing the need for repeated experiments. Their stable pharmacological responses enable longitudinal and chronic treatment paradigms over several weeks. A validated four-week use window allows for flexible assay workflows while maintaining data quality.



(A) CNS-3D organoids are batch-produced in monthly builds and exhibit exceptional uniformity in diameter—both within individual plates and across plates in a single batch—overcoming the morphological variability that typically plagues organoid biology. (B) The remarkable batch-to-batch reproducibility in organoid diameter has been demonstrated across both 384-well and 96-well lots historically, with CVs across batches guaranteed under 5%.

## Quality Control Of Organoid Production

Organoid production undergoes rigorous quality control, including functional characterization and brightfield imaging to verify consistent organoid size (500–800  $\mu\text{m}$  diameter with <5% CV within plate) and a high presence rate (98%). Immunocytochemistry (ICC) is used to confirm the expected neuron-to-astrocyte ratio, and sterility is ensured through testing for mycoplasma, anaerobic, aerobic, and fungal contaminants. Each shipment includes a certificate of analysis detailing these metrics, along with EC/IC50 values and robust Z-prime scores for FLIPR-based functional screening.



384-wells of functional activity can be captured simultaneously using high-throughput kinetic plate readers such as FLIPR. Shown above are the dose-dependent changes in waveform activity in response to 8 neuromodulators including positive control K<sup>+</sup> blocker, 4-AP, and GABA analog, muscimol. The waveforms demonstrate remarkable reproducibility between replicates, enabling the use of as few as 4 organoids per condition.

## Technical Specifications

Specification	Details
Formats	96-well and 384-well microplates
Organoid Size	500–800 $\mu\text{m}$ diameter, CV <5% within and between plates
Cell Composition	Excitatory (glutamatergic) neurons and supporting astrocytes
Use Window	Validated assay window from week 6 to week 10 of maturation
Assays	Neuromodulation (FLIPR), viability (CellTiter-Glo), cytotoxicity (LDH-Glo), high-content imaging, transcriptomic profiling
Compatible Modalities	Small molecules, siRNAs, antisense oligonucleotides (ASOs), adeno-associated viruses (AAVs), and antibody therapeutics

## Ordering Information

CNS-3D Organoids are available in ready-to-use 96- and 384-well microplate formats, CNS-3D organoids are compatible with automated liquid handling systems, seamlessly integrating into compound screening workflows and streamlining time to data acquisition.

A proprietary media hibernation system allows for shipping to any lab around the world.

Product Name	Description	Modality Type	Catalog Number
CNS-3D Organoids	96-well, assay ready microplates	Human	500096
CNS-3D Organoids	384-well, assay ready microplates	Human	500384

→ To learn more, visit: [28bio.com/cns-3d-technology](https://28bio.com/cns-3d-technology)